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Influence of short-term hydrographic variations during the north-east monsoon on picophytoplankton community structure in the eastern Arabian Sea



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ABSTRACT

The eastern Arabian Sea over the continental margin is a dynamic region subjected to short-term variability in hydrography as a result of various physical forcing such as coastal advection and vertical mixing. In order to assess the influence of hydrography on the picophytoplankton community, a temporal high resolution (every 3 h for nine days) study was carried out at a fixed location (15° 18' 46"N, 72° 41' 53"E) in the eastern Arabian Sea during the early north-east monsoon (November 2011). The picophytoplankton community comprised of Synechococcus, Prochlorococcus, and picoeukaryotes. Based on the temperature and salinity distribution, the study period was divided into phase I representing a stratified water column and phase II representing a vertically mixed water column. Phase I had higher picophytoplankton abundance with the initial dominance of Prochlorococcus which was later taken over by picoeukaryotes. Towards the end of phase I, with the initiation of vertical mixing, picoeukaryotes were the first to respond to the nutrient influx. As the vertical mixing intensified during phase II, the picophytoplankton abundance declined. Picophytoplankton carbon biomass and their contribution to total phytoplankton biomass was relatively higher during phase I with picoeukaryotes as the major contributors. These transient variations in picophytoplankton abundance highlights the importance of high frequency observations at the single cell level for better understanding the population dynamics in such environments.

1. Introduction

The Arabian Sea (AS) constitutes the north-western part of the Indian Ocean and its semi-enclosed feature leads to an unusual climate, hydrography, and biogeochemical processes (Naqvi et al., 2003). As a result of the semiannual reversal of monsoon winds, seasonal variation of water column characteristics is also observed (Oasim, 1982; Banse, 1987). During the south west monsoon (SWM), coastal upwelling is a common phenomenon (Banse, 1968; Banse and McClain, 1986; Shetye et al., 1994) whereas during the north-east monsoon (NEM) convective mixing is prominent in the northern AS (Madhupratap et al., 1996) with decreasing intensity of mixing towards the southern AS (Prasanna Kumar et al., 2000). Additionally, during this period downwelling is also observed (Rao et al., 2008). Monsoonal forcing results in seasonal variations in the mixed layer depth, flux of nutrients to the upper mixed layer and thereby on pelagic food web structure and production (Madhupratap et al., 1996; Morrison et al., 1998; Prasanna Kumar et al., 2000; Wiggert et al., 2000; Shankar et al., 2005).

In the AS, phytoplankton biomass and primary productivity are high

during the SWM and the NEM (Marra et al., 1998; Prasanna Kumar et al., 2000). During the NEM, interannual variations in the phytoplankton biomass and primary production are significant along the eastern (Bhattathiri et al., 1996; Sawant and Madhupratap, 1996; Parab et al., 2006; Ahmed et al., 2016) and the western AS (Campbell et al., 1998; Brown et al., 1999; Garrison et al., 2000). Only a few of the above studies have dealt with the smaller sized phytoplankton groups in the eastern AS (Roy et al., 2015; Ahmed et al., 2016).

During the last decades, the importance of picophytoplankton (PP; $< 3 \mu m$) has been demonstrated in the marine environment as major contributors to the phytoplankton biomass (Worden et al., 2004; Richardson and Jackson, 2007). PP comprises of three major groups, Prochlorococcus (PRO), Synechococcus (SYN) and picoeukaryotes (PEUK). PRO dominates the oligotrophic waters and is encountered down to 150 m depth (Chisholm et al., 1988; Partensky et al., 1999; Johnson et al., 2006; Fuller et al., 2006). SYN is abundant in mesotrophic waters with a shallower vertical distribution than that of PRO (Bouman et al., 2006; Zwirglmaier et al., 2007; Flombaum et al., 2013). PEUK, along with SYN dominate the nutrient-rich coastal ecosystems

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(Blanchot et al., 2001; Jiao et al., 2005; Sharples et al., 2009). Although limited information is available on the PP community from the eastern AS, short-term variability which can impart an in-depth understanding of the PP dynamics still remains unknown.

In this study, we focused on the short-term vertical variations (every 3 h for 9 days) in the PP community structure at a fixed location over the continental slope in the eastern AS, off Goa, India. The objective of this study was to assess the influence of different hydrographic conditions such as stratification and mixing on the PP distribution pattern. We hypothesize that in the eastern AS, the PP community structure and carbon biomass varies in response to the short-term hydrographic variability resulting from coastal advection and vertical water column mixing. This study will also serve as a basis for understanding the response of phytoplankton community as a whole with chlorophyll biomass as the proxy.

