



## Spatial and temporal dynamics of phytoplankton and bacterioplankton biomass in Sanya Bay, northern South China Sea

ZHOU Weihua<sup>1,2,3,4,\*</sup>, LI Tao<sup>1,2</sup>, CAI Chuanghua<sup>1,2</sup>, HUANG Liangmin<sup>1,2</sup>,  
WANG Hankui<sup>1,2</sup>, XU Jirong<sup>1,2</sup>, DONG Junde<sup>1,2</sup>, ZHANG Si<sup>1,2</sup>

1. LED and LMB, South China Sea Institute of Oceanology, Chinese Academy of Sciences, Guangzhou 510301, China. E-mail: [whzhou@scsio.ac.cn](mailto:whzhou@scsio.ac.cn)

2. Tropical Marine Biological Research Station in Hainan, Chinese Academy of Sciences, Sanya 572000, China

3. State Key Laboratory of Organic Geochemistry, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences, Guangzhou 510640, China

4. Laboratory of Marine Ecosystem and Biogeochemistry, SOA, Second Institute of Oceanography, SOA, Hangzhou 310012, China

Received 10 June 2008; revised 25 July 2008; accepted 07 August 2008

### Abstract

The composition of phytoplankton and the dynamics of phytoplankton and bacterioplankton biomass (PB and BB, respectively) of Sanya Bay, South China Sea, were determined. A total of 168 species (67 genera) phytoplankton were identified, including Bacillariophyta (diatom, 128 species), Pyrrophyta (35 species), Cyanophyta (3 species), and Chrysophyta (2 species). Annual average abundance of phytoplankton was  $1.2 \times 10^7$  cells/m<sup>3</sup>, with the highest abundance in autumn, and the lowest in summer. Annual average diversity index ( $H'$ ) and evenness ( $J$ ) values were 3.96 and 0.70, respectively. Average chlorophyll-*a* was 2.5 mg/m<sup>3</sup>, and the average PB was 124 mg C/m<sup>3</sup>, with the highest value in autumn. Surface PB was higher than the bottom, except for summer. Annual mean bacterioplankton abundance and BB were  $6.9 \times 10^{11}$  cells/m<sup>3</sup> and 13.8 mg C/m<sup>3</sup>, respectively. The highest BB was found in summer, followed by winter, spring, and autumn. Surface BB was higher than bottom all year round. The spatial distribution patterns of PB and BB were very similar with the highest biomass in the estuary, and decreased seaward, primarily due to the terrestrial input from the Sanya River and influx of oceanic water. The main factor influencing PB and BB was dissolved inorganic nitrogen (DIN). Other factors such as temperature, which is above 22°C throughout the year, had a negligible impact. The correlation between BB and PB was significant ( $P < 0.01$ ). The annual average ratio of BB/PB was 0.12 (0.06–0.15). Phytoplankton primary production was one of the most important factors in controlling the distribution of bacterioplankton.

**Key words:** biomass; phytoplankton; bacterioplankton; Sanya Bay; northern South China Sea

**DOI:** 10.1016/S1001-0742(08)62313-X

### Introduction

Phytoplankton are the main primary producers and they are the base of the marine food chain and require light and nutrients to synthesize organic matter. Chlorophyll *a* (chl-*a*) is the main photosynthetic pigment, and is an index reflecting the amount of phytoplankton biomass (Zhou *et al.*, 2004). Bacterioplankton are the primary component of the microbial food loop, and provide a trophic link to higher organisms (Fuhrman and Azam, 1980). They occupy a range of ecological niches. Many saprotrophic bacterioplankton mineralize organic matter that was released by living plankton or decomposed from particulate materials such as marine snow (Ducklow, 1993). Many other bacterioplankton are autotrophic, and derive energy from either photosynthesis or chemosynthesis (Fukami *et al.*, 1996). It has been shown that bacterioplankton play a central role in the carbon flux in aquatic ecosystems, and also play roles in ecological pathways such as nitrogen

fixation, nitrification, denitrification, and methanogenesis (Häder *et al.*, 1998).

Sanya Bay (18° 11'–18° 18'N, 109° 20'–109° 30'E), a typical bay with a tropical coral reef, is located in the northern part of South China Sea and the southernmost coast of Hainan Island, China. It covers an area of 120 km<sup>2</sup>. Dongmao Island, Ximao Island and Luhuitou Peninsula compose most of the coastal coral reef area. Sanya River (length 31.3 km, drainage area 337 km<sup>2</sup> and annual flow of  $2.11 \times 10^9$  m<sup>3</sup>), the main river flowing into the bay, is located in the eastern part of the bay (Wang *et al.*, 2002; Huang *et al.*, 2003). With the rapid development of the tourism and the sudden increase in the population in the past 10 years, the bay is facing many ecological problems (Chen *et al.*, 1999). The previous ecological and biological studies were mainly focused on the survey of traditional biological resources (Huang *et al.*, 2001), such as phytoplankton (Wang *et al.*, 2002; Huang *et al.*, 2003), zooplankton (Yin *et al.*, 2004a, 2004b; Tan *et al.*, 2004), benthos and fishes (Huang *et al.*, 2003). However, little is known about the dynamics of phytoplankton and bacterio-

\* Corresponding author. E-mail: [whzhou@scsio.ac.cn](mailto:whzhou@scsio.ac.cn)

plankton biomass in the bay since the study of Ning *et al.* (1999) who described the distribution of chl-*a* and bacteria. The objectives of this study were: (1) to assess the status of phytoplankton and bacterioplankton in Sanya Bay; (2) to determine the physical and chemical factors which govern the phytoplankton and bacterioplankton biomass; (3) to provide the background information for the energy flow and material circulation of food chains and the microbial food loop in this bay with coral reefs.

## 1 Materials and methods

### 1.1 Study area and sampling methods

Four seasonal sampling in Sanya Bay, northern South China Sea were conducted in winter (January 12–13), spring (April 19–20), summer (July 10–11), and autumn (November 5–6) in 2005, respectively. The location of sampling stations is shown in Fig. 1. Four sections with two additional stations (Station 1 at the estuary and Station 12 near Sanya Harbor) were included in the study area. Station 12 was a contrast station, and it had no contribution to the statistics tables and distribution figures, since we only took samples in the surface layer. The water depth was < 20 m at all stations, except Station 4 (26 m) and Station 9 (28 m).

