## Optical Characteristics of Pollen Grains in Coastal Waters of the Gulf of Maine

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## LONG-TERM GOALS

In coastal areas, terrestrial material, including dissolved substances, can absorb a substantial fraction of the incoming light. We believe that seasonal inputs of pollen to coastal waters may contribute significantly to this terrestrial signal. We are describing the optical characteristics of pollen grains and determining if pollen leachates contribute to colored DOM. Eventually, we hope to assess the contribution of pollen to the optical signature of coastal waters on a seasonal basis and determine how light attenuation is altered in Case II waters where they are abundant.

#### **OBJECTIVES**

1. To characterize the optical signature of pollen grains.

2. To assess the confounding effects of pollen on typical phytoplankton absorption spectra and backscattering properties.

3. To quantify the contribution of pollen material to the optics of coastal waters. Attainment of this objective will require an eventual field effort to determine pollen's *in situ* properties.

## APPROACH

In Year 1, we collected mixed pollen samples throughout the flowering season for use in subsequent analyzes of optical properties. Based on the identification of the tree species that are the main source of pollen arriving at the shore, we collected monospecific pollen samples in year 2, by placing the pollen collector adjacent to the dominant species.

We determined absorption characteristics using the glass fiber filter pad technique (Phinney and Yentsch, 1991 and refs. therein) to measure spectral particulate absorption of mixed and monospecific pollen samples and mixtures of phytoplankton species and pollen. Absorbance is recorded between 250-750 nm on a Bausch and Lomb Spectronic 2000 spectrophotometer. Backscattering properties of the pollen will be evaluated using a Wyatt Industries Dawn laser,

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light-scattering photometer. This instrument measures the volume scattering function between 28 -  $160^{\circ}$  at 632 nm. These data can be used to estimate the backscattering coefficient over a range of pollen concentrations. We also examined *in vivo* fluorescent characteristics of the pollen grains using a Baird-Atomic spectral fluorometer.

To evaluate relative changes in phytoplankton absorption spectra due to the presence of pollen, we prepared mixtures of different coastal phytoplankton species (Table 1) and pollen in various ratios (0, 25, 50, 75, 100% pollen: phytoplankton cells). The species which were chosen were those in the same size range as pollen grains, those known to form surface populations, those known to occur at the same time of the year as the pollen releases, and/or with distinct optical signatures (e.g. coccolithophores).

We will also be evaluating the release of dissolved organic material (DOM), specifically yellow substances, by pollen grains, with and without exposure to UV wavelengths of light. Known quantities of pollen grains in filtered seawater, as well as a seawater control, will be placed in polycarbonate-screened and unscreened quartz bottles and placed under a UV light source at a constant temperature. Dark controls will also be included. Samples will be taken over time to evaluate the release of DOM and determine its absorption and fluorescent characteristics. Samples will be filtered through a  $0.2 \,\mu m$  filter and placed in a 10 cm spectrophotometer cuvette and measured on the instruments described above.

## WORK COMPLETED

We set up a passive aerosol collection unit at Bigelow to obtain mixed pollen samples throughout the flowering season, and also collected material from surface slicks in shoreline waters. Pine and birch pollens were identified as the two major sources and these types of pollen were collected directly from nearby trees. The pollen samples were stored frozen until they were filtered and examined microscopically. Pollen grains were identified and enumerated, and optimal concentrations for optical work were determined.

We determined absorption characteristics, using the glass fiber filter pad technique (Phinney and Yentsch, 1991 and refs. therein) to measure spectral particulate absorption of the mixed and monospecific pollen samples, and prepared mixtures of phytoplankton and pollen for analysis. Absorption spectra have been run on these mixtures and are currently being analyzed.

#### RESULTS

Samples were collected over the main flowering season (May - July) on a weekly basis. When pollen levels increased significantly, sampling was done more frequently. Samples were collected in two size fractions,  $20 - 53 \mu m$  and  $> 53 \mu m$ . In late June, the major release occurred and was dominated by pine pollen. Prior to that, there were smaller, defined releases of several other species. Pollen counts obtained from Eastern Maine Medical Center indicate that the pattern and magnitude of release were very consistent with those of the past five years. We knew

previously that pine pollen was a major contributor, but now know that earlier in the spring, the pollen of multiple hardwood species are also significant components. Pine pollen grains ranged from 60-75  $\mu$ m in size, while pollen grains of the other major species, birch, were considerably smaller (25-35  $\mu$ m).

Before beginning the optical analyses, we essentially performed a quench curve for the absorption spectra, determining the optimal amount of material to place on the filters. Due to their highly hydroscopic nature, we were also concerned about obtaining an even distribution of particles on the filters to be used in the analyses. Absorption and fluorescence spectra revealed that pine pollen grains appear to act as a long-pass filter, absorbing strongly in the UV and somewhat in the blue, and allowing the passage of longer wavelengths. There was a very long tail on the absorption spectrum, resulting in a fluorescent signal in all emission channels. The grains do have some autofluorescence, similar to flavin-like compounds. The absorption spectra of the mixed pollen samples were very similar, with most absorption occurring at less than 450 nm. In some of the surface water slick samples, there was a small peak at 680 nm, which we suspect may have come from contaminating phytoplankton. By careful, sequential screening and washing, we have been able to eliminate this peak to get the specific signal from the pollen material. We also supplied pollen material to C.S. Yentsch for analysis on a hyperspectral spectroradiometer (Analytical Spectral Devices) to determine the reflectance patterns of the pollen grains. The grains are highly reflective above 550 nm which is compatible with the absorption spectra (Figure 1). The ratio of wavebands 450:550, which is often used to distinguish phytoplankton, will be confounded significantly by the presence of pollen.

