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Workshop Proceedings

TRACE METAL SENSORS FOR COASTAL MONITORING

Seaside, California

April 11-13, 2005

Funded by NOAA's Coastal Services Center through the Alliance for Coastal Technologies (ACT)

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An ACT 2005 Workshop Report

A Workshop of Developers, Deliverers, and Users of Technologies for Monitoring Coastal Environments:

Trace Metal Sensors for Coastal Monitoring

Seaside, California

April 11-13, 2005

Sponsored by the Alliance for Coastal Technologies (ACT) and NOAA's Center for Coastal Ocean Research in the National Ocean Service.

Hosted by ACT Partner organization the Moss Landing Marine Laboratories (MLML) and the Monterey Bay Aquarium Research Institute (MBARI).

ACT is committed to develop an active partnership of technology developers, deliverers, and users within regional, state, and federal environmental management communities to establish a testbed for demonstrating, evaluating, and verifying innovative technologies in monitoring sensors, platforms, and software for use in coastal habitats.

TABLE OF CONTENTS

ACT WORKSHOP: TRACE METAL SENSORS FOR COASTAL MONITORING

EXECUTIVE SUMMARY

The Alliance for Coastal Technologies (ACT) Workshop on **Trace Metal Sensors for Coastal Monitoring** was convened April 11-13, 2005 at the Embassy Suites in Seaside, California with partnership from Moss Landing Marine Laboratories (MLML) and the Monterey Bay Aquarium Research Institute (MBARI).

Trace metals play many important roles in marine ecosystems. Due to their extreme toxicity, the effects of copper, cadmium and certain organo-metallinc compounds (such as tributyl tin and methylmercury) have received much attention. Lately, the sublethal effects of metals on phytoplankton biochemistry, and in some cases the expression of neurotoxins (Domoic acid), have been shown to be important environmental forcing functions determining the composition and gene expression in some groups. More recently the role of iron in controlling phytoplankton growth has led to an understanding of trace metal limitation in coastal systems. Although metals play an important role at many different levels, few technologies exist to provide rapid assessment of metal concentrations or metal speciation in the coastal zone where metal-induced toxicity or potential stimulation of harmful algal blooms, can have major economic impacts. This workshop focused on the state of on-site and *in situ* trace element detection technologies, in terms of what is currently working well and what is needed to effectively inform coastal zone managers, as well as guide adaptive scientific sampling of the coastal zone. Specifically the goals of this workshop were to: 1) summarize current regional requirements and future targets for metal monitoring in freshwater, estuarine and coastal environments; 2) evaluate the current status of metal sensors and possibilities for leveraging emerging technologies for expanding detection limits and target elements; and 3) help identify critical steps needed for and limits to operational deployment of metal sensors as part of routine water quality monitoring efforts.

Following a series of breakout group discussions and overview talks on metal monitoring regulatory issues, analytical techniques and market requirements, workshop participants made several recommendations for steps needed to foster development of *in situ* metal monitoring capacities:

1. Increase scientific and public awareness of metals of environmental and biological concern and their impacts in aquatic environments. Inform scientific and public communities regarding actual levels of trace metals in natural and perturbed systems.

2. Identify multiple use applications (e.g., industrial waste steam and drinking water quality monitoring) to support investments in metal sensor development. Technologies with

broad performance capabilities (e.g., functional in both fresh and saltwater along with ease of deployment customization) were identified as critical design targets.

3. Promote development of *in situ* or field deployable analytical packages for rapid screening and/or pollutant source tracking as well as follow up analytical sensor development. Water quality management efforts would be enhanced by robust affordable user-friendly sensor packages and analytical software that could improve budget allocation for analytical services to support QA/QC requirements.

4. Encourage research and development of new ionophores, metal-specific fluorochromes and ligands which would enable development of metal specific sensors leveraging existing metal detection platforms (e.g., ion selective electrodes, flow injection analysis, biosensors, diffusive gradients thin films).

5. Develop sensors or *in situ* analytical packages for detection of labile metal species. Consensus on a ***standardized definition of this class is critical*** (e.g., free ion activity, vs. inorganic species vs. weak ligand bound). Further refinement and legal standing of water quality criteria requires a clear definition in regards to the connection between labile metal species and biological activity measures.

ALLIANCE FOR COASTAL TECHNOLOGIES

There is widespread agreement that an Integrated Ocean Observing System (IOOS) is required to meet a wide range of the Nation's marine product and information service needs. There also is consensus that the successful implementation of the IOOS will require parallel efforts in instrument development and validation and improvements to technology so that promising new technology will be available to make the transition from research/development to operational status when needed. Thus, the Alliance for Coastal Technologies (ACT) was established as a NOAA-funded partnership of research institutions, state and regional resource managers, and private sector companies interested in developing and applying sensor and sensor platform technologies for monitoring and studying coastal systems. ACT has been designed to serve as:

- An unbiased, third-party testbed for evaluating new and developing coastal sensor and sensor platform technologies,
- A comprehensive data and information clearinghouse on coastal technologies, and
- A forum for capacity building through a series of annual workshops and seminars on specific technologies or topics.

