Optical detection and assessment of algal blooms

John J. Cullen, Aurea M. Ciotti, Richard F. Davis, and Marlon R. Lewis

Center for Environmental Observation Technology and Research, Department of Oceanography, Dalhousie University, Halifax, Nova Scotia B3H 4J1

Abstract

Concerns about harmful algal blooms (HABs) have grown in recent years. There is a pressing need for robust, quantitative, and cost-effective methods to detect and characterize algal blooms. Critical applications of these methods include long-term monitoring of coastal waters to indicate the degree to which present trends of HABs and human activities are linked, early warning systems to protect aquaculture sites and to signal the need for further investigations, and systems to characterize synoptically the distributions and physiological state of phytoplankton in an oceanographic context. Because phytoplankton strongly influence the fate of light in the ocean, optical methods are well suited for HAB applications. Passive optical systems can measure ocean color or the penetration of solar irradiance in surface waters; both properties can be related to the constituents of natural waters, including phytoplankton. The sensors measure radiometric quantities: consequently, rigorous calibration is possible and measurements can be compared between sites and over long periods of time. One passive optical system—a radiometer buoy—is shown to be useful for characterizing biological variability in surface waters over scales from minutes to months. A red tide was easily observed in measures of ocean color from the buoy; sensors for downwelling irradiance detected a subsurface bloom. Some optical instruments use artificial illumination to determine optical properties such as the coefficients of absorption and scattering. These measures can be made continuously in situ and effectively related to phytoplankton. Several types of fluorometers can be used to characterize the abundance, pigmentation, and physiological state of phytoplankton. All of these optical technologies should be useful for the study of HABs, but biological interpretations of many optical measurements need further testing.

Environmental and economic impacts of harmful algal blooms (HABs) have increased in recent decades (Smayda 1995; McGowan 1995). The possible influences of human activities on estuarine and coastal ecosystems, during these same years, some marine ecosystems have undergone fundamental changes that are apparently associated with natural variability of climate (Roemmich and McGowan 1995; Sugimoto et al. 1995). The possible influence of anthropogenic changes in greenhouse gases is also a concern (Fraga and Bakun 1993; Tester et al. 1993). In order to ensure sustainable development of critical coastal resources, it is imperative to know the degree to which present trends of HABs and human activities are linked (e.g. Kahru et al. 1994), if those trends will lead to unacceptable consequences, and if the means can be developed to mitigate harmful effects. Essential to this effort will be reliable, quantitative, and practical means to detect and characterize HABs. Detection is required not only to describe the frequency of blooms and long-term trends, but also for the protection of aquaculture sites and for implementation of measures to protect public health.

Historically, algal blooms have been noticed because the water turns red, brown, or yellow. Now we have relatively simple optical instruments to tell us quantitatively what a trained observer can easily see: that the water has changed color, and it is probably due to algae (Gower et al. 1984; Carder and Stewart 1983; Cullen et al. 1994; Gitelson et al. 1994). In addition, phytoplankton pigment can be detected effectively with in situ fluorometers (see Falkowski and Kolber 1995). If such optical observations can be made autonomously and interpreted reliably, they would be ideal for the detection of HABs in coastal waters, even in remote locations. Species composition of blooms cannot yet be determined reliably from optical measurements (discussed below), and some harmful species can profoundly affect coastal resources without dominating the phytoplankton (Anderson 1983) or the optical characteristics of the water, so there are limitations to the usefulness of optical instruments for detecting HAB phenomena. Nonetheless, continuous optical measurements in coastal waters would be extremely useful for describing bloom dynamics and long-term trends as well as for detecting threats to public health or aquaculture.

Although critical factors for bloom formation have been described for some species (Seliger et al. 1970; Anderson and Morel 1979; Tyler and Seliger 1981; Franks and Anderson 1992b; Honjo 1993; Figueiras et al. 1994; Watanabe...
et al. 1995), we have only a crude understanding of the eco-
physiological mechanisms that promote and maintain harm-
ful algal blooms. What adaptations and hydrographic or tro-
ic conditions favor one species over another? Can the
success of toxic vs. nontoxic species be predicted on the
basis of some general principles, at least for some species
groups? At this time, we simply do not know. More research
is needed, but it must be focused on integratingphysiologi-
ical ecology with modern oceanographic field research.
Physiological characterization of harmful phytoplankton in
an oceanographic context is key to studying the environ-
mental regulation of toxic and other harmful effects, the
challenge is to develop effective methods for synoptic de-
scriptions of abundance and physiological condition of phy-
toplankton. We feel that optical techniques are particularly
well suited to this task.

Without a doubt, there is a pressing need for robust, quan-
titative, and cost-effective methods to detect and characterize
algal blooms. There are many different applications, how-
ever, and no single approach is appropriate for all. With that
in mind, we review several different technologies for the
optical detection and characterization of algal blooms, pro-
viding an introduction to the literature and emphasizing in
situ measurements. Then we focus on our own research, de-
scribing one system that has good potential for autonomous
detection of blooms and for long-term monitoring of coastal
systems. Finally, we discuss the future of optical measure-
ments in HAB research, identifying areas for further research
and suggesting which approaches may be well suited for
particular applications.

Critical applications for HAB research

Long-term monitoring—Long, quantitative time series of
the abundance of phytoplankton or zooplankton are essential
to discerning trends in planktonic systems (Tont 1976; Harris
1986; Falkowski and Wilson 1992; Harding 1994; Roem-
mich and McGowan 1995). Ancillary data, such as temper-
ature, salinity, and nutrient concentrations are required for
discriminating the environmental factors which have the
strongest influence on biological patterns. Long-term records
from the Seto Inland Sea of Japan provide an excellent ex-
ample of how a continuous series of key, albeit simple, mea-
surements can illustrate a clear link between coastal eutro-
phication and the occurrence of harmful algal blooms (Fig.
1). For HAB research, then, it would be desirable to collect
long-term, quantitative records of bloom occurrence along
with ancillary oceanographic measurements.

