Pewaukee Lake
Phosphorus Monitoring
2003-2004

Wisconsin Lutheran College, Chemistry Dept.
Technical Bulletin 001, March 2005
Prepared by
Daniel D. Ebeling, PhD

Funding provided by
Lake Pewaukee Sanitary District

Significant sampling analysis performed by
Wisconsin Lutheran College chemistry majors:
Michael Poston, Anatole Wiik, Aaron Kittell, and Jeffrey Potratz

Special thanks to Mr. Charles Shong for technical support and
willingness to teach teachers and students alike.

Table of Contents

ABSTRACT

Phosphorus load and concentration:

Water and sediment samples were collected from Pewaukee Lake (Pewaukee, WI) and analyzed for total phosphorus (P) concentration. Water flow rates were measured to determine P loading from the inflowing streams. P loading calculations are available for the summer of 2004, while only P concentration data is available for the summer 2003.

Method analysis:

The sediment samples were digested with sulfuric acid and persulfate in an autoclave. The water samples were digested with persulfate in an autoclave. Phosphorus concentrations were determined using a uv-vis spectrometer and analyzing the 625 nm absorption peak based on the association of malachite green with phosphomolybdate under acidic conditions. Lower detection limit results from this complex having a molar absorptivity constant more than three times greater than the phosphoantimonylmolybdenum blue in the standard Murphy-Riley method. While the malachite green method does provide signal at lower concentrations, the Murphy-Riley method provided greater dynamic range and a more reliable standard curve.

INTRODUCTION

The most important nutrients in aquatic ecosystems are phophorus (P) and nitrogen (N). In Pewaukee Lake, plant and algae growth is limited by P as it is a body of fresh water.[1] In marine areas the opposite is true, and N is the most likely limiting nutrient.[2]

Elevated nutrient levels create a eutrophic environment with increased plant and algae growth. When the plants and algae die and decompose, dissolved oxygen levels in lakes and streams are depleted, which can lead to odors, death of fish, and a general degradation of the aesthetic and recreational value of the environment.[3] Pewaukee Lake has been classified as a eutrophic lake by Aqua Tech, Inc. in 1972 and by Southeastern Wisconsin Reagional Planning Commission (SEWRPC) in 1984.[4,5]

Phosphorus measurement background.

There are several spectrometric methods to determine phosphorus concentration in water and soil extracts. Methods based on the formation of phosphoantimonylmolybdenum blue with ascorbic acid as a reductant are commonly suggested for the determination of P in water and in soil-water extracts. These are based on the procedure of Murphy and Riley for orthophosphate analysis.[6] Alternatively, a method has been developed in which the molybdophosphate is reacted with the cationic dye, malachite green, to produce absorbance at 625 nm.[7]

In order to measure total P, the condensed and organic forms of phosphorus must be converted into orthophosphate for spectrometric analysis. Commonly, fusion or boiling acid digestion techniques are used to convert the phosphorus.[8] A more easily automated digestion by persulfate and autoclave was chosen for this step.[9]

Note: Acceptable units for reporting P concentration in sediment are in terms of parts per million (ppm), in water, it is common to use either ppb or µg/L. Some comparisons are made between the different unit systems, so it will be helpful to keep in mind: 1 ppm = 1 mg/L = 1 µg/mL = 1,000 ppb = 1,000 µg/L.

MATERIALS AND METHODS

Phosphorus concentration measurements were made at several central lake locations. Measurements of total P in water samples from several central lake locations and the outflow water in the Pewaukee River have been made. Measurement of P concentration and flow rates from the inlets Coco Creek, Meadowbrook, and Zion Creek provided the data in order to calculate total P loads. This is a portion of the P budget for the lake, and can indicate overall lake trends and clarify a picture of major P contributors. A comprehensive P budget would take into consideration estimates of inputs from direct drainage, precipitation, and dry fallout. The necessary outputs would include measurements of the outgoing river flow and estimates of the weed and fish harvests.

Flow Measurements

A rotating-cup Price AA current meter was used in a velocity-area method to measure inflow. The velocity-area method involves measuring the channel area and water velocities of a stream at a cross section that is perpendicular to the main flow of the channel. The channel is divided into a number of vertical "subsections." The area and mean velocity in each subsection is measured and the subsection discharge is computed. The total discharge within the stream is the sum of the individual subsection discharges.

Sample collection

Water samples from the lake were taken with an integrated sampler. This is a 2 meter PVC pipe (collection tube) with a PVC ball that acts as a water locking mechanism. The contents of the tube were transferred to 100 ml HDPE bottles. Water samples from the inlets were gathered by submerging the bottles just under the water surface.

The sediment samples were collected with an Eckman dredge. Most samples were taken in triplicate. All standard deviations in the tables or error bars represent the replicate measurements for water and sediment samples.

Persulfate Digestion and Malachite Green (Chemical Analysis Protocol)

Reagent 1: 28.4 mmol/L ammonium molybdate tetrahydrate in 3.15 Molar H_2SO_4

Reagent 2: 3.5 g/L polyvinyl alcohol (PVA) and 0.35g/L malachite green

Standard P solutions made by KH_2PO_4 and then successive dilutions

Persulfate solution for digestion: 5% $K_2S_2O_8$ (solubility is 5.2%)

Soil digestion: In 15 ml glass incubation tubes
- 50 mg of dried and ground sediment
- 2 ml persulfate solution
- 0.50 ml of concentrated sulfuric acid
- Autoclaved 1hr. at 121°C

Added 10 ml of DI and shake for 1 min.
Centrifuged and removed 10 ml of liquid for analysis

Water digestion: In 15 ml glass incubation tubes
10 ml of water sample (including all standards, and blank)
1.5 ml persulfate 5%
Autoclaved 30 min
Cooled to near room temp

Malachite green and spectrometric analysis:
Added 2 ml of reagent 1 to digested samples
Capped and mixed
Added 2 ml of reagent 2
Waited 30 min. before measuring absorbance at 625nm
with 1 cm pathlength

Ascorbic Acid (Murphy-Riley)
Modified Ascorbic Acid method from Dick and Tabatabai.[10]
Absorbance measured at 882 nm.

