Characterization and Evaluation of Phosphate Microsensors to Monitor Internal Phosphorus Loading in Lake Erie Sediments

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Abstract

Monitoring phosphate concentration is very important to prevent and control eutrophication in natural waters. In this study, cobalt-based microsensors were modified, characterized, and tested to monitor internal soluble phosphorous (SRP) loading in lakes with improved detection limits. The effectiveness of surface modification on the performance of a cobalt-based microelectrode was fully examined by determining detection limit, response time, selectivity, interference with ions (sulfate, nitrate, and nitrite) and dissolved oxygen (DO). To assess their performance, phosphate sensors were applied to sediment samples collected from Lake Erie. SRP loading from sediments was determined under different DO conditions. After increasing the phosphate sensing area and modifying the surface, phosphate microsensors showed an increased detection limit of up to 10^{-8} M concentration of phosphate ion. The phosphate microsensor also showed its ability to measure sediment SRP profiling without disturbing sediment structure, and diffusion coefficients of phosphate in sediment could be determined under both oxic and anoxic conditions. Modified phosphate sensors showed improved sensitivity and could be applied to both water and sediment samples with high spatial resolution; however, signal interferences (especially with oxygen) required consideration during sample analysis. Overall, obtained results showed that phosphate microsensors can be an effective tool for measurement of phosphate in lake water and sediment samples for SRP monitoring.

Key Words

Eutrophication; microsensors; soluble reactive phosphorous; phosphate; sediment; internal phosphorus loading

1. Introduction

As a result of anthropogenic impacts, including sewage effluent and agricultural fertilizer release into the water or soil, phosphorus is considered a universal concern for environmental managers, as it contributes to the eutrophication of water bodies (Ritter et al., 2002; Sharpley et al., 1994). In a water system, phosphorus loading is comprised of two major parts: external and internal loading. External phosphorus loading refers to the phosphorus load deposited into the water column, while internal phosphorus loading refers to the accumulation of phosphorus in the sediment (Ekholm et al., 1997). Studies show that phosphorus becomes accumulated in sediment when external phosphorus loading is high (Granéli, 1999; Søndergaard et al., 2003). Furthermore, the required recovery periods in lakes after the reduction of the external phosphorus loading are lengthened due to release of the internal phosphorus load (Garber and Hartman, 1985; Molen and Boers, 1994; Søndergaard et al., 1999; Steinman et al., 2004). As a major influence on the duration of recovery, reports indicate that phosphorus content in sediment can be approximately 1000 times higher than that found in the water column (Rivas et al., 2000). Thus, even small phosphorus release from sediment may significantly affect lake water phosphorous concentration (Pettersson, 1998).

Total phosphorus (TP) has been accepted as a good indicator of a lake's nutrient status to prevent eutrophication, as it remains more stable than soluble reactive phosphorus (SRP) in lakes. However, SRP has received more attention than total phosphorus (TP) recently as an approach for managing eutrophication. SRP is largely comprised of the inorganic orthophosphate (PO_4), and this labile phosphorus variant is a great concern as it is directly ingested by algae and accelerates eutrophication (Hilton et al., 2006). To monitor SRP release to water bodies from

different sources, various analytical methods have been used. The common analysis of phosphorus in sediment involves two general procedures. First, digestion of phosphorus is performed by oxidizing various forms of phosphorus to dissolved phosphate in a strong acidic media (Colina et al., 1996). After digestion, the digested phosphate is determined using various detection techniques such as spectrometry and ion-chromatography (Chen et al., 1998; Colina et al., 1996). Although these techniques provide very accurate results, such analytical methods require complex and time-consuming sample pretreatment procedures. In addition, the instruments are prohibitively expensive and inappropriate for field sample analysis (Kim et al., 2007).

