

# Phosphorus Fractionation in Sediment Cores Collected In 2005 Before and After Onset of an *Aphanizomenon flos-aquae* Bloom in Upper Klamath Lake, OR, USA

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**Abstract** We tested the hypothesis that there would be measurable losses of phosphorus (P) from surficial sediments of Upper Klamath Lake (UKL), Oregon, if sediments were a source of P during an algal bloom. We compared concentrations of total and forms of P at various depths in cores collected before and after the onset of a large *Aphanizomenon flos-aquae* bloom. Concentrations of inorganic P were determined in extraction solutions of MgCl<sub>2</sub> (1 M, pH 8), citrate-dithionite-bicarbonate, and 1 M HCl. Sediments below 2 cm were dominated by residual P which is defined as total P minus inorganic P. During the study period, data from the top 2-cm of sediment indicated (a) significant decrease in total P concentration, primarily associated with iron oxyhydroxides at one site, and (b) significant increase in total P concentration associated with residual P at a second

site. Data from two other sites indicated no net changes in concentrations of total P.

**Keywords** Phosphorus fractionation · Residual phosphorus · Cyanophyte · Eutrophic · Shallow lake · Metals

## 1 Introduction

The pollutant target in the total minimum daily load (TMDL) for Upper Klamath Lake is the load of total phosphorus (P) to the lake. The Oregon Department of Environmental Quality has determined that reduction in the load of total P to UKL will decrease the algal blooms which in turn will decrease pH and increase median concentrations of dissolved oxygen (DO) in the water column (Oregon Department of Environmental Quality 2002).

A recent National Research Council report found that large-scale adult mortality of two endangered sucker species in Upper Klamath Lake (UKL), in south-central Oregon (Fig. 1) was attributable to large zones of lake water where DO concentrations were low. To avoid hypoxic conditions, the TMDL for UKL requires that the concentration of DO in the lake be  $\geq 4$  ppm (Oregon Department of Environmental Quality 2002).

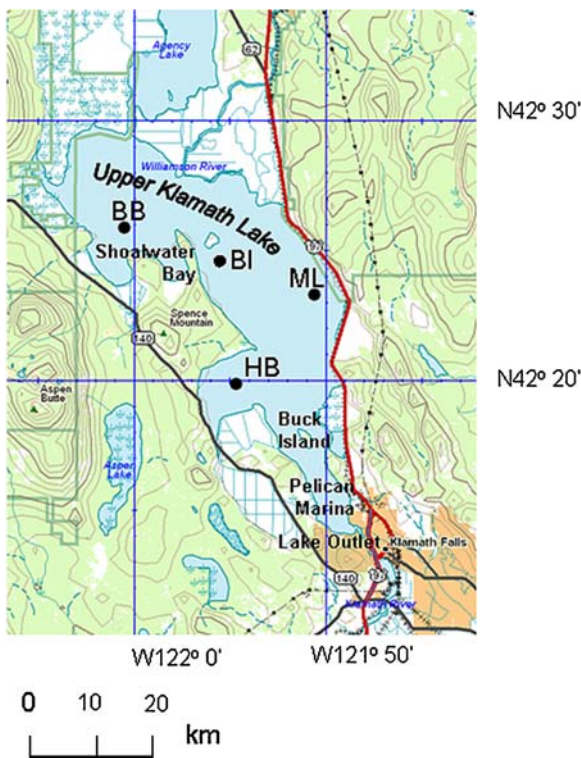
Low concentrations of oxygen in the water column occur after large blooms of *Aphanizomenon flos-aquae* (AFA) senesce (National Research Council

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**Fig. 1** Map of Upper Klamath Lake, Oregon. Sampling sites include *BB* Ball Bay, *HB* Howard Bay, *BI* Bare Island, *ML* Mid Lake. Sites were sampled on 29 April and 7 July 2005

2004). AFA is now the monoculture algae in UKL (Wood et al. 2006). Large blooms of AFA are attributed to elevated concentrations of total P in UKL (Oregon Department of Environmental Quality 2002). AFA is nitrogen (N)-fixing cyanophyte, and its growth is considered independent of nutrient forms of N. In the period of 2002 to 2005, DO concentrations in UKL were commonly less than 4 ppm at deeper sites, in the lower part of the water column, and for short periods during the day (Wood et al. 2006; Hoilman et al. 2008). In 2003, widespread low DO concentrations throughout the water column which persisted through the whole day preceded a low DO event which resulted in a large fish die-off (Wood et al. 2006). The low oxygen event occurred when elevated chlorophyll *a* concentrations began to decrease indicating that the algal population was declining rapidly. The purpose of the TMDL for UKL is to decrease growth of AFA and increased median concentrations of oxygen by decreasing loads of total P to the lake.

Upper Klamath Lake is a large, shallow lake with an average depth of 2.8 m full pool (Wood et al.

2006). Records indicate that the lake was eutrophic when European settlers arrived in the mid-19th century (Colman et al. 2004b). Settlement activities including the draining of marshes, farming, cattle ranching, and lumber harvesting appear to have contributed to nutrient enrichment (pollution) of the lake (Bradbury et al. 2004b). An increase in external loading of P has supported a noticeable increase in the intensity of AFA blooms in UKL in recent decades (Bradbury et al. 2004b; Colman et al. 2004b).

The internal load of P to UKL is larger than the external load and calculated to be approximately two thirds of the yearly average total load of P to UKL (Oregon Department of Environmental Quality 2002). Internal loading of P can occur during both anaerobic and aerobic conditions. When sediment conditions are anaerobic, the dominant mechanism for release of P to the water column is understood to be dissolution of poorly crystalline iron (Fe) oxides and release of sorbed P (Mortimer, 1972). It also has been suggested that P can be added to the water column when sediment conditions are anaerobic by (a) microbial cells at the surface of the sediment releasing P (Brunberg and Boström 1992) or (b) microbial cells at the surface of the sediment releasing polyphosphate (Hupfer and Rube 2004). Mechanisms for the release of P from the sediment during aerobic conditions include resuspension of surface material (Laenen and LeTourneau 1996; Lennox 1984), dissolution of P minerals by bacterial organic acids which also sequester metallic ions (Harrison et al. 1972), bioturbation, and consumption and digestion of surficial organic matter by benthic organisms including tubificid worms and chironomid larvae (Andersson et al. 1988; Fukuhara and Sakamoto 1987). The importance of each of these factors on the internal loading of P in a lake is related to the redox cycle in the lake. For the period of this study (April to July 2005), DO concentrations in the water column were reported to be  $\geq 9$  ppm (Hoilman et al. 2008). If aerobic conditions existed at the sediment–water interface during the period of this study, internal loading of P from bottom sediment would be expected from biological and physical processes rather than redox reactions.