RESULTS AND DISCUSSION

Following digestion, phosphorus concentrations were determined using a uv-vis spectrometer and analyzing the 625 nm absorption peak based on the association of malachite green with phosphomolybdate under acidic conditions. Lower detection limit results from this complex having an absorptivity more than three times greater than the phosphoantimonyl-molybdenum blue in the standard Murphy-Riley method (Table 1). While the malachite green method does provide signal at lower concentrations, the Murphy-Riley method provided greater dynamic range and a more reliable standard curve (Figures 1 and 2). This would indicate that either method can be used, but if some concentrations fall to the ten ppb range, malachite green would be preferred. The malachite green method was used for all the Pewaukee Lake data that is presented here.

The 2003 sample collections were all made from a boat on the lake. The Meadowbrook and Coco Creek sampling was done at the mouth of those streams at the gps coordinates shown in Table 7. The 2004 sampling of interior lake sites was done at the same locations as 2003, but Coco Creek, Meadowbrook, and Zion Creek were sampled at locations upstream from Pewaukee Lake. The only sampling that followed a significant rain event was on June 18, 2004. In the previous two days 0.72 inches of rain had fallen. On this day increased flows and generally higher P concentrations were observed. It would be desirable to have more data after rain events and snow-melts.

The concentration of P in sediment samples taken on July 2, 2003 is shown in Figure 4. The 2003 total P concentrations in the water samples is shown in Figure 5 and Table 3. The 2004 P measurements were accompanied by flow readings for Coco Creek, Meadowbrook, and Zion Creek. For these inlets the P loading and P concentrations are displayed (Figure 7). The last six measurements of Meadowbrook registered no measurable flow, and therefore the P loads are zero. An example of the flow calculations by sectioning is shown in Table 6. This type of table was reproduced for all of the inflow calculations. From the P loading data an idea of relative P loads could be obtained (Figure 5). Although the total P concentrations of Coco Creek were lower, the larger flow made it a greater P contributor. The interior lake and the Pewaukee River P concentrations are shown in Figure 6, and all the P concentrations with standard deviations are in Table 4.

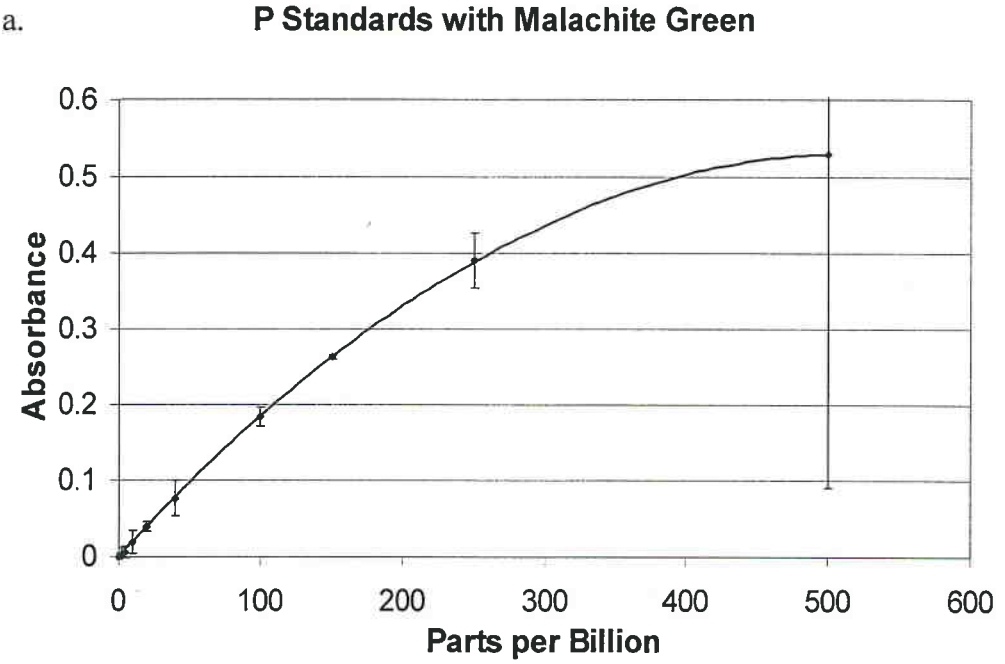
Absorptivity Comparison

	Absorptivity (absorbance units/ppb)	Average Standard Deviation of standards (ppb)
Malachite Green	0.0020	5.0
Ascorbic Acid	0.0006	1.5

Table 1. Comparison of two P analysis methods. The greater absorptivity of the malachite green demonstrates the ability of the technique to show measurable signal at lower concentrations of P. Ascorbic acid did have a better average standard deviation which makes it a better technique for most measurements over 10 ppb.

Comparison of Standard Curves for Ascorbic Acid and Malachite Green

a.



b.

