9-2005

Final Report: Evaluation of the Morphoedaphic Index and Sediment Diatoms for Inference of Pre-European Settlement Total Phosphorus Concentration in EPA Region 10 Lakes

Rich Miller
Portland State University

Aaron Hook
Portland State University

Richard Petersen
Portland State University

Mark D. Sytsma
Portland State University, sytsmam@pdx.edu

Final Report
Evaluation of the Morphoedaphic Index and Sediment Diatoms for Inference of Pre-European Settlement Total Phosphorus Concentration in EPA Region 10 Lakes

Prepared for:
US Environmental Protection Agency
and the
Pacific States Marine Fisheries Commission

PSMFC Contract Number:04-14

Prepared by:
Rich Miller, Aaron Hook, Richard Petersen, and Mark Sytsma
Center for Lakes and Reservoirs
Portland State University
Portland, OR

September 2005

Final Report
Evaluation of the Morphoedaphic Index and Sediment Diatoms for Inference of Pre-European Settlement Total Phosphorus Concentration in EPA Region 10 Lakes

Introduction

Water quality in many Northwest lakes has declined over the past century due, in part, to increased anthropogenic nutrient loading (Edmonson and Lehman, 1981). Under the Clean Water Act, resource managers such as the Washington Department of Ecology, Oregon Department of Environmental Quality, and tribes are responsible for restoring and protecting the integrity of these waters. Targets for restoration or criteria for impairment are not well defined, however, and may naturally vary by geology, hydrology, morphometry, and climate.

One way to determine whether lakes are impaired and to identify restoration targets is to assess lake reference conditions (EPA 2000). Reference conditions are the water quality conditions that would exist in the absence of anthropogenic perturbation. Reference conditions are not nutrient criteria but can be useful in establishing realistic criteria that reflect natural regional variations in water quality. The EPA suggests three general approaches for assessing reference conditions (EPA 2000):

- Observation of reference lake or entire lake population distributions
- Paleolimnological reconstruction of past conditions.
- Estimation of past conditions from mass balance nutrient loading models, morphoedaphic index models, or other mathematical models.

The lake population distribution approach involves observing data from either a set of lakes that are relatively unimpacted or from a random selection of all lakes in a region. The lower percentile of each population distribution is assumed to represent reference conditions. The lower 75th percentile of the relatively unimpacted population distribution has been used as a cutoff for reference conditions while the 25th percentile has been used for the lake population distribution as a whole. The Washington Department of Ecology has used this approach to develop phosphorus criteria (Moore and Hicks 2004). This approach assumes that the percentiles in a distribution represent reference conditions.

Paleolimnological models are based on the observation that sediments continually incorporate information from the overlying water column and that sediment cores provide a history of past conditions. Sedimented diatom-based water quality reconstructions are based on two main observations (Hall and Smol 1992): (1) individual diatom species have restricted nutrient requirements and are sensitive indicators of lake trophic status, and (2) the cell walls of diatoms are abundant and well preserved in most lake sediments and can usually be identified to a specific or subspecific level. Weighted-averaging and calibration statistical models have been used to relate sediment surface diatom assemblages to contemporary water quality parameters. These species-environment relationships, known as transfer functions, can then be used to infer past water quality conditions from diatom remains preserved deep in sediments. This approach has been successfully applied in several geographic regions including the northeastern United States (Dixit et al. 1999), Minnesota (Ramstack et al. 2003), and British Columbia (Reavie et al. 1995).

The morphoedaphic index (MEI) was originally developed to predict fisheries yield in lakes based on lake morphometry and alkalinity or conductivity (Ryder 1965). Vighi and Chiaudani (1985) suggested the utility of the index for inferring natural background phosphorus levels in lakes. Their major assumption was that human activities have not altered lake morphometry and ionic strength to the degree that the same activities have increased nutrient loading. While the model is simple, it has been successfully used in to infer background phosphorus concentration in Midwest lakes (Ramstack et al. 2003).

We used the MEI and the sedimented diatom total phosphorus inference models to assess lake reference conditions for Puget Lowlands and Northern Willamette Valley Ecoregion Lakes.

Materials and methods

Study sites

Seventy lakes throughout the Puget Lowlands and the northern section of the Willamette Valley Level III Ecoregions of Oregon and Washington State were included in the study (Figure 1). Low elevations, mild climate, mixed forests, and significant human development characterize these Ecoregions (Omernik 1987; Omernik 1995). The Puget Lobe of the Vashon Glacier covered the most of the Puget Lowlands Ecoregion during previous continental glaciations but not the Willamette Valley Ecoregion. The study area was limited to these regions in order to minimize variation in environmental parameters other than phosphorus that can affect diatom species composition such as pH and conductivity (Hughes et al. 1994). We selected 70 lakes within the region based on two primary criteria: one, maximum depth greater than five meters, and two, the availability of recent water quality data. A subset of twenty lakes was selected for application of the historic total phosphorus reconstruction models. The twenty lakes were chosen based on the criteria that the cores collected were long enough to capture early or pre-European settlement sediment material. Thirty cm of sediment accumulation in Western US and Canadian lakes generally corresponds to over 100 years of accumulation (Birch 1980; Brugham and Vallarino 1989; Reavie et al. 1995; Macdonald et al. 1999; Paulson 2004).