2. Materials and methods

2.1. Sampling

Sampling was conducted for 9 days (D) at an interval of 3 h at one fixed location over the continental slope of the eastern AS (15° 18' 46"N, 72° 41' 53"E) (Fig. 1), onboard the ocean research vessel ORV Sindhu Sankalp (SSK-27) during the early NEM season (18-26 November 2011). Temperature, salinity and dissolved oxygen (DO) data profiles were taken from precalibrated CTD. Mixed layer (ML) depth was derived from the sigma-t criteria, as the depth at which a change from the surface sigma-t of 0.125 has occurred. Water samples for nutrients and PP analyses were collected from 7 to 10 depths in the upper 0-200 m water column with 12 dm³ Niskin bottles (General Oceanics, Miami, FL, USA) mounted on a CTD (Sea- Bird electronics) rosette sampler. Nutrient samples were analyzed [nitrite (NO₂⁻) nitrate (NO₃⁻), orthophosphate (PO_4^{3-}), ammonium (NH_4^+), and silicate (SiO_4^{4-})] with a SKALAR SAN^{plus} auto-analyzer. For analysis of PP, water samples were preserved in paraformaldehyde (final concentration 0.2%; Campbell, 2001). After 15 min dark incubation, the samples were quick-frozen in liquid nitrogen and stored at -80 °C until analysis. For chlorophyll a analysis, water samples ($\sim 3-4$ L) were filtered through GF/F filter followed by extraction of pigments in 90% acetone which were measured using High Performance Liquid Chromatography (LC 1200 series, Agilent Technologies, USA; Acharyya et al., 2012).

2.2. Flow cytometric analyses of picophytoplankton

In the laboratory, samples were thawed and analyzed through FacsVerse flow cytometer configured with 20 mW and 40 mW air-

cooled lasers exciting at 488 nm (blue) and 664 nm (red), respectively. Sample acquisition was set for 10,000 events. Flow rates were calibrated before analysis using the equation, $R = (W_i - W_f)/(T \times d)$ where W_i = initial weight of the sample (mg), W_f = final weight of the sample (mg), T = time (minutes), and d = density of the sample (seawater = 1.03) (Marie et al., 2005). Forward angle light scatter (FALS; proxy for cell size) and right angle light scatter (RALS), orange (564-606 nm) and red (> 650 nm) fluorescence intensities were collected from each particle and analyzed with Facs Suite[™] software. Yellow-green fluoresbrite fluorescent beads (Polysciences co., USA) of 2 um diameter were added to the samples as an internal standard. Cell fluorescence emission and light scatter signals of each PP group were normalized to that of the beads (mean cell values/mean bead values) to distinguish the different PP groups based on their autofluorescence properties and size. Three major groups of PP were determined (Fig. 2a-c). SYN was identified based on the orange fluorescence of the pigment phycoerythrin. PEUK were differentiated from PRO based on the relatively higher red chl a fluorescence and largest FALS.

2.3. Carbon biomass estimation

Phytoplankton carbon biomass was derived from chl *a* using a carbon: chl *a* ratio of 140 and 83 μ g C: μ g chl⁻¹ for the phase-I and phase-II ML depths, respectively (Shalapyonok et al., 2001). A C: chl *a* ratio of 52 was used for depths below the ML (Brown et al., 1999; Garrison et al., 2000). Carbon biomass of *SYN*, *PRO* (Garrison et al., 2000; Shalapyonok et al., 2001) and PEUK (Shalapyonok et al., 2001) were estimated using conversion factors as given in Table 1. For PEUK, the biovolume calculated from FALS was converted to carbon per cell (Shalapyonok et al., 2001).

2.4. Data analyses

The depth of euphotic zone ($Z_{1\%}$) was estimated from chl *a* concentration of surface layer using the chl centered approach (Lee et al., 2007). The percentage contribution of PP to total phytoplankton carbon biomass and individual PP group contribution to total PP carbon biomass were estimated from the phytoplankton and PP carbon values. PP carbon biomass was depth integrated by trapezoidal method and its percentage contribution to the total phytoplankton biomass was also calculated.

PERMANOVA analysis (Wood et al., 2016) was performed using PRIMER 6 software to assess the variation in the biotic (*SYN*, *PRO*, PEUK, PP carbon biomass, chl *a*, phytoplankton carbon biomass, and PP carbon contribution) and abiotic (nutrients, temperature, salinity, and DO) parameters between phase I (stratified water column) and phase II





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