STATE OF MAINE
DEPARTMENT OF ENVIRONMENTAL PROTECTION

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GOVERNOR

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December 27, 2017

Ms. Kimberly Bose, Secretary
Federal Energy Regulatory Commission
888 First Street, N.E.
Washington, D.C. 20426

Subject: FERC 4784 – Pejepscot Hydroelectric Project
Pre-Application Document Comments
Study Request Submission

Dear Ms. Bose:

The Maine Department of Environmental Protection (MEDEP or Department) received and reviewed a Notice of Intent to File License Application, Filing of Pre-Application Document (PAD), Commencement of Pre-Filing Process, and Scoping; Request for Comments on the PAD and Scoping Document, dated October 30, 2017. MEDEP staff attended a scoping meeting on November 28, 2017 and reviewed appropriate project documents to prepare the following comments and recommendations.

The proposed relicensing is subject to Water Quality Certification provisions of Section 401 of the Federal Water Pollution Control Act (a.k.a. Clean Water Act). By Executive Order of the governor of the State of Maine, the Maine Department of Environmental Protection is the State certifying agency for projects located wholly or in part in organized towns and cities, and as such, has jurisdiction over the Pejepscot Hydroelectric Project.

The existing Pejepscot Hydroelectric facilities consist of a 560-foot-long, 48-foot-high, rock and gravel filled timber crib overflow structure that is topped with a 5-foot-thick reinforced concrete slab; a spillway consisting of five 96-foot-long by 3-foot -high bascule gates; a 225-acre impoundment at full pool elevation of 67.5 feet; a powerhouse containing three horizontal Francis turbine-generator units with a combined rated capacity of approximately 1,580 kW and maximum hydraulic capacity of approximately 1,050 cfs; a powerhouse containing a vertical-shaft propeller type (Kaplan)turbine rated at 17,000 hp with a maximum hydraulic capacity of approximately 7,100 cfs and a generator rated at approximately 12,300 kW; an upstream fish passage facililty consisting of a vertical fish lift; a downstream fish passage facility consisting of two-four-foot wide entry weirs that pass fish through 30-inch and 24-inch outlet pipes respectively; and appurtenant facilities.

Formal and informal recreation facilities associated with the Project, include impoundment boat launches and fishing access; parking for the recreational facilities is limited.

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Topsham Hydro Partners Limited Partnership (Topsham Hydro) proposes no changes in project operation, and plans to continue to operate and maintain the Pejepscot Hydroelectric Project as required under its current license (P-5362). No new development is proposed.

Comments on PAD

The Department appreciates the effort that Topsham Hydro and their consultants have made to prepare a Pre-Application Document. The PAD provides an understanding of the Project, the surrounding resources, and proposed dam operation. The PAD highlights the issues related to dam operations and relicensing that should be investigated to ensure that operations do not have a negative impact on the resources of the Androscoggin River.

1. The Department understands that in order to obtain a federal license or permit, the applicant is required to obtain water quality certification from states where a discharge occurs. Sufficient high quality data is necessary for the Department to assess whether the presence and operation of the Pejepscot Hydroelectric Project impacts water quality, and whether water quality standards, including designated uses, narrative criteria and numeric criteria, are met in the Androscoggin River, including in impoundments created by the Pejepscot Hydro dam. Water quality studies to collect such data are necessary to provide such data as needed by the agencies. To ensure that studies are conducted in a manner to collect the requisite data, a number of water quality studies are required.

Water Quality Classifications and Standards

Water Quality Standards and the water quality classifications of all surface water of the State have been established by Maine Legislature (Title 38 M.R.S. §§ 464-467). The following classifications apply to the water affected by Pejepscot Hydroelectric Project:

Androscoggin River from its confluence with the Ellis River to a line formed by the extension of the Bath-Brunswick boundary across Merrymeeting Bay in a northwesterly direction - Class C

Class C waters must be of such quality that they are suitable for the designated uses of drinking water supply after treatment; fishing; agriculture; recreation in and on the water; industrial processes and cooling water supply; hydroelectric power generation; navigation; and as habitat for fish and other aquatic life.

The dissolved oxygen content of Class C waters shall be not less than 5 parts per million or 60% of saturation, whichever is higher, except that in identified salmonid spawning areas where water quality is sufficient to ensure spawning, egg incubation and survival of early life stages, that water quality sufficient for these purposes must be maintained. In order to provide additional protection for the growth of indigenous fish, the following standards apply.

(1) The 30-day average dissolved oxygen criterion of a Class C water is 6.5 parts per million using a temperature of 22 degrees centigrade or the ambient temperature of the water body, whichever is less, if:
    a. A license or water quality certificate other than a general permit was issued prior to March 16, 2004 for the Class C water and was not based on a 6.5 parts per million 30-day average dissolved oxygen criterion; or
    b. A discharge or a hydropower project was in existence on March 16, 2005 and required but did not have a license or water quality certificate other than a general permit for the Class C water.
    This criterion for the water body applies to licenses and water quality certificates issued on or after March 16, 2004.
(2) In Class C waters not governed by subparagraph (1), dissolved oxygen may not be less than 6.5 parts per million as a 30-day average based upon a temperature of 24 degrees centigrade or the ambient temperature of the water body, whichever is less. This criterion for the water body applies to licenses and water quality certificate3s issued on or after March 16, 2004.

Discharges to Class C water may cause some changes to aquatic life, except that the receiving waters must be of sufficient quality to support all species of fish indigenous to the receiving waters and maintain the structure and function of the resident biological community.

Antidegradation

The State's antidegradation policy provides that water quality certification may be approved only if the applicable standards of classification of the affected water body are met and existing in-stream uses and the level of water quality necessary to protect those existing uses are maintained and protected. The policy also provides that, where the actual quality of any classified water exceeds the minimum standards of the next highest classification, that higher water quality classification shall be maintained and protected.

Water Quality Certification Data Requirements

To certify that the hydropower project does not cause or contribute to non-attainment of Maine's Water Quality Standards, under section 401 of the Federal Water Pollution Control Act, the applicant must demonstrate that designated uses, numeric criteria and narrative criteria are met in the Project impoundments and downstream of the project tailrace. The applicant proposed a number of water quality studies or other resource studies for the relicensing of the Pejepscot Hydroelectric Project. Water quality studies of the impoundments and tailrace are necessary to evaluate the impact of proposed project operations on the Androscoggin River and to determine if continued operations under a new project license can be expected to meet Maine's water quality standards. Such assessment is provided by issuance of a water quality certification, pursuant to authority delegated by the United States Environmental Protection Agency to the States where a discharge occurs; in this case the discharge is the waters affected by the operation of a hydropower facility and the discharge over the spillway occur in the State of Maine. It has

been the Department's practice to determine the metrics, methods, timing, and duration of water quality monitoring necessary to ensure that the water quality studies meet data quality objectives for certification. Therefore, the Department requests that the applicant conduct water quality studies that include the following parameters and adhere to the Department's established sampling protocols in support of water quality certification.

Impoundment Tropic State Study –The applicant has proposed to conduct an Impoundment Trophic State Study to demonstrate that the impoundment exhibits a steady or improving trophic state, and that Maine's water quality standards are met; therefore, the Department is requesting that the Impoundment Trophic State Study be conducted in accordance with established sampling protocols, including sample collection and analysis parameters, as provided under "Lakes, Ponds, and Impoundments" in DEP Sampling Protocol for Hydropower Studies – November 2014, which is attached to this comment letter.

Impoundment Aquatic Habitat Study – The purpose of this study is to determine the effect of impoundment drawdowns on the impoundment's littoral zone and the ability of the impoundment to support fish and other aquatic life. The Pejepscot Hydroelectric Project is operated in run-of river mode and there is no significant impoundment drawdown during normal operations; therefore, no impact to littoral habitat in the impoundments is expected and no Impoundment Aquatic Habitat Study is necessary.

