Dynamics of dissolved nutrients in the aquaculture shrimp ponds of the Min River estuary, China: Concentrations, fluxes and environmental loads

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GRAPHICAL ABSTRACT
HIGHLIGHTS

- NH$_4^+$-N was the predominant species of dissolved inorganic nitrogen in shrimp ponds.
- Dissolved inorganic nutrients varied greatly among different shrimp growth stages.
- Aquaculture pond effluent is a key contributor to China’s coastal water pollution.
Dynamics of dissolved nutrients in the aquaculture shrimp ponds of the Min River estuary, China: Concentrations, fluxes and environmental loads

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ABSTRACT

Dissolved inorganic nutrients (NO₂⁻-N, NO₃⁻-N, NH₄⁺-N, and PO₄³⁻-P) play a critical role in the effective management of water quality and prevention of fish and shrimp diseases in aquaculture systems. In this study, dissolved inorganic nutrient concentrations in the water column and sediment porewater and the fluxes across the sediment-water interface (SWI) were investigated in three intensive shrimp ponds with zero water exchange to examine nutrient cycling during the different growth stages of shrimps. We found distinct changes in the dissolved inorganic nutrient concentrations in both the water column and sediment porewater among the three growth stages. Average NO₂⁻-N, NO₃⁻-N, NH₄⁺-N, and PO₄³⁻-P concentrations in the sediment porewater were 3.53, 2.81, 29.68, and 6.44 times higher, respectively, than those in the water column over the study period, indicating that the pond sediment acted as a net source of nutrients to the water column. This was further supported by the net release of nutrients from the sediments to the water column observed during the incubation experiment. Nutrient fluxes were dominated by NH₄⁺-N, while NOₓ⁻-N (NO₂⁻-N and NO₃⁻-N) and PO₄³⁻-P fluxes remained low. The high rates of NH₄⁺-N release from the sediment highlight the need of taking into account the biogeochemical role of sediments in mitigating the problem of water quality degradation in coastal shrimp ponds. Based on a total water surface area of mariculture ponds and a total mariculture production of 2.57×10⁶ ha and 2.30×10⁹ kg, respectively, we estimated conservatively that approximately 4.77×10⁴ ton of total nitrogen and 3.75×10³ ton of total phosphorus are being discharged annually from the mariculture ponds into the adjacent coastal zones across China. Our results demonstrated the importance of aquaculture pond effluent as a major contributor of water pollution in the coastal areas of China, and
called for actions to properly treat these effluents in alleviating the eutrophication problem in the Chinese coastal zones.

**Keywords:** Porewater; Nutrient dynamics; Sediment-water interface; Prawn culture; Eutrophication; Estuary
1. Introduction

Global aquaculture production has increased dramatically over the past 50 years, with an average annual increase rate of 8.3% during the period of 1970–2008, to meet the rising demand around the world for protein (FAO, 2010). Intensive shrimp aquaculture (FAO, 2016), in which shrimps are raised at very high densities in closed or semi-closed systems with constant supply of oxygen, water, and feeds, is seen as an important component in sustaining a steady aquaculture production because of its short production cycles and high product values (Silva et al., 2013; Molnar et al., 2013). According to data from the Food and Agriculture Organization (FAO), global food shrimp (prawn) culture reached a total annual production of about 2.1 million tonnes in 2015 (FAO, 2016). Although intensive aquaculture has been very effective in responding to the ever-growing global demand for aquaculture food, it has also been linked to serious environmental problems.

One of the key environmental concerns regarding intensive aquaculture is the accumulation of nutrients (especially inorganic nitrogen and phosphorus), which can cause water quality problems within ponds (Hargreaves and Tucker, 2004; Castillo-Soriano et al., 2013; Hu et al., 2014) and subsequently shrimp diseases. In general, intensive aquaculture shrimp ponds are maintained through daily supply of feeds. However, only a small proportion of these nutrient inputs are being converted into shrimp biomass, as the feed utilization efficiency is only about 4.0–27.4% (Su et al., 2009; Chen et al., 2016). Consequently, the majority of the nutrients in the residual feeds are retained in the pond water. Once the accumulation of nutrients in pond water exceeds the tolerance threshold, adverse effects in the form of harmful algae blooms and water quality deterioration could be seen in the aquaculture ponds (Huang et al., 2016; Yang et al., 2017a). More importantly, high concentrations of nutrients
(especially ammonia and nitrite) in the water column can stimulate the release of corticosteroid hormones into the venous circulation of shrimps (Hu et al., 2014), which may be hazardous to shrimp health and thus cause a reduction in shrimp productivity. Understanding the nutrient dynamics of intensive aquaculture ponds therefore is critical for proper pond management and improvement of shrimp yield.

Another key environmental concern regarding intensive aquaculture is the discharge of pond effluents into the water bodies of the nearby coastal zones. In general, the water column is the main habitat for animals in aquaculture ponds, and its conditions are closely associated with the healthy growth of fish, shrimps, and other organisms (Yang et al., 2017b). Complete drainage of pond water is typically done at the end of each aquaculture production cycle, in order to aerate the bottom soils and discard the water polluted with nutrients and wastes in preparation of the next round of aquaculture production (Wang and Wang, 2007; Wu et al., 2014). During this process, large quantities of water enriched with nutrients are discharged into the adjacent ecosystem over a very short period of around a month (Wu et al., 2014). Such practice can rapidly alter the nutrient levels and quality of nearby waters, and thus create eutrophication problems in the coastal ecosystems. Impacts of the discharge of aquaculture effluents on the water quality in coastal creeks (Wolanski et al., 2000; Burford et al., 2003; Costanzo et al., 2004) and mangrove swamps (Molnar et al., 2013; Cardoso-Mohedano et al., 2016a, 2016b) have already received great attention. However, very few studies have investigated the effects of aquaculture pond effluents on the trophic status of receiving coastal waters (Herbeck et al., 2013).

