

# Spatiotemporal variations of inorganic nutrients along the Jiangsu coast, China, and the occurrence of macroalgal blooms (green tides) in the southern Yellow Sea



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## ABSTRACT

Large macroalgal blooms (i.e. green tides of *Ulva prolifera*) occurred in the southern Yellow Sea, China, yearly from 2007 to 2016. They were among the largest of such outbreaks around the world, and these blooms likely originated along the coast of the Jiangsu Province, China. Understanding the roles of nutrients in the onset of these macroalgal blooms is needed to identify their origin. This study analyzes the spatiotemporal variations in dissolved inorganic nitrogen and phosphorus (DIN and PO<sub>4</sub>-P) and the N/P ratio along the Jiangsu coast from 1996 to 2014 during late-March to April, the months which corresponds to the pre-bloom period of green tides since 2007. A zone of high DIN and PO<sub>4</sub>-P concentrations has developed along the Jiangsu coast, between the cities of Sheyang and Nantong, since 1996. There was an 18-year trend of increasing DIN concentrations during the pre-bloom period as well as a positive correlation between the *U. prolifera* biomass and DIN concentrations. Nutrient inputs from rivers and mariculture in the Jiangsu Province may have provided nitrogen that contributed the magnitude of macroalgal blooms that subsequently spread into the southern Yellow Sea.

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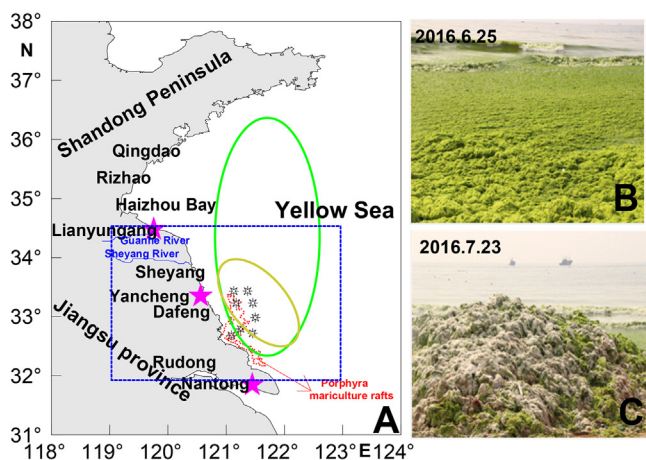
## 1. Introduction

Blooms of green macroalgae are occurring worldwide, and can have large negative ecological and economic impacts (Choi et al., 2006; Guidone and Thornber, 2013; Smetacek and Zingone, 2013; Hu et al., 2014; Perrot et al., 2014). These blooms are commonly associated with coastal eutrophication caused by inputs of nutrients (Valiela et al., 1997; Morand and Merceron, 2004; Ye et al., 2011; Jiang et al., 2014; Li et al., 2014a), such as nitrogen (N) and phosphorus (P) resulting from rapid human population growth, excessive agricultural fertilization, intensive mariculture, and other anthropogenic perturbations (Glibert et al., 2006; Heisler et al., 2008; Zhou et al., 2008), but not inputs of silicon.

Based on nutrient enrichment experiments conducted in Maui (Hawaii, USA), Dailer et al. (2012) found a correlation between macroalgal blooms and increased nutrient concentrations. Nelson et al. (2003) also indicated the strong positive correlation between ulvoid algal biomass and additional anthropogenic N concentrations in the coastal waters of Washington State (USA), and showed that high nutrient supply had a crucial role in macroalgal growth. Perrot et al. (2014) reported that massive green-tide events that have recurred annually in Brittany (France) since the 1970s were related to increasing anthropogenic N input. Similar events occurred in the Ios Island, Greece (Tsagkamilis et al., 2010), Biscayne Bay, USA (Collado-Vides et al., 2013), Knysna Estuary, South Africa (Allanson et al., 2016), Narragansett Bay, USA (Conover et al., 2016), and coastal seas of both Korea (Choi et al., 2006) and Japan (Ogawa et al., 2015), where nutrient inputs from adjacent land use and green macroalgal blooms have increased over the past 14 years (i.e. 2003 to present).

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**Fig. 1.** Maps of the study area. (A) Yellow Sea, China. The green and yellow ellipse denote the areas of macroalgal blooms between 27 April and 9 June 2012 and the initiation period between 27 April and 6 May 2012, respectively (Wang et al., 2015). The blue dashed rectangle represents the study area along the Jiangsu coast, where temporal variations and the horizontal distribution of surface nutrients were studied (the locations of sampling stations are provided in Supplementary Fig. S1). The red irregular dashed frame in this panel corresponds to the area with *Porphyra mariculture* rafts, and the pink solid stars indicate pond-based animal mariculture area. The eight open asterisks indicate the sampling stations that were investigated monthly from October 2010 to October 2011. (B, C) Pictures of the macroalgal blooms in the southern Yellow Sea and the Qingdao coastal area, respectively, in 2016.

