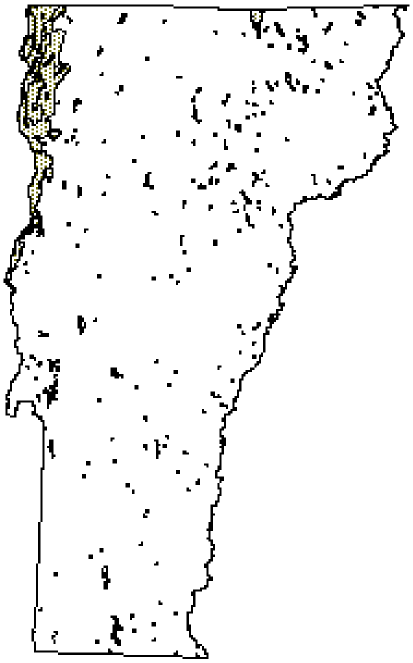
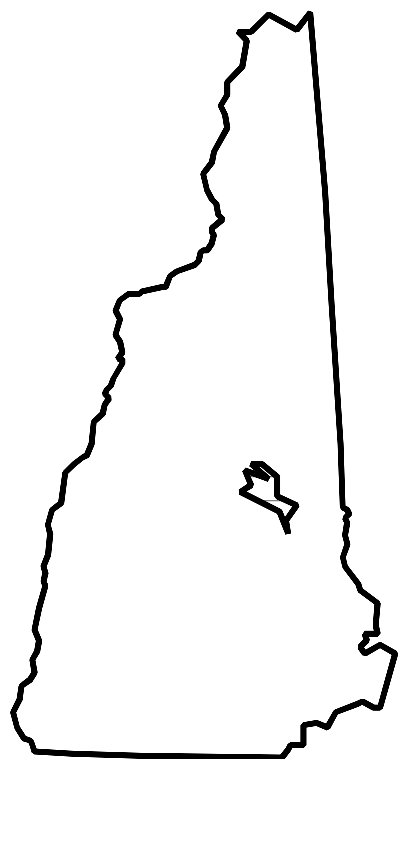


Paleolimnology and Bioassessment of Vermont and New Hampshire Lakes

Quarterly Report, 07/01/99-10/30/99

Bioassessment Project Component



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Project: Bioassessment of Vermont and New Hampshire Lakes Project and Development of Biological Criteria for Vermont Lakes Project

Funding Source: § 104(b)(3) of the Clean Water Act and EPA-Office of Water, Biocriteria Program

Reporting Period: 07/01/99-10/30/99

Criteria Development - Phytoplankton:
In preparation for a comprehensive project reporting, all available phytoplankton data and previous analyses were re-visited. The discriminant function analysis reported in the 04/01/99 quarterly report suggested that significant changes to the previously developed trial criteria were warranted given the additional 1998 data. Thus, a series of statistical analyses were performed to reclassify the lakes, verify the reclassification, and optimize the ability to discriminate conditions which depart from reference.

To reclassify the lakes, a canonical correspondence analysis (CCA) was first performed on all lakes using percent composition metrics. CCA is a useful tool which permits the visualization of phytoplankton-driven lake groups in multidimensional ordination space (Figure 1). This analysis, while explaining only 25 percent of the variance in the phytoplankton data, nonetheless suggested the existence of three phytoplankton-derived lake classes; low alkalinity lakes, well buffered lakes, and large lakes.

These three CCA-inferred classes were then used as a-priori groups within a discriminant classification analysis, which is an extension of multivariate ANOVA. Discriminant classification analysis is similar to multiple regression in that it seeks to generate linear equations using the original data which capture the maximum amount of variance in the dataset. Unlike multiple regression, which is used to predict a specific response variable, discriminant classification analysis identifies every lake (or other sampling unit) in the analysis as belonging to one of a given number of pre-established classes. In the process, all of the variance in the dataset is accounted for. The discriminant classification procedure will generate one mathematical equation for each pre-established class. In this case, there are three classes: 'low-alkalinity lakes;' 'well-buffered lakes;' and 'large lakes.' For each lake, the result of each of the three equations is calculated, and the equation producing the largest value will identify the class to which the lake should belong. For the equations (discriminant functions) to be useful, the mean class values for the classification variables must differ significantly as determined by multivariate ANOVA. As with most parametric statistical methods,