The ACT workshops are designed to aid resource managers, coastal scientists, and private sector companies by identifying and discussing the current status, standardization, potential advancements, and obstacles in the development and use of new sensors and sensor platforms for monitoring, studying, and predicting the state of coastal waters. The workshop goals are to both help build consensus on the steps needed to develop and adopt useful tools while also facilitating the critical communications between the various groups of technology developers, manufacturers, and users.

ACT Workshop Reports are summaries of the discussions that take place between participants during the workshops. The reports also emphasize advantages and limitations of current technologies while making recommendations for both ACT and the broader community on the steps needed for technology advancement in the particular topic area. Workshop organizers draft the individual reports with input from workshop participants.

ACT is committed to exploring the application of new technologies for monitoring coastal ecosystem and studying environmental stressors that are increasingly prevalent worldwide. For more information, please visit http://www.act-us.info/.

ACT, Headquartered at the UMCES Chesapeake Biological Laboratory, has eight Partner Institutions around the country that provide a variety of habitats and a range of technical expertise for testing sensor sensor/platforms for use in coastal observing systems.

The Stakeholder Council provides input into ACT priorities from private sector companies and resource managers involved in sensor technology development and use.

The regional Alliance Member Chapters organized by each ACT partner assures input from the broader coastal observing community stakeholders.

GOALS FOR THE WORKSHOP

The workshop was designed to:

1) Summarize current regional and national requirements and future targets for metal monitoring in freshwater, estuarine and coastal environments;

2) Evaluate the current status of metal sensors and possibilities for leveraging emerging technologies for expanding detection limits and target elements; and

3) Help identify critical steps needed for establishing limits for the operational deployment of metal sensors as part of routine water quality monitoring efforts.

WORKSHOP STRUCTURE

The Alliance for Coastal Technologies (ACT) Workshop on **Trace Metal Sensors for Coastal Monitoring** was convened April 11-13, 2005 at the Embassy Suites in Seaside, California. The workshop was sponsored by ACT-Pacific Coast headquartered at MLML and maintaining a collaborative partnership with MBARI. Invited participants were selected to include equal representation from three population segments concerned with water resource quality and included individual from academic research institutes, private sector companies and local, state and federal resource managers. An opening reception was held for participants the first evening, and G. Jason Smith, Technical Coordinator for the ACT-Pacific Coast Chapter provided an introduction to the workshop and programmatic overview of the national ACT program.

The following morning, during the opening plenary session Co-Chairs, Drs. Al Hanson and Kenneth Coale, provided an overview of the workshop goals. This introduction was followed by three plenary talks (Appendix A) to set the stage for subsequent breakout session discussions. Dr. Bobbye Smith, Regional Science Liaison to the USEPA Office of Research and Development (ORD) for Region 9, provided an overview of EPA ORD's organization and research focus areas as well as funding opportunities available through USEPA's extramural research programs STAR (Science To Achieve Results) and SBIR (Small Business Innovation Research). While no programs directly target funding for aquatic metal sensor development, several programmatic technology needs were identified that may offer basic funding opportunities. Dr. Samuel Kounaves, Tufts University Department of Chemistry, provided an overview of the history and development future for *in situ* electrochemical sensors, providing much needed background for the participants inexperienced in trace metal analysis. The session's final plenary talk was provided by Dr. Al Hanson who discussed private sector interests in trace metal sensor development. This emphasized the critical distinction between chemical **sensors** where the analyte is measured directly without sample processing and chemical **analytical systems** which incorporate a sample processing stream on board, and the associated R&D costs for commercialization of *in situ* instrumentation.

For the remainder of the day two breakout working group discussion and summary sessions focused on the role of *in situ* metal sensor systems for aquatic monitoring programs. After the working sessions, a tour and dinner was hosted at MLML. Dr. George Luther, University of Delaware, College of Marine Studies, provided an informative and entertaining after dinner talk summarizing real world deployments of *in situ* voltammetric trace metal analysis in both terrestrial and deep sea extreme environments. The data provided certainly indicated that a robust and proven technology base is available for routine on site if not *in situ* metal speciation analysis.

A final plenary session was convened the following morning at the Embassy Suites. The session was opened with a talk by Bridget Hoover, Monterey Bay National Marine Sanctuary, who described the structure, training efforts and monitoring objectives of the Citizens Watershed Monitoring Network program. This program represents an example of expanding nationwide efforts to provide high quality regional WQ snapshots based completely on volunteer sampling efforts. These types of programs can potentially provide public demonstration opportunities for emerging onsite metal analysis. The remainder of the final working session was spent openly discussing the consensus recommendations derived from the working group discussions.