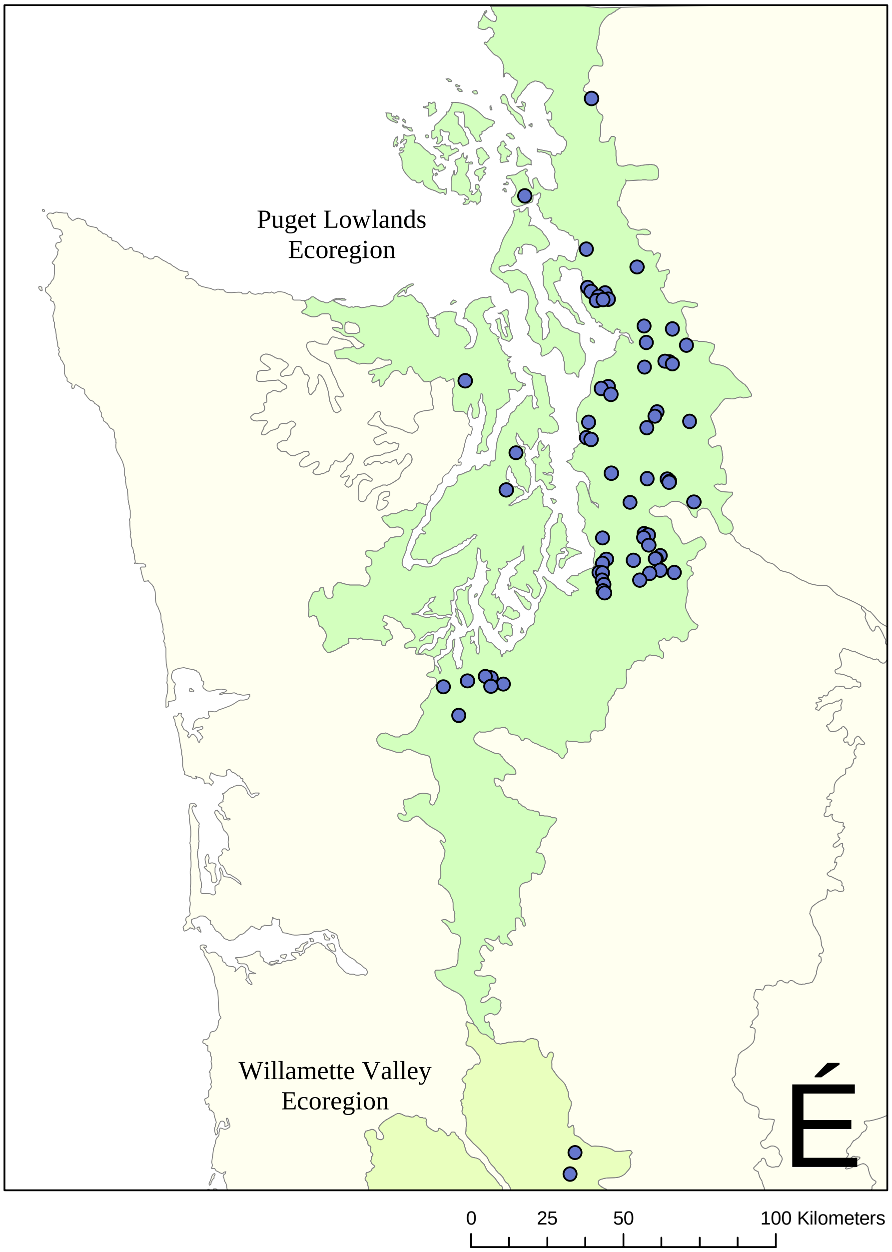

Figure 1. Study sites in the Puget Lowlands and the northern section of the Willamette Valley Ecoregions.

Sedimented diatom dataset

The sedimented diatom dataset consisted of diatom valve counts from surface sediment samples of the 70 lakes and core bottom samples from 20 of the 70 lakes. Cores were collected in 2004 from the deepest portion of each lake using a modified KB-type gravity corer equipped with a 3.8 cm inner diameter core tube (Glew 1991). Cores lengths ranged from 15.5 – 53.5 cm with a median length of 30.5 cm. Cores were sectioned on shore using a portable extruding device (Glew 1988). Surficial 0.5 cm sections of each core and 1 cm sections thereafter were stored on ice in Whirlpak® bags. Bags were labeled by coring location, date, and section depth.

Sediment subsamples (0.2-0.5 g wet weight) were heated for six hours in a mixture of concentrated sulfuric and nitric acid to digest organic matter (Battarbee et al. 2001). Samples were settled overnight, aspirated, washed with deionized water, and left to settle again overnight. The washing process was repeated at least ten times or until sample pH was neutral.

Permanent microscope slides were prepared by filtering subsamples onto a 0.45 μm filters (McNabb, 1960). A section was cut out and placed on a glass slide with immersion oil added to make the filter transparent, followed by placing a cover slip on top, with nail polish applied to the periphery for permanency. Four hundred diatom valves were counted along transects with a Zeiss standard microscope (1000X, phase contrast). Diatoms were identified to species when possible. Broken valves were counted if at least half of the valve was present. Diatom counts were expressed as relative abundance. Diatom species that occurred in at least two lakes with at least one lake with greater than 0.5 % abundance were included in subsequent analyses. A total of 122 species were included in analyses.

Environmental dataset

The environmental dataset consisted of lake surface area, maximum depth, mean depth, and water chemistry variables. Water chemistry variables included surface water total phosphorus (TP), pH, conductivity, alkalinity, tannic acid equivalents, and dissolved organic carbon. TP, pH, conductivity, and Secchi disk depth were measured by Clark, King, Kitsap, Snohomish, and Thurston Counties as part of their lake monitoring programs. Water chemistry data were also provided by Western Washington University, Pacific Ecological Institute, the Washington Department of Ecology, and the Oregon Department of Environmental Quality. TP, pH, and conductivity values were reduced to 2002 - 2004 summer (May – October) averages for each lake. TP values consisted of averages of at least 5 dates per lake and a median of 18 dates. Conductivity and pH values were available for 38 of the 70 lakes. All other environmental parameters were available for all lakes (Table 1).

Dissolved organic carbon (DOC), tannic acid equivalents, and alkalinity samples were collected on the dates of sediment core collections. Samples were filtered through GF/F

Table 1. Location, size, depth, and water quality values for the 70 lake dataset.

Lake	Latitude	Longitude	Level III Ecoregion	Area (acres)	Mean depth (m)	Maximum depth (m)	Mean TP (µg/l)	Secchi depth (m)	Tannic acid equiv. (mg/l)	DOC (mg/l)	Alkalinity (mg CaCO3/l)	Cond. (µS/cm)	pH (units)
Blue	45.5536	122.4529	Willamette Valley	61	3.4	7.3	31	0.9	0.5	3.6	56	113	8.4
Lacamas	45.6205	122.4321	Willamette Valley	315	7.3	19.8	28	1.5	0.6	2.8	33	87	8.3
Deep	46.9082	122.9124	Puget Lowlands	66	3.7	5.2	10	4.1	0.8	3.1	20	77	7.8
Black A	46.9869	122.9770	Puget Lowlands	570	5.8	8.8	28	2.0	1.0	3.6	26	84	7.8
South Pattison	46.9937	122.7738	Puget Lowlands	190	4.0	5.8	36	3.2	0.4	3.2	53	150	8.6
St. Clair	46.9947	122.7271	Puget Lowlands	268	9.8	33.5	19	1.8	1.6	6.1	43	115	7.7
North Pattison	47.0010	122.7847	Puget Lowlands	81	4.3	6.7	20	3.2	0.4	2.6	51	149	7.8
Ward	47.0093	122.8770	Puget Lowlands	65	10.1	20.4	8	6.4	0.3	2.6	3	26	7.9
Long	47.0216	122.7767	Puget Lowlands	311	3.7	6.4	28	2.4	0.6	4.4	49	130	8.0
Hicks	47.0241	122.7975	Puget Lowlands	160	5.5	10.7	15	4.6	0.6	3.9	7	36	7.6
Trout	47.2660	122.2800	Puget Lowlands	18	5.2	8.2	24	1.8	2.1	9.9	29	-	-
Fivemile	47.2714	122.2865	Puget Lowlands	38	5.5	9.8	19	1.2	3.2	12.2	17	-	-
Geneva	47.2916	122.2806	Puget Lowlands	29	5.8	14.0	13	4.3	0.6	5.4	30	-	-
Neilson	47.3032	122.1274	Puget Lowlands	19	5.5	9.4	14	1.6	1.9	7.7	15	-	-
North	47.3057	122.2885	Puget Lowlands	55	4.3	10.4	13	4.4	0.9	7.2	21	-	-
Morton	47.3235	122.0827	Puget Lowlands	66	4.6	7.0	9	3.7	0.6	5.4	16	-	-
Dolloff	47.3244	122.2859	Puget Lowlands	21	3.0	5.8	34	1.8	2.6	12.5	33	-	-
Twelve	47.3254	121.9775	Puget Lowlands	43	4.0	8.5	10	3.5	0.6	4.5	35	-	-
Steele	47.3274	122.3034	Puget Lowlands	46	4.0	7.3	13	3.8	0.5	4.3	28	-	-
Sawyer	47.3338	122.0362	Puget Lowlands	279	7.9	17.7	10	2.7	0.7	4.1	57	-	-
Star	47.3545	122.2867	Puget Lowlands	34	7.6	15.2	9	4.0	0.4	3.9	25	-	-
Meridian	47.3633	122.1543	Puget Lowlands	150	12.5	27.4	8	3.5	0.4	3.6	33	-	-
Pipe	47.3649	122.0615	Puget Lowlands	52	8.2	19.8	10	6.6	0.4	4.0	31	-	-
Fenwick	47.3675	122.2711	Puget Lowlands	17	4.0	9.4	22	3.0	0.8	5.1	30	-	-
Lucerne	47.3680	122.0515	Puget Lowlands	16	5.5	11.3	9	5.3	0.5	4.2	32	-	-
Wilderness	47.3746	122.0349	Puget Lowlands	67	6.4	11.6	17	4.2	0.4	2.5	44	-	-
Shadow	47.4063	122.0858	Puget Lowlands	50	6.7	13.7	14	2.3	1.5	7.0	20	-	-
Angle	47.4274	122.2879	Puget Lowlands	102	7.6	15.8	10	3.4	0.4	3.4	12	-	-
Shady	47.4295	122.1074	Puget Lowlands	21	6.4	12.2	8	4.0	0.7	5.4	34	-	-
Spring	47.4382	122.0895	Puget Lowlands	68	5.8	9.8	13	2.1	1.7	8.0	19	-	-
Desire	47.4435	122.1073	Puget Lowlands	72	4.0	6.4	24	2.1	1.5	7.0	21	-	-
Alice	47.5328	121.8878	Puget Lowlands	32	2.4	9.1	12	3.8	0.8	4.6	2	-	-
Boren	47.5330	122.1652	Puget Lowlands	15	5.5	10.4	17	4.3	0.7	5.2	63	-	-
Kitsap	47.5758	122.7035	Puget Lowlands	238	5.7	8.8	21	3.7	0.5	3.3	47	94	7.8
Sammamish	47.5778	122.0888	Puget Lowlands	4893	17.7	32.0	9	4.1	0.3	2.7	41	106	8.5
Pine	47.5887	122.0423	Puget Lowlands	88	6.1	11.9	9	4.7	0.5	4.1	18	-	-

