# Coastal Benthic Optical Properties (COBOP): Characteristics and Processes Related to Optical Properties of Benthic Marine Organisms and Substrates

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## LONG-TERM GOALS

The long-term goal of this research is to gain an understanding of the nature and significance of fluorescence and reflectance characteristics of benthic marine organisms in general, and coral reef cnidarians in particular. We wish to determine both how biological processes act to determine the optical properties and how optical measurements can be used to provide insight into biological state or process.

## OBJECTIVES

The objectives for this year's work were to:

- Evaluate several methods for separating the fluorescence and reflectance components contributing to spectral signatures under daylight illumination;
- Locate specimens that contain only one of each of the coral fluorescent pigments and make excitation and emission measurements for use as prototypes for a spectral unmixing algorithm.

## APPROACH

This work is part of the Coastal Benthic Optical Properties (CoBOP) program. The main effort in FY98 was conducted as part of the first large-scale CoBOP field campaign at the Caribbean Marine Research Center, Lee Stocking Island, Bahamas. The work described here was carried out by Eran Fux (doctoral candidate, Department of Ocean Engineering, MIT) under the supervision of the Principal Investigator.

*In situ* measurements of fluorescence and reflectance were made with the Benthic SpectroFluorometer (BSF)[1], a diver-operated instrument for measurement of spectral signatures from discrete benthic features. Laboratory measurements of fluorescence excitation and emission spectra were made with a FluoroMax-2 spectrofluorometer.

From the *in situ* measurements we identified a number of specimens that contained only one of the coral host fluorescent pigments. These specimens were brought to the laboratory for measurement of excitation and emission spectra. Several different methods were used for separating the fluorescence and reflectance components of spectral exitance: dual monochromator method; filter replacement method[2]; and our own modification of the filter replacement method.

# Report Documentation Page

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## WORK COMPLETED

Participation in the 1998 CoBOP field campaign was successful. Measurements were made to address the stated objectives. Data and sample analysis are well advanced. Some has been reduced to final form, while the remainder has been examined for quality and completeness.

## RESULTS

Prototype fluorescence excitation and emission spectra (figure 1) were collected for each of the previously described[3] coral fluorescent pigments. Figure 2 is an example of the use of prototypes to decompose a composite spectrum into the contributions of the individual pigments. A manuscript on the algorithm has been accepted for publication[4].

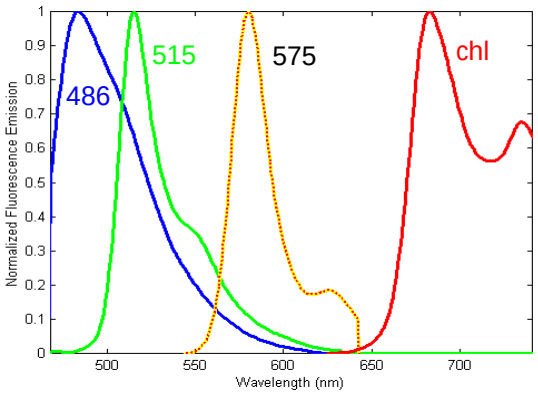


***Figure 1. Prototype fluorescence emission spectra***

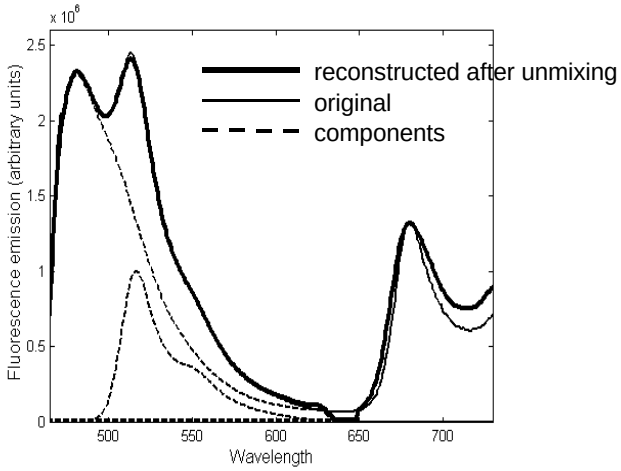


***Figure 2. Composite emission spectrum unmixed to components.***

Our modification of the filters method proved to be the most useful for separating spectral signatures into fluorescence and reflectance components (figure 3). The method can only be used for samples containing a single pigment in addition to chlorophyll in the zooxanthellae. Once the components have been separated the true reflectance curve clearly shows a minimum corresponding to the absorption (excitation) peak of the fluorescent pigment. These results will be presented at the Ocean Optics XIV conference in November 1998[5].

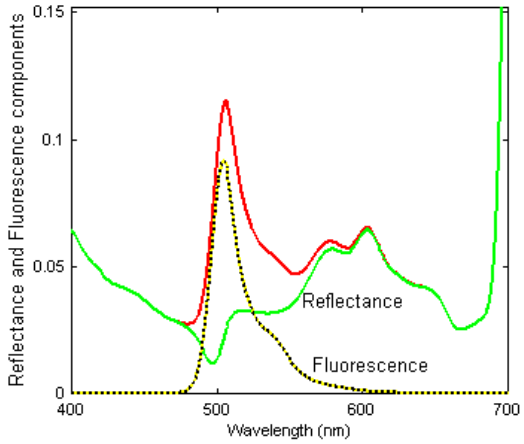


***Figure 3: Separation of the total spectral radiance factor (red) into contributions of fluorescence and reflectance. Note the reflectance minimum at about 500 nm due to absorption by the fluorescent pigment.***

## IMPACT/APPLICATIONS

The identification of spectral prototypes for the individual pigments supports an algorithm developed to unmix composite emission spectra from specimens with multiple pigments[4]. The unmixing technique will aid in the analysis of field data by helping us to identify the relative amounts of each of the pigments. Application of the algorithm has already indicated that there appears to be energy coupling between two of the coral fluorescent pigments. If correct, this implies that there is a more significant relationship between these pigments than has so far been expected.

The separation of the true reflectance and fluorescence contributions to the spectral signatures of some subjects coupled with the measurements of fluorescence yield provide the data needed to predict how those signatures will vary as a function of ambient illumination. This will be useful for interpretation of remote sensing spectral data and will be explored for the ecological implications of color and contrast change as a function of time of day.

## TRANSITIONS

We will work collaboratively with Curt Mobley to use Hydrolight™ to analyze the relative contributions of fluorescence and reflectance to spectral signatures under varying lighting conditions.

## RELATED PROJECTS

No related projects.

## REFERENCES

1 Mazel, C. H. "Diver-operated instrument for in situ measurement of spectral fluorescence and reflectance of benthic marine organisms and substrates", Opt. Eng. 36:2612-2617 (1997).

2 Allen, E. "Separation of the spectral radiance factor curve of fluorescent substances into reflected and fluoresced components", Appl. Opt. 12: 289-293 (1973).

3 Mazel, C. H. "Coral fluorescence characteristics: excitation-emission spectra, fluorescence efficiencies, and contribution to apparent reflectance", In Ocean Optics XIII, S. G. Ackleson, R. Frouin, Eds. Proc. SPIE 2963, 240-245 (1997).

4 Fux, E., and C. H. Mazel. "Unmixing coral fluorescence emission spectra and predicting new spectra under different excitation conditions", Applied Optics, accepted for publication.

5 Fux, E., and C. H. Mazel. "An experimental method to separate the fluorescence and reflectance components of the spectral signatures of corals", to be presented at Ocean Optics XIV, 1998.