Temperature and Dissolved Oxygen Study – The applicant will need to conduct a temperature and dissolved oxygen study in the impoundment and in the tailwater of the Pejepscot Hydroelectric Project to demonstrate compliance with Maine water quality standards. Data must be collected in the Androscoggin River below the Pejepscot dam in accordance with the Department's "Temperature and Dissolved Oxygen Study" protocol under "Rivers and Streams" in DEP Sampling Protocol for Hydropower Studies – November 2014, and at the deepest location within the impoundment in accordance with the Department's protocol for Lakes, Ponds, and Impoundment Trophic State Study, which is attached to this comment letter. As noted in the protocol, the applicant will need to consult with the Department to verify representative sampling locations as the study plans are developed.

Benthic Macroinvertebrate (BMI) Monitoring – Assessment of the macroinvertebrate community is critical to determine whether current in-stream flow releases affect attainment of classification standards for habitat and aquatic life in the Androscoggin River below the Project. A BMI study is proposed by the applicant, to determine the current structure of the community and to evaluate any impacts caused by project operations. To ensure data meets water quality certification compliance objectives, the study plan must be developed in accordance with the Department's Methods for Biological Sampling and Analysis of Maine's Rivers and Streams, which is attached to this comment letter. Similar to the Temperature and Dissolved Oxygen Study, the applicant will need to consult with the Department to verify representative sampling locations as the study plan is developed.

Aquatic Habitat Cross-Section Flow Study - This study evaluates whether current in-stream flow releases are affecting attainment of habitat standards for fish and other aquatic life in the Androscoggin River below the Project dam. It is the Department's position that there must be both sufficient quality and quantity of habitat for aquatic organisms to meet aquatic life and habitat standards. The Pejepscot Hydroelectric Project is operated in a run-of-river mode, with a continuous minimum flow of 1,710 cfs, or inflow, whichever is less, below the Project. The applicant is not proposing any changes to existing operations, therefore continued operations are expected to provide and maintain aquatic habitat and so no cross-section flow study is necessary.

Mercury Study - This study assesses the impact of impoundment drawdown on the bioavailability of mercury. Mercury contamination in Maine lakes is well documented. The largest source of mercury appears to be atmospheric deposition from out of state sources, however study suggests that there may be a correlation between lake drawdowns and the bioavailability of mercury in the form of methyl mercury. Operating under run-of-river mode requires no significant drawdown under normal operating conditions; therefore no Mercury Study is necessary.

In addition to meeting requirements of the water quality certification process, MDEP supports study requests prepared by other natural resource agencies, including but not limited to, Maine Department of Inland Fish and Wildlife (MDIFW), Maine Department of Marine Resources (MDMR), and the US Fish and Wildlife (USFWS).

Thank you for the opportunity to comment on the Pre-Application Document for the Pejepscot Hydroelectric Project. If you have any questions, please contact me by phone at (207) 446-2642 or by email at Kathy.Howatt@maine.gov.

Sincerely,

Kathy Davis Howatt
Hydropower Coordinator
Bureau of Land Resources

Encl: DEP Sampling Protocol for Hydropower Studies (November 2014)
DEP Methods for Biological sampling and Analysis of Maine's Rivers and Streams

DEP SAMPLING PROTOCOL FOR HYDROPOWER STUDIES November 2014

LAKES, PONDS, AND IMPOUNDMENTS

Trophic State Study

Sampling personnel must be certified annually for this sampling protocol by DEP's Division of Environmental Assessment Lakes Section.

Each basin shall be sampled at the deepest location twice each month for at least five consecutive months during one open water season as follows.

Parameter	Sampling method	Detection limits
Secchi disk transparency	water scope	0.1 meter
Temperature	profile*	0.1 C
Dissolved oxygen	profile*	0.1 mg/l
Total phosphorus	epilimnetic core	0.001 (DEP method)
Chlorophyll a	epilimnetic core	0.001
Color	epilimnetic core	1.0 SPU
pH	epilimnetic core	0.1 SU
Total alkalinity	epilimnetic core	1.0 mg/l

*Profiles shall consist of temperature and dissolved oxygen measurements taken every meter up to 15 meters, every other meter to 25 meters, then every 5 meters thereafter.

In addition, during late summer (mid to late August depending on latitude and weather conditions), water samples shall be collected and analyzed from up to three depths in the water column for the parameters below except Chlorophyll *a*. If the waterbody is thermally stratified ($\Delta T \geq 1°C/m$ at any depth below the top 3 m depth), samples will be collected from an epilimnetic core, at the top of the hypolimnion, and at one meter above the sediment. If the waterbody is not thermally stratified, only one sample is needed, that being from an integrated core from the surface to two times the Secchi disk depth or within 1 m of the bottom whichever is less.

Parameter	Detection limit
Total phosphorus	0.001 mg/l
Nitrate	0.01 mg/l
Chlorophyll a (uncorrected)	0.002 mg/l (trichromatic determination)
Color	1.0 SPU
DOC	0.25 mg/l
pH	0.1 SU
Total alkalinity	1.0 mg/l
Total iron	0.005 mg/l
Total dissolved aluminum	0.010 mg/l
Total calcium	1.0 mg/l
Total magnesium	0.1 mg/l
Total sodium	0.05 mg/l
Total potassium	0.05 mg/l
Total silica	0.05 mg/l
Specific conductance	1 ms/cm
Chloride	1.0 mg/l
Sulfate	0.5 mg/l

Additional sampling may be required due to the hydraulic or physical characteristics of a given waterbody or to the presence of significant water quality problems.

Habitat Study

For lakes, ponds, and riverine impoundments, determination of attainment of the designated use 'habitat for fish and other aquatic life' will be determined as follows. Using a depth of twice the mean summer Secchi disk transparency, determined from the Trophic State Study or historic DEP data, as the bottom of the littoral zone, the volume and surface area dewatered by the drawdown will be calculated to determine if at least 75% of the littoral zone remains watered at all times. Alternatively, studies of fish and other aquatic life communities, including freshwater mussels, may be conducted to demonstrate that the project maintains 'structure and function of the resident biological community' despite a drawdown that results in less than 75% of the littoral zone remaining watered at all times.

Fishing (Mercury Contamination) Study

To ensure that the project does not contribute to the Statewide Fish Consumption Advisory due to mercury, projects with excessive drawdowns (generally >10 feet) may be required to analyze sport fish from the project waterbody and one or more reference waters for mercury. Contact DEP for specific requirements for each project.

RIVERS AND STREAMS

Temperature and Dissolved Oxygen Study

Applicability

This rivers and streams sampling protocol shall apply to tailwater areas that are not impoundments where existing data are insufficient to determine existing and future water quality.

Sampling Stations

Sampling shall occur in the tailwater downstream from the turbine/gate outlet or dam at a location representative of downstream flow as agreed by DEP on a case by case basis. Initially, measurements of temperature and dissolved oxygen should be made along a transect across the stream at the first, second and third quarter points across the width. If there is no violation of dissolved oxygen criteria and no significant (<0.4 mg/l) difference in concentrations among the quarter points, subsequent measurements may be made at the location shown to be representative of the main flow. Otherwise, measurements should be made at the location of the lowest concentration and the location of the main flow. Sampling should also occur in any bypassed segment of the river created by the project. Additional sampling stations may be required in the upstream or downstream areas where significant point or nonpoint sources exist or where slow moving or deep water occurs. The number and spacing of any additional stations will be determined by DEP on a case-by-case basis.

Parameters

Temperature and dissolved oxygen shall be sampled at mid-depth in rivers less than 2 m deep or in a profile of 1 meter increments of depth in rivers greater than 2 m deep. In rivers where it is already known that attainment of required statutory dissolved oxygen criteria is questionable, sampling for additional parameters (e.g. BOD, nitrogen, phosphorus) may be necessary.

Frequency and Timing

Sampling should be conducted during the summer low flow high temperature period, with the ideal conditions being the 7Q10 flow (the 7 day average low flow with a 10 year recurrence interval) combined with daily average water temperatures exceeding 24 ^{o}C. Measurements of temperature and dissolved oxygen shall be made every hour with a datasonde in remote unattended mode continuously during July and August, unless high flows well above seasonal median flows occur.

Alternatively, with concurrence by DEP, sampling could be undertaken one day per week for a minimum of ten weeks throughout the summer low flow, high temperature period. Each discrete grab sampling event for temperature and dissolved oxygen would consist of a minimum of two daily runs, the first of which should occur before 7 AM and the second of which should occur after 2 PM. Sampling results will not be considered complete unless a minimum of 5 sampling days meets the following conditions: The product of the water temperature (^{o}C) and the flow duration (the percentage of the time a given flow is statistically exceeded) at the time of sampling exceeds 1500. For cycling hydropower projects, in addition to twice daily monitoring, continuous monitoring may be required at some locations for a duration equivalent to the period of one cycle of the storage and the release of flow.