OVERVIEW

THE IMPORTANCE OF AND CHALLENGES FOR METAL MONITORING IN WATER QUALITY PROGRAMS

Metal elements are ubiquitous and diverse components of the earth's geochemistry and play critical roles in ecosystem function. While several metals (e.g., Cu, Co, Fe, Mn, Mo, Zn) are essential micronutrients at sub-nanomolar concentrations, some (e.g., Cu, Zn) at higher concentrations can become potent toxins to a range of biological systems. Metals are highly active in redox chemistry, contributing to their dual biological function, but this inherent reactive character also presents significant challenges to the measurement of biologically active species and interpretation of bulk metal concentration data. Complexation reactions control the bioavailability of metal cations:

$$\mathbf{M^{+n} + L^{-n} \Leftrightarrow ML} \quad \textit{and} \quad \mathbf{[M]_{tot} = M^{+n} + ML}$$

where $\mathbf{M^{+n}}$ represents the free metal species of interest and $\mathbf{L^{-n}}$ represents a complexing agent (dissolved inorganic anions, oxides, dissolved organics, detrital surfaces, mineral particles). The study of environmental metal distributions becomes even more daunting when one considers biologically catalyzed covalent modifications and metallo-organic compounds produced by human industrial activities (e.g., methyl mercury and tributyl tin). Indeed, even the act of sampling can induce rapid transitions in metal species distributions (see for example, Nuzzio et al, 2002) and at minimum requires simultaneous measurement of ambient physiochemical properties (pH, salinity, DO, temperature, alkalinity) for rigorous interpretation of offsite measures. For these reasons metals represent an important target for development of *in situ* analytical tools.

The distribution of metals in the environment has changed continuously since their routine adoption into human society and even more dramatically with global spread of industrialization. The changes in metal availability have had pronounced effects on plant and animal distributions. In large part, they are determined by the organisms' capacity to acclimate to either limiting or superabundant metal availabilities. Water quality and underlying sediment habitat quality have also been significantly impacted by the anthropogenic changes in metal distributions. The U.S. Clean Water Act (CWA) and similar international regulations mandate federal and state agencies to develop programs to protect the chemical, physical and biological integrity of the nation's waters. To date, the USEPA has published (EPA-8230R-03-010 2003) 165 water quality criteria. Of these 165 criteria, 10% define upper limits to the specific metal content of fresh and saltwater habitats. Recommended metal criteria maximum concentrations, above which which significant negative biological effects may occur, all fall in the nanomolar range of the total recoverable metal fraction (Table 1). Twelve percent (12%) of all documented water quality impairments in 2002 were attributed to metal contents above the recommended criteria level, indicating that metal contamination of water resources is a nation-wide problem. While these WQ criteria guidelines are designed to minimize negative impacts to aquatic ecosystems and water resource users, they represent derived proxies for metal biogeochemistry and provide no assessment of the contribution of metal speciation to their vital or toxic biological activity.

TABLE 1. USEPA Recommended Water Quality Criteria (RWQCs) for priority toxic metal pollutants and non-priority metals. Priority metals are indicated in **bold**. Values represent total dissolved concentrations below which no significant detriment to aquatic life is predicted. Source: USEPA-8230R-03-010 2003. Values converted to nanomolar (nM) to facilitate comparison with method detection limits. Assumes 100 mg $CaCO_3$ /L hardness for freshwater.

			Freshwater		***Saltwater***	
Element	***Symbol***	***Atomic Wt***	***CMC***[a]	***CCC***[b]	***CMC***	***CCC***
Arsenic	**As**	74.92	4538.2	2002.1	921.0	480.5
Cadmium	**Cd**	112.40	17.8	2.2	355.9	78.3
Chromium (VI)	**Cr**	52.00	307.7	211.5	21153.8	961.5
Copper	**Cu**	63.55	204.6	141.6	75.5	48.8
Lead	**Pb**	207.20	313.7	12.1	1013.5	39.1
Mercury	**Hg**	200.60	7.0	3.8	9.0	4.7
Nickel	**Ni**	58.69	8008.2	886.0	1260.9	139.7
Selenium	**Se**	78.96		63.3	3672.7	899.2
Silver	**Ag**	107.90	29.7		17.6	
Zinc	**Zn**	65.39	1835.1	1835.1	1376.4	1238.7
Aluminum	*Al*	26.98	27798.4	3224.6		
Iron	*Fe*	55.85		17905.1		
Manganese	*Mn*	54.94				

[a] ***CMC***: criteria maximum concentration, acute exposure acceptable limit. [b] ***CCC***: criterion continuous concentration - maximum acceptable chronic exposure concentrations.