The goal of this study was to determine whether temporal differences in geochemical phases of P in UKL sediments indicate internal loading of P from the sediment column during an algal bloom. Rapid,

large increases in water column concentrations of total P during large AFA bloom were observed in 2002–2004 (Wood et al. 2006). In 2005, increases in water column concentration of total P ranged from 0.2 to 0.8 mg L<sup>-1</sup> (Hoilman et al. 2008) during the AFA bloom observed during this study. Four sites were sampled before and after the onset and development of the bloom. Cores were analyzed for total P, P associated with geochemical phases, metals known to bind with phosphate, and organic carbon (C) and nitrogen (N). If the hypothesis is correct, there should be a net change (decrease) in the concentrations of total P in bottom sediments during a period of bloom development.

## 2 Methods

### 2.1 Study Sites

On April 29, 2005, and July 7, 2005, cores were collected manually by divers using self-contained underwater breathing apparatus (SCUBA) at sites located at Ball Bay (BB), Bare Island (BI), Howard Bay (HB), and Mid Lake (ML) in UKL (Fig. 1). There was no apparent shortening (compaction) of core material during the manual collection process. Replicate cores were collected approximately 10 m apart at each site in both April and July 2005. The cores were 30 and 10 cm in length. The cores provided replicate sediment material from the interface between the sediment and the water column to a depth of 10 cm beneath the sediment surface. Processes in sediment between sediment–water interface and a depth 10 cm below the sediment–water interface will have more effect on water quality during a 10-week study than processes at depths greater than 10 cm. Sediment at depths greater than 10 cm provide information about the depositional history of P in the lake.

### 2.2 Sediment Geochemistry

Colman et al. (2004a) reported an average sedimentation rate of 0.172 cm year<sup>-1</sup> for sediments in UKL. Using this rate, the time required to accumulate 1 cm of sediment would be approximately 6 years. To quantify processes in which surface sediment and the overlying water column interact, cores were sectioned at 1 cm intervals near the sediment–water interface

(<5 cm). Two-centimeter intervals were collected between depths of 5 and 11 cm; 3-cm intervals were collected between depths of 11 and 20 cm; 4-cm intervals were collected between depths of 20 cm and the bottom of the core. There were 14 samples from each 30-cm core and eight samples from each 10-cm core. Samples were shipped on ice on the day of collection for overnight delivery to the laboratory.

Samples were weighed, frozen, and freeze-dried. The most labile form of P, exchangeable P, was shown by Andrieux and Aminot (1996) to be stable during the freeze-drying process. In preparation of sediment samples, dry-sieving is considered preferable to wet sieving to prevent leaching of exchangeable P by the sieving water (Andrieux and Aminot 1996). Each sample of freeze-dried sediment was ground with mortar and pestle until it passed through a 60-mesh (250 µm) sieve.

Samples were digested in concentrated nitric and hydrofluoric acids using microwave digestion equipment (Simon et al. 2005). Standard reference material 2710 was run with every set of digest sediments. If the concentration of total P in the digest of SRM 2710 did not fall within the range reported by the certificate of analysis, the sample set was discarded. Digests were diluted 1:20 with 0.5% nitric acid and analyzed for aluminum (Al), calcium (Ca), and Fe with a Perkin-Elmer Model 5100 ICP-AES. Total P in digests was analyzed using the molybdenum blue method (Rand et al. 1976).

Although originally developed for use with marine sediments, the Ruttenberg method (SEDEX; Ruttenberg 1992) has been used for sequential extraction of P from freshwater sediments (Ruban et al. 1999; Brunberg et al. 2002; Filippelli et al. 2006) and from freshwater and estuarine sediments (Andrieux-Loyer et al. 2008). In this method, phosphorus is fractionated into (1) loosely sorbed P and interstitial water P using a MgCl<sub>2</sub> (1 M, pH 8), (2) easily reducible Fe-bound P using citrate dithionite bicarbonate solution (pH 7.6), and P associated with carbonates and acid-soluble minerals using 1 N HCl. The loosely sorbed P and the P associated with poorly crystalline Fe-oxyhydroxides are operationally defined as bioavailable (Ruban et al. 1999).

For this study, 0.1 g sediment samples were extracted using the SEDEX method (Ruttenberg 1992) with the modifications of Anderson and Delaney (1999). The sodium acetate step was omitted, and the extraction time for the CDB extraction was

increased to facilitate the dissolution of poorly crystalline Fe-oxyhydroxides. Dissolved reactive P (DRP) concentrations in all fractions were determined by the molybdenum blue method (Rand et al. 1976). Colorimetric P analysis of CDB extracts was done without butanol extraction because concentrations of P in the CDB extracts were within detection limits of the colorimetric method.

Residual P concentrations were calculated by subtracting the sum of the concentrations of P recovered in the sequential extraction procedure (inorganic P) from the concentrations of total P. Concentrations of Fe were determined in CDB fractions by ICP-AES.

Freeze-dried sediments, collected in July 2006 at the ML site from intervals 0–1, 1–2, and 2–3 cm were extracted with 20 ml of 0.25 M NaOH–0.05 M EDTA (Cade-Menun and Preston 1996) for 16 h at a temperature of approximately 22°C. Samples were centrifuged, pHs of supernatants were adjusted to approximately 7 (Cade-Menun et al. 2006), and 50 µg of P as methylene diphosphonic acid (MDPA) were added before freezing and lyophilization. Dried samples were transferred to 2-cc centrifuge tubes and dissolved in a combination of 0.675 ml of 0.25 M NaOH–0.05 M EDTA and 0.2-ml of 10 M NaOH in D<sub>2</sub>O (99.9% D<sub>2</sub>O, Sigma-Aldrich) immediately before analysis. The solution was mixed for ~2 min using a vortex shaker, centrifuged and pipetted into 5 mm NMR tubes. The <sup>31</sup>P NMR measurements were carried out on a Bruker Avance, 600 MHz spectrometer, equipped with a 5-mm broad band tunable probe operating at 242.9 MHz frequency for <sup>31</sup>P. The proton decoupled spectra were acquired with a 30° pulse width (3.25 µs), acquisition time 1.35 s, relaxation delay 1 s, and sweep width 24,350 Hz (100 ppm). The sample temperature was maintained at 293°K throughout the acquisition of 8,000 to 25,600 pulses, as required to achieve acceptable signal to noise ratio. The 64 K data point fid was processed with 3 Hz line broadening and transformed to produce a 32-K data point spectrum. The chemical shifts were indirectly referenced to external 85% H<sub>3</sub>PO<sub>4</sub> (at δ=0.0) via the lock signal. Spectral processing was performed with Bruker-BioSpin's TopSpin 1.3 software.

The loss of organic P during the sequential extraction procedure for inorganic P was estimated using sediments collected in July 2006 at the ML site from intervals 0–1, 1–2, and 2–3. These samples were sequentially extracted with MgCl<sub>2</sub> (1 M, pH 8), CDB,

and 1 M HCl using the procedure described above. The sediment remaining after extraction with 1 M HCl was extracted with 0.25 M NaOH–0.05 M EDTA as described in the previous paragraph. Preparation of the supernatants from the NaOH–EDTA extractions and the operating conditions for obtaining <sup>31</sup>P NMR spectra were the same for both samples not subjected to sequential extraction, and for samples that were subjected to sequential extraction.