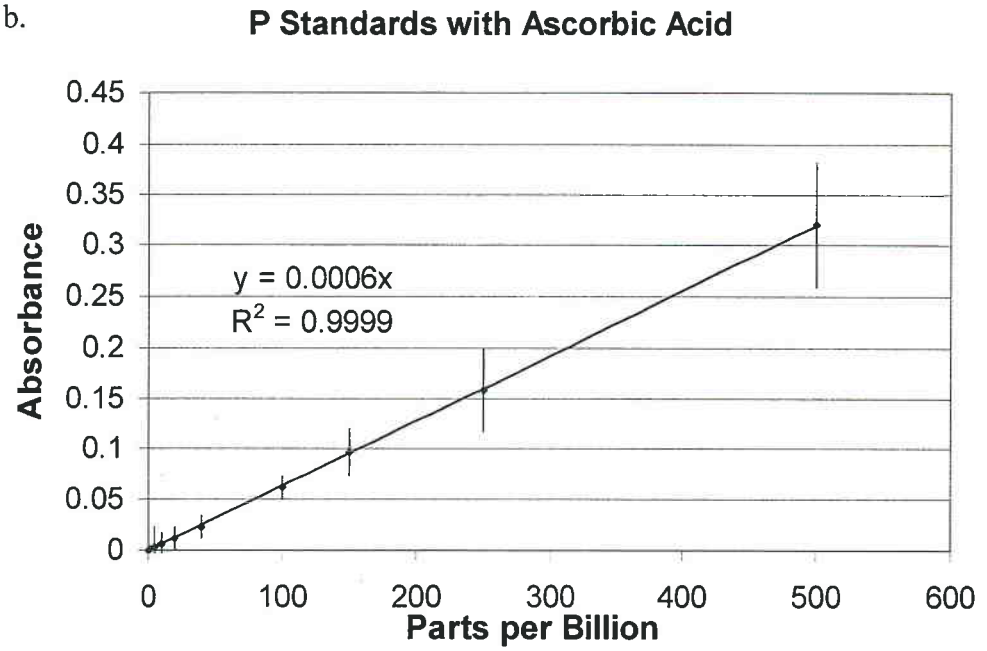
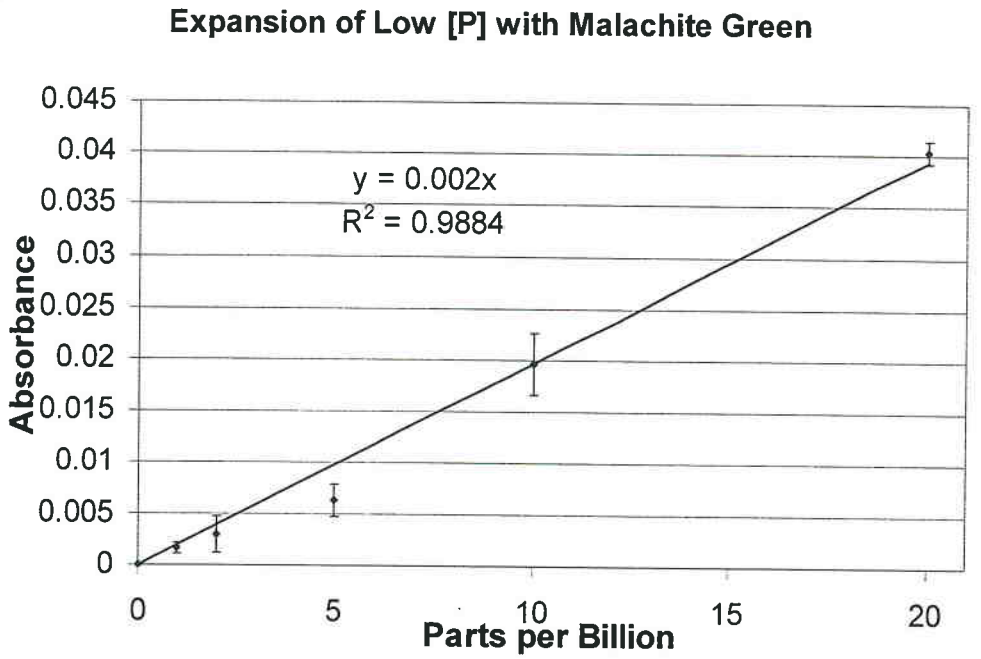


Figure 1. Plot of phosphorus water standards 0, 1, 2, 5, 10, 20, 40, 100, 150, 250, 500 ppb (µg/L) a) With malachite green analysis and a curve fit, the error bars represent +/- 5x the standard deviation and b) with ascorbic acid analysis and a linear fit, the error bars represent +/- 20x the standard deviation in order to make them visible on the graph.

a.



b.