Microsensors, also called microelectrodes, are useful tools that have been applied for measurements of various analytes in micro-environments, such as soil pores, biofilm, and activated sludge floc (Dalsgaard and Revsbech, 1992; Li and Bishop, 2004; Sexstone et al., 1985). This technology, which has drawn increased attention in recent studies, offers several advantages over standard phosphate detection methods, including low cost, simple fabrication procedures, portability, and ability to be applied in both direct and indirect detection of phosphorus (Korostynska et al., 2013). However, to date, few microelectrodes are used for direct determination of phosphate since they are not commercially available (Carey and Riggan, 1994; Chen et al., 1998). In a previous study, a cobalt wire based phosphate sensor with small tip diameter (5-10 μm) was successfully fabricated and tested to directly monitor phosphate ion concentration changes in activated sludge floc during the anaerobic phase of an enhanced biological phosphorus removal (EBPR) process (Lee et al., 2009). In comparison with other colorimetric techniques, such as wet-chemical oxidation and microfluidic technology, this cobalt wire based microelectrode showed a much quicker response time and a wide detection range (Al-

Dasoqi et al., 2011). However, the sensor showed limited field application capability beyond samples from wastewater with high phosphate concentration, due to its detection limit. In addition, phosphate detection interferences from co-existing ions, common to fresh water systems (lake water and sediment), have not yet been evaluated. Because of deficiencies in sensing devices for directly monitoring SRP in sediments, internal SRP loading from lake sediment is not well understood.

The main goal of this study was to assess performance of the phosphate microsensor and its applicability for *in-situ* SRP monitoring. To achieve this goal, cobalt-based microsensors were modified, characterized and tested to improve detection limits for phosphate ($< 10^{-5}$ M). The effectiveness of surface modification of a cobalt-based microelectrode was examined by determining detection limit, response time, selectivity, reproducibility, lifetime, interference with ions (sulfate, nitrate, and nitrite), and dissolved oxygen (DO). Then, the microsensor was further characterized and tested on sediment samples collected from Lake Erie to assess its applicability to detect internal SRP loading under different redox conditions.

2. Materials and Methods

2.1 Microsensor fabrication

The basic detection mechanism of the cobalt based phosphate sensor involves measurement of the voltage output response to phosphate concentrations. When the cobalt first makes contact with water, a cobalt oxide film is formed on the surface. This oxide layer subsequently interacts

with phosphate ions in solution and creates a unique cobalt phosphate complex layer on the surface, depending on the pH level of the solution (Engblom, 1999) [equations (1-3)].

In order to construct the phosphate sensor, two fabrication methods were considered. First, glass pipette barrels were used and tested for phosphate sensor fabrication. Then, a new type of phosphate sensor was fabricated using polymer based sealants (see supporting information figure S1). For glass pipette based sensor fabrication, glass pipette barrels (O.D.: 1.2mm, I.D.: 0.69mm, 15cm length, Sutter instrument Co.) were purchased, heated, and pulled over the flame. After pulling the barrels, a section of cobalt wire (0.1mm diameter, 99.995% pure, Aldrich Chemical Company) was inserted into the pulled glass micropipette. Without the use of an etching process, the cobalt wire maintained its initial tip size. The micropipette was then melted in the middle section using a trough heating filament (Sutter instrument Co.) to completely seal the cobalt wire in the stretched glass pipette barrel. The tip of the sensor was then beveled using a diamond abrasive plate (BV10-1684, Sutter instrument Co.) to hone the sensor tip to a 45° angle and expose the cobalt surface. Then, the microelectrode was connected to copper wire by melting a small section of bismuth alloy (44.7% bismuth, 22.6% lead, 19.1% indium, 8.3% tin and 5.3% cadmium) (Lee et al., 2009). To fabricate a new type of phosphate sensor, cobalt wires were dipped in a polymer solution, and the polymer layer was solidified and beveled to expose the cobalt surface. To measure electrical sensor response during calibration and sensor performance experiments, the potential between the working microelectrode and reference electrode was

monitored using a millivolt (mV) meter (Model 250, Denver instruments). An Ag/AgCl reference mini-electrode (MI-401, Microelectrodes Inc.) was used as the reference electrode. The pH/ mV meter was connected to a computer and data was acquired with a spread data logger (Balance Talk SL TM, Labtronics Inc.) to record responses at five second intervals.