In the Asia-Pacific region where approximately 90% of the world’s aquaculture production takes place, land-based aquaculture pond culture is the most important method of shrimp production in freshwater and brackish water systems (FAO, 2010).
China has the world’s largest mariculture industry, with a total mariculture pond area and total aquaculture production of $2.57 \times 10^6$ ha and $2.30 \times 10^9$ kg, respectively, in 2015 (Chen et al., 2016). In view of this, studying the nutrient dynamics of aquaculture ponds in China is essential for promoting the sustainable development of the aquaculture industry and assessing the likely risks of eutrophication of coastal water bodies. The main objectives of this study were to: (1) investigate the temporal variability of dissolved inorganic nutrients in the water column and sediment porewater of aquaculture shrimp ponds in China at different growth stages of shrimps, (2) assess the nutrient fluxes across the sediment-water interface (SWI) of these ponds, and (3) evaluate the impact of effluent discharge from these aquaculture ponds on the trophic status of receiving coastal waters.

2. Materials and Methods

2.1. Study area description

The study site is located in Shanyutan wetland (ca. 3120 ha) of the Min River estuary in southeast China (Fig. 1). The area has a subtropical monsoon climate, which is relatively warm and wet, with a mean annual temperature of 19.6 °C and a mean annual precipitation of about 1,350 mm (Tong et al., 2010). Shrimp ponds are a dominant landscape feature in the Min River estuary (Yang et al., 2015). The total area of shrimp ponds in the Shanyutan wetland is about 234 ha. These ponds were converted in 2011 by complete removal of all marsh vegetations. Aquaculture production in the majority of the shrimp ponds occurs between June and November.

2.2. Shrimp pond system and management

As the optimal water temperatures for the growth of shrimps (*Litopenaeus vannamei*) are 22–35°C, only one crop of shrimps can be produced per year in our study site. Prior to shrimp production, the ponds were filled with estuarine water from the
Min River estuary using a submerged pump. The water first passed through a 2 mm mesh bag in order to prevent the entry of predators and competitors (Guerrero-Galván et al., 1999). Additional input of freshwater into the ponds took place occasionally during rainfall events. No water was discharged from the spillways of the ponds until the end of shrimp harvesting. The water depth in the shrimp ponds over the culture period ranged between 1.1 and 1.5 m, with a mean of 1.4 m.

To assess nutrient cycling during the culture period, water and sediment samples were collected from three commercial shrimp ponds in the Shanyutan wetland (26°01′49″ N, 119°37′39″ E) of the estuary (Fig. 1). The basic details of the three ponds are shown in Table 1. Before stocking, each shrimp pond was fertilized with 37.5 kg ha⁻¹ of urea (46% N) and 5.2 kg ha⁻¹ of phosphate fertilizer (38% P₂O₅). Postlarval (PL) Litopenaeus vannamei shrimps of approximately 0.9–1.2 cm in length were successively stocked in the ponds from mid- to late-May, and feeding was initiated simultaneously. The shrimp production cycle began on May 15, and lasted for about 163 days. L. vannamei were fed with artificial feeds containing 42% of crude protein (Yuehai™, Guangzhou, China) twice per day at 07:00 and 16:00 (local standard time), respectively, by direct application from a small boat. Based on the management practices (e.g. feeding rate, water depth, etc.), water salinity, and shrimp weight, the shrimp grow-out cycle was divided into three different stages (Table 2), which was similar to the classification scheme adopted by Páez-Osuna et al. (1997). The feeding rate was maintained at approximately 10–16, 50–55, and 40–45 kg ha⁻¹ d⁻¹ during the initial, intermediate, and final stages, respectively. The exact amount of feed added was determined based on the response of shrimps to the previous feeding (Casillas-Hernández et al., 2006; Liu et al., 2015). On average, a total of 3.5 ton of feeds were applied to each of the three ponds over the culture period (personal
communications). At each pond, three 1500-W paddlewheel aerators were activated
four times a day between 07:00–09:00, 12:00–14:00, 18:00–20:00, and 00:00–03:00
(local standard time). Pond water was completely drained and surface sediment (0–10
cm) was removed after shrimp harvest.

2.3. Collection and analysis of water samples

Water sample were collected in the middle of June, August, and October of 2015
that represented the conditions during the initial, middle, and final stages of shrimp
production, respectively.

2.3.1. Collection and analysis of nutrient concentrations in the water column

Three replicate sampling sites were established in each pond for water sampling.
Water samples were collected at three different depths of the water column, including
the bottom layer at approximately 5 cm above the soil, the surface layer at
approximately 10 cm below the water surface, and the middle layer at approximately
70–85 cm below the water surface. Water samples from each of the three depths were
collected four times at 08:00, 11:00, 14:00, and 17:00 (local time) on each sampling day
with a 5 L Niskin bottle, and then transferred to 250 mL polyethylene bottles.
Approximately 0.2 mL of saturated HgCl₂ solution was injected into each bottle to
inhibit microbial activity (Zhang et al., 2013). The water samples were subsequently
stored in a 4 °C cooler, transport to the laboratory, and analysed within one week. For
the analysis of dissolved inorganic nutrients, the water samples were filtered through a
0.45 μm cellulose acetate filter (Biotrans™ nylon membranes), and the filtrates were
analysed for the concentrations of dissolved inorganic nutrients using a flow injection
analyser (Skalar Analytical SAN⁺⁺, Netherlands). NO₃⁻-N was calculated as the
difference between NOₓ⁻-N and NO₂⁻-N, while dissolved inorganic nitrogen (DIN) was
determined by summing up the concentrations of NO₃⁻-N, NO₂⁻-N, and NH₄⁺-N
The detection limits for NO$_2^-$-N, NO$_X^-$-N, NH$_4^+$-N, and PO$_4^{3-}$-P were 0.02, 0.1, 0.1, and 0.05 μmol L$^{-1}$, respectively. The relative standard deviations of all analyses were in the range of 0.1-4.0%.