Table 1 (continued). Location, size, depth, and water quality values for the 70 lake dataset.

Lake	Latitude	Longitude	Level III Ecoregion	Area (acres)	Mean depth (m)	Maximum depth (m)	Mean TP (µg/l)	Secchi depth (m)	Tannic acid equiv. (mg/l)	DOC (mg/l)	Alkalinity (mg CaCO3/l)	Cond. (µS/cm)	pH (units)
Beaver 2	47.5916	121.9972	Puget Lowlands	62	6.4	16.5	12	2.7	1.0	6.1	10	-	-
Beaver 1	47.5948	121.9908	Puget Lowlands	12	6.7	16.8	20	1.7	2.2	9.1	7	-	-
Washington	47.6280	122.2626	Puget Lowlands	21500	32.9	65.2	9	6.4	0.4	2.5	38	98	8.1
Island	47.6818	122.6606	Puget Lowlands	43	6.1	10.7	14	4.0	0.8	5.1	26	59	7.5
Haller	47.7195	122.3337	Puget Lowlands	15	5.2	11.0	16	2.0	0.9	3.9	12	-	-
Bitter	47.7271	122.3529	Puget Lowlands	19	4.9	9.4	14	2.3	0.5	4.1	7	-	-
Cottage	47.7528	122.0876	Puget Lowlands	63	4.6	7.6	27	2.7	1.0	5.5	45	-	-
Margaret	47.7685	121.8992	Puget Lowlands	53	5.5	13.1	9	3.8	0.6	3.4	9	-	-
Echo (King Co.)	47.7711	122.3434	Puget Lowlands	12	4.3	9.1	31	2.2	0.8	4.8	12	-	-
Echo (Sno. Co.)	47.7866	122.0511	Puget Lowlands	16	5.2	15.2	9	2.7	0.7	6.3	20	60	7.5
Lost	47.8008	122.0430	Puget Lowlands	12	7.0	13.7	18	1.7	2.0	8.3	37	71	7.2
Martha S.	47.8530	122.2430	Puget Lowlands	57	7.3	14.6	9	6.4	0.5	4.5	26	89	7.6
Serene	47.8689	122.2831	Puget Lowlands	43	4.3	7.0	10	4.9	0.5	4.5	27	80	7.1
Stickney	47.8750	122.2546	Puget Lowlands	24	4.6	10.4	13	3.7	1.3	8.4	40	110	7.4
Leland	47.8977	122.8807	Puget Lowlands	107	4.0	6.1	42	1.4	1.9	10.9	31	70	7.3
Blackman	47.9322	122.0944	Puget Lowlands	57	4.3	8.8	16	3.3	0.8	5.5	25	77	7.0
Storm	47.9385	121.9727	Puget Lowlands	73	6.7	14.0	10	2.1	1.3	5.6	6	26	7.1
Flowing	47.9468	121.9881	Puget Lowlands	131	8.5	21.0	10	2.7	0.7	4.7	11	35	7.1
Panther	47.9477	122.0047	Puget Lowlands	45	7.0	11.0	12	2.4	1.0	5.7	14	37	7.2
Roesiger	47.9930	121.9070	Puget Lowlands	340	11.3	33.5	6	4.4	0.4	3.0	7	24	7.1
Stevens	48.0037	122.0876	Puget Lowlands	1040	19.2	47.2	12	3.8	0.4	3.9	32	-	-
Bosworth	48.0433	121.9698	Puget Lowlands	102	10.7	24.1	7	3.5	0.6	3.8	9	34	7.9
Cassidy	48.0515	122.0938	Puget Lowlands	123	3.4	6.1	46	1.0	4.9	18.0	10	45	6.9
Crabapple	48.1317	122.2733	Puget Lowlands	35	5.5	14.9	10	3.0	0.8	6.6	10	43	7.3
Shoecraft	48.1321	122.3053	Puget Lowlands	132	5.5	10.7	8	3.4	0.5	4.8	32	96	7.6
Loma	48.1335	122.2544	Puget Lowlands	21	3.4	8.5	33	1.5	2.9	12.9	7	44	6.7
Goodwin	48.1382	122.2922	Puget Lowlands	535	7.0	15.2	7	4.4	0.4	4.3	28	74	7.8
Ki	48.1533	122.2625	Puget Lowlands	96	10.1	21.3	10	6.2	0.3	4.6	7	37	7.2
Howard	48.1572	122.3270	Puget Lowlands	26	8.8	15.2	10	5.0	0.6	5.7	40	106	7.6
Martha N.	48.1676	122.3384	Puget Lowlands	59	10.1	21.3	10	4.3	0.9	7.0	26	91	7.4
Armstrong	48.2255	122.1233	Puget Lowlands	30	4.6	7.3	19	2.9	1.0	6.4	23	54	7.3
Ketchum	48.2821	122.3452	Puget Lowlands	24	3.7	6.4	289	1.3	1.1	9.0	47	133	8.2
Campbell	48.4373	122.6167	Puget Lowlands	367	2.4	4.9	28	1.4	0.7	11.3	94	244	8.3
Whatcom	48.6857	122.3080	Puget Lowlands	5000	46.9	100.6	7	4.7	0.3	2.2	18	56	8.0

glass fiber filters (0.7 µm pore size) and stored in the dark at 4°C prior to analysis. DOC filtrate was acidified to pH < 2 with hydrochloric acid. DOC was analyzed by combustion and infrared detection with a Shimadzu model 5000A TOC-V CHS Total Organic Carbon Analyzer. Tannic acid equivalents (TAE), an estimate of dissolved humic substances in the water, were calculated from 440 nm absorbance readings on a Milton Roy Spectronic spectrophotometer (Cuthbert and del Giorgio, 1992). Alkalinity was measured by Gran titration (Wetzel and Likens 1991). Ten percent of newly collected water chemistry samples were replicated for quality assurance.

