

Sampling Workshop Report

1. Introduction

A. Why the Workshop

Collecting water samples and transporting them to the laboratory for analysis will remain a critical activity for the ocean sciences for the foreseeable future. While methods for ship-based sampling are well understood, the growing use of autonomous platforms, both fixed and mobile, raises both opportunities and technical challenges. MBARI hosted a workshop in spring 2010 for the purpose of reviewing the needs of autonomous water sampling systems in marine science. Topics discussed included:

- 1) Scientific drivers for water sampling and resulting requirements
- 2) Available systems and methodology
- 3) Needs and opportunities for future development.

The goal of the workshop was to develop a detailed understanding of sampling needs, including requirements for sample size, spatial and temporal scales of sample intake, sample container sterility, management of ambient/container pressure differentials before, during and after sample acquisition, onboard sample processing or preservation methods appropriate for specific organisms, and subsequent sample analyses.

Workshop participants included science users and technology developers of water sampling systems.

B. What was the Workshop

- a. Presentations
- b. Breakout groups
- c. Discussion

C. Primary focus on sample collection by autonomous platforms

- a. Not sensors per se, adaptive sampling, data communications, processing or management etc

D. Workshop and report focus on physical sample capture (c. and d. above)

- a. Physical samples typically required for (i) high value measurements (genomics, primary production) and (ii) sensor calibration (nutrients, species composition)
- b. Sensors typically used to target sample capture
- c. Technological innovation often flows from sample capture → sensor development.

E. Report outline

2. Case studies --- Collection of physical samples by autonomous platforms

A. HABs

- a. Scientific problem

- b. Current autonomous samplers
 - c. Future autonomous samplers
- B. Mesoscale eddys
 - a. Scientific problem
 - b. Current autonomous samplers
 - c. Future autonomous samplers
- C. Thin layers
 - a. Scientific problem
 - b. Current autonomous samplers
 - c. Future autonomous samplers
- D. Zooplankton
 - a. Scientific problem
 - b. Current autonomous samplers
 - c. Future autonomous samplers
- E. Low oxygen
 - a. Scientific problem
 - b. Current autonomous samplers
 - c. Future autonomous samplers

3. Synthesis

- A. Continuum of methodologies for obtaining environmental information
 - a. Sensors not needing water handling
(sonar, adcp, fluorescence, backscatter)
 - b. Sensors needing water ducting
(LOPC, digital imaging)
 - c. Sensors with temporary sample (water or filtrate) capture, not returned to lab (cytometer, autoanalyzer)
 - d. Samplers that capture sample (commonly water) and utilize sensors (oxygen, cytometer, DIC, etc.) to measure rates of change over time, sample not returned
 - e. Permanent sample (water or filtrate) capture, returned to lab
(genomics, primary production, salt, nutrients)
- B. Common and unique sampling requirements for case studies
- C. Unsolved sampling requirements
- D. Future developments
- E. Revisit general issues raised in Introduction

The Controlled, Agile, and Novel Observing Network

Growth and death of microorganisms in the sea occur as ephemeral features that move over time in many directions. These “booms and busts” reflect the interplay between physics, chemistry, and biology, and represent the very base of the oceanic food chain. But not all of this activity is benign. Some blooms can be harmful to humans and wildlife, and can bring negative economic consequences. Our ability to achieve mechanistic understanding of these phenomena has eluded us because of technical difficulties tracking water masses and the rapidly changing life they carry. The CANON team aims to overcome that limitation by creating new ways to remotely interrogate oceanic conditions and collect samples of microorganisms for in situ analysis as well as for return to shore side laboratories.

How does the marine pelagic food web respond to changes in the natural environment? Our ability to answer this question is central to predicting how the oceans will respond to human perturbations such as increasing atmospheric CO₂ levels, nutrient runoff from agricultural and aquacultural activities, dispersal of various pollutants, and climate change. Yet present-day models are not capable of accurately predicting future consequences because very basic questions about the food web are still unanswered. Further, unexpected but significant events will continue to appear requiring an observing system capable of rapidly responding to these new challenges as well as investigating the basic processes required for accurate environmental prediction.