Peak assignments in the <sup>31</sup>P spectra that were obtained were made using chemical shift data reported by Koopmans et al. (2003) and Turner et al. (2003) and from <sup>31</sup>P NMR experiments in our lab. Integrated peak areas were compared with the integral value of MDPA to calculate concentrations of P compounds. Peak areas were calculated and compared with the integrated area (set at 1.0) for the internal standard MDPA peak at approximately 18.3 ppm.

Sediments were analyzed for organic C and N using 20 mg of freeze-dried, ground samples. Flash oxidation of each sample was followed by separation of the gaseous products using a Carlo Erba EA 1108 Elemental Analyzer. Two aliquots of sediment samples from the ML site were analyzed; one aliquot was analyzed without treatment with HCl to determine the total C concentration; a second aliquot was treated with HCl vapors to remove carbonates before analysis to determine the concentration of organic C. This experiment was done to test the statement in Bradbury et al. (2004a) that UKL sediments are carbonate free. We used sediment from the ML site where concentrations of Ca in the sediment were determined to be the larger than in sediment from other sites sampled in this study.

### 2.3 Statistical Analysis

Paired Student's *t* tests were used to compare concentration data from 0–1 and 1–2 cm intervals in the 30- and 10-cm cores collected in April with concentration data for the same intervals in cores collected in July, using a significance level of *P*<0.05.

## 3 Results

### 3.1 Study Sites

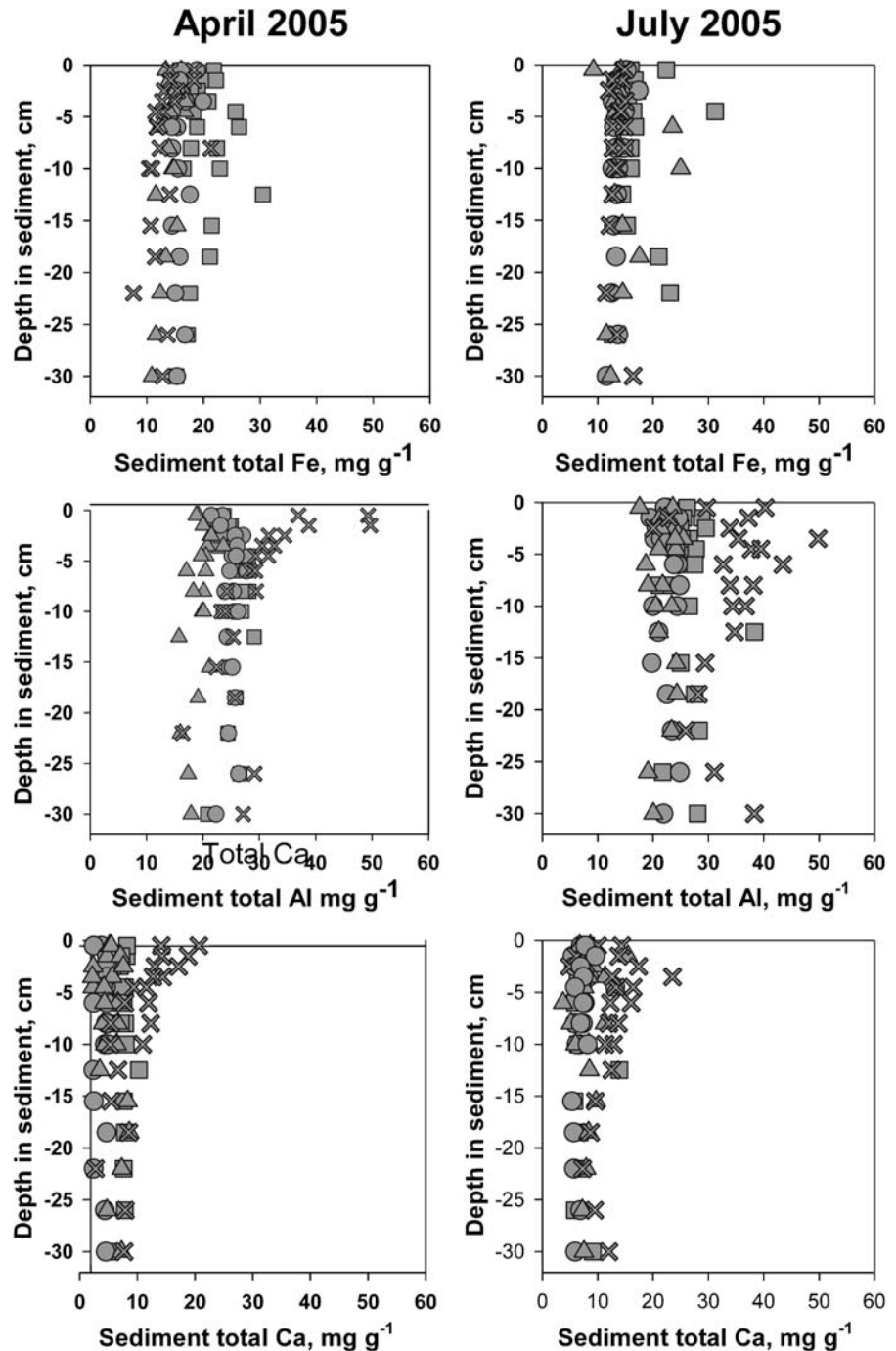
All sites in UKL for which concentrations of chlorophyll *a* are available between April 29 and July

7, 2005 indicated a first maximum in chlorophyll *a* concentration during, or before, the week of July 11–15, 2005 (Hoilman et al. 2008). Chlorophyll data indicate that because collection dates for the cores were 29 April and 7 July, the sediment cores were

collected before and after the onset of the first bloom of AFA in UKL in 2005.

Water column depths on 29 April 2005 were 3.0, 3.3, 1.8, and 4.2 m for the sites BB, BI, HB, and ML, respectively. The water column depths at all sites in

**Fig. 2** Concentrations of total Fe, total Al, and total Ca in sediment cores from Upper Klamath Lake collected 29 April and 7 July 2005. Concentration units are  $\text{mg g}^{-1}$  DW sediment. Symbols for study sites:  $\square$  Ball Bay,  $\triangle$  Howard Bay,  $\circ$  Bare Island,  $\times$  Mid Lake



July were approximately 0.2 m lower than measured in April.

### 3.2 Sediment Geochemistry

Chemical data for all samples are presented in Kuwabara et al. (2007). The average water content of these samples was  $90 \pm 2\%$ .

Concentrations of total Fe, total Al, and total Ca were similar in sediments from all cores at all depths (Fig. 2) with the following exceptions. Several samples from the sediment cores collected at the Ball Bay and Howard Bay sites had concentrations of total Fe that larger than the concentrations of total Fe in cores from the other two sites. Concentrations of total Al and total Ca in the top 5 cm of sediment were larger in cores from ML than in the cores from other study sites.