Data Analysis

Many of the statistical analyses require or suggest that distributions of environmental variables be normally distributed. Total phosphorus, dissolved organic carbon, tannic acid equivalents and lake surface area were log transformed to correct non-normal skewed distributions. Maximum and mean depth were inverse transformed to achieve normality. Additionally, ecological datasets contain redundancies in environmental information, unusual samples, and environmental variables that are not well related to diatom distributions (Birks et al. 1990). Data were examined for these problems prior to further analysis.

Redundancies in environmental data were assessed with Pearson-moment correlations. Within the complete 70 lake dataset, inverse mean depth and inverse maximum depth were highly correlated as were tannic acid equivalents and dissolved organic carbon (Table 2). Alkalinity and conductivity were highly correlated (Pearson-moment correlation = 0.96) in the 38 lake dataset that contained conductivity and pH values. Mean depth, tannic acid equivalents, and conductivity were removed from the dataset because of the redundancies with other variables. pH was removed from the dataset because it was not available for all lakes.

Unusual or outlier lakes were evaluated using the Grubbs test (Zar, 1984) and were removed from further analysis. The mean log total phosphorus concentration in Ketchum Lake was an outlier from the rest of the lakes with a value 4.9 standard deviations from the population mean. Lakes Washington, Sammamish, and Whatcom were outliers in terms of log surface area as was Campbell Lake for alkalinity. The remaining environmental dataset consisted of 65 lakes and 6 environmental parameters: $\log_{10}$surface area, inverse maximum depth, $\log_{10}$total phosphorus, Secchi disk depth, $\log_{10}$dissolved organic carbon, and alkalinity.

The program C2 was used to generate species-environment transfer functions and perform environmental variable reconstructions (Juggins 2003). Several types of weighted-averaging and calibration models are available but all require a strong environment-diatom species relationship (Birks 1998). The strength of relationships between environmental variables and diatom species assemblages were assessed with canonical correspondence analysis (CCA) using the program CANOCO (ter Braak 1987). Canonical correspondence analyses as well as weighted averaging regression and calibration models have the underlying assumption that diatom taxa respond to environmental parameters in a unimodal rather than a linear manner, i.e., they have optima and tolerances across an environmental gradient. Detrended correspondence analysis was used to determine whether the assumption of unimodal response was suitable for our dataset (ter Braak and Prentice 1988). Root mean square errors of the predictions from the weighted – averaging and calibration models were determined by leave-one-out jackknifing. Rare diatom species were downweighted in the weighted averaging models.

Morphoedaphic index inferred TP concentrations were calculated according to Vighi and Chiaudani's (1985) equation derived empirically from North American reference lakes:

$$\text{Log [TP]} = 1.44 + 0.33\ (\text{Log MEI}_{alk})$$

where:

$$\text{MEI}_{alk} = (\text{alkalinity in milliequivalents/l}) \div (\text{mean depth in meters}).$$

Table 2. Pearson product-moment correlations between environmental variables.

	$\log_{10}$area	mean depth^{-1}	max. depth^{-1}	$\log_{10}$TP	Secchi	$\log_{10}$TAE	$\log_{10}$DOC	Alkalinity
$\log_{10}$area	1.00	-	-	-	-	-	-	-
mean depth^{-1}	-0.42	1.00	-	-	-	-	-	-
max. depth^{-1}	-0.32	0.88	1.00	-	-	-	-	-
$\log_{10}$TP	-0.19	0.55	0.61	1.00	-	-	-	-
Secchi	0.24	-0.45	-0.43	-0.62	1.00	-	-	-
$\log_{10}$TAE	-0.35	0.34	0.33	0.48	-0.67	1.00	-	-
$\log_{10}$DOC	-0.47	0.46	0.40	0.46	-0.56	0.85	1.00	-
Alkalinity	0.20	0.20	0.30	0.32	-0.09	-0.18	-0.02	1.00

Results

Strength of species-environment relationships

The gradient length of the first DCA axis (3.1 standard deviation units) indicated that use of analyses such as CCA and weighted-averaging regression that are based on unimodal response were appropriate for our dataset (ter Braak and Prentice 1988). The gradient length of the second DCA axis was 2.4 standard deviation units, a value indicating that either unimodal or linear methods were applicable. CCA axis 1 and axis 2 explained 14.9 percent of the variance in diatom species in the 65-lake data set constrained to the six environmental variables (Figure 2). A larger proportion (47.3 percent) of the species-environment correlations were explained by the first two CCA axes. The forward selection option in CCA analysis identified four environmental variables as significantly and independently related to the diatom assemblages (Table 3). Alkalinity was the most closely related variable to the diatom assemblages followed by surface area, total phosphorus, and dissolved organic carbon. Maximum depth and Secchi disk depth were not significantly independently related to the diatom assemblages. A series of CCA analyses with each environmental variable constrained to CCA axis 1 confirmed the significance and relative strength of each of the variables in the species environment relationships (Table 3). Development of a numerical transfer function between diatom species and an environmental variable has been successful when the ratio of the first two CCA axes is high (>0.5), the correlation to the first axis is high (>0.5), and the variable contained a significant and high (>5 %) proportion of the total explained variance of species in a constrained CCA (Schönfelder et al. 2002). Alkalinity met all the criteria for a good transfer function candidate while TP did not meet all criteria but was marginal (Table 3). Partial CCA analysis provided other evidence that development of a TP transfer function was appropriate. Like a constrained CCA, a partial CCA constrains each environmental variable to the first axis. Unlike the constrained CCA, however, other variables are treated as co-variables. Environmental variables that explain greater than two percent of the variation in diatom species are good candidates for transfer function development (Schönfelder et al. 2002). Total phosphorus and alkalinity accounted for 4.6 and 4.3 percent of the variation in diatom species respectively. Overall, analysis of the strength of the species environment relationships suggests that while development of alkalinity and TP transfer functions were warranted, results of a TP transfer function would not be as robust.

