# Sensor Technology for Remote, Interactive Aquatic Experiments Workshop and Special Session

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## ASLO Copenhagen Meeting

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## WORKSHOP REPORT

The coming decades will witness a rapid growth in moored, cabled, and autonomous observatories to investigate a spectrum of basic processes in aquatic environments. In anticipation of the need to develop or re-engineer sensors to measure physical, chemical, biological, and geological processes *in situ*, a one-day workshop and special session on sensor technology was held during the June meeting of the American Society of Limnology and Oceanography (ASLO) in Copenhagen, Denmark. The goal of the workshop was to exchange ideas on new experimental approaches and methodology, to define strategic themes, and to formulate specific recommendations related to sensor development. The 25 participants from North America and Europe represented academic and industry sensor developers and users, as well as a broad spectrum of scientific interests. Herein are the recommendations resulting from that meeting in hope that they will be useful as a catalyst for further development of sensor systems.

Oceans, lakes, rivers, and groundwater are complex, dynamic environments in which physical, chemical, and biological processes occur over varying temporal and spatial scales (e.g., eddies, nutrient fluxes, patchiness of organisms, benthic processes, and pollution). In addition, deep, remote, or hostile systems, such as hydrothermal vents and polar regions, traditionally are poorly sampled, but are important to understanding global biogeochemical and hydrological cycles. These cycles are critical to maintaining Earth's habitability. The new insights on marine and freshwater processes obtained from satellite remote sensing and time-series *in situ* sensors underscore the scientific and societal need for an improved synoptic view of aquatic systems.

There was consensus among the workshop participants that development and validation of chemical and biological sensors were urgently needed. Lack of inexpensive and reliable sensors generally limit chemical and biological observations. For example, 3,000 profiling floats will be deployed as part of the Argo Program (www.argo.ucsd.edu) to monitor global changes in ocean temperature and salinity as part of a climate observing system. The inability of biogeochemists to utilize these floats was perceived as a tremendous missed opportunity to link physical, chemical, and biological processes to climate variability.

Information exchange is important to ensure that the development and use of sensors will progress efficiently. The primary recommendation was that workshops involving scientists, engineers, and technologists were essential to foster information exchange and to provide advice on community priorities for sensor development. More than one workshop would be warranted because of the specialized needs of different habitats and the varying research focus of different scientific programs. A coordinating committee could be beneficial for tracking the common themes among these groups and finalizing cross-cutting recommendations in a document for funding agencies, sensor developers, and user groups. There also was a consensus that some areas of sensor development/use required community agreement (e.g., hardware/software compatibility issues, precision issues, calibration standards) and that other areas needed strong encouragement for continued development (e.g., $O_2$ sensors, profiling moorings).

The first working group recommended the following criteria to prioritize the chemical and biological sensors needed to address fundamental science questions during the next decade.

- Sensors that are now operational, but could be better utilized.
- Individual sensors or suite of sensors that require additional development.
- Sensors that need to be developed.

Some sensors, for example $pCO_2$, pH, nitrate, fluorometers, and spectral radiometers, are currently operational on moorings, but long deployments may be limited by biofouling. Biofouling came up repeatedly as a problem that must be resolved. A combination of optical (i.e., absorption, transmissometers, and fluorometers), $O_2$, and $pCO_2$ sensors was given as an example of a suite of sensors that would be useful to address a broad array of questions related to aquatic productivity and biogeochemical cycles. However, instrumental drift of $O_2$ sensors in marine systems was a concern. The wish list for new sensors was as long as the number of participants. Examples of chemical sensors that must be developed included particulate and dissolved organic carbon, nitrogen, and phosphorus, phosphate and acetate, and sensors for speciation of elements. The need for robust, stable sensors at extreme temperatures was discussed. The development of sensors for microbial activity also was strongly endorsed. Our understanding of microbial ecology is far behind all other biota. Recent developments in microfabrication provide the foundation for developing high-density arrays of biologically-based detection elements (e.g., nucleic acid, enzymatic, or immunochemical). For example, DNA microarrays could be used to monitor both abundance and activity level variations among natural microbial populations.

Participants noted that the accuracy, precision, and interpretation of sensor data must be improved. They recommended that calibration protocols be developed for all sensors, especially *in situ* calibrations, that standards for calibrations of sensors and analyzers be developed and maintained, and that training workshops should be encouraged to provide instruction on the proper use of equipment. Workshop participants noted the success of the global ocean carbon dioxide survey was enabled by the development of easily distributed standards for total inorganic carbon. Interpreting the carbon data, however, has proven problematic due to the lack of similar standards for nutrients. Biological sensors have suffered from a lack of rigorous field validation and must be accorded sufficient funding to complete this essential development phase. Too often biosensor validation has been done in an *ad hoc* fashion during field research, resulting in a lack of confidence in data interpretation.

The second working group discussed the problem of moving from prototype sensors to mass production. The example of the TAO/Triton mooring array across the Tropical Pacific Ocean was used as a focus for the discussion. About 70 ATLAS and TRITON moorings, with physical sensors at 11 depths, telemeter oceanographic and meteorological data to shore in realtime via the Argos satellite system. The chemical and biological oceanography communities must develop strategies to deploy a comparable number of sensors in order to achieve a similar synoptic coverage. In addition to conceptual hurdles, sensor development and mass production was viewed as being limited by funding, lack of a trained workforce (users and repair), poor long-term stability and reliability of sensors, and inadequate follow-up on calibration and data quality control. It was clear that community acceptance of a sensor technique was necessary before mass production could occur.

Several directions for broadening the application and use of sensors were considered. Sensor designs could be simplified so that non-experts can use them. Sacrificing precision should be evaluated in terms of the process being measured and whether it increases instrument reliability or reduces the level of expertise needed to maintain the instrument. Alternatively, sensor designs could be made more complex, whereby an intelligent sensor would perform the function of the expert technician. Smart sensors also could be designed to detect natural scales of variability and respond in some pre-programmed way to collect data more intensively during or near the phenomenon of interest. Smart sensors would be easier to transport to different environments that operate on different scales of variability (e.g., hydrothermal vents, freshwater, and sediments). Dedicated scientific/engineering centers were suggested for intensive development of certain sensors and to facilitate the broad use, validation, and mass production of sensors. Cooperation between scientists and industrial partners should be encouraged for the final development. Finally, there must be a broad effort to inform and train users to interpret results. Support groups should be set up to provide advice to all users.

The second working group also discussed problems associated with hardware and software compatibility, the so-called “Plug-N-Play” issue. Everyone agreed that this problem continued to be a tremendous time- and money-consuming challenge. The most flexible instrument drivers utilize low-level C programming language. Investigators wishing to combine observations from multiple instruments are forced to either limit their sampling options to those supported by pre-programmed drivers or invest significant time and resources into electronic and software programming themselves. Mutual compatibility is an increasingly difficult problem as serial instruments are each programmed and interrogated separately. This is a community problem that could benefit from standardization of power and communication, while recognizing that power requirements and data output rates vary among sensors. One solution suggested was the use of Master-Slave processors, which would have the capability of distinguishing three modes of sensor operation; autonomously driving itself, autonomously driving other sensors, and being fully driven by another processor. Another option would be to develop an identification reference system allowing the "smart" central processor to talk with individual sensors. Currently these systems are custom-designed and maintained by only a few hardware/software experts.