For either method, a summer in which low flows and high temperatures are not experienced may result in additional sampling requirements for the next summer. Low flow conditions may occur naturally, as an unregulated river or may be artificially induced, as in the case of upstream flow regulation or flows downstream from a cycling or peaking power project or in the case of a bypassed segment which receives flow only by spillage, leakage or specific releases.

Available Data

The use of data already available is encouraged provided that adequate QA/QC procedures have been followed. Old data may not be acceptable for considerations of meeting minimum sampling requirements, but could still provide useful information. Acceptance/rejection of data will be determined on a case by case basis, but generally data more than 10 years old may be rejected.

Habitat and Aquatic Life Studies

For rivers and streams, determination of attainment of the designated use 'habitat for fish and other aquatic life' will be determined as follows. A Cross-Section Flow Study is required that measures width and depth at various flows to determine the flow at which at least 75% of the bank full cross-sectional area of the river or stream is continuously watered. At least three cross-sections representative of the river or stream must be measured. Alternately, a combination of ambient measurements in one cross-section, flow data from existing flow gages, and/or modelling may be approved by DEP.

In addition, to determine if the project 'attains the aquatic life criteria, i.e. 'maintains the structure and function of the resident biological community', biological monitoring of the benthic macroinvertebrate community must be conducted following DEP's standard protocol in Methods for Biological Sampling and Analysis of Maine's Rivers and Streams, DEP LW0387-B2002.
A copy can be found at www.maine.gov/dep/water/monitoring/biomonitoring/material.html

Methods for Biological Sampling and Analysis of Maine's Rivers and Streams

Susan P. Davies
Leonidas Tsomides

DEP LW0387-C2014
Revised April, 2014

MAINE DEPARTMENT OF ENVIRONMENTAL PROTECTION

METHODS

FOR

BIOLOGICAL SAMPLING AND ANALYSIS OF

MAINE'S RIVERS AND STREAMS

Susan P. Davies

Leonidas Tsomides

Maine Department of Environmental Protection
Bureau of Land and Water Quality
Division of Environmental Assessment
Augusta, Maine 04333
January, 1987

Revised April, 2014

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CONTENTS

FOREWORD

This manual describes the field, laboratory and data preparation methods required by the Maine Department of Environmental Protection to collect and analyze benthic macroinvertebrate samples for the River and Stream Biological Monitoring Program. The biological classification of Maine's inland waters was authorized by the Maine State Legislature with the passage of Public Law 1985 Chapter 698 - The Classification System for Maine Waters. This law states that it is the State's objective "to restore and maintain the chemical, physical and biological integrity" of its waters, and establishes a water quality classification system to enable the State to manage its waters so as to protect their quality. The classification system further establishes minimum standards for each class, which are based on designated uses, and related characteristics of those uses, for each class of water.

Each water quality class contains standards that, among other things, describe the minimum condition of the aquatic life necessary to attain that class. The Maine Department of Environmental Protection (the Department) has developed numeric criteria in support of the narrative aquatic life standards in the Water Quality Classification Law. The Department has collected a large, standardized database consisting of benthic macroinvertebrate samples from above and below all significant licensed discharges in the State, from areas impacted by non-point sources, as well as from relatively unperturbed areas. These sampling locations were chosen to represent the range of water quality conditions in the State. This information has been used to develop numeric criteria which are specific to the natural biotic community potential of the State of Maine (see Davies et al., 1995 and 1999 for a description of the development and application of numeric criteria) and is established in DEP regulation Chapter 579 : Classification Attainment Evaluation Using Biological Criteria for Rivers and Streams.

Standardization of data collection and analytical methods is fundamental to the consistent, unbiased and scientifically sound evaluation of aquatic life impacts. This manual sets forth the standardized practices and procedures used by the Department to acquire or accept benthic macroinvertebrate data for use in regulation, assessment or program development.

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I GENERAL METHODS FOR RIVER AND STREAM AQUATIC LIFE CLASSIFICATION ATTAINMENT EVALUATION

Each water quality class is defined by standards that describe the minimum condition of the aquatic community necessary to attain that class. The benthic macroinvertebrate community is used as an indicator community of the general state of the aquatic life in flowing waters for the purpose of assessment of classification attainment. Standardized sampling techniques and sample analysis are required for assessment of biological attainment of stream water quality classification. This manual presents the standard practices and procedures that have been adopted by the Department to acquire benthic macroinvertebrate data for purposes of aquatic life classification attainment evaluation.

Purpose:

To determine the water quality class attained by a particular river or stream reach in terms of the aquatic life standards set forth in 38 MRSA Sec. 465 (The Classification System for Maine Waters).

Requirements:

All samples of aquatic life that are collected for purposes of classification attainment evaluation, whether collected by the Department or by any party required to make collections by the Department, must be collected, processed and identified in conformance with the standardized methods outlined in this manual. Selection of appropriate sampling sites and micro-habitat to sample, as well as procedures for quantitative analysis of the sample must conform to methods set forth in this manual. Data submitted by any party required to make collections by the Department must be accompanied by a Quality Assurance Plan, approved by the Commissioner.

1. Qualifications of Sampling Personnel

Biological sampling must be performed by a professional aquatic biologist or by qualified personnel under the supervision of a professional aquatic biologist. The professional aquatic biologist must have, as a minimum, a Bachelor of Science degree in biological sciences with aquatic entomology, invertebrate zoology, fisheries or closely related specialization, and greater than 6 months experience working with macroinvertebrate sampling methods and taxonomy. (See also Qualifications of Laboratory Personnel, Sec. II-1.)

2. Apparatus, Equipment, Supplies, Instruments

(1) Sampling devices

a) Rock-filled wire basket introduced substrate

Use: flowing wadeable, eroded, mineral-based bottom rivers and streams.

Description: cylindrical plastic coated or chrome wire, baskets with at least 1.5 cm spaces between wires, a hinged opening, and secure closure (Klemm, D.J. et al, 1990).

Substrate material: clean, washed, bank-run cobble, graded to uniform diameter range of 3.8 to 7.6 cm (1.5 to 3 inches) in size (#2 roofing stone).

Baskets must be filled to 7.25 +/- 0.5 kg (16 lbs +/-1 lb) of substrate material.

b) Rock-filled mesh bag introduced substrate

Use: small flowing streams, too shallow for rock baskets to be fully submerged.

Description: mesh bags of sufficient size to hold 7.25 +/- 0.5 kg of cobble substrate as described above, with at least 2.54 cm aperture mesh, and secure closures.

c) Closing introduced substrate cone

Use: deep, non-wadeable rivers having sufficient flow to have an eroded, mineral based bottom.

Description: cone shaped wire, or plastic coated wire basket filled with substrate material and closed by means of an inverted, weighted funnel (Courtemanch, 1984).

Substrate material: (see above Rock-filled wire basket substrate material).

(2) Sieves, sieve buckets, nets

Samples are concentrated on sieves having a mesh size between 500 - 600 microns (USA Standard Testing Sieve ASTM-E-11 Specification size No. 30 or No. 35).

(3) Optical equipment

a) Binocular microscope: Magnification range from 10x or less to 30x or greater.

b) Compound microscope: Magnification range from 10x to at least 400x; 100x with oil immersion lens is advisable.

3. Sampling Season, Sampler Exposure Period, Placement and Retrieval

(1) Sampling season

The standard sampling season upon which all macroinvertebrate classification criteria are based is the late summer, low flow period (July 1 to September 30). All baseline data for the biological classification program has been collected during this time period. This period often presents conditions of maximal stress to the biological community due to decreased dilution of pollutional material and increased stream water temperatures. Furthermore, because the composition of the benthic macroinvertebrate community changes with season, due to natural life history features, this period defines a standardized seasonal community.

As noted, the Department's linear discriminant models define biological classification criteria derived from a macroinvertebrate community defined by the specific sampling methods and index season under which they were collected. Samples collected at other times of year may yield valuable water quality related information, however classification attainment may not be assigned solely on the basis of results of the linear discriminant models for these non-standard samples.