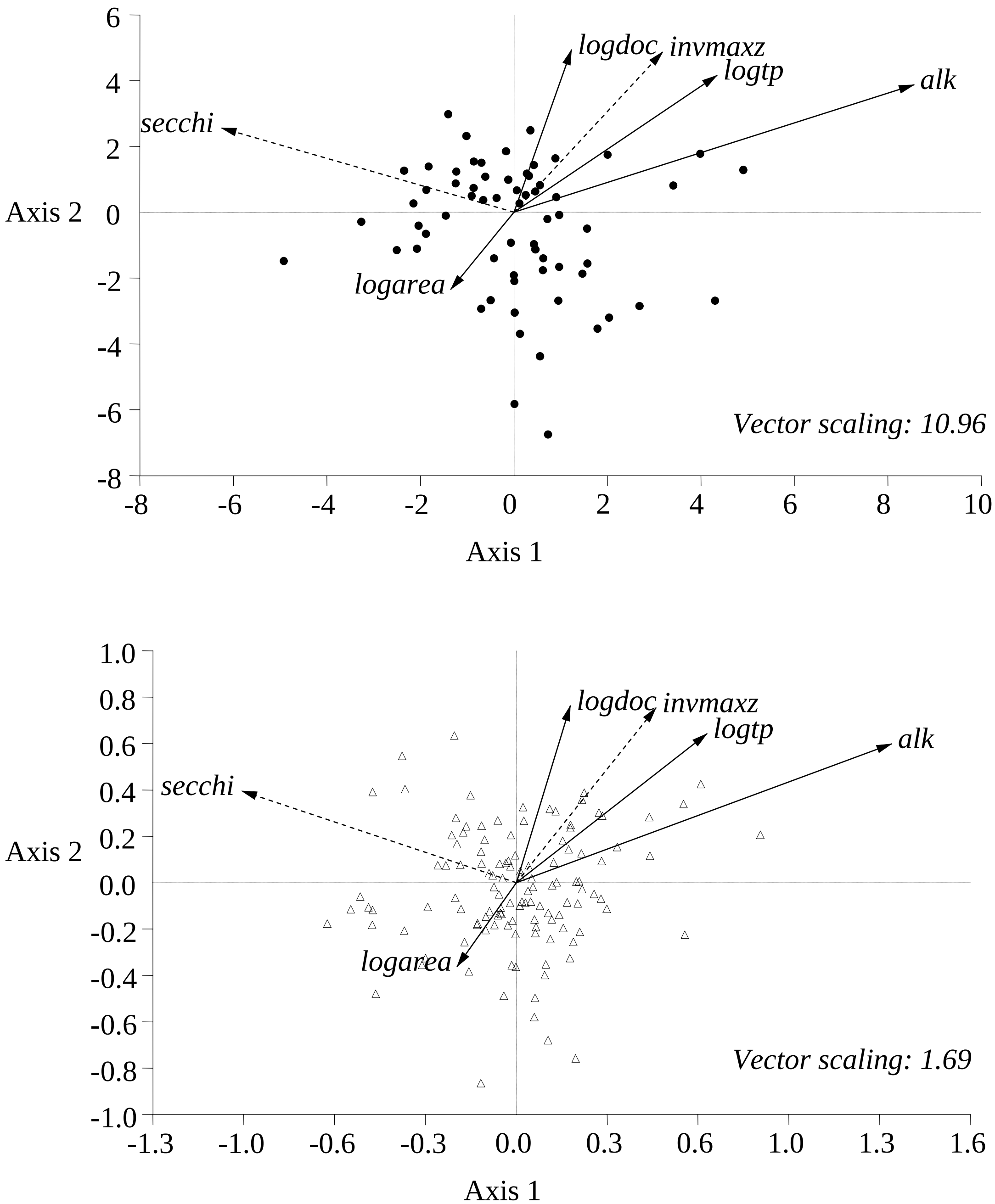

Figure 2. Canonical correspondence analysis of the 65 lake dataset. Circles represent lake scores and triangles represent diatom species scores. Dashed environmental vectors were not significantly related to the diatom species. Note the environmental variable vectors are on different scales on the two plots.

Table 3. Results of canonical correspondence analyses to determine the strength of multivariate species-environment relationships (forward selected CCA) and univariate species environment relationships (series of CCA's with each environmental variable constrained to CCA axis 1).

	Multivariate species-environment relationships		Univariate species-environment relationships			
Variable	Additional explained total diatom variance	Monte-Carlo significance (Bonferoni adjusted)	Marginal explained variance	Monte-Carlo significance (Bonferoni adjusted)	Eigenvalue 1 to 2 ratio	Correlation with axis 1
Alkalinity	6.9	< 0.008	7.0	< 0.008	0.67	0.76
$\log_{10}$area	3.5	< 0.008	4.7	< 0.008	0.42	0.67
$\log_{10}$TP	3.8	< 0.008	4.5	< 0.008	0.39	0.68
$\log_{10}$DOC	3.5	< 0.008	4.0	< 0.008	0.32	0.64
Maximum depth^{-1}	-	not significant	-	not significant	-	-
Secchi disk depth	-	not significant	-	not significant	-	-

Transfer function development

We developed transfer functions for alkalinity, total phosphorus, and dissolved organic phosphorus. Although the relationship between surface area and diatom species assemblages was sufficiently strong for developing a transfer function, we were not interested in reconstructing historic lake surface area. Several transfer function regression models were evaluated including those based on unimodal species responses to environmental variables (weighted-averaging with classical deshrinking (WACD), weighted-averaging with inverse deshrinking (WAINV), weighted-averaging partial least squares(WA-PLS)), and those based on linear responses to environmental variables (partial least squares (PLS)). The WA-PLS model provided the best fit between inferred values and measured values for all parameters; however, there was a significant pattern in the model residuals. The model overestimated values at the low end of the observed value range and underestimated values at the high end of the range. This pattern of residuals was present in all models with the exception of the WACD model (Figure 3). The jackknifed regression statistics for the WACD were best for alkalinity followed by total phosphorus and dissolved organic carbon (Table 4). WACD model regression parameters were used for all subsequent environmental parameter reconstructions. *Coscinodiscus spp*. and two *Melosira* species had the highest model inferred total phosphorus optima and tolerances while a *Navicula* species has the lowest TP optima and tolerance.

Table 4. Performance statistics of the weighted-averaging and calibration model with classical deshrinking

Parameter	r^2 (jackknifed)	RMSEP (jackknifed)
Alkalinity	0.425	13.97 mg $CaCO_3$/l
Log_{10}TP	0.375	0.214 $\log_{10}$TP
Log_{10}DOC	0.282	0.234 $\log_{10}$DOC

