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Distribution of nanoflagellates in five water masses of the East China Sea in autumn and winter

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ABSTRACT

The variations of abundance, biomass and trophic structure of nanoflagellates (NF) among five typical water masses in the East China Sea were investigated in autumn (November 19–December 23, 2006) and winter (February 22–March 11, 2007). It was found that water mass had a significant impact on the distribution of NF. Either in autumn or in winter, the highest abundance and biomass of NF were recorded in the East China Sea Shelf Mixing Water (ECSSMW), and the lowest in the Kuroshio Subsurface Water (KSSW). While in the East China Sea Coastal Water (ECSCW), the abundance and biomass of both heterotrophic nanoflagellates (HNF) and pigmented phototrophic nanoflagellates (PNF) were only slightly higher than that in Taiwan Strait Water (TSW) and Kuroshio Surface Water (KSW). In respect to the seasonal variation, the abundance and biomass of NF in TSW declined in winter, while in other 4 water masses, they showed an increasing trend from autumn to winter, mainly due to the decrease (in TSW) or increase (in ECSCW, ECSSMW, KSW and KSSW) of HNF. The distribution pattern of abundance- or biomass-based PNF/HNF ratio was found to be correlated to the nutrient level of the water mass. Results of Pearson correlation analysis and principle component analysis indicated that PNF was mainly constrained by nutrient supply, and HNF was controlled by food availability in the East China Sea.

1. Introduction

Nanoflagellates (NF) dominate the nanoplankton community in pelagic habitats, and they are the integral part of microbial food web (Caron et al., 2009). In the past decades, many researches had been carried out to study the distribution and bacterivory of NF in the world ocean, e.g., those reported by Patterson and Lee (2000), Dennett et al. (2001) and Massana et al. (2006). These studies have significantly pushed forward our understanding of NF distribution and their roles in marine food web (Leadbeater and Green, 2000). Now, it has been widely accepted that NF play important roles in transferring carbon and in nutrient regeneration within the marine microbial food web (Chiang et al. 2013). For pigmented phototrophic nanoflagellates (PNF, including autotrophs and mixotrophs), they constitute the main component of oceanic phytoplankton biomass and sometimes account for major proportion of primary production (Falkowski et al., 2003). Heterotrophic nanoflagellates (HNF) are identified as the main grazer on picoplankton (including heterotrophic bacteria, cyanobacteria and picoeukaryote), which provide an essential link in the transfer of carbon to higher trophic levels (Pernthaler, 2005).

The ecological function of nanoflagellates depends on their size and quantity feature (abundance and biomass), PNF/HNF ration, interaction with prey and predator, etc., which differed from each environment. Many studies have shown a distinct functional role of NF among areas. For example, HNF grazing can account from 1.1% (Sherr et al., 1997) to 430% (Wikner et al., 1990) of bacterial production and differs greatly among habitats. To date, this knowledge is mainly derived by the comparative study among large habits (Bong and Lee, 2011). Comparative study between the different water masses in the coastal region has been rarely reported. Actually we have got no idea about the effect of different water masses on the distribution of the NF in marginal seas. Here we chose the East China Sea as an ideal ecosystem to reveal this uncertainty.

The East China Sea (ECS), a marginal sea east of China, is a part of the Northwest Pacific Ocean and covers an area of 770×10^3 km^2 (Chen, 2009). Seasonal variations of air temperature, river runoff and wind stress in the continental shelf area of ECS result in high variability of temperature, salinity and nutrient concentrations in the near-shore waters. However, outside the continental shelf, as a result of intruding by Kuroshio water, the above variations are rather mild. Based on a broad criteria, water masses in the ECS can be distinguished mainly as Changjiang Diluted Water (CDW), East

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China Sea Coastal Water (ECSCW), East China Sea Shelf Mixing Water (ECSSMW), Taiwan Strait Water (TSW), Kuroshio Surface Water (KSW), Kuroshio Subsurface Water (KSSW), etc. (Chen, 2009; Hu et al., 2010; Su and Weng, 1994). The aim of this study is to probe how these water masses of different nature affect the distribution of NF, and to figure out what are the main factors influencing seasonal variation of NF in different water masses in such a hydrologically complicated marginal sea.

2. Materials and methods

The abundance and trophic structure of nanoflagellates (NF) in the East China Sea were investigated on two cruises in autumn (November 19–December 23, 2006) and winter (February 22–March 11, 2007), respectively (Fig. 1). Samplings were collected from 3 to 6 depths including 2 m, 10 m, 30 m (no sample if water depth larger than 100 m), 50 m, 75 m, 100 m, and bottom layer in the upper 200 m with 10 L Niskin bottles on a Seabird SBE 9/11 CTD rosette.

Water temperature and salinity were measured with probes equipped in the Seabird CTD. Chlorophyll *a* (Chl *a*), Nitrate and Phosphate concentrations were measured following the methods of Parsons et al. (1984).

For enumeration of NF, samples were pre-filtered through 20 μm nylon mesh by gravity and then fixed with cold glutaraldehyde (final conc. 0.5% (V/V)). Subsample (20 ml) was stained with DAPI (final conc. 10 μg ml^{-1}) and filtered onto a 0.2 μm pore size black-stained polycarbonate membrane filters (Millipore) at a low pressure (< 100 mm Hg) (Sherr et al., 1993). Filters were mounted to glass slides and preserved in −20 °C dark condition until laboratory analysis as described in Lin et al. (2013). At least 30 fields of view were counted. In order to estimate the biovolume, cell dimensions of flagellate were measured with Lecia DM4500 self-carried software and transformed to biovolume by analogy to geometrical forms (Sun and Liu, 2003). Conversion to carbon biomass was made using a factor of 220 fg C μm^{-3} (Børsheim and Bratbak, 1987).