After fabricating the microsensors, the sensors were pretreated prior to calibration. In detail, sensors were first immersed into DI water to form a cobalt oxide (CoO) layer on the surface of sensor tips. Upon reaching a stable potential, the sensors were removed from DI water and immersed into potassium phosphate (KH_2PO_4 , 10^{-4}M) solution at pH 7 until a new steady-state potential was obtained. Following the pretreatment process, phosphate sensors were calibrated. Several phosphate standard solutions were prepared using KH_2PO_4 at a range of 10^{-1}M to 10^{-8}M , where the solution pH was maintained at 7. Sensor calibration and experiments were conducted under condition of ambient oxygen levels and room temperature (20°C). To monitor sensor performances, tested sensors were calibrated again after being applied to samples.

2.2 Ion and DO (dissolved oxygen) interference tests

Considering the co-presence of other ions in the natural environment and their potential interference with sensor performance, sulfate, nitrite, and nitrate ions were used to investigate potential ion interferences on the phosphate sensor response. For each ion interference experiment, interfering ions were kept constant while phosphate ion concentrations were varied from 10^{-3}M to 10^{-6}M . Ion interference tests were also conducted by keeping the phosphate ion concentration constant while the other ion concentrations were varied. Table S1 shows the tested ions and the ranges of tested ion concentration. The concentrations of three interfering ions were selected based on reported ion concentrations in sediments (Boström et al., 1988; Boström and

Pettersson, 1982; Cowan and Boynton, 1996; Holmer and Storkholm, 2001; Jensen and Andersen, 1992; Lovley and Klug, 1983; Søndergaard et al., 1993; Xie et al., 2003).

Typical benthic DO concentration in lakes is about 0.1 mM (Archer and Devol, 1992). Based on this reference point, two different oxygen concentrations [0 mM and 0.13 mM (4.2 mg/L)] were tested to monitor dissolved oxygen interference on the performance of phosphate sensor. Other test conditions such as phosphate concentration, pH, temperature, and distance between reference electrode and the phosphate sensor remained fixed. Nitrogen gas mixed with varying amounts of oxygen gas was bubbled into the solution via a diffuser to achieve relevant DO concentrations (0 and 0.13mM DO). A micro-oxygen electrode (MI730, Microelectrodes, Inc.) was used to monitor the dissolved oxygen levels in the solution.

2.3 Water and Sediment Samples

Performance of fabricated sensors was first evaluated using water samples including effluent samples from a wastewater treatment plant and Lake Erie water. Next, sensors were applied to monitor SRP release from sediment samples. Sediment samples were collected from a site in Lake Erie in June by Dr. Thomas Bridgeman's research group. Upon collection, the sediment samples were sealed in plastic containers and transferred directly to a laboratory, where they were refrigerated prior to sensor tests and phosphorus analysis. The total phosphorus (TP) and soluble reactive phosphorus (SRP) concentrations were measured as 0.9698 mg/g dry weight and 0.096 mg/g dry weight, respectively, by the National Center for Water Quality Research at Heidelberg University.

In order to monitor SRP release from sediments with the phosphate sensors, sediment samples were carefully transferred into a beaker and then saturated with Lake Erie water. The depth of

the sediment sample was approximately 5 cm, and the depth of lake water was approximately 10 cm (5 cm above sediment surface). A microscope was used to identify the sediment-water boundary location for sensor application. The phosphate sensors were connected to a manipulator, which enabled precise vertical movement of sensors through the depth of the sample. After a stable potential was acquired, the position of the sensors was slowly adjusted to avoid disturbing the sediment structure. During the profiling process, the Ag/AgCl reference electrode was kept in a static position. The experiments were conducted in ambient air and room temperature (20 °C). The concentration of phosphate in surface water was also measured using ion chromatography (IC-1200, Dionex Company) to verify the performance of the microsensors.