One outcome of this workshop will be to establish a sensor network and information exchange on the ASLO website. The exchange will include an interactive, searchable directory where individuals and industry representatives will be able to submit or update statements about their research activities, interests, and basic contact information. Other features will include links to sensor-related websites, and a discussion forum. We encourage anyone interested in sensor technology to watch the ASLO website for further developments early this fall. We hope that this report will serve to stimulate a continuing dialogue on these topics and provide a focus for future sensor development.

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## Acknowledgments

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## WORKSHOP DISCUSSION TOPICS

Participants were asked to send three priority issues on sensor development prior to the workshop to serve as a basis for discussion. These issues are collated below.

### I. Sensors for Measurements on Different Space and Time Scales

- More observations of chemical and biological variables.
- Integrate a variety of sensors to tackle problems/questions in a concerted effort.
- Develop better and more reliable chemical sensors for short term and long term monitoring.
- Integration of micro-scale chemical sensors with biosensors.
- Development of techniques which allow fine scale measurements in heterogeneous systems, and which allow for simultaneous measurements of structure and solute distribution/dynamics.
- Development of chemical sensors with fast response time to enable measurements of more parameters with high spatial resolution.

### II. Technology Issues

- New ideas to achieve long term stability.
- Development of long-term remote deployment and data transfer capabilities.
- Data telemetry.
- Cost efficiency (purchase vs logistics vs data transmission costs).
- Specifications for unattended measuring equipment on buoys (kind of sensors, lifetime, power consumption, data transmission, ...).
- Scientists' agreement on performance specification/data rate.
- Interfacing of sensors to platforms.
- Dependency of modern sensors on Microsoft Windows-based systems (are we setting ourselves up?).
- Expendable biological sensors as an alternative to expensive submersible operations (e.g. expendable bouyancy-controllable cameras).
- Dynamical properties of sensors.

### III. Enabling New Technology, Collaborations, Funding Development

- New technology.
- Enabling technologies - new "nanno sensors" or at least much smaller, less expensive, robust sensors (able to withstand hurricanes!).
- Keep an eye on new technology developments and how they can be applied for marine instrumentation.
- Sharing of information / technologies.
- The impossibility of keeping abreast of current technological opportunities - in our experience oceanography is out of touch with the commercial computational and

technological breakthroughs coming along. Often, the appropriate technology exists but we have no idea that it's out there.

- And a related issue, integration/connectivity of new technologies to oceanographic applications - Most applications of new technology to oceanography is coincidental, being developed for much more large-scale, lucrative enterprises. Oceanographic development is thus a hodge-podge of sophisticated tools reapplied along with a lot of duct tape (the adaptation by an ocean mechanical or electronics engineer costing often ten times the original technology).
- Collaborations with new scientific and commercial partners.
- The two-three year timescale of funding is often incompatible with development - testing - scientific use - writeup. It is often impossible to report results of sensors (or even fully develop them) before one must abandon them for new avenues of funding.

## IV. Calibration Issues

- In situ calibration facilities for sensors intended to be used in moorings etc. over long periods of time.
- Intercalibration of various sensor systems (e.g. optical and acoustic biological sensors).

## V. Biofouling (this issue was listed by many people)

## VI. Determine Specific Sensors Needed for Science-Driven Questions (Participant Wish List)

- Which sensors are really needed (specification, detection range, ...) for marine monitoring?
- Measurement of microbial signaling molecules (e.g., homoserine lactones).
- Reliable sensors for simple organic compounds like acetate and VFA's.
- Chemical sensors that can provide speciation (oxidation state, specific ions/compounds) of elements.
- Development of reliable phosphate sensors for use in natural waters.
- Development of chemical and bio-sensors that can operate at extreme temperatures.
- Create a new generation of unattended samplers that will concentrate phytoplankton and bacteria from seawater, extract target biochemicals, and analyze them in situ. Desired analyses include molecular probes, pigments, tracer chemicals, etc.
- Expand suite of marine organisms amenable to genetic manipulation.
- Sequencing of additional ecologically-relevant organisms.
- In situ, real-time monitoring of reporter activity.
- On-line measurement of metabolic activities at the single cell level.
- Measurement of metabolic activities of uncultured microorganisms.
- Improved bioinformatics to relate complex biological and chemical/physical data sets.
- Improve the interpretation of optical data as biological information.

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Over the last 18 years of hydrothermal vent biology we have made significant strides in understanding the composition and physiology of the resident macro and microfauna. What has been lacking in these studies has been defining the link between the geochemistry and the local primary productivity it supports. The dynamic diffuse flow environments where most of the vent taxa reside are extremely difficult to sample at a spatial and temporal scale relevant to the biology. The dissolved reactive components are extremely labile and may significantly change during the transport to the surface - a current requirement for shipboard analysis. My interests lie in the development of in situ sensors that will provide real time assessment of physiochemical factors that have a direct influence on macro and micro fauna community composition and structure. Recently, we have employed voltametric micro-electrodes to provide rapid in situ measurements of most of the redox species from the DSRV Alvin. These electrodes when integrated to a micro-water sampler (SIPPER) and thermocouple array provided fine resolution measurements over short time scales. During our most recent cruise we were able to assess the chemical composition of fluid passing through the tube of vent polychaete, *Alvinella pompejana*. These analyses provided clear evidence that chemical speciation is a key component in governing fauna distribution in vent systems.

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The Oceanographic Technology and Interdisciplinary Coordination (OTIC) Program supports a broad range of research and technology development activities. Unsolicited proposals are accepted for instrumentation development that has broad applicability to ocean science research projects and that enhance observational, experimental or analytical capabilities of the ocean science research community.

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My research interests encompass marine population dynamics, the influence of physical and biological factors on spatial heterogeneity of zooplankton, zooplankton predator-prey interactions, phytoplankton-grazer interactions, and the influence of plankton on biogeochemical cycles. Although I have worked in a number of different systems, much of my research has focused on polar marine systems. These systems are difficult to sample and would benefit from sensor-based in situ observations. My general interest in sensors is the application of new technologies to improve plankton sampling and to quantify processes.

For many years I have used high-frequency acoustic systems to assess the abundance, distribution, and behavior of marine zooplankton and fishes. Although both acoustic hardware and software have improved significantly during the last 30 years, researchers still face serious technical issues in relating the detected returned signal to a biologically useful measure. I would like to see the continued improvement of current technology, as well as the development of new sensors for synoptic sampling of physical, chemical, and biological parameters to improve our understanding of processes in marine systems.

