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Journal of Marine Systems 43 (2003) 51–59

JOURNAL OF MARINE SYSTEMS
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Fine-scale variability in phytoplankton community structure and inherent optical properties measured from an autonomous underwater vehicle

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Received 6 December 2002; accepted 26 May 2003

Abstract

The relationship between inherent optical properties (IOPs), phytoplankton community structure and the abundance of suspended particles in the size range 3–500 μm was studied near the Isles of Scilly (UK) in May 2000. Autosub, an autonomous submersible vehicle specifically designed for science missions, was used as an instrument-positioning platform. It carried a CTD system, an ac-9+ dual tube spectrophotometer, a prototype submersible flow cytometer and an Aqua-monitor water sampler. The vehicle made a 10-km transect at constant depth across a boundary between water masses with contrasting remote sensing reflectance, which was located using SeaWiFs ocean colour imagery. This boundary corresponded to a sharp (1 km) transition between one phytoplankton community consisting of coccolithophores, flagellates and dinoflagellates, and a second community dominated by diatoms and flagellates. Inherent optical properties measured along the autonomous underwater vehicle (AUV) track showed marked changes in magnitudes, ratios, spectral shapes and fine-scale spatial variability. These changes were well correlated with variations in the composition of the suspended particle assemblage measured by microscopy and in situ flow cytometry.

Keywords: AUV; Flow cytometry; Optical water types; Optical classification; Scattering coefficient; Phytoplankton optics; UK; English channel; Isles of Scilly

1. Introduction

The inherent optical properties (IOPs) of seawater are influenced by the size and shape of phytoplankton cells as well as by their numerical abundance (Morel, 1991, 1994), and it is possible to calculate the optical properties of model phytoplankton assemblages with reasonable accuracy (Ciotti et al., 1999; Stramski et al., 2001). On the other hand, inverse methods for deriving information on phytoplankton communities from optical measurements are still at an early stage of development (Kirkpatrick et al., 2000). One practical problem hindering the validation of these methods is

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doi:10.1016/S0924-7963(03)00088-5

the difficulty of achieving spatial coincidence between IOP measurements (which are best made in situ) and particle analyses (which are usually carried out in the laboratory). The usual technique is to attach optical instruments to a rosette sampler/CTD system, but the flow pattern through the instrument cage may disturb the sampling regime and the time taken to perform instrument casts limits the spatial coverage that can be achieved. For near-surface waters, spatial surveys can be carried out by pumping a sample stream through on-board instruments (Claustre et al., 2000). Surveys deeper in the water column usually involve mounting instrument packages on towed bodies (Owens et al., 1993; Barth and Bogucki, 2000). An alternative platform, which has recently become available for scientific use is the autonomous underwater vehicle (AUV). These vehicles can operate far from the disturbing influence of their support ship, are manoeuverable in three dimensions, carry relatively large payloads and are able to position instruments at precise depths and locations. They can be programmed to survey features identified from satellite or airborne images, or to alter course and sampling strategy in response to the conditions which are encountered. The successful use of an AUV as a platform for optical measurements has recently been reported by Yu et al. (in press). This paper explores the feasibility of using a specific AUV (Autosub) to combine optical measurements with in situ particle analyses and water sampling. Trials were carried out in the English Channel south of the Isles of Scilly, in an area where tidal mixing promotes phytoplankton growth (Simpson et al., 1982) and SeaWiFS ocean colour images often indicate strong spatial gradients in remote sensing reflectance.

2. Methods

2.1. Autosub

Autosub is an AUV developed by Southampton Oceanography Centre (UK Natural Environment Research Council) specifically for scientific missions and survey work (Griffiths et al., 2000). It has a length of 7 m, diameter of 0.9 m and a mission range of approximately 500 km. Autosub is pressure-rated to 1600 m and can carry a scientific payload of 1 m^3 volume weighing up to 100 kg in air. Navigation information is acquired by bottom tracking using a Doppler log from a 150-kHz ADCP with a range of 500 m, and an inertial navigation system provides an accuracy of 0.2% of the distance travelled (McPhail and Pebody, 1998).