Temperature and salinity were measured with Quanta Water Quality Monitoring System (Hydrolab, Texas, USA). Water samples were collected with 5 L Niskin bottles attached to a nylon cable. Sub-samples for bacterioplankton were preserved with 3.7% formaldehyde (V/V, final concentration) in the sterilized tube, and then stored at 4°C. Phytoplankton were sampled with a standard Shallow III Microplankton Net (diameter 37 cm, mesh fiber JF62, mesh size 0.077 mm) that was hauled vertically from the bottom to the surface layer, strictly followed the standard method (China State Bureau of Technical Supervision, 1991a). Samples were mixed with buffered formaldehyde to obtain a final concentration of 2.5%. Enumeration and identification of phytoplankton were performed using an Olympus IX51 Inverted Microscope (Olympus, Japan). Water samples (250 mL) for chl-*a* measurement were passed through 0.7 µm of Whatman GF/F glass fiber filters (25 mm). The filters were stored in aluminum foil and immediately kept at –20°C until laboratory analysis within half a month.

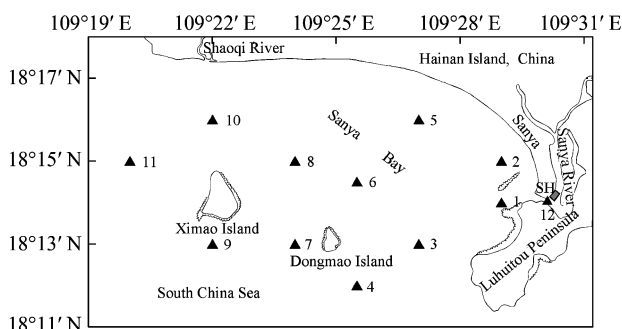


Fig. 1 Survey stations in Sanya Bay, Hainan Island, and South China Sea. The gray area in Sanya River is the location of Sanya Harbor (SH).

### 1.2 Bacterioplankton, chl-*a* and nutrients analysis and data processing

Bacteria were counted by the technique of 4',6-diamidino-2-phenylindole (DAPI)-staining with DNA-intercalating dye DAPI (Velji and Albright, 1986; Zheng and Cai, 1993). Attached bacterioplankton were dislodged by dispersing them in a suspension medium using a combination of chemical treatment and ultrasound. Details are as follows: 1 mL sample was taken from a sterilized tube, 100 µL of 0.01 mol/L sequestering and deflocculating agent (e.g., tetrasodium pyrophosphate) was added, and then incubated for at least 15 min. After that, samples were sonicated (300 r/min) for 40 s in an ice-cold circumstance. The suspension samples were stained with 200 µL (100 µg/mL) DAPI, and kept in the dark for at least 5 min, then filtered through 0.2 µm black polycarbonate membranes (Millipore, Isopore™, 25 mm). Membranes were mounted on microscope slides in immersion oil, and were counted immediately using an epifluorescence microscope (Olympus BX41, Japan). DNA-DAPI complex fluorescence appeared as a bright blue color, while unbound DAPI and non-DNA complex appeared as yellow. Ten fields with about 15–30 bacterioplankton per field were counted. Cell density ( $D_c$ , cells/m<sup>3</sup>) was determined by Eq. (1) (Zheng *et al.*, 2002):

$$D_c = A \times S_1 / (S_2 \times V) \quad (1)$$

where,  $A$  is the mean cells of 10 fields;  $S_1$  is the effective filtration area of the membrane;  $S_2$  is the area of visual field of the microscope;  $V$  is the volume of filtrated water sample. Bacterioplankton biomass (BB) was calculated with a carbon conversion factor of 20 fg C/cell (Lee and Fuhrman, 1987).

Phytoplankton biological diversity ( $H'$ ) and evenness ( $J$ ) were calculated according to Eq. (2) (Shannon and Wiener, 1949; Pielou, 1966):

$$H' = - \sum_{i=1}^S P_i \log_2 P_i \quad (P_i = n_i/N); \quad J = \frac{H'}{\log_2 S} \quad (2)$$

where,  $n_i$  is individual amount of the species organism;  $N$  is total individual amount;  $S$  is the total species.

Filters with chl-*a* were extracted with 10 mL of 90% acetone and sonicated for 10 min in an ice-cold water bath in the dark, and then, extracted at 4°C in the dark for 24 h. The fluorescence of the extract was measured with a Turner Designs model 10-AU fluorometer (Parsons *et al.*, 1984; Zhou *et al.*, 2004). Phytoplankton biomass (PB, mg C/m<sup>3</sup>), was determined from a conversion factor of 50 mg C/mg chl-*a* (Krempin and Sullivan, 1981; Harris, 1986). DIN (dissolved inorganic nitrogen, NO<sub>3</sub><sup>-</sup>-N + NO<sub>2</sub><sup>-</sup>-N + NH<sub>4</sub><sup>+</sup>-N), phosphate (PO<sub>4</sub><sup>3-</sup>-P), DO and BOD<sub>5</sub> were analyzed by the standard methods (China State Bureau of Technical Supervision, 1991b).

The statistical analysis software SPSS 11.0 ANOVA test was used to examine the differences in seasonal changes of PB and BB. Pearson correlation analysis was used to test the relation between environmental parameters and

biomass (PB and BB). Probabilities ( $P < 0.05$ ) were considered to be significant.

## 2 Results and discussion

### 2.1 Physical and chemical environment in Sanya Bay

Seasonal variations of environmental parameters in Sanya Bay are shown in Table 1. Salinity was high in the bay all around the year, with an annual average of 34.3. The lowest salinity appeared in autumn and the highest appeared in winter, and the difference between the two seasons was only 0.8. The annual temperature was 25.8°C, and the seasonal variation was opposite with salinity. Stratification in summer was well developed due to the temperature and salinity differences between the surface and the bottom waters. The bay was affected by cold-water upwelling during summer, which was also reported by others (Huang *et al.*, 2003). The waters were mixed well in winter owing to the Coriolis effect (force) and the north-eastern monsoon winds.

Nutrients concentrations increased shoreward and clearly demonstrated the impact from the terrestrial input and the Sanya River. The nutrients in the bay were low, especially  $\text{PO}_4^{3-}\text{-P}$ , according to the National First Class Water Quality Standards. The highest DIN and  $\text{PO}_4^{3-}\text{-P}$  concentrations occurred in autumn and winter, respectively. The average  $\text{BOD}_5$  over all seasons was  $< 0.4$  mg/L and ranged from 0.29 to 0.37 mg/L. The highest DO occurred in winter due to the strong mixing and the lowest was detected in autumn, while the DO in spring was similar to that in summer (ca. 7.0 mg/L). The DO level was 5.1–7.9 mg/L in 2005, with the average of 6.9 mg/L. As Station 12 is located downstream of Sanya Harbor, the water was already eutrophied and polluted seriously. The average DIN in station 12 was 21.5  $\mu\text{mol/L}$ , with the highest value in autumn (54.3  $\mu\text{mol/L}$ ). The annual average  $\text{PO}_4^{3-}\text{-P}$  was

1.60  $\mu\text{mol/L}$ , DO was  $< 5$  mg/L and  $\text{BOD}_5$  was 1.92 mg/L in the water column in the surface of station 12.