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My research interests focus on understanding the causes and consequences of mass and energy flux through the global ridge crest system; hydrothermal, magmatic and tectonic processes which form and modify young oceanic crust; and interplay among physical, chemical and biological processes at spreading centers. Currently the NEPTUNE project is being planned to establish a network of underwater observatories within the depths of the northeastern Pacific Ocean. NEPTUNE's 2000 miles of fiber-optic cable will provide power and communications to scientific instruments. For the first time, researchers, as well as shore-based learners of all ages, will participate in detailed studies and experiments on a wide area of seafloor and ocean for decades rather than just hours or days. NEPTUNE will be located in the northeastern Pacific and will be spatially associated with the Juan de Fuca tectonic plate. The NEPTUNE network will provide a coherent system of high-speed, submarine communication-control links using fiber-optic/power cables. Remote, interactive experimental sites will be connected with land-based research laboratories and classrooms. The system will provide real-time flow of data and imagery to shore-based Internet sites. It will permit interactive control over robotic vehicles on site and will provide power to the instruments and vehicles. We anticipate the need for the development of technologies and sensors for use in continuous experiments. NEPTUNE also may serve as a unique testbed for sensor and robotic systems designed to explore other oceans in the solar system.

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I am interested in benthic metabolism including oxygen consumption and denitrification. Also, I am working with Steve Emerson and John Dunne on developing a profiling mooring for Puget Sound. This is one of EPA's CISnet projects. With respect to the benthic stuff I am interested in ways to measure benthic flux remotely especially in the Arctic where ice cover makes these kind of measurements difficult in the early part of the growing season. With respect to Puget Sound we are interested in putting almost any sensor on the mooring. By the time of the ASLO meeting we should have completed our testing and actually have the mooring out in the field with a limited number of sensors (T, S, O2, light). These data are sent back by cell phone.

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I am a bio-physical oceanographer interested in biological-physical interactions and plankton patch dynamics. Patchiness is a nearly universal characteristic of the distribution of plankton in the coastal ocean. It is also a nearly universal characteristic of the physical, chemical and biological structures, processes and interactions that control plankton dynamics. This is particularly true in the coastal ocean where concentrations can vary over many orders of magnitude, where gradients can be steep, and where physical, chemical and biological forcing can be extremely strong and episodic. These conditions create major challenges in the design of sensors and techniques for their deployment. I have worked on 5 aspects of this problem: (1) I have developed deployment techniques that allow simultaneous sampling of vertical physical, biological and chemical structure down to centimeter scales (Donaghay, et al., 1992, Hanson and Donaghay, 1998); (2) I have collaborated with optical oceanographers and engineers in the development, testing and calibration of spectral optical sensors that have the dynamic range and spectral sensitivity to quantify and characterize plankton layers (Twardowski, et al., 1999); (3) I have collaborated with chemists in the development of techniques for in situ finescale profiling of nutrients down to sub-nanomole levels (Hanson and Donaghay, 1998); (4) I have collaborated with engineers in the design, development and testing of an underwater holocamera that can quantify in situ particle distributions, characteristics and motions over scales from 10 microns to 10 centimeters (Katz et al., 1999); and (5) I have developed deployment techniques that allow simultaneous 3-dimensional bottom-up sampling of finescale physical and optical structure (Donaghay, et al., 1998, 1999). I am currently coordinating a National Ocean Partnership Program project . The goal of this project is to develop an array of small, rapidly deployable profilers that will allow the real-time, 3-dimensional physical, chemical, biological and optical response of the coastal ocean to episodic events. This project involves the development of autonomous bottom-up profilers that use a new generation of highly integrated physical, chemical and optical sensors to profile the water column and telemeter and display the data in real time.

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Over the last ten years the main topic was the development of optical sensors and sensor instrumentation, developing new sensing schemes like fluorescence decay time sensing, nonlinear-optical sensing or capillary waveguide sensing. Much labour has been devoted to develop inexpensive and reliable instruments to measure luminescence decay times in the laboratory as well as in the field, for application in medicine, biotechnology, food technology, and environmental and marine monitoring.

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My interests in sensor design are in the autonomous monitoring of biogeochemical cycles and water quality. I am currently spearheading efforts to develop an autonomous, moored profiler called the Oceanic Remote Chemical/optical Analyzer (ORCA). The purpose of this mooring is to monitor water quality in south Puget Sound - an area potentially at risk to eutrophication due to urbanization. This project is part of the US EPA Coastal Intensive Site Network (CISNet). ORCA has three main components: 1) a three-point moored ATLAS toroidal float 2) a profiling assembly on the float with a computer, marine winch, cellular system, meteorological sensors, batteries and solar panels and 3) an underwater sensor package at the end of a hydro-wire with a CTD, transmissometer, chlorophyll fluorometer and oxygen electrode. At regular sampling intervals, ORCA profiles the water column using a winch driven by pressure information from the CTD. The data is recorded on the computer and transmitted back to the lab automatically via cellular communications to a host computer. Future additions will include a chemical nitrate analyzer, total gas tension and oxygen tension system and spectral optics package.

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I have been head of the Ocean Engineering Division since April 1993. Before that, my career was spent as a development engineer within the Applied Physics Group of the Institute of Oceanographic Sciences (1976-89) and working on the interface between physical oceanographers and ocean instrument developers at the James Rennell Centre for Ocean Circulation (1989-93). As well as managing the Division, I am responsible to the Director for the Autosub autonomous underwater vehicle development at the SOC.

Current research interests:

Acoustic methods for velocity measurement. I have been involved in developing, using, evaluating and improving acoustic Doppler velocity measurement techniques for over a decade. More recently I have been working with colleagues at RD Instruments evaluating a long range acoustic correlation current profiler (ACCP). This instrument can also be used as a deep ocean bottom track velocity log. Operating at 22 kHz it can provide current profiles to 1200 m and bottom tracking has been proven in 4800 m of water.

Bioacoustics: Zooplankton abundance and behaviour observations through sonar. Arising out of my work on velocity measurement using sonar, I became interested in what was causing the backscatter that the ADCP and similar instruments relied upon. Classical patterns of backscatter due to animal behaviour were observable in our early records, e.g. diurnal migration, but ADCP records also showed that patchiness in backscatter coincided with physical features such as fronts and eddies. We now use a Simrad EK500 scientific echo sounder operating at 38, 120 and 200 kHz to gather well-calibrated backscatter and relate the observations to physical features identified by towed undulators such as SeaSoar. Together with colleagues in the George Deacon Division we are working on developing a better understanding of the animal populations that give rise to the backscatter. A new instrument - TUBA -is being developed to make high frequency (250 - 2500 kHz) observations. TUBA will be mounted underneath SeaSoar, thereby providing the depth coverage we need.

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Our interest focuses on development of in situ sensing technology for nutrient elements in the upper ocean and hydrothermal environments. We have been developing and using in situ chemical analysis systems for 15 years. Our work began with hydrothermal sulfide, silicate and oxygen sensors (Johnson et al. 1986. In situ measurements of chemical distributions in a deep-sea hydrothermal vent field. Science, 231, 1139-1141), which we used to study biogeochemical interactions between vent organisms and their chemical environment (Johnson et al., 1994. Biogeochemistry of hydrothermal vent mussel communities: the deep-sea analogue to the intertidal zone. Deep-Sea Research, 41, 993-1011.). We extended this to measurements of metals in hydrothermal vent plumes (Coale et al., 1991. In situ chemical mapping of dissolved iron and manganese in hydrothermal plumes. Nature, 352, 325-328). More recently, we have focused on measurments of nitrate in the upper ocean (Johnson, 1989. Continuous determination of nitrate concentrations in situ. Deep-Sea Research 36, 1407-1413; Jannasch et al., 1994. A submersible, osmotically pumped analyzer for continuous determination of nitrate. Analytical Chemistry, 66, 3352-3361; Johnson and Jannasch. 1994. Analytical chemistry under the sea surface: monitoring ocean chemistry in situ. Naval Research Reviews, XLVI, #3, 4-12). All of the mentioned above was accomplished using wet chemical analysis systems that were adapted to operate at full ocean depths. We are now working on direct chemical sensing of species such as nitrate and hydrogen sulfide using ultraviolet spectrophotometry, as well as several new generations of chemical analyzers, including microfluidic systems.

