The Effects of Water Depth on Short-term Biofouling of Introduced Substrates

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LONG-TERM GOALS

Our goal is to interpret changes in the characteristics of elastic and inelastic light scatter from submerged substrates colonized by biological organisms. We wish to determine the changes in optical properties that occur with the settlement and succession of sessile organisms and their associated biofilms and to develop an optical model that can predict the magnitude and characteristics of the biota and their ability to disguise man-made objects placed on the seafloor.

OBJECTIVES

As this is the first year of the project, we have concentrated on the design and logistics of our experiments as well as development and evaluation of the bio-optical methods (flow cytometry, reflectance, flash fluorescence) we will use for measurements of short term fouling on panels of varying composition and surface roughness. To this end, we have initiated two studies in coastal Gulf of Maine waters: 1) a near-surface study of color (black vs. white) and texture (rough vs. smooth), and 2) a study of fouling as a function of depth (light levels) that included horizontally and vertically oriented panels of roughened PVC and glass. The primary optical property under consideration is spectral reflectance, fluorescence yield and absorption are also measured on panels and fouling communities brought back to the laboratory.

APPROACH

Our overall approach will encompass studies in temperate and subtropical coastal waters in the Gulf of Maine and off Key West, Florida. This summer's work was restricted to the Gulf of Maine where glass and plastic fouling panels were suspended from floats for several weeks. In the near-surface study, a panel set (Figure 1) was retrieved at time points and optical measurements were made in the laboratory by Charles Yentsch and Sara Woodman Yentsch using a flow cytometer, spectral reflectometer, spectrophotometer and pulse flash fluorometer. Fouling as a function of depth was determined on panel sets suspended from an optical mooring in 20m of water in the mouth of the New

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14. ABSTRACT
Our goal is to interpret changes in the characteristics of elastic and inelastic light scatter from submerged substrates colonized by biological organisms. We wish to determine the changes in optical properties that occur with the settlement and succession of sessile organisms and their associated biofilms and to develop an optical model that can predict the magnitude and characteristics of the biota and their ability to disguise man-made objects placed on the seafloor.

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Meadows River. Panel sets (Figure 2) were fixed at the 50, 15 and 5% light levels and remained in place for the duration of the experiment, David Phinney and Douglas Phinney documented fouling photographically and spectral reflectance was measured using a diver operated reflectometer.

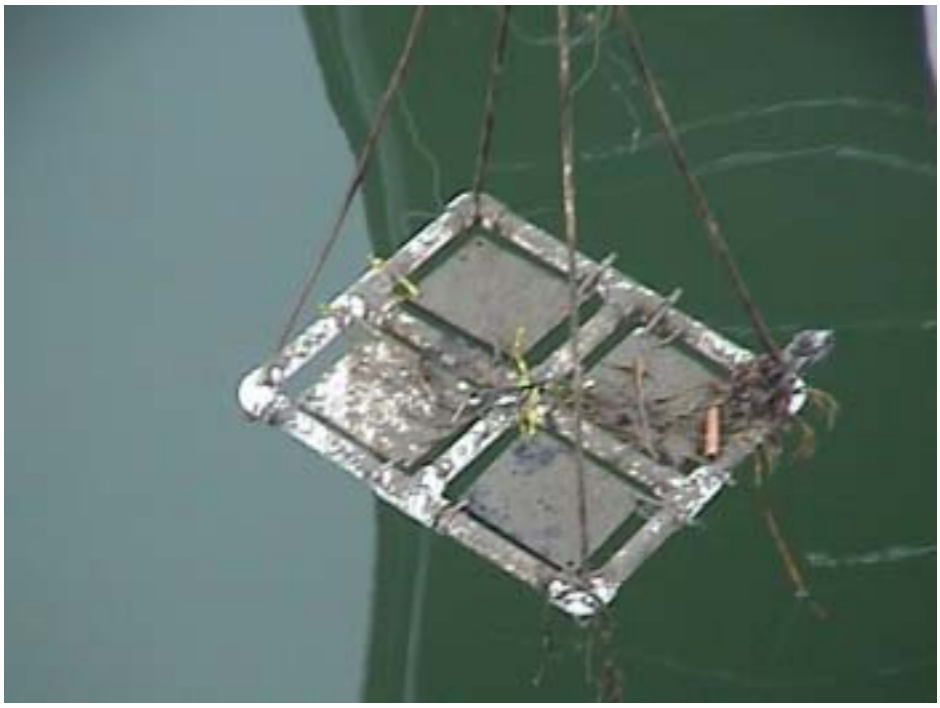

Figure 1. Fouling panel set for surface study in Gulf of Maine coastal waters. Black and white plastic panels were deployed for six weeks at 1m below the surface.

Figure 2. Panel set used for study of fouling as a function of depth in the mouth of the New Meadows River. Plastic and glass panels were deployed horizontally and vertically at the 50, 15 and 5% light levels. Experimental surfaces were set within a larger surface to reduce edge effects.