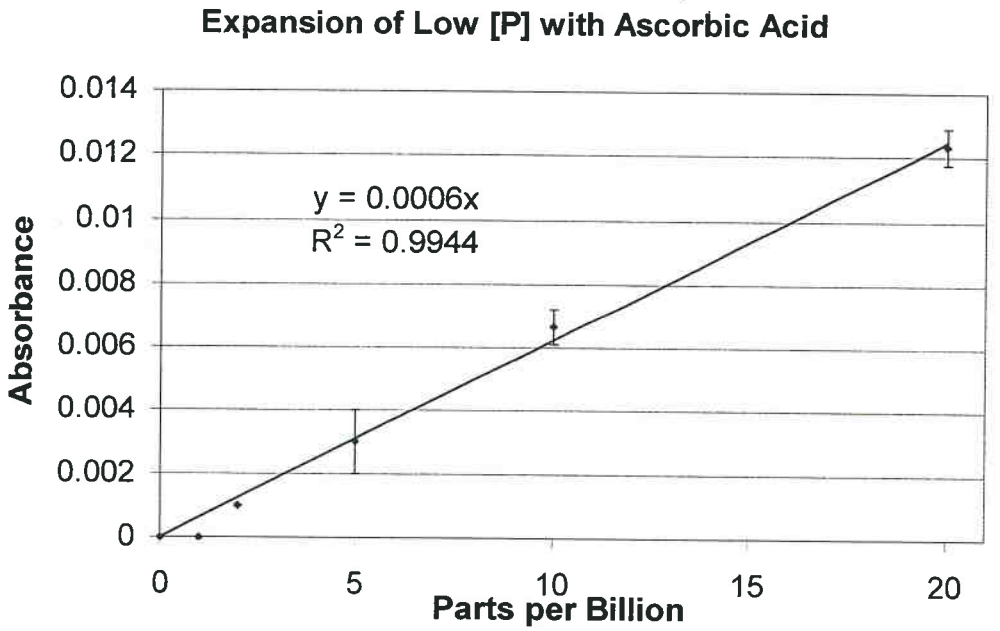


Figure 2. Plot of same phosphorus water standards 0, 1, 2, 5, 10, 20 ppb (μg/L) a) With malachite green analysis and a linear fit b) with ascorbic acid analysis and a linear fit. The error bars represent +/- a single standard deviation for n=3.

Water samples were sent to the University of Wisconsin soils lab for inductively coupled plasma (ICP) analysis. The phosphorus measurements are below a reliable concentration in this ICP analysis. The routine ICP analysis provided concentrations of other elements (Table 2.)

Concentration Values for Elemental Analysis

Sample ID	P ppm	K ppm	Ca ppm	Mg ppm	S ppm	Zn ppm	B ppm	Mn ppm	Fe ppm	Cu ppm	Al ppm	Na ppm
E2 1	0.02	1.96	46.6	38.6	9.15	< 0.001	0.03	0.004	0.015	0.010	<0.02	48.4
E2 2	0.02	2.03	46.5	39.8	9.12	< 0.001	0.02	0.004	0.013	0.013	<0.02	49.8
E2 3	<0.02	2.00	46.6	39.8	9.17	< 0.001	0.02	0.003	0.013	0.006	<0.02	49.7
E2 4	<0.02	1.94	46.7	38.3	9.15	< 0.001	0.02	0.003	0.013	0.009	<0.02	47.7
W2 1	0.07	1.99	47.7	37.4	9.03	< 0.001	0.02	< 0.001	0.002	0.011	<0.02	46.3
W2 2	0.05	2.06	51.4	39.0	11.87	< 0.001	0.02	< 0.001	0.001	0.010	<0.02	48.3
W2 3	<0.02	1.96	48.0	37.2	9.04	< 0.001	0.02	< 0.001	0.002	0.011	<0.02	45.8
W2 4	0.03	1.98	47.9	37.5	9.03	< 0.001	0.02	< 0.001	0.002	0.008	<0.02	46.1

Table 2. June 12, 2003 Analysis of 4 replicates of water samples from the East and West locations, performed by UW soil lab ICP.

In order to establish confidence in the method and P concentration data, eight samples were sent to the State Lab of Hygiene for comparison (Figure 3).

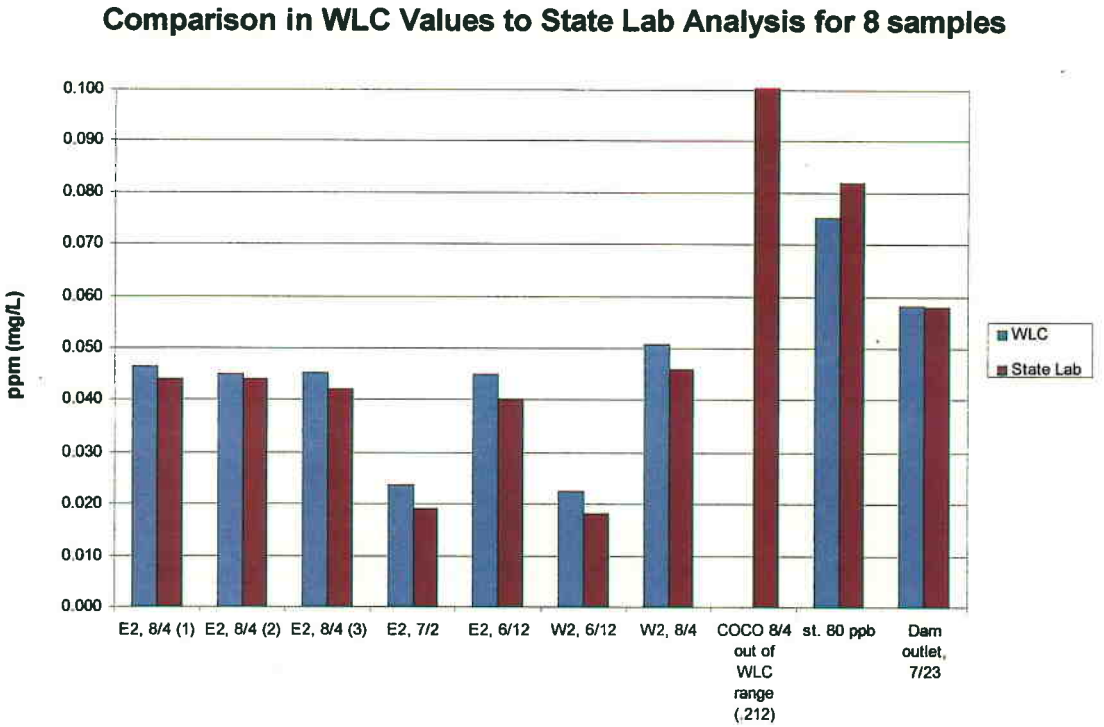


Figure 3. State Lab of Hygiene (Murphy Riley) comparison to WLC (Mal. Green).