2.3.2. Physicochemical and biological parameters of the water column

In the field, physicochemical variables including water temperature, pH, and salinity were determined via a pH/mV/Temp system (IQ150, IQ Scientific Instruments, USA) and a salinity meter (Eutech Instruments-Salt6, USA). The dissolved oxygen (DO) level at various depths of the water column was measured in situ using a multi-parameter controller (HORIBA, Japan). All these instruments were calibrated in the laboratory every time before actual field measurement according to the instruction manuals. Chlorophyll $a$ (Chl $a$), which served as a proxy of phytoplankton biomass (Lacoste and Gaertner-Mazouni, 2016), was determined by filtering 250 mL of water samples through GF/F glass microfibre filters. Following the extraction of filters at 4°C in 90% (V/V) acetone for 24 h and the centrifugation of the mixture at 4500 rpm for 15 min, the absorbance of the supernatant at 665 and 750 nm was measured using a UV-visible spectrophotometer (Shimadzu UV-2450, Japan) (Zhang et al., 2016; Yang et al., 2017b). The relative standard deviations of this analysis were in the range of 5-8%. At each growth stage, 50 shrimps were captured from each sampling location using a small bow net. The shrimps were then weighed individually with a portable electronic scale up to a precision of 0.01 g for determining the shrimp biomass (g PL$^{-1}$) at each stage.

2.4. Collection and analysis of sediment samples

On each sampling day, nine intact sediment cores were collected in each pond by means of 30 cm long Plexiglas tubes (internal diameter of 6.0 cm) that were inserted into a surface-operated coring device (Core-60, Austria) equipped with a core cylinder
and a one-way check valve to preserve the sediment and the overlying water integrity (Han et al., 2014). The intact cores consisted of 15 cm of sediment overlaid by 15 cm of water column. These sediment cores were sealed and stored vertically in a 4 °C cooler and transported to the laboratory within 4 h.

2.4.1. Physicochemical parameters of the sediment

Triplicate sediment samples (0–15 cm) from each pond were freeze-dried, homogenized, and then grounded to a fine powder for the analyses of total nitrogen (TN), pH, grain composition, and porosity (Zhou et al., 2016). Sediment TN was analysed with a CHN elemental analyser (Elementar Vario MAX CN, Germany) on oven-dried and grounded subsamples that passed through a 2 mm sieve. Sediment pH was determined using a pH meter (Orion 868, USA) with a soil-to-water ratio of 1:2.5. In addition, sediment porosity ($\Phi$) was estimated based on the water content in surface sediment samples (from a subsample taken before freeze drying) that was determined by weight differences after drying in an oven at 105 °C for 24 h (Zhang et al., 2013).

2.4.2. Collection and analysis of nutrient in the sediment porewater

Triplicate sediment samples (0–15 cm) from each pond were used to determine the porewater nutrient concentrations. Sediment porewater was collected following the methods described by De Vittor et al. (2012) and Zhang et al. (2013). Briefly, upon return to the laboratory, the sediment cores were extruded and then sliced at 3 cm intervals. Porewater from each depth interval was extracted from the sediment by centrifugation (5000 rpm, 10 min, Cence® L550) at in situ temperature and then filtered (0.45 μm acetate fibre membranes) (De Vittor et al., 2012). The filtered porewater samples were collected in vials pre-cleaned with acid, and 0.2 mL of saturated HgCl$_2$ solution was injected into each vial to inhibit microbial activity (Zhang et al., 2013). To prevent oxidation during handling, all porewater samples were
processed in a nitrogen-filled glove bag (Matos et al., 2016). All filtered porewater samples were stored at 4 °C until analysis. Concentrations of NO$_2^-$-N, NO$_3^-$-N, NH$_4^+$-N, and PO$_4^{3-}$-P in the filtered porewater were analysed using a flow injection analyser (Skalar Analytical SAN++, Netherlands).

2.4.3. Nutrient fluxes across the SWI

Triplicate sediment samples (0–15 cm) from each pond were used in an incubation experiment to study nutrient fluxes across the SWI. The incubation device (Fig. 2) was constructed according to Cowan et al. (1996) and Chen et al. (2014). Approximately 15 cm-long sediment cores were retrieved, along with 15 cm deep of overlying water. Bottom water samples at each site were also taken and stored under the same conditions as the incubated cores. The sediment cores were covered using a Teflon plunger with inlet and outlet tubes, and sealed to the core cylinder with an O-ring. The overlying water above the sediments was bubbled with air to simulate the *in situ* bottom water O$_2$ levels (Mu et al., 2017). Thereafter, the cores were incubated in a temperature-regulated incubator device (QHZ-98A, China) for a period of 9 h. The incubated temperature was chosen to be the same as the field *in situ* temperature. Based on previous researches (Yang et al., 2017a) and the pilot experiments of this study, we selected shaking velocities of the incubator to be 20, 40, and 80 rpm, to simulate the intensity of shrimp disturbance during the initial, middle, and final stages, respectively. Sampling of overlying water was performed using a plastic syringe at the beginning and the end of the 9 h incubation period. During each sampling, 60 mL of overlying water was taken and then replaced by 60 mL of overlying water collected *in situ* in the field. Water samples were filtered through 0.45 μm acetate fibre membranes immediately and the nutrient concentrations were measured with a flow injection analyser (Skalar...
Analytical SAN++, Netherlands). Fluxes of nutrients across the SWI were quantified according to the following equation (Cheng et al., 2015):

\[ F = (C_{W,E} - C_{W,B}) \times V / S / T \]

where \( F \) (mg m\(^{-2}\) h\(^{-1}\)) is the flux of nutrient at the SWI (positive and negative values indicate a net release from, and a net uptake by the sediment, respectively); \( C_{W,E} \) and \( C_{W,B} \) are the nutrient concentration (mg L\(^{-1}\)) in the overlying water at the end and the beginning of incubation, respectively; \( V \) is the volume of overlying water (L); \( S \) is the cross-sectional area of the sediment column (m\(^2\)); and \( T \) is the incubation time (h).