Especially in less populated areas where ephemeral events
might not be noticed and where discrete sampling programs
are costly and inappropriate for characterizing short-term vari-
ability, bloom occurrence would be better quantified with
autonomous, continuous measurements supplemented with
satellite observations (see Kahrn et al. 1994). Even simple
observations (e.g. temperature, Secchi depth, zooplankton
volume) are extremely valuable if made for long enough
periods (Eppley and Holm-Hansen 1986; Falkowski and
Wilson 1992; Roemmich and McGowan 1995). Multiple
measurements are better yet, especially if they can describe
physical as well as biological variability (Chelton et al.
1982). It would also be extremely useful to compare long-
term records from a variety of coastal environments to look
for similarities and differences in the relationships between
hydrographic processes and the occurrence of blooms. For
example, if the frequency of blooms can be linked to coastal
eutrophication on the basis of quantitative analysis rather than
on qualitative impressions about deterioration of an ecosys-
tem. Long time-series of quantitative measurements are thus
central to management decisions.

Critical requirements for long-term monitoring, then, are
that measurements are quantitative, comparable between
sites, and will not be compromised as science advances. App-
propriate measurements include temperature, conductivity,
and Secchi depth (see Lewis et al. 1988; Falkowski and Wil-
son 1992). Unfortunately, methods for making important bi-
ological observations (e.g. cell counts, chlorophyll concen-
tration, primary productivity) have changed over the years
(through the introduction of epifluorescence microscopy,
flow cytometry, filters with smaller pores, high performance
liquid chromatography, radioactive tracers, and trace-metal
clean techniques; making it problematic to discern long-term
trends. We will show below that in situ optical observation
of the ocean is particularly well suited for long-term moni-
toring because properly designed instruments can measure
radiometric quantities that will retain their validity indefi-
nitely, even if the biological interpretations of those mea-
surements change.

Systems for long-term monitoring must be cost-effective,
so it is important to develop observation technologies that yield useful, quantitative, and affordable information. Ultimately, it is appropriate to compare the expense of autonomous systems to the high cost associated with conventional sampling programs.

**Early-warning systems**—The benefits of long-term records are so great that a strong case can be made for establishing well instrumented moorings in strategic locations (Dickey 1991; Chavez et al. 1994; Hansen 1995). It is not likely, however, that such systems will ever be plentiful enough to provide the spatial coverage to determine when and where blooms are initiated. This information is crucial to process studies, which should be set into motion as soon as possible after a bloom commences, and also to fish-farmers, whose enclosures might be threatened by a bloom. Hence, there is a need for inexpensive but robust early-warning systems. If one is interested in detecting explosive increases in algal biomass, then relatively simple and imprecise detection systems should be adequate. However, some toxic phytoplankton can have strong impacts when they are present in relatively low concentrations and are therefore undetectable as dominant populations (Anderson 1983). Immunological approaches might be useful for their detection (Scholin et al. 1997). Here, we will discuss optical measurements that could be used for early warning of algal blooms.

**Oceanography and physiological ecology of HABs**—Laboratory studies provide the foundation for oceanographic research on the physiological ecology of HABs (e.g. Harrison 1976; Anderson et al. 1990; Flynn et al. 1994), but oceanographic insight can be developed best if the characteristics of cultured phytoplankton can be related quantitatively to natural populations (Cullen and Horrigan 1981; Tyler and Seliger 1981; Watras et al. 1982; see also Watanabe et al. 1995). The critical requirement for oceanographic research on HABs is synoptic descriptions of planktonic processes in relation to important environmental factors (irradiance, nutrients, water-column stability, etc.). Analysis of historical records can be productive (Franks and Anderson 1992a), but what we really need are tools to describe the distribution and physiological state of phytoplankton in three dimensions, along with characteristics of the physical environment. Optical methods are ideally suited to synoptic and quasi-synoptic characterization of surface waters, and, as we show below, some of these methods are particularly good for describing the distributions and physiology of phytoplankton.

**Rationale for detection of algal blooms with passive optical measurements**

Measurements of ocean color (Fig. 2) and the fate of light in the ocean are extremely useful for describing biological dynamics in surface waters (Yentsch 1960; Lorenzen 1972; Smith et al. 1989); thus, the oceanographic community has made a substantial commitment to remote sensing of ocean color from space (Aiken et al. 1992; Mitchell 1994) and the development of new technologies for in situ optical observation of the sea (Dickey 1991). The fields of hydrological optics and bio-optical oceanography have advanced rapidly as a consequence (Gordon et al. 1988; Spinrad 1989; Mobley 1994; Ackelson 1995), and optical systems have figured prominently in a number of oceanographic studies (Dickey 1991; Marra et al. 1992; Abbott et al. 1995). Means, a great deal of research has been conducted on toxic phytoplankton and harmful algal blooms (e.g. Okaichi et al. 1989; Smayda and Shimizu 1993). Although optical technologies have been applied to HAB research (e.g. Carder and Steward 1985; Uno and Yokota 1989; Millie et al. 1992; Johnsen et al. 1994), and optical monitoring systems have been proposed (Yentsch 1989) and implemented (Pettersson et al. 1993), recent literature (e.g. Okaichi et al. 1989; Smayda and Shimizu 1993) suggests that many research and monitoring programs throughout the world would benefit from having greater information about and access to optical technologies. Oceanographers and limnologists have for many years studied the influence of particles (especially phytoplankton) on the transmission of light in aquatic systems (Yentsch 1960; Talling 1981). Because satellites provide unprecedented opportunities to describe the distributions of phytoplankton in the open ocean, and because phytoplankton are the dominant influence on the color of these waters, much of the research on phytoplankton and ocean optics has been done for waters isolated from terrestrial influences (Morel 1988). The potential for using passive optical observations to characterize coastal waters has been recognized for some time, however, and studies have demonstrated clearly that optical measurements can provide useful information about the constituents of coastal waters, including algal blooms, despite the complication of dealing with the influence of terrestrial runoff (Carder and Steward 1985; Sathyendranath et al. 1989; Tyler and Stumpf 1989). A central objective of

![Fig. 2. Measures of ocean color (upwelling radiance at wavelength $\lambda$, $L_u(\lambda)$, normalized to downwelling irradiance at 490 nm, $E_d(490)$) as recorded with a tethered spectral radiometer buoy (described by Cullen et al. 1994) in coastal waters off the coast of Oregon. Medians ± upper and lower quartiles for records of 5 min recorded at 1 s$^{-1}$: 139 km offshore, 63 km offshore, and during a diatom bloom in the plume of the Columbia River, ~8 km offshore. Patterns are very similar to those that stimulated interest in remote sensing of ocean color (Hovis et al. 1980).](image-url)
retrieving more and better information from measurements of ocean color have been described many times, and methods for deriving chlorophyll concentration from measurements of ocean color are being constantly improved (see Morel et al. 1989; Abbott et al. 1995). Many approaches is to detect and quantify the characteristic spectral absorption of phytoplankton in the presence of other absorbing and scattering constituents of surface waters. Much ocean-color research has been directed toward understanding and interpreting measurements from radiometers positioned above the surface, but in-water optical measurements figure heavily in the development and validation of theory and algorithms (e.g. Austin and Petzold 1981; Smith et al. 1989; Abbott et al. 1995).