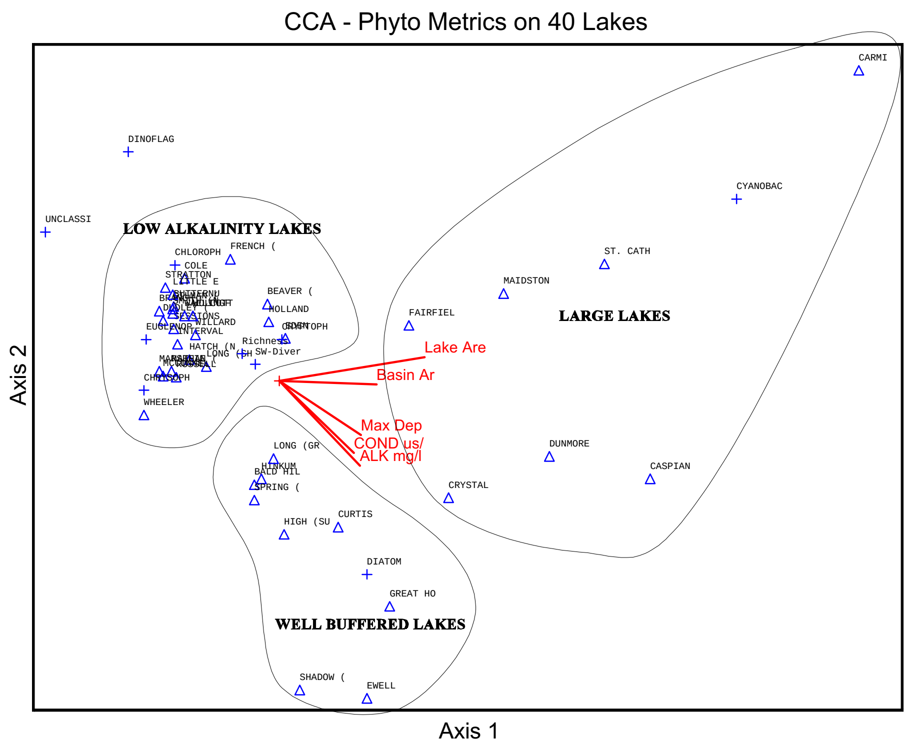

Figure 1. Canonical correspondence analysis triplot of 40 lakes and 8 compositional measurements of the lakes' phytoplankton community, in relation to physico-chemical attributes. 25% of the total phytoplankton variance is explained in this ordination.

discriminant classification analysis requires multivariate normality and equality of covariance matrices.

In the present case, the 40 lakes were allocated to the three classes discussed above, and the following log-transformed physico-chemical variables were used as classification variables: lake area; basin area; basin area/lake area; maximum depth; alkalinity; and conductivity. The overall analysis yielded significant class separation based on mulitvariate ANOVA(Wilkes' Λ = 0.09, F= 12.21, p=0.001). Accounting for prior class size, the classification error rate was determined by crossvalidation (jack knifing) to be 85 % correct allocation to a class. The equations generated by the analysis are presented in Table 1.

Table 1. Classification coefficients and constants which constitute the classification functions used to allocate lakes into one of three classes.

Lakes are classified to the largest solution of each linear function.			
Lake Class ➜	Well Buffered	Large	Low Alkalinity
Coefficient ⇓			
CONSTANT	-214.12	-240.40	-217.32
Log-Lake Area (ac)	889.09	879.35	884.97
Log-Basin area (ac)	-871.96	-856.23	-866.60
Log-Basin/Lake Area Ratio	938.44	922.31	935.72
Log-Maximum depth (m)	-0.01	2.08	0.18
Log-Alkalinity (mg/l)	-22.51	-23.38	-27.55
Log-Conductivity (us/cm^3)	36.03	34.82	37.22

Due to the low percent variance explained in the CCA, a multivariate ANOVA procedure was then used to verify that the phytoplankton metrics themselves vary between classes for all sampled lakes. Based on available phytoplankton metrics (appropriately transformed to normality), the three lake classes were found to differ significantly from each other (Wilks' Λ=0.168, F=2.26, p=0.0076). Percent diatom by density (p=0.05), % chrysophyte by density (p=0.0035), and % cyanobacteria by density (p=0.023) varied significantly between one or more classes. Percent cryptophytes by volume also varied across the classes, but at a reduced significance level (p=0.08).