### 2.5. Collection and analysis of adjacent coastal waters of the study area

In order to evaluate the impact of shrimp pond effluent on the water quality of receiving coastal waters, surface water samples (10 cm depth) were collected from adjacent coastal waters of the study area (Fig. 1) both before and after the discharge of shrimp pond effluents upon the completion of shrimp harvesting. Sampling was carried out between 09:00 and 10:30 by boat in sequence from site 1 to site 6 (Fig. 1). At each sampling site, one water sample at a depth of 10 cm was collected by a 5 L Niskin bottle. Water samples were then stored in 500 ml polyethylene bottles kept at 4 °C, transported to the laboratory, and analysed within 72 h. Water samples were filtered and analyzed for dissolved inorganic nutrients following the same method as previously described. The unfiltered seawater samples were also analyzed for the concentrations of total nitrogen (TN) and total phosphorus (TP) by the peroxydisulfate oxidation method (Ebina et al., 1983), with the NO\(_3\)-N and PO\(_4^{3-}\)-P concentrations in the digestate being determined by ion chromatograph (Dionex 2100, American) (Han et al., 2014). Detection limits for TN and TP were 0.7 and 0.3 \(\mu\)mol L\(^{-1}\), respectively. The relative standard deviations of these analyses were in the range 2-5%.

### 2.5. Statistical analyses
The calculations of basic statistical parameters (e.g. mean, standard error (SE), etc.) were carried out using Microsoft Excel 2003. Differences in the studied variables among the three growth stages of shrimps were assessed by analysis of variance (ANOVA). Pearson correlation analysis was conducted to examine the relationships between nutrient fluxes/levels and the environmental variables. All data were reported as mean ± 1 SE. ANOVA and correlation analysis were carried out using SPSS (Version 17.0) with the significance level set at 0.05.

3. Results and discussions

3.1. Physicochemical and biological properties of the shrimp ponds

The measured physicochemical variables and shrimp biomass in the three shrimp ponds are presented in Table 3. The water and sediment temperatures over the study period ranged between 24.02 and 30.95 °C, and 22.49 and 28.16 °C, respectively, with significantly higher temperatures during the middle stage (Table 3). Water pH varied significantly among the three growth stages (p<0.05), with considerably lower values during the middle stage (Table 3). Both sediment TN content and porosity were significantly higher during the middle stage than the other two stages, which was similar to the temperature trend (Table 3). Water DO level and shrimp biomass also varied significantly (p<0.05) among the growth stages, with the highest and lowest values during the final and initial stages, respectively (Table 3).

The results of Chl a concentrations in the water column are presented in Fig. 3. Average Chl a concentrations ranged between 130.1±2.5 and 258.3±3.1 μg L⁻¹ during the study period. Significant temporal variations in Chl a were observed, with considerably lower concentrations observed during the initial stage than the other two stages (Fig. 3). However, no significant difference in Chl a concentration was seen among the three water depths (p>0.05; Fig. 3).
3.2. Nutrient concentrations in the water column

The results of dissolved inorganic nutrients in the water column are shown in Fig. 4. The NO$_2^-$-N, NO$_3^-$-N, and NH$_4^+$-N concentrations during the study period ranged between 7.18 and 49.75 µg L$^{-1}$, 15.66 and 81.02 µg L$^{-1}$, and 0.10 and 0.81 mg L$^{-1}$, respectively, with mean values of 21.45±1.47 µg L$^{-1}$, 56.47±2.24 µg L$^{-1}$, and 0.44±0.02 mg L$^{-1}$, respectively. The mean concentration of NH$_4^+$-N was significantly higher than that of NO$_2^-$-N and NO$_3^-$-N ($p<0.05$), which showed that NH$_4^+$-N was the predominant species of DIN in the water column of shrimp ponds. Our findings were inconsistent with those of Silva et al. (2013) and Li et al. (2014), in which NO$_3^-$-N was the dominant form of DIN in a super-intensive biofloc technology culture system and the semi-intensive shrimp ponds, respectively. The high NH$_4^+$-N concentrations in the water column observed in this study might be attributed to the high rates of NH$_4^+$-N release from the shrimp pond sediment.

Significant temporal variations in NO$_x^-$-N and NH$_4^+$-N concentrations in the water column of shrimp ponds were observed over the study period (Fig. 4). The NO$_x^-$-N concentrations in the pond water showed an increasing trend over time (Fig. 4a, b). A plausible reason for this could be due to the continuous input of protein-rich feeds that had stimulated the degradation of organic matter and nitrification, thereby favouring the accumulation of NO$_x^-$-N in the water column. In contrast to NO$_x^-$-N, we found lower NH$_4^+$-N concentrations during the final stage but higher concentrations during the middle stage (Fig. 4c). These temporal patterns were different from those seen in other results in which NH$_4^+$-N concentrations in aquaculture systems generally increased over time in response to an increase in shrimp excretion rates and the accumulation of residual feeds (Yu et al., 2013; Castillo-Soriano et al., 2013). In our study, the significantly higher water temperature (Table 3) during the middle stage
could have stimulated microbial mineralization of organic matter (Hopkinson et al., 2001; Zhang et al., 2013) and hence the release of NH$_4^+$-N from the sediment into the overlying water. On the other hand, the significantly lower NH$_4^+$-N concentrations detected during the final stage might, to some extent, be explained by a combination of low water temperature (Table 3) and high consumption rate of NH$_4^+$-N by phytoplankton (as represented by Chl a) (Fig. 3).

Dissolved PO$_4^{3-}$-P concentrations in the water column also differed significantly among the three stages, ranging between 36.12 and 99.55 µg L$^{-1}$ (Fig. 4d). In contrast to DIN, the PO$_4^{3-}$-P concentrations in pond water showed a decreasing trend over the study period (Fig. 3d). Some previous researches have indicated that, P assimilation in aquaculture production system is tightly linked to biomass growth (Montoya et al., 2000; Mai et al., 2010). Furthermore, Hu et al. (2015) found that the high phytoplankton amount accounted for the low PO$_4^{3-}$-P concentrations in the coastal upwelling ecosystem of the southern Taiwan Strait. Hence, the decrease in PO$_4^{3-}$-P concentrations in the water column of our shrimp ponds over time (Fig. 4d) was probably associated with an increase in the assimilation of P by shrimps and phytoplankton. This was further supported by the observation that both shrimp biomass and Chl a were negatively correlated with PO$_4^{3-}$-P concentrations in the water column over time (Table 3, Figs. 2 and 3d).