The principles behind optical detection of near-surface pigments have been described many times, and methods for retrieving more and better information from measurements of ocean color are being constantly improved (see Morel 1990; Kirk 1994; Bukata et al. 1995 for overviews). Here we describe some general principles that are directly relevant to detecting phytoplankton with a variety of optical measurement systems, beginning with the basis for estimating chlorophyll concentration from measurements of ocean color (see Lewis and Cullen 1991).

### Light absorption by phytoplankton and the optical properties of surface waters

A satellite ocean-color sensor measures spectral radiance \( L_{\text{rs}}(\lambda) \) (terms defined and units given in list of notation). This property depends on solar irradiance, scattering from and transmission through the atmosphere, reflection from the sea-surface, and the optical properties of surface waters. After elimination of surface reflection and atmospheric factors, what remains is upwelling radiance \( L(\lambda) \) just beneath the surface, the spectral variation of which defines ocean color (see Gordon et al. 1988). Because upwelling radiance is essentially reflected light, much of the variability in \( L(\lambda) \) is due to changes in downwelling solar irradiance \( E_{\text{d}}(\lambda) \). A property of principal interest, then, is radiance reflectance \( R_{\text{r}}(\lambda) \), the ratio of upwelling radiance to downwelling irradiance.

Radiance reflectance can be described as a ratio of two optical properties, irradiance reflectance, \( R \) (the dimensionless ratio of upwelling irradiance, \( E_{\text{u}} \) to \( E_{\text{d}} \)), and \( Q \), the ratio of upwelling irradiance to upwelling nadir radiance. From here on wavelength dependence is implicit:

\[
R_{\text{r}} = \frac{L(\lambda)}{E_{\text{d}}(\lambda)} = \frac{R}{Q},
\]

In turn, \( R \) depends on inherent optical properties (IOPs) of surface waters, i.e. the absorption coefficient, \( a \), and backscattering coefficient, \( b_{\text{b}} \):

\[
R = \frac{b_{\text{b}}}{a + b_{\text{b}}},
\]

Inherent optical properties are characteristics of the water that are independent of the spatial distribution of the radiance field. Although irradiance reflectance is strongly dependent on IOPs, it is also influenced by the natural light field and is thus considered an apparent optical property (AOP) because it depends not only on the IOPs of the water itself (\( a \) and \( b_{\text{b}} \)), but also on the radiance field (see Gordon et al. 1988).

Note that water, phytoplankton, other suspended particles, and dissolved organic matter (DOM) contribute to \( a \) and \( b_{\text{b}} \). The key to remote sensing of algal biomass is that phytoplankton have characteristic absorption bands in the visible whereas the absorption by water or DOM as well as backscatter spectra lack strong spectral features in the same range (Kirk 1994). Consequently, under many conditions, algal absorption is detectable in reflectance. Algorithms for the CZCS (Coastal Zone Color Scanner), and other remote-sensing algorithms consider ratios of wavelengths (e.g. Harding et al. 1995 and references therein), in part because spectral variation of backscatter is more conservative than that of absorption, hence variation of algal absorption dominates the ratios (see also Stumpf and Tyler 1988 for an approach that exploits contributions of absorption vs. backscatter to red vs. infrared reflectance). If we could measure reflectance in absolute units (i.e. near-surface in situ, or from above the surface at \( E_{\text{d}} \) and \( E_{\text{u}} \) can be well characterized; see Bukata et al. 1988), then constituents of surface waters could be determined by inverse modeling (Duerffert and Fischer 1994; Roesler and Perry 1995). For a more general illustration of how absolute values of \( R \) can be more informative than ratios of \( L \) at different wavelengths, consider that the enhanced backscatter associated with a coccolithophore bloom (Akeleison et al. 1988) would cause \( R \) to increase in all visible wavebands, but it would reduce the degree to which chlorophyll absorption is manifest in the ratio of blue : green upwelling radiance (see Aiken et al. 1992 and references therein).

The relationship between algal absorption and spectral reflectance is such that contributions from backscatter (e.g. Ulloa et al. 1992) must be accounted for to quantify the contribution of phytoplankton. A different AOP of surface waters, the diffuse attenuation coefficient for downwelling
irradiance \([K_d(\lambda)]\) is also closely related to absorption, but in a different way (Gordon 1989):

\[
K_d = \frac{(a + b_h)}{\bar{\mu}_d}
\]

where \(\bar{\mu}_d\) is the average cosine of the downwelling radiance field and spectral dependence is again implicit. Generally, \(b_h\) is much less than \(a\) and its contribution to \(K_d\) can be ignored. Thus, \(K_d \approx (a/\bar{\mu}_d\)\). Variations in \(\bar{\mu}_d\) can be characterized well enough to permit retrieval of \(a\) from measurements of \(K_d\) (Kirk 1994; Berwald et al. 1995). Diffuse attenuation cannot be measured directly from airborne sensors (but see Austin and Peltzold 1981); however, it is readily determined with profiling radiometers (Fig. 3), or it can be recorded continuously with sensors that measure \(E_d\) at depth. Variability of \(K_d\) can then be related to the constituents of water, including chlorophyll (Aiken 1990; Smith et al. 1991; Abbott et al. 1995).

To recapitulate: apparent optical properties of surface waters can be measured and analyzed to estimate the absorption of light due to constituents of the water. Absorption of light by phytoplankton \(a_{np}\) can be discerned because the characteristic absorption bands are in many cases readily distinguishable in the presence of other constituents of the water. Once \(a_{np}\) is measured or estimated, it is still necessary to relate absorption to algal biomass and species composition (see also Morel 1990; Hoepffner and Sathyendranath 1992; Garver et al. 1994). Applications specific to HABs are discussed elsewhere in this issue (Millie et al. 1997).