WORK COMPLETED

Our observations of biofilms covered the period from July 18 to August 28, 2002, when conventional glass microscope slides were suspended during four five-day sequences. Autotrophs and heterotrophs contained in biofilms were measured using flow cytometry to provide a time sequence of the appearance of micro-organisms on the glass slides. We have assessed the possibility of chemical preservation so that various locations can be sampled and sent to the laboratory. Flow cytometric counts were compared to fluorescent microscopic counts with good results. Two deployments of the panel set in Figure 1 were completed between July 8 and August 22, 2002, when smooth and roughened black and white plastic panels were retrieved after 10 and 20 days. Spectral reflectance, fluorescence yield, pigment concentration and cell counts by flow cytometry were measured on each panel at each time point. The depth study was deployed on August 21, 2002, with a planned duration of four weeks. Measurements were obtained after one week, but it was discovered after two weeks that fishing gear had become entangled with the mooring line that the panels sets were suspended on, the fisherman cut our gear to retrieve his sending our experiment to the bottom. Panel sets were recovered but the experiment was disrupted too late in the season to repeat.

RESULTS

For biofilm measurements, slides were retrieved daily and the film was brushed off into a scintillation vial. Suspensions were analyzed using a Cytomation MO-FLO flow cytometer. During the July to August period, photosynthetic autotrophs increased in the five day exposure from approximately ten thousand to two million cells per millimeter while DAPI-stained bacteria increased from seven hundred to nine million per millimeter. The fluorescent yield on all slides ranged from 0.5-0.6 indicating a highly efficient photosynthetic biofilm community. The submerged plastic panels showed the effects of fouling after ten days of submergence at one-meter depth when the reflectance spectra appeared to be dominated by suspended sediment settling on the horizontal surfaces. Figure 3 shows the spectral reflectance after three weeks of exposure. One-centimeter square scrapings from the

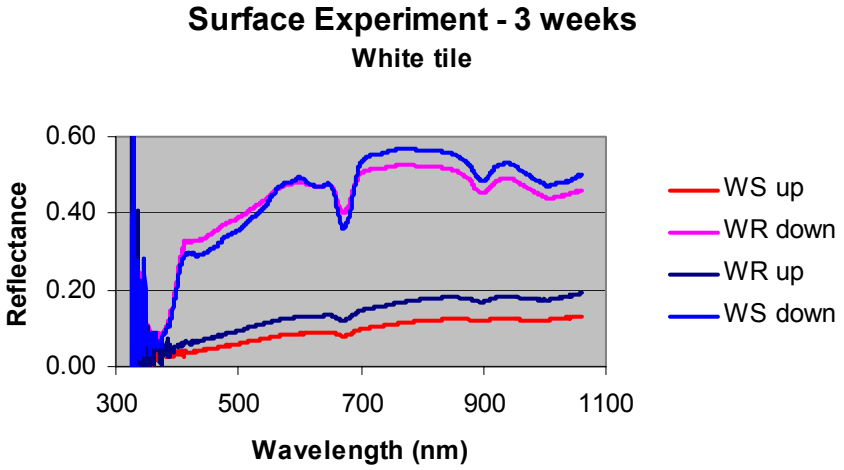

Figure 3. Spectral reflectance from white panels after three weeks at 1 meter. S – denotes smooth, R – denotes rough surface. Marked differences can be seen between upward and downward facing panels, all spectra show reduced reflectance at 687nm due to chlorophyll absorption.

panels contained large numbers of autotrophs, bacteria and ciliates. The fluorescence yield ranged from 0.2 to 0.6 and was variable. Pigment extracts showed that most of the pigment was in the degraded form of phaeophorbide. Even though the depth study was shortened to one week, differences were already apparent as a function of depth and between horizontal and vertical panels. The largest reduction in spectral reflectance was seen in horizontal panels at the 5% light level (Figure 4). In general, less fouling occurred at higher light levels and vertically panels were always more reflective than horizontal panels. This was an unexpected result as we had hypothesized that more fouling by autotrophic organisms would occur higher in the water column where more light was available for photosynthesis. This experiment was timed to occur at the end of the productive season in Gulf of Maine waters, perhaps resulting in a different pattern of fouling as a function of depth.

Figure 4. Fouling panel set at 5% light level (25 feet) after one week in Gulf of Maine coastal waters. Plastic panels can be seen on the left, glass panels over masonite on the right.

IMPACT/APPLICATIONS

The use of modern bio-optical tools, to our knowledge, has not been vigorously applied to biological fouling problems. In our opinion, flow cytometry will be especially useful to pinpoint the early sequence of biofilm succession and subsequent development of macro communities. This area of research needs some standard methods so that regional comparisons can be made.

TRANSITIONS

It is too early to say how effective and, hence, how useful these techniques will be. We will revisit this next year.

RELATED PROJECTS

We have developed close contacts with Dr. Mike Sieracki and colleagues at the Center for Flow and Imaging Cytometery at Bigelow Laboratory for Ocean Sciences, their expertise in sample preparation and analysis has been extremely helpful. Dr. Charles Mazel of Psicorp, Inc., has assisted with measurements of *in situ* reflectance using the DiveSpec.

PUBLICATIONS

D'Sa, E.J., J.B. Zaitzeff, C.S. Yentsch, J.L. Miller and R. Ives. 2001. Rapid remote assessment of salinity and ocean color in Florida Bay. In: The Everglades, Florida Bay and Coral Reefs of the Florida Keys: An Ecosystem Source Book. J.W. Porter and K.G Porter (eds.), CRC Press, NY, pp. 451-460.

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