Macroalgal blooms in China threatened the 2008 Olympic sailing competition in Qingdao (Jiang et al., 2008) and since 2007 they have occurred annually in the southern Yellow Sea (Fig. 1B,C), with no overall decreasing trend in bloom extent or duration (Liu et al., 2013a; Li et al., 2015; Liu et al., 2016). These macroalgal blooms usually originate in the Jiangsu coastal area during late-April or early May (Wang et al., 2015; Zhou et al., 2015) and start as small patches of floating algae (Liu et al., 2009; Hu et al., 2010), which expand when growth is favorable to large area of the southern Yellow Sea (Fig. 1A). Along the Jiangsu coast, *Porphyra mariculture* has expanded over a large region between the cities of Yancheng and Nantong since 2006, and covers an area of >20,000 ha where 126,000 t of fresh *Porphyra* are harvested annually (Liu et al., 2010; Xing et al., 2015). Mariculture activities have rapidly expanded in the Jiangsu province over the last decade (Pang et al., 2010; Wang et al., 2011). In Lianyungang, Yancheng and Nantong, which are three coastal cities of Jiangsu province, there are large coastal areas of mariculture ponds where fish, shrimp and shellfish, are farmed in ponds. The discharge of nutrient-rich wastewater from the intensive mariculture ponds may contribute to eutrophication along the Jiangsu coast (Liu et al., 2013a; Li et al., 2015). Moreover, the main rivers that flow through Jiangsu province, i.e. the Guanhe and Sheyang Rivers, discharge large quantities of inorganic and organic nutrients into coastal areas in the western Yellow Sea (Liu et al., 2013a). This region is characterized by a sand plateau approximately 200 km long and 100 km wide (Keesing et al., 2011), forming an extended intertidal mudflat region along the Jiangsu coast. The combination of this physical environment and the inputs of nutrients and macroalgal propagules creates near-optimal conditions for the development of macroalgal blooms (Wang et al., 2015).

The importance of nutrients for green-tide blooms in the southern Yellow Sea have been previously studied (Li et al., 2015; Shi et al., 2015; Zhou et al., 2015). Li et al. (2015) reported that increased concentrations of dissolved inorganic nitrogen (DIN) may be one of the most important factors contributing to the large extent and long duration of green tides. Nevertheless, the role of

nutrients in initiating the green-tide bloom along the Jiangsu coast has not been investigated previously.

The present study addresses the hypothesis that changes in nutrient concentrations of waters along the Jiangsu coast since 2007 are correlated with the initial development of blooms of green tides in the southern Yellow Sea. A combination of published data, for years 1996, 1998, 2001, 2005, 2007, 2010, 2011, 2012 and field-collected information, for years 1997, 2003, 2006, 2008, 2014 were used to examine the relationship between nutrient concentrations and bloom onset for 1996–2014.

## 2. Materials and methods

### 2.1. Study area

The study area (119–123°E, 32–34.5°N) is located in the southwestern Yellow Sea and encompasses waters off the Jiangsu coast (Fig. 1A).

### 2.2. Data sources

This study reports the temporal variations of dissolved inorganic nitrogen ( $\text{DIN} = \text{NO}_3\text{-N} + \text{NH}_4\text{-N} + \text{NO}_2\text{-N}$ ), inorganic phosphate ( $\text{PO}_4\text{-P}$ ) and the N/P (i.e.  $\text{DIN}/\text{PO}_4\text{-P}$ ) ratio along the Jiangsu coast during the pre-bloom period late-March to April 1996–2014. The data were from both previously published studies (see Supplementary Table S1 for the dates, stations and nutrients sampled, and references) and five cruises conducted in 1997, 2003, 2006, 2008 and 2014 (previously unpublished data, Supplementary Fig. S1).

In addition to the 1996–2014 nutrient data series described above, in order to analyze nutrient conditions before (from late March to April), during (from May to August) and after (from September to early March of the following year) green-tide blooms, DIN and  $\text{PO}_4\text{-P}$  concentrations were measured monthly for one year (October 2010 to October 2011) at eight sites located near *Porphyra mariculture* rafts along the Jiangsu coast (Fig. 1A). Surface nutrient conditions were also determined during the pre-bloom period, from 25 March to 16 April 2012 along the Jiangsu coast (first bloom detected on 16 April 2012). In order to compare the nutrient characteristics of the stations in the cruise of 25 March to 16 April 2012, the 34 sampling stations were divided in two groups according to the biomass of *U. prolifera* in surface waters (0–5 m) during the initial period (2–6 May; Supplementary Fig. S2) of the 2012 macroalgal bloom, biomass i.e.  $>5 \text{ g wet weight m}^{-2}$  and biomass  $<5 \text{ g wet weight m}^{-2}$ , respectively (Liu et al., 2015; Wang et al., 2015).

The temporal patterns of surface nutrients during the entire macroalgal bloom period in the southern Yellow Sea were measured during nine cruises between 12 March and 4 September 2012. Information about sample locations, stations and general conditions during on these cruises and general biomass distributions are in Supplementary Fig. S3 and Table S2.

### 2.3. Sampling analysis

Surface water samples (0–5 m) were collected with 12-L Niskin bottles and nutrient concentrations were measured on seawater that was passed through GF/F filters, which were pre-combusted at 450 °C for 4 h. Dissolved inorganic nitrogen and  $\text{PO}_4\text{-P}$  were measured by spectrophotometry (Grasshoff et al., 1999) on board the ship. Total dissolved nitrogen (TDN) and phosphorus (TDP) were measured in the laboratory on samples frozen at  $-20^\circ\text{C}$ , using persulfate oxidation (Valderrama, 1981). The analytical precisions of the  $\text{NO}_3\text{-N}$ ,  $\text{NO}_2\text{-N}$ ,  $\text{NH}_4\text{-N}$ ,  $\text{PO}_4\text{-P}$ , TDN and TDP

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