3. Results and discussion

3.1 Calibration of microelectrodes

Figure 1(a) shows a phosphate sensor calibration curve. Eight different concentrations of standard phosphate solutions were prepared (10^{-8} to 10^{-1} M KH_2PO_4) for sensor calibration. The sensors demonstrated a detection limit of up to 10^{-8} M (typically between 10^{-6} and 10^{-7} M) concentration of phosphate ion. Also, the phosphate microsensors exhibited good linear response to phosphate concentration change with a slope of -32.9 mV/decade. Figure 1 (b) shows the response changes of microsensors over time. During the calibration, the response time of sensors for each standard point was between 5 seconds and 1 minute, except at 10^{-8} M phosphate concentration, which required approximately 100 seconds for stabilization.

3.2 Ion interference

Three ions (sulfate, nitrite, and nitrate) commonly found in freshwater samples were tested to evaluate possible interference in the performance of the phosphate microsensors. Figures 2 (a)-(c) show the ion interference tests results. Compared to calibration curves generated in the absence of an interfering ion, the presence of ions did impact the electrode signal responses for phosphate ion detection. The effects of nitrite and nitrate ions on signal variations were not statistically significant (ANOVA test, P-value = 0.28 and P-value = 0.22, respectively), while the effect of sulfate was statistically significant with a P-value less than 0.05. Please note, however, that the tested sulfate concentration range in the experiment is much wider than the other two ions [0 to 3.12×10^{-1} mM (30 mg/L) of sulfate]. There was no significant effect between regression curves from 0 to 5.2×10^{-2} mM (5 mg/L) of sulfate (P value = 0.24). This suggests that it is better to operate the sensors under lower sulfate concentration; however, if the sulfate concentration is high, the sensors should be calibrated in accordance with the ambient sulfate concentration. Despite the signal shifts, all calibration curves retained a linear response to the changes of phosphate ion concentrations, and the signal response shift resulting from tested ion interference appeared to be minor.

3.3 DO (Dissolved Oxygen) interference

Figure 3 shows signal response changes of the phosphate microsensor under different DO concentrations. From experimental results, overall, a 40 mV signal response difference was observed between 0 mM and 0.13 mM DO. While ion interferences on phosphate sensor performance were not significant (signal shifts were relatively large only at very low phosphate concentration), phosphate sensor signals were shifted for all phosphate solution concentrations under 0.13 mM DO. These results indicated that oxygen interferes with the binding mechanism

between cobalt oxide and phosphate, subsequently decreasing the sensitivity of sensors to phosphate ions (Lee et al., 2009). At high phosphate concentrations, signal shift may cause a significant difference in phosphate concentration measurement. These findings indicate that calibration curves should be developed for varied DO concentrations, and that DO measurements are necessary for phosphate sensor applications.

3.4 Aqueous sample measurement

After calibrating the phosphate microsensor, measurement of phosphate concentration was tested with various environmental samples. To validate the accuracy of the phosphate sensor, the phosphate concentration in the samples was also measured by ion chromatography (IC). Table S2 provides a comparison between phosphate measurement by microsensor and by IC. Despite interference in one sample with a very high ion concentration (with 5.64 mM sulfate concentration), obtained results from various water samples revealed that the microsensor results correlated well with IC determination and that the microsensors can accurately detect phosphate in the samples tested, especially after correcting for DO interference.

3.5 Influence of reference electrode position on phosphate measurement

Before measuring phosphate concentration in the sediment with the phosphate microsensor, the influence of the location of a reference electrode on the phosphate measurement was determined. The distance between phosphate microelectrode and Ag/AgCl reference electrode, as well as the location (in sediment or in bulk solution) of the Ag/AgCl reference electrode, were evaluated by monitoring the signal changes of the phosphate microelectrodes. According to the experimental results, varying distances between a phosphate microsensor and a reference electrode resulted in up to ± 30 mV signal difference during sediment phosphate profiling. However, sensor signal

shift (± 2 mV) was insignificant when the reference electrode was buried in sediment samples, as compared to when it was located in bulk solution. In order to obtain reproducible and consistent sensor responses for sediment application, the position of the reference electrode was kept constant during all sediment profiling experiments. Also, the phosphate microelectrode was only allowed to have vertical movement in order to maintain a fixed positional distance between the Ag/AgCl reference electrode and the phosphate microelectrode during SRP profiling.

