

# ARE THE INHERENT OPTICAL PROPERTIES OF PHYTOPLANKTON RESPONSIBLE FOR THE DISTINCT OCEAN COLORS OBSERVED DURING HARMFUL ALGAL BLOOMS?

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## 1. ABSTRACT

Harmful algal blooms are becoming more frequent phenomena in the coastal environment. As this escalating trend continues, an early warning system based upon non-invasive rapid detection of harmful algal blooms is desired. A discoloration of water is often associated with these blooms, suggesting the feasibility of detection via ocean color measurements. Previous research has focused on absorption characteristics and pigment compositions of the algae as being responsible for the unique optical signatures observed. Harmful algal species do not contain unique pigments, and thus absorption alone can not explain the distinct changes in water color. However, taxon-specific optical properties, in particular scattering and backscattering, in combination with high concentrations of a monodispersed population may be responsible for the significant changes in ocean color during bloom events. We measured the inherent optical properties (IOPs) of several harmful algal species to determine the source for the frequently observed changes in ocean color during blooms. Four common harmful algal species (*Prorocentrum minimum*, *Gymnodinium splendens*, *Heterosigma akashiwo*, and *Aureococcus anophagefferens*) were investigated under controlled growth conditions to determine IOPs and particle size distributions (PSD) for exponential and stationary phase cells in order to understand how ocean color might change over the course of a bloom. Only slight distinctions in the shape of the absorption spectra were observed between species or growth phase, indicating that pigments are not responsible for the distinct ocean colors associated with blooms. The exception was *G. splendens* which was in a heterotrophic mode, and therefore contained few pigments, resulting in low absorption. The data demonstrate that absorption is not the source of distinct ocean color during harmful algal blooms, but rather the algal scattering and backscattering properties. The scattering and backscattering spectra are affected by cell size and growth phase, providing a major contribution to the changes in water color associated with blooms as the PSD changes from a polydispersed to a monodispersed population and as the phytoplankton physiology changes. Therefore our ability to detect these blooms optically will depend upon our ability to determine and interpret accurately scattering and backscattering spectra.

## 2. INTRODUCTION

There are approximately 60-80 phytoplankton species that comprise the category harmful algae. Their modes of destruction include the production of potent biotoxins, physical damage imparted to other organisms (i.e. grazers), the creation of eutrophic and anoxic conditions in their surrounding environment, as well as numerous other problems (Smayda, 1997; Taylor, 1990). The occurrence of these toxic outbreaks is increasing

worldwide and is the result of both an absolute increase in harmful algal blooms and the development of more advanced instrumentation with which to detect these toxic outbreaks. The increasing prevalence of toxic algae appears to be the result of such changing environmental conditions as enhanced temperature, nutrients, and storm-induced mixing events (e.g. Franks and Anderson, 1992; Smayda, 1990; Hallegraeff, 1993). These devastating blooms are usually initiated by eutrophication due to point source (e.g. industrial wastes and sewage disposal) and non-point source (e.g. agricultural runoff) inputs, a direct result of regional population growth. Although current technology has improved our ability to detect harmful algal blooms, most of the methods currently employed require laborious analyses of discrete water samples, resulting in a time lag between the onset of bloom formation and detection. Therefore it is necessary to develop non-invasive techniques that will allow faster determination of harmful algal cells on extensive spatial scales such that early warning systems can be implemented.

The fact that most harmful algal blooms impart distinct colors to the water suggests that optical methods may provide better detection and prediction of these blooms and can be employed as an early warning system such that the adverse effects of bloom events can be minimized. It has already been postulated that optical properties of the algae can be used to distinguish harmful algal blooms from mixed assemblages; although most of the studies thus far have been concerned with absorption characteristics (Cullen, 1997; Millie et al., 1997). Given that harmful algal species contain the same pigments as do their non-toxic counterparts; it is unlikely that absorption spectra alone can distinguish harmful species from a mixed algal assemblage. One species of toxic algae, *Gymnodinium breve*, does contain a unique photopigment, gyroxanthin diester, however, Millie et al. (1997) suggest that the presence of this different pigment still does not distinguish its absorption spectrum from that of other species; therefore it is not feasible to detect the occurrence of this species based solely upon absorption. If pigment compositions and absorption signatures are not responsible for the distinct ocean color often accompanying blooms, what is? The goal of this study was to determine the IOPs as a function of growth stage for a variety of algal species which comprise the group commonly referred to as red or brown tides to determine the source of the distinct color imparted to the water and how it may change over the course of the bloom.