3.3. Nutrient concentrations in the sediment porewater

The results of dissolved inorganic nutrients in the sediment porewater of the shrimp ponds are presented in Fig. 5. The NO$_2^-$-N, NO$_3^-$-N, NH$_4^+$-N, and PO$_4^{3-}$-P concentrations in the sediment porewater over the study period ranged between 36.57 and 108.10 µg L$^{-1}$, 30.00 and 384.10 µg L$^{-1}$, 4.82 and 24.95 mg L$^{-1}$, and 0.28 and 0.88 mg L$^{-1}$, respectively, with mean values of 75.66±5.02 µg L$^{-1}$,158.41±21.15 µg L$^{-1}$,
13.01±1.02 mg L\(^{-1}\), and 0.44±0.03 mg L\(^{-1}\), respectively. Porewater DIN was dominated by NH\(_4^+\)-N as indicated by the low NO\(_x\)-N and high NH\(_4^+\)-N concentrations (Fig. 5), which was similar to the case of the water column. This could be attributed to the highly reducting environment in the sediment that favoured the presence of NH\(_4^+\)-N over the oxidized forms of nitrogen (Zhang et al., 2013).

A significant difference in dissolved inorganic nutrient concentrations was recorded across the three shrimp growth stages during the study period (Fig. 5). The concentrations of NO\(_x\)-N and PO\(_4^{3-}\)-P showed similar temporal patterns, with lower and higher values during the initial and final stages, respectively (Fig. 5). The nutrients in porewater are typically assumed to be derived from the degradation of organic matter (Lee et al., 2008; Zhang et al., 2013). A large supply of organic matter may ensure a steady supply of substrates for microbial mineralization of soil N and P (Kristensen et al., 2008), leading to an overall increase in porewater nutrient concentrations. Hence, the increase in porewater NO\(_x\)-N and PO\(_4^{3-}\)-P concentrations observed in the present study over time (Fig. 5a, 5b, and 5d) was probably associated with an increase in the accumulation of degradable organic matter in sediment (Jackson et al., 2003; Lacoste and Gaertner-Mazouni, 2016; Matos et al., 2016).

The porewater NH\(_4^+\)-N concentrations demonstrated a different temporal trend from those of NO\(_x\)-N and PO\(_4^{3-}\)-P, with the highest values being observed during the middle stage than the final stage (Fig. 5c). This temporal pattern was consistent with that of temperature and TN content in the pond sediment (\(p<0.01\); Fig. 6). These results further supported the idea that both high sediment temperature and great supply of organic matter contributed to the elevated porewater NH\(_4^+\)-N concentration found during the middle stage as compared to the initial stage. Meanwhile, at the final stage, the low porewater NH\(_4^+\)-N concentration could be attributed to the combination of low
sediment temperature and intense bioturbation, which enhanced nitrification through increasing the oxygen levels in the sediment (Henriksen et al., 1983). This hypothesis could be further supported by the higher porewater NO$_x$-N concentration observed in our ponds during the final stage (Fig. 5a and 5b).

3.4. Fluxes of nutrients across the sediment-water interface

The results of our laboratory incubation experiment on the fluxes of dissolved inorganic nutrients across the SWI are shown in Fig. 7. The NO$_x$-N fluxes were quite variable over the study period, ranging from negative values (-0.20 and -0.84 mg m$^{-2}$ h$^{-1}$ for NO$_2$-N and for NO$_3$-N, respectively) during the initial stage to positive values (0.76 and 1.57 mg m$^{-2}$ h$^{-1}$ for NO$_2$-N and NO$_3$-N, respectively) during the final stage (Fig. 7a and 7b). In contrast, the NH$_4$+-N and PO$_4^{3-}$-P fluxes across the SWI were always positive, which corresponded to net nutrient releases from the sediment to the water column (Fig. 7c and 7d). The NH$_4$+-N and PO$_4^{3-}$-P fluxes over the study period ranged between 10.17 and 140.86 mg m$^{-2}$ h$^{-1}$, and 0.25 to 14.84 mg m$^{-2}$ h$^{-1}$, respectively, with average values of 41.42±6.69 and 2.74±0.71 mg m$^{-2}$ h$^{-1}$, respectively. These results show that NH$_4$+-N was the main form of DIN being released from the sediment to the water column, while the magnitude of DIN fluxes was higher than that of PO$_4^{3-}$-P.

Fig. 7 also shows that DIN fluxes from the shrimp pond sediment varied significantly among the three stages, which was in agreement with the results reported in other aquaculture systems (e.g. Nizzoli et al., 2011; Holmer et al., 2015; Zhong et al., 2015; Yang et al., 2017a). Many of these studies suggested that sediment temperature, organic matter content, DO, and benthic organisms were the dominant factors governing the temporal variations of nutrient fluxes across the SWI. In this study, NO$_x$-N fluxes were positively correlated with shrimp biomass (Table 4). An increase
in shrimp biomass might imply a greater bioturbation of sediment by shrimps, which
could enhance the oxygen supply from the overlying water to the sediment (Henriksen
et al., 1983), and subsequently increase nitrification rates (Nicholaus and Zheng, 2014;
Zhong et al., 2015) and the release of NO$_x$-N from the pond sediment over time (Fig.
7a and 7b). This hypothesis was further supported by the strong correlation observed
between NO$_x$-N and DO concentrations (Table 4). In addition, Jiang et al. (2000) and
Zhong et al. (2015) reported that the excretion of *Litopenaeus vannamei* could enrich
the sediments and contributed to the enhancement of NO$_x$-N release from the sediment.
The positive relationship between NO$_x$-N fluxes and shrimp biomass found in this
study suggested that the temporal dynamics of NO$_x$-N could at least partly be
associated with the excretion of urea and other nitrogenous compounds from shrimps.
On the other hand, previous studies have suggested that the enhancement of NH$_4$+-N
release across the SWI could be attributed primarily to animal excretion, bioturbation,
and the transport of accumulated NH$_4$+-N in the anoxic sediment (Svensson, 1997;
Zhang et al., 2011a, b; Zhong et al., 2015). In the present study, the temporal patterns of
sediment temperature, TN content, and NH$_4$+-N fluxes in the shrimp ponds were highly
similar (Table 3 and Fig. 7c), which implied that the effects of temperature on the
mineralization rates of organic matter were more significant than that of shrimp
excretion or other environmental variables. Moreover, the temporal change in the rate
of NH$_4$+-N release across the SWI was similar to that of shrimp biomass from the initial
to the middle stage (Fig. 7c and Table 3), indicating that the influence of shrimp
excretion and bioturbation was also important in controlling the variations of NH$_4$+-N
flux over time during the initial and middle stages of shrimp growth.
PO$_4^{3-}$-P fluxes across the SWI of shrimp ponds also greatly varied among different
stages, with minimum and maximum fluxes occurred at the initial and final stages,
respectively (Fig. 7d). Similar temporal patterns were observed by Nicholaus and Zheng (2014), who found a significant increase in \( \text{PO}_4^{3-} \)-P fluxes across the SWI over time for clam aquaculture ponds. Zhong et al. (2015) also reported similar temporal variations of \( \text{PO}_4^{3-} \)-P fluxes from the aquaculture ponds of *Litopenaeus vannamei*. These studies found that bioturbation played a primary role in controlling the rate of \( \text{PO}_4^{3-} \)-P release from the aquaculture ponds. Waldbusser et al. (2004) reported that the effects of bioturbation on \( \text{PO}_4^{3-} \)-P release could be attributed to the changes in oxygen level and sediment redox potential. Nicholaus and Zheng (2014) further proposed that bioturbation could influence \( \text{PO}_4^{3-} \)-P release through altering the bacterial decomposition of biodeposits and the resuspension of sediments. In the present study, we found that \( \text{PO}_4^{3-} \)-P fluxes were significantly and positively correlated with shrimp biomass \((r^2 = 0.657, p < 0.001, n = 27)\), which pointed to the potential of shrimp bioturbation on sediment \( \text{PO}_4^{3-} \)-P fluxes. Yet, as \( \text{PO}_4^{3-} \)-P fluxes in this study were determined based on laboratory incubation experiments only, in situ measurements might be needed to quantify \( \text{PO}_4^{3-} \)-P fluxes in a field setting simulating the actual conditions, and the exact mechanisms of bioturbation effects on \( \text{PO}_4^{3-} \)-P release deserve further investigation.

