Long-term in situ chemical sensors for monitoring nutrients: phosphate sensor commercialization and ammonium sensor development

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LONG-TERM GOALS

The long-term goals of this project are to 1) transition the CYCLE-Phosphate sensor from a prototype to a fully functional commercial product to enable sustained observations of phosphate concentration for detailed investigations of biogeochemical variability in open ocean and coastal environments, and 2) develop an in situ ammonium sensing capability along with the phosphate sensor, thereby allowing comprehensive research to be conducted on the impacts and controls of nutrient cycling in coastal and nearshore ecosystems.

OBJECTIVES

The overarching objective of our effort is to develop a reliable, accurate, long-term in situ nutrient sensing sensor system employing reagent-based colorimetric methods using fluidics (the so called “wet” chemistry method) that is easy to use and can be readily adapted for several nutrients. WET Labs has been developing a core set of technologies to utilize optical detection techniques for determination of nutrient concentrations using low volume, reagent-based reaction chemistry. Specifically in this work we focus on the commercialization of the CYCLE phosphate prototype in situ sensor. Evaluation of the *in situ* data obtained using this system has shown that the measurement technology is robust, and that the sensor provides stable and accurate readings. In Phase I of this project, we seek to transition this technology from prototype status to a commercial product. Our efforts will broadly encompass design issues for simplifying manufacturing, maintenance, service, customer easy of use, and enhancements for integrating the sensor with other observing systems. We propose to build wet chemistry based sensors with easy to use cartridges, removing the need for users to deal with the chemistry. By accomplishing these tasks, we not only intend to produce a commercially viable sensor, but also to set the technology foundation from which development or adaptation of the system for other nutrients can occur. In Phase II of this project, we aim to modify the reagent based detection methodology for the *in situ* determination of ammonium concentration.

APPROACH AND WORK PLAN

Based on the excellent results obtained during our extensive field testing conducted over the prior year with the CYCLE Phosphate sensor, we have defined a set of goals necessary to accomplish a successful transition of these prototype sensors to a commercial product. The primary focus of these goals is to provide the user an easy to use, wet chemistry-based *in situ* phosphate sensor.

Goals for Phase I – Commercialization of the in situ CYCLE Phosphate sensor

- Utilize easy to replace reagent cartridges to allow user to keep "hands off" chemicals
- Instrument will be relatively small (16 x 6inches) and light weight (less than 15 pounds).
- Improved reliability of internal calibration.
- On board calculation of Phosphate concentration.
- Easy exchange of intake filters, covers and other expendable items.
- Automation of laboratory dilution series.
- Seamless integration of sensor with other data acquisition/control systems (interoperability).
- Employ anti-biofouling measures to allow for unattended deployment for three months, thereby reducing service and maintenance costs while ensuring data quality.
- Extensive field testing by WET Labs and the project partners.

The focus of our efforts in Phase II of this project will be to develop and test an in situ ammonium sensor based upon the reagent-based chemical analytical system developed for the CYCLE Phosphate sensor. Having addressed several of the key fluidic, electrical, and ease of use issues through the work associated with the transition of the CYCLE-Phosphate sensor, we will be well positioned to focus on the development of a stable, reliable, accurate wet chemistry methodology to employ on the CYCLE Ammonium sensor.

Goals for Phase II – Development of the in situ CYCLE Ammonium sensor

- Identification and testing of the most suitable method for the determination of dissolved ammonium.
- Development of ammonium chemistry for long-term storage.
- Design new manifold based on the above.
- Obtain precision and detection limit (0.1 and 1 μM, respectively).
- Extensive testing by the Project Team.

WET Labs project team consists of Dr. Andrew Barnard (lead PI), Casey Moore (corporate lead), Dr. Ron Zaneveld (Science liaison), Bruce Rhoades (senior engineer and project manager), John Koegler (senior application engineer), Alex Derr (senior mechanical engineer), Cale Wetzel (production engineer), and Marvin Johnson (production engineer). As the goal of phase I of this project is to transition the existing prototype from engineering to production, several WET Labs production personnel are also involved in the fabrication of the commercial sensor. One of the major foci in the early stages of our work has been to stage the release of the commercial sensor to production (personnel, training, manufacturing, machining, etc) in tandem with the necessary engineering. As such, we have involved from the onset, our applications and production engineering teams to help facilitate the design of the system and to ensure that the system is inherently manufacturable by our staff.