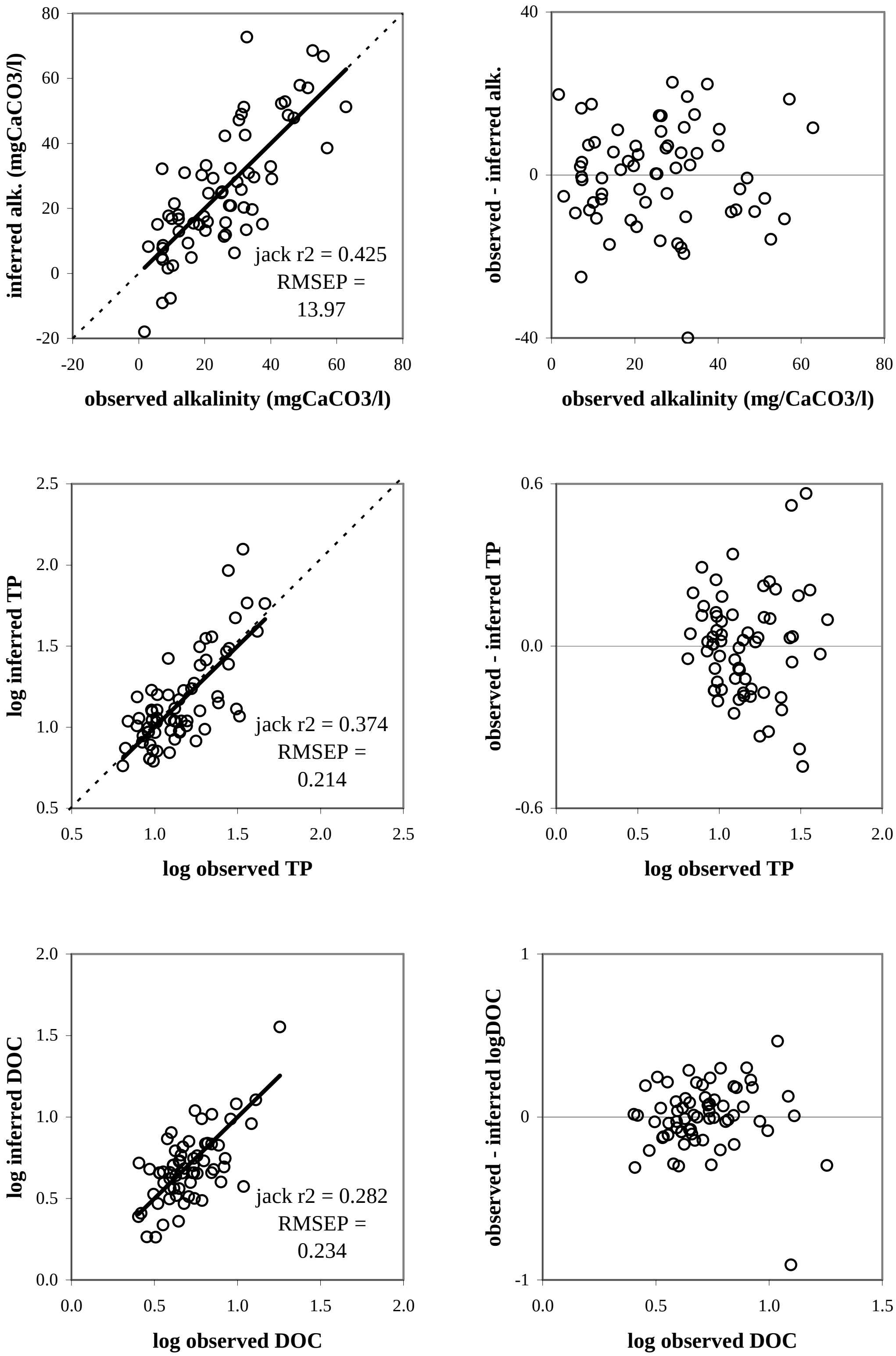

Figure 3. Plots of observed versus inferred alkalinity, log total phosphorus, and dissolved organic carbon (left panels) and regression residuals (right panels) based on a weighted-averaging and calibration models using classical deshrinking.

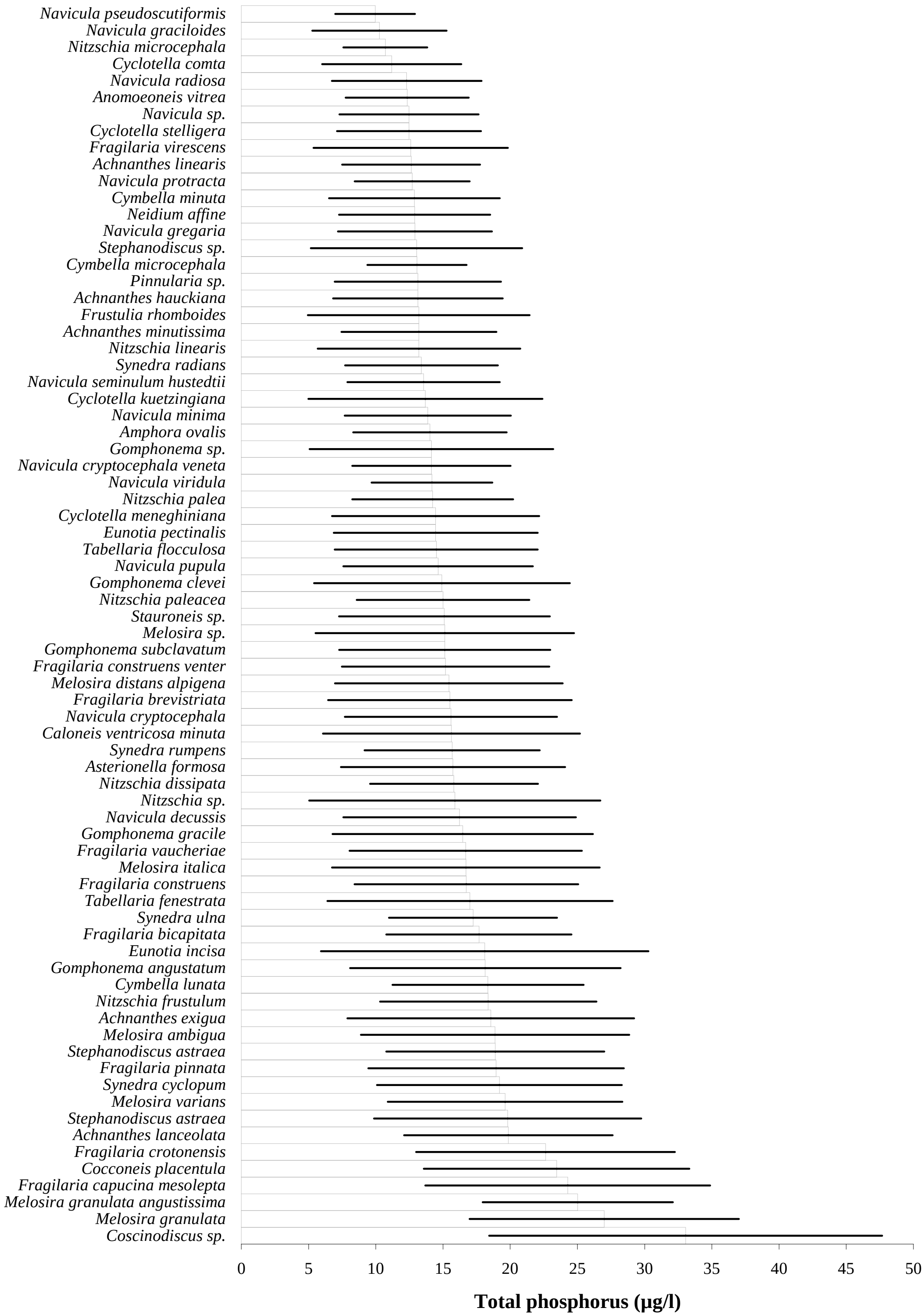

Figure 4. Species optima and tolerances for surface water total phosphorus for species present in 10 or more study lakes.

Diatom based reconstructions

Modern observed TP concentrations in were significantly higher than WACD model inferred pre- or early Europeans settlement TP concentrations for three of the 20 lakes with core bottom diatom counts (Figure 5). One lake had a significantly lower modern TP concentration than historical concentration. Alkalinity changed significantly in eight of the 20 lakes; six increased and two decreased. Dissolved organic carbon concentrations changed in nine of the 20 lakes; five higher and four lower. Across the 20 lake population, however, modern observed alkalinity, TP, and DOC values did not significantly increase from historic values (t-test $p = 0.22$, $p = 0.49$, and $p = 0.76$ respectively).