(2) Exposure period

Standard methods require that substrate samplers be exposed in the water body for a period of 28 days +/- four days within the above-specified sampling season. However, extended exposure periods may be necessary to allow for adequate colonization in the case of assessments of low velocity or impounded habitats. If such conditions exist a 56 days +/- four days exposure period may be used.

(3) Sampler placement

Rock Baskets/Bags

The actual sampler location should be approached so as to avoid any disturbance in, or upstream of, the sampled site. Position baskets in locations of similar habitat characteristics. Orient baskets with the long axis parallel to stream flow. Provide for relocation of baskets by flagging trees in the vicinity and/or by drawing a diagram with appropriate landmarks indicated.

Cones

Cone samplers should be marked with individual marker buoys (milk jugs or other suitable float) leaving about 5 extra feet of line to allow for water level changes and to provide for easy retrieval. They should be placed on the substrate with a minimum of disturbance, in an apex-up position, and located in the approximate middle fifty percent of the channel. (Note however, care should be taken not to create an obstruction to boat traffic.) In areas subject to vandalism, or in rivers having extensive macrophyte beds, it may be necessary to attach the sampler lines to a common anchor and thence to one unobtrusive surface float. Retrieval funnels will not properly close when lines are fouled with drifting macrophytes.

(4) Sampler retrieval

Rock Baskets/ Bags

Baskets are approached from downstream. Excessive accumulations of macrophytes, algae or debris clinging to the outside of the basket should be carefully removed, taking care to avoid jarring the basket itself. An aquatic net or drift net (mesh size 500 - 600 microns) is positioned against the substrate immediately downstream of the basket which is then quickly lifted into the net. The contents of the basket and all net washings are emptied into a sieve bucket (500 - 600 microns); the basket wires are carefully cleaned first, then rocks are hand washed and inspected and returned to the basket. All sieve bucket contents are placed in sample jars. A small amount of stream water and 95% ethyl alcohol is added to yield an approximately 70% solution of alcohol. Especially dense samples should be re-preserved in the laboratory, with fresh 70% ethyl alcohol. Rock baskets should be thoroughly cleaned and allowed to desiccate prior to re-use.

Cones

Cone samplers should be retrieved with the boat anchored directly upstream of the samplers. Once the float is retrieved and removed, the line should be held as vertically as possible while the weighted funnel is released down the line to enclose the cone. Cone and funnel should be retrieved quickly and smoothly from the bottom, and released directly into a sieve bucket or tub. Field processing should then proceed as described above for rock baskets.

4. Site Selection Criteria

Classification criteria apply to a strictly defined sample of the benthic macroinvertebrate community. Habitat type from which the community is obtained is a significant determinant of the make-up of the target community. Benthic macroinvertebrate communities of flowing streams and rivers having a hard, eroded substrate comprise the majority of samples in the baseline data set. This habitat is characteristic of the majority of the river and stream waters of the State. Exceptions to these conditions may require special consideration and the exercise of professional judgment. (Note: See Section III-2. (3) "Classification attainment evaluation of waters subjected to flow regulation" page 13, for procedures relating to the assessment of regulated flow sites.) While it is useful to obtain both an upstream and downstream sample to evaluate the effect of a pollution source, classification attainment evaluation does not require data from a matched reference site in order to arrive at a determination of aquatic life class. Analytical methods for classification attainment evaluation are described in Section III.

(1) Site attributes

a) The area selected should be generally representative of the habitat of the stream reach as a whole;
b) Where there is alternating riffle/pool habitat, the riffle/run is the habitat of choice;
c) A location should be selected where there is a high degree of certainty that the rock basket samples will remain fully submerged even if the water level drops significantly.

(2) Precautions

a) Avoid atypical influences such as bridges, entering culverts, channelized areas such as road crossings, culverts, or obstructions to flow;
b) Avoid bank effects: samplers should be located in the middle 50% of the bank to bank width, or in an area with a flow regime typical of the overall character of the stream segment;
c) Avoid slackwater areas and eddies immediately upstream or downstream of large rocks or debris.

(3) Matching reference and effluent impacted sites

If possible both stream reaches should be viewed prior to selection of sampling sites. Efforts should be made to sample habitats which are comparable in the following characteristics:

a) Water velocity;
b) Substrate composition (i.e., size ranges and proportions of particles making up the substrate);
c) Canopy coverage;
d) Depth;
e) Other upstream influences except the pollution source in question (for example, use caution when one site is just below a lake outfall and the other is not).

(4) Factors to be considered in site selection below point sources

The area of initial dilution of an effluent should be determined by visual observation of the plume pattern; by observations of biotic effects attributable to the plume, if evident (periphyton growth, die-off patterns); and by transects of specific conductance measurements from the outfall, in a downstream direction. The site selected should be in an area where reasonable opportunity for mixing of the effluent has occurred. If a mixing zone has been defined in a license, sampling should occur immediately downstream of it. In cases where the effluent plume channels down one bank for great distances (>1 km), or where localized effluent impact is expected to be severe for a distance beyond the zone of initial dilution, it is advisable to have a sampling site upstream of the source, one or more in the plume, and at least two farther downstream. One downstream site should be located at the point of presumed bank to bank mixing and subsequent sites should be located to assess the extent of impact downstream.

5. Sample Size

The biological community is evaluated on the basis of benthic macroinvertebrates obtained from at least three samplers which yield an average of at least 50 organisms per sampler. Matched upstream and downstream sites must be sampled using identical methods and level of effort, preferably by the same personnel.

Subsampling may be performed on samples if the mean number of organisms in a sampler exceeds 500 and subsampling will yield at least 100 organisms per rock/cone sampler. All samplers in a site should be treated consistently. Subsampling methods are described in Section II-5. Note: Subsampling will

reduce sample richness by an indeterminate amount. This may affect the outcome of linear discriminant analysis. See Section III-2. (2).

6. Physical Habitat Evaluation

A field data sheet (Appendix A) is to be completed at the time of sampler placement. This form records site specific information concerning natural variables that may affect community structure. Items addressed include exact site location (latitude and longitude, narrative description of the mapped location and/or a topographic map with site indicated); substrate composition; canopy coverage; land use and terrain characteristics; water velocity, temperature, dates of exposure and investigator name. The form is to be completed by observation as well as instrument measurement of water velocity, specific conductance, dissolved oxygen, global positioning device, temperature, etc.

II LABORATORY METHODS

1. Qualifications of Laboratory Personnel

Sample processing and taxonomy in the laboratory must be performed or supervised by a professional freshwater macroinvertebrate taxonomist who is certified by the Society of Freshwater Science in the identification of eastern US taxa. Certification must include Genus level categories, such as Ephemeroptera, Plecoptera and Trichoptera (EPT), General Arthropods and Chironomidae taxa. Taxonomic data will not be accepted without verification that the supervising laboratory taxonomist has been certified in relevant categories.

2. Sample Preservation, Sorting

All sample material collected in the field, as described in Section I, is preserved in 70% ethyl alcohol. Samples are stored in airtight containers until sorted. Sorting of macroinvertebrates from detritus and debris should follow methods described in Appendix B. One out of every ten samples is evaluated by a biologist for sorting completeness.

After sorting, recommended storage for macroinvertebrates is in 70% ethyl alcohol with 5% glycerin, in vials sealed with tightly fitting rubber stoppers.

3. Sample Labeling

All samples are labeled in the field immediately upon collection. The label must include the following information:

Date of sample retrieval
Waterbody
Town or target discharge
Whether above or below the discharge (if applicable)
Replicate number

4. Sample Log Book

In the laboratory, the samples from each sampled site are to be assigned a sample log number, written on all items generated by the sample (e.g., sample vials, slides, records, count sheets, etc.). Log numbers are sequentially recorded in a master log book. The log book shall also contain site identification, date of placement and retrieval, investigator name, sampler type and any comments regarding sampler retrieval or data quality.