The food web of interest is much more complex than it might initially appear. Take the marine microorganisms at its base—not composed of one entity, but rather they span a tremendous range of physiological capabilities and mediate many different biogeochemical processes as functionally diverse as that of the full biota on land. Thus, changes such as increased atmospheric CO₂ levels, which will reduce ocean pH, will have different consequences for the many organisms in the marine environment, and these in turn will propagate through the food web. While the last decade has seen a revolution in understanding of the diversity and importance of microbial populations in the ocean, our understanding of how those populations respond to changes in their environment—and to other microbes—is rudimentary at best. Developing a better understanding of microbial dynamics through the development of new ocean observation capabilities is the initial thrust of the Controlled, Agile, and Novel Observing Network program, known as “CANON”.

A key initial premise of CANON is to provide a new class of observation systems that will be able to autonomously follow and facilitate the study of communities of organisms and the transitions they undergo in the ocean environment. This is critical because current spatial and temporal sampling resolutions are not relevant to the spatial and temporal scales on which microbial processes occur. A fundamental driving principle for the initiative, and for further technology development, is that processes and microbes must be studied at scales relevant to the organisms’ adaptive strategies to determine how metabolism influences larger-scale ecosystem dynamics.

The CANON team aims to merge observation, modeling, and prediction to cast projections of biological, chemical, and physical gradients within a defined region. By directing small fleets of mobile sensors within that domain, they will detect specific phenomena remotely, collect physical samples of microbes, algae, and small invertebrates autonomously, and track the evolution of biological patches over time. The system in its final form will be able to integrate information from remote sensing, in situ arrays, and models and navigate autonomously while providing scientists and managers with the information required to interpret the mechanisms detected and make decisions as to how to proceed.

Environmental Sample Processor

From a technological perspective, CANON has its roots in a convergence of sensing and autonomous platform technologies developed at MBARI over the last decade. New “ecogenomic sensors”, like the Environmental Sample Processor (ESP), provide an unprecedented possibility for in situ molecular detection of organisms such as harmful algal species.

The ESP employs molecular probe techniques to autonomously assess the abundance of microorganisms, their genes, and metabolites in near real-time. Coupled with sustained environmental measurements provided by ocean observatories, assessment of microbial community structure and function will soon be possible in situ on spatial and temporal scales not previously achievable. To date, tests proving the feasibility of this approach were largely limited to coastal waters. In 2009, the MBARI team proved for the first time that the ESP could also be operated for extended periods at depth (Figure H7). Deployed to 900 meters, the instrument successfully used the same DNA probe array and sample archival functionality as that proven to work in shallow waters, revealing time-varying alterations in microbial community structure as a function of changing environmental conditions. The instrument ran as a stand-alone system as well as networked via the MARS cabled observatory.

Long-Range Autonomous Underwater Vehicle

Over the last three years MBARI engineers have developed a new class of autonomous underwater vehicles (AUVs) to support chemical and biological sensing missions covering ranges of 1,000 kilometers or more. The size and power consumption of desired chemical and biological sensors precluded the use of existing long-range gliders, and the endurance requirement precluded the use of more traditional propeller-driven AUVs. The concept for the new vehicle was a highly energy efficient propeller-driven AUV capable of operating at speeds between 0.5 and 1.0 meter per second. The new vehicle, named *Tethys*, conducted its first brief autonomous missions in December 2009, just offshore of Moss Landing. [Figure H8]

The range and endurance of the new long-range AUV greatly expands the types of observations and experiments possible with autonomous platforms. For example, one of the institute's AUVs carries a comprehensive suite of sensors out to MBARI's M2 mooring and back. *Tethys* will carry a smaller, but still impressive suite of sensors 10 times further, extending the reach of MBARI's shore-launched AUVs into the California Current system. This will expand researcher's non-ship observational capability beyond the upwelling shadow, well into the oligotrophic (nutrient poor) ocean. The capability also provides a foundation for studying phytoplankton blooms from boom to bust, by providing a mobile platform which can survey a bloom continuously through the two weeks to monthlong lifetime of a bloom.