The quantity of total Fe determined to be poorly crystalline Fe-oxyhydroxides ranged from 0.4% to 11%; the median value was  $3.0 \pm 2.0\%$ . In sediment cores collected at the same site, 85% of the intervals in cores collected in April contained larger concentrations of poorly crystalline Fe-oxyhydroxides than matching intervals in July cores.

Sediment concentrations of total P ranged from 0.23 to 1.11  $\text{mg g}^{-1}$  dry weight (DW) and 0.24 to 1.07  $\text{mg g}^{-1}$  DW in April and July, respectively (1 and 2 of Fig. 3).

Concentrations of P in operationally defined sediment phases are shown in Figs. 3–a and 3–b. Approximately 3–15% of total P in the surface sediment was loosely sorbed P, extractable with  $\text{MgCl}_2$  (1 M, pH 8). At depths  $>2$  cm, concentrations of loosely sorbed P were small, with little variation. The largest percentage (15%) of loosely sorbed P was observed in surface sediment collected at the BI site in July.

The sum of the concentrations of  $\text{MgCl}_2$  (1 M, pH 8) extractable P, CDB-extractable P, and P extracted with 1 M HCl is defined as the concentration of inorganic P. Inorganic fractions of P were dominant in the top 1–2 cm of sediment in cores from BB, BI, and HB. Concentrations of poorly crystalline Fe-oxyhydroxides in these cores were largest near the sediment–water interface. The largest contributor to the inorganic fraction is CDB-extractable P. The sum of  $\text{MgCl}_2$  (1 M, pH 8) extractable P and CDB-extractable P, the most bioavailable forms of extractable P (Ruban et al. 1999), ranged from 10% to 60% of total P concentrations in surface ( $<2$  cm) sediment

**Fig. 3 a** Cores from the Ball Bay and Howard Bay sites in UKL collected on 29 April and 7 July 2005. Concentrations of total P and sequentially extracted P are  $\text{mg g}^{-1}$  DW sediment. The symbol for each fraction of P extracted follows the description of the extraction solution; (1)  $\text{MgCl}_2$  (1 M, pH 8) (loosely sorbed phosphate)  $\blacksquare$ , (2) dithionite citrate bicarbonate (phosphate associated with poorly crystalline Fe-oxyhydroxides),  $\square$  and (3) 1 M HCl (phosphate associated with carbonates and P minerals),  $\boxtimes$ . (4) Residual P: The difference between the concentration of total P and the sum of extractable phosphate is defined as residual P  $\boxdot$ . **b** Cores from the Bare Island and Mid Lake sites in UKL collected on 29 April and 7 July 2005. Concentrations of total P and sequentially extracted P are  $\text{mg g}^{-1}$  DW sediment. The symbol for each fraction of P extracted follows the description of the extraction solution; (1)  $\text{MgCl}_2$  (1 M, pH 8; loosely sorbed phosphate)  $\blacksquare$ , (2) dithionite citrate bicarbonate (phosphate associated with poorly crystalline Fe-oxyhydroxides),  $\square$  and (3) 1 M HCl (phosphate associated with carbonates and P minerals),  $\boxtimes$ . (4) Residual P: The difference between the concentration of total P and the sum of extractable phosphate is defined as residual P  $\boxdot$

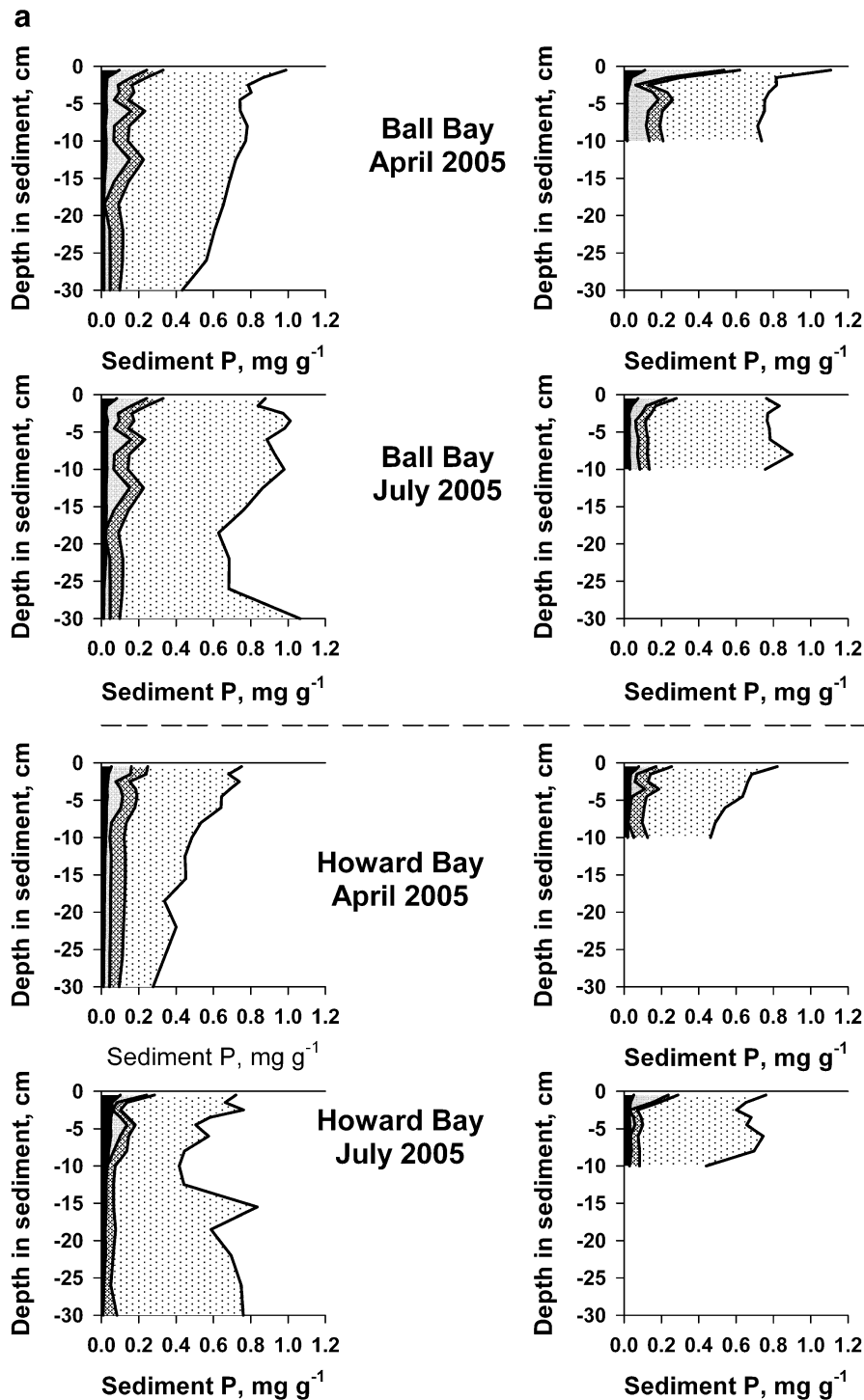
from sampling locations at BB, BI, and HB but was less than 10% of total P concentrations in surface sediment from the ML site.