5. Subsampling

(1) Methods

If it is determined that a sample should be subsampled (see criteria in Section I-5 Sample Size) methods of Wrona et al, (1982) are followed. These are summarized below:

a) Fit a plastic or glass Imhoff-type settling cone with an aquarium air stone sealed in the bottom and connected to a compressed air supply.

b) Place the sorted macroinvertebrate sample in the cone and fill the apparatus with water to a total volume of one liter.

c) Agitate gently for 2 to 5 minutes with the air stone.

d) Remove 25% of the sample in 5 aliquots with a wide-mouth 50 ml dipper and combine into one sample vial. The dipper should be submerged and withdrawn over a five second interval.

e) Ascertain whether or not the required 100 organisms have been obtained in the subsample.

f) Indicate clearly on the sample label and on the data sheet the fraction of the sample that the subsample represents.

(2) Precautions

a) Especially large or dense organisms such as crayfish, molluscs or caddisflies with stone cases, which do not suspend randomly in the sample, should not be included in the subsample. They should be counted separately.

b) When removing aliquots, the subsampler should be careful to avoid biased capture of organisms in the cone. Avoid watching the cone as the dipper is withdrawn.

This method has been tested by the Department and has been found to randomly distribute the sample. The five separate counts conform to a Poisson series and thus can be combined into one sample (Elliott, 1979).

(3) Chironomidae subsampling

A subsampling plan for Chironomidae shall be approved by the Department. A Department recommended subsampling plan follows the following criteria:

a) For samples having less than 100 midges, all midges will be identified to genus/species level.

b) For samples having 100 to 199 midges, a subsample of one half (0.5) will be removed by randomly selecting the specimens to be identified and identified to genus/species level. Remaining unsampled midges will be examined for unusual or rare specimens, which will be removed and identified to genus/species level separate from the subsample of the sample.

c) For samples having 200 to 499 midges, a subsample of one quarter (0.25) will be removed by randomly selecting the specimens to be identified and identified to genus/species level. Remaining unsampled midges will be examined for unusual or rare specimens, which will be removed and identified to genus/species level separate from the subsample of the sample.

d) For samples having 500 or more midges, midges will be grouped by genus for those for which it is possible to confidently identify them to genus level without mounting. For remaining midges not grouped by genus, a subsample of 100 specimens will be randomly selected and identified to genus/species level. Remaining unsampled midges will be examined for unusual or rare specimens, which will be removed and identified to genus/species level separate from the subsample of the sample.

e) Reporting of the subsample of the sample will be as follows. Numbers reported on the Excel spreadsheet will be converted to reflect the sample total. Any round-off errors between the subsample total and the sample total will be equalized by adding or deducting the difference from the most numerous taxon. If unusual or rare specimens are removed from the sample following the subsample removal, the conversion of the subsample total to a "partial" sample total will be based on the sample total minus the number of unusual or rare specimens. Following this procedure, the number of unusual or rare specimens will be added to the "partial" sample total to bring it back to the sample total.

6. Sample Taxonomy

All taxonomic data submitted to the Department must be accompanied by the name(s) of the individual(s) actually performing the identifications. A list of taxonomic references used, and a reference collection of organisms must also be submitted (see below).

(1) Taxonomic resolution

Macroinvertebrate organisms are identified to genus in all cases where possible. If generic keys are not available or taxonomic expertise is lacking for a taxon it should be identified to the lowest level possible. Identification of organisms to species is highly recommended whenever possible. Although quantitative analysis of benthic macroinvertebrate samples by the Department is based on counts adjusted to the generic level of resolution, species designations are recorded in the Department database and can contribute to the final stage of data analysis, Professional Judgment Evaluation of the model outcome. This is especially important for Class Insecta. Taxonomists submitting data for use by the Department must use current taxonomic references.

(2) Identification of Chironomidae

Specimens of chironomid midges are identified from slide mounts of the cleared head capsule and body parts. Euparol or Berlese mounting medium is recommended for preparation of slides. CMCP-9 is recommended for the preparation of permanent slide mounts of reference material, for voucher specimens or for permanent collections. These slides should be prepared under a fume hood. Instructions for preparation and slide mounting may be found in Wiederholm, (1983). In samples in which a given taxon is represented by a large number of individuals, the identification to genus may be made from slide mounts of a sufficient proportion of the individuals to give a high degree of certainty that they are all the same (10-50% depending on

the distinctiveness of the taxon visible under binocular microscope). A subsampling plan for Chironomidae is described in Section II-5. Each permanent slide mount is to be fully labeled or coded in a manner which positively associates the slide with the sample from which it originated.

(3) Quality control

All organisms and records from any sampling event intended to serve regulatory purposes must be preserved for a period of at least ten years. In the course of identifying taxa collected as part of the Department's biological monitoring program, or in other collection activities, a special reference collection of separate taxa is established. This collection allows subsequent identifications of the same taxon to be confirmed and thus serves to standardize taxonomy for the program.

Each contracted taxonomist, working for the Department or working for anyone submitting data to the Department, will be required to submit a reference collection of taxa identified, as well as a list of the taxonomic references used in the identifications. Organism identifications will be checked against the Department's collection by a Department taxonomist.

III ANALYTICAL METHODS

In general, it is the responsibility of the Department, or its agents, to conduct sampling for the purpose of making decisions on the attainment of water quality classification. Under certain conditions, sampling may be required of applicants for waste discharge licenses, or applicants requiring Section 401 Water Quality Certification. Sampling may be performed by corporations, businesses, organizations or individuals who can demonstrate their qualifications and ability to carry out the Department's sampling and analytical protocol, described in this manual. Such monitoring will be conducted according to a quality assurance plan provided to the Department and approved by the Commissioner.

Classification attainment evaluation is established in DEP regulation Chapter 579: Classification Attainment Evaluation Using Biological Criteria for Rivers and Streams. Davies et al, 1995 details the conceptual and technical basis for the State’s application of linear discriminant analysis to assess attainment of aquatic life standards. A synopsis of Chapter 579 follows in this section.

1. Minimum Provisions

Properly collected and analyzed samples that fail to achieve the following criteria are unsuitable for further analysis through the numeric criteria statistical models:

- Total Mean Abundance must be at least *50* individuals (average per basket/bag/cone);
- Generic Richness for three replicate basket/bag/cone samplers must be at least *15*.

Samples not attaining these criteria shall be evaluated by Professional Judgment. A determination will be made whether the affected community requires re-sampling or whether the community demonstrates non-attainment of minimum provisions of the aquatic life standards.

2. Aquatic Life Statistical Decision Models

The four statistical decision models consist of linear discriminant functions developed to use quantitative ecological attributes of the macroinvertebrate community (Appendix C-1) to determine the strength of the association of a test community to any of the water quality classes (Appendix D). The coefficients or weights are calculated using a linear optimization algorithm to minimize the distance, in multivariate space, between sites within a class, and to maximize the distance between sites between classes.

(1) Linear discriminant models

The discriminant function has the form:

$$Z = C + W_1X_1 + W_2X_2 + ...W_nX_n$$

Where: Z = discriminant score
C = constant
W_i = the coefficients or weights
X_i = the predictor variable values

Association values are computed, using variable values from a test sample, for each classification using one four-way model and three two-way models. The four-way model uses nine variables pertinent to the evaluation of all classes and provides four initial probabilities that a given site attains one of three classes (A, B, or C), or is in non-attainment (NA) of the minimum criteria for any class. These probabilities have a possible range from 0.0 to 1.0, and are used, after transformation, as variables in each of the three subsequent final decision models. The final decision models (the three, two-way models)

are designed to distinguish between a given class and any higher classes as one group and any lower classes as the other group (i.e., Classes A+B+C vs. NA; Classes A+B vs. Class C+NA; Class A vs. Classes B+C+NA). The equations for the final decision models use the predictor variables relevant to the class being tested (Appendix E). The process of determining attainment class using association values is outlined in Appendix F.

(2) Application of professional judgment

Where there is documented evidence of conditions which 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 DEP regulation Chapter 579: Classification Attainment Evaluation Using Biological Criteria for Rivers and Streams. 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.