At this point, all available phytoplankton metrics were plotted for reference and test lakes, for all three identified classes (Figure 2), and these distributions examined. A correlation matrix of the metrics was also generated to determine statistical independence of the metrics. The goal of this evaluation was to select metrics which simultaneously vary between classes, appear to discriminate reference from test conditions, and are statistically independent. Five metrics were found to satisfy these criteria. For metrics which 'appear' to discriminate reference from test conditions, the interquartile coefficient was calculated. The interquartile coefficient is defined as the interquartile range of the candidate reference metric, divided by the scope for detection of the same metric.

It is expressed as:

$$IC = IQ/ScD$$

where: IC= Interquartile coefficient
IQ= 75th percentile - 25th percentile of the metrics distribution within reference lakes;
ScD= The absolute value of the difference between the upper (or lower) quartile of the reference range, and the maximum (or minimum) value for the test distribution.

The Lake and Reservoir Bioassessment and Biocriteria Technical Guidance Document indicates that metrics with interquartile coefficients of > 1 are typically considered overly variable to detect deviation from reference conditions. The selected phytoplankton metrics, along with their interquartile coefficients, are presented in Table 2 and are highlighted in Figure 2.

Table 2. Interquartile coefficients for five phytoplankton metrics which discriminate between reference and test lakes, across three VT and NH lake classes.

Lake Class	Total Density cells/ml	% Cryptophytes by Volume	% Chrysophytes by Density	% Diatoms by Density	Percent of Aphanizomenon spp., Anabaena spp., Microcystis spp., by Volume
Clear Lakes	0.42	>1	0.74	>1	0.05
Tannic Lakes	0.74	0.42	>1	>1	0.56
Large Lakes	0.05	>1	>1	0.47	0.04

To develop a phytoplankton index, the 'bisection' scoring algorithm outlined in the Lake and Reservoir Bioassessment and Biocriteria Technical Guidance Document was used to identify numeric scores corresponding to deviation from the reference range for each metric. This algorithm allocates a score of five for all metric values which falls within the best 75 percent of the reference range for that metric. The range of metric values from the lower quartile of the reference range to the 'worst-case' test lake value is then bisected, with a score of three and one being allocated to the ranges of values corresponding to 'better' and 'worse' values, respectively. Table 3 identifies the scoring ranges for the five selected metrics.

To calculate a given lakes' overall phytoplankton index score, the scores corresponding to the actual metric value measured for the test lake are summed. For this index, actual scores for the 40 lakes ranged from a maximum of 15 for lakes fully meeting reference conditions for all three measures, to three for lakes not meeting reference expectations for any metric. Only one lake, the 303(d) listed Lake Carmi, has received a score of three.

The Lake and Reservoir Bioassessment and Biocriteria Technical Guidance Document suggests that a designation of 'deviating from reference conditions', should be made for lakes whose overall index score is below the 75th percentile of the reference range of the composite index. This a-priori assumes that 25 percent of the reference lakes measured in the development of the index are actually not reference. Vermont is at present uncomfortable with such a designation, although such a method does indeed account for mis-designation of reference lakes (see discussion below). The degree of departure, and thus potential impairment, should be contingent on an evaluation of all individual metrics. Figure 3 identifies the actual scores and Tukey box-plot ranges of reference and test lakes for the overall phytoplankton index..