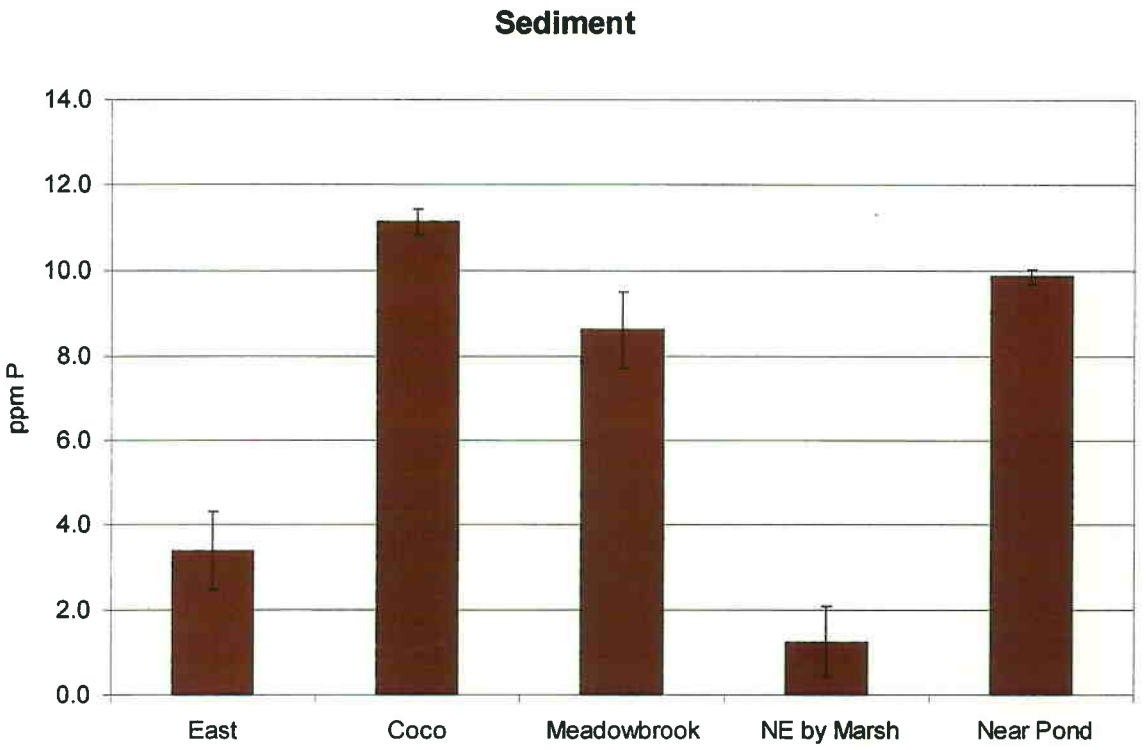


Figure 4. Total P concentrations in sediment 2003.

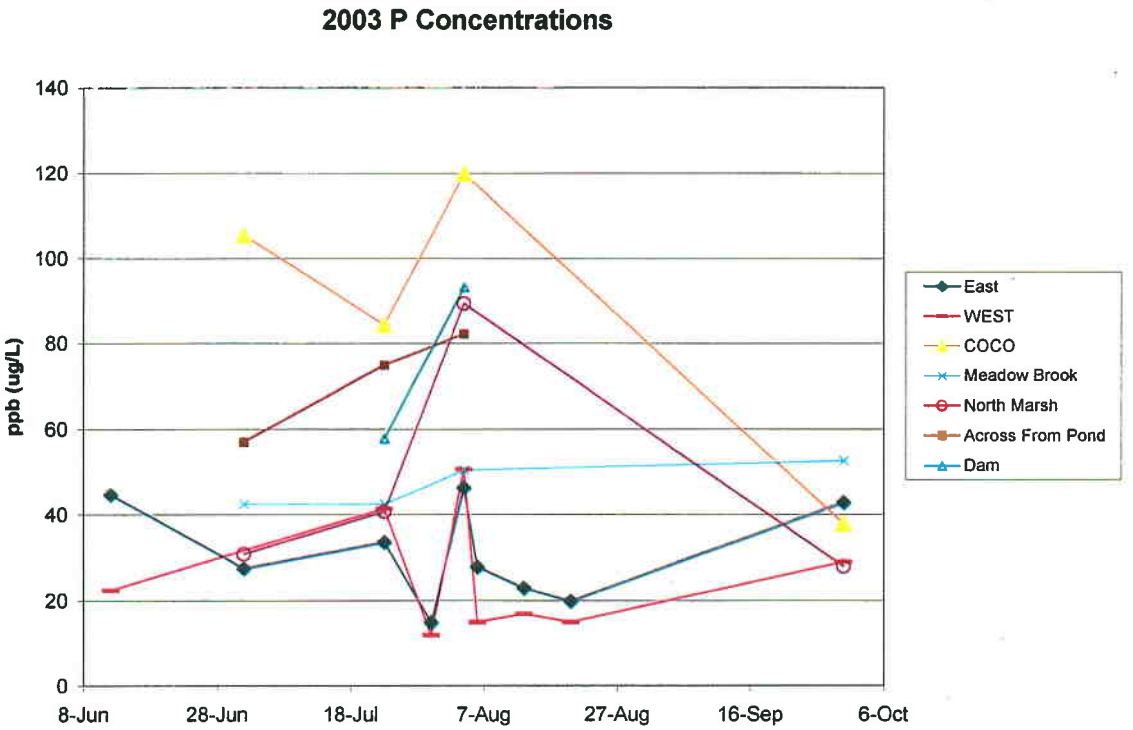


Figure 5. June 12 to September 30 phosphorus concentrations 2003.