2.2. Cruise details

Work was carried out from the *MV Terschelling*, an engineering support vessel fitted for this cruise with the Autosub launch and recovery system and additional laboratory facilities. Information derived from SeaWiFS images by the NERC Remote Sensing Data Analysis Service at Plymouth Marine Laboratory was relayed to the ship and used to guide the programming of Autosub missions. The images showed an extensive coccolithophore bloom in the Western Approaches and a smaller patch of high reflectance south of the Isles of Scilly, which appeared to be actively increasing in area. The data presented here were obtained on the morning of May 25th, 2001, when Autosub made a 10-km transect at a depth of 9 m across the northern boundary of this smaller patch (Fig. 1). Vertical profiles of optical properties were measured from the ship at two stations (15 and 16) located at the beginning and end of the transect.

2.3. Autosub instrumentation

In addition to navigation and communication equipment, Autosub carried a CTD system (Seabird SBE 19), an ac-9+ recording dual-tube spectrophotometer (WetLabs), an Aqua-monitor triggerable water sampler (WS Ocean Systems) and a prototype submersible flow cytometer on its first sea trial. Measurements of single-particle and bulk optical properties were made at closely spaced positions by sampling a water stream pumped from an inlet which pierced the Autosub hull. The ac-9+ was run continuously throughout the mission with data subsequently bin averaged in 1-s increments. Calibration of the two optical channels was regularly checked using ultrapure Millipore water and remained within the manufacturer's specifications of ± 0.005 m^{-1} throughout the cruise. Absorption and attenuation signals at 715 nm were corrected for temperature dependent water

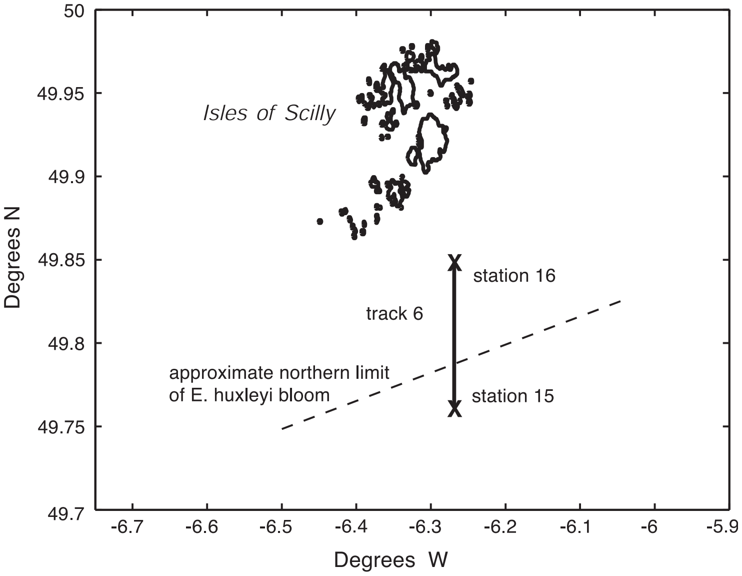

Fig. 1. Map of the Isles of Scilly (English Channel) showing the Autosub track and stations referred to in this paper, and the approximate boundary between distinct phytoplankton communities.

absorption (Pegau and Zaneveld, 1993) and a scattering correction algorithm was applied to the absorption data (Zaneveld et al., 1994). The values of absorption and beam attenuation quoted in this paper were measured relative to a pure water blank. The water sampler was fitted with plastic transfusion bags arranged in pairs containing either neutralised formaldehyde or Lugol's iodine preservative and collected approximately 450 ml of sea water per bag. Preserved samples were transferred to glass bottles and transported to Plymouth Marine Laboratory for further analysis.