The implementation of such metals criteria is not straightforward due to the site-specific nature of metals toxicity in aquatic environments. Compounding the challenges to metal monitoring imposed by their inherent chemical properties are the plethora of monitoring goals (i.e., total maximum daily loads (TMDLs), Maximum Daily Effluent Limits (MDEL) wastewater effluent monitoring, compliance and ambient monitoring), many of which are expressed as site-specific objectives, criteria (total recoverable versus total dissolved), sampling constraints (clean technique, volume and frequency) and ex situ analytical turnaround. Furthermore regional variation in metal contaminant loads in underlying sediments may contribute to continued WQ degradation even when direct discharges have been mitigated. While powerful analytical tools are available for high precision quantification of metal ion content in environmental samples (e.g., graphite furnace atomic absorption spectroscopy, GFAA; inductively coupled plasma mass spectroscopy, ICP-MS; atomic emission spectrometry, AES; X-ray spectroscopy, neutron activation analysis) their utility in monitoring programs is ultimately constrained by the nature of ex-situ sampling and potential for sample alteration and contamination during handling. While these established technologies offer the advantage of high precision analysis of an array of elements with parts per trillion detection limits, they suffer the critical limitations of high operational costs, low portability and only provide measurement of total metal concentration in the processed sample fraction. Analysis of metal speciation by these methods, a parameter important to the interpretation of biological effects of metal loading, can only be accomplished by coupling upstream separation and extraction procedures, thereby not only adding cost, but also increasing the risk of altering sample composition from the ambient state. The development of *in situ* tools for monitoring ambient metal loads would clearly enhance all aspects of current monitoring efforts.

Technologies Available For In Situ Metal Analysis

Desirable features for *in situ* and on site metal analysis should include the capacity to measure not only total dissolved or extractable metal content, but also metal concentrations within the dynamic or labile species fractions (potentially representing biologically available forms) and free metal ion species (representing the most biologically active form). In order to provide the most accurate assessment of the true chemical state, the targeted technologies should provide analytical results with minimal sample manipulation. It is useful to distinguish technological approaches based on whether they function as sensors, directly measuring the metal analyte of interest or analytical systems which incorporate sensor types into a sample processing stream. In the following, technology base summaries for: Spectroscopic Techniques, Electrochemical Techniques, Voltammetric Techniques, Potentiometric Sensors, Flow Injection Analysis, Diffusive Gradients in Thin Gels (DGT), and Biosensors, are presented in order of standard usage in WQ studies and application history in the field of metal analysis.

Spectroscopic Techniques

While existing spectroscopic techniques (e.g., mass spectrometry, emission spectrometry, atomic absorption spectrometry) offer high precision and sensitive analysis of total metal content orders of magnitude below existing WQ criteria, the portability and infrastructure costs of these analytical systems need to benefit from advances in nanofabrication; ultimately their utility in monitoring metal bioactivity is limited by the requirement to couple these detection systems with additional analytical separation procedures (e.g., chemical modification and extraction, HPLC, ion chromatography). Numerous systems are available through a variety of laboratory equipment vendors.

Electrochemical Techniques
Associated with their compatibility with redox chemistries, metal detection has been a target for development of electrochemical analytical techniques for over 30 years. Two broad analytical approaches have been applied to the problem of metal detection in environmental samples: voltammetric and potentiometric systems. The low power requirements and flexible design with standard electronic components make electrochemistry a flexible and relatively low cost target for field deployable metal analysis systems.

Voltammetric Techniques
Monitor the current (i) flowing between an auxiliary electrode (AE) and a working electrode (WE) resulting from the reduction or oxidation of a metal species at applied potentials (E) supplied to the WE relative to a reference electrode (RE). Depending on the electrode composition and configuration, each metal analyte will have unique i vs. E profiles and the current magnitude is proportional to analyte concentration. Due to the flexibility of this approach, a range of protocols have been developed based on the potential scan rate format (linear sweep, cyclic voltammetry, differential pulse or square wave) and WE configuration (macro- vs. stirring independent microelectrodes ($r < 10\ \mu m$)). Direct reduction methods generally have higher detection limits ($>10^{-8}$ M) than adsorptive methods such as ASV (anodic stripping voltammetry) and AdSV (adsorptive SV), which employ a high WE potential to pre-concentrate the target metal(s) in the mercury film on the WE surface, thereby lowering the detection limits in environmental samples ($< 10^{-12}$ M). While the inclusion of a metal ion specific ligand in AdSV techniques can expand the range of metal analyte species targets, this required addition of exogenous co-analytes will limit its applicability for *in situ* monitoring applications. Electrode sensor design has also been improved for field applications by refinement of techniques for fabrication of microWEs, which eliminate dependence on sample stirring. Further improvements in electrode durability have been achieved by inclusion of agarose gel impregnated surface microelectrodes (GIME), which help to stabilize the Hg films and reduce sensitivity to macromolecular fouling components (Buffle and Tercier-Waeber 2005). While microWEs incorporate small amounts of Hg, acceptance of these systems would be enhanced by further research on development of Hg-free electrodes.

Field deployable voltammetric analytical systems have been developed for biogeochemical research applications in extreme environments (Luther et al, 2001; Nuzzio et al, 2002; see G. Luther presentation). Several voltammetric analytical systems are now commercially available for on-site metal analysis (GAT TEA 4000 MP, www.rudolphinst.com/trace_metal_analyzer.html ; Nano-BandTM Explorer, www.tracedetect.com) and a single system is available with capacity for *in situ* trace element profiling and monitoring in aquatic systems with nanomolar sensitivity (VIP System www.idronaut.it). The VIP System incorporates a sensor package for concurrent measurements of critical ambient WQ parameters of conductivity, temperature and depth (CTD), dissolved oxygen, pH and redox. Thus, enabling more rigorous interpretation of the metal speciation data.