3.5. Impact of biogeochemical cycling of nutrients

3.5.1. Impact of sediment nutrient release on the survival rates of shrimp

We observed the emergence of shrimp diseases in the ponds of our study area, with a mass mortality of shrimps particularly during the middle and late stages of shrimp growth. The shrimp survival rates in the three ponds ranged between 60 and 70% (personal communication), with a mean value of 65% that was significantly lower than the normal level of 80% (Lai, 2014). The low survival rates of shrimps in our ponds might be related to the high rates of sediment nutrient release. Over the study
period, we observed predominantly positive NOx^-N fluxes, and consistently positive 
NH4^+-N and PO4^{3-}-P fluxes across the SWI in the coastal shrimp ponds (Fig. 7), 
indicating that the shrimp pond sediment was generally a net source of dissolved 
inorganic nutrients. We found that the rates of nutrient (especially NH4^+-N and PO4^{3-}-P) 
release from the shrimp pond sediment were much higher than those reported by Yang 
et al. (2017a) from the estuarine and marine sediments in various aquaculture systems 
(e.g. blue crab, grouper, tuna, oysters) in China. The nutrient fluxes from our shrimp 
pond sediment were also much greater than those from the pearl oyster culture in 
Tuamotu Archipelago (Lacoste and Gaertner-Mazouni, 2016), mussel cultures (Mytilus 
edulis) in Skive Fjord of Denmark (Holmer et al., 2015), and shrimp (L. vannamei) 
ponds in Shandong Province of China (Zhong et al., 2015). In particular, the sediment 
NH4^+-N fluxes in our shrimp ponds were significant higher than the range of 
0.01-31.25 mg N m^-2 h^-1 reported for a range of freshwater, estuarine, and marine 
sediments in aquaculture ponds by Hargreaves (1998). The large release of NH4^+-N 
from sediment could lead to significant ammonia accumulation in the water column, 
which would subsequently be oxidized to nitrites (NO2^-) by nitrite bacteria under oxic 
conditions (Kou et al., 2014). Generally, the acceptable or safe levels for nitrite and 
ammonia in prawn aquaculture systems under low-salinity water conditions are under 
10 µg L^-1 and 0.20 mg L^-1, respectively (Lai, 2014). Yet, the mean NO2^-N and NH4^+-N 
concentrations from the shrimp ponds water in Min River estuary were 21.45±1.47 µg 
L^-1 and 0.44±0.02 mg L^-1, respectively, which were substantially greater than the 
acceptable levels. This could impose toxicity problems to shrimps, leading to a 
reduction in both survival and growth rates, as well as an increase in a variety of 
physiological dysfunctions of shrimps (Hu et al., 2012; Kou et al., 2014).
The low survival rates of shrimp were also probably related to the interactions of large sediment nutrient releases and harmful algal blooms. In the present study, high rates of sediment nutrient release to the overlying water might support a significant proportion of the total nutrient requirements for primary productivity in shrimp ponds, which in turn lead to the formation of harmful algal or cyanobacterial blooms. It has long been known that cyanobacterial blooms in aquaculture ponds can cause acute and massive shrimp deaths (Alonso-Rodriguez and Páez-Osuna, 2003; Ajinet et al., 2016; Gao et al., 2017). The stoichiometric ratio of N and P plays an important role in determining the dominant phytoplankton species and the potential formation of harmful algal blooms in the water column of shrimp ponds (Alonso-Rodriguez and Páez-Osuna, 2003). In the Gulf of California, Barraza-Guzmán (1994) found that a N/P ratio of 6.8 in shrimp pond waters would favor the dominance of cyanobacteria over diatoms, dinoflagellates and phytoflagellates. In the present study, the molar ratios of DIN/PO$_4^{3-}$-P of our shrimp ponds water were estimated to be 4.49±0.79, 11.30±2.11, and 6.43±1.13 during the initial, middle, and final stages, respectively. The overall mean N/P ratio in this study was 7.41±0.98, which was close to that reported by Barraza-Guzmán (1994), which implied that cyanobacteria was dominant in our study ponds that caused an adverse effect on shrimp growth.