With the expansion of the tourism and the rapid population growth of Sanya City (more than  $5 \times 10^5$  inhabitants), the impact of human activities on the bay is increased significantly, which are causing more nutrients inputs into the bay. For example, in summer, DIN reached 26  $\mu\text{mol/L}$  in Sanya Harbor in 1999 (He *et al.*, 2000), and increased to ca. 50  $\mu\text{mol/L}$  in 2005 (this study). But in 2007, DIN exceeded 60  $\mu\text{mol/L}$  at the harbor waters (Zhou *et al.*, unpublished data).

### 2.2 Phytoplankton species diversity

A total of 168 species of phytoplankton were identified over four seasons. These belong to 67 genera of the Bacillariophyta (128 species), Pyrrophyta (35 species), Cyanophyta (3 species) and Chrysophyta (2 species). Diatoms dominated the phytoplankton community all year around, and almost 80% of the total phytoplankton were diatoms. Of the 128 diatom species, the main genera were *Chaetoceros* (25 species), *Coscinodiscus* (24 species) and *Rhizosolenia* (12 species). The ecotypes of phytoplankton were mostly marine warm water species and eurytopic species.

The seasonal dominant species of phytoplankton are shown in Table 2. *Bacillaria paradoxa* and *Rhizosolenia styliformis* dominated throughout the year in the bay. *B. paradoxa* was the predominant species in summer and winter, and made up 15% and 25% of the total phytoplankton cells, respectively. *R. styliformis*, made up 26% of the total cells, ranked first in abundance in spring. While in autumn, *Thalassionema nitzschioides* became the key species with 15% of the total phytoplankton abundance. *Trichodesmium thiebautii*, one of the cyanobacteria, bloomed in spring and summer, and comprised 16% and 10% of the total cells, respectively.

**Table 1** Seasonal physical and chemical parameters, phytoplankton biomass (PB) and bacterioplankton biomass (BB) in Sanya Bay

Parameter	Spring	Summer	Autumn	Winter
Temperature (°C)	26.7 ± 0.3 (26.2–27.1)	26.6 ± 2.4 (22.0–28.9)	27.2 ± 0.4 (26.9–28.5)	22.8 ± 0.2 (22.4–23.0)
Salinity	34.4 ± 0.1 (34.1–34.5)	34.4 ± 0.2 (34.2–34.8)	33.8 ± 0.1 (33.6–34.0)	34.6 ± 0.1 (34.4–34.7)
DIN ( $\mu\text{mol/L}$ )	1.8 ± 1.2 (1.0–5.8)	1.5 ± 0.3 (1.1–2.1)	3.7 ± 8.3 (1.0–40.4)	1.8 ± 1.4 (1.1–7.6)
$\text{PO}_4^{3-}\text{-P}$ ( $\mu\text{mol/L}$ )	0.06 ± 0.02 (0.04–0.09)	0.11 ± 0.12 (0.04–0.56)	0.11 ± 0.31 (0.04–1.48)	0.28 ± 0.12 (0.04–0.61)
DO (mg/L)	7.0 ± 0.2 (6.4–7.5)	6.9 ± 0.2 (6.6–7.2)	6.5 ± 0.5 (5.1–6.9)	7.2 ± 0.3 (6.6–7.9)
$\text{BOD}_5$ (mg/L)	0.37 ± 0.34 (0.03–1.19)	0.33 ± 0.17 (0.03–0.65)	0.29 ± 0.25 (0.01–0.95)	0.29 ± 0.27 (0.02–1.06)
Chl- <i>a</i> (mg/m <sup>3</sup> )	1.9 ± 4.0 (0.3–16.8)	2.1 ± 1.3 (0.6–6.6)	3.6 ± 2.0 (1.3–8.0)	2.3 ± 3.6 (0.7–15.5)
PB (mg C/m <sup>3</sup> )	96.3 ± 200 (14.8–842)	105 ± 66.0 (28.9–328)	182 ± 98.2 (63.0–400)	112 ± 181 (36.8–773)
BA (×10 <sup>11</sup> cells/m <sup>3</sup> )	6.6 ± 2.4 (3.5–13.7)	8.1 ± 2.4 (4.9–13.8)	5.3 ± 2.4 (2.8–10.9)	7.6 ± 3.7 (3.8–21.9)
BB (mg C/m <sup>3</sup> )	13.2 ± 4.9 (6.1–27.4)	16.3 ± 4.7 (9.8–27.6)	10.5 ± 4.7 (5.5–21.9)	15.3 ± 7.5 (7.6–43.9)

DIN: dissolved inorganic nitrogen; BA: bacterioplankton abundance. Data shown as mean ± SD (range),  $n = 22$ .

**Table 2** Relative seasonal abundance of phytoplankton in Sanya Bay

Species	Spring	Summer	Autumn	Winter
<b>Bacillariophyta</b>				
<i>Bacillaria paradoxa</i>	+	+++	+	+++
<i>Bacteriastrium comosum</i>				+
<i>Bacteriastrium varians</i>			++	
<i>Biddulphia sinensis</i>		+	+	+
<i>Chaetoceros affinis</i>			+	+
<i>Chaetoceros coasteacanei</i>		+	+	
<i>Chaetoceros lorenzaianus</i>	+			
<i>Coscinodiscus asteromphalus</i>		+		
<i>Guinardia slacida</i>	+	+	+	
<i>Melosira nummuloide</i>		+		+
<i>Nitzschia longissima v. reversa</i>				+
<i>Pseudonitzschia pungens</i>			+	++
<i>Rhizosolenia alata</i>	+			
<i>Rhizosolenia calcar-avis</i>	+	+		
<i>Rhizosolenia stouterforthii</i>				+
<i>Rhizosolenia styliformis</i>	+++	+	+	+
<i>Thalassionema nitzschioides</i>		+	+++	+
<i>Thalassiosira subtilis</i>		+		
<i>Thalassiothrix fransfeldii</i>		+	+	+
<b>Cyanophyta</b>				
<i>Trichodesmium thiebautii</i>	++	++		

+++: predominant species; ++: second dominant species; +: dominant species.