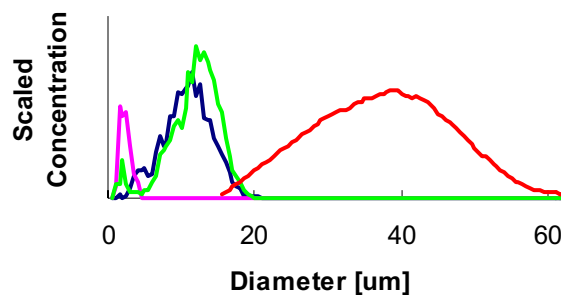
### 3. METHODS

Algal cultures were grown in *f/2* media (Guillard, 1975) at 17°C on a 14:10 hour light-dark cycle ( $100 \mu\text{E m}^{-2} \text{s}^{-1}$ ). Particle size distributions were determined using a Galai CIS 100 laser particle analyzer (Roesler and Iturriaga, 1994). Absorption and attenuation of each sample were measured using a WETLabs Histar (provided by S. Ackleson). Blanks of filtered culture media served as a correction for fluorescence signals and temperature and salinity effects (Pegau, 1997), and absorption was corrected for spectral scattering. The samples were then filtered onto a GF/F to measure filter pad absorption via spectrophotometry to ensure validity of data obtained from the Histar (Yentsch, 1962; Roesler, 1998). All of these measurements were made on cultures in exponential and stationary phase.

## 4. RESULTS

### 4.1 Optical properties for various algal species

The algal species selected for this study provided a range of cell sizes from approximately 1-50  $\mu\text{m}$  (Fig. 1). In addition to size differences, the general shape and morphology of the algae are also unique and may influence IOPs. *Prorocentrum minimum* is a 12  $\mu\text{m}$  dinoflagellate that is oval in shape with a flattened side view. Pores and spines are commonly observed on the surface of this alga. The Rhaphidophyte *Heterosigma akashiwo* is 13  $\mu\text{m}$  and is also oval. *Aureococcus anophagefferens* is a 1  $\mu\text{m}$  spherical Chrysophyte, and *Gymnodinium splendens* is a 46  $\mu\text{m}$  dinoflagellate oval to spherical in shape.

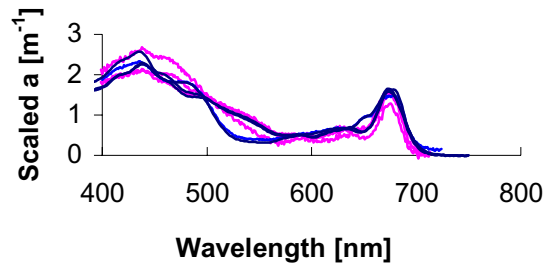


**Figure 1** Particle size distributions for algal cultures (magenta = *A. anophagefferens*, blue = *P. minimum*, green = *H. akashiwo*, and red = *G. splendens*).

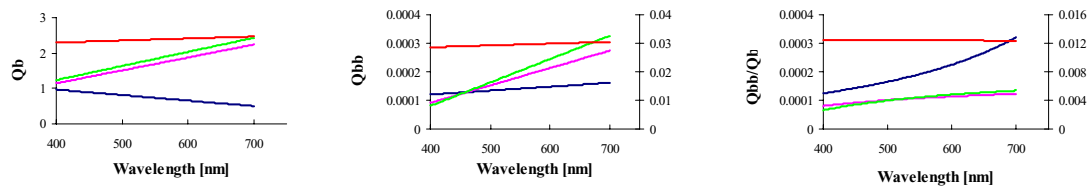
Not only did these algal species provide a broad range in PSD, their pigment compositions are also different (Jeffrey et al., 1997; Millie et al., 1997). Absorption spectra of various phytoplankton representatives (including several species that are not associated with harmful algal blooms), scaled to the area to exclude biomass effects, demonstrate that even with unique pigment compositions, the resulting shapes in absorption are not distinguishable from a mixed assemblage (Fig. 2). Spectral shape in absorption between species in this study (Fig. 4a) are similar and further demonstrate that absorption alone can not be responsible for the typical ocean colors observed during blooms.

