Available online at www.sciencedirect.com
SCIENCE DIRECT®
Estuarine, Coastal and Shelf Science 69 (2006) 189–195
ESTUARINE COASTAL AND SHELF SCIENCE
www.elsevier.com/locate/ecss

Spatial variation of phytoplankton pigments along the southwest coast of India

Rajdeep Roy*, Anil Pratihary, Gauns Mangesh, S.W.A. Naqvi

National Institute of Oceanography, Dona Paula, Goa 403004, India

Received 23 December 2005; accepted 10 April 2006
Available online 15 June 2006

Abstract

Phytoplankton composition and abundance were studied along the southwestern Indian coast toward the end of the upwelling season in October 2004. Phytoplankton pigment analyses, complemented by limited microscopic counts, were carried out to determine the community structure. Chlorophyll *a* was the most abundant of all pigments, followed by fucoxanthin. Zeaxanthin was abundantly found in the southern part of the study region (off Trivandrum), whereas fucoxanthin was the dominant marker pigment in the north (off Goa). The inferred shift in the community structure from a dominant picoplankton fraction and Prymnesiophytes to diatom-dominated microplankton toward the north is ascribed to differences in the physico-chemical environment.

Keywords: phytoplankton; prymnesiophytes; pigment; chlorophyll; zeaxanthin; picoplankton; west coast of India; Arabian Sea

1. Introduction

Accounting for approximately one-fourth of all plants in the world (Hallegraeaff and Jeffrey, 1985), marine phytoplankton are important contributors to global carbon fluxes (Falkowski et al., 1998). Phytoplankton communities in the ocean comprise many different taxonomic groups, which together determine primary production and various trophic level interactions. Quantification of phytoplankton biomass and community composition is important for understanding the structure and dynamics of marine ecosystems.

Phytoplankton biomass is often quantified through chlorophyll *a* (Chl *a*), which has long been measured following spectrophotometric (Lorenzen, 1967) or fluorometric (Holm-Hansen et al., 1965) methods. These methods suffer from spectral interference from other degradation products of chlorophyll (chlorophyllides, phaeophoribides and paheophytins). The application of high-performance liquid chromatography (HPLC) has been found to be more accurate and reliable for estimating not only Chl *a* but other pigments as well. This technique allows quantification of additional 50 phytoplankton pigments and carotenoids in marine phytoplankton (Wright et al., 1991; Jeffrey et al., 1997).

Phytoplankton identification and enumeration is usually done through microscopic examination. This procedure is time-consuming and also requires a high level of taxonomic skill. Moreover, smaller organisms such as picoplankton cannot be identified or counted with this approach. Alternatively, photosynthetic pigments can easily be studied to know the phytoplankton composition and their physiological status. Most of these pigments (Table 1) have chemotaxonomic association. For example, fucoxanthin is considered to be a marker of diatoms; zeaxanthin of cyanobacteria; 19′-hexanoyloxyfucoxanthin of Prymnesiophyceae; alloxanthin and crocoxanthin of Cryptomonads; prasinoxanthin of prasinophytes; peridinin and chlorophyll c_2 of dinoflagellates (see Jeffrey et al., 1997, for details of signature pigments of various phytoplankton). However, it should be noted that marker pigments are not

* Corresponding author.
E-mail address: rrajdeep@nio.org (R. Roy).

0272-7714/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.ecss.2006.04.006

Table 1
Chemotaxonomic relation used in the study of phytoplankton taxonomy (Gibb et al., 2001; Jeffrey et al., 1997)

Pigment	Abbreviation	Occurrence
Chlorophyll *a*	Chl *a*	Total algal biomass (including cyanobacteria)
Chlorophyll *b*	Chl *b*	Chlorophytes, prasinophytes
Chlorophyll c_1c_2	Chl c_2	Diatoms, prymnesiophytes, cryophytes, dinoflagellates
Chlorophyll c_3	Chl c_3	Prymnesiophytes, cryophytes
Alloxanthin	ALLO	Cryptophytes
19′-Butanoyloxyfucoxanthin	BUT	Chrysophytes, prymnesiophytes
Diadinoxanthin	DIADINO	Diatoms, dinoflagellates, prymnesiophytes, chrysophytes
Diatoxanthin	DIATO	Diatoms, dinoflagellates, prymnesiophytes, chrysophytes
Divinyl chlorophyll *a*	DvChl *a*	Prochlorococcus sp.
Fucoxanthin	FUC	Diatoms, prymnesiophytes, chrysophytes
19′-Hexanoyloxyfucoxanthin	HEX	Prymnesiophytes
Lutein	LUT	Chlorophytes, prasinophytes
Peridinin	PER	Autotrophic dinoflagellates
Violaxanthin	VIO	Chlorophytes, prasinophytes
Prasinoxanthin	PRAS	Prasinophytes
Zeaxanthin	ZEA	Cyanobacteria, *Prochlorococcus* sp.

exclusive of any one group of algae. In natural environment pigment composition may well vary with prevailing light condition and photoadaptive state (Falkowski and LaRoche, 1991).