The phytoplankton species showed a clear seasonal succession with an average annual abundance of  $1.2 \times 10^7$  cells/m<sup>3</sup>. The highest phytoplankton abundance appeared in autumn, while the lowest was in summer (Table 3), with a second peak in spring. The average  $H'$  and  $J$  values were 3.96 (ranged 2.44–4.76) and 0.70 (ranged 0.44–0.87), respectively.

Phytoplankton species in Sanya Bay were much less than that collected in 2000 (Huang *et al.*, 2003). Phytoplankton species, diversity index ( $H'$ ) and evenness ( $J$ ) in the tropical Sanya Bay were much higher than those in the sub-tropical Daya Bay in Southeast, China (Wang *et al.*, 2004) and the temperate of Jiaozhou Bay (Li *et al.*, 2005) in North of China, respectively.

### 2.3 Spatial-temporal variations of phytoplankton biomass

The chl-*a* concentration in Sanya Bay ranged from 0.3 to 10.8 mg/m<sup>3</sup> and the average of 2.5 mg/m<sup>3</sup> was much higher than the concentration at the end of 1990s (Huang *et al.*, 2003). The seasonal variation in chl-*a* was in the following order from the highest to the lowest: autumn, winter, summer, and spring, with averages of 3.6, 2.3, 2.1, and 1.9 mg/m<sup>3</sup>, respectively (Table 1). The chl-*a* in the surface layer was higher than in the bottom layer, except in summer. The annual average PB was 124 mg C/m<sup>3</sup>.

Chl-*a* was the highest in the Sanya River estuary, and it decreased offshore (Fig. 2). The lowest phytoplankton biomass appeared near Dongmao Island in the bay, with chl-*a* < 1 mg/m<sup>3</sup> in winter, spring and summer. In autumn, the contour lines were close together in the estuarine zone, parallel with the shoreline and formed a tongue like distribution westward in the other three seasons. The annual average chl-*a* at Station 12 near the Sanya Port was 26.3 mg/m<sup>3</sup>, with the annual PB value of 1316 mg C/m<sup>3</sup>. It was 10 times higher than the value in the bay, due to severe pollution and excessive nutrient inputs. The highest chl-*a* at Station 12 appeared in winter with the value 37.8 mg/m<sup>3</sup>, and the lowest occurred in autumn (ca. 10 mg/m<sup>3</sup>). These concentrations indicated that serious eutrophication occurred in the port waters, which should be investigated further by the local government.

There was higher phytoplankton abundance in the coast near the city and its adjacent water than in the bay. The diatom biomass ( $B_d$ , mg/m<sup>3</sup>) showed a clear linear relationship with chl-*a* concentration ( $P_{chl-a}$ , mg/m<sup>3</sup>).  $P_{chl-a}$  can be expressed as a function of the  $B_d$  shown as follows (Eq. (3)):

$$P_{chl-a} = 1.12 \times B_d + 0.21 \quad (R^2 = 0.98, P < 0.001) \quad (3)$$

### 2.4 Spatial-temporal variations of bacterioplankton biomass

The annual mean BA and BB was  $6.9 \times 10^{11}$  cells/m<sup>3</sup>, and 13.8 mg C/m<sup>3</sup>, respectively, and the value in the surface layer was higher than that in the bottom layer. The average value of BA in summer was  $8.1 \times 10^{11}$  cells/m<sup>3</sup>, which was the highest over the four seasons. The second highest value occurred in winter with a mean value of  $7.6 \times 10^{11}$  cells/m<sup>3</sup> and the lowest occurred in autumn with a mean value of  $5.3 \times 10^{11}$  cells/m<sup>3</sup> (Table 1). The annual mean BA and BB of Station 12 was  $42.8 \times 10^{11}$  cells/m<sup>3</sup> and 85.6 mg C/m<sup>3</sup>, respectively, which was almost 6 times higher than in Sanya Bay.

The seasonal horizontal distribution pattern of bacterioplankton was similar to that of chl-*a* (Fig. 3). The highest abundance appeared near the river mouth and gradually decreased to the west. Outside the Sanya River mouth, BA formed a dome-shaped pattern except in autumn. The contours were intensive near the estuary and were sparse in the open sea (Fig. 3).

### 2.5 Environmental regulation of phytoplankton and bacterioplankton biomass

Spatial-temporal variation characteristics of phytoplankton and bacterioplankton can be controlled by some key processes, which can be divided into two groups: bottom-up and top-down control (Li and Fan, 2004). Phytoplankton growth was significantly controlled by environmental factors, such as light, temperature, and nutrients, which are the bottom-up controlling factors. Some processes, such as the grazing by zooplankton, sinking and lysis by biological pathogens can play important roles in the top-down control of phytoplankton (Sun *et al.*, 2004). This study focused on the bottom-up control processes, which affected the distribution pattern of phytoplankton and bacterioplankton.

Sanya Bay is a shallow water bay with an average depth of 16 m. In spring and summer, the transparency could reach the depth of 6.1 and 7.0 m, respectively. There was sufficient sunlight for phytoplankton in the bottom layer to carry out photosynthesis because sunlight could pass through the whole water column. Light was not the limiting factor for phytoplankton growth. The bay is also affected by cold-water upwelling occurring during June to August (Dong *et al.*, 2002; Huang *et al.*, 2003). The stable water in summer can help nutrients to be released from the sediment to the overlying water supplying algal growth in the bottom layer (Wang *et al.*, 2003). These were the main factors that induced the high PB levels in the bottom layer in summer. The concentrations of chl-*a* in surface and bottom layer

**Table 3** Summary of phytoplankton species and seasonal abundance in Sanya Bay

Parameter	Spring	Summer	Autumn	Winter
Total species	114	114	115	116
Abundance ( $\times 10^6$ cells/m <sup>3</sup> )	10.3 $\pm$ 12.9 (3.1–38.9)	7.6 $\pm$ 0.8 (6.5–9.1)	18.5 $\pm$ 11.3 (6.1–41.1)	9.4 $\pm$ 5.6 (2.6–17.9)
Diversity index ( $H'$ )	3.65 $\pm$ 0.62 (2.44–4.06)	4.13 $\pm$ 0.50 (3.57–4.76)	4.15 $\pm$ 0.30 (3.69–4.47)	3.90 $\pm$ 0.43 (3.10–4.20)
Evenness ( $J$ )	0.65 $\pm$ 0.11 (0.44–0.73)	0.79 $\pm$ 0.07 (0.67–0.87)	0.73 $\pm$ 0.06 (0.65–0.80)	0.64 $\pm$ 0.08 (0.57–0.76)

Data shown as mean  $\pm$  SD, except total species,  $n = 22$ .