2003 P Concentrations

LOC.	12-Jun	±	2-Jul	±	23-Jul	±	4-Aug	±	30-Jul	6-Aug	13-Aug	20-Aug	30-Sep
East	45	6	28	6	34		46	5	15	28	23	20	43
							45	3					
							45	2					
West	22	3			41		51	1	12	15	17	15	29
COCO			106	16	85		out of range						38
Meadow brook			43	3	43		51	15					53
North end By Marsh			31	4	41		90	20					28
Across from pond			57	5	75		82	5					
Outlet at the Dam					58	2	93	6					

Table 3. June 12 to September 30 phosphorus concentrations 2003 in µg/L.

2004 Interior Lake P Concentrations

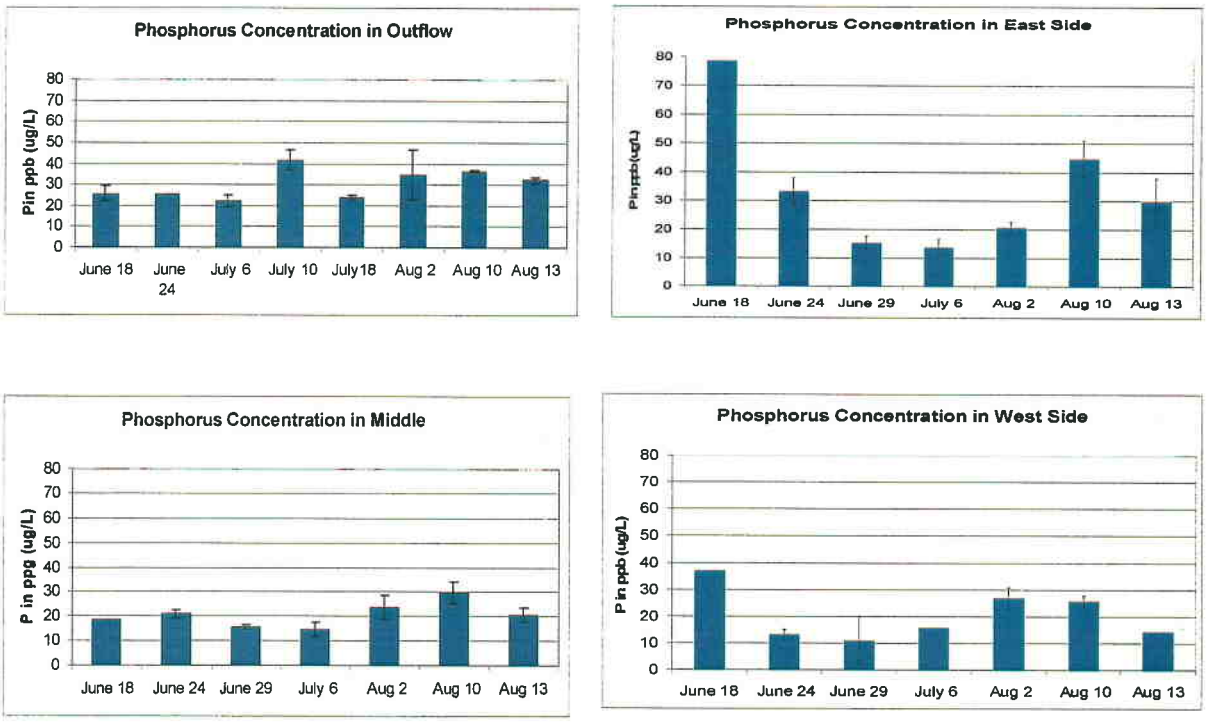


Figure 6. Concentrations of P in the lake and the outflow of the Pewaukee River.