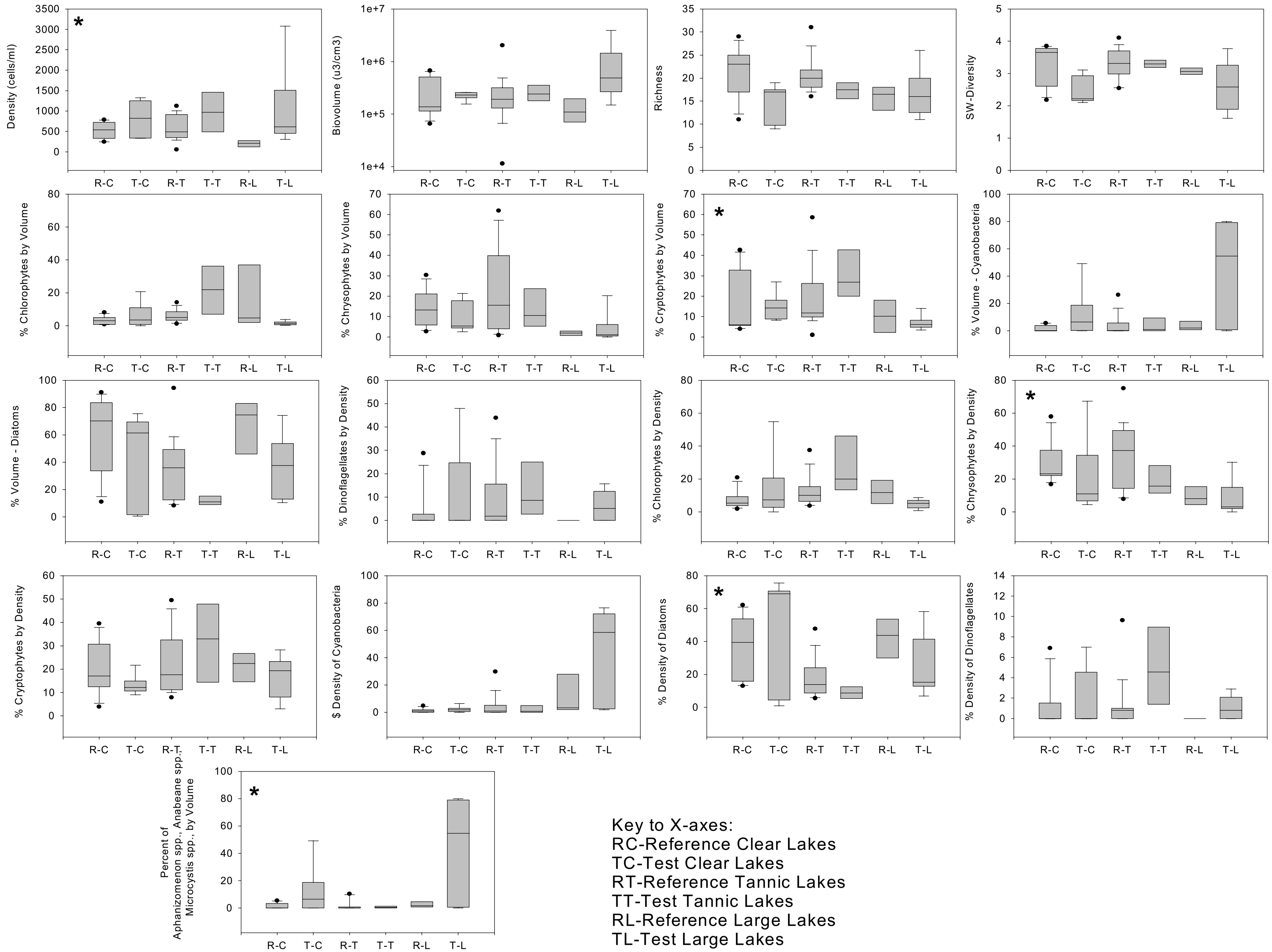

*Figure 2. Box-and-whisker plots for 16 phytoplankton metrics, measured on 40 VT and NH lakes. An * located in the upper left corner of the box-plot indicates that the metric was retained for development of the phytoplankton index.*

Table 3. Index scoring algorithms for five phytoplankton community measures metrics measured on 40 Vermont and New Hampshire lakes. Metrics which do not discriminate between reference and test lakes are not included in the overall index score calculation.

Clear Lakes:				
Metric under evaluation:	*Score Attributed:*			
	1	3	5	IQ Coefficient
Total Density	>1001	675-1001	<674	0.40
%Cryptophytes by Volume	Non-discriminating metric			-29.1
% Chrysophytes by Density	<11%	11-22%	>22%	0.70
% Diatoms by Density	Non-discriminating metric			2.1
% of the Cyanobacteria *Aphanizomenon spp., Anabaena spp., and Microcystis spp.* by Volume	>28%	2.3% - 28%	<2.3%	0.05
Tannic Lakes:				
	1	3	5	IQ Coefficient
Total Density	>1275	903-1275	<903	0.70
% Cryptophytes by Volume	>39%	23% - 39%	<23%	0.4
% Chrysophytes by Density	Non-discriminating metric			5.3
% Diatoms by Density	Non-discriminating metric			3.6
% of the Cyanobacteria *Aphanizomenon spp., Anabaena spp., and Microcystis spp.* by Volume	>5%	1-5%	<1%	0.56
Large Lakes:				
	1	3	5	IQ Coefficient
Total Density	>1404	269 - 1404	<269	0.05
% Cryptophytes by Volume	Non-discriminating metric			-4.2
% Chrysophytes by Density	Non-discriminating metric			1.2
% Diatoms by Density	<21.4%	21.4% - 34.9%	>34.9%	0.5
% of the Cyanobacteria *Aphanizomenon spp., Anabaena spp., and Microcystis spp.* by Volume	>41.8%	3.6% - 41.8%	<3.6%	0.04