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Although we have been interested in biosensor applications for a few years, our hands-on involvement with biosensors started with an 1999 NSF grant. My collaborators in this work are Drs. Josephine Aller, a marine benthic ecologist interested in benthic biogeochemal cycling, and Harbans Dhadwal, an optical engineer. We have contructed a prototype biosensor designed to detect hybridization of extracted nucleic acids to the surface of an optical fiber. The target molecules are primarily ribosomal RNA and messenger RNA. Both targets provide a natural signal amplification, relative to the number of copies of the equivalent DNA targets. Based on our past work, we believe we can use the biosensor as an extremely sensitive method for quantifying rRNA as a surrogate for direct measurements of bacterial growth. However, we are especially interested in relating the expression of mRNA to measured metabolic processes in marine water and benthic environments.

The following paragraph, taken from a special symposium proposal, describes our philosophy with regard to the use of molecular and biotechnological tools to study bacterial processes:

"By necessity, microbial ecology once focused on "black box" approaches that were biogeochemical in nature. Lacking the means to track specific microorganisms, microbial ecologists instead measured their consumption and production of organic and inorganic compounds, and from that inferred the biogeochemical role of microbes in ecosystems. The introduction of molecular-biology tools generated great excitement over the prospect of opening the microbial black box and learning exactly which taxa were involved in biogeochemical cycles; of tracking changes in the abundance, distribution and activity of biogeochemically important microbes; and of finally having the capacity to predict the impact of specific microorganisms on biogeochemical and ecological processes. However, something very different has happened instead. Molecular tools are commonly used to construct phylogenetic trees and lists of microorganisms present in a particular location at a particular time. Few studies make the bold leap from constructing a phylogenetic list, to making any inference about the biogeochemical role of these microorganisms. Instead of providing new and exciting insights into biogeochemical processes, molecular tools have tended to lead the study of microorganisms (especially bacteria) off on a separate and increasingly disconnected side track, in which microbial diversity is the primary focus and the biogeochemical role of microorganisms almost ignored. To a considerable extent, this evolution in the field of microbial ecology is driven by a reluctance to infer metabolism when the organisms are represented only by gene sequences. We particularly wish to invite debate over the potential for gene-sequence data to contribute to our understanding of microbial biogeochemistry, and to take the first steps in reintegrating microbial "molecular ecology" into the mainstream of aquatic biogeochemistry. "

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Research interests:
Development and application of electrochemical and fiber-optic microsensors in aquatic ecology and biogeochemistry. Development and application of imaging techniques for mapping solute distribution in surface associated microbial communities.

Background:
M.Sc. and Ph.D. in microbial ecology from the University of Århus, Denmark. Thesis: "Micro-optics and carbon turnover in dense microbial communities as studied with optical and electrochemical microsensors. From 1992-1998 I established and headed the Microsensor Research Group at the Max-Planck-Institute for Marine Microbiology. Since 1998 I have directed a group at the Marine Biology Laboratory, University of Copenhagen that works on development and application of microscale techniques in aquatic systems.

Some recent publications and reviews on sensor developments:

Kühl, M., and Jørgensen, B. B. (1994). The light field of microbenthic communities: radiance distribution and microscale optics of sandy coastal sediments. Limnology and Oceanography 39: 1368-1398.

Kühl, M., Lassen, C., and Jørgensen, B. B. (1994). Light penetration and light intensity in sandy sediments measured with irradiance and scalar irradiance fiber-optic microprobes. Marine Ecology Progress Series 105: 139-148.

Klimant, I., Meyer, V., and Kühl, M. (1995). Fiber-optic oxygen microsensors, a new tool in aquatic biology. Limnology and Oceanography 40: 1159-1165.

Kühl, M., Lassen, C., and Revsbech, N. P. (1997). A simple light meter for measurements of PAR (400-700 nm) with fiber-optic microprobes: application for P vs. I measurements in microbenthic communities. Aquatic Microbial Ecology, 13: 197-207.

de Beer, D., Glud, A., Epping, E., and Kühl, M. (1997). A fast responding CO2 microelectrode for profiling in sediments, microbial mats and biofilms. Limnology and Oceanography, 42: 1590-1600.

Amman, R. and Kühl, M. (1998). In situ methods for assessment of microorganisms and their activities. Current Opinion in Microbiology, 1: 352-358.

Kühl, M., Steuckart, C., Eickert, G., and Jeroschewski, P. (1998). A H2S microsensor for profiling sediments and biofilms: Application in acidic sediment. Aquatic Microbial Ecology 15: 201-209.

Glud, R.N., Kühl, M., Kohls, O., and Ramsing, N. B. (1999). Heterogeneity of oxygen production and consumption in a photosynthetic microbial mat as studied by planar optodes. Journal of Phycology 35: 270-279.

Kühl, M. and Steuckart, C. (2000). Sensors for in-situ analysis of sulfide in aquatic systems In: J. Buffle and G. Horvai (eds.), In-situ analytical techniques for water and sediments. IUPAC Environmental Analytical Chemistry Series Vol. 6, Wiley Pub., Chichester, 121-159.

Holst, G., Klimant, I., Kohls, O., and Kühl, M. (2000) Optical microsensors and microprobes. In: M. Varney (ed.), Chemical sensors in oceanography. Gordon & Breach, pp. 143-188.

Thar, R., Blackburn, N., and Kühl, M. (2000) A new system for three-dimensional tracking of motile microorganisms. Applied and Environmental Microbiology 66: 2238-2242.

Kühl, M. and N. P. Revsbech (2000). Microsensors for the study of interfacial biogeochemical processes. In: B. P. Boudreau and B. B. Jørgensen (eds.),The benthic boundary layer. Oxford University Press, Oxford. in press

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We are working with voltammetric sensors or (micro)electrodes. Voltammetry is the scanning of a potential scan while measuring the current and is analogous to spectrophotometry (scan wavelength while measuring absorbance). We presently work with gold-amalgam electrodes (Au/Hg) so that we can measure dissolved $O_2$, $H_2S/HS^-$, $S_x^{2-}$, $S_2O_3^{2-}$, $Fe^{2+}$, $Mn^{2+}$, $FeS_{aq}$, $Fe^{3+}$, $I^-$. We have used the electrodes to make *in situ* measurements in sediments from a ROV and at hydrothermal vents from DSV *Alvin*.