3.6 Reversibility of profiling measurement

To determine the versatility and reproducibility of phosphate sensors for SRP profile in sediment, phosphate profile measurements were conducted during both insertion into sediments and withdrawal out of sediments, after which signal responses were compared. Figure S2 shows the measured phosphate profile in sediment during both insertion and withdrawal processes. The trends are very similar, especially as the sensor moved through the depth of the sediment. Overall, there were no significant differences between the inserting process and withdrawing process for phosphate measurements. This finding indicated that the phosphate microsensors are able to monitor local phosphate concentration changes in sediment samples without disturbing sediment structure, resulting in accurate *in situ* sediment profiles.

3.7 Sediment SRP profiling under different oxygen concentrations

Sediment SRP profiling experiments were conducted under controlled DO conditions (0 mM DO and 0.13 mM DO concentration). Phosphate concentrations were monitored from 5 mm above the sediment surface to 25 mm within the sediment sample, as shown in Figure 4.

First, a SRP profile was generated under 0.13 mM DO concentration. The SRP concentration was $10^{-7.8}$ M in bulk solution, which was the same as the concentration detected by IC. Before the sensor reached the sediment surface, there was a diffusive boundary layer (DBL), approximately 0.3 mm thick, where SRP concentration slightly increased. The flux of the SRP across this area is considered to be dominated by diffusion (House and Denison, 2002). Then, the sensor reached the surface of the sediment where the phosphate concentration was 10^{-7} M. As the depth increased, the concentration of phosphate increased sharply. At a depth of approximately 3 mm from the surface, the concentration of phosphate was stable and determined as approximately $10^{-5.7}$, which was nearly 20-fold the surface concentration.

The upper 3 mm layer of the sediment where SRP concentration changed sharply is the oxygenated layer, which is called the surface oxic layer and contains dissolved oxygen in the pore water. According to previous studies, oxygen only penetrates 3-5 mm deep in sediment (Jorgensen and Revsbech, 1985; Revsbech et al., 1980), which was consistent with our study (measurement by DO microsensor, data not shown). Numerous studies have shown that the surface oxic layer plays an important role in preventing phosphorus from leaching into bulk water (Anthony and Lewis, 2012). Under oxic conditions, phosphorus is insolubly bound in sediment through chemical and biological processes. Chemically, phosphorus is retained in sediment by fixation to Fe (III), which absorbs phosphorus in an oxidized form. Biologically, some polyphosphate accumulating bacteria groups, like *Acinetobacter spp.*, will store phosphorus as polyphosphate under oxic conditions, while using oxygen as a terminal electron acceptor. Thus, the surface oxic layer of the sediment acts like a boundary to phosphorus leaching into the water column. Under oxic conditions, the profile generated during this

experiment exhibited agreement with the theoretical profile generated using a Boundary-Layer model and a Triple Zone Model, proposed in a previous study (House and Denison, 2002).