It is clear from Figure 3A that, while eight of 14 test lakes fail to meet reference expectations, considerable overlap exists between individual index scores for reference and test lakes. This is not surprising, given that the majority of lakes in Vermont and New Hampshire are in relatively good condition. Since an appropriate cut point for determining impact to the community is not yet established, a proposed cut point for these criteria is the bottom of the reference range of index scores.

Using this cut point, it is clear that one reference lake within each class falls significantly below the scores for the remaining lakes. These lakes are Long Pond (Greensboro, VT), Little Elmore Pond (Elmore, VT), and Shadow Lake (Glover, VT), for the 'well-buffered,' 'low-alkalinity,' and 'large lakes' classes respectively.

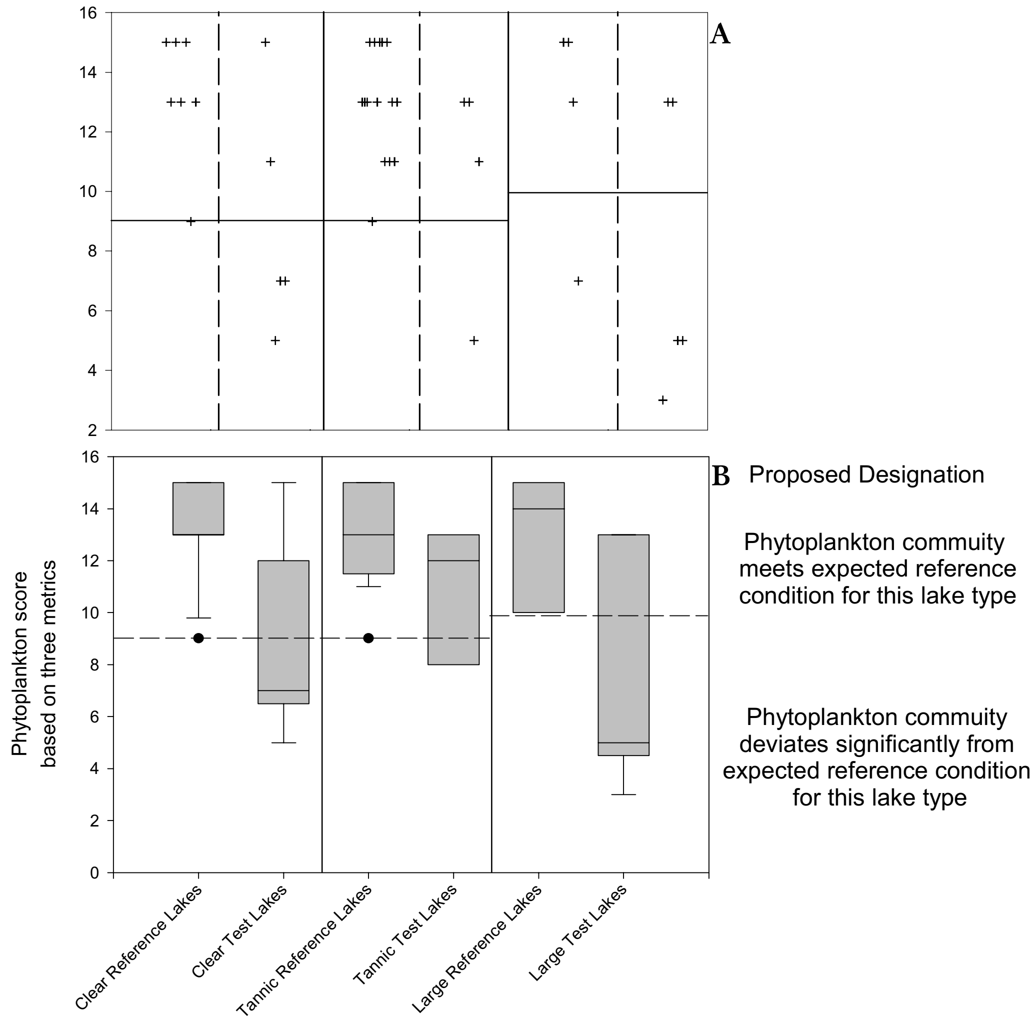

Figure 3. Phytoplankton index scores for 40 Vermont and New Hampshire bioassessment project lakes. Actual individual lake scores (A), and Tukey box-plots and proposed cutpoints for determining impact to the phytoplankton community (B) are shown.

Regarding Long and Little Elmore Ponds, these are relatively pristine lakes which can accurately be described as best presently available despite some prior logging in the lakes' respective watersheds. Accordingly, the low scores for these lakes likely identify conditions which are simply at the bottom of the reference range of the phytoplankton index.

The situation is different for Shadow Lake. This lake is characterized by a low overall algal density, but a high volume of Cyanobacteria in concert with a low density of diatoms. Despite very low mean total phosphorus concentration, Shadow Lake is in fact well settled with shoreline dwellings, and is ringed by unpaved roads. Based on the available phytoplankton data, Shadow Lake may best be reallocated to the test lake category. This, however, would result in a very low sample of reference-class 'large lakes' (n = 3). Given the low number of this lake type sampled to date, application of the Lake and Reservoir Bioassessment and Biocriteria Technical Guidance Document's 'trisection' scoring method may be warranted. By this scoring algorithm, the range of scores from the zero to 95th percentilefor all lakes within a class is trisected, and the best third of each metric's range is considered reference. This approach is problematic in that it a-priori rejects nearly two-thirds of the lakes in a class as not meeting reference conditions.