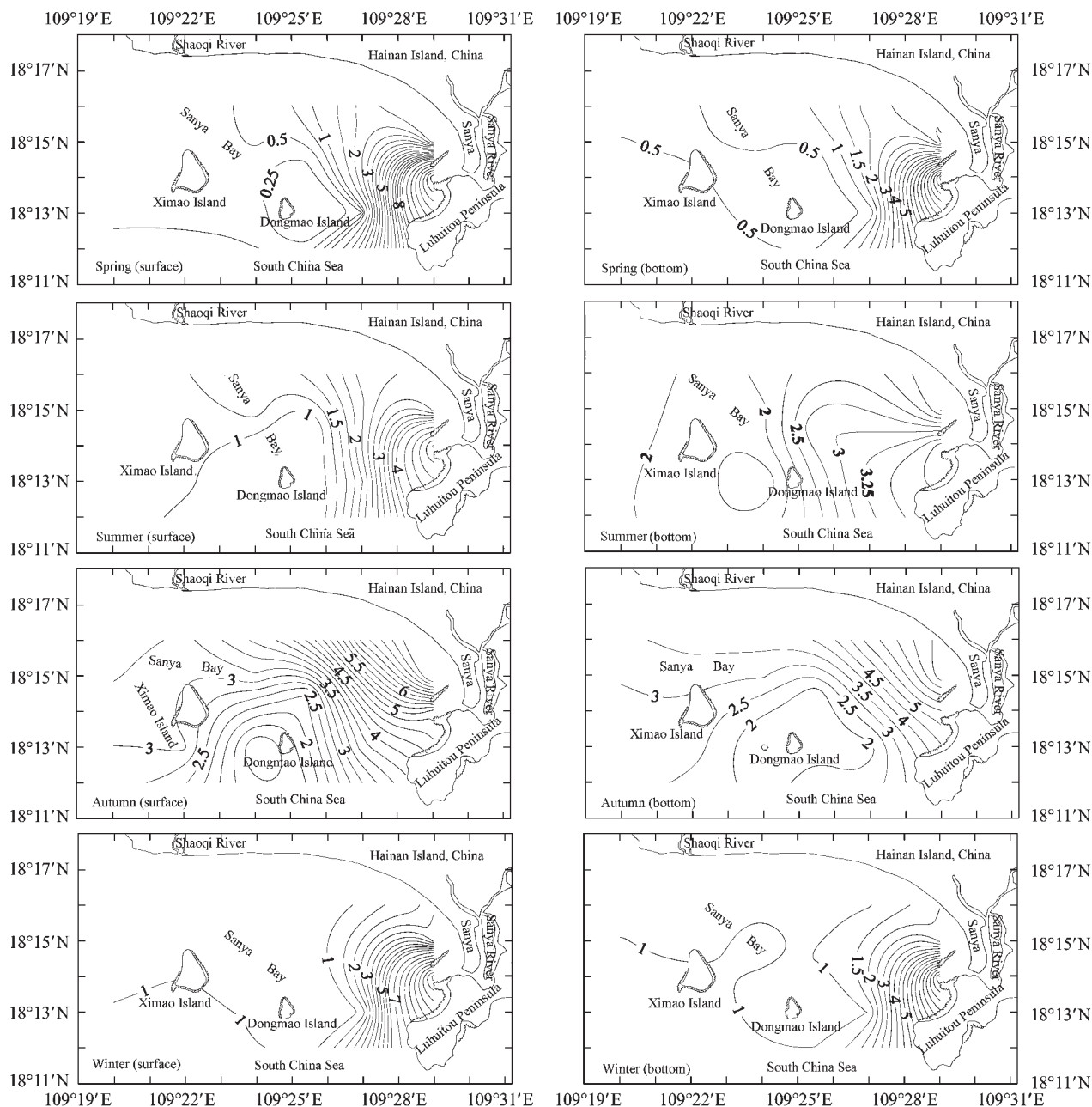


Fig. 2 Seasonal horizontal distribution of chl- $\alpha$  ( $\text{mg}/\text{m}^3$ ) in the surface and bottom layer in Sanya Bay.

were similar in autumn and winter due to the strong mixing that occurred in the dry season by the northeast monsoon winds.

Shiah and Ducklow (1994) reported that temperature played a major role in regulative functions and became the main regulating factor for bacterioplankton growth

than nutrients, except in summer (temperature > 20°C). In summer, temperature (> 20°C) was less important than nutrients. Since Sanya Bay is a tropical, the temperature of sea water was > 22°C all year round, and the seasonal temperature differences were < 5°C. The correlations between temperature and biomass (PB and BB) were not significant

Table 4 Correlation coefficient ( $R$ ) between environmental factors and biomass (PB and BB)

Parameter	Spring		Summer		Autumn		Winter	
	PB	BB	PB	BB	PB	BB	PB	BB
Temperature	-0.19	-0.20	-0.16	-0.26	-0.04	-0.01	-0.20	-0.29
DIN	0.96**	0.90**	0.76**	0.74**	0.69**	0.87**	0.97**	0.90**
PO <sub>4</sub> <sup>3-</sup> -P	0.51*	0.42	0.18	0.17	0.17	0.57**	0.73**	0.60**
DO	0.38	-0.38	0.08	-0.04	0.24	-0.55*	0.54*	-0.41
BOD <sub>5</sub>	0.55*	0.53*	0.23	0.13	0.16	0.20	0.71**	0.76**

\*\* and \* represent correlation significance at  $P < 0.01$  and  $P < 0.05$  level, respectively.