P Loading and Concentrations in Inlets

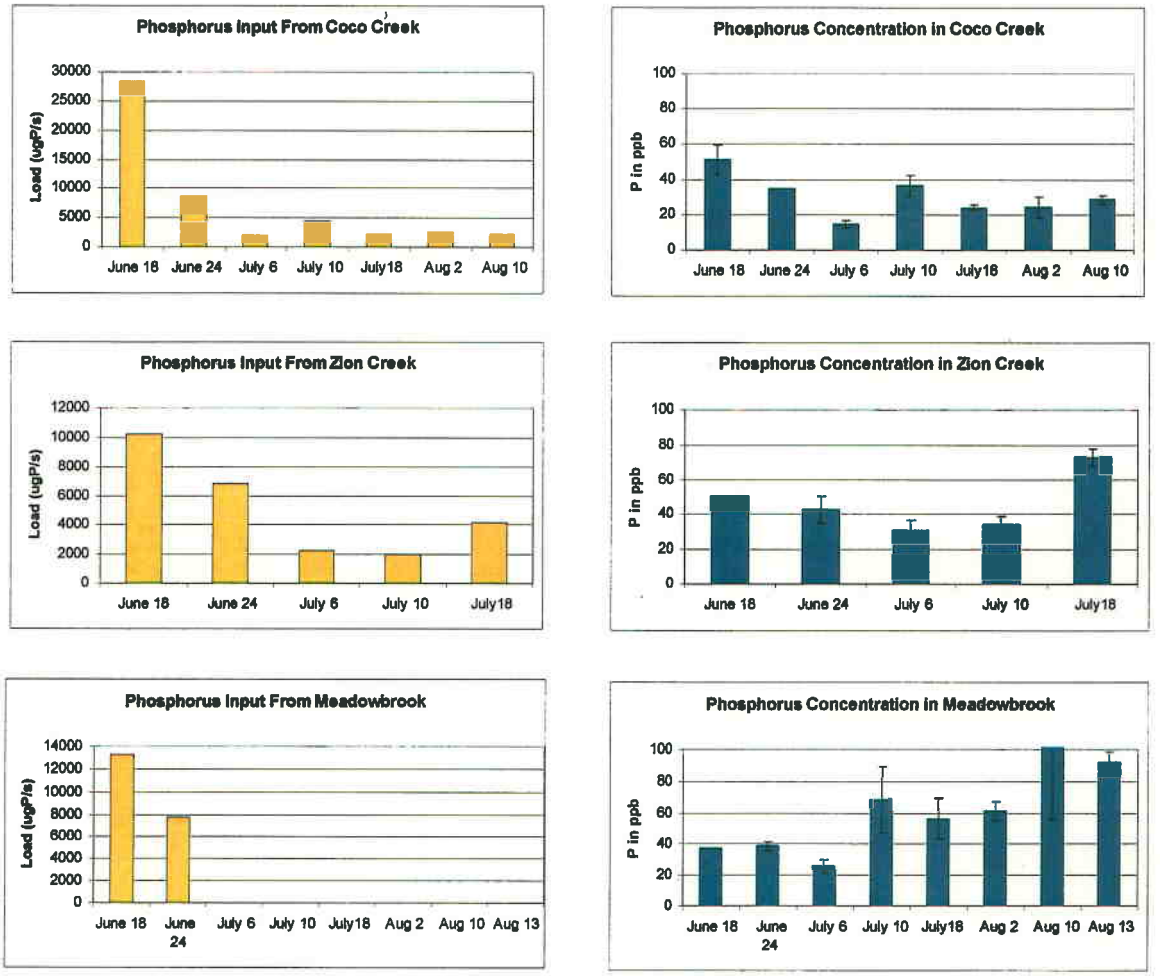


Figure 7. Phosphorus loads and concentrations from main inflows in 2004.

	Pew. R.		Coco Creek		Meadowbrook		Zion Creek		East Lake		Middle Lake		West Lake	
2004	ppb P	st dev	ppb P	stdev	ppb P	st dev	ppb P	stdev	ppb P	stdev	ppb P	stdev	ppb P	stdev
18-Jun	25.80	3.56	51.12	8.23	37.50		50.00		78.75		18.75		36.88	
24-Jun	25.48		34.60		38.85	2.89	42.57	7.35	33.35	4.54	21.08	1.78	13.06	2.00
29-Jun									15.26	2.45	15.73	0.89	11.01	8.90
6-Jul	22.13	2.85	14.94	2.00	25.90	4.41	30.62	6.05	13.74	2.92	14.58	2.82	15.73	
10-Jul	41.65	4.64	36.67	5.67	68.18	21.02	33.72	5.47						
18-Jul	23.91	1.33	23.91	1.44	56.39	12.46	72.79	5.31						
2-Aug	34.64	11.81	24.51	5.75	60.99	5.91			20.82	1.78	23.49	5.08	26.90	3.92
10Aug	36.49	0.55	28.56	2.09	103.4	47.08			44.78	6.38	29.48	4.50	25.80	2.09
13Aug	32.06	1.46			91.77	7.04			29.79	7.96	20.63	2.86	14.37	

Table 4. Phosphorus concentration measurements for 2004 in ug/L (ppb).

	Coco Creek	Meadowbrook	Zion Creek
Average Flow (cu ft/s)	6.3	2.4	3.9
Average [P] ppb	30.6	60.4	45.9
Average load (ugP/s)	7253	2628	5090
% contribution	48.4%	17.6%	34.0%

Table 5. Relative phosphorus contributions.

		cu ft/sec	ug/L		ugP/s	
Date	Location	total flow	P conc	stdev	P load	
6/18/2004	Coco	19.52	51.12	8.23	28260	
section	rev	seconds	rev/sec	depth ft	width ft	cu ft/sec
a	40	46	1.92	1	2.5	4.79
b	40	45	1.96	1.1	2	4.31
c	40	46	1.92	1.25	2	4.79
d	40	45	1.96	1.15	2.5	5.63

Table 6. Example of P load calculation spreadsheet.

Sampling locations

(E) East	43° 04.887 N 88° 17.132 W
(W)West	43° 04.136 N 88° 19.199 W
(C) Coco 2003	43° 05.241 N 88° 17.116 W
(Mb)Meadow brook 2003	43° 03.658 N 88° 18.239 W
(NM)North by marsh	43° 05.241 N 88° 18.057 W
(P)Across from Pond	43° 04.247 N 88° 17.699 W
(D) Dam	Just on lake side

Table 7. GPS coordinates for 2003 sampling locations

A water quality index (Table 8) and trophic classification table (Table 9) provide context for phosphorus concentration values in Pewaukee Lake. A combination of parameters such as Secchi disc readings, total phosphorus concentrations, and chlorophyll-α concentrations can provide stronger evidence for the trophic classification.[11]

The average *in-lake* P concentration measurements in 2003 was 28 μg/L, and the values ranged between 12 and 51. In 2004 the average was 25 μg/L, with a range from 14 to 79. These values place it at average for "natural lakes" and in the category of mesotrophic (according to the P criteria in figures 8 and 9).