Approximately 35–50% of the P in surface sediment at the BI site was bound with poorly crystalline Fe-oxyhydroxides in both April and July. Concentrations of P sorbed to poorly crystalline Fe-oxyhydroxides decrease sharply at depths  $>2$  cm. Concentrations of P bound to poorly crystalline Fe-oxyhydroxides represented 15–50% and 10–25% of total P concentrations in surface sediment from the BB and HB sites, respectively. Although Fe was extracted from ML sediment using the CDB solution, only the 1-cm interval sample in July contained P in this fraction.

One molar HCl extracted approximately 10–20% of total P in the April and July cores collected at the BB, BI, and HB sites. One molar HCl extracted approximately 10–30% of the total P in both April and July cores collected at the ML site.

Concentrations of residual P in sediment samples tended to increase between onset and development of the AFA bloom. Average values for the percent of total P represented by residual P was  $71 \pm 10\%$  for all samples collected in April 2005 and  $82 \pm 12\%$  for all samples collected in July 2005.

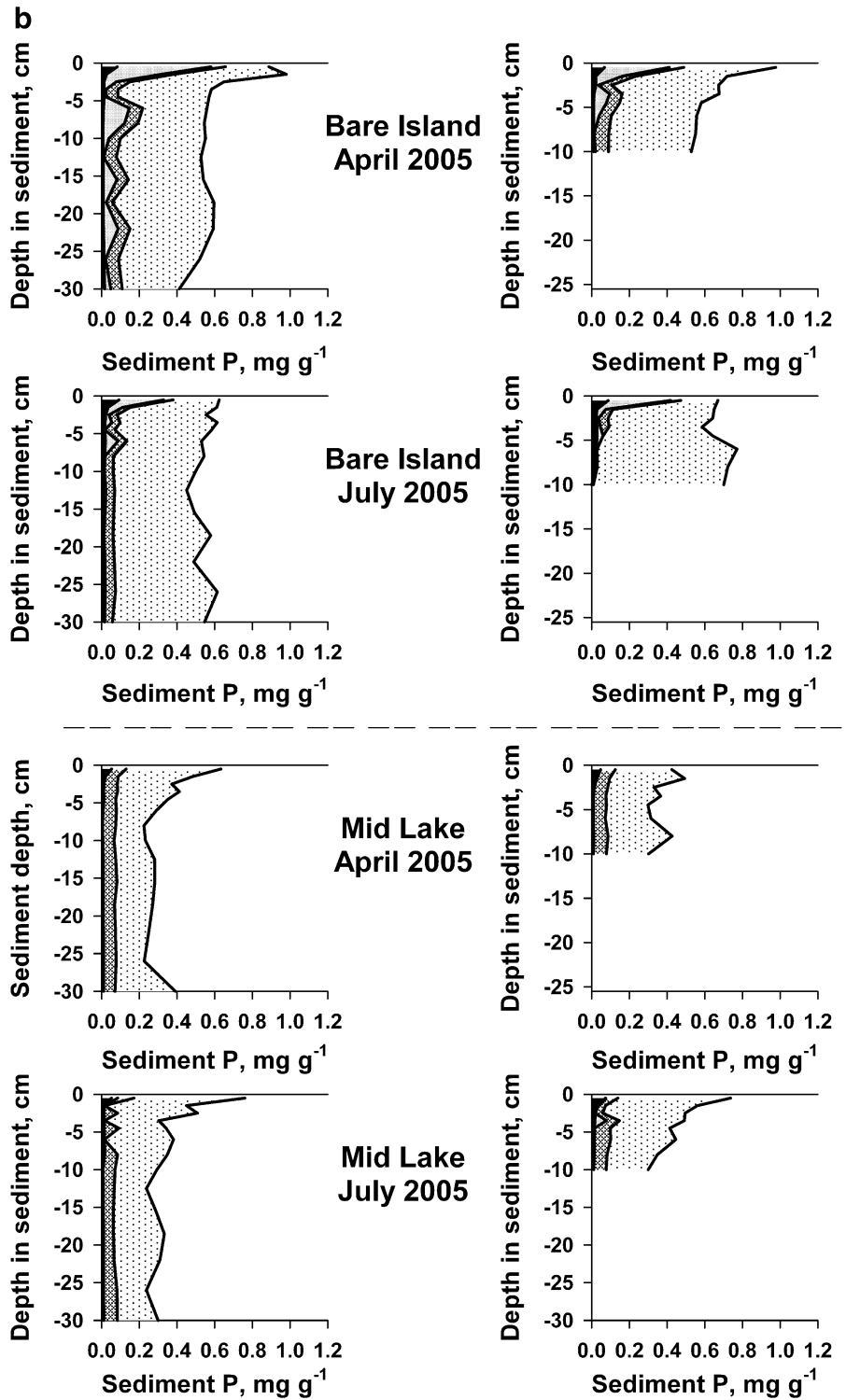
$^{31}\text{P}$  NMR spectra for sediment samples collected from the ML site in July 2005 are presented in Fig. 4. Signals were assigned to P compounds based on values reported by Koopmans et al. (2003) and Turner et al. (2003) for 0.25 M NaOH–0.05 M EDTA extracts of



soil samples. A peak for orthophosphate appears at approximately 6.8 ppm in all spectra. Concentrations of orthophosphate were larger in unextracted samples than in sequentially extracted samples. Signals appear-

ing between 5.5 to 4.4 ppm were assigned to orthophosphate monoesters. Signals appearing at approximately 3.03, 1.86, and -0.17 ppm were assigned to orthophosphate diesters. The peaks for orthophos-

Fig. 3 Continued.





phate monoesters and orthophosphate diesters are smaller in spectra for the sequentially extracted sediments than in the spectra for the sediments that were not subjected to sequential extraction. The signal at approximately  $-2.78$  ppm was assigned to pyrophosphate. This peak was present in the spectra of the sediments that were not extracted but did not appear in the spectra for the sequentially extracted sediments. Peak areas were calculated and compared with the integrated area (set at 1.0) for the peak at approximately  $18.30$  ppm, the internal standard, MDPA.

For ML, the sampling site with the largest Ca concentrations, the similarity in C concentrations before and after HCl treatment indicated that total C was organic C, with no detectable carbonates present (data not shown). The average concentration of organic C in all of the sediment samples was  $6.0\% \pm 1.5\%$  ( $n=112$ ). The largest concentrations of organic C were determined in the core from the BB site which had an average value of  $7.8 \pm 0.5\%$  for organic C. The smallest concentrations of organic C were determined in sediments from the ML site. The average value of organic C in the core from this site was  $3.9 \pm 0.4\%$ .