3.5.2. Impact of aquaculture pond effluent on water quality of receiving coastal waters

We further estimated the effects of effluent discharge from the shrimp ponds on the adjacent coastal water column, by comparing the nutrient levels in the coastal waters of the study area (Shanyutan wetland) before and after the discharge of shrimp pond effluents following harvesting (Table 5). Based on an estimated total area of 234 ha and an average depth of 1.4 m for aquaculture ponds in this region, we calculated that a total of $3.3 \times 10^6$ m$^3$ of effluents were released from the ponds into the adjacent
waters without any prior treatment on an annual basis. As shown in Table 5, following
the discharge of shrimp pond effluents, the nutrient levels in the coastal waters around
the study area changed dramatically. The NO$_x$-N, NH$_4^+$-N, TN, PO$_4^{3-}$-P, and TP
concentrations in the adjacent receiving coastal waters of the study area increased
significantly by 350%, 146%, 270%, 415%, and 234%, respectively, following the
discharge of pond effluents. Our results suggested that the traditional practice of annual
drainage of shrimp ponds could potentially increase the nutrient levels of the receiving
waters in the surrounding area within a relatively short time period.

Previous studies have shown that the export of aquaculture pond effluents is an
important anthropogenic source of nutrient pollution in the coastal areas (Seitzinger et
al., 2005; Cardoso-Mohedano et al., 2016a). Lacerda et al. (2008) estimated that a total
of 827 t N yr$^{-1}$ of nitrogen and 69.2 t P yr$^{-1}$ of phosphorus were exported by aquaculture
production from shrimp ponds with an area of 3,279 ha in the Ceará State coast of
northeastern Brazil. In the Gulf of California, the nutrient loads from shrimp
aquaculture were 9044 t N yr$^{-1}$ and and 3078 t P yr$^{-1}$, which were estimated with a mass
balance model assuming an export of 110.2 kg N and 37.5 kg P yr$^{-1}$ ha$^{-1}$, and a total area
of 82,068 ha of shrimp farms (Páez-Osuna et al. 1997; 2013; 2017). On the other hand,
the inputs of nitrogen and phosphorus from shrimp aquaculture was estimated to be
1.5% and 0.9% of the main sources from municipal and agricultural activities,
respectively (Páez-Osuna et al. 1999). In the Mekong Delta, De Silva et al. (2010)
estimated a discharge of 31,602 t of N and 9,893 t of P from a pond-based striped
catfish production system with an area of 7000 ha. In this study, we estimated that the
discharges of TN and TP arising from shrimp aquaculture (1639 ha for area and 1.4 m
for water depth) into the adjacent seawater of the Min River estuary in 2015 were 30.45
t and 2.40 t, respectively. Assuming that our data are representative of the aquaculture
ponds across China with a total area of $2.57 \times 10^6$ ha and a mean water depth of 1.4 m, we calculated that approximately $4.77 \times 10^4$ t N y$^{-1}$ and $3.75 \times 10^3$ t P y$^{-1}$ were exported from the direct discharge of mariculture pond effluents into the adjacent seawater in China. This represents about 5% of the total nutrient loadings from the main rivers of China into the sea (State Oceanic Administration, People’s Republic of China, 2015). Given that the estimated contribution of industrial and municipal sewages to the nutrient loadings in the coastal sea is 32% and 41%, respectively (State Oceanic Administration, People’s Republic of China, 2015), the total nutrient loading from the effluent discharge of aquaculture ponds is rather small in comparison with other anthropogenic nutrient sources. Yet, aquaculture production might still be an important contributor to the problem of water pollution locally in the coastal areas of China, leading to potential negative effects (e.g. cause eutrophication, red tides, and biodiversity loss) on the adjacent coastal ecosystems (Herbeck et al., 2013; Cardoso-Mohedano et al., 2016a; Páez-Osuna et al., 2017). Long-term field monitoring and sampling with multiple frequencies in various regions of China should be carried out in future in order to obtain a better and more precise picture on the influence of aquaculture pond effluent on coastal ecosystems.

4. Conclusions

Based on the analysis of the samples collected from the sediment and water column at three representative shrimp ponds in the Min River estuary at three growth stages over the production cycle, we made some key findings on the biogeochemical cycling of nutrients in the mariculture ponds of China as follows:

1) NH$_4^+$-N was the main form of DIN in both the water column and sediment porewater.

Also, the concentrations of DIN were significant higher than those of PO$_4^{3-}$-P, which implied that P could become a limiting nutrient to phytoplankton growth in the shrimp
ponds.

2) Nutrient (DIN and PO$_4^{3-}$-P) fluxes across the sediment-water interface of shrimp ponds greatly varied among the different growth stages of shrimps, with considerably higher values during the middle and late stages. These results suggested that water quality deterioration caused by high rates of sediment nutrient releases could potentially lead to mass mortality of shrimps during the middle and late stages.

3) The discharge of aquaculture pond effluents could be an important contributor to the problem of water pollution and eutrophication in the receiving waters of the coastal zones in China. Effective treatment of aquaculture pond effluents before discharge will become an important challenge in the future in alleviating the pressures of eutrophication in the coastal zone.

Acknowledgements

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References


**Table 1**

Basic details of the three shrimp ponds in the Min River estuary

<table>
<thead>
<tr>
<th></th>
<th>Pond-1</th>
<th>Pond-2</th>
<th>Pond-3</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Shrimp species</strong></td>
<td><em>Litopenaeus vannamei</em></td>
<td><em>Litopenaeus vannamei</em></td>
<td><em>Litopenaeus vannamei</em></td>
</tr>
<tr>
<td>Mean water depth (m)</td>
<td>1.3</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>Surface area (ha)</td>
<td>0.85</td>
<td>0.70</td>
<td>0.72</td>
</tr>
<tr>
<td>Stocking density (PL m(^{-2}))(^a)</td>
<td>35</td>
<td>45</td>
<td>45</td>
</tr>
<tr>
<td>Survival rate (%)(^a)</td>
<td>62</td>
<td>63</td>
<td>70</td>
</tr>
<tr>
<td>Yield (kg pond(^{-1}) crop(^{-1}))(^a)</td>
<td>2300</td>
<td>2500</td>
<td>3000</td>
</tr>
<tr>
<td>Feed conversion rate(^b)</td>
<td>1.72</td>
<td>1.30</td>
<td>1.11</td>
</tr>
</tbody>
</table>

\(^a\) The data for the stocking density, survival rate, and yield were provided by the farmers.