Our project partners include SubChem Inc (Dr. Al Hanson) who will assist with reagent production, stability, and verification, as well as provide assistance with the ammonium sensing methodology development, and Dr. James Ingle (Professor Emeritus, Oregon State University) who is acting as a consultant to the project on the ammonium sensing wet chemistry methodology and sensing design. Several other groups are involved with this project including Dr. Burt Jones (University of Southern California), Dr. Ru Morrison (University of New Hampshire), Dr. Dan Doolittle (Kachemak Bay Research Reserve), Robert Campbell (Prince William Sound Science Center), and Doug Wilson (NOAA Chesapeake Bay Office). These groups will be acting as CYCLE sensor evaluators for the project, and as such will be provided with CYCLE sensors to deploy on there established monitoring platforms. Their primary role is to provide the project with nutrient validation data, assist with issues related to integration with data systems, and to act as independent evaluators of the CYCLE sensing capabilities.

At the time of writing this annual report we have just started our 3 year effort (< 90 days into the first year of funding). In the first 3 months of this project, we have focused on several key engineering tasks to transition the prototype CYCLE phosphate sensor to a commercial product. These tasks are outlined in the following section. During this period, we have developed a 12 month project schedule for the CYCLE phosphate sensor that includes a pre-commercial build of 5 CYCLE phosphate sensors, a rigorous testing period, including field evaluations by our project partners, and finally release of the CYCLE phosphate sensor to production (commercial product). In early 2009 we are planning on holding a project partner kick-off meeting hosted by WET Labs, where we will demonstrate and train the partners on the use and operation of the CYCLE phosphate system, discuss deployment and testing activities (including issues of sensor integration and communication), as well as discuss the long-term testing and evaluation plans associated with the CYCLE ammonium sensor development.

In separate short paragraphs, Please: 1) describe your proposed scientific and/or technical approach including data quality requirements as applicable, 2) identify the key individuals participating in this work at your own or other organizations and the roles they play and 3) describe your work plans for the upcoming year (if applicable).

WORK COMPLETED

Although we have just begun our three year NOPP project (< 90 days into the first year of funding), we have already accomplished several tasks. These include:

- Assembled the engineering team, assigned roles, held kick-off meeting, and developed a detailed project schedule for the WET Labs personnel.
- Produced a detailed description and specification for the CYCLE phosphate sensor (External Reference Specification)
- Worked with a micropump vendor to modify existing micropumps to specifically work with the CYCLE phosphate system. The vendor has agreed to make these pups, and has integrated them into their existing product line. We have also set up a blanket order with the vendor to supply for the next year to ensure adequate and timely supply of these pumps for the CYCLE phosphate product release in the next year.
- We have redesigned the electronics for the CYCLE phosphate sensor to enhance the capabilities and functionality of the sensor, and to reduce the cost of the components, thereby making increasing the manufacturability for production. The first run of the production level PCBs have been completed and are undergoing performance testing currently.
- Firmware/Software: added features, to increase functionality and reliability. These include on board phosphate calculation, interruptible, two way communications during any period of the measurement cycle, programmable and adjustable standard measurement cycles,

- Flow cell modifications and testing to reduce the impact of bubbles within the system and to aid in efficient flow of sample through the fluidic components.
- We have conducted several tests to improve the onboard phosphate calculation and to reduce the measurement period, thereby extending the deployment duration of the sensor.
- Several mechanical design improvements have been made to facilitate easy changing of the prefilter, reduce the number of plumbing connections, and produced several potential reagent cartridge prototype designs.
- Initiated a postdoctoral candidate search to aid in the development of the chemical analytical techniques, specifically the ammonium sensing methodologies.

In the initial 3 months of the project, we have focused on the engineering efforts requited to transition the CYCLE phosphate sensor from prototype to commercial product. As part of this process, we have initiated a production build of 5 CYCLE phosphate sensors. A gage study will be conducted with these sensors to determine performance metrics. Sensors will then the distributed to the NOPP project team members for field testing and integration with observations systems.