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Development of a Cyanobacterial Fe-Responsive Biosensor by Genetic Fusion Methods

We are seeking to develop strategies to assess bioavailable Fe using a cyanobacterial biosensor engineered to contain an iron-responsive promoter-reporter gene fusion. The fresh water cyanobacterium Synechococcus sp. PCC7942 has been routinely employed to probe mechanisms of gene regulation in cyanobacteria and is being used in this study for construction of the biosensor. Our laboratory has adapted genetic fusion methods already developed for Synechococcus to develop a fresh water biosensor prototype. Gene constructs have employed the luxAB/luxCDE reporter system and are driven by a novel Fe-dependent promoter that controls dpsA, which encodes a DNA binding protein of the ferritin superfamily. luxAB reporter constructs will also employ the iron-responsive isiA and irpA promoters, whose promoter structure is distinct from dpsA. Such work may yield a family of biosensors responsive to different transition metals and metal concentrations. Biosensor response is being calibrated using a luminometer over a range of Fe-limited conditions maintained in chemostat culture. In addition to the steady-state response, biosensor response during short-term Fe addition is also being characterized. Not only is the biosensor expected to provide valuable insight into Fe-phytoplankton interactions in lucustrine systems, its development can also be viewed as a necessary first step toward constructing a similar tool to be applied to the marine environment.

Dwivedi, K., A. Sen and G.S. Bullerjahn. 1997. Expression and mutagenesis of the dpsA gene of Synechococcus sp. PCC7942, encoding a DNA-binding protein required for growth during oxidative stress. FEMS Microbiol. Lett. 155: 85-91.

Sen, A., K. Dwivedi, K.S. Rice and G.S. Bullerjahn. 2000. Growth phase and metal-dependent regulation of the dpsA Gene in Synechococcus sp. strain PCC 7942. Arch. Microbiol., In press.

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*In situ* Measurements in Lake Sediments with Chemical Sensors

The main nutrient turnover in lakes occurs in the microbial mats at the sediment-water interface. The study of these biogeochemical processes is intricate due to the delicate structure and hindered access of the sediment surface. Our work is focused on the construction and *in situ* application of PVC based liquid membrane ion-selective electrodes (ISEs) such as pH, nitrate, ammonium, calcium, carbonate, and sulfide in the aquatic environment. A submersible device, LISA (Lander for Ion-Selective Analysis) was built to allow measurements in place directly at the sediment. A vertical stepping motor drives 12 sensors through the sediment-water interface where steep concentration gradients are measured with sub-millimeter resolution. The device is controlled from aboard ship, and the position of the sensors is observed with an endoscope. Reaction rates of early diagenetic processes and fluxes across the interface are calculated from the shape of concentration profiles with a model that takes into account reactions and vertical diffusion.

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Main activities:
open ocean moorings (sediment trap, sensors etc.)
moored sensor networks
underwater acoustic communication
bidirectional satellite communication (OrbComm)

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Looking for life 3 km underground in South Africa. Need to instrument boreholes for microbial experiments.

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My long-term goals are to understand the mechanisms controlling the distribution and productivity of phytoplankton. I have experience in the areas of nitrogen and phosphorus dynamics, photosynthesis, immunological methods, ocean color remote sensing, and in-water optics. I directed the Optical Oceanography course that was formerly taught at the Friday Harbor Laboratories (and will now be taught at the University of Maine in summer 2001).

Optical sensors provide information that can be related to phytoplankton biomass and physiology. One of my interests is to use multiple sensors to improve extraction of biological information from optical data. A second interest is to measure optics on the same space and time scales as the physics. I am working with Charlie Eriksen at the University of Washington to miniaturize and incorporate optical sensors on an underwater glider. A second project with John Dunne, Al Devol, and Steve Emerson, also at Washington, is to incorporate optical sensors on their profiling mooring.

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After my PhD at Kiel University, Germany, about an in situ calibration method for short pathlength transmissometers I started working in the Ocean Engineering Division of Southampton Oceanography Centre about one year ago. We are currently working on a new Oxygen sensor, a fluorometer for a wet-chemical Ammonia sensor and are just about to start working on some other wet-chemical sensors. I'm also working on an in situ calibration technique for conductivity cells.

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Challenger Oceanic is an R&D and manufacturing company of oceanographic samplers and sensors for biogeochemical studies. The main core business is equipment to monitor $CO_2$ and carbon fluxes using:
1. underway, unmanned $pCO_2$ sensors (air-sea exchange),
2. in situ pumps, with particle extraction onto filters or cartridges and dissolved phase extraction onto columns. Included in this method is disequilibria of uranium series isotopes to gain information on particle dynamics.
3. sediment/seawater interface by time series multi-coring and in situ profiling for oxygen, sulphide, pH and temperature.

Instrumentation for the real-time measurement of in situ anthropogenic gamma emitters in sediments is also supplied, including HPGe detection at 20K at the sea floor.

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Research in my laboratory integrates studies in microbial ecology and evolution. Specific research programs include the study of microbially catalyzed sulfur and nitrogen cycling. anaerobic microbial transformation of aromatic and chlorinated pollutants, control of metal speciation by anaerobes, and the structure and activity of biofilms. In order to study highly complex microbial systems (both in the open environment and as used in applied biotechnology ) our laboratory has worked to develop a conceptual and analytical framework using comparative nucleic acid sequencing for direct measurement of abundance, distribution, and activities of microorganisms in the environment. Full understanding of the microbial ecology of many natural processes will require information at many levels of biological organization - from the level of single cells, to populations, to communities. Some of our most recent research in technology development has been in the area of DNA microarrays. We are now developing microarrays for the comprehensive monitoring of microbial population abundance in the environment using ribosomal RNA sequence signatures. Longer-term goals include expression studies using microarrays incorporating key functional genes.

D.A. Stahl. Molecular approaches for the measurement of density, diversity, and phylogeny. *In:* Manual of Environmental Microbiology, ASM Press, Washington, D.C. (1997)

Guschin, D.Y., B.K. Mobarry, D. Proudnikov, D.A. Stahl., B.E. Rittmann, and A.D. Mirzabekov. Oligonucleotide microchips as genosensors for determinative and environmental studies in microbiology. Appl. Environ. Microbiol. **63**: 2397-2402 (1997).

Stahl, D.A. Small instruments for the study of small life. Env. Microbiol. **2**: 10 (2000).

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My interest in oceanographic sensor development centers on the application of new technologies to the problems of plankton sampling. The biggest problem facing the planktologist is one of resolution. The more samples one can process, the higher the resolution of data. However, microscopical analysis of a single plankton sample can take hours to days. Thus, time and funding often define the resolution of a study. Recent advances in digital technology now allow us to "see" plankton distributions on time scales relevant to their existence. My current line of research involves the application of these new technologies to plankton sampling to better understand plankton dynamics. I have experience with a wide range of oceanographic sensors and would like to keep abreast of recent developments as well as learn from other people's experiences.