Potentiometric Sensors

Are the simplest analytical systems in that they are comprised of a simple galvanic cell between a WE and RE. The millivolt potential output from the cell is proportional to the analyte of interest as long as the WE electrolyte chamber is separated from the ambient medium by an ion selective (IS) coating. ISE based sensors provide a direct measure of ion activity and may therefore offer the best proxy for bioactivity. However, reliable interpretation of such metal ion activity data requires precise concurrent CTD measurement and their utility in open ocean systems may be limited. Availability of field deployable and *in situ* pH and redox electrodes indicates that robust fabrication technologies are in place to support electrode development. Additionally leveraging technical advances in fabrication of solid state ISE for deployment on interplanetary missions would enable developing compact field deployable ISE arrays for multiple metal ion analysis systems (see S. *Kounaves presentation*). These electrode arrays could also be leveraged as screening to facilitate and support needed research into the synthesis of novel ion-selective coatings for critical metal species, the availability of which is currently the major obstacle limiting the use of ISEs for metal monitoring. Investment in development of new IS coatings could also benefit development of more sensitive and durable voltammetric microelectrodes.

Flow Injection Analysis

Traditional wet chemical analysis has a long history in aquatic science, particularly for monitoring macro and micronutrient dynamics. Advent of continuous flow analysis (CFA) and flow injection analysis (FIA) instrumentation was quickly adopted to promote high resolution profiling and surface mapping (Johnson et al. 2000). FIA based analytical systems are comprised of one to several pumps feeding samples, blanks and reagents to a mixing manifold and subsequent passage of the analytical stream through a flow through detector to measure either absorbance, fluorescence or chemiluminescence of the specific analyte+reagent reaction product. Continuing advances in microfluidics and nanofabrication have enabled further size and power reductions in these design of FIA system components making them compatible not only with on site, but also *in situ* monitoring efforts. Application of FIA technology to metal analysis by the research community has required inclusion of trace metal clean Teflon tubing reagent reservoirs and on board cleaning capacities. Additionally, sensitivity for specific metal species has been improved in oceanic systems by including solid phase preconcentration, long optical pathlength detectors, or incorporation of detection reagents, yielding metal ion specific chemiluminescent or fluorescent products for ambient level detection (10-12 - 10-9 M) of several metals (Fe, Co, Cu, Mn, Zn) influencing biological activity. Alternation of acidifying and non-acidifying process streams into the sample flow may in some cases, be used to distinguish between the labile and total dissolved metal fractions. As with ISE and voltammetric techniques, investment in identification of metal ion-specific detection reagents would enhance the adoption of this technology by the monitoring community. As this approach is reagent dependent, *in situ* or on site deployment intervals will ultimately be limited by reagent storage volumes and stability under field conditions.

At present a FIA system for *in situ* measurement of dissolved iron II is commercially available from SubChem Systems (www.subchem.com).

Diffusive Gradients in Thin Gels (**DGT**)

While not considered metal sensors or analytical systems, diffusive equilibrium based sampling devices provide a low cost means to obtain temporally integrative samples of metal distributions over a range of spatial scales. **DGT** devices are comprised of a metal chelating resin underlying a thin (<1mm) hydrogel film. Loading of metal ions onto the resin is limited by its rate of diffusion through the hydrogel (a function of metal species diffusivity, gel pore size and thickness). For a given deployment time, accumulated metal concentration in the resin will be proportional to the average concentration of the labile metal fraction in the ambient water or sediment, and can be derived using the Fick equation (Davison et al. 2000). Concentration measurements based on diffusive metal accumulation are comparable to direct measurement by ASV representing the dynamic metal fraction. While metal analysis is performed off site generally using spectroscopic methodology, these sampling devices do provide a low cost, sensitive and reliable method to passively obtain multi-element samples. As their deployment does not require expert users, these devices may be ideal to incorporate into regional volunteer WQ monitoring efforts (*see B. Hoover presentation*) and enable much higher spatial and temporal sampling schemes. DGT samplers are commercially available (www.windsor-ltd.co.uk/dgt.html), and are relatively straightforward to construct (Davison et al, 2000; Twiss and Moffett, 2002). It would also appear that specificity of the samplers could also benefit from recommended IS coating developments as well as metal species specific ligands.