Morphoedaphic index based total phosphorus reconstructions

Because there were significant changes in alkalinity in several of the lakes, morphoedaphic index inferred TP values were calculated based on measured alkalinity as well as diatom inferred alkalinity (Figure 6). MEI inferred TP concentrations were not significantly different than diatom inferred concentrations using either measured alkalinity ($p = 0.64$) or inferred alkalinity ($p = 0.14$) values

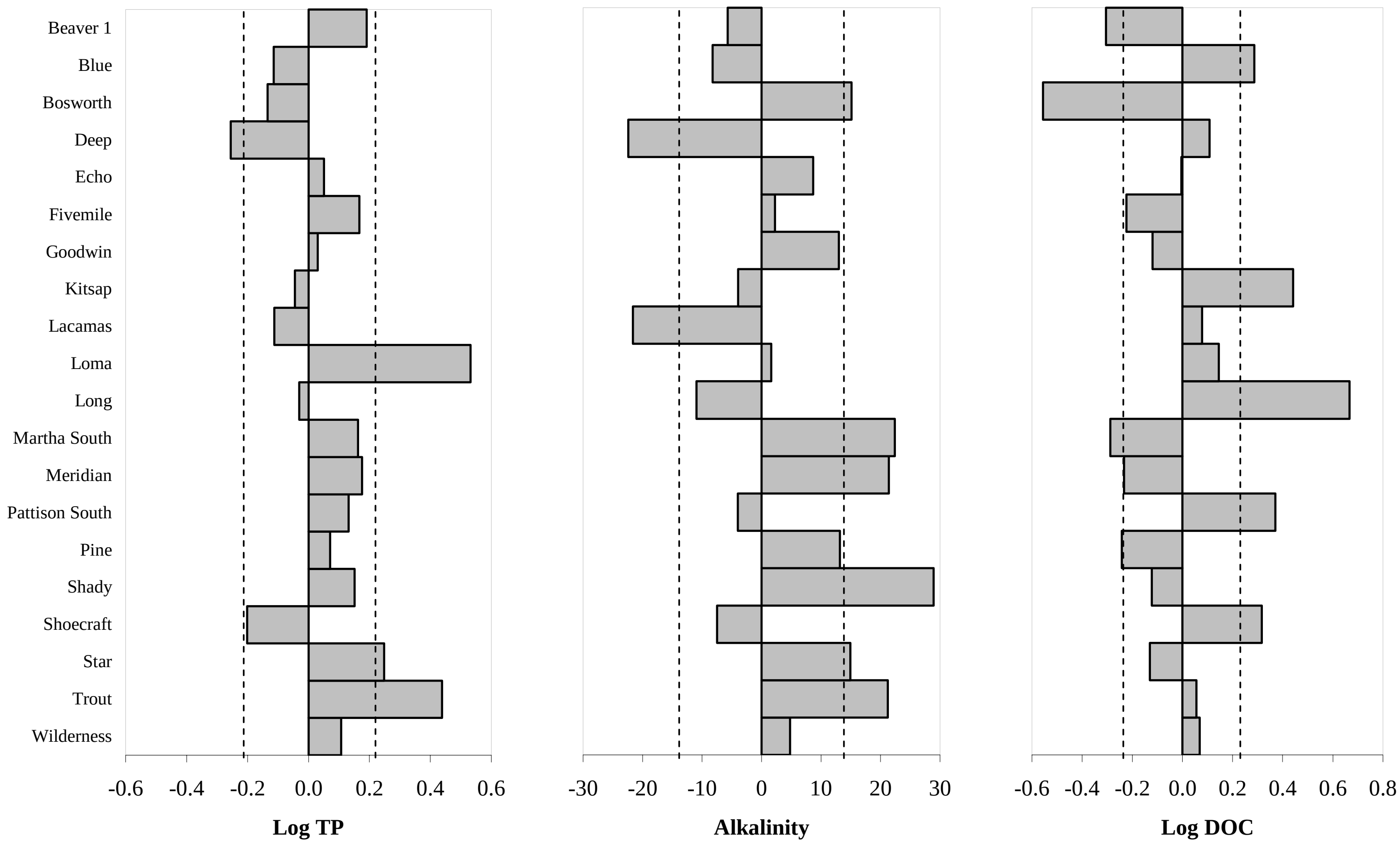

Figure 5. Differences between modern measured and inferred historical background water quality variables. Dashed lines represent the jackknifed root mean square errors of the prediction.

Inference of Total P Concentration in Region 10 Lakes

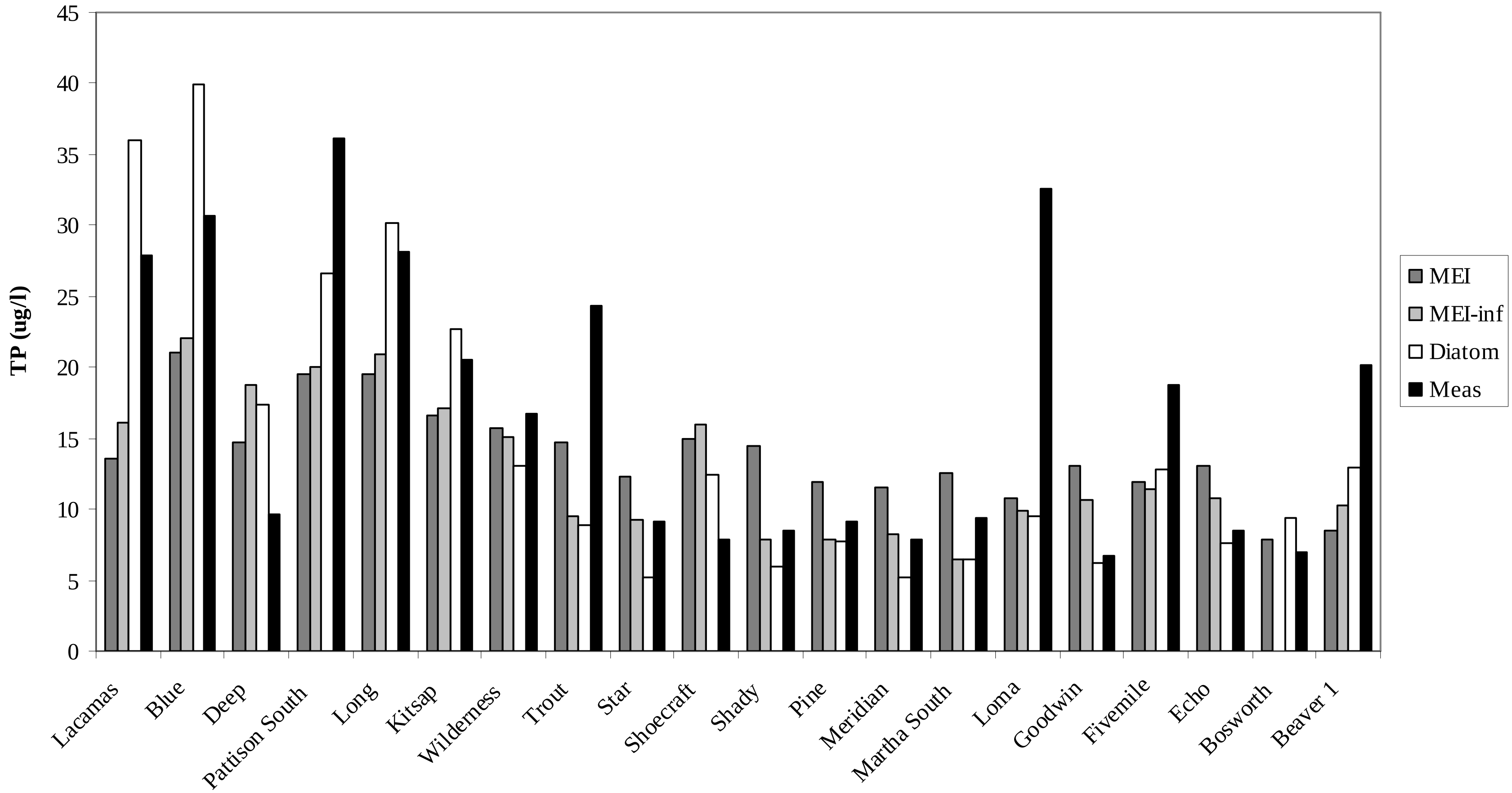

Figure 6. Measured modern and model inferred reference total phosphorus concentrations for 20 Puget Lowlands and Northern Willamette Valley Ecoregion lakes