### 3.3 Statistical Analysis

Paired Student's *t* tests were used to determine whether UKL sediments acted as a source or sink for P during an AFA bloom in 2005. The largest differences for concentrations of total P and fractionated forms of P were apparent in the top two intervals of the cores. Therefore, data from the 0–1 and 1–2 cm intervals from the two cores collected at each site in April were compared with data from the 0–1 and 1–2 cm intervals in the two cores collected at each site in July. There was a significant difference between data for concentrations of total P in surface sediment samples collected in April and July at the BI and ML sites. In sediment from the BI site concentrations of P in the CDB extraction solutions from the intervals 0–1 and 1–2 cm were statistically larger (95% confidence) in the two cores collected in April than in the two cores collected in July. In sediment from the ML site, concentrations of residual P in the extraction solutions from the intervals 0–1 and 1–2 cm were statistically larger (95% confidence) in cores collected in July than in cores collected in April. Statistical analyses indicated that there was no net loss or gain of P in surface sediment from BB or HB during the study period.

## 4 Discussion

Concentrations of Al and Ca in surface sediments at the ML site are larger than concentrations of these metals at the other study sites. This might be explained by deposition of suspended particulates transported by circulation currents from the Williamson River, the primary tributary to UKL (Wood et al. 2006). A clay component in UKL and UKL watershed sediments has been described in earlier reports (Klamath Consulting Services, Inc 1983; Reynolds et al. 2004). Preliminary data obtained in our laboratory for surface sediment collected from UKL indicates that as much as 30% of the bottom material is clay particle size ( $\leq 0.01$   $\mu\text{m}$ ; unpublished data). Clays are a source of Al and sorbed cations, including Ca.

The largest concentrations of total Fe in UKL sediments are similar to the smallest concentrations of total Fe in the 66 lakes in eastern North America (Ostrofsky, 1987) and the smallest concentrations of total Fe in sediments of four lakes in Oregon (Spyridakis and Welch 1973). The median concentration of total Fe in surface sediment (top 2.5 cm) in 66 lakes in eastern North America was  $43 \pm 18$   $\text{mg g}^{-1}$ . The median concentration of total Fe in surface sediment (top 10 cm) in four Oregon lakes was  $56 \pm 26$   $\text{mg g}^{-1}$ . The median concentration of total Fe in the top 10-cm of cores from UKL collected for this study was  $15 \pm 7$   $\text{mg g}^{-1}$ . The correlation between total P and total Fe was not strong for these three sets of sediment samples. The  $R^2$  value for the relation between total P and total Fe in surface sediment was 0.39, 0.22, and 0.19, four in the eastern North American lakes, four lakes in Oregon, and UKL, respectively.

Concentrations of easily reducible Fe-oxyhydroxides were small and make up a small percentage of the total Fe concentrations in UKL sediments. The median value was  $3.0 \pm 2.0$  for the percent of the concentration of total Fe determined to be the concentration of poorly crystalline Fe-oxyhydroxides in sediments collected for this study. Concentrations of total P were well correlated with the concentration of dithionite-extractable Fe for all samples collected in April; the  $R^2$  was 0.60. The correlation between concentrations of total P concentrations of dithionite-extractable Fe for all samples collected in July was poor; the  $R^2$  was 0.05. The correlation between concentrations of total P and concentrations of dithionite-extractable Fe in sediments from the BI site had calculated  $R^2$  values of 0.91 and

0.04 in April and July, respectively. Concentrations of total P and dithionite-extractable P decreased in surface sediment at BI between the onset and full development of the AFA bloom in 2005.

Sorption capacity coefficients from Torrent et al. (1992) and Schwertmann and Cornell (2000) were used to calculate the maximum concentration of P that could be sorbed by the amount of poorly crystalline Fe-oxyhydroxides in UKL sediments. It was assumed in the calculations that the poorly crystalline Fe-oxyhydroxides extracted from UKL sediments were a combination of goethite and, to a lesser extent, ferrihydrite and that these poorly crystalline Fe-oxyhydroxides had a sorption capacity within the range of  $2.62 \pm 0.52 \mu\text{mol P m}^{-2}$  (Torrent et al. 1992). Using these assumptions, the calculated sorption capacity attributable to poorly crystalline Fe-oxyhydroxides in surface sediments from UKL is approximately  $0.08 \text{ mg P g}^{-1} \text{ DW}$ . The average concentration of P sorbed to poorly crystalline Fe-oxyhydroxides in UKL sediment was  $0.08 \pm 0.08 \text{ mg P g}^{-1} \text{ DW}$  ( $n=97$ ). These calculations indicate that the poorly crystalline Fe-oxyhydroxides in UKL sediments collected for this study were saturated with respect to P. The small concentrations of poorly crystalline Fe-oxyhydroxides and the resulting small concentrations of phosphate sorbed to them indicate that between April and July 2005, reduction of Fe-oxyhydroxides was not a major process for transfer of phosphate from the sediment to the water column in UKL.

A possible partial explanation for the small concentrations of poorly crystalline Fe-oxyhydroxides in UKL sediment might be complexation of iron. Complexation of Fe could inhibit the formation of amorphous Fe-oxyhydroxides. Brantley et al (2004) reported a high affinity of siderophores to complex with Fe (III) (formation constants between  $10^{-30}$  and  $10^{-51}$ ). Similarly, Robert and Chenu (1992) reported that microbial siderophores, in particular, have stability constants as Fe complexors that can be as large as  $10^{-40}$ . Development of complexes of iron and biogenic siderophores would be thermodynamically favored over Fe-oxide formation.

The median concentration of total P in the surface sediments (top 2.5 cm) of 66 lakes in eastern North America (Ostrofsky 1987) was  $2.42 \pm 1.20 \text{ mg g}^{-1} \text{ DW}$  sediment. The median concentration of total P in the top 10 cm of sediments of four lakes located in the state of Washington (Spyridakis and Welch 1973) and one lake located in the state of Oregon (Eilers et al.

**Fig. 4**  $^{31}\text{P}$  NMR spectra of nonsequentially extracted and sequentially extracted sediment from intervals 0–1, 1–2, and 2–3 cm in cores from the ML site collected in July 2005. In preparation for  $^{31}\text{P}$  NMR analysis, the samples were extracted for 16 h with 0.25 M NaOH–0.05 M EDTA. The extraction solution was neutralized and 50  $\mu\text{g P}$  as methylene diphosphonic acid (MDPA) added before freeze drying. Freeze-dried samples were prepared for  $^{31}\text{P}$  NMR analysis by adding 0.675 ml of 0.25 M NaOH–0.05 M EDTA and 0.2 ml 10 M NaOH in  $\text{D}_2\text{O}$ . The chemical shifts were indirectly referenced to external 85%  $\text{H}_3\text{PO}_4$  (at  $\delta=0.0$ ). Signals were assigned to P compounds based on values reported by Koopmans et al. (2003) and Turner et al. (2003). Chemical shifts for peaks in the spectra of UKL sediments included 6.8 ppm for orthophosphate, 5.5 to 4.4 ppm for orthophosphate monoesters, 3.03, 1.86, and  $-0.17$  ppm for orthophosphate diesters and  $-2.78$  ppm for pyrophosphate. Peak areas were calculated and compared with the integrated area (set at 1.0) for the peak at 18.30 ppm, the internal standard MDPA