The comprehensive project report will provide details behind the analysis presented above. It is anticipated that the analytical approach outlined above will be applied to the macrophyte and macroinvertebrate assemblage data, as well as for trophic indicators.

Reporting:

No new reporting (beyond quarterly) is presently available. A comprehensive analysis and writeup of all project data is in progress.

Synopsis of Progress:

Activities undertaken in conjunction with the Bioassessment component of this bi-state initiative are behind schedule. The following is a list of Bioassessment component milestones (a ✓ indicates completion of task):

✓Task 1) *Review and reassess the lake classification and biological metrics employed by the 1995 104(b)3 lake bioassessment.*

✓Task 2) *Review and tailor methods presented in the draft Lake and Reservoir Bioassessment and Biocriteria Technical Guidance Document to be appropriate for Vermont and New Hampshire lakes.*

✔Milestone: Preparation of workplan detailing specific sampling methodologies and biometrics under evaluation. Completed workplan submitted to EPA Region 1 project contact (Peter Nolan). Expected date of completion 6/15/96. Completed 6/25/96

✓Task 3) *Conduct biological and chemical sampling on six lakes/State in 1996, an additional six lakes/State in 1997, and 10 Vermont lakes in 1998.*

✔Milestone: Conduct field bioassessment operations. Expected date of completion for year 1, 10/01/96. Expected date of completion for year 2, 10/01/97. Expected date of completion for year 3, 10/01/98. Year 1 completed 10/3/96; Year 2 completed 10/17/97.

Task 4) *Pick(✔), sort(✔) and identify benthic macroinvertebrates to lowest taxonomic level as determined under task 2 (above). Identify phytoplankton to lowest taxonomic level as determined under task 2 (above). Calculate biometrics for all biological data. Analyze physico-chemical data(✔) and calculate appropriate physico-chemical metrics(✔).*

Milestones:
- *Macroinvertebrate taxonomy -1996 lakes, completed 10/1997* ✔
- *Macroinvertebrate taxonomy - 1997 lakes* ☐
- *1996 mid-project presentation, completed 3/1997* ✔
- *1997 mid-project presentation, completed 3/1998*✔
- *Trial criteria development, 1996-1997 lakes, completed 3/1998* ✔
- *Redraft phytoplankton criteria, completed 10/1999* ✔
- *Redraft of macrophyte criteria* ☐
- *Redraft of trophic criteria* ☐
- *Draft trial macroinvertebrate criteria* ☐

Task 5) Prepare a Bioassessment Component comprehensive final report.