2.4. Supporting measurements

Niskin bottle samples for pigment measurements were collected at three depths from each station. One liter of water was filtered through Whatman GF/F filters and these were stored immediately in liquid nitrogen. HPLC analysis was performed within 1 month of collection following the method of Barlow et al. (1993). Suspended particulate material (SPM) and coloured dissolved organic matter (CDOM) were measured following the procedures described in McKee et al. (2002). Depth profiles of inherent optical properties were obtained using a frame lowered from a hydrographic winch carrying a second ac-9 and a Hydroscat 2 backscattering meter (HobiLabs). The manufacturer's calibration was employed for the Hydroscat 2.

3. Results

3.1. Station characteristics

Station 15 exhibited a thermocline at 15 m with a density anomaly (σ_t) difference of 0.1 kg m^{-3}, while station 16 was rather well mixed. The stratification at station 15 was associated with a layer of elevated attenuation and backscattering extending from the surface to around 15 m, while the optical properties of station 16 were more uniform with depth. These characteristics are illustrated by profiles of the backscattering coefficient at 470 nm and the beam attenuation coefficient at 555 nm for both stations in Fig. 2. The Autosub track depth was set at 9 m to allow the surface layer to be sampled while minimising the possibility of collision with shipping. Pigment, CDOM and SPM concentrations for samples taken from a nominal depth of 10 m at both stations are shown in Table 1.

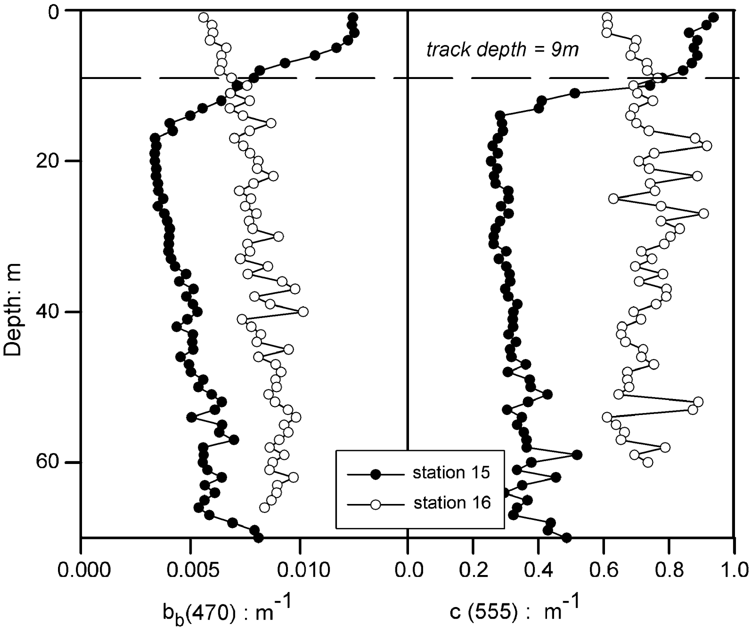

Fig. 2. Profiles of the coefficients of backscattering (b_b) at 470 nm and beam attenuation (c) at 555 nm for stations at the Autosub launch and recovery positions. The dashed line indicates the depth of the AUV transect.

3.2. Pigments

Pigment concentrations determined by HPLC were expressed in terms of their ratio to chlorophyll *a*. The resulting histograms for stations 15 and 16 show marked differences at the two ends of the transect (Fig. 3). The pigment of greatest relative abundance changed from 19′-hexanoyloxyfucoxanthin (occurring mainly in prymnesiophytes, including coccolithophores) at the southern end to fucoxanthin (occurring mainly in diatoms) in the north. Peridinin, a marker for dinoflagellates, was found only in the southern station. The pattern of occurrence of chlorophyll C3 and 19′-butanoyloxyfucoxanthin is consistent with a decrease in the abundance of prymnesiophytes and dinoflagellates from south to north. The photoprotective carotenoid diadinoxanthin occurred mainly in the southern population, which was confined by stratification to a well-illuminated surface layer.