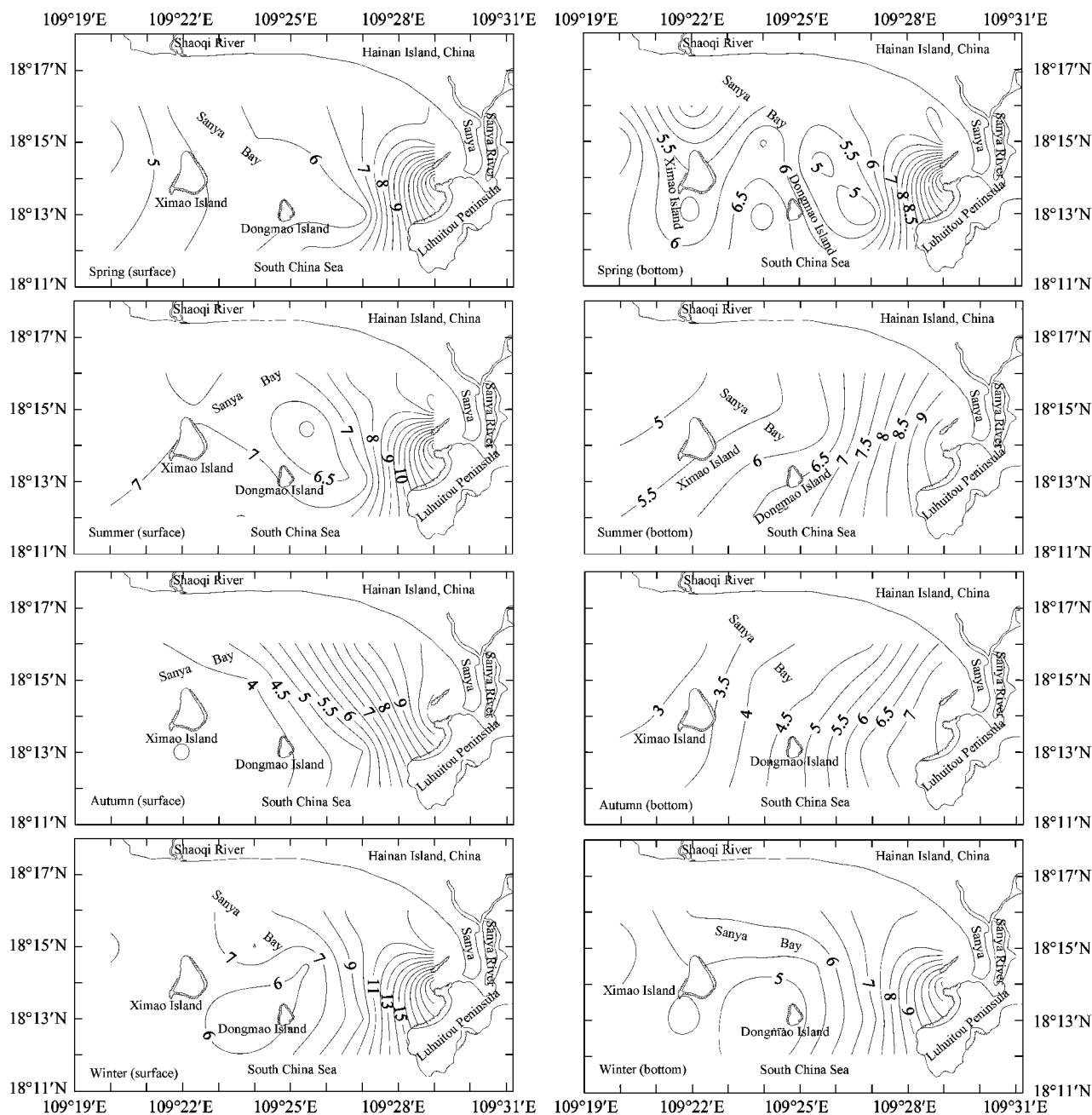


Fig. 3 Seasonal horizontal distribution of bacterioplankton abundance ( $\times 10^{11}$  cells/m<sup>3</sup>) in the surface and bottom layer in Sanya Bay.

(Table 4). Temperature was not the controlling factor of the biomass, and the distribution pattern of phytoplankton and bacterioplankton.

The main terrestrial nutrient source for the bay was from the Sanya River, which directly controlled the distribution of PB and BB, e.g., gradually decreased from the river mouth to the open coastal waters. This significantly demonstrated the fact that the influence of land-based nutrients sources to the biomass distribution pattern was diluted by seawater. The harbor at the mouth of the Sanya River was highly eutrophicated with chl-*a* > 10 mg/m<sup>3</sup> all year. The abundance of bacterioplankton was an important indicator of water quality, and BB was always higher in the seriously polluted waters (Ning *et al.*, 1999). The abundance of bacterioplankton in the port was 10 times higher than the average value in Sanya Bay.

The correlations of environmental factors with PB, as well as with BB are shown in Table 4. DIN had highly significant correlations with PB and BB ( $P < 0.01$ ) all year, which obviously indicated that DIN was the leading factor among all inorganic nutrients that controlled the biomass of phytoplankton and bacterioplankton. PO<sub>4</sub><sup>3-</sup>-P had highly significant correlation with PB ( $P < 0.01$ ) in winter, and the correlation was significant ( $P < 0.05$ ) in spring, whereas no correlations were found in other two seasons. PO<sub>4</sub><sup>3-</sup>-P and BB showed highly significant correlations ( $P < 0.01$ ) in autumn and winter. Phytoplankton photosynthesis and bacterial respiration are the main biological factors that affects DO levels in seawater. However, DO had a significant positive correlation with PB ( $P < 0.05$ ) in winter and significant negative correlations with BB ( $P < 0.05$ ) in autumn, which showed that DO

was also regulated by other environmental factors (e.g., mixing), in addition to phytoplankton photosynthesis and bacterial respiration. BOD<sub>5</sub> had highly significant positive correlations with both PB and BB ( $P < 0.01$ ) in winter, and the correlations were significant ( $P < 0.05$ ) in spring. There was no correlation in summer and autumn.

## 2.6 Relationship between phytoplankton and bacterioplankton biomass

Bacterial abundance can be correlated with phytoplankton growth since bacteria can utilize phytoplankton photosynthetic products (Zheng *et al.*, 1994). On the other hand, the bacteria can inhibit the growth of phytoplankton through biological competition, secreting special compounds and even causing cell lysis (Zhou *et al.*, 2001). The positive correlations between PB and BB were highly significant in Sanya Bay (Fig. 4). The annual correlation coefficient ( $R$ ) was 0.68 ( $n = 88$ ,  $P < 0.01$ ), and the  $R$  value in winter, spring, summer and autumn was 0.90, 0.90, 0.71 and 0.67 ( $n = 22$ ,  $P < 0.01$ ), respectively. They are consistent with the results obtained from Chesapeake Bay, USA (Shiah and Ducklow, 1994) and from the subarctic Pacific Ocean (Kirchman *et al.*, 1993), where processes were determined to be regulating mechanisms for bacterioplankton. Bai *et al.* (2003) also reported a strong relationship between phytoplankton and bacterioplankton in the Bohai Sea, and further considered the fact that the dissolved organic carbon (DOC) and particulate organic carbon (POC) were produced by phytoplankton and were the main sources of nutrition for bacterioplankton. Therefore, it can be concluded that the primary production of Sanya Bay was likely an important factor which influenced the distribution of bacterioplankton.

