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Environmental impact on diversity and distribution of tintinnid (Ciliata: Protozoa) along Hooghly Estuary, India: A multivariate approach

Dibyendu Rakshit [a,*], Kadarkarai Murugan [b], Jayanta Kumar Biswas [c], Kamala Kanta Satpathy [d], P. Sankar Ganesh [e], Santosh Kumar Sarkar [a]

[a] Department of Marine Science, University of Calcutta, 35 Ballygunge Circular Road, Calcutta 700019, India

[b] Thiruvalluvar University (State University), Serkkadu, Vellore 632 115, Tamil Nadu, India

[c] Department of Ecological Engineering & Environmental Management, International Centre for Ecological Engineering, University of Kalyani, Nadia, 741235, India

[d] Environment and Safety Division, Indira Gandhi Centre for Atomic Research, Kalpakkam 603 102, Tamil Nadu, India

[e] Department of Biological Sciences, Birla Institute of Technology and Science, Pilani, Hyderabad Campus, Jawahar Nagar, Hyderabad 500078, Telangana State, India

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ABSTRACT

The spatiotemporal distribution, diversity and biomass of the choreotrich ciliate tintinnid, the ubiquitous planktonic protist, were analysed from nine sampling sites ($n = 252$) of diverse environmental stresses along the Hooghly Estuary, eastern part of India during March 2012 to August 2014. Among 32 identified tintinnid species, the agglomerated genera *Tintinnopsis* (20 sp), dominated the community (~62%) followed by *Tintinnidium* (2 sp), *Leprotintinnus* (2 sp), *Codonellopsis*, *Stenosemella*, *Helicostomella*, *Favella*, *Eutintinnus*, *Metacylis*, *Dadayiella*and *Wangiella* (each comprising single species). A wide range of seasonal variations in tintinnid abundance was recorded maximum (2067 ± 893 ind. l^{-1}) for *Tintinnopsis beroidea* and minimum (11 ± 4 ind. l^{-1}) for *Metacylis* sp. during the investigation period. The biomass and daily production rate of tintinnid ranged from 0.004–2.764 μg C l^{-1} and 0.04–3.54 μg C l^{-1} day^{-1} respectively. An overall dominance and diversity of the small-sized tintinnid (lorica length < 76 μm) belonging to the genera *Tintinnopsis* sp., *Tintinnidium* sp., *Codonellopsis* sp., *Wangiella* sp., *Eutintinnus* sp., *Metacylis* sp. and *Helicostomella* sp. was pronounced, accounting ~66% of the total tintinnid abundance. K-dominance curves were plotted against log rank *k*, showed species dominance over the investigated sites. The multidimensional scaling (MDS) Canonical Analysis of Principal coordinates (CAP) highlighting a significantly different spatial distribution of tintinnid. Principal Component Analysis (PCA) map showed clustering of core species with chl *a* and nitrate and could be considered as the crucial factors controlling the distribution and seasonal patterns of tintinnid. Biota-environment (BIOENV) analyses also reveal that these two parameters were the significant causative factors, suggesting that tintinnid may be used as a bioindicator for discriminating water quality in this estuarine system. The study provided detailed information of microzooplankton which enhances our understanding regarding its crucial role in marine ecosystem and complex biotic interactions for maintaining the ecological and economic stability.

1. Introduction

Tintinnid (Ciliata: Protozoa) are ubiquitous and significant components of microzooplankton communities in the marine and estuarine water and play a crucial role in microbial food webs. They are important consumers of the pico- and nano-sized fractions of the plankton and the linkage in transferring energy from the microbial loop to higher trophic levels in the sea (Azam et al., 1983; Jiang et al., 2011). By virtue of their rapid growth rate and sensitive reaction to environmental changes, they have been considered as effective bioindicator of water quality and environmental contamination (Jiang et al., 2011; Kim et al.,

* Corresponding author.
E-mail address: rakshit_dibyendu@yahoo.co.in (D. Rakshit).

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2012; Xu et al., 2008). They are characterized by species specific loricae, which have various shapes ranging from tubular to vase- or bowl-shaped (Dolan, 2013). Lorica morphology is not only a valuable taxonomic characteristic but has also been linked to ecological characteristics of tintinnid especially in terms of feeding activity. The abundance, biomass and composition of tintinnid communities in coastal waters should be much more variable than those in the open sea, due to the greater variability of influencing factors (Vaqué et al., 1997). Tintinnid have many advantages as a favourable bioindicator to evaluate environmental stress and anthropogenic impact in aquatic ecosystems (Jiang et al., 2011, 2013a,b; Xu et al., 2011a,b,c, 2014). With short life cycle, rapid response to environmental changes and the easy comparison on temporal and spatial scales, planktonic ciliates have been successfully used to indicate water quality status in marine ecosystems (Jiang et al., 2011, 2014, 2016; Feng et al., 2015; Xu et al., 2015, 2016a,b). Many tintinnid microbiota can tolerate extremes of environmental conditions to macrofauna (Xu et al., 2011a,b). Thus, ciliated protozoa have been used as favourable bio-indicators of water quality in many aquatic environments (e.g., Jiang et al., 2011, 2013a,b; Xu et al., 2011a, 2014; Rakshit et al., 2017). So far, there have been several studies of marine planktonic tintinnid assemblages (Jiang et al., 2011; Zhang and Wang, 2000; Dolan et al., 2012, 2013a,b). Although there have been a number of recent investigations on bio-assessment using ciliated protozoa, the ability of marine tintinnid for discriminating water quality status is yet to be studied (Jiang et al., 2011; Xu et al., 2011a, 2014). The main objectives of this study were: (1) to characterize the systematic taxonomic composition distribution, abundance, biomass and biodiversity of planktonic ciliates; (2) to reveal the spatial patterns in community structures and determine relationships between tintinnid and abiotic factors in Hooghly Estuary; and (3) to investigate the potential of using estuarine tintinnid communities as an indicators to study the effects of environmental impact.