Similarly, a SRP profile was then generated under 0 mM DO concentration. With 0 mM DO concentration in the bulk water, there was not sufficient oxygen to generate a surface oxic layer; therefore, the surface of sediment and pore water was anoxic. Accordingly, phosphorus was released from the sediment as a result of Fe (III) reduction and subsequent dissolution of iron-bound phosphorus complexes. In conjunction, the reduced iron reacts with H_2S , as the product of simultaneous sulfate reduction in the sulfur cycling process and results in the formation of insoluble iron sulfide under anoxic conditions. In this case, chemical reduction of iron complexes and sulfate in the sediment result in a more favorable end product. Therefore, the established competition between sulfide and phosphorus results in the indirect release of phosphorus (Holmer and Storkholm, 2001). In biological processes, the stored polyphosphate was used by polyphosphate bacteria to provide energy in order to store organic carbon as polyhydroxybutyrate (PHB), resulting in a phosphorus release (Davelaar, 1993). The SRP concentration in bulk water increased to $10^{-3.4}$ M due to the phosphorus release from the sediment under anoxic conditions, where the results are consistent with the concentration measured using IC ($10^{-3.5}$ M). As the sensor reached the sediment surface, the SRP concentration decreased. A stable concentration ($10^{-6.1}$ M) was obtained around a depth of 6 mm, although the concentration was lower than the stable concentration under oxic condition ($10^{-5.7}$ M) between 6 mm and 20 mm. This was because phosphorus was released and subsequently transported to the upper sediment layer. The SRP concentrations under both conditions finally reached the same concentration at depths greater than 20 mm.

3.9 Diffusivity calculations

The analysis of mass transport in sediment is very important in order to understand the mechanisms and rate of SRP loading from sediment. As such, the effective diffusion coefficient of phosphate release in sediment must be determined. Based on obtained phosphate profile data under 0 mM DO concentration and 0.13 mM DO concentration, diffusivities of SRP in sediment were calculated and compared to each other as described previously (Abrahamson et al., 1996).

The calculation is based on an assumption that the flux (J_{f,X_s}) of phosphate diffusion from the sediment to the surface of the sediment is equal to the phosphate flux (J_{w,X_s}) across the surface of the bulk solution, using Fick's law (Lee and Bishop, 2010; Lewandowski, 1994).

In both sediment and bulk solution,

$$J_{f,X_s} = D_f \left(\frac{dC}{dx} \right)_{f,X_s} = D_w \left(\frac{dC}{dx} \right)_{w,X_s} = J_{w,X_s} \quad (4)$$

In sediment,

$$\left(\frac{dC}{dx} \right)_{f,X_s} = A(C_{sedi} - C_s) \quad (5)$$

$$-A(x - x_s) = \ln \left(1 - \frac{C - C_s}{C_{sedi} - C_s} \right) \quad (6)$$

In bulk solution,

$$\left(\frac{dC}{dx} \right)_{w,X_s} = B(C_s - C_b) \quad (7)$$

$$-B(x - x_s) = \ln\left(1 - \frac{C - C_s}{C_s - C_b}\right) \quad (8)$$

, where sediment surface $x_s = 0$ cm, x = local depth (cm), C_s = SRP concentration at the surface (mg/L), C = local SRP concentration (mg/L), C_{sed} = stable SRP concentration in sediment, C_b = stable SRP concentration in bulk water. A and B are constants, which can be determined from the slopes presented in Figure 5.

First, the diffusion coefficient under 0.13 mM DO concentration (D_{f1}) was calculated. Figure 5 (a) shows $\ln[1-(C-C_s) / (C_{\text{sed}}-C_s)]$ versus $(x-x_s)$ inside the sediment. The slope is the coefficient A in equation (4), where $C_{\text{sed}} = 0.20$ mg/L as phosphate, $C_s = 0.01$ mg/L as phosphate, and the calculated coefficient $A = 7.3454 \text{ cm}^{-1}$. Figure 5 (b) shows $\ln[1-(C-C_s) / (C_s-C_b)]$ versus $(x-x_s)$ in bulk solution. The slope is the coefficient B in equation (8), where $C_b = 0.002$ mg/L as phosphate,

and the calculated coefficient $B = 16.146 \text{ cm}^{-1}$. Therefore, both $\left(\frac{dC}{dx}\right)_{f,x_s}$ and $\left(\frac{dC}{dx}\right)_{w,x_s}$ can be calculated using equations (5) and (7). The phosphate ion diffusivity (D_w) in water is $7 \pm 0.5 \times 10^{-6} \text{ cm}^2/\text{s}$ at 20°C (Krom and Berner, 1980). Thus, the phosphate ion diffusivity in sediment under oxic condition (D_{f1}) was calculated to be $6.48 \times 10^{-7} \text{ cm}^2/\text{s}$ under oxic conditions.