Table 1
Concentrations of total chlorophylls, total carotenoids, SPM and CDOM (as absorption at 440 nm) at 10 m depth for the stations indicated in Fig. 1

Station	Chlorophylls (mg m^{-3})	Carotenoids (mg m^{-3})	SPM (mg l^{-1})	*a*(440) (m^{-1})
15	0.67	0.6	1.47	0.05
16	1.82	0.8	3.41	0.06

3.3. Particle analysis: microscopy and flow cytometry

Species identification by optical microscopy was carried out on samples preserved with Lugol's iodine, and numbers reported as single cells for flagellates and dinoflagellates and colonies for diatoms. The 'flagellate' counts included coccolithophores, which were difficult to distinguish due to the loss of calcite in this preservative. Separate counts of coccolithophores in neutral formaldehyde indicated that they contributed less than 3% to the 'flagellate' totals. The coccolithophores were identified as *Emiliania huxleyi* by electron microscopy. *Phaeocystis* colonies were also observed in the northern community, but their numerical density that was too low to be estimated by microscopy. Fig. 4 shows the abundance of the main phytoplankton groups along the transect. There was a clear change in taxonomic composition at approximately 3 km, where the southern community consist-

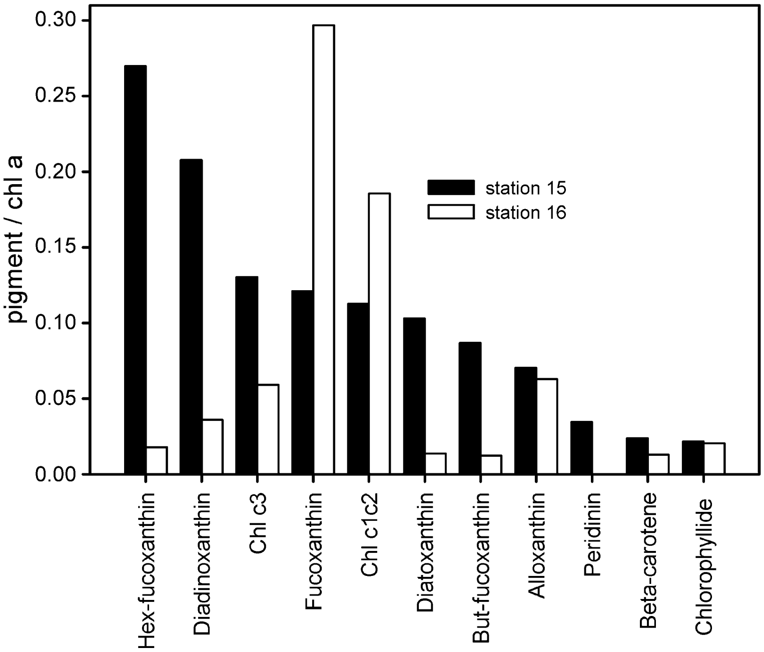

Fig. 3. Abundance (normalised relative to chlorophyll *a*) of phytopigments in samples taken from a depth of 10 m from stations 15 and 16.