Generally, the ratio of BB/PB is determined to compare the primary production and the bacterial secondary production, which reflects the importance of bacterioplankton in the marine biogeochemical cycling and the microbial food loop (Cherrier *et al.*, 1996). The average BB/PB ratios of Sanya Bay were 0.14, 0.14, 0.15 and 0.06 in winter, spring, summer and autumn, respectively, and the annual average was 0.12. The BB/PB ratios in Sanya Bay were far below those in the East China Sea (1.27) and the Yellow

Sea (0.97) (Zhao *et al.*, 2003), and the Yellow Sea Cold Water Mass (0.85) (Li *et al.*, 2006). This is mainly caused by the shallow water depth and the high transparency of the seawater. The entire water column was almost in the euphotic zone. However, in the East China Sea and the Yellow Sea, phytoplankton decreased with depth in the euphotic zone, meanwhile and the predators of bacteria and bacteriophage in the coastal are higher than in the offshore areas (Suttle and Chen, 1992). The composition of phytoplankton and the dominant species may change with the changing of the seasons under different environmental factors, such as nutrients, sunlight, and temperature. This would, in turn, lead to certain differences between the ratios of PB and chl-*a* (C/chl-*a*). Zhang *et al.* (2001) has found that, in general, the C/chl-*a* ratios of phytoplankton ranged from 10 to 150. In this study, we used the empirical conversion factor of 50 to roughly estimate PB. In order to exactly calculate the value of PB, the C/chl-*a* ratio should be determined move directly by estimating the carbon content for the dominant species in future research.

## 3 Conclusions

In summary, Sanya Bay was rich in phytoplankton species diversity. In all, 168 species, including 128 species of diatoms, were identified. Phytoplankton species and diversity index ( $H'$ ) in the tropical Sanya Bay were much higher than those in the sub-tropical and the temperate bay in China.

This study systematically synthesized the possible mechanisms affecting phytoplankton and bacterioplankton biomass in Sanya Bay, Hainan Islands. Our results demonstrated that DIN played a major role in determining the spatial and seasonal patterns of PB and BB. Temperature ( $> 22^\circ\text{C}$  all year round) had a negligible impact on BB. Highly significant correlations between PB and BB were found, which indicated that algal organic substrate sources were important for bacterial growth in Sanya Bay waters. The photosynthetic exudation coupled with protozoan egestion and cell lysis provided a plausible basis for the particular linkage of phytoplankton and bacterioplankton (Ning *et al.*, 2005). This inference, arising from this study, should be tested in future by direct experimentation.

## Acknowledgments

This study was part of the Young Scientist Fund of NSFC (No. 40806050), Knowledge Innovation Program of CAS (No. SQ200803) and continually supported by Special Basic Research Funds (No. 2008FY110100), the Open Research Program Fund of the LMEB, SOA (No. 200806), SKLOG, Institute of Geochemistry, CAS (No. OGL200605), MEL, Xiamen University (No. MEL0502). Authors would like to thank Mr. Hu Youmu for his help with part of the field and laboratory work. We also would like to thank Dr. Lin Jianping (University of Melbourne, Australia) and Prof. Paul J Harrison (AMCE Program, Hong Kong University of Science and Technology) for their valuable comments and appreciable efforts in improving English text.

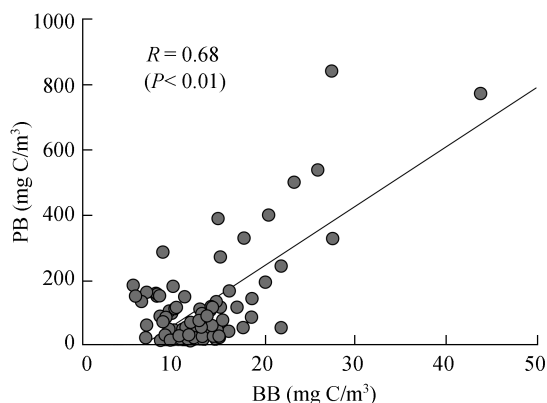


Fig. 4 Relationship between and PB and BB in Sanya Bay.