After elucidating phosphate diffusivity in sediment under oxic conditions, the diffusion coefficient under 0 mM DO concentration (D_{f2}) was calculated. Similar to the previous calculations, figure 5(c) shows $\ln[1-(C-C_s) / (C_{\text{sed}}-C_s)]$ versus $(x-x_s)$ inside the sediment, where $C_{\text{sed}} = 0.057$ mg/L and $C_s = 5.854$ mg/L as phosphate. The slope in figure 5(c) is $A = 8.956 \text{ cm}^{-1}$. Figure 5(d) shows $\ln[1-(C-C_s) / (C_s-C_b)]$ versus $(x-x_s)$ in bulk solution where $C_b = 34.87$ mg/L as

Phosphate. The slope in figure 5(d) is $B = 5.427 \text{ cm}^{-1}$, and the phosphate ion diffusivity in sediment under anoxic condition (D_{f2}) was calculated to be $2.12 \times 10^{-5} \text{ cm}^2/\text{s}$.

When comparing the two diffusivity values, the phosphate ion diffusivity under 0 mM DO concentration (D_{f2}) is larger than the phosphate ion diffusivity under 0.13 mM DO concentration (D_{f1}). Thus, the release rate of SRP is larger under anoxic condition, which confirmed the result obtained by sediment profiling. Also, considering variability in sediment samples, the calculated diffusivity under 0 mM DO concentration ($D_{f2} = 2.12 \times 10^{-5} \text{ cm}^2/\text{s}$) is within an acceptable range compared to the diffusivity value in anoxic sediments ($D_s(\text{PO}_4) = 3.6 \pm 1.1 \times 10^{-6} \text{ cm}^2/\text{s}$) reported by a previous study (Krom and Berner, 1980).

4. Conclusions

In this study, cobalt-based microsensors were modified, characterized, and performance tested for phosphate detection limits, response time, selectivity, interference with ions (sulfate, nitrite, and nitrate), and DO (dissolved oxygen). After increasing phosphate sensing area and re-modifying the surface, phosphate sensors possessed improved detection limits to 10^{-8} M concentration of phosphate ion. The measurements for environmental samples also revealed that the phosphate microsensor performance correlated well with IC results and possessed a high level of accuracy to detect phosphate in solutions. However, signal interferences (especially with oxygen) needed to be considered and properly addressed for sample analysis with phosphate microsensors. For *in-situ* SRP profiling in lake sediment samples, the phosphate sensor could

monitor local phosphate concentration changes with high resolution and sensitivity. In using the phosphate sensor, researchers should ensure that obtained sediment samples are fresh, because ion concentrations may change over time. Diffusion coefficients for SRP in sediments under different oxygen levels could be determined for the first time based on actual sediment profiling data. Overall, the successful characterization and application of the phosphate microelectrode showed that the cobalt-based phosphate microsensor can be a very useful tool to not only monitor phosphate concentration in solutions but also study SRP dynamics in sediments under various environmental conditions.

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Figure 1. Performance of phosphate sensor: (a) calibration curves, (b) response changes of sensors over time

Figure 2. Ion interference: (a) sulfate, (b) nitrite, (c) nitrate

Figure 3. Dissolved oxygen interference (at 10^{-4}M KH_2PO_4 concentration)

Figure 4. Sediment SRP profiling under 0 mM and 0.13 mM DO

Figure 5. Diffusivity calculations: (a) $\ln[1-(C-C_s)/(C_f-C_s)]$ versus (x-x_s) in sediment (DO is 0.13mM); (b) $\ln[1-(C-C_s)/(C_s-C_b)]$ versus (x-x_s) in bulk solution (DO is 0.13mM); (c) $\ln[1-(C-C_s)/(C_f-C_s)]$ versus (x-x_s) in sediment (DO is 0 mM); (d) $\ln[1-(C-C_s)/(C_s-C_b)]$ versus (x-x_s) in bulk solution (DO is 0 mM)