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For several years I have been working together with a colleague on the development of a new optical in situ method to determine the salinity and density of seawater. This method is based on the measurement of the index of refraction. Three methods to realize the measurement are favoured which is the refractometric, measuring the changes of the angle of refraction, the interferometric, measuring changes of the optical path length and methods that measure intensity changes at interfaces. Only the first two has been pursuit where the refractometric method has some advantages for in situ applications. Although there are still some technical problem showing up with the current existing prototype of an in situ going refractometer there is a good chance to transfer the design into a commercial product. The potential of such kind of instrument is not alone limited to the range of physical variables. By applying special coatings on the sensitive element it will be as well possible to determine other biological or chemical parameters. My current interest is to find application areas for this kind of sensor where it could fill a gap in urgently needed observations. Examples might be investigations of double diffusion processes or coastal areas where river outflows can severely disturb the composition of salts in the water.

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# AGENDA

**Sensor Workshop/ ASLO Copenhagen 2000**
**4 June 2000**

**Bella Center**
**9:00 AM – 4:30 PM**
**Room 19**

0900 Welcome and Introduction — Kendra Daly

0910 Opportunities for Sensor Technology — Larry Clark

0920 Microbial sensors: George Bullerjahn, Mike McKay, Paul Kemp, David Stahl

0945 Vents/sediment boundary sensors: George Luther, Craig Cary, Beat Mueller, Christian Dinkel, Michael Kuehl

1010 Optical sensor technology: Michael Kuehl

1025 Break

1045 Hyperspectral sensors/AUV: Mary Jane Perry

1110 Mooring/sensors: Ken Johnson, Sonja Draxler, Tommy Dickey, Percy Donaghay, John Dunne, Al Devol, Gerrit Meinecke, Christoph Waldmann, Bill Simpson

1135 Southampton Ocean Centre sensor activities: Ralf Prien

1150 University of South Florida/Center for Ocean Technology activities: Tracey Sutton

1200 Questions/discussion

1215 Lunch

1315 Discussion Topic: Current challenges to the development of chemical and biological sensors.

1330 Breakout groups

1500 Break

1520 Group reports/ general discussion

1630 Adjourn workshop

# ABSTRACTS FOR SPECIAL SESSION

**Development of a Cyanobacterial Fe-Response Biosensor by Genetic Fusion Methods**
Bullerjahn, G., Rice, K., Kochendoerfer, S., McKay, R.

We are seeking to develop strategies to assess bioavailable Fe using a cyanobacterial biosensor engineered to contain an iron-responsive promoter-reporter gene fusion. The freshwater cyanobacterium *Synechococcus* sp. PCC7942 has been routinely employed to probe mechanisms of gene regulation in cyanobacteria and is being used in this study for construction of the biosensor. Our laboratory has adapated genetic fusion methods already developed for *Synechococcus* to develop a freshwater biosensor prototype. Gene constructs have employed the luxAB/luxCDE reporter system and are regulated by a novel Fe-dependent promoter that controls dpsA, a gene which encodes a DNA-binding protein of the ferritin superfamily. Reporter constructs will also employ the Fe-responsive isiA and irpA promoters, whose structure is distinct from dpsA. Biosensor response is being calibrated using a luminometer over a range of Fe-limited growth rates achieved in chemostat culture. In addition to the steady-state response, the biosensor is being evaluated during short-term Fe addition. Not only is the biosensor expected to provide valuable insight into Fe-phytoplankton interactions in lacustrine systems, its development is also viewed as a necessary first step toward constructing similar tools to be applied to the marine environment.

**In Situ Determination of Sulphide Speciation Correlates with Macrofaunal Distribution in Deep-Sea Hydrothermal Vent Communities**
Cary, S., Rozan, T., DiMeo, C., Luther, G.

The distribution of macrofaunal species at hydrothermal vents appears to be under the influence of physical and chemical parameters; however, this has not been documented. The vent tubeworm, *Riftia pachyptila*, and the vent polychaete, *Alvinella pompejana*, inhabit clearly different environments within a given vent site on the East Pacific Rise (9 deg. N). *In situ* voltammetry measurements were coupled with measurements from discrete samples to determine what phyico-chemical parameters may influence this distribution. Both the *in situ* voltammetry and discrete measurements show that free $H_2S$ is the major sulfide species found at fields of *Riftia*. However, dissolved FeS not free $H_2S$ is the major component found on areas where *Alvinella* reside. *In situ* temperature does not correlate with total sulfide concentration. However, as pH (measured onboard ship) increases, total sulfide content decreases. Pyrite is also detected by Cr(II) reduction of unfiltered discrete samples from several sites containing these organisms. In areas where *Riftia* have died, no free $H_2S$ or FeS is detected, Only $0_2$ is detected and the dead *Riftia* have Fe(III) solid phases on their outer tubes.

**Nitrogen Transformation in Lake Sediments Measured *In Situ* with Ion-Selective Sensors**
Dinkel, C., Müller, B., Stierli, R., Wehrli, B.

The LISA system (lander for ion-selective analysis) has been used in Baldeggersee (Switzerland) to measure the chemical gradients of ammonia, nitrate, pH and oxygen *in situ* at the sediment-water interface on the sub millimeter scale. A video endoscope and a submersible micromanipulator allowed to position the sensors at different stations down to the deepest part of the lake (64 m). Two sensors of the same kind recorded profiles in parallel. The high-resolution profiles from a field study in May 1999 were analyzed with a simple diagenetic model in order to extract diffusion rates across the sediment-water interface. The diffusion rates of oxygen, nitrate and ammonia were in the range of 12-67, 0.6-9.8 and 1.4-12 mmol/$m^2$d, respectively. Several nitrate profiles showed peaks at the $0_2$ boundary indicative of coupled nitrification-denitrifi cation.

**4-D Systems for Assessing Episodic Events in Coastal Waters**

Donaghay. P.L., Dekshenieks, M.M., Hanson, A.K., Holliday, D.V., Sullivan, J.M., Moore, C., and Zaneveld, J.R.

The determination of the coastal environmental response to episodic events such as storms, nutrient inputs, hypoxia and algal blooms requires the development of a system of ship-deployed and autonomous moored bottom-up profilers for coherent, real-time monitoring of multiple physical, biological, chemical, optical and acoustical parameters within the ocean, in three-dimensional space, as well as in time. These systems must be readily deployable, yet able to provide the real-time data necessary to modify sampling strategies and design experiments. Recent tests of prototype sensors and deployment systems indicate that these systems must be able to sample at far finer vertical scales and with far greater sensitivity than has been possible with conventional ship-based systems. Herein, we will use results of initial prototype tests to illustrate both the need for such capabilities and extent of recent improvements in optical and nutrient sensors and techniques for their deployment. This research was supported by ONR biological and chemical oceanography, ONR physical oceanography, NSF Oceanographic Instrumentation, and the National Ocean Partnership Program.

**New Measurements of Bio-Optical and Chemical Properties from the Bermuda Testbed Mooring**

Dickey, T, Bates, N., Boyle, E., Degrandpre, M., Jannasch, H., Merlivat, L., Moore, C., Taylor, C., Turner, K., Wanninkhof, R.