**Solar-induced fluorescence of phytoplankton**—Theory notwithstanding, it can be very difficult to estimate the abundance of phytoplankton in optically complex coastal waters, where DOM and suspended sediment produce a brown color to which the contribution of phytoplankton is hard to discern. Under such circumstances, and whenever phytoplankton are present in moderate concentrations, the presence of chlorophyll can be detected in solar-induced chlorophyll fluorescence near 683 nm (Neville and Gower 1977; Gordon 1979; Kiefer et al. 1989; Cullen and Lewis 1995; see also Fig. 2). Careful consideration of fluorescence emission spectra and hydrological optics is required for accurate measurement of solar-induced fluorescence (Roesler and Perry 1995), and interpretation of those measurements in terms of algal biomass and photosynthesis requires basic information on algal physiology that is presently sketchy at best (Chamberlin et al. 1990; Chamberlin and Marra 1992; Kiefer and Reynolds 1992; Lizotte and Priscu 1994), especially for irradiance levels characteristic of the sea surface (Cullen and Lewis 1995). Regardless, field measurements of solar-induced fluorescence have been related to chlorophyll and photosynthesis under a variety of conditions (e.g. Gower et al. 1984; Kiefer et al. 1989; Stegmann et al. 1992; Gitelson et al. 1994; Abbott et al. 1995).

**Measurements requiring artificial illumination**

So far we have discussed optical properties that can be measured passively and used to estimate the absorption or fluorescence of phytoplankton. Passive observation is attractive for many reasons: it can be used in remote sensing; the radiometric quantities that are measured (e.g. radiance and irradiance) are very well defined, so instruments can be accurately calibrated with standard procedures (Mueller and Austin 1995); and passive instruments have very low power requirements, hence they are well suited for long deployments in remote locations. The relationships between algal absorption and easily observable optical properties of surface waters are imprecise, however, and can be strongly influenced by environmental conditions. The interpretation and calibration of solar-induced fluorescence signals is also uncertain. Thus, there is good reason to consider instrumentation that uses artificial illumination to measure the interaction of phytoplankton with light in the ocean. Instruments using artificial illumination have the added advantage of being able to measure optical variability at night and below the photic zone. Some measurements require the concentration of particles or other manual procedures and are thus not suitable for continuous measurements in situ. They can be adapted for use in situ, so they are discussed here along with measurements that are made with submersible instruments.

**Measurement of absorption**—We have shown that estimation of absorption is key to optical detection of phytoplankton in surface waters. The absorption coefficient of a suspension can be measured spectrophotometrically if modifications are made so that photons scattered out of the light path (principally in the forward direction) are detected with the same efficiency as those that are neither scattered nor absorbed (see Shibata 1958). However, spectrophotometry is too insensitive for use on unconcentrated seawater (cf. Nelson et al. 1993). Instead, absorption by particles can be measured by collecting them on a glass-fiber filter, which serves the dual purpose of concentrating the sample and acting as a diffuser for ensuring the detection of forward-scattered light. After absorbing with chlorine or nitric acid, the samples are digested and measured spectrophotometrically.
light (Yentsch 1962). One can also use a highly portable spectroradiometer to measure the reflectance of samples on filter pads, and the spectra can be analyzed to estimate absorption (Balch and Kilpatrick 1992). Filter-pad methods provide a great deal of useful information (Bricaud et al. 1995 and references therein), especially when their excellent spectral resolution is exploited (e.g. Johnsen et al. 1994). However, the measurements must be corrected for the toruous path of light through the sample (Mitchell and Kiefer 1988a; Cleveland and Weidemann 1993). Methods are being improved (Tassan and Ferrari 1995), and a freeze-transfer technique eliminates the need for a pathlength correction (Allali et al. 1995), but still, the measurements require hands-on manipulation of samples.

Systems have recently been developed for continuous in situ measurement of the absorption coefficient of natural waters (Hakvoort et al. 1994; Saito et al. 1994; Pegau et al. 1995), and absorption meters that use a quartz cylinder to reflect scattered photons toward the detector (Zaneveld et al. 1990) are now commercially available. If beam attenuation ($c$, an IOP: $c = a + b$) is measured at the same wavelength as $a$, the scattering coefficient $b$ can be calculated by difference. The scattering coefficient is also used in the calculation of a correction factor for photons scattered away from the detector of the absorption meter (Zaneveld et al. 1994). To retrieve the IOPs of surface waters and relate them to biological constituents and AOPs, one would also measure $b$, using an optical backscatter detector (Voss and Smart 1994). However, algal blooms can be detected more simply by measuring absorption at three key wavelengths to characterize the red peak of chlorophyll absorption; such a system has been successfully deployed for up to 6 months in the Bering Sea, with encouraging results (Fig. 4). Fouling of the reflecting tube was retarded by periodic flushing with bromine dissolved in seawater (Davis et al. 1997). As with most measurement systems (e.g. Abbott et al. 1995), validation of instrument performance during extended autonomous deployments is difficult.

We should recognize that the technology for measuring absorption in situ is still emerging. Despite the sound theoretical foundations of the measurement systems, accuracy and precision is not assured; for example, although several methods show the same general patterns in absorption, measured ratios between wavelengths, as well as absorption coefficients, differ between instruments (Pegau et al. 1995). Thus, it may be some time before detailed analysis of in situ absorption spectra (e.g. Johnsen et al. 1994; Millie et al. 1997) will be practical (see also Garver et al. 1994). However, improvements are being made rapidly and further research on the optical characteristics of marine particles is surely warranted.