Heterotrophic bacteria (HB) and *Synechococcus* abundance were counted using a FACSVantage SE (Becton Dickinson) flow cytometer. Protocols were adapted from Marie et al. (2000). HB biomass was calculated with a carbon conversion factor of 20 fg $cell^{-1}$ (Lee and Fuhrman, 1987). *Synechococcus* cell carbon content was derived from biovolume by using an empirical regression equation: $C\,(pg) = 0.433 \times (Biovolume)^{0.863}$ (Verity et al., 1992). For ciliate enumeration, 1000 ml samples were taken and fixed with acid Lugol's solution (final conc. 1%). The samples were analyzed at 400× with an Olympus IX-51 inverted microscope. Ciliate abundances were converted into biomass using appropriate geometric formulae (Zhang et al., 2002) and a carbon conversion factor of 190 fg C μm^{-3} (Putt and Stoecker, 1989).

The stepwise cluster analysis (Kim, et al., 1991) and temperature–salinity (*T–S*) diagram method (Hur et al., 1999) were applied to delineate water masses. A one-way ANOVA was used to assess differences of NF abundance and biomass within water masses, and then a posteriori Fisher LSD test of mean comparison among water masses was performed. Pearson correlation analysis and principal component analysis were employed to analyze the main factors that influence distribution of NF. For KSSW, due to fewer samples ($n < 3$), no statistical analysis was conducted. All the above statistical analyses were processed with the SPSS software (Version 16, SPSS Inc.).

3. Results

3.1. Water masses in the study area

By stepwise cluster analysis and *T–S* method, five water masses were recognized both in autumn and winter, including East China Sea Coastal Water (ECSCW), East China Sea Shelf Mixing Water (ECSSMW), Taiwan Strait Water (TSW), Kuroshio Surface Water (KSW) and Kuroshio Subsurface Water (KSSW) (Fig. 2). In autumn, ECSCW and ECSSMW had low salinity and temperature, but high Chl *a* concentration and HB abundance. TSW was characterized by higher water temperature, HB and *Synechococcus* abundance, lower in nutrient. Kuroshio water (including KSW and KSSW) was marked by low HB abundance and Chl *a* concentration, but high in salinity. While in winter, water temperature decreased sharply in the coastal area, the highest water temperature appeared in KSW. Higher Chl *a* content and lower salinity were found in the near-shore water mass (ECSCW and ECSSMW). TSW had a moderate temperature, salinity, HB abundance and Chl *a* concentration but the lowest nutrient among the five water masses in winter. The general parameters of above mentioned water masses in autumn and winter are listed in Tables 1 and 2, respectively.

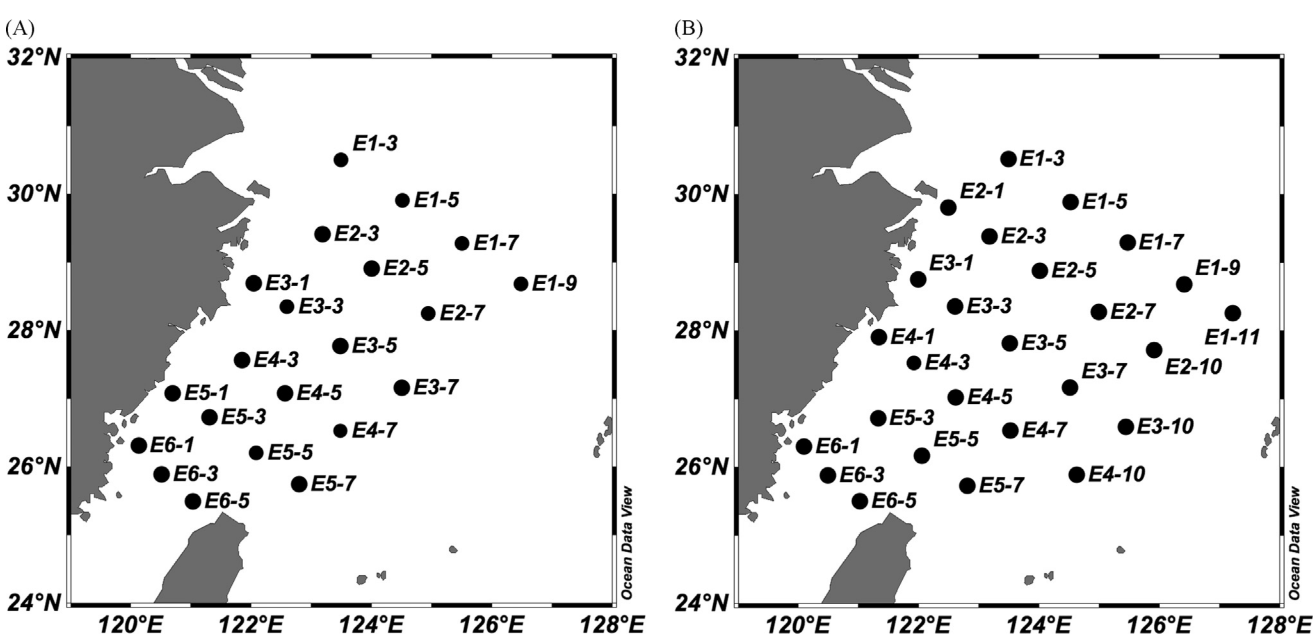

Fig. 1. Locations of sampling stations in the East China Sea in autumn (A) and winter (B).