The Bermuda Testbed Mooring (BTM) project was initiated in June 1994 and has provided the first deep-sea mooring available for long-term testing of interdisciplinary oceanographic sensors and systems.  The standard BTM data suite includes measurements of meteorological, physical, and optical variables.  The BTM allows investigators to regularly exchange instruments in order to evaluate sensor and system performance.  Several new sensors and systems have been tested and scientific results have been published from the data sets.  Instrumentation has included: 1) bio-optical systems for measuring spectral inherent and apparent optical properties and spectral fluorescence, and 2) chemical systems for measuring nitrate, dissolved oxygen, and $pCO_2$.  Serial sampling devices have measured trace elements (e.g., lead and iron) and primary production using $C^{14}$ incubations.  In this presentation, we describe several of the systems and show highlight data sets illustrating the capability to observe a diverse set of variables, some of which are sampled on time cales of a few minutes.  In addition, we describe some promising future directions for measuring a broader suite of chemical, biological, and optical variables.

**Optical Sensors for Marine Measurements**

Draxler, S.

To meet the future requirements for ocean observing systems, a new generation of marine sensors for chemical parameters specially designed for the use on autonomous long-term measuring stations has to be developed. In the last years most of the efforts in the development of marine sensors were focused on the improvement of the accuracy of the measurements, or to complete the spectrum of the parameters which can be measured with in situ instrumentation. However, less attention was paid to the development of sensors specially designed and qualified for long-term observation stations.  Various optical sensing techniques used so far for sensing chemical parameters in marine environments will be presented, and their suitability for maintenance on unattended observation facilities with long-term autonomous measuring capability will be discussed. Special attention will be paid to devices that continuously and reversibly exhibit a change in some of its properties as a function of the concentration of a respective analyte.

**An Autonomous, Moored Profiler: The Oceanic Remote Chemical/Optical Analyzer (ORCA)**
Dunne, J., Emerson, S., Devol, A.,

We have developed an autonomous, moored profiler called the Oceanic Remote Chemical/Optical Analyzer (ORCA) that senses a variety of chemical and optical properties. The purpose of this mooring is to monitor water quality in south Puget Sound - an area potentially at risk to eutrophication due to urbanization. This project is part of the US EPA Coastal Intensive Site Network (CISNet). ORCA has three main components: 1) a three-point moored **ATLAS** toroidal float, 2) a profiling assembly on the float with a Tattletale-8 microcomputer, marine winch with level wind and slip rings, cellular system for remote programming and data download, meteorological sensors (wind, temperature, humidity, irradiance) batteries and solar panels for battery recharge, and 3) an underwater sensor package at the end of a hydro-wire with Seabird CTD profiler, Moss Landing Marine Labs chemical nitrate analyzer, Beckman dissolved oxygen electrode, Pro-Oceanus chemical dissolved oxygen sensor, Pro-Oceanus total dissolved gas sensor, Wetlabs transmissometer for particle concentration, Wetlabs Chlorophyll fluorometer for phytoplankton concentration and spectral optics (Wetlabs AC-9). At regular sampling intervals, ORCA profiles the water column using the winch driven by pressure information from the CTD. The data is recorded on the computer and transmitted back to the lab automatically via cellular communications to a host computer. Data will be presented for the spring of 2000 from a preliminary deployment with CTD, transmissometer, fluorometer and oxygen sensor.

**In Situ Chemical Sensors for Sea Water: A Review**
Johnson, K.

The interactions of ocean chemistry with biological and physical processes are exceedingly complicated. Dissolved chemicals that may act as nutrients limiting phytoplankton growth at low levels can be toxic and inhibit growth rates when present at higher concentrations. Variations in the concentration of organic ligands that bind trace metals can produce large changes in bioavailability with no change in dissolved metal concentration.

These interactions demand large numbers of observations of dissolved chemicals to understand their impact on ecosystems. However, the concentrations of most biologically essential chemicals in the ocean are chronically undersampled. There are few instruments, which are routinely available, that allow chemical concentrations to be remotely monitored in situ. Chemical measurements can, generally, be made only when ships are present on station to collect water samples for analysis in shipboard or land-based laboratories. As a result, the biogeochemical cycles that control ecosystem structure in the ocean are not well understood and they elude reliable prediction.

The pressing need for in situ chemical analyses has led many research groups to focus on this topic. As a result, there have been many significant advances made during the past 10 years in the development of systems for in situ chemical analyses in sea water. These advances bring
the promise of routine, in situ chemical analysis in the future. In my talk, I will focus on the work that is being done by the oceanographic community, the problems that still face us, and the potential solutions that should make in situ analysis a routine procedure in the future.

**Optical Biosensors for Marine Microbial Process Studies**
Kemp, P.F., Aller, J.Y., Dhadwal, H.S.

The biosensor consists of a flow cell containing a DNA-coated quartz rod; laser excitation source; photoncounting PMT detector; optical connection fibers; fluid and temperature-control systems. Unlabelled target rRNA, mRNA or DNA molecules are brought into proximity by hybridization to the immobilized probe DNA, in competition with fluorochrome-labelled synthetic targets. The concentration of target molecules is inversely proportional to fluorescence yield. Emission-collection fibers are oriented at 90', greatly reducing background and permitting independent optimization of the excitation and emission optics. Chemical regeneration allows for repeated measurements. Our goal is a biosensor with sufficient sensitivity to detect: specific bacteria (rRNA target); the expression of a gene associated with a metabolic function (mRNA target); or multiple-copy DNA targets, including viruses, in small, easily processed samples. In aquatic environments, realistic sample sizes currently would be <100 ml for a moderately slow-growing bacterial species comprising 10% of the total community; <100 ml for relatively abundant viruses; or ca. 250 ml for an mRNA target. Much smaller sample sizes would be required for sediment porewater. We anticipate further increases in sensitivity with later generation instruments.

**A Hydrogen Sulphide Microsensor for Use in Aquatic Environments, Sensor Characteristics and Appliction**
Kühl, M.

Fine scale measurements of sulfide have mostly relied on the use of Ag/Ag2S ion-selective electrodes, which often exhibit non-ideal measuring characteristics when applied in natural waters. We present an amperometric microsensor for $H_2S$ measurements in aquatic environments. The microsensor exhibits a linear response to $H_2S$ over the range of 1-1000 micromol $H_2S$, low stirring sensitivity of <1-2 percent, and fast response time of <0.5-1 s. The sensitivity of the $H_2S$ microsensor is typically 0.2-2 pA per micromol $H_2S$. The microsensor has proven a better alternative to Ag/Ag2S electrodes in both laboratory and *in situ* applications. We present technical details of the $H_2S$ microsensor and show examples of application in studies of anoxygenic photosynthesis, sulfide oxidation, and sulfate reduction of sediments.

**In Situ Sensors Using Voltammetric Solid State Microelectrodes Detect Sulfur Speciation at Hydrothermal Vents**
Luther, G., Taillefert, M., Cary, S., Nuzzio, D.

An in situ submersible electrochemical analyzer was designed by Analytical Instrument Systems, Inc. for use from the deep sea submersible Alvin and used with up to four gold/amalgam electrodes to study the diffuse flow chemistry of hydrothermal vents. The analyzer and electrodes will be described. Two electrodes were deployed directly onto sampling locations at ambient temperatures and two others were housed in a flow cell to make measurements at 2 deg. C. We present data from dives on recent cruises in May 1999 (9 deg. N East Paciflc Rise; 2500 meters depth) and January 2000 (Guaymas basin; 3500 m depth). The data from the May 1999 cruise show that total dissolved sulfide is primarily due to aqueous FeS and $H_2S$. Polysulfldes and thiosulfate are not major components of the sulfur species detected. The geochernical implications of this speciation will be discussed.