Optical efficiency factors were modeled based on the PSD and measured absorption to investigate how spectral scattering efficiencies varied among species (Fig. 3). These efficiency factors were weighted for polydispersion (see Roesler and McLeroy-Etheridge, this issue), and they demonstrate a spectral dependence of scattering and backscattering on cell size. The larger cells, for example *G. splendens*, exhibited spectrally flat scattering characteristics, whereas the small cells, *A. anophagefferens*, showed a spectral decrease in scattering. Although the spectral shape of scattering and backscattering appear similar, the backscattering ratio ( $Q_{bb}/Q_b$ ) is different. Smaller cells tend to be more efficient at backscattering. High scattering values reported for the heterotrophic *G. splendens* are due its low pigment composition, thus making it behave

like a non-absorbing particle. The similarities in scattering profiles between *H. akashiwo* and *P. minimum* are the result of being similar in size. This suggests that harmful algal blooms formed by cells of the same size would be difficult to discern optically unless other information was available to distinguish them.



**Figure 2** Spectral shape of absorption for various phytoplankton species (magenta spectra represent species associated with harmful blooms, and blue spectra represent non-harmful species).



**Figure 3** Modeled spectral efficiency factors for a) scattering, b) backscattering, and c) backscattering ratio ( $Q_{bb}/Q_b$ ) (red = *G. splendens*, magenta = *P. minimum*, blue = *A. anophagefferens*, and green = *H. akashiwo*) Note that the scale on the right corresponds to *G. splendens* (see Roesler and McLeroy-Etheridge, this issue).

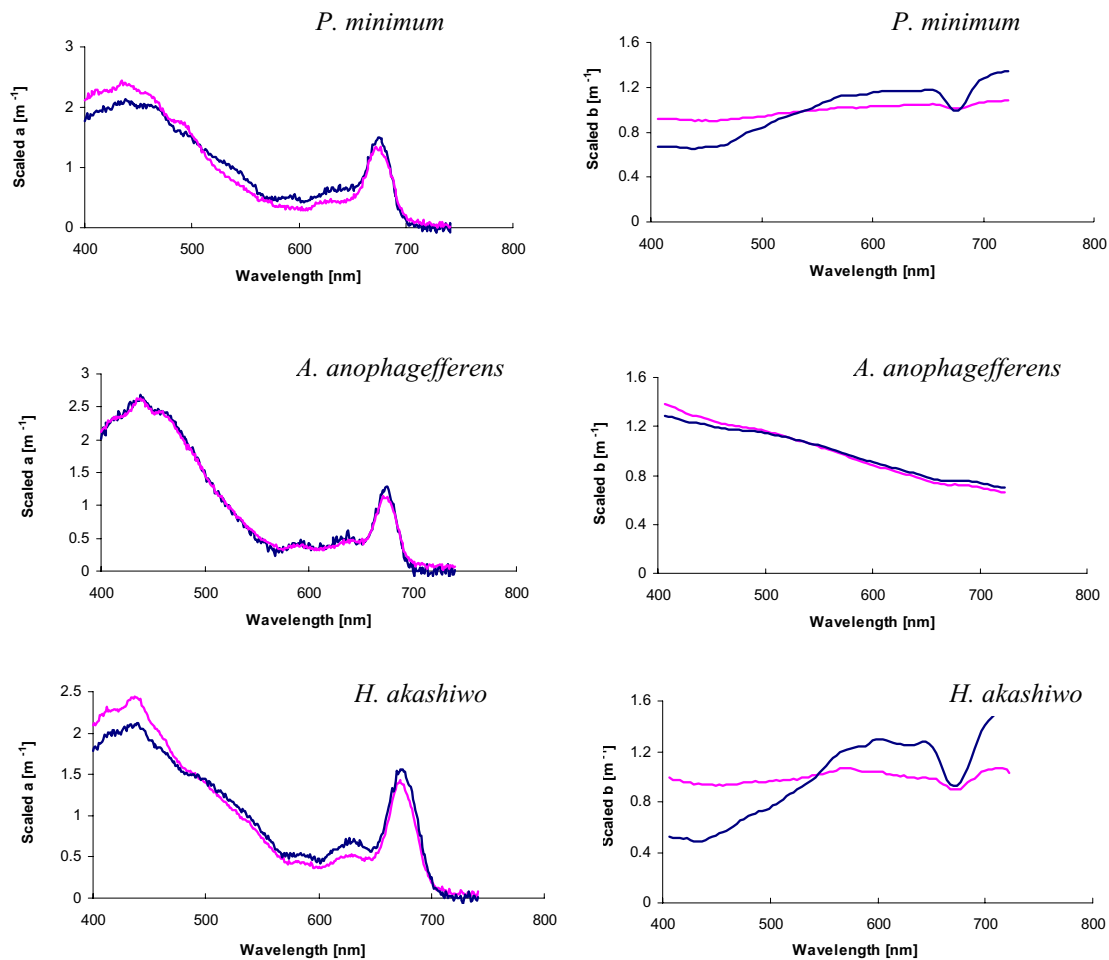
#### 4.2 Optical properties as a function of algal growth phase