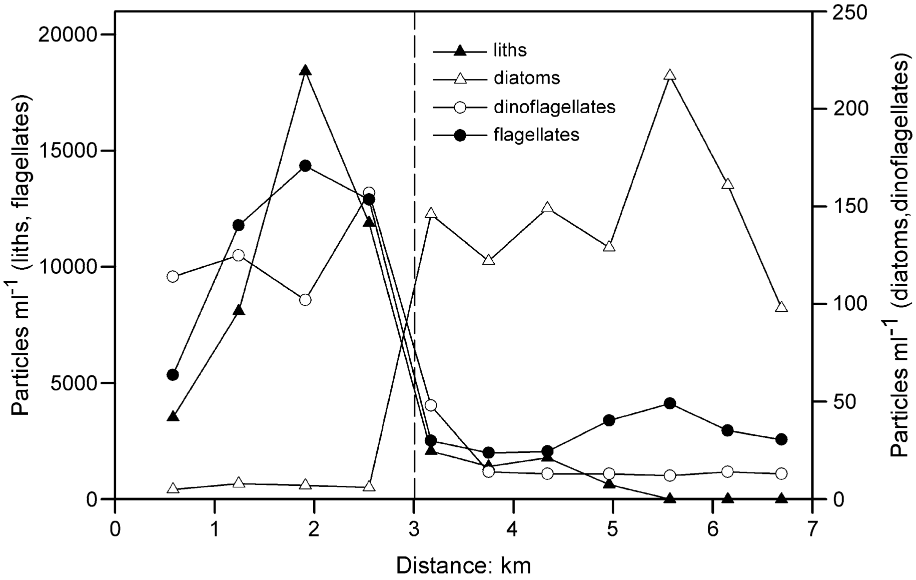

Fig. 4. Variation in the numerical density of the most common phytoplankton taxa, determined by optical microscopy of preserved samples acquired in situ. The dotted line indicates the approximate boundary between communities and is repeated in subsequent figures. Distance is measured in a northerly direction from station 15.

ing of small flagellates, dinoflagellates and coccolithophores was replaced by the northern one consisting of diatoms with lower flagellate numbers. These microscope observations are in very satisfactory agreement with the taxonomic information provided by the pigment data. Single particle counts from the in situ flow cytometer (Fig. 5) also showed spatial patterns consistent with those derived from microscopy. Coccolithophores were identifiable in the cytometric data as a distinct cluster in a scatter plot of particle length against side scattering amplitude, and the numbers measured correlated fairly well with those obtained by microscopy ($r^2 = 0.73$). The proportion of particles detected in the size range 3–500 μm which exhibited red fluorescence (indicating the presence of chlorophyll) ranged from around 2% in the southern community to around 12% in the north. Such low percentages need further investigation, but they suggest that a large number of non-phytoplankton particles contribute to optical properties in these coastal waters.

The changes in the suspended particle assemblage at the 3-km boundary were more subtle than a simple transition between coccolith-dominated and coccolith-free water. The maximum concentration of detached liths south of the 3-km boundary was comparable with the concentration of nanoflagellates, and intact *E. huxleyi* cells never accounted for more than 3% of the total cell counts.

3.4. Horizontal variations in IOPs

Fig. 6 shows the coefficients of absorption and scattering at 676 nm measured by the ac-9+ along the Autosub track. The step-change in signal characteristics at 3 km, which was evident in all instrument wavebands, consisted not only of a shift in the mean level of the signal but also a significant increase in the magnitude of the fluctuations about the mean. Examination of raw ac-9+ data (before bin averaging) indicated that the absorption spectrum remained characteristic of phytoplankton pigments throughout these fluctuations. A similar increase in signal variability occurred in other transects between the two phytoplankton communities, when it was also observed in beam transmission and backscattering measurements. It was probably due to the presence of aggregated phytoplankton material.

3.5. Optical indicators of community change

One unexpected feature of the data set was the degree to which relationships between optical properties changed along the AUV transect. Since the magnitudes of the inherent optical properties are generally taken to be linearly related to the concentrations of seawater constituents (Mobley, 1994), variations in their ratios indicate changes in the quality of the constituents rather than their abundance.