Professional Judgment may be utilized when conditions are found that are atypical to the derivation of the linear discriminant model. Factors that may allow adjustments to the model outcome include but are not limited to:

a) Habitat factors
- Lake outlets
- Impounded waters
- Substrate characteristics
- Tidal waters

b) Sampling factors
- Disturbed samples
- Unusual taxa assemblages
- Human error in sampling

c) Analytical factors
- Subsample vs. whole sample analysis
- Human error in processing

(3) Classification attainment evaluation of waters subjected to flow regulation

The Maine State Legislature, in 38 MRSA Article 4-A Sec. 464 (9)-(10), *The Water Classification Program*, acknowledges that changes to aquatic life and habitat occur as the result of the impoundment of riverine waters and has modified the standards of waters so affected. The habitat and aquatic life criteria of riverine impounded waters of Class A, Class B or Class C are

deemed to be met if the impoundment attains the standards of Class C (e.g., maintenance of structure and function of the resident biological community). Impoundments managed as Great Ponds must also attain Class C aquatic life standards. If the actual water quality attains any more stringent characteristic or criterion than the Class C standards dictate, then the waterbody must be managed so as to protect those higher characteristics. Class C standards also apply to the *downstream* waters below certain specified riverine impoundments on the Kennebec River and the Saco River (Wyman Dam, Moosehead East Outlet Dam, West Buxton Dam and Skelton Dam) that are classified as A or B. All other waters subjected to flow regulation are managed according to standards of the water quality classification assigned by the Legislature.

(4) Adjustments of a decision

It is the responsibility of the Department to decide if adjustments of a decision should occur. The following adjustments may be made to correct for these conditions:

a) Resample
The Department may require that additional monitoring of the test community be done before a determination of class attainment can be made, based on documented evidence of specific sampling factors that may have influenced the results.

b) Raise the finding
   i. The Department may raise the classification attainment outcome predicted by the model from non-attainment of any class to indeterminate or to attainment of Class C, based on documented evidence of specific conditions, as defined above.

   ii. The Department may raise the classification attainment outcome predicted by the model from attainment in one class to attainment in the next higher class, based on documented evidence of specific conditions, as defined above.

c) Lower the finding
The Department may decide to lower the classification attainment finding, on the basis of documented, substantive evidence that the narrative aquatic life criteria for the assigned class are not met.

d) Determination of non-attainment: minimum provisions not met
Samples having any of the ecological attributes not attaining the minimum provisions, and where there is no evidence of conditions which could result in uncharacteristic findings, as defined above, must be determined to be in non-attainment of the minimum provisions of the aquatic life criteria for any class.

e) Determination of attainment: minimum provisions 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.

(5) Sampling procedures do not conform

For classification attainment evaluation of test communities that do not conform to criteria provided in Section I General Methods, or Section III-1, Minimum Provisions, of this manual, and are therefore not suitable to be run through the linear discriminant models, 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 shall be approved by the department.

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. Soft-bottomed substrates shall, whenever ecologically appropriate and practical, be sampled by core or dredge of known dimension or volume.

ii. The preferred method for sampling hard-bottomed substrates shall be the rock basket/cone/bag as described in Section I-2.

iii. Other methods may be used where ecologically appropriate and practical.

c) 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 38 MRSA Section 465 and found in Appendix D. The decision is based on established principles of water pollution biology and must be fully documented.

d) 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.

e) 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.

Appendix A

Maine DEP Biological Monitoring Unit
Stream Macroinvertebrate Field Data Sheet

Log Number _____ Directions_____ Type of Sample_____
Station Number_____ _____ Date Deployed_____
Waterbody_____ _____ Number Deployed_____
River Basin_____ Lat-Long Coordinates (WGS84, meters) Date Retrieved_____
Municipality_____ Latitude_____ Number Retrieved_____
Stream Order_____ Longitude_____ Agency/Collector(s)_____

1. Land Use (500 m radius upstream)
- ☐ Urban
- ☐ Cultivated
- ☐ Pasture
- ☐ Upland hardwood
- ☐ Upland conifer
- ☐ Swamp hardwood
- ☐ Swamp conifer
- ☐ Marsh

2. Terrain (500 m radius upstream)
- ☐ Flat
- ☐ Rolling
- ☐ Hilly
- ☐ Mountains

3. Canopy Cover (upstream view)
- ☐ Dense (75-100% shaded)
- ☐ Partly open (25-75% shaded)
- ☐ Open (0-25% shaded)

(% daily direct sun) _____

4. Physical Characteristics of Bottom (estimate % of each component over 12 m stretch of site; total = 100%)

[ ] Bedrock [ ] Rubble (3" – 10") [ ] Sand (<1/8") [ ] Boulders (<10") [ ] Gravel (1/8" – 3") [ ] Silt-clay-muck [ ] Detritus

5. Habitat Characteristics (immediate area)

Time _____AM PM	Time _____AM PM
Width (m) _____	Width (m) _____
Depth (cm) _____	Depth (cm) _____
Flow (cm/s) _____	Flow (cm/s) _____
Diss. O_2 (ppm)_____	Diss. O_2 (ppm)_____
Temp (°C) _____	Temp (°C) _____
pH _____	pH _____
SPC (µS/cm) _____	SPC (µS/cm) _____
TDS (ppm) _____	TDS (ppm) _____

Temperature Probe # _____

☐ deployed ☐ retrieved

6. Observations (describe)
Fish_____
Algae_____
Macrophytes_____
Habitat quality_____
Dams/impoundments_____
Discharges_____
Nonpoint stressors _____

7. Water Samples
- ☐ Standard
- ☐ Metals
- ☐ Pesticides

Lab Number

8. Photographs

9. Landmarks of Sampler Placement (illustrate or describe landmarks to be used for relocation)

Appendix B

Instructions for Macroinvertebrate Sorters

1. Pick the sample **in small portions** (1-2 TBS of material) at a time.

2. Pick all organisms you can see. If in doubt it's usually best to include it.

3. Some types of samples can be easily floated by adding a saturated solution of Epsom salt or sugar to the water. Maintain the saturated solution for the lab by adding enough salt or sugar to water to maintain a thick layer of crystals on the bottom of the storage jar. Use the supernatant solution for picking. Large numbers of organisms can be removed with a sieve spoon from the water surface. After the floaters have been removed, proceed to pick the rest of the sample as usual. A significant portion of the sample will not float and must be picked out with forceps.

4. The sample can be considered done when a careful 45 second search, after swirling the sample, yields no further organisms.

5. The samples are picked in water but should not remain unpreserved for more than 8 hours. Be certain that the final sample vial is preserved with 70% alcohol and 5% glycerin solution when done.

6. Return the detrital material to the original sample jar and preserve with 70% alcohol.

7. Write on the sample jar label "Picked X1 (your initials)".

8. Include in the vial of organisms a slip of index card label in hard pencil (No. 2) including **all information appearing on the original jar label**:

   Log Number — River
   Date - month/day/year — Location (Town or industry name)
   whether above or below
   Basket or Cone number
   Vial number if more than 1 vial is needed per basket

   ex. Log 621 Sandy R. 9/5/97
   Below Farmington (disturbed)
   Basket 2 vial #1 of 2

9. Complete all samples from one log number before beginning a new log number.

10. Keep a record of samples picked including log number

    Basket number — Time spent per basket
    Your name — Date

Appendix C-1

Methods for the Calculation of Indices and Measures of Community Structure Used in the Linear Discriminant Models

Variable Number

1 **Total Mean Abundance**

Count all individuals in all replicate samples from one site and divide by the number of replicates to yield mean number of individuals per sample.

2 **Generic Richness**

Count the number of different genera found in all replicates from one site.

Counting rules for Generic Richness:

a) All population counts at the species level will be aggregated to the generic level.

b) A family level identification which includes no more than one taxon identified to the generic level is counted as a separate taxon in generic richness counts.

c) A family level identification with more than one taxon identified to generic level is not counted towards generic richness. Counts are to be divided proportionately among the genera that are present.

d) Higher level taxonomic identifications (Phylum, Class, Order) are not counted toward generic richness unless they are the only representative.

e) Pupae are ignored in all calculations.

3 **Plecoptera Mean Abundance**

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**

Count all individuals from the order Ephemeroptera in all replicate samplers from one site and divide by the number of replicates to yield mean number of Ephemeropteran individuals per sampler.