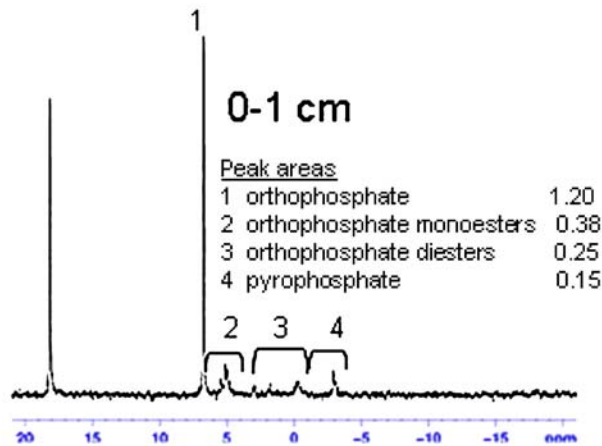
1996) was  $1.56 \pm 0.8 \text{ mg g}^{-1} \text{ DW}$  sediment. Concentrations of total P in the top 10 cm of UKL sediments have a median concentration of total P of  $0.61 \pm 0.19 \text{ mg g}^{-1} \text{ DW}$  sediment. The concern about internal loading in UKL is based more on the amount of P which is stored in its sediments rather than the concentration of total P per unit weight of sediment. UKL has an area of approximately  $232 \text{ km}^2$ . Using a median value of  $0.6 \text{ mg g}^{-1} \text{ DW}$  for the concentration of total P, the calculated amount of P in the top 10 cm of sediment in UKL is approximately 1,392 metric tons.

Larger concentrations of total P were determined in sediments near the top of the cores (<5 cm) than in sediment at depths of 10–30 cm. This could be explained by increased external loading (pollution) to the lake over time. It might also be explained by diffusion of P from depth in the sediment where microbial mineralization of P-containing organic compounds releases P. Released P diffuses from depth and can be retained by geochemical phases (e.g., poorly crystalline Fe-oxyhydroxides) near the sediment surface or stored as polyphosphates by bacteria in surface sediment.

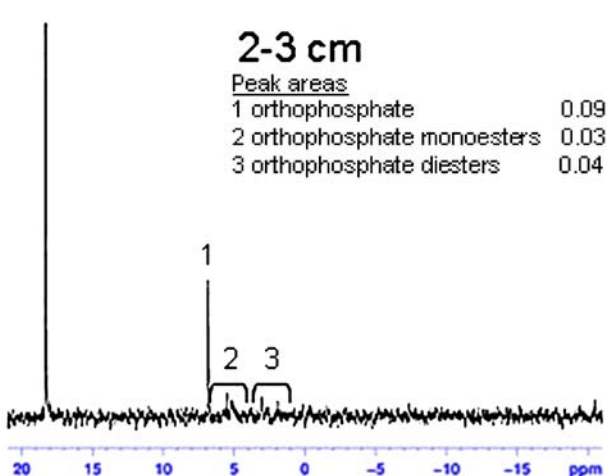
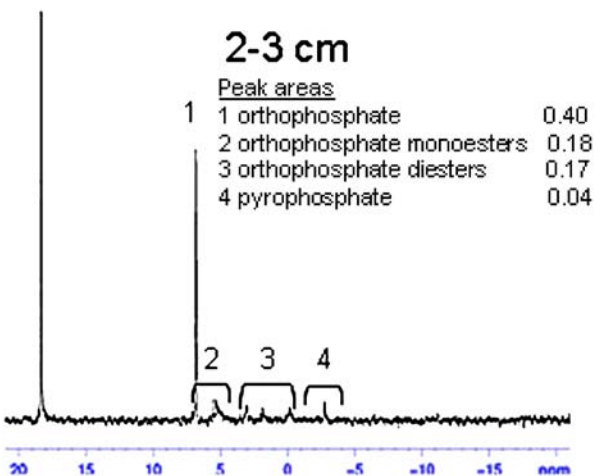
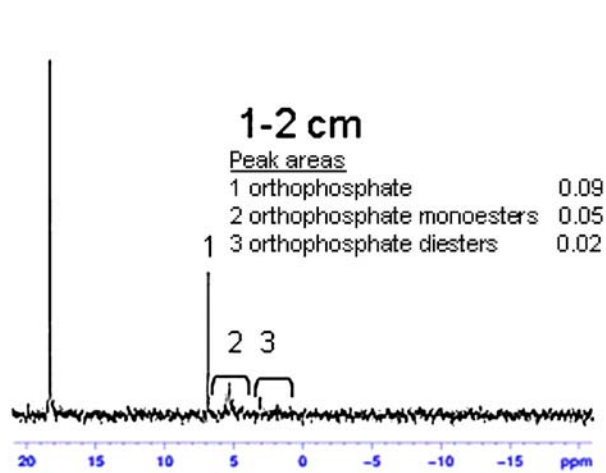
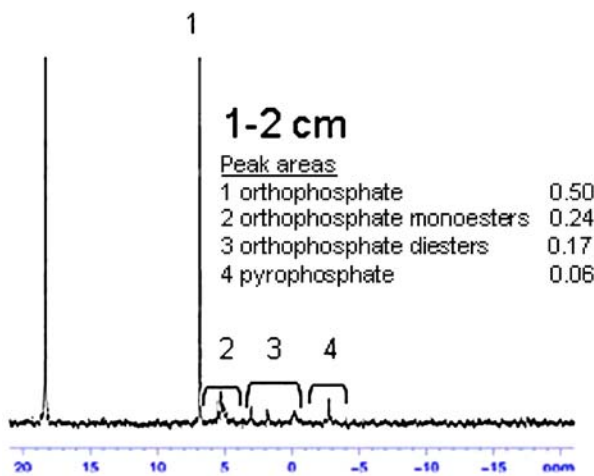
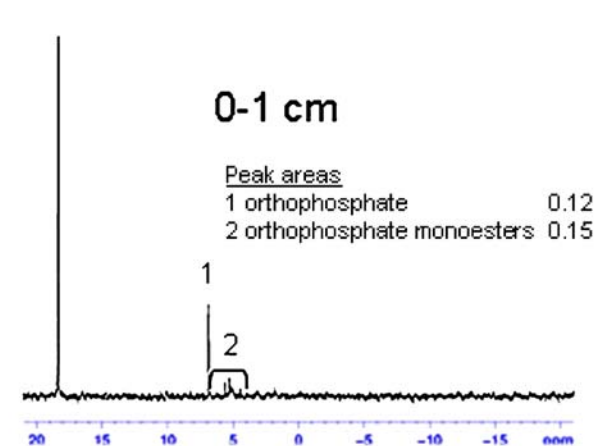
One molar HCl will extract P-containing salts of Ca and Al, and some P-containing Fe minerals (Aspila et al. 1976; Hieltjes and Lijkema 1980). Because UKL sediments are considered carbonate free (Bradbury et al. 2004a), HCl extractions of UKL sediments are unlikely to contain P associated with carbonates and are more likely to contain P associated with Al phases, acid labile P minerals, and some acid hydrolyzable organic P.