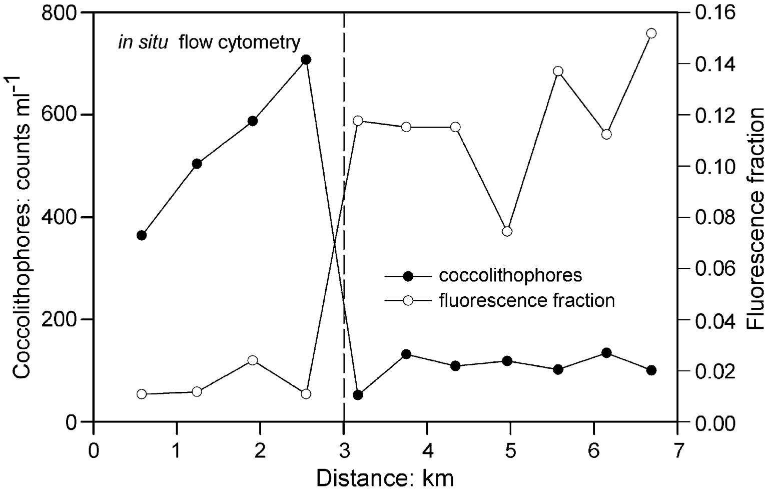

Fig. 5. Data from the submersible flow cytometer showing changes in the numerical density of intact coccolithophore cells and in the fraction of detected particles (above the sensitivity threshold of 3 μm), which exhibited chlorophyll fluorescence.

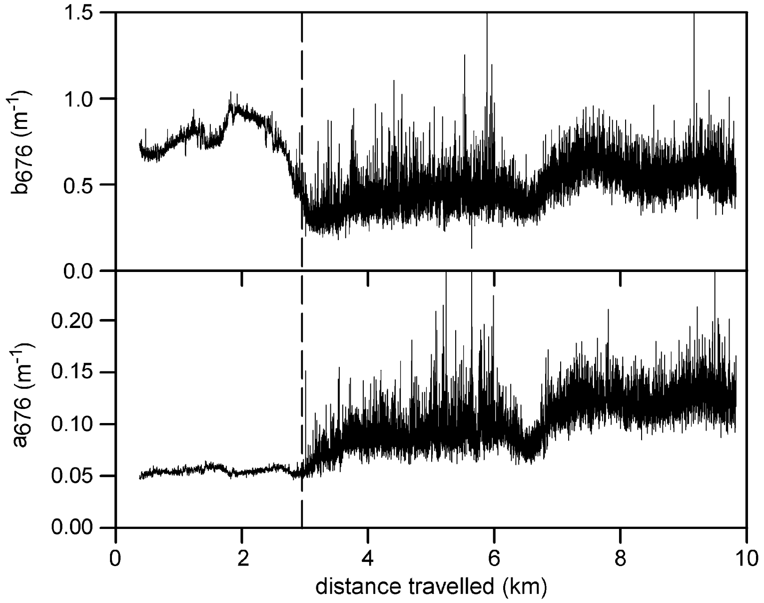

Fig. 6. Variations in the coefficients of scattering and absorption at 676 nm along the AUV transect.

In this transect, the ratio of scattering at the shortest (412 nm) and longest (715 nm) wavebands, the ratio of green (555 nm) to red (676 nm) absorption and the ratio of scattering to absorption at chlorophyll absorption peak (676 nm) all showed step changes at the 3-km boundary between the two phytoplankton communities (Fig. 7). The change in the ratio of scattering at two wavelengths may have been a response to