Measurement of beam attenuation and scattering—Measurements of spectral beam attenuation have been made in a variety of environments (Voss 1992; Volent and Johnsen 1993), and transmissometers that measure $c$ at 660 nm are now used routinely in oceanographic research. The data have proved to be extremely useful in several contexts (Pak et al. 1988; Siegel et al. 1989; Stramska and Dickey 1992). In surface waters, the scattering coefficient is generally much greater than the absorption coefficient, so measures of $c$ are dominated by $b$, which for phytoplankton has weaker spectral features in the visible range than does absorption (Morel 1990; Stramski and Reynolds 1993; Roesler and Zaneveld 1994). Thus, although it is feasible to measure spectral $c$ to detect phytoplankton (Volent and Johnsen 1993), on first principles the approach is less sensitive than absorption-based techniques (cf. figures 6A and 6C of Stramski and Reynolds 1993). The same conclusions apply to measurements of spectral backscatter (Voss and Smart 1994): they are very important for inverse modeling and for characterizing particle assemblages (Stramski and Kiefer 1991; Ulloa et al. 1992), but spectra of $b$ for phytoplankton have very weak features in the visible region (Stramski et al. 1995), and are thus not very useful on their own for discerning the contribution of phytoplankton.

Fluorometry to estimate the biomass of phytoplankton—Since the introduction of flowthrough in vivo fluorometry 30 yr ago (Lorenzen 1966), fluorometry has been used routinely as an extremely sensitive and specific method for describing the distributions of marine phytoplankton (see Cullen 1982; Falkowski and Kolber 1995). Fluorometry is useful not only because it measures a signal that is dominated by the contribution of phytoplankton, but also because measurements can be made continually during transects, vertical profiles, or long deployments. In situ fluorometers (Derenbach et al. 1979) and fiber-optic fluorometers (Cowles et al. 1989; D'Sa et al. 1994) provide extremely good spatial resolution.
Calibration of fluorescence as a measure of chlorophyll has been an issue from the outset, and studies have shown that the relationship between fluorescence and chlorophyll \( a \) (Chl) is influenced by irradiance (Kiefer 1973a,b; Vincent 1980; Marra 1992; Falkowski and Kolber 1995), species composition (Strickland 1968; Heaney 1978), nutritional state of phytoplankton (Kiefer 1973a; Sakshaug and Holm-Hansen 1977), and the type of fluorometer used for the measurement (Harris 1980; Neale et al. 1989). Regardless, measurements of chlorophyll fluorescence in situ are extremely useful for describing spatial and temporal variability of phytoplankton (Denman and Platt 1975; Dickey 1991; Marra et al. 1992; Abbott et al. 1995; Falkowski and Kolber 1995). For accurate estimation of chlorophyll in surface waters, stimulated fluorescence should be regularly calibrated with measurements of Chl, and the effects of bright light on fluorescence yield (i.e., fluorescence quenching, see Falkowski and Kolber 1995) should be considered explicitly (Cullen and Lewis 1995).

**Spectral fluorometry**—For studies of phytoplankton, most in situ fluorometers use broad-band blue irradiance for excitation and they detect the red emission of Chl near 683 nm. If more wavelength combinations are measured, it is possible to discern groups of phytoplankton on the basis of differences in accessory pigments that can be detected in spectra of excitation and emission of fluorescence (Yentsch and Phinney 1985; see also the interpretation of fluorescence induced by an airborne laser, Hoge and Swift 1983). Pigment-specific spectral fluorescence yield is also useful in determining light utilization efficiency for photosynthesis (Sosik and Mitchell 1991, 1995). Consequently, measurements of spectral fluorescence on filtered (e.g. Mitchell and Kiefer 1988b), concentrated (e.g. Nelson et al. 1993) or unconcentrated (Sosik and Mitchell 1995) samples have been used very effectively in oceanographic research. Spectral fluorometry is also an important feature of flow cytometry, which is discussed below.

Until recently, the requirement for hands-on manipulation of samples limited the utility of spectral fluorometry for studying the variability of species assemblages or photosynthetic physiology with depth and time. Now, both experimental (Cowles et al. 1993) and commercially available systems can make in situ measurements of spectral excitation or emission of fluorescence. Such systems should be effective in describing the distributions of phytoplankton groups, and might be useful for the detection of HAB species.

**Fluorometry to examine the physiology of phytoplankton** For some time, it has been recognized that the fluorescence yield of phytoplankton, before and during dark adaptation or exposure to bright light, can be an informative indicator of physiological state as influenced by environmental irradiance (Kiefer 1973a,b; Loftus and Seliger 1975; Harris 1980; Vincent 1980; Demers et al. 1985) and nutrition (Kiefer 1973a; Welschmeyer and Lorenzen 1981; Cleveland and Perry 1987). Likewise, oceanographers have known that even more can be learned by making some well-defined measurements (see Falkowski and Kolber 1995): \( F_o \) fluorescence induced by illumination immediately after dark adaptation when the quenching of fluorescence by photosynthetic electron flow is maximal, and \( F_n \) fluorescence when photosynthetic quenching is eliminated by fully reducing photosystem 2 (PS2) reaction centers using exposure to saturating irradiance or by blocking noncyclic photosynthetic electron flow with a photosynthetic inhibitor such as DCMU [3-(3, 4-dichlorophenyl)-1, 1 dimethyl urea]. Variable fluorescence, \( (F_v = F_o - F_n) \) is a measure of maximum photosynthetic quenching, hence the capacity for noncyclic photosynthetic electron flow through PS2; because \( F_v \) is related to the transfer of absorbed quanta to active PS2 reaction centers, the ratio \( F_v : F_o \) is a measure of the quantum efficiency for photochemistry of PS2. The rate at which fluorescence rises to \( F_o \) from \( F_n \) during exposure to bright light (fluorescence induction) can be related to the absorption cross section of PS2. Although valid and useful measurements of variable fluorescence can be made inexpensively with conventional fluorometry employing DCMU (e.g. Vincent 1980, 1981; Cullen et al. 1986; Neale and Richerson 1987; Neale et al. 1989), methods that use active manipulation of irradiance are more versatile, informative, and adaptable for use in situ. These measurements have great potential for noninvasive estimation of photosynthetic rate (Kolber and Falkowski 1993) and for inferring the physiological state (e.g. relative growth rates) of natural phytoplankton (Falkowski et al. 1992). Also, active fluorometry should be very useful in trying to understand the mechanisms that determine the quantum yield of passively measured solar-stimulated fluorescence (Kiefer and Reynolds 1992). The most commonly used active fluorescence techniques have been pump-and-probe fluorometry, fast-repetition-rate (FRR) fluorometry (both reviewed by Falkowski and Kolber 1995), and pulse-amplitude-modulated (PAM) fluorometry (Schreiber et al. 1986; Kroon et al. 1993). Unfortunately, active fluorometers sensitive enough for use on natural samples of phytoplankton have only recently been marketed.