Biosensors

This represents a nascent technology area but one with great promise, as it leverages the precise molecular recognition interactions inherent in biological systems and tremendous advances in site directed mutagenesis and *in vitro* protein expression protocols to manipulate binding site characteristics. Sensor development strategies include manipulating native metal binding site properties to alter metal selectivity or developing molecular mimics of binding site structure. Transduction of metal-macromolecule interactions can be achieved by reporters responding to metal-induced conformational changes via quenching or enhancement of site-specific fluorescent or luminescent signals. Metaloproteins provide obvious targets for metal biosensor development. Zinc-finger proteins or their consensus metal binding motifs and human carbonic anhydrase have provided highly selective binding sites with picomolar affinities for Zn(II), Cu(II) and low interference from abundant Mg(II) and Ca(II) cations in biological or environmental fluids (Thompson et al. 1999). Site directed covalent attachment of a variety of fluorophores near the native metal binding sites can yield a signal transducing system yielding metal concentration enhanced or quenched fluorescence intensity, fluorescence lifetime shifts and polarization shifts depending upon the metal dye-combination and molecular proximity. These molecular transduction signals report free metal ion binding and provide a means to measure free metal ion concentrations *in situ*, a parameter generally beyond the detection limits of current analytical techniques in absence of preconcentration. Use of time resolved fluorescence signals to monitor bind site activity makes this sensor design compatible with fiber optic measurement systems, enabling spatially precise free metal ion measurements in near real time. In terms of cross-over into WQ monitoring applications, proof of concept for the human carbonic anhydrase based fiber optic sensor has been reported for picomolar detection of cupric ion (Cu(II)) in natural seawater (Zheng *et al*. 2003). Current technological impediments to operational deployment of these true biosensors relate to slower metal ion off rates and either denaturation or poisoning / fouling of binding site capacity by dissolved seawater components; this may require future sensor designs to incorporate flushing and cleaning/chelating functionality in the analytical system. Again coupling this sensor transduction base with IS coating may offer *in situ* performance enhancements.

SUMMARY OF BREAKOUT GROUP DISCUSSIONS

Breakout Session I.
This breakout session sought to develop sector-specific viewpoints on the following aspects of *in situ* metal monitoring:

I. **WHAT ARE THE CURRENT MONITORING NEEDS *(target prioritization, regulatory metrics)?***

II. **WHAT IS THE CURRENT STATUS OF METAL SENSOR DEVELOPMENT FOR APPLICATION IN ENVIRONMENTAL MONITORING *(strength and weaknesses regarding application to coastal environmental research, monitoring and management)?***

Management Viewpoint: Group Chair, Michael Lyons; Rapporteur, Keith Maruya

Participants representing the management sector felt that from a regulatory perspective the following metals should be priority targets for development of *in situ* or on site analytical systems: Hg, Se, As, Cu, Pb, Ag, Cr, Cd, Ni. These elements already have defined WQ criteria goals and impact water resources globally (Table 1). In terms of development of new metal sensor technologies, this group felt that desirable performance features of such analytical systems should include the capacity to measure not only total metal concentrations, but also provide information on metal speciation. Identification of the proportion of total metal content present in the labile or dynamic dissolved metal fraction which would serve as a better proxy for bioactivity than the current total dissolved concentration measures targeted by WQ criteria. Desirable performance features would include sub- parts per billion (ppb) limits of detection, functionality in a range of aquatic environments (i.e., freshwater to marine). The participants felt that availability of portable, low cost and rugged on site analytical systems would greatly enhance their WQ monitoring efforts by enabling them to increase temporal sampling frequency and spatial resolution. A critical point for adoption of new technologies into their monitoring programs would be whether they enabled more efficient allocation of resources for targeted lab-based metal analysis required to support regulatory decisions regarding compliance (e.g., TMDLs and MDELs), drinking water supply monitoring, waste processing management, storm water source tracking and ambient recreational water quality.

Research Viewpoint: Group Chair, Zanna Chase; Rapporteur, Maeve Lohan

Participants representing the academic research and development sector are motivated to adopt and/or design new tools to help them understand and elucidate the biogeochemistry of metals in natural and perturbed systems. They felt that at present there is a general lack of public awareness as to the importance of metals in ecosystem function and in particular what metal ion species exhibit bioactivity. As such, their priority monitoring needs for which *in situ* sensor systems would be useful were identified as: the study of benthic boundary layer processes (sediments are important metal source/sinks and can influence overlying WQ, e.g., Sundby et al, 2005), developing capacity for event response (storm runoff, dredging and erosion effects on metal mobilization), compatibility with adaptive sampling programs to enhance spatial and temporal resolution of metal dynamics and continuous near real time monitoring capacities in order to study biotic and abiotic effects on metal dynamics. This group recognized substantial differences in the regulatory compared to science monitoring needs, but felt that developing instrumentation with inherently low limits of detection would benefit both applications. Capacities for robust and reliable measurement of free ion activities was essential for demonstrating their bioactivity, and ultimately increasing reliable databases of this information could be leveraged to influence development of future regulatory targets and standard methods of metal analysis. Ideally, new generations of analytical systems will provide simple, cost effective, portable and secure analysis of total, labile and free ion concentrations of targeted dissolved metals. Multi-element detection capabilities (e.g. Luther et al. 1998) would also enhance biogeochemical research efforts. Concurrent measurement of ambient conditions (temperature, conductivity, pressure, pH, dissolved oxygen, redox) was viewed as necessary to fully interpret the habitat-specific control of dissolved metal speciation processes. This information could also be used to support development of improved computation chemistry based modeling of metal toxicity.