**Remote Control of Deep-Sea Moored Instruments**
Meinecke, G., Ratmeyer, Wefer

The DOMEST project was started to develop a moored sensor network in the deep ocean, north off the Canary Islands. The implementation of an bidirectional satellite link in combination with an underwater acoustic link provide communication from land into the deep sea and vice versa, on request. With the DOMEST project, a remotely controlled measurement of element and particle transport in the deep sea is possible. Remote control includes access on a variety of without recovering sensors from the deep ocean. Sampling intervals can be changed interactively from land, status data from the instruments can be checked and it is possible to download the data subsequently. These possibilities allow an advanced sampling and probing of parameters depending on various environmental parameters, such as satellite derived ocean colouir or particle input during dust storms. Communication underwater is based on 4 independent acoustic modem clients, combined with different sensors like sediment trap, ADCP, CTD and autonomous particle camera system. Bidirectional data transmission between these modems is possible up to 2400 baud.

**Monitoring Subsurface Microbial Activity *In Situ* by Instrumenting Bore Holes in Ultradeep South African Au Mines**
Onstott, T., Moser, D., Takai, K., Fredrickson, J., Pfiffner, S., White, D.

Narrow boreholes; frequently penetrate up to 100 meters into the country rock of the deep South African Au mines. PLFA and 16SrDNA analyses of an open, dripping borehole located 3.2 km below surface revealed a rich and diverse thermophilic microbial community present in saline, subneutral water. When the borehole was isolated from the mining air the borehole water became anaerobic and alkaline and community structure changed. *In situ* rates of microbial activity could potentially be assessed by instrumenting fresh mine boreholes with robust sensor arrays.

**Incorporation of Sensors into Autonomous Gliders for 4-Dimensional Measurements of Bio-optical and Chemical Parameters**
Perry, M.J., Eriksen, C.

A partnership of universities, industries, and local government agencies (sponsored by U.S. National Oceanographic Partners Program) is extending development of an existing long-range autonomous glider vehicle. The goal is to measure bio-optical, optical, and chemical parameters - in addition to physics -continuously, in real time, and in diverse environments. The Virtual Mooring glider, developed by Eriksen, is a small, light-weight vehicle that alternately dives and climbs as it glides forward under buoyancy control. Because of its low power consumption, it is capable of extended missions of several thousand kilometers over several months. The CTD-equipped glider with a cell phone for transmitting data and receiving new commands has already proven successful in collecting physical data on coastal deployments. Miniaturized sensors, under development by SeaBird for dissolved oxygen and by WET Labs for chlorophyll fluorescence and inherent optical properties, are being incorporated into the glider. The newly instrumented glider will allow us to measure biologically important properties on the same time and space scales as the physics. It will provide a mechanism for ground-truthing ocean color sensors on the appropriate scales.

**Hyperspectral Remote Sensing as a Monitoring Tool for Coastal and Near-Shore Marine Habitats**
Siliciano, D. siliciano@biology.ucsc.edu

Hyperspectral imaging involves the acquisition of image data in many contiguous spectral bands in the visible and infrared range, to produce reflectance spectra for each pixel in the image. These spectral measurements investigate biological, chemical and physical properties of surface matter, enabling applications over the regional scale of the image. We are using hyperspectral imaging to map and monitor intertidal and benthic habitats, and to identify key oceanographic processes synoptically over large scales. Elkhorn Slough is one of California's largest remaining tidal wetlands and its waters are part of Monterey Bay National Marine Sanctuary. Hyperspectral imagery was acquired for this area in October 1999. Simultaneous measurements with a portable hyperspectral radiometer characterized a variety of intertidal organisms and substrates. The resulting maps of the key biotic and abiotic components of the region and key oceanographic processes provide a synoptic, large-scale assessment at the time of acquisition. These methods provide a promising tool for environmental analysis and monitoring for many marine ecosystems. Preliminary analyses of field spectroscopy from coral reef habitats illustrate the potential applications for coral reef management.

**The Use of DNA Microarrays to Characterize Environmental Microbial Diversity**
Stahl, D., Kelly, J., Sappelsa, L., Bavykin, S., Mirzabekov, A.

The combination of molecular systematics and molecular methods for recovering DNA sequence directly from environmental samples has shown that far less than 1% of environmental diversity has been described. Thus, the most general of monitoring strategies should encompass all microbial diversity, both known and unknown. This is now possible using the phylogenetic framework provided by comparative sequencing of ribosomal RNAs in combination with DNA probes targeting them. Since probes can be designed to capture groups of different phylogenetic depth, species not yet identified can be identified. Since DNA microarrays provide a format for massively parallel hybridization reactions, simultaneous identification of hundreds or thousands of different microbial populations is now possible. We are currently working with a microarray format in which oligonucleotide probes are immobilized within individual polyacrylamide gel elements affixed to a glass slide, and target nucleic acids are fragmented and fluorescently labeled before hybridization with the immobilized probes.

**A Towed Array for High-Resolution Plankton Studies**
Sutton, T., Hopkins, T., Langebrake, L., Burghart, S., Remson, A.

Traditional plankton sampling methods involve the use of nets and/or bottles to determine faunal composition and abundance. Modern electronic sensors have been developed which count and size suspended particles, allowing detection of particle distributions on time scales relevant to physical oceanographic processes. Drawbacks of the former methods include sampling bias and the need for time-consuming microscopical analysis, while the latter are subject to high noise-to-signal ratios and provide little taxonomic information. Here we present a towed instrument array, the High Resolution Sampler (HRS), which enables simultaneous particle analysis and verification. The HRS electronics array bears commercially-available sensors as well as two particle analyzers developed in-house. The verification system consists of a 20-net carousel assembly, in-line with particle analyzers, and a Niskin bottle array, with bottles triggered during net index. Analysis of electronic particle distributions can then be used to direct microscopical analysis of selected net samples. In this manner, sampling bias can be minimized and the appropriate space scales of biophysical interactions can be determined. Data obtained with the HRS at an ecosystem modeling site will be presented.

**Advantages of Using Buoyancy Driven Autonomous Instrument Carriers to Measure Physical and Biochemical Parameters – Current Results and Future Perspectives**
Waldmann, C.

Within the framework of the national funded project DOMEST a further development of a buoyancy driven instrument carrier has been undertaken. This carrier is operating fixed to a mooring line and serves as platform for different sensors. The main goal of this development was to extend the range of this type of carrier to full ocean depth. The results of the tests were very encouraging. The dynamic behaviour is comparable to free falling or rising devices like profiling floats. Therefore a continous flushing of attached sensors is guaranteed. This makes principle very attractive especially for biochemical sensors. The quality of the physical measurements are improved as well. Within this presentation the different approaches for fixed profiling measurements will be discussed. The latest results of the on-going *in situ* tests and future developments for instance the bidirectional communication to the moored instrument via a satellite link and the extension of the lifetime of the system and the number of cycles will be presented