RESULTS

As mentioned above, most of the work performed in the short 3 months of this project have focused on the engineering aspects of transitioning the CYCLE phosphate sensor to a commercial product. The design modifications incorporated into the pre-commercial product version (listed in previous section) have been completed, resulting in a completed bill of materials, machine drawings, and assembly documentation. A CAD rendition of the completed pre-commercial product version is shown in figure 1. The method for the determination of soluble reactive phosphate in natural waters is based on the reaction of phosphate ions with an acidified molybdate reagent to yield a phosphomolybdate complex, which is then reduced with ascorbic acid to a highly colored blue compound, molybdenum blue. The sensor uses four micropumps that deliver the two reagents (ascorbic acid and acidified molybdate), ambient water, and a phosphate standard. The flow system incorporates an integrated pump manifold and fluidics housing that includes a controller and mixing assemblies virtually immune to bubble interference. Our efforts have led to a four to one volumetric reduction in total reagent per sample required. A miniature reflective tube absorption meter measures the color change associated with variable phosphate concentrations. A substantial decrease in reaction time is also accomplished, thus battery power and reagent supply are sufficient for 1000 samples without recharging reagents.

The newly designed electronics for the CYCLE phosphate sensor have resulted in a 30% part count reduction (increasing manufacturability and decreasing cost), as well as provided for improved pump control and metering, ability to interrupt the sample cycle, and enable two way communications (retrieval of data and samples) during a measurement sequence. The electronics for the CYCLE contain 3 boards (PCBs); microprocessor and data storage, analog/amplification board, and a pump and power control board. These boards have been fabricated and are currently undergoing testing and integration with our existing CYCLE prototypes.

Firmware and host software development is also ongoing. The primary development activities in our firmware development have been focused on the on board calculation of phosphate concentration. We are currently testing and evaluation two separate methods for the calculation, one based on a reaction completion and stabilization and one based on the initial slope of the colorometric reaction. Both have shown initial promise in dilution series testing. In addition to reporting the phosphate concentration, the CYCLE phosphate sensor will store the complete colorometric reaction (absorption) data on board for user retrieval and processing through the host software.

As mentioned above, we are currently building 5 pre-commercial phosphate sensors, to be completed by early February 2009. These systems will undergo a complete gage study to determine performance metrics as well as define the reproducibility of the CYCLE phosphate product. Early next year, we will also host a project team kick-off meeting to train our partners on the use and operation of the CYCLE system as well as identify issues of interoperability with other data systems.