**Flow cytometry**—A flow cytometer can be an extremely powerful tool for characterizing algal assemblages with respect to species composition as well as physiological status. A suspension of particles is directed into a very narrow stream, such that one particle at a time encounters a focused beam of light. Detectors measure properties such as forward light scatter and side-scatter (to characterize cell size and, to some extent, refractive index; Ackleson and Spinrad 1988), as well as the fluorescence of chlorophyll \( a \), phycoerythrin, or perhaps a fluorescent stain (Yentsch and Phinney 1988). Flow cytometry was a powerful tool for identifying taxa and quantifying important cell constituents (see Scholin et al. 1997). The method can also be used to estimate particle dynamics on the basis of diel changes in cell properties as related to growth and cell division (Durand and Olson 1996). Flow cytometers are expensive and not suitable for use on small boats. However, many types of
analyses can be done on preserved samples (Vaulot et al. 1989), so routine monitoring or surveys could be practical.

Practical evaluation of an emerging technology:
Spectral radiometer buoy

We have shown that a wide variety of optical observation technologies may be useful for describing spatial and temporal variability of phytoplankton in surface waters. Optical systems have been used effectively in open-ocean experiments (Dickey et al. 1991; Marra et al. 1992; Abbott et al. 1993) and in a reservoir (White et al. 1991). However, this work is only partially relevant to the detection and assessment of harmful algal blooms. Here we describe a recently developed passive optical detection system that has considerable potential for use in long-term monitoring of coastal waters, early warning of algal blooms, and in oceanographic research on optical variability associated with coastal phytoplankton.

**Description of the instrument**—A Tethered Spectral Radiometer Buoy (TSRB; Satlantic, Inc.) is a calibrated radiometer system deployed from a research vessel to drift at the end of a near-neutrally buoyant conducting cable. It records downwelling solar irradiance at 400 nm \( E_d (400) \) and upwelling radiance in seven wavebands that correspond to six detectors of the SeaWiFS ocean color satellite and a channel for the fluorescence of Chl near 683 nm (Fig. 5). A SeaWiFS waveband, such as 510 nm, can be replaced with one at 700 nm to provide a baseline correction for fluorescence at 683 nm (i.e. a straight line between 670 nm and 700 nm, cf. Neville and Gower 1977). The TSRB is similar to an expendable instrument (McLean and Lewis 1991; Landry et al. 1997) which drifts with ocean currents and transmits through the ARGOS satellite a statistical summary of measurements over the previous 1 h. With a sampling frequency of 1 s\(^{-1}\), the TSRB is suited for examining the influence of short-term variability on those 1 h averages (Cullen et al. 1994).

**Objectives**—A principal use for the TSRB is in the development and validation of algorithms for estimating Chl from remotely sensed ocean color. These efforts require the estimation of near-surface radiance reflectance simultaneous with measurements of near-surface pigments, a task for which the TSRB is ideally suited. Rigorous comparison of observations from the TSRB with measurements from satellite sensors requires the consideration of many factors associated with propagation of light through the atmosphere and sea surface. With respect to the detection of harmful algal blooms or coastal eutrophication, however, a prime objective is to evaluate the TSRB as a system for quantifying biologically induced variability in coastal waters. Simply: can measurements of upwelling radiance in seven wavebands, normalized to above-surface downwelling irradiance in one waveband, be used to describe spatial and temporal variability of phytoplankton in coastal waters? If so, on what temporal and spatial scales would these optical methods be valid?

**Potential problems**—Although the TSRB is a precisely calibrated optical instrument, it is deployed as a tethered buoy that moves with the waves and the tug of the cable. Consequently, optical measurements can be highly variable on the time scale of seconds, as the sensors tilt toward and away from the sun (Cullen et al. 1994) and as waves modify the optical field near the surface (see Stramski et al. 1993 and references therein). We have found that this extraneous variability can be greatly reduced by adjusting the mechanical attachments so the buoy floats vertically when the tether is taugh and by averaging data over one to several minutes. On sunny days in choppy seas, extreme values elevate the means for \( L_s (\lambda) \), relative to the median. Thus, a running median is better than the mean as a measure of central tendency.

Deployment of the TSRB on station during research cruises presents little problem: useful data have been obtained in 3-m seas and when the research vessel was maintaining headway at \(-50\) cm s\(^{-1}\). Long-term measurements from moorings and drifters present challenges, however. In particular, biofouling of the sensors must be considered. Also, drifters with attached drogues may eventually sink under the weight of the macrofaunal fouling community (M. R. Abbott pers. comm.). Further, both moorings and drifters are attractive to the curious, so they are subject to disturbance.

To achieve our objective of relating optical measurements to biological variability, we would like to estimate radiance reflectance \( R(\lambda) = \frac{L_s (\lambda) / E_d (\lambda)}{L_s (\lambda) / E_d (\lambda)} \) in the seven wavebands for which upwelling radiance is measured and to relate variability in \( R(\lambda) \) to the constituents of surface waters. However, \( E_d \) is measured in one waveband only (490 nm), so we measure variability in \( L_s (\lambda) / E_d (490) \), which will be referred to as \( R_\text{TSRB} (\lambda) \). It is thus necessary to consider the degree to which changes in the spectral shape of solar irradiance [i.e. \( E_d (\lambda) / E_d (490) \)] contribute to patterns in the data. The shape of the solar spectrum at sea level changes with elevation of...
Optical detection of blooms

Fig. 6. Detection of a dinoflagellate bloom with passive optical measurements, 18 August 1993 in Bedford Basin. A. Spectra of reflectance \( R_{\text{TSRB}} = L_o(\lambda)/E_d(490) \) during patchy discoloration of surface waters by high concentrations of the nontoxic dinoflagellate *Gonyaulax digitale* (see Amadi et al. 1992). The red-water spectrum is the average for 1345–1350 hours, and the green-water spectrum is for 1354–1359 hours. Error bars are SD for measures recorded 1 s⁻¹. B. Measurements of diffuse attenuation at 490 nm (obtained with a profiler consisting of two sensors for \( E_d(490) \) separated by 1 m), along with ancillary observations (Cullen et al. 1994), show that a subsurface population was being entrained into the mixed layer: the profile at 1320 hours detected the subsurface layer, which was fully mixed by wind into the surface layer by 1710 hours. (From Cullen et al. 1994.)