Absorption spectra varied little between exponential and stationary phase cells (Fig. 4a). This is consistent with previous research that has shown harmful algae to be resilient to the changes in light environment over the course of a bloom (Harding, 1988). These algae appear to have rapid photoacclimation responses thereby maintaining constant relative pigment compositions and absorption spectral shape as the bloom progresses. This is in contrast to the spectral shape of scattering over the course of a bloom. The scattering coefficients vary with growth phase suggesting that the physiological changes in the phytoplankton induce variations in the index of refraction (Fig. 4b). Coats and Harding (1988) demonstrated that other cellular components (e.g. starch granules) change during a bloom as a function of light and nutrients. Therefore

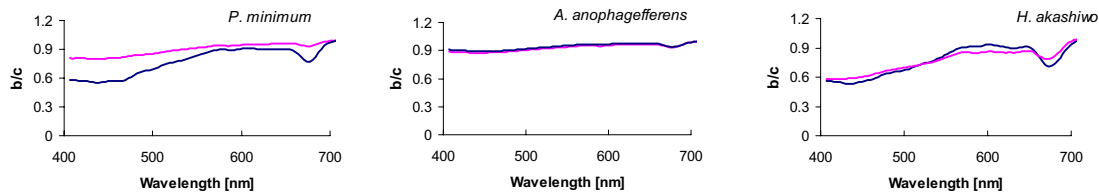
scattering characteristics measured remotely may provide information about the growth stage and algal physiology throughout the bloom.

Single scattering albedo (b/c), the probability that a photon will be scattered rather than absorbed, was determined for each of the three absorbing algal species in exponential and stationary phase (Fig. 5). As expected, scattering was a major component in the attenuation of light by the small phytoplankton, *A. anophagefferens*, and the scattering albedo was near one. The values did not vary with algal growth phase since the absorption and scattering spectra for this species was the same in exponential and stationary phase. Values for *P. minimum* and *H. akashiwo* were lower which is consistent with their larger size which would provide longer pathlengths over which more absorption could take place. Single scattering albedo for *P. minimum* was higher for the cells in exponential phase than those in stationary phase.

Spectra of IOPs as a function of growth stage are not shown from *G. splendens*, as this species exhibited very low absorption. This species was in a heterotrophic mode and contained few pigments; therefore, most of the attenuation of light by this species was due to scattering.



**Figure 4** Spectral shape of absorption and scattering versus algal growth phase (magenta = exponential and blue = stationary phase).



**Figure 5** Single scattering albedo for exponential (magenta) and stationary (blue) phase cells.

## 5. CONCLUSIONS

The present data suggest that absorption is not unique for harmful bloom-forming algae and is not a function of algal growth stage. Scattering coefficients varied between species and with growth phase, thereby contributing to the changes in ocean color during bloom events. Since ocean color can change over the course of the bloom depending on these properties; optical detection of harmful algal blooms is going to depend on the ability to measure and interpret accurately scattering and backscattering. Because the IOPs, in particular scattering characteristics, provide information about cell size and growth phase, we can use optical methods not only for the detection of these blooms but also to lead to a better understanding of the processes involved in bloom dynamics.

Although the combination of PSD and optical properties allow for better detection and prediction of bloom formation, limitations exist in that non-harmful algal blooms or other factors leading to changes in ocean color may generate false alarms. Therefore optical monitoring will not replace the necessary analysis of discrete water samples for toxin detection; however, it can serve as a warning system such that precautions can be taken to minimize the potential destruction that may occur if the bloom is toxic. Once the bloom status has been determined, continued use of optical methods will allow the progress of those existing blooms to be tracked thereby providing an early warning to other areas likely to be impacted by the advection of these waters containing harmful species.

## 6. ACKNOWLEDGEMENTS

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