Private Sector Viewpoint: Group Chair, Tom Mitchell; Rapporteur, Andrea Zappe

Representatives from companies developing and marketing environmental monitoring equipment focused their discussion on identification of market drivers required to direct investment in research and development towards field deployable trace metal analytical systems. Ultimately potential market size will determine their risk to R&D investment. For small companies, the group felt that an identified market of at least $1M was needed. Larger companies, with capacity to enter into OEM agreements with smaller companies, would require an identified market of at least $3M. Market definition will be dependent upon identification of priority metal species and governmentally mandated WQ criteria goals for those metal species. It was recognized that a disconnect exists between mandated goals and detection levels for metal species of interest to the research community and that limited investment may be available to support a bottom up influence on modifying regulatory guidelines. Additionally streamlined pathways for approval of new methodology would spur development.

The group also identified possible development times for the analytical systems discussed during the plenary sessions. Near horizon targets (present to 3 years) for commercial development of field deployable analytical systems include both voltammetric and flow injection analyzers.

Medium term (5-10 years) technologies would include robust microelectrode voltametric systems, ion selective electrodes or electrode arrays, fluorescent biosensors and microfluidic based FIA systems. Long term (>10years) development targets would include nanotechnology based sensor systems like MEMS (microelectromechanical systems), lab chip based chemical systems (see ACT Workshop report WR04-03: Developing Technologies for Environmental Micro-Chemical Sensors) and micro-total analytical systems (Buffle and Tercier-Waeber 2005). As a means to facilitate this development cycle, the group formulated the following table (Table 2) summarizing information required to peak industries' interest in R&D investment in metal sensor development. It was also suggested that this could be incorporated into an ACT-sponsored user survey.

Parameter	***Provided by Company***	***Provided by End Users***
Target Metal(s) & Species		¤
Detection range for each		¤
Limit of Detection		¤
Response Time		¤
Sampling Frequency		¤
Minimum sample volume		¤
Standard Method		¤
Deployment time		¤
Power Options	¤	¤
Telemetry Options	¤	¤
Physical Constraints		¤
Ancillary Sensors		¤
Acceptable Unit Cost		¤
Acceptable Cost per Sample	¤	¤
QA/QC constraints (reagen ts, samples, calibrations)	¤	¤
Operator Training Level	¤	¤
Market Size	¤	
Internal Development Cost to Commercialize	¤	
Cost of Goods Solds	¤	
Certification Requirements (e.g. EPA)	¤	
Ancillary Applications (e.g. Homeland Security)	¤	

TABLE 2: Information needed to support R&D investment decisions by the private sector for development of field compatible trace metal sensors.

Breakout Session #2

Cross-sector groupings of the participants were formed to discuss prospects and barriers to adoption of field compatible trace metal analytical systems as framed by the following questions:

III. WHAT ARE THE MAJOR LIMITATIONS TO CURRENT AND FUTURE APPLICATIONS OF *IN SITU* METAL SENSORS? ARE THERE ALTERNATE TECHNOLOGIES THAT COULD BE LEVERAGED FOR THIS PURPOSE?

Group Chairs, Eric Crecelius, Bob Byrne, Drew Sweetak
Rapporteurs: Michael Callahan, James Downing, Ian Walsh

Plenary session reports from the second breakout session revealed consensus on several obstacles to the adoption of new technologies facilitating high resolution monitoring of trace metals in aquatic environments. First, the group strongly felt that low societal awareness of the importance of metal speciation to biological systems needs to be overcome, mainly by increased communication within the scientific community and regulatory agencies. This is a critical issue since development of regulatory standards is not based on scientific study alone. Public demand also has a roll in this process. Increasing investment in outreach activities from expert scientific groups like GeoTraces (www.geotraces.org) could be used to enhance public awareness of not only the toxic, but also the vital functions of metals in the earth ecosystem. Investment in additional targeted studies of metal speciation behavior, directly coupled to multiple level ecological assessments (i.e., microbes, phytoplankton and higher trophic levels) would help strengthen the database, identifying free and liable metal fractions as the bioactive forms (including positive and negative effects). This information is critical to disseminate to regulatory decision makers governing the adoption of new analytical technologies into compliance monitoring programs. Without the impetus of regulatory guidelines and standards, as well as acceptance of methods, the likelihood of extensive investment in new R&D for commercialization by the private sector is low. An alternative strategy would be to identify cross-over uses for the technology such as biomedical diagnostics and industrial process control and waste processing.

A second related obstacle is that the analytical approaches developed and employed by the scientific community are highly customized, difficult to use and rarely cross-standardized; this in part is evidenced by the plethora of voltammetric techniques in practice. Technology demonstrations could serve in part to overcome these misperceptions. Incorporation of some these analytical approaches into regional citizen WQ monitoring events (e.g. MBNMS Citizen Watershed Monitoring Network) would serve the dual function of increasing public awareness and perhaps widespread adoption.