the sun, surface albedo, and atmospheric conditions; ratios of \( E_d(\lambda) \) to \( E_d(490) \) can be modeled with adequate precision using relatively simple models for clear-sky conditions (e.g. Gregg and Carder 1990). Uniform cloud cover does not change the blue:green ratio of the solar spectrum much (Webb 1991). Partial clouds present a problem, however. When a cloud occludes the sun while much of the sky is clear, direct sunlight is attenuated to a greater extent than diffuse skylight, which is relatively blue (Bird and Riordan 1986). As a result, the longer wavelengths of solar spectral irradiance are differentially attenuated when patchy clouds block the sun, and upwelling radiance ratios shift toward the blue for reasons independent of biological variability. The effect (e.g. a 20% increase of \( L_o(443)/L_o(555) \); Cullen et al. 1994) is not overwhelming, but it is rapid and could conceivably be misinterpreted as biological variability if only \( L_o \) ratios were measured.

**Detection of an algal bloom**—As shown in Fig. 2, the TSRB, like other radiometers (e.g. Carder and Steward 1985; Tyler and Stumpf 1989; Gower and Borstad 1990; Harding et al. 1995), can readily distinguish waters of different colors and brightness. The fluorescence signal near 683 nm should help in distinguishing the contributions of phytoplankton vs. suspended sediment to red or brown water. Detection of discolored waters associated with algal blooms should thus be no problem. Indeed, the TSRB has effectively recorded the entrainment of dinoflagellates into the surface mixed layer of Bedford Basin (Fig. 6A). It can be seen that the red water is distinguished not only by different color, but also by reduced reflectance throughout the visible region and a strong fluorescence signal at 683 nm. A simple array of sensors for downwelling irradiance detected the population of *Gonyaulax digitale* prior to the red-water event, when the dinoflagellates resided in a subsurface layer (Fig. 6B), demonstrating that passive measurements could detect a subsurface bloom. Several days after the red water was observed, anoxia led to a fish kill in Bedford Basin. Optical measurements were helpful in tracing the cause to the decline of the dinoflagellate bloom. Profiles of beam attenuation and fluorescence from earlier in summer suggested that the bloom had persisted for weeks in a subsurface layer.

**Quantitative measurements of short-term variability**—It has been shown that radiometer buoys can detect short-term optical variability in coastal waters associated with patchiness of phytoplankton and changes in ocean color consistent with diurnal accumulation of phytoplankton (Cullen et al. 1994). Coordinated sampling of surface waters is required for quantitative interpretation of such optical variability, however. Recent observations off the coast of Oregon demonstrate that the TSRB can detect biological variability with extremely good precision: both a green:blue reflectance ratio and a measure of solar-induced fluorescence tracked changes in surface Chl almost exactly (Fig. 7). We expect the best possible fit between optical measurements and biological properties because the observations were made at one location where the dominant phytoplankton (a nontoxic *Pseudo-nitzschia* sp.; L. Shapiro pers. comm.) did not change. Also, the scaling of axes in Fig. 7 represent a best-fit for local conditions. Nonetheless, the data indicate that the TSRB can track short-term biological variability in surface waters accurately, given an appropriate calibration.
Regional calibration—It is well recognized that the contribution of phytoplankton to optical variability in coastal waters cannot be described adequately with simple ratios of radiance reflectance in two wavebands (Sathyendranath et al. 1989; Carder et al. 1991; Doerffer and Fischer 1994); the confounding influences of DOM and suspended sediment are too great (but see Stumpf and Tyler 1988; Tyler and Stumpf 1989). Thus it is no surprise that the strong correlation between simple measures of reflectance and Chl for one locality (Fig. 7) fails to hold in a regional comparison that includes environments with vastly different contributions of DOM and suspended sediment (Fig. 8). Work is underway to evaluate more sophisticated algorithms for retrieving Chl from measurements of \( L_\lambda \). The reflectance signal at 683 nm was well related to Chl for data off Oregon and poorly related to Chl in the muddy waters near Prince Edward Island (data not shown); however, a plot of solar-induced fluorescence as related to Chl, similar to Fig. 8, could not be produced because \( L_\lambda \) was not measured in Bedford Basin. There, a crude measure of fluorescence, \( L_\lambda(683) L_\lambda(670) \) was fairly well related to Chl (log : log regression, \( R^2 = 0.81, n = 12; \) Cullen et al. 1994).

Extended deployment of a drifting buoy—Upward- and downward-looking passive optical sensors have been deployed in a drifter several meters below the surface to track biological variability in the California Current (Abbott et al. 1995). Likewise, a downward-looking radiometer deployed as a surface drifter has detected similar variability in a remote location (Fig. 9). Calibration is a problem, and fouling is a concern. Nonetheless, observations from drifting radiometers provide extremely useful information on temporal variability of near-surface phytoplankton, and concurrent measurement of temperature help to describe the physical causes of that variability (e.g. ice-melt and stratification; Fig. 9B). Temporal changes of surface Chl, as estimated with a drifting radiometer, have been used in a calculation of food-web dynamics and advection in the equatorial Pacific Ocean (Landry et al. 1997). Such applications are likely to increase as more information is obtained on variability of the relationships between optical properties and biological processes in the surface layer (Cullen and Lewis 1995).
Optical observation technologies and HABs: status and prospects

This report is being prepared at a particularly interesting time: optical observation technologies are being developed at a rapid pace, and several approaches have been successfully employed in open-ocean applications. New ocean-color sensors are scheduled to be launched on satellites, and research on the optical properties of coastal waters is accelerating. Meantime, concerns about and interest in harmful algal blooms is arguably at an all-time high. It seems appropriate to follow a path that has been established (Carder and Steward 1985; Uno and Yokota 1989; Yentsch 1989; Pettersson et al. 1993) and to pursue a greater integration of optical oceanography with research and monitoring of harmful algal blooms.

We identified three critical applications for HAB research: long-term monitoring of coastal environments, systems for early warning of algal blooms, and optical measurements for oceanographic research. Then we described different approaches: passive observation of radiometric quantities in natural waters; measurement of inherent optical properties using instruments employing artificial light sources; and fluorometric methods having a broad range of capabilities. Which approaches are best suited for each critical application?