Discussion

Results of diatom inferred alkalinity reconstructions suggested that the MEI inferred TP model's assumption of the conservative nature of alkalinity may not be valid for the soft-water lakes in the Puget Lowlands Ecoregion. MEI reconstructed TP concentrations agreed well with diatom inferred TP concentrations for most lakes, however, agreement was not as strong for lakes in the Willamette Valley Ecoregion and the southern part of the Puget Lowlands Ecoregion (Figure 6). The southern lakes generally had higher TP concentrations than the rest of the population and were at the upper end of the transfer function model's range. Several northern lakes such as Loma and Trout, however, also had measured TP values towards the upper end of the model but good agreement between diatom inferred and MEI inferred TP values. One difference between the southern and northern lakes is the degree of glaciation. During the last glaciation, the Willamette Valley was not glaciated, the southern Puget Lowlands were at the edge of Puget Lobe of the Vashon Glacier, and the rest of the Puget Lowlands was fully glaciated.

Statistically, there was no population-wide difference between modern measured, diatom-reconstructed, and MEI-reconstructed TP concentrations. This suggests that population distribution approaches based on modern measured TP values are valid for determining reference conditions in the Puget Lowlands. The Washington Department of Ecology used this approach to flag lakes high in TP that require restoration or further study. Their flag or "action" value for TP in the Puget Lowlands Ecoregion is 20 µg/l. This compares well with the range of TP values derived from the MEI and sediment diatom-based models (Table 5).

Table 5. Comparison of pre- or early European settlement TP estimates derived with modern measured TP values.

	MEI inferred (measured alkalinity)	MEI inferred (inferred alkalinity)	Sediment diatom inferred	Modern measured
TP ± 95% c.i. (µg/l)	14 ± 2	13 ± 2	15 ± 5	17 ± 4

References

Battarbee, R. W., L. Carvalho, V. J. Jones, R. J. Flower, N. G. Cameron, H. Bennion, and S. Juggins. 2001. Diatoms. In Smol, J. P., H. J. B. Birks and W. M. Last (eds), Tracking Environmental Change Using Lake Sediments, Volume 3, Terrestrial, Algal, and Siliceous Indicators. Kluwer Academic Publishers, Dordrecht, The Netherlands.

Birch, P. B., R. S. Barnes, and D. E. Spyridakis. 1980. Recent Sedimentation and Its Relationship with Primary Productivity in Four Western Washington Lakes. Limnology and Oceanography 25:240-247.

Birks, H. J. B, D.G. Frey and E.S. Deevey. 1998. Review #1: Numerical tools in palaeolimnology - Progress, potentialities, and problems. Journal of Paleolimnology 20:307-332.

Brugam, R. B., and J. Vallarino. 1989. Paleolimnological investigations of human disturbance in western Washington lakes. Archiv fur Hydrobiologie 116:129-159.

Cuthbert, I. D., and P. Del Giorgio. 1992. Toward a standard method of measuring color in freshwater. Limnology and Oceanography 37: 1319–1326.

Edmondson, W. T., and J. T. Lehman. 1981. The Effect of Changes in the Nutrient Income on the Condition of Lake Washington. Limnology and Oceanography 26:1-29.

Glew, J. R. 1988. A portable extruding device for close interval sectioning of unconsolidated core samples. Journal of Paleolimnology 1:235 – 239.

Glew, J. R. 1991. Miniature gravity corer for recovering short sediment cores. Journal of Paleolimnology 5:285 – 287.

Hall, R. I., and J. P. Smol. 1992. A weighted - averaging regression and calibration model for inferring total phosphorus concentration from diatoms in British Columbia (Canada) lakes. Freshwater Biology 27:417-434.

Hughes, R. M., S. A. Heiskary, W. J. Matthews, and C. O. Yoder. 1993. Use of ecoregions in biological monitoring. Lewis Publishers, Boca Raton, FL.

Juggins. S. 2003. C2 User guide. Software for ecological and palaeoecological data analysis and visualisation. University of Newcastle, Newcastle upon Tyne, UK. 69pp.

Macdonald, R. W., D. P. Shaw, and C. Gray. 1999. Contaminants in lake sediments and fish. In: Gray, C. and T. Tuominen (eds.), Health of the Fraser River Aquatic Ecosystem: A Synthesis of Research Conducted under the Fraser River Action Plan. Environment Canada Pub. No. DOE FRAP 1998-11. p 23-45.

McNabb, C. D. 1960. Enumeration of freshwater phytoplankton concentrated on the membrane filter. Limnology and Oceanography 5:57-61.

Moore, A. and M. Hicks. 2004. Nutrient criteria development in Washington State: phosphorus. Washington State Department of Ecology. Pub No. 04-10-033.

Omernik, J. M. 1987. Ecoregions of the conterminous United States. Map (scale 1:7,500,000). Annals of the Association of American Geographers 77(1):118-125.

Omernik, J. M. 1995. Ecoregions: A spatial framework for environmental management. In: Biological Assessment and Criteria: Tools for Water Resource Planning and Decision Making. Davis, W.S. and T.P. Simon (eds.) Lewis Publishers, Boca Raton, FL. pp. 49-62.

Paulson, A. J. 2004. Sources of mercury in sediments, water, and fish of the Lakes of Whatcom County, Washington. U.S. Geological Survey Scientific Investigations Report 2004-5084. 98 p.

Ramstack, J., S. Fritz, D. Engstrom, and S. Heiskary. 2003. The application of a diatom-based transfer function to evaluate regional water-quality trends in Minnesota since 1970. Journal of Paleolimnology 29:79-94.

Reavie, E. D., R. I. Hall, and J. P. Smol. 1995. An expanded weighted-averaging model for inferring past total phosphorus concentrations from diatom assemblages in eutrophic British Columbia (Canada) lakes. Journal of Paleolimnology 14:49-67.

Reavie, E. D., J. P. Smol, and N. B. Carmichael. 1995. Postsettlement eutrophication histories of six British Columbia (Canada) lakes. Canadian Journal of Fisheries and Aquatic Sciences 52:2388-2401.

Ryder, R. A. 1965. A method of estimating the potential fish production of north-temperate lakes. Trans. Am. Fish. Soc. 111:154-164.

Schönfelder, I., J. Gelbrecht, J. Schönfelder, and C. E. W. Steinberg. 2002. Relationships between littoral diatoms and their chemical environment in Northeastern German lakes and rivers. Journal of Phycology 38:66-89.

Smol et al. 2001 – lab manual

Smol, J. P. 2002. Pollution of Lake and Rivers: A Paleoenvironmental Perspective. Oxford University Press, New York, NY.

ter Braak, C. J. F. 1987. The analysis of vegetation-environment relationships by canonical correspondence analysis. Vegetatio 69:69-77.

ter Braak, C. J. F. and I. C. Prentice, 1988. A theory of gradient analysis. Advances in Ecological Research 18: 271–317.

Vighi, M. and G. Chiaudani. 1985. A simple method to estimate lake phosphorus concentrations resulting from natural background loadings. Water Research 19: 987–991.

Wetzel, R. G. and G. E. Likens. 1991. Limnolgical Analyses. Springer-Verlag, New York, NY.

Zar, J.H. 1999. Biostatistical Analysis. Prentice-Hall, Upper Saddle River, NJ.