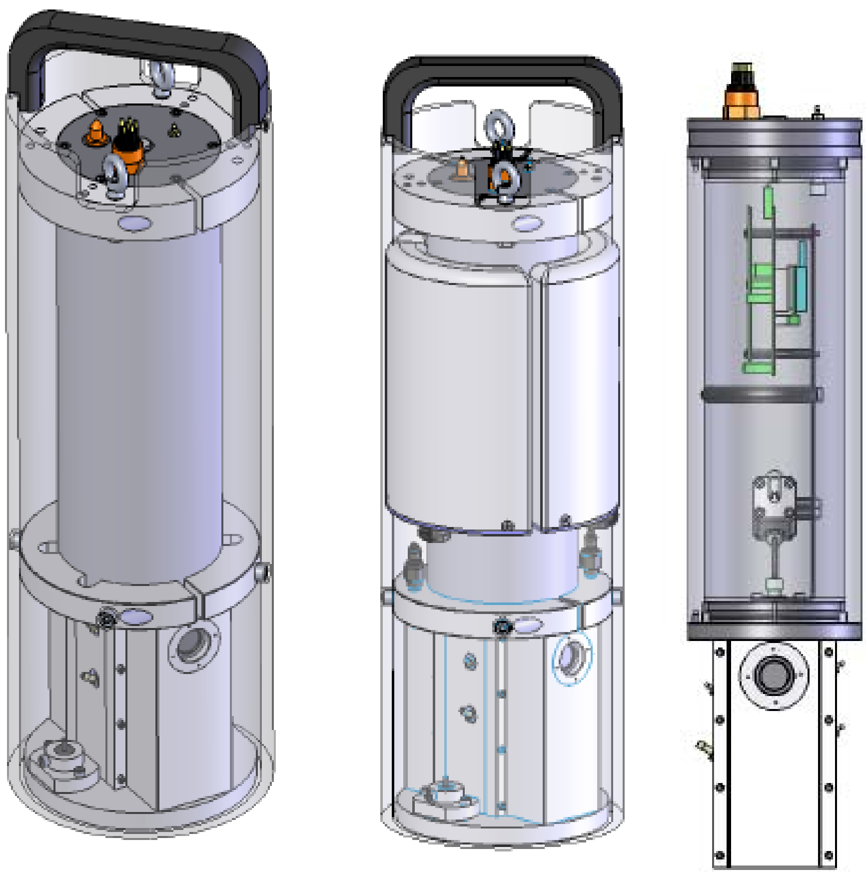

Figure 1 – Drawings of the pre-commercial CYCLE phosphate in situ sensor. Left panel shows the CYCLE sensor without the reagent cartridges. Middle panel shows with the reagent cartridges (upper portion of the body). Right panel shows the CYCLE sensor without the protective housing, and with the electronics housing shown transparent.

IMPACT AND APPLICATIONS

Economic Development

The CYCLE in situ chemical sensor development will result in a new product line for WET Labs. The expectation is that initial sales of the phosphate will result in $500,000 in new revenue over the first year of sales, with sales doubling in the ensuing 1-2 years. As the customer acceptance of the CYCLE product builds, we expect that sales of the product will double within 2 years of release of the product fulfilling the in situ phosphate chemical sensing need in the oceanographic, freshwater, and water quality. As we develop the ammonium sensor, we expect that the chemical analytical skills and manufacturing capabilities will experience further growth, thus resulting in an expansion of our production bas. Finally, this result of this project will establish new technology basis from which to expand and develop new products from.

Quality of Life

Dissolved nutrient dynamics broadly affect issues related to public health, ecosystem status, and resource sustainability, including impacts of climate variability, eutrophication, harmful algal blooms, carbon cycling, and species composition among others. The need for *in situ*, autonomous, real-time nutrient monitoring capabilities has been clearly documented in several national reports on ocean observing. Providing a commercial in situ nutrient sensor to the broader ocean observing, resource management, and freshwater water quality communities that is easy to use, reliable, accurate and long-term deployable, will allow for improved monitoring of public and ecosystem health, coastal resource management, and science research on anthropogenic impacts to be conducted on a routine basis.

Science Education and Communication

Combined with other real time data systems such as the LOBO, the CYCLE phosphate and ammonium sensors will offer the opportunity to learn and teach about nutrient cycles. WET Labs has been involved in an education and outreach project over the past year, through financial and technical support of a marine resource management masters student from Oregon State University, to improve the public's understanding of water quality monitoring in coastal estuaries. As part of this project, the student is building in a module to describe the impact of riverine nutrients on coastal/estuarine ecosystems utilizing data from our water quality monitoring station (http://yaquina.satlantic.com). The addition of a CYCLE to this system will expand the discussion and presentation on nutrients by including real time observations of phosphate concentration.

TRANSITIONS

Economic Development

As mentioned above, the commercialization of the CYCLE phosphate sensor will set WET Labs on a definitive new product pathway focused on in situ nutrient measurement capabilities. This will not only expand our core suite of technologies, but will also facilitate economic growth of the company through market expansion. This in turn will increase our production capacity (through increased staff and production personnel), thus contributing to the economic health and viability of our region.

Quality of Life

As described in this report, several of the partners involved in this project are currently maintaining, or are a part of active resource management, coastal ecosystem health, and/or water quality monitoring programs. By involving these partners as active participants in the development and commercialization process, we will ensure effective transition of the CYCLE phosphate sensor into key local, state, federal monitoring programs as well as ocean observing and science research projects.

RELATED PROJECTS

This project will significantly overlap with the Coastal Margin Ocean Prediction (CMOP) National Science Foundation Science and Technology Center, lead by Dr. Antonio Baptista, Oregon Health and Science University. As part of this project, WET Labs has built and maintained a water quality monitoring station (termed Saturn04) within the lower Columbia River estuary. The monitoring station is providing real time hourly observations of surface temperature, salinity, pressure, chlorophyll concentration, turbidity, dissolved nitrate and dissolved oxygen (http://columbia.loboviz.com). In the next 8 months, a CYCLE phosphate sensor will be integrated with this system, and thus will be providing real time phosphate observations. Through this project, will also be addressing issues of sensor integration/interoperability with the WET Labs WQM (www.wetlabs.com) and Satlantic Inc LOBO (www.satlantic.com) instrumentation systems.