Improving autonomous water sample collection

The development of novel water samplers for autonomous platforms is another focus of CANON. In 2009, major advances were made in sampling intensity and screening of biodiversity from water samples obtained with the AUV "gulper" water sampling system. Previously, only tens of samples were collected and processed, but during 2009, nearly 100 were obtained and processed, thanks to a shift in project collection priorities and the success of AUV missions. New molecular probes were designed to assay a greater diversity of zooplankton, including copepods, to complement those already available for microorganisms. Together, these improvements are yielding data with sufficient resolution to reveal patterns in the distribution of plankton across oceanographic fronts, layers, and other features identified in the AUV surveys. A major national workshop is being organized at MBARI in 2010 to develop a detailed understanding of sampling needs, including requirements for sample size; spatial and temporal scales of sample intake; sample container sterility; management of ambient and container pressure differentials before, during, and after sample acquisition; onboard sample processing or preservation methods appropriate for specific target species; and subsequent sample analyses.

Decision support system

One principal problem continues to be where to position the AUVs and where they should sample to resolve key ecological questions. This also involves deriving an optimal path for sampling "at the right place and time" based on science requirements. To solve this problem the team envisions the capability to capture data from diverse sources to target AUV deployment as well as to provide situational awareness to the scientist on shore. Current efforts take a number of complementary approaches. First, to use a wide range of data sources including satellite observations, mooring data, and high-frequency radar to statistically project patch advection. Second, to build decision-making capability using artificial intelligence techniques onboard AUVs to adaptively sample dynamic coastal phenomenon. CANON proposes to systematically use these capabilities to provide a shore-side Decision Support System (DSS) tool for scientists (Figure H9) to target the placement of AUVs close to and within bloom patches. Such a tool will enable

scientists on shore to generate “what if” scenarios for potential changes to AUV transects, changes to environmental conditions or alternative sample sites for hypothesis testing. The aim will be to encapsulate predictive capabilities in one tool that generates advice for scientists on shore.

Data and information management

In the first year of the CANON initiative, one goal was to demonstrate that the whole is more than the sum of its parts, especially in the area of data and information management, which involves several ongoing efforts at MBARI. These include:

- A project to organize and visualize MBARI upper-water-column oceanographic data;
- Data systems and collaborative tools developed initially for multi-platform experiments in Monterey Bay, such as the Autonomous Ocean Sampling Network experiments in 2003 and 2006, for physical processes on scales ranging from one to 100 kilometers;
- Development of the decision support system described above;
- Data management and communications systems developed by our close partner, the Central and Northern California Coastal Ocean Observing System (CeNCOOS), a regional association that is part of the U.S. Integrated Ocean Observing Systems (IOOS); and
- Emerging genomic data management systems.

Figure H10 shows a conceptual diagram of how these efforts might be integrated such that as the initiative progresses, its information management needs will be met.

The initial focus

CANON brings together a broad group of researchers committed to tackling the sampling, experimental, and analysis issues behind more sensitive investigation of communities, and the dynamics of individual components. Thus, CANON will bring the biological insights to the requirements of new sensor development, based on cutting-edge approaches to genomics, transcriptomics, and physiology studies, and the technological innovations necessary to access the environment in unprecedented resolution, letting environmental conditions guide sampling decisions in real time and in situ.

Field work in 2010 will tackle two fundamentally different ocean biomes—the coastal environment, which is highly dynamic in terms of physical and chemical properties—and more oligotrophic waters representative of the open ocean environments that cover much of the planet. Each environment provides a different set of challenges. Coastal systems harbor the majority of ocean life, are in direct contact with human populations, and are easier to access but highly dynamic, triggering resolution and time response issues for instrumentation and adaptive sampling processes. The coastal experiments will center on observing and following harmful algal blooms. The primary coastal microorganisms are diatoms and dinoflagellates, part of the microphytoplankton.

Open ocean waters tend to be more stable and drive global biogeochemical budgets but provide intense technological challenges, requiring long-range autonomous devices and sample preservation techniques for samples that may be collected weeks or months prior to analysis. Organisms in these environments are very different from coastal communities and are dominated by very tiny phytoplankton (picophytoplankton). We still have limited understanding of which picophytoplankton are the key players in photosynthetic uptake of carbon in open ocean environments. Progress will require measuring not only abundance of individual groups, but also measuring their activities, growth and mortality.

By contrasting field work in these two systems we hope to tease apart rules of community assembly for each system, which can then be applied to more predictive modeling of how communities might transition under climate change.