**Long-term monitoring**—The quantification of a trend in transparency of Japanese waters (Fig. 1) clearly demonstrates that simple but robust optical measurements can be extremely useful for describing long-term changes in coastal environments. For monitoring long-term changes in trophic status and bloom frequency, it would be desirable to make a comprehensive set of continuous measurements from moorings, complemented with observations from airborne sensors or satellites (c.g. Kahru et al. 1994) as well as essential sampling from ships or boats. Costs for instruments, routine maintenance and calibration must be considered, though. We suggest that a moored radiometer system optically compatible with satellite radiometers would be well suited for long-term monitoring of biological variability. A spectral absorption-attenuation meter would be an extremely useful adjunct, with the added advantage of working at night. A chain of sensors for downwelling irradiance in one waveband could detect subsurface blooms that would be missed by airborne sensors as well as the surface radiometer, and which might not be sampled by active in situ optical instruments deployed at one or more depths. To validate performance, each of these systems would require routine checks, including well-defined radiometric calibrations; in principle, comparison between sites and over long periods of time would not be confounded by differences in methodology that, for example, could influence the determination of Chl as would be required to calibrate a moored fluorometer.

**Early warning systems**—With respect to early warning systems, important criteria include cost, temporal continuity of measurements, and spatial coverage. In situ fluorometers are very useful for making continuous records related to phytoplankton abundance (Falkowski and Kolber 1995); transmissometers (Volent and Johnsen 1993) and spectral absorption meters can serve a similar purpose. Because many algal blooms can be easily detected in spectra of water-leaving radiance (e.g. Figs. 2, 6; see also White et al. 1991), it is clear that spectrometers should also be extremely useful in the detection of HABs. High precision is unnecessary for the detection of water discolored by phytoplankton, so inexpensive versions of currently available oceanographic instruments might be useful for early warning systems. Remote sensing of ocean color provides excellent spatial coverage (Brown et al. 1985), and even a system designed to measure temperature can detect some blooms (AVHRR,
established and it has been shown that fluorescence measures in situ can be related to photosynthesis (Kolber and Falkowski 1993), aquatic primary productivity estimated from active fluorescence has not been compared extensively to direct measures of photosynthesis. Such comparisons are relatively straightforward in stratified waters, but in surface layers that are mixing vertically, static incubations can yield artifically low results (Cullen and Lewis 1995 and references therein). If active fluorescence techniques are to be used to estimate photosynthesis in HAB research, it will be necessary to improve our understanding of the relationship between photosynthesis and fluorescence during vertical mixing (Ibelings et al. 1994). Improved access to active fluorometers capable of in situ measurements is essential. Also, specialized methods for observing phytoplankton during simulated vertical mixing (Kirkpatrick et al. 1990) could be useful.

One of the most promising oceanographic applications of active fluorescence techniques is as a diagnostic of nutrient limitation in natural phytoplankton (Falkowski et al. 1992), and the approach would surely be useful in studying the physiological ecology of HABs. It is crucial, of course, that fluorescence-based diagnostics of nutrient limitation (e.g. dark-adapted $F_o : F_m$, an index of the quantum efficiency of PS2) can be verified for HAB species in the natural environment. For example, a high $F_o : F_m$ (~0.65) must be an unambiguous indication of nutrient-replete growth, and a low $F_o : F_m$ must be observed only when specific growth rate is strongly depressed due to nutrient limitation (Falkowski 1992). Indeed, pump-and-probe and FRR-fluorometry-based observations from the laboratory support these assumptions (e.g. Kolber et al. 1988; Greene et al. 1992), and patterns in the open ocean seem consistent as well (Kolber et al. 1990; Greene et al. 1994; Kolber et al. 1994). On this basis, one might accept that fluorescence measures such as $F_o : F_m$ are robust indicators of nutrient limitation for all oceanographic applications. Caution is warranted, however. Laboratory studies have been conducted on only a few species, and very little is known about fluorescence parameters of phytoplankton grown on light-dark cycles at irradiance levels characteristic of surface layers, when high growth rates could be maintained despite reduced photosynthetic efficiency (and perhaps depressed $F_o : F_m$). Also, although it is well established that fluorescence parameters and several measures of photosynthetic performance decline during nutrient starvation in batch culture, there are stark, unexplained inconsistencies between published studies in the relationships between steady-state nitrogen-limited growth and photosynthetic performance. Reviewing these differences, Cullen et al. (1992) stated (p. 75)...

We feel that no set of results can be considered “right” or “wrong”; rather, they differ for reasons that have not been identified or explicitly studied. It is curious and unsettling to discover that the laboratory experiments central to our understanding of nitrogen limitation and photosynthetic physiology can have such disparate results, for no obvious reason. Resolution of uncertainties will require further experiments with carefully monitored continuous cultures, during which fluorescence parameters and photosynthetic performance are
measured. Fieldwork with active fluorometers should proceed as interpretations of the measurements are being refined.

Conclusions

Optical observation technologies are well suited for research and monitoring of HABs. There is a wide variety of approaches, each of which has particular strengths, and all of which have limitations. So far, only a few optical methods have been applied to HAB research, in part because instrumentation has only recently been developed, and also because many researchers, aquaculturists, and managers concerned with HABs in coastal waters have had little reason to focus on advances in hydrological optics or fluorescence techniques. There are now good reasons to apply optical oceanography to problems associated with HABs: needs for monitoring and characterization of HABs are greater than ever, optical measurements and their interpretations are sufficiently well developed to be useful, and satellites sensors will soon provide unprecedented views of optical variability in coastal waters. For progress to be most effective, we should understand the principles of optical methods, appreciate cost-benefit for each approach, and develop robust techniques for turning raw data from sensors into information that is useful to all who are concerned with coastal ecosystems and resources.

References


DENMAN, DAVIS, - , D'SA, E. J., FIGUEIRAS, E
EI'PLL:Y, DOERFFER, DICKBY, DIJRENBACI-I,
1236 Cullen et al.


---, E. C. ITSWEIRE, AND W. E. ESAIAS. 1995. Algorithm development for recovering chlorophyll concentrations in the