## References

- Bai J, Li K R, Li Z Y, Sun J, Wei H, Liu S M, Wu Z M, 2003. Relationship between the environmental factors and distribution of bacterioplankton in the Bohai Sea. *Journal of Ocean University of Qingdao*, 33(6): 841–846.
- Chen Z Q, Zhang H S, Liu X Y, 1999. Distributions of the dissolved trace metals (Cu, Pb, Zn, Cd and Cr) in Sanya and Yulin Bay, Hainan. *Marine Environmental Science*, 18(2): 31–37.
- Cherrier J, Bauer J E, Druffel E R M, 1996. Utilization and turnover of labile dissolved organic matter by bacterial heterotrophs in eastern North Pacific surface waters. *Marine Ecology Progress Series*, 139: 267–279.
- China State Bureau of Technical Supervision, 1991a. The Specification for Oceanographic survey: Marine Biology Survey (GB12763.6-91). 17–22.
- China State Bureau of Technical Supervision, 1991b. The Specification for Oceanographic survey: Observations of Chemical Parameters in Sea Water (GB12763.4-91). 1–20.
- Ducklow H W, 1993. Bacterioplankton distribution and production in the northwestern Indian Ocean and gulf of Oman, September 1986. *Deep-Sea Research*, 40: 753–771.
- Dong J D, Wang H K, Zhang S, Huang L M, 2002. Vertical distribution characteristic of seawater temperature and DIN in Sanya Bay. *Journal of Tropical Oceanography*, 21(1): 40–47.
- Fuhrman J A, Azam F, 1980. Bacterioplankton secondary production estimates for coastal waters of British Columbia, Antarctica, and California. *Applied and Environmental Microbiology*, 39(6): 1085–1095.
- Fukami K, Muriyuki N, Morio Y, Nishijima T, 1996. Distribution of heterotrophic nanoflagellates and their importance as the bacterial consumer in a eutrophic coastal seawater. *Journal of Oceanography*, 52: 399–407.
- Häder D P, Kumar H D, Smith R C, Worrest R C, 1998. Effects on aquatic ecosystems. *Journal of Photochemistry and photobiology B: Biology*, 46(1): 53–68.
- Harris G P, 1986. Picoplankton Ecology: Structure, Function and Fluctuations. London: Chapman and Hall Press. 221–235.
- He X Q, Wen W Y, He Q X, 2000. Evaluate on the water quality of the Sanya Bay waters. *Research and development of the South China Sea*, 2: 18–21.
- Huang L M, Wang H K, Dong J D, Zhang S, 2001. Research status on marine ecological and biological resources in the recent years. *Research and development of the South China Sea*, 2: 52–54.
- Huang L M, Tan Y H, Song X Y, Huang X P, Wang H K, Zhang S et al., 2003. The status of the ecological environment and a proposed protection strategy in Sanya Bay, Hainan Island, China. *Marine Pollution Bulletin*, 47: 180–186.
- Kirchman D L, Keil G K, Simon M, Welschmeyer N A, 1993. Biomass and production of heterotrophic bacterioplankton in the oceanic subarctic Pacific. *Deep-Sea Research*, 40: 967–988.
- Krempin D W, Sullivan C W, 1981. The seasonal abundance, vertical distribution, and relative microbial biomass of chroococcoid cyanobacteria at a station in southern California coastal waters. *Canadian Journal of Microbiology*, 27: 1341–1344.
- Lee S, Fuhrman J A, 1987. Relationships between biovolume and biomass of naturally derived marine bacterioplankton. *Applied and Environmental Microbiology*, 53(6): 1298–1303.
- Li G G, Fan Z G, 2004. *Marine Ecology*. Beijing: High Education Press. 247–282.
- Li H B, Xiao T, Ding T, Lu R H, 2006. The distribution of bacterioplankton in the Yellow Sea Cold Water Mass. *Acta Ecologica Sinica*, 26(4): 1012–1020.
- Li Y, Li R X, Wang Z L, Zhu M Y, Sun P X, Xia B, 2005. A preliminary study on phytoplankton community structure and its changes in the Jiazhou Bay. *Advances in Marine Science*, 23(3): 328–334.
- Ning X R, Chen J Z, Liu Z L, 1999. Distributions of chlorophyll a concentration and abundance of total bacteria and coliform in the waters of Sanya Bay and Yulin Bay, Hainan. *Donghai Marine Science*, 17(4): 51–54.
- Ning X R, Li W K W, Cai Y M, Shi J X, 2005. Comparative analysis of bacterioplankton and phytoplankton in three ecological provinces of the northern South China Sea. *Marine Ecology Progress Series*, 293: 17–28.
- Parsons T R, Maita Y, Lalli C M, 1984. *A Manual of Chemical and Biological Methods for Seawater Analysis*. New York: Pergamon Press. 101–112.
- Pielou E C, 1966. Species-diversity and pattern-diversity in the study of ecological succession. *Journal of Theoretical Biology*, 10: 370–383.
- Shannon C E, Wiener W, 1949. *The Mathematical Theory of Communication*. Urbana: University of Illinois Press. 125.
- Shiah F K, Ducklow H W, 1994. Temperature regulation of heterotrophic bacterioplankton abundance, production and specific growth rate in Chesapeake Bay. *Limnology and Oceanography*, 39(6): 1243–1258.
- Sun J, Liu D Y, Wang Z L, Zhu M Y, 2004. The effects of zooplankton grazing on the development of red tides. *Acta Ecologica Sinica*, 24(7): 1514–1522.
- Suttle C A, Chen F, 1992. Mechanisms and rates of decay of marine viruses in seawater. *Applied and Environmental Microbiology*, 58: 3721–3729.
- Tan Y H, Huang L M, Dong J D, Wang H K, Song X Y, 2004. Species composition of copepods and feeding impact on phytoplankton. *Journal of Tropical Oceanography*, 23(5): 17–24.
- Velji M I, Albright L J, 1986. Microscopic enumeration of attached marine bacteria of seawater, marine sediment, fecal matter, and kelp blade samples following pyrophosphate and ultrasound treatments. *Canadian Journal of Microbiology*, 32: 121–126.
- Wang H K, Dong J D, Zhang S, Huang L M, 2002. Distribution of N/P ratio and its limitation to growth of phytoplankton in Sanya Bay. *Journal of Tropical Oceanography*, 21(1): 33–39.
- Wang H K, Dong J D, Zhang S, Huang L M, 2003. A study on release of phosphorus from sediments in Sanya Bay. *Journal of Tropical Oceanography*, 22(3): 1–8.
- Wang Y S, Wang Z D, Huang L M, 2004. Environment changes and trends in Daya Bay in recent 20 years. *Journal of Tropical Oceanography*, 23(5): 85–95.
- Yin J Q, Zhang G X, Huang L M, Li K Z, 2004a. Diel vertical migration of zooplankton in Sanya Bay, Hainan Province, China. *Journal of Tropical Oceanography*, 23(5): 25–33.
- Yin J Q, Zhang G X, Tan Y H, Huang L M, Li K Z, 2004b. Species composition and quantitative distribution of zooplankton in Sanya Bay, Hainan Province, China. *Journal of Tropical Oceanography*, 23(5): 1–9.
- Zhang W C, Zhang F, Wang K, 2001. The ratio of the phytoplankton carbon and chlorophyll in the sea. *Marine Sciences*, 25(3): 28–29.



- Zhao S J, Xiao T, Yue H D, 2003. The distribution of heterotrophic bacteria and some related factors in the East China and Yellow Seas during fall. *Oceanologia et Limnologia Sinica*, 34(3): 295–305.
- Zheng T L, Cai L Z, 1993. The role of marine bacteria in upwelling ecosystem. *Acta Oceanologica Sinica*, 12(2): 261–267.
- Zheng T L, Wang F, Xu M Z, Hong H S, 2002. Bacterial production, biomass and role in microbial loop in Taiwan Strait. *Oceanologia et Limnologia Sinica*, 33(4): 415–423.
- Zheng T L, Wang H L, Hong H S, 1994. Role of microorganisms in biogeochemical cycling of carbon in oceans. *Chinese Journal of Ecology*, 13(4): 47–50.
- Zhou W H, Yuan X C, Huo W Y, Yin K D, 2004. Distribution of chlorophyll a and primary productivity in the adjacent sea area of Changjiang River Estuary. *Acta Oceanologica Sinica*, 26(3): 143–150.
- Zhou Y H, Pan J M, Ye Y, Zhou H Y, 2001. Relationship between bacterium, virus and phytoplankton and their effects to geochemical cycling in ocean. *Journal of Oceanography in Taiwan Strait*, 20(3): 340–345.