2. Materials and methods

2.1. Study sites

Hooghly Estuary (87°55′01″N–88°48′04″N latitude and 21°29′02″E–22°09′00″E longitude), the first deltaic offshoot the River Ganges, is a funnel-shaped positive estuary with ever changing geomorphologic and hydrodynamic characteristics. The estuary (shallow depth of ∼6 m and catchment of 6×10^4 km^2) is located in the head Bay of Bengal region is a part of the highly dynamic deltaic environment. Tidal variations are pre-dominant in this estuary, and tides propagate considerable distance through a complex network of various riverine systems, inlets, bays and creeks having vital implications on water mass exchange, reworking of deltaic sediments and the mixing process (Rose et al., 2015). The mean monthly rainfall showed that more than 74% of the annual rainfall occurred during monsoon months (mean annual rainfall ∼1700 mm). Hence, the climate of the area could be classified into 3 seasons as pre-monsoon (March–June), dry season with occasionally higher temperature, southwest monsoon (July–October) accompanied by heavy rainfall and post-monsoon (November–February) characterized by lower temperatures and lower precipitations (Khan, 1995).

The study was conducted on monthly basis from nine sampling sites (covering ∼150 km) along Hooghly Estuary. The sites were almost equidistant from each other and were chosen on the basis of different environmental features, such as tidal environments, wave energy fluxes and distances from the sea (Bay of Bengal) (Fig. 1). The sites can be sub-divided into 3 distinct ecological zones mainly on the basis of salinity regime as follows; Oligohaline (fresh water; salinity <0.5 psu) zone [Barrackpore (S_1), Dakhineswar (S_2), Babughat (S_3) and Budge budge (S_4)]; Mesohaline (brackish water; salinity <5 psu) zone [Nurpur (S_5) and Diamond Harbour (S_6)] and Mixoeuhaline (Estuarine; salinity 18–30 psu) zone [Lot 8 (S_7), Phuldubi (S_8) and Gangasagar (S_9)]. Global positioning system (GPS) was used to fix the geographic position of the sampling sites, which have a mean elevation of 13.7–16.7 m, belonging to a lower deltaic plain experiencing intense semidiurnal tides and wave action.

2.2. Sampling and sample processing

During March 2012 to August 2014, surface water samples were collected monthly from nine sampling sites during high tide in morning hours along estuary. Surface water temperature (°C) and salinity were measured in-situ thermometer (0–110 °C, mercury) and refractometer (0–35 psu) respectively. Turbidity, total dissolved solids (TDS) and pH were measured by Water analyser-371 (Systronics). Dissolved oxygen (DO), biochemical oxygen demand (BOD) and inorganic nutrients (nitrate, silicate and phosphate) were analysed by following the standard protocol devised by Strickland and Parsons (1972). The presence of faecal coliforms in samples was determined by using the most probable number (MPN) procedure of Vanderzant (1992). To analyse chlorophyll *a* (chl *a*), 1 L surface water was collected in amber colour bottles and immediately restored in deep freeze (−20 °C). Samples were filtered through GF/F Whatman filter paper (0.45 μm) and the residue (filter paper) was preserved in 90% acetone solution and was incubated for 24 h. Later, chl *a* concentration was analysed by adopting the technique of Strickland and Parsons (1972) using spectrophotometry (UV–Vis spectrophotometer 117, Systronics). Water quality index (WQI) values were calculated following the equation procured by National Sanitation Foundation (NSF) using 8 water quality parameters as referred above (Yisa et al., 2012).

For collection of tintinnid, 1000 mL from each selected site was collected in pre-cleaned plastic bottles and fixed with 2% Lugol's iodine solution and stored in a cool dark place until analysis (Dolan et al., 2002). In the laboratory, water samples were placed in measuring cylinders of 1000 mL with 2 special outlets at the level of 500 ml and 250 ml respectively which were blocked by clumps and incubated for at least 48 h (Godhantaraman, 2002). After that when almost all the planktons were settled at the bottom of the cylinder, the clumps were opened and the water from the upper portion of the measuring tube was allowed to flow out without disturbing the last 250 mL sample and kept incubated for another 24 h. Finally the sample was concentrated to a volume of 25 mL by syphoning out from rest of the volume and stored in a sterile storage vial (Godhantaraman, 2002). From the remaining 25 mL, 0.1 mL of well-mixed concentrated sample was taken and analysed on a microscope slide. 1 mL sample has been used for identification of tintinnid and counted under a phase contrast microscope at 40× magnification (NIKON Trinocular Microscope, Model E-200). For each species, a minimum of 20 typical individuals were examined for morphological measurements and species identification according to literatures (Kofoid and Campbell, 1929, 1939; Zhang et al., 2012). The dimensions of about 30 individual cells for each taxon were measured and average biovolume of each taxon was estimated from appropriate geometric shapes. The LOD is an important taxonomic characteristic of tintinnid as they use lorica for ingestion of food (Kršinić, 2010). Lorica volume (LV, μm^3) was calculated and transformed to biomass using the regression equation (Verity and Langdon, 1984): Biomass (μgC l^{-1}) $= 444.5 + 0.053\,LV$. Production rate (P, μgC l^{-1} day^{-1}) was estimated from biomass (B, μgC l^{-1}) and empirically determined specific growth rate (g, day^{-1}): $P = B \times G$. Multiple regression equation of Müller and Geller (1993) was used for estimation of growth rate of tintinnid: Growth rate $= 1.52 \ln T - 0.27 \ln CV - 1.44$; where T is the temperature (°C) and CV is the cell volume (μm^3).