Water Quality Index	Total Phosphorus (µg/l)
VeryPoor	150
	130
Poor	90
	70←Average for impoundments
	50
Fair	
	40
	30
Good	
	25←Average for natural lakes
	20
	10
Very Good	
Excellent	

Table 8: Total Phosphorus concentrations for Wisconsin's Natural Lakes and Impoundments.[12,13]

Trophic class	Total Phosphorus µg/l	Chlorophyll a µg/l	Secchi Disc ft.
Oligotropic	3	2	12
	10	5	8
Mesotrophic	18	8	6
	27	10	6
Eutrophic	30	11	5
	50	15	4

Table 5: Trophic classification of Wisconsin lakes based on Chlorophyll a, water clarity measurements, and total phosphorus values. Only total phosphorus is presented in this report.[12,13]

The measurements of inflows that were taken in 2004 are not compared to the same standards as that of lakes or impoundments. The USEPA describes a method for identifying appropriate nutrient criteria for rivers and streams based on data collected from lakes within an ecoregion. Pewaukee Lake falls into ecoregion VII, which is "mostly glaciated dairy region including all or parts of the states of New York, Pennsylvania, Michigan, Wisconsin, Minnesota, Ohio, Indiana, Illinois, and Iowa."[14] Within each ecoregion, reference streams may be identified to represent the least culturally impacted waters. Monitoring data from these streams are then used to identify recommended nutrient criteria. The criterion for a particular parameter (e.g., phosphorus) is calculated as the 75th percentile of the values measured in the reference lakes. When reference streams are not identified, the second method is to determine the lower 25th percentile (P25) of the population of all streams within a region. Data analyses to date indicate that the lower 25th percentile from an entire population roughly approximates the 75th percentile for a reference population and are assumed to represent minimally impacted conditions and be protective of designated uses.[14]

The 25th percentile data from ecoregion VII are reported as an aggregate of all locations throughout the region, and then it is also reported for a smaller zone (53) that is essentially the southeastern quadrant of Wisconsin. For the entire ecoregion VII, the total phosphorus P25 is 33.0 µg/L (ppb) based on a population of 910 streams. For southeastern Wisconsin, the total phosphorus P25 is 80 µg/L (ppb) based on a population of 136 streams.[14] The 2004 P concentration measurements of Pewaukee Lake inlets averaged 46 µg/L, with a range of 15 to 103. These values give some context as to the quality of the inflows into Pewaukee Lake. When combined with flow rates, this P monitoring provides a relative picture of the sources of P in the lake from each of the inlets.

Literature Cited

[1] Schindler D.W. 1977. Evolution of Phosphorus Limitation in Lakes. *Science*, 195, 260-262.

[2] Ambio 1990. Special issue. *Marine Eutrophication*, 19, 102-176.

[3] Ebeling, A. M. et al. 2002. Dairy Diet Phosphorus Effects on Phosphorus Losses in Runoff from Land-Applied Manure. *Soil Sci. Soc. Am. J.* Vol. 66, 284-291.

[4] Auqa Tech, Inc. 1972. Limnological Survey of Pewaukee Lake for the Determination of Water Quality.

[5] Southeastern Wisconsin Regional Planning Commission. 1984. A Water Quality Management Plan for Pewaukee Lake Waukesha County, Wiscsonsin, Community Planning Assessment Report No. 58, Madison, WI.

[6] Murphy, J., and J.R. Riley. 1962. A modified single solution method for the determination of phosphate in natural waters. *Anal. Chem.* 27:37-36.

[7] Ohno, T., and Zibilske, L.W. 1977. An alkaline oxidation method for determination of total phosphorus in soils. *Soil Sci. Soc. Am. J.* 41:511-514.

[8] SSA Book Series: 5. 1996. Methods of Soil Analysis. Editor: Bigham J.M. *Soil Sci. Society of America*, Inc. Madison, WI.

[9] Nelson, N. S. 1987. An acid-Persulfate digestion procedure for determination of phosphorus in sediments. *Commun. In Soil Sci. Plant Anal.* V. 18 no.4 p. 359-69.

[10] Dick, W.A., and M.A. Tabatabai. 1977. An alkaline oxidation method for determination of total phosphorus in soils. *Soil Sci. Soc. Am. J.* 41:511-514.

[11] Koch, P. and Anderson. R.C. 2003 Southeast Wisconsin's Pewaukee Lake Biological Evaluation 2002. Wisconsin Lutheran College, Biology Dept. Technical Bulletin.

[12] Lillie, R. A. and J. W. Mason. 1983. Limnological Characteristics of Wisconsin Lakes. WI Dept. of Natural Resources Tech. Bulletin. 138, Madison.

[13] Shaw, Byron, Christine Mechenich, and Lowell Klessig. 2002. Understanding Lake Data. University of Wisconsin Cooperative Extension Publishing, Madison, Wisconsin. (G3582) SR-02/2002-1M-525 http://cecommerce.uwex.edu/pdfs/G3582.PDF

[14] USEPA. Ambient Water Quality Criteria Recommendations: Information Supporting the Development of State and Tribal Nutrient Criteria. Lakes and Reservoirs in Nutrient Ecoregion VII. EPA 822-B-00-018. U.S. Environmental Protection Agency, Office of Water, Office of Science and Technology, Washington, DC. 2000.