The  $^{31}\text{P}$  NMR data indicate that organic P lost from sediment during sequential extraction represents

## No sequential extraction



## Sequentially extracted



14–21% of residual P in ML sediment collected in July. If phosphate released from organic compounds in the sequential extraction procedure reacts with molybdate in the colorimetric analysis for phosphate, P concentrations in extracts for inorganic P will overestimate inorganic P, and calculated residual P concentrations will be underestimated. For this reason, the residual P concentrations calculated for these UKL sediments are considered minimum values.

$^{31}\text{P}$  NMR spectra of sediments from the ML site collected in July 2005 indicate that organic P (orthophosphate mono- and diesters) represents 20%, 13%, and 11% of total P, and 29%, 16%, and 14% of residual P in the 0–1, 1–2, and 2–3 cm intervals in ML surface sediments. Carman et al. (2000) used  $^{31}\text{P}$  NMR to determine organic P concentrations in intervals 0–5 cm (two lakes) and 0–7 cm (one lake) from cores collected in three Swedish lakes. They reported that the concentrations of organic P represented 4–44% of the residual P in their samples. Residual P concentrations and concentrations of organic P determined by  $^{31}\text{P}$  NMR in samples from both Carman et al. (2000) and this study indicate that concentrations of residual P cannot be assumed to be the same as concentrations of NaOH–EDTA extractable organic P. The median value for all samples collected in this study for the ratio of the concentration of residual P to the concentration of total P was  $0.79 \pm 0.11$ . Residual P was an important fraction of total P in these sediments.

Below a depth of approximately 2 cm at sites BB, BI, and HB, and at depths from 0–27 cm in ML sediment, the dominant form of P was residual P. The relative importance of the concentrations of residual P in relation to concentrations of total P in UKL sediment at depths greater than 2 cm is demonstrated by the following. The  $R^2$  values for the relation between total P concentrations and residual P concentrations are 0.76 and 0.82 for data from April and July, respectively. If data from intervals 1 and 2 are eliminated from each data set, the  $R^2$  values increase to 0.96 and 0.97 for data from April and July, respectively.

The median concentration of organic C in UKL sediments was  $6.0 \pm 1.6\%$ . There was a positive correlation between concentrations of organic C and total P in UKL sediments; the  $R^2$  was 0.6. The observation that there was a positive relation between concentrations of total P and organic C in UKL sediments complements the observation that the concentration of residual P (which includes organic P) in

UKL sediments is the dominant form of P in the sediment cores at depths greater than 2 cm. Future work should include studies to evaluate the bioavailability of the P in this fraction of the bottom sediments of UKL.

Statistical analyses indicated that at two sites, there were significant differences in the concentrations of total P in the surface sediment between the onset and full development of the bloom. The decreases in concentrations of total P and CDB-extracted P indicated that the net loss of P from sediment at the BI site between the months of April and July 2005 was the result of reduction of poorly crystalline Fe-oxyhydroxides in the sediments and release of associated P. The daily lake wide median concentration of DO in the water column was  $\geq 9$  ppm between April and July 2005 (Hoilman et al. 2008). The concentrations of DO at the sediment–water interface at each study site are unknown. Anoxic conditions at the sediment–water interface could develop at the BI site because the elevation of the island buffers the water column from mixing effects of prevailing WNW winds (Wood et al. 2006).

The net gain in concentrations of residual P and total P in the upper 2 cm of sediment from ML between April, before development of an AFA bloom, and July, after development of an AFA bloom, might be explained by accumulation of algal debris. Kleeberg (2002) attributed substantial concentrations of organic P in the sediments of Lake Petersdorf, Germany, to the large amounts of algal-derived P depositing on surficial sediments.

Internal loading of P has been identified as a major source of P to the water column in UKL (Walker, 2001; Oregon Department of Environmental Quality 2002). The amount of P that would have to come from surface sediments to increase the concentrations of total P in the water column to levels measured in 2005 could be determined by the following calculation. If we estimate the lake wide amount of P in the top 2 cm of sediment using the average concentration of total P ( $0.7 \text{ mg g}^{-1} \text{ DW}$ , 90% water) in the top 2 cm of sediment at the four sites sampled in this study, about 30% of the total calculated P in the top 2 cm of sediment would be required to increase water column concentrations of total P  $0.2 \text{ mg L}^{-1}$ , an increase observed at peak AFA development in 2005 (Hoilman et al. 2008). The median loss of P from the top 2 cm of sediment at the BI site was approximately 30%. Data from the other three sites in this study did

not indicate a net loss of P; therefore, the internal load of P in UKL in 2005 during the period of this study could not be accounted for by measured changes in P concentrations in surface sediment at the sites sampled for this study.

Our findings are not inconsistent with current understanding of the importance of internal loading of P in UKL. If the process responsible for addition of P from the sediment to the water column occurs (a) in the top 1 or 2 mm of sediment at the interface with the water column, (b) at depth in the sediment, or (c) is influenced by benthic activity, the techniques used in this study are not designed to confirm the process. To observe differences in the top millimeter of sediment, more detailed sectioning of sediment cores would be required. The contribution of P from depth in the sediment would be difficult to measure using P analysis of sediment cores collected during a short study period because percent changes in total P concentrations in the large volumes of sediment would be small. Comparison of cores collected at the same location over extended periods of time or diffusive flux measurements could address the question of loss of P from sediments at depth. To evaluate the importance of biological mechanisms for transporting P in the sediment to the water column, studies will be required to determine the populations of benthic organisms in UKL and the processes by which these organisms influence the P cycle. All future work should include more study sites within UKL.

## 5 Conclusions

Net changes in total P concentrations in surface sediment were detected at some sites during an algal bloom in UKL in 2005. Processes that contribute to P cycling between bottom sediment and the water column in UKL were not uniform throughout the lake. Statistically significant differences in concentrations of total P were measured in cores from two of the four sites in the period between onset and full development of an AFA bloom. Loss of P from Fe-oxyhydroxides was significant at one of the four study sites. Sediment samples analyzed in this study indicated that the concentrations of poorly crystalline Fe-oxyhydroxides in UKL sediments were small. Gain in residual P was significant at one of the four study sites. The dominant form of P in these

sediments at depth greater than 2 cm was residual P. Residual P concentrations cannot be equated with extractable organic P concentrations in these sediments.  $^{31}\text{P}$  NMR analyses indicated that the extractable organic P was no more than 30% of residual P in surface sediment collected at the ML site in July. There was a positive correlation ( $R^2=0.6$ ) for the relationship between total P and organic C in our samples. Continued studies of P cycling at more sites in UKL are indicated. Biological and physical processes should be investigated as factors in internal loading of P in UKL.

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