The semi-annual reversal of coastal currents in the Arabian Sea introduces a high degree of seasonality in the physico-chemical environment (Banse, 1959, 1968; Naqvi et al., 2000, 2006). The consequent strong biological response includes marked changes in the composition of phytoplankton, a subject of several previous studies (e.g., Subrahmanyan, 1959; Dehadrai and Bhargava, 1972; Devassy and Goes, 1988), all of which, however, have been confined to microplankton. In general, phytoplankton have been found to be most abundant during the upwelling period that lasts from May–June to October–November. Diatoms constitute the bulk of microplankton exhibiting rich diversity. Dinoflagellates are the next abundant group, occasionally forming blooms sometimes associated with fish kills (Naqvi et al., 1998 and references therein), and in rare cases resulting in paralytic shellfish poisoning (Karunasagar et al., 1984). In September 2004 an incident of fish mortality also accompanied by the release of an obnoxious gas (presumably H_2S) from the sea that caused sickness to children led to a public health alarm around Trivandum (Lat. 8.5° N). Investigations carried out just after this incident suggested a bloom of holococolithophore and oxygen (O_2) depletion in the upwelled water to be the cause of fish mortality (Ramaiah et al., 2005). Such blooms, not recorded previously, are quite significant in the context of coastal ecosystem dynamics and biogeochemical processes. In this study we provide additional data on the plankton composition offshore of the sampling sites of Ramaiah et al., based on observations made approximately 3 weeks after the bloom. To our knowledge this is the first report on the spatial distribution of phytoplankton marker pigments along the Indian coast.

2. Material and methods

Water samples were collected at a number of stations off the coast of Kerala (Fig. 1) that was affected by the above-mentioned obnoxious bloom and at few other stations along the southwest coast of India during a cruise of the coastal research vessel (CRV) *Sagar Sukti* during the period 2–10 October, 2004. Niskin bottles (5 L) fixed on polyvinyl chloride (PVC)-coated hydrowire were used for sampling. Continuous profiles of salinity and temperature were also obtained at all stations except Sta. 13 using a Sea-Bird Conductivity-Temperature-Depth (CTD) profiler (Sea-Bird Electronics 25). Dissolved O_2 was measured immediately after collection following the Winkler procedure. Sub-samples for nutrients were deep-frozen for analysis in the shore laboratory performed soon after the cruise with a Skalar analyser following standard methods (Grasshoff et al., 1983).

For the pigment analysis, samples were immediately filtered on a GF/F filter (pore size 0.7 μm) avoiding exposure of the filter paper to direct light and high temperature. The filter paper was stored in liquid nitrogen until analysed in the shore laboratory as follows. The frozen filters were immersed in 3 ml of 95% acetone (v/v in deionized water) for extraction using a sonicator probe (5 s, 25 kHz) under low light and temperature (4 °C) followed by storage at −20 °C for 4 h. The extract was passed through a Teflon syringe cartridge (Millipore) having a glass fibre pre acrodisc filter (pore size 0.45 μm, diameter 25 mm) to remove the cellular debris. The clarified extract was collected in a 5 ml amber colour glass vial and placed directly into the temperature controlled (5 °C) autosampler tray for the (HPLC) analysis.

Pigments were separated following a slight modification of the procedure of Van Heukelem (2002), which provides quantitative analysis of 20 pigments and qualitative analysis of several others. The HPLC system was equipped with an Agilent 1100 pump together with online degasser, an Agilent diode array detector connected via guard column to an Eclipse XDB C8 HPLC column (4.6 × 150 mm) manufactured by Agilent Technologies. The column was maintained at 60 °C. Elution at a rate of 1.1 ml/minute was performed using a linear gradient program over 22 min with 5/95% and 95/5% of solvents B/A being the initial and final compositions of the eluant, where solvent B was methanol and solvent A was (70:30) methanol and 1 M ammonium acetate (pH 7.2) instead of 28 mM solution as recommended in the protocol. An isocratic hold on 95% B was necessary from 22 to 27 min for the elution of the last pigment (α- or β-carotene) at approximately 27 min. After returning to the initial condition (5% solvent B) by 31 min, the column was equilibrated for 5 min prior to next analysis. The eluting pigments were detected at 450 and 665 nm (excitation and emission) by the diode array detector. All chemicals used were of HPLC-grade, procured from E. Merck (Germany).