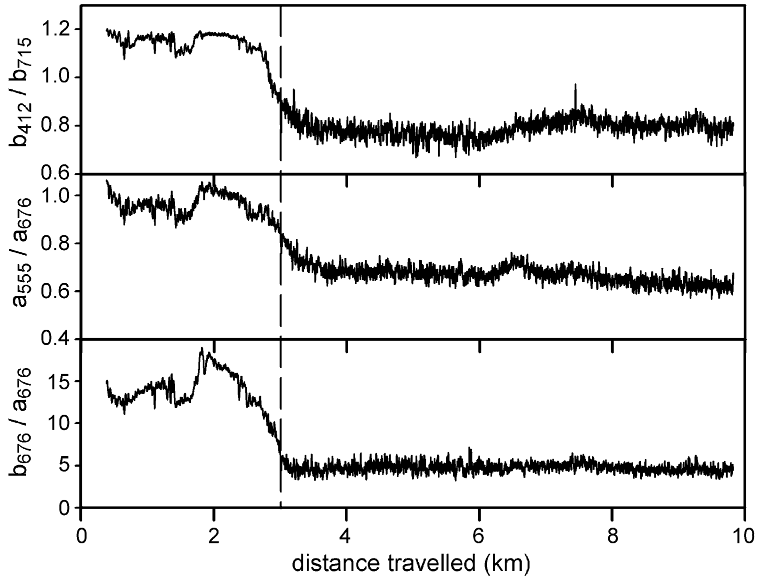

Fig. 7. Ratios of inherent optical properties: scattering coefficients at 412 and 715 nm (top.), absorption coefficients at 555 and 676 nm (middle) and scattering to absorption at 676 nm (bottom).

variations in particle size distribution (Stramski et al., 2001). The 30% change in the green/red absorption ratio along the transect was probably related to a reduction in the carotenoid/chlorophyll ratio from 0.89 in the southern community to 0.44 in the northern one. The ratio of scattering to absorption at 676 nm, which is often used as a discriminator between phytoplankton and detritus, proved in this case to be a useful indicator of the transition between different phytoplankton communities.

The mass-specific cross section for total scattering, calculated by dividing the scattering coefficient at 676 nm by the dry weight of suspended particles, fell from 4.8×10^{-4} m^2 mg^{-1} at station 15 to 1.8×10^{-4} m^2 mg^{-1} at station 16. This was probably due to the absence of detached liths, which have a high scattering efficiency (Balch et al., 1999) north of the boundary. Estimates of in vivo chlorophyll-specific absorption cross sections at 676 nm (a_{676}*) were obtained by dividing ac-9 absorption measurements by chlorophyll concentrations. This procedure assumes that the absorption coefficient measured at 676 nm could be attributed almost entirely to phytoplankton cells because CDOM levels were very low and inorganic particles absorb weakly at red wavelengths. The a_{676}* values fell from 0.15 m^2 mg^{-1} at station 15 to 0.06 m^2 mg^{-1} at station 16. In contrast, the mean absorption cross section per algal particle (estimated using microscope counts) increased from 1×10^{-11} m^2 at station 15 to 3×10^{-11} m^2 at station 16. The figures therefore indicate the occurrence of larger cells with lower chlorophyll-specific absorption efficiencies in the northern section of the transect. This result is consistent with pigment packaging theory, with the diatom-dominated community structure revealed by the microscope and with the analysis of Ciotti et al. (2002).

4. Discussion

Several conclusions can be drawn from these trials. First, the large payload capacity of an AUV such as Autosub makes it possible to deploy instrument combinations which would be difficult to fit on a towed body. AUVs have been used for surveys of zooplankton (Brierley et al., 2002) and turbulence (Levine and Luck, 1999), and it is apparent that they have much to contribute to the study of frontal structures, thin layers and other fine-scale features in shelf seas (Dekshenieks et al., 2001). Second, it is likely that the labour-intensive microscopic analysis of water samples will be replaceable by in situ flow cytometry as the technology matures. Current development work in this area is being carried out by groups in The Netherlands (Dubelaar et al., 1999; Dubelaar and Gerritzen, 2000) and Woods Hole Oceanographic Institution (Olson and Sosik, 2001). Third, the use of satellite images to guide AUV missions is a powerful tool for maximising sampling efficiency in spatially heterogeneous waters. A very time-consuming survey would have been required to locate the interface between phytoplankton communities studied in this paper by ship observations alone.

Acknowledgements

We thank the Autosub engineering team from Southampton Oceanography Centre for making the cruise possible. The submersible flow cytometer was constructed by George Dubelaar and Peter Gerritzen, and Peter Burkill gave advice and encouragement throughout the project. Funding was provided by the UK Natural Environment Research Council.

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