## **IMPACT/APPLICATIONS**

Remote sensing of ocean color from both aircraft and satellite platforms has the capability to quantitatively measure upper water column phytoplankton biomass if the signals can be quantitatively interpreted. The coastal waters of the Gulf of Maine are optically classified as extreme Case 2 waters, which implies that water-leaving radiance contains information on a complex mixture of optically-active substances. These include phytoplankton chlorophyll, phaeophytins and other pigments, phytoplankton and other detritus, suspended sediment, and colored DOM. To determine chlorophyll concentration with a high degree of accuracy, absorption due to competing substances must be accounted for and removed to isolate and quantify correctly the signal due to the phytoplankton. The accuracy of satellite systems cannot be tested without in-water validation, which must be done over a wide range of conditions - both seasonal and spatial - to come up with the best correction factors for the Case 2 algorithms under development. The seasonal wind-borne and river-borne contribution of pollen material to Maine and other coastal areas is significant. This material is likely to alter in situ attenuation in these waters and confound attempts to achieve an optical budget during the spring period. This work will provide information on pollen-specific optical properties and the relative importance of these yellow-colored particles in Case II waters.

### TRANSITIONS

None at this time.

### **RELATED PROJECTS**

Two other projects in which Keller is participating are of relevance to this project:

1. NOAA/NESDIS Project: Development and Validation of Regional Time-Varying Coastal Marine Algorithms - Gulf of Maine - A Case Study. (C.S. Yentsch et al, Bigelow; Keller - Associate Investigator)

Using the NOAA MARMAP cruises as a platform, optical properties of surface waters in diverse regions in the Gulf of Maine, including areas around river mouths, areas in which different phytoplankton blooms tend to occur, and areas where there are concentrations of yellow substances, are being measured. The direct goal of this project is to provide validated Case 2 algorithms for implementation into the NESDIS ocean color operational products. Keller is providing information on phytoplankton community composition and size structure.

2. NOAA/NESDIS Project: Penobscot Bay Marine Resource Collaborative: Applications of remote sensing and geographical information systems for marine resources management in Penobscot Bay, ME. Task 7: Characterization of phytoplankton communities, primary production and detrital components in Penobscot Bay and their relationships to hydrography. (M.D. Keller, PI, A.C. Thomas, U. Maine, co-PI)

The primary objective of this project, which is composed of ten distinct tasks, is the creation of a Penobscot Bay GIS which will contain layers of information essential to understanding the ecological character of the Bay. Our eventual objective is to contribute to the development of remote sensing techniques to monitor the Bay's primary production and for use in the management of marine resources. We are measuring optically active substances within the Bay on a seasonal basis and these data will become part of the larger NOAA/NESDIS ocean color data files being compiled to develop coastal algorithms.

## REFERENCES

Phinney, D.A. and C.S. Yentsch. 1991. On the contribution of particles to blue light attenuation in the sea. J. Plankton Res. 13, 143-52.

CCMP #	Species Siz	æ range (µm)	Isolation Data
118	Alexandrium tamarense	ca. 30	Gulf of Maine, 8/86
356	Ditylum brightwellii	10 x 90	Gulf of Maine, 12/86
628	Phaeocystis globosa	colonial, up to 1 mm	North Atlantic, 6/65
1321	Gymnodinium sanguineum	40-50	Long Island, US, 7/58
1330	Rhizosolenia setigera	20 x 300	Vineyard Sound, US, 1/59
1660	Pseudonitzchia multiseries	chain-former	Gulf of St. Lawrence, 12/93
1731	Oscillatoria sp.	colonial	North Atlantic, 4/96
1770	Ceratium longipes	180-250	Gulf of Maine, 1/97
1818	Coscinodiscus wailesii	115	Gulf of Maine, 3/95
376	Emiliania huxleyi	5	Gulf of Maine, 7/86

# Table 1. Phytoplankton isolates used in pollen experiments

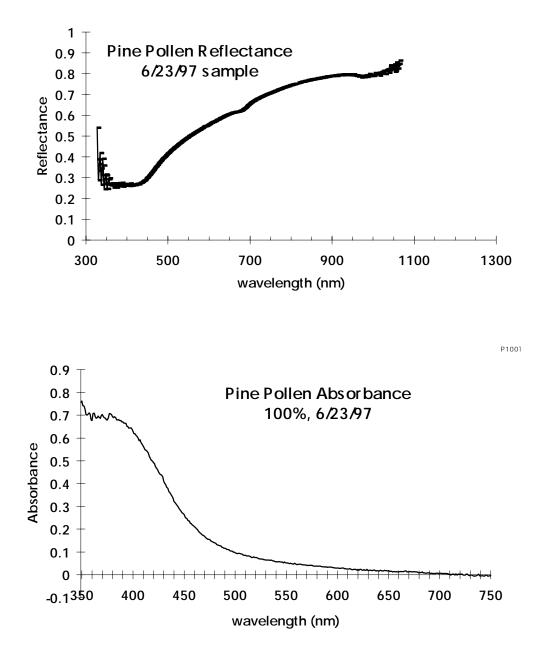


Figure 1. Examples of pine pollen absorption and reflectance spectra.