Additionally, development of robust and user friendly instrument control and data analysis algorithms would facilitate the transfer of these technologies to operational use. Related to the issue of customization, is the fact that few of the available technologies are multi-element capable without sacrificing analytical precision and cannot analyze critical elements like Hg. Promoting advancements in analytical chemistry for the synthesis of novel metal-species specific ligands, ion selective coatings and fluorometric reporters could find wide application in development of multianalyte systems.

These vigorous discussions led to development of a series of recommendations that the group felt were needed to facilitate continued development of field deployable metal analysis tools. These recommendations were subsequently prioritized by the results of workshop polling.

WORKSHOP RECOMMENDATIONS

The enthusiastic and rigorous discussions among the workshop participants led to their development of the following top ten recommendations that they as a community, felt would help promote the regulatory acceptance and commercialization of tools for *in situ* metal analysis.

1. Increase scientific and public awareness of metals of environmental and biological concern, impacts in aquatic environments, and the distinction between biologically useful and toxic concentrations.

2. Identify multiple use applications (e.g. industrial waste steam and drinking water quality monitoring) to support investments in metal sensor development. Technologies with broad performance capabilities (e.g. functional in both fresh and saltwater along with ease of deployment customization) were identified as critical design targets.

3. Promote development of *in situ* or field deployable analytical packages for rapid screening and/or pollutant source tracking as well as follow up analytical sensor development. Water quality management efforts would be enhanced by robust affordable user-friendly sensor packages and analytical software that could improve budget allocation for analytical services to support QA/QC requirements.

4. Encourage research and development of new ionophores, metal-specific fluorochromes and ligands, which would enhance development of sensors for additional metal-species by leveraging existing metal detection platforms (e.g., ion selective electrodes, flow injection analysis, biosensors, diffusive gradients thin films).

5. Develop sensors or *in situ* analytical packages for detection of labile metal species. Consensus on a ***standardized definition of this class is critical*** (e.g. free ion activity, vs. inorganic species vs. weak ligand bound). Further refinement and legal standing of water quality criteria requires clear definition of connection between labile metal species and biological activity measures.

6. Define ancillary environmental measurements (e.g. pH, temp, salinity, dissolved O2, redox, alkalinity/hardness, TSS, CDOM) required to facilitate accurate interpretation of

in situ metal speciation data sets, and couple these with computational toxicology tools to help describe metal bioactivity.

7. Development of particulate phase, total, and pore water detection capabilities also should be continued.

8. Robust and consistent data processing (including QA/QC) algorithm development needs to continue, and would enhance marketability of all *in situ* sensor packages.

9. Work with regulators to accept developing standards for metal detection.

POTENTIAL EFFORTS FOR PROGRAMS, INCLUDING ACT TO CONSIDER

Participants discussed how programs, including ACT, could best support future efforts towards continued development and refinement of existing trace metal sensor technologies. It was suggested that they:

1. Provide a forum to increase awareness of metal issues in environment.

2. Help develop a database of metal related "smoking guns" or compelling scientific evidence of metal effects to leverage funding agency and investment support. This could be hosted on the ACT TMS Discussion Website Forum.

3. Support an assessment of science (time / space scales / speciation / targets) and management needs (source /sink tracking, sampling frequency, regulatory metrics) for metal monitoring capabilities. Help identify research and educational opportunities at the environmental chemistry / oceanographic interface.

4. Facilitate continued discussions among management, industry, & scientists to develop sensor requirements and performance specifications and deployment limitations (e.g. LODs and WQ criteria for freshwater and saltwater).

5. Assess user needs for on site metal analysis (expand user survey formate derived from Nutrient sensor model).

6. Maintain a list of sensor developers and source suppliers.

7. Develop and maintain a list of funding opportunities targeting sensor development.

CONCLUSIONS

There was a general agreement among the workshop participants that a variety of robust, sensitive and field compatible techniques are in place to enable at minimum, on site if not *in situ* analysis of metal speciation in a variety of aquatic environments. Future commercialization of these technologies was viewed as largely market, rather than base technology limited. This market limitation is based in part on the lack of societal awareness and even apathy regarding the significance of metal biogeochemistry to ecosystem function. Sound science coupled with public outreach describing the habitat specific nature and influence of dissolved metal speciation on biological processes is encouraged. In particular, a cross technology standardized definition of bioactive labile metals needs to be developed to facilitate acceptance of speciation measurements into regulatory criteria. Additional R&D investment may be encouraged by identifying cross-market applications (e.g. biomedical, industrial process control) of these analytical techniques.

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APPENDIX A: PLENARY SESSION TALKS

Speaker	**Topic**
Bobbye Smith	***US EPA's Research Areas for Metals in Aquatic Systems***
Sam Kounaves	***Electrochemical Sensors Past and Present***
Al Hanson	***Private Sector Interests in Metal Sensors***
George Luther	***Applications of in situ Voltammetry for Metal Measurements: Sediments to Vents***
Bridget Hoover	***Citizen Monitoring: Real Science, Real People, Real Results***

Copies of the presentation files are available upon request from jsmith@mlml.calstate.edu or the ACT Trace Metal Workshop Discussion forum at www.act-us.info/forum

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