LONG-TERM GOALS

My long term goal is to evaluate controls on nutrient limitation in the coastal ocean using an interdisciplinary approach combining geochemical, biochemical, and biological assays. I am particularly interested (i) in assessing the role of phosphorus (P) as a limiting nutrient in the coastal ocean; (ii) in the phenomenon of seasonal shifts in limiting nutrient(s) (e.g. between nitrogen (N) and P); and (iii) how such shifts influence phytoplankton species distributions.

OBJECTIVES

Establish whether phosphorus acts as a limiting nutrient in coastal systems, how pervasive P-limitation is in both space and time, and how nutrient limitation dynamics may affect phytoplankton population distributions. Despite mounting evidence that nutrients other than N limit primary production in marine systems, the assumption of N-limitation is still pervasive. By simultaneously determining all bioavailable forms of nutrients in a system, and coupling this information to data which constrain the identity of limiting nutrient(s), such as enzymatic assays and bioassays, we can begin to critically evaluate nutrient limitation. This study takes such a multi-faceted approach, and where possible views these data within the context of phytoplankton species distributions. Indications thus far from this study are that nutrient limitation in the coastal ocean is a dynamic process, with the identity of the limiting nutrient(s) shifting on seasonal or even shorter time-scales.

APPROACH

Contrast biogeochemical and physical factors on the LA and Eel River Shelves to determine which factors force the observed, opposing, seasonal shifts in limiting nutrient status of these systems.

Extend the data base for coastal systems exhibiting phosphate limitation by taking advantage of funded field programs in which participating scientists are willing to collect samples for me to assay for Alkaline Phosphatase (APase) and dissolved organic nutrients, in order to document presence/absence of P-limitation.

Pursue the ELF methodology, for evaluation of cell-specific APase activity, in collaboration with Dr. Sonya Dyhrman in the WHOI Biology Department.
### Controls on Nutrient Limitation in the Coastal Ocean

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WORK COMPLETED

Completed cytofluor method development for rapid determination of Alkaline Phosphatase (APase) on water samples, including head-to-head comparison with standard fluorometric method and statistical analysis of methods comparison.

Successfully utilized the cell-specific Enzyme Labeled Fluorescence (ELF) method for tagging and imaging phytoplankton cells that are P-stressed and are therefore producing APase. This work has been done with samples from the Oregon Coast and from Ashumet Pond, Cape Cod.

RESULTS

The cytofluor method has been validated as a robust and rapid method for determination of APase in natural waters.

![Comparison of traditional fluorometer and newly developed cytofluor method for APase activity determinations. Black circles represent unfiltered Ashumet Pond samples; red squares represent 0.2 µm filtered Ashumet Pond water spiked with commercial APase (E. coli). Error bars represent standard deviation over triplicates. The field within the dashed blue lines represents 1-sigma about the 1:1 line. Most samples fall within this field (Ruttenberg and Haupert, unpubl.).](image)

APase activity has been observed now in two coastal ocean regions, the Eel River shelf and the Oregon shelf, indicating that phytoplankton become stressed by low phosphate levels in both these environments.

On the Oregon shelf, analysis of ELF-labeled diatom cells indicates that there may be species differences in expression of APase.
IMPACT/APPLICATIONS

Demonstration of P-limitation in coastal ocean systems, and seasonal shifts in limiting nutrient dynamics in the coastal ocean, contributes to the growing appreciation of the complexity of nutrient limitation of primary production.

The optimized method for APase assays by Cytofluor will be highly valuable in field studies of enzymatic parameters such as APase. The high degree of spatial heterogeneity common in aquatic systems demands broad sample coverage and, therefore, large numbers of analyses. The Cytofluor increases sample through-put by 50x to 100x over traditional methods providing a significant advance in our abilities to accurately characterize natural ecosystems.

Utilization of the ELF-APase assay in tandem with APase activity measurements will provide new insights in to species specific response to the nutrient environment by permitting us to examine individual cell response to P-stress.

TRANSITIONS

RELATED PROJECTS

I am participating in an AFCEE-funded project to determine the nutrient limitation status of a lake on the Otis Air Force Base Superfund site which is being impacted by a groundwater sewage plume. In order to determine the extent of the potential impact, the nutrient limitation status of the lake must first be verified. I am using the methods developed in this and previous ONR-funded work (APase and DOP analyses) in the AFCEE project.

I have just begun work on an NSF-OCE funded project to study DOP cycling and P-limitation on the Oregon Shelf. I have begun applying the methods developed as part of my ONR-funded work to evaluate DOP cycling and the presence/absence of Alkaline phosphatase in these samples.

PUBLICATIONS