5 **Shannon-Wiener Generic Diversity (Shannon and Weaver, 1963)**

After adjusting all counts to genus following counting rules in Variable 2:

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

where: $\bar{d}$ = Shannon-Wiener 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^{th} taxon

6 **Hilsenhoff Biotic Index (Hilsenhoff, 1987)**

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

where: HBI = Hilsenhoff Biotic Index
n_i = number of individuals in the i^{th} taxon
a_i = tolerance value assigned to that taxon
N = total number of individuals in sample with tolerance values.

7 **Relative Chironomidae Abundance**

Calculate the mean number of individuals of the family Chironomidae, following counting rules in Variable 4, and divide by total mean abundance (Variable 1).

8 **Relative Diptera Richness**

Count the number of different genera from the Order Diptera, following counting rules in Variable 2, and divide by generic richness (Variable 2).

9 ***Hydropsyche* Mean Abundance**

Count all individuals from the genus *Hydropsyche* in all replicate samplers from one 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**

Sum of probabilities for Classes A, B, and C from First Stage Model.

11 ***Cheumatopsyche* Mean Abundance**

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 Generic Richness (Variable 19) divided 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**

Calculate the mean number of individuals from the Order Oligochaeta, following counting rules in Variable 4, and divide by total mean abundance (Variable 1).

14 **Probability (A + B) from First Stage Model**

Sum of probabilities for Classes A and B from First Stage Model.

15 **Perlidae Mean Abundance (Family Functional Group)**

Count all individuals from the family Perlidae (Appendix C-3) 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 (Family Functional Group)**

Count all individuals from the subfamily Tanypodinae (Appendix C-3) 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 (Family Functional Group)**

Count all individuals from the tribe Chironomini (Appendix C-3) 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**

Variable 4 divided by Variable 1.

19 **EPT Generic Richness**

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 Abundances of**: ***Dicrotendipes, Micropsectra, Parachironomus* and *Helobdella***

Sum the abundance of the 4 genera and divide by the number of replicates (as performed in Variable 4).

22 **Probability of Class A from First Stage Model**

Probability of Class A from First Stage Model.

23 **Relative Plecoptera Richness**

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 Abundances of *Cheumatopsyche, Cricotopus, Tanytarsus* and *Ablabesmyia***

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 Abundances of *Acroneuria* and *Stenonema***

Sum the number of individuals in each genus in all replicate samplers and divide by the number of replicates (as performed in Variable 4).

27 **Variable Reserved**

28 **Ratio of EP Generic Richness**

Count the number of different genera from the order 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**

Count the number of Class A indicator taxa as listed in Appendix C-2 that are present in the community and divide by 7 (total possible number).

Appendix C-2

Indicator Taxa: Class A

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

Appendix C-3

Family Functional Groups

PLECOPTERA

Perlidae
Acroneuria
Attaneuria
Beloneuria
Eccoptura
Perlesta
Perlinella
Neoperla
Paragnetina
Agnetina

CHIRONOMIDAE

Tanypodinae
Ablabesmyia
Clinotanypus
Coelotanypus
Conchapelopia
Djalmabatista
Guttipelopia
Hudsonimyia
Labrundinia
Larsia
Meropelopia
Natarsia
Nilotanypus
Paramerina
Pentaneura
Procladius
Psectrotanypus
Rheopelopia
Tanypus
Telopelopia
Thienemannimyia
Trissopelopia
Zavrelimyia

Appendix C-3

Family Functional Group (continued)

Chironomini
Pseudochironomus
Axarus
Chironomus
Cladopelma
Cryptochironomus
Cryptotendipes
Demicryptochironomus
Dicrotendipes
Einfeldia
Endochironomus
Glyptotendipes
Goeldichironomus
Harnischia
Kiefferulus
Lauterborniella
Microchironomus
Microtendipes
Nilothauma
Pagastiella
Parachironomus
Paracladopelma
Paralauterborniella
Paratendipes
Phaenopsectra
Polypedilum
Robackia
Stelechomyia
Stenochironomus
Stictochironomus
Tribelos
Xenochironomus

Appendix D

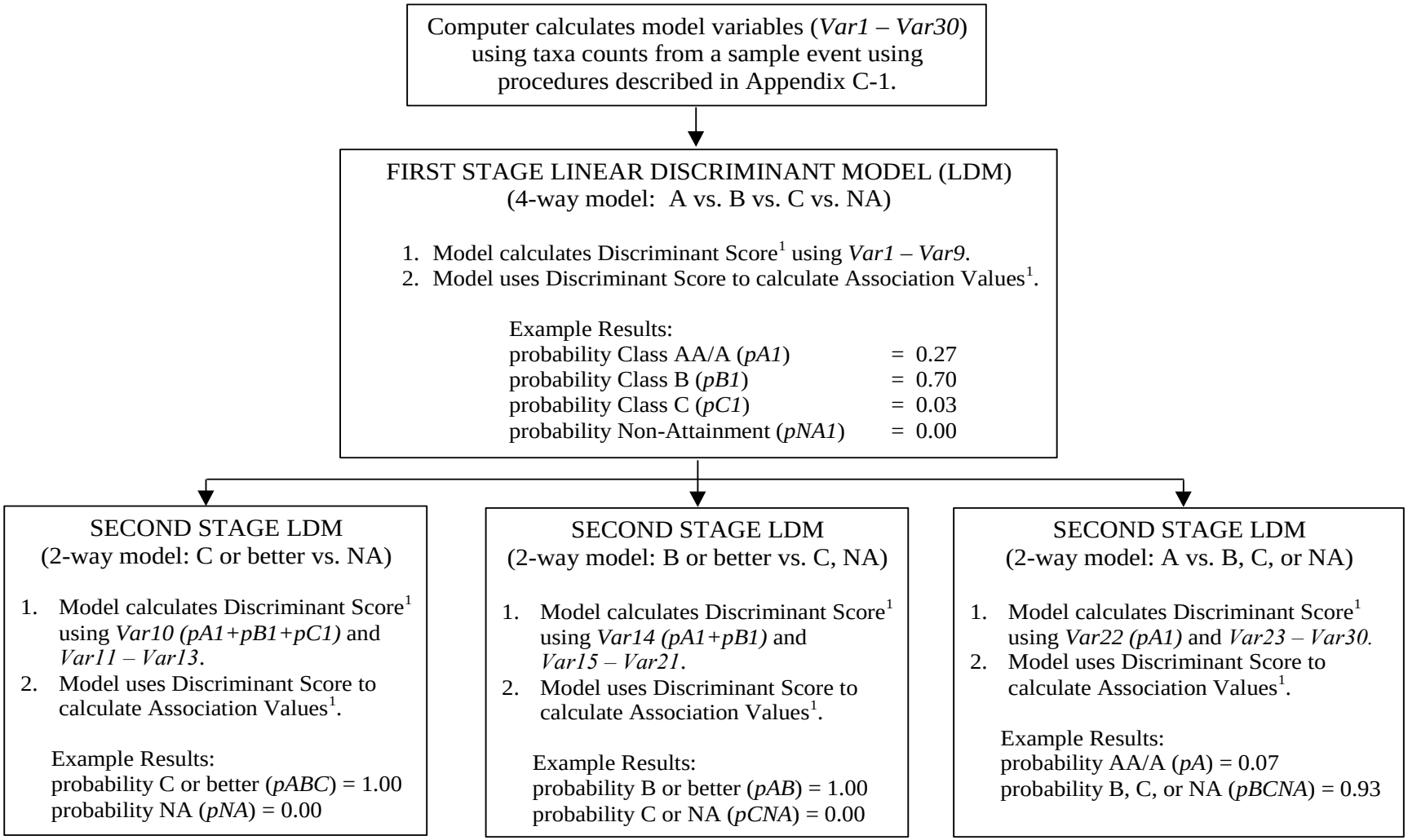
MRSA 38, 4-A Sec 464-465

Aquatic Life Standards for the State of Maine

Classification	Biological Standards
AA	No direct discharge of pollutants; aquatic life shall be as naturally occurs.
A	Natural habitat for aquatic life; aquatic life shall be as naturally occurs.
B	Unimpaired habitat for aquatic life; discharges shall not cause adverse impact to aquatic life in that the receiving waters shall be of sufficient quality to support all aquatic species indigenous to the receiving water without detrimental changes in the resident biological community.
C	Habitat for 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 waters and maintain the structure and function of the resident biological community.

Appendix E

Process of Calculating Model Variables and Association Values Using Linear Discriminant Models

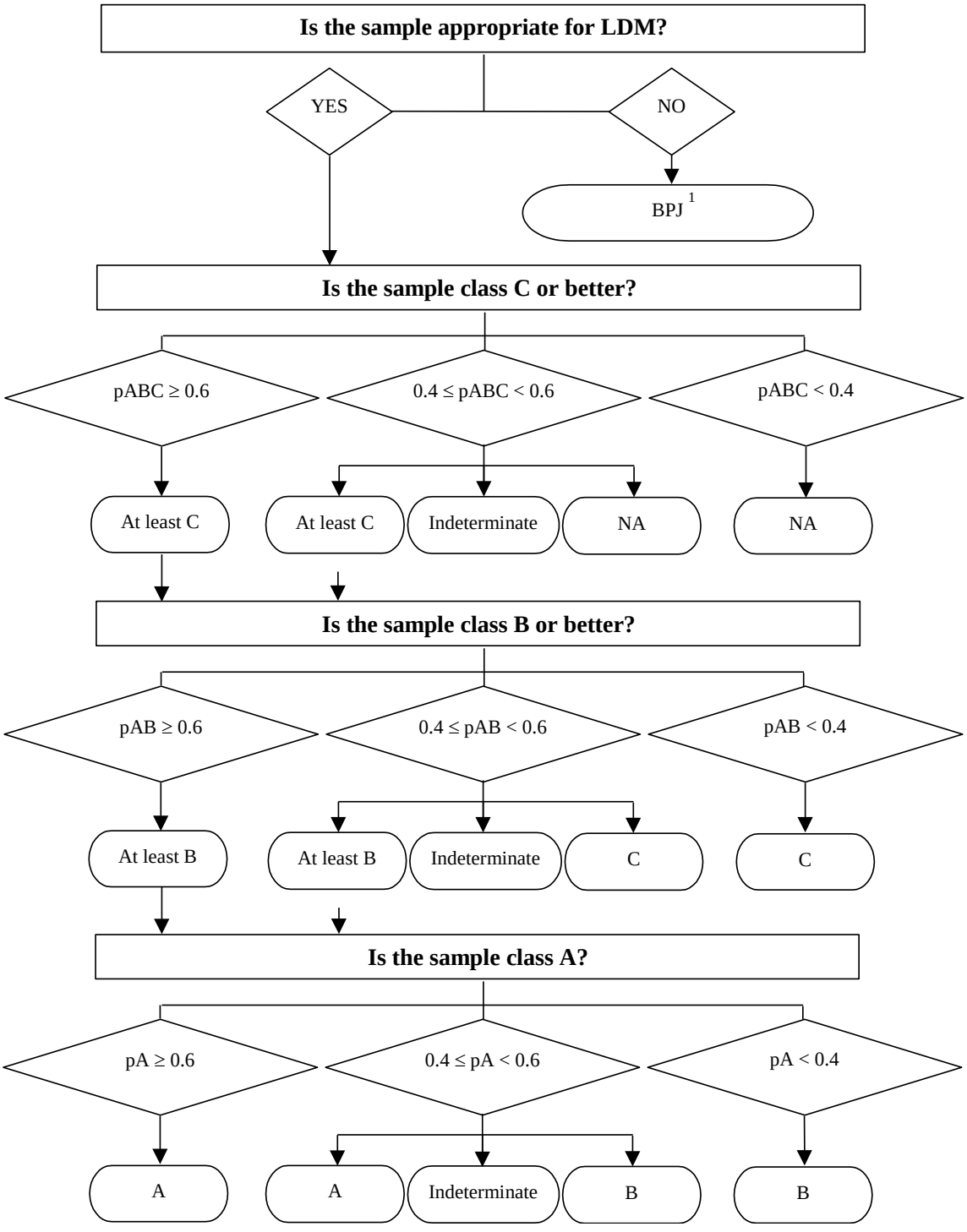


[1] Discriminant Score and Association Values are defined in Section III-2.(1).

Chart by Thomas J. Danielson

Appendix F

Process for Determining Attainment Class Using Association Values



[1] Best Professional Judgment (BPJ) is defined in Section III-2. (2), (4), and (5)

Chart by Thomas J. Danielson

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Maine DEP Biological Monitoring Unit
Stream Macroinvertebrate Field Data Sheet

Location: _____

_____

Potential Stressor: _____

_____

Log Number _____
Station Number_____
Waterbody_____
River Basin_____
Town_____
Stream Order_____

Directions_____
_____
_____
Lat-Long Coordinates (WGS84, meters)
Latitude_____
Longitude_____

Type of Sampler_____
Date Deployed_____
Number Deployed_____
Date Retrieved_____
Number Retrieved_____
Agency/Collector(s) Put-In:
Take-Out:

1. Land Use (surrounding watershed)
- ☐ Urban
- ☐ Cultivated
- ☐ Pasture
- ☐ Upland hardwood
- ☐ Upland conifer
- ☐ Swamp hardwood
- ☐ Swamp conifer
- ☐ Marsh

2. Terrain (surrounding watershed)
- ☐ Flat
- ☐ Rolling
- ☐ Hilly
- ☐ Mountains

3. Canopy Cover (surrounding view)
- ☐ Dense (75-100% shaded)
- ☐ Partly open (25-75% shaded)
- ☐ Open (0-25% shaded)

(% daily direct sun) _____

4. Physical Characteristics of Bottom (estimate % of each component over 12 m stretch of site; total = 100%)

[ ] Bedrock [ ] Cobble (2.5" – 10") [ ] Sand (<1/8") [ ] Clay
[ ] Boulders (>10") [ ] Gravel (1/8" – 2.5") [ ] Silt [ ] Muck [ ] Detritus

5. Habitat Characteristics (immediate area)

Flag location where measured

Time _____ AM PM	Time _____ AM PM
Wetted Width (m)_____	Wetted Width (m) _____
Bank Full Width (m) _____	Bank Full Width (m) _____
Depth (cm) _____	Depth (cm) _____
Velocity (cm/s) _____	Velocity (cm/s) _____
Diss. O_2 ___ (ppm) ___ (%)	Diss. O_2 ___ (ppm) ___ (%)
Temp (°C) _____	Temp (°C) _____
SPC (µS/cm) _____	SPC (µS/cm) _____
pH _____	pH _____
DO Meter #_____ Cal? Y / N	DO Meter #_____ Cal? Y / N
SPC Meter # _____ Cal? Y / N	SPC Meter # _____ Cal? Y / N

Temperature Probe # _____
☐ deployed ☐ retrieved

6. Observations (describe, note date)

7. Water Samples
- ☐ Standard
- ☐ Other

Lab Number:

8. Photograph #

Put-In
- Up
- Down

Take-Out
- Up
- Down

9. Landmarks of Sampler Placement (illustrate or describe landmarks to be used for relocation)

Options for Potential Stressor:

Agricultural Runoff
Altered Habitat
Altered Hydrology
BOD (Low DO)
Bog Headwaters
Chlorine
Gravel Pit
Impounded
Inorganic Solids
Lake Outlet
Logging
Low Gradient
Low pH
Metals
NPS Pollution
Nutrients
Organic Solids
Pesticides
Regulated Flows
Sedimentation
Superfund Site
Thermal
Tidal/Estuary
Toxic Organics
Urban Runoff

Options for 6. Observations:

Fish
Algae
Macrophytes
Habitat quality
Dams/impoundments
Discharges
Nonpoint stressors

Options for Location:

Above Road Crossing
Below Road Crossing
Above Town
Below Town
Above Fish Hatchery
Below Fish Hatchery
Above POTW
Below POTW
Above Landfill
Below Landfill
Below Airport
Below In-Place Contamination
Above In-Place Contamination
Above Point Source
Below Point Source
Above Urban NPS
Below Urban NPS
Above Agriculture NPS
Below Agriculture NPS
Above Forestry NPS
Below Forestry NPS
Above Dam
Below Dam
Impoundment
Lake Outlet
Main Stem (only for larger systems)
Above Confluence
Below Confluence
Below Falls
Pristine Landscape
Designated Ecoreserve
Minimally Disturbed