\(^b\) Feed conversion rate = dry weight of feeds added/wet weight of shrimps produced
Table 2

The three main stages of the grow-out cycle during the aquaculture period in intensive shrimp ponds in the Min River estuary

<table>
<thead>
<tr>
<th>Stage</th>
<th>Duration (days)</th>
<th>Remarks*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial stage</td>
<td>0 – 47</td>
<td>Pond filled, fertilized and stocked. Feeding rate 10 – 16 kg ha⁻¹ d⁻¹. Water depth 1.0 – 1.1 m. Water salinity 3.0 ‰ – 3.6 ‰. Shrimp weight 0.01 – 2.43 g PL⁻¹.</td>
</tr>
<tr>
<td>Middle stage</td>
<td>48 - 114</td>
<td>Feeding rate 50 – 55 kg ha⁻¹ d⁻¹. Water depth 1.4 – 1.5 m. Water salinity 2.2 ‰ – 2.7 ‰. Shrimp weight 2.43 – 9.67 g PL⁻¹.</td>
</tr>
<tr>
<td>Final stage</td>
<td>115-163 (or more)</td>
<td>Harvest, and effluent discharge. Feeding rate 40 – 45 kg ha⁻¹ d⁻¹. Water depth 1.2 – 1.3 m. Water salinity 1.7 ‰ – 2.0 ‰. Shrimp weight 9.67 – 13.25 g PL⁻¹.</td>
</tr>
</tbody>
</table>

* The data for the feeding rate, shrimp weight and water salinity were provided by the farmers.
<table>
<thead>
<tr>
<th></th>
<th>Initial stage</th>
<th>Middle stage</th>
<th>Final stage</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Water physicochemical properties</strong>*</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Temperature (°C)</td>
<td>24.02±0.02&lt;sup&gt;b&lt;/sup&gt;</td>
<td>30.95±0.18&lt;sup&gt;a&lt;/sup&gt;</td>
<td>24.17±0.10&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Dissolved oxygen (mg L&lt;sup&gt;-1&lt;/sup&gt;)</td>
<td>8.31±0.17&lt;sup&gt;b&lt;/sup&gt;</td>
<td>11.88±0.61&lt;sup&gt;a&lt;/sup&gt;</td>
<td>12.39±0.37&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>pH</td>
<td>8.95±0.02&lt;sup&gt;c&lt;/sup&gt;</td>
<td>8.64±0.04&lt;sup&gt;b&lt;/sup&gt;</td>
<td>10.00±0.03&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>Sediment physicochemical properties</strong>&lt;sup&gt;**&lt;/sup&gt;</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Temperature (°C)</td>
<td>22.49±0.03&lt;sup&gt;b&lt;/sup&gt;</td>
<td>28.16±0.12&lt;sup&gt;a&lt;/sup&gt;</td>
<td>22.52±0.22&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Total nitrogen (g kg&lt;sup&gt;-1&lt;/sup&gt;)</td>
<td>1.70±0.08&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.03±0.10&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.86±0.11&lt;sup&gt;ab&lt;/sup&gt;</td>
</tr>
<tr>
<td>Porosity (%)</td>
<td>61.02±2.18&lt;sup&gt;b&lt;/sup&gt;</td>
<td>83.25±8.24&lt;sup&gt;a&lt;/sup&gt;</td>
<td>75.31±4.78&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>Biological parameters</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Shrimp biomass (g shrimp&lt;sup&gt;-1&lt;/sup&gt;)</td>
<td>0.92± 0.01&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.01±0.06&lt;sup&gt;b&lt;/sup&gt;</td>
<td>12.30±0.11&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

* Values are means (±S.E.) of water samples (n = 108) collected at three depths, at four sampling times (08:00, 11:00, 14:00, 17:00 h) and in three ponds for each stage. ** Values are means (±S.E.) of sediment samples (n = 9) collected from three ponds for each stage. Mean values in a row with the same lowercase letter are not significantly different by the Fischer's Least Significant Difference (LSD) test (p < 0.05).
Table 4

Relationship between NO$_x$-N fluxes, dissolved oxygen, and shrimp biomass in the three shrimp ponds.

<table>
<thead>
<tr>
<th>Nutrients fluxes (y)</th>
<th>Variables (x)</th>
<th>Regression equation</th>
<th>$r^2$</th>
<th>Correlation coefficient</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>NO$_2$-N fluxes</td>
<td>Dissolved oxygen</td>
<td>$y = 0.0866x - 0.7249$</td>
<td>0.6993</td>
<td>0.836**</td>
<td>27</td>
</tr>
<tr>
<td></td>
<td>Shrimp biomass</td>
<td>$y = 0.0427x - 0.1393$</td>
<td>0.6480</td>
<td>0.805**</td>
<td>27</td>
</tr>
<tr>
<td>NO$_3$-N fluxes</td>
<td>Dissolved oxygen</td>
<td>$y = 0.2043x - 1.9428$</td>
<td>0.6314</td>
<td>0.795**</td>
<td>27</td>
</tr>
<tr>
<td></td>
<td>Shrimp biomass</td>
<td>$y = 0.1145x - 0.6545$</td>
<td>0.7565</td>
<td>0.870**</td>
<td>27</td>
</tr>
</tbody>
</table>

** denote correlation coefficients for significant relationships at the 0.01 level.
Table 5

Nutrient concentrations in adjacent the coastal waters of the study area before and after the discharge of shrimp pond effluents during the low tide after the completion of shrimp harvesting.

<table>
<thead>
<tr>
<th></th>
<th>NO\textsubscript{3},-N (µg L\textsuperscript{-1})</th>
<th>NH\textsubscript{4},+ -N (mg L\textsuperscript{-1})</th>
<th>TN (mg L\textsuperscript{-1})</th>
<th>PO\textsubscript{4},\textsuperscript{3},-P (mg L\textsuperscript{-1})</th>
<th>TP (mg L\textsuperscript{-1})</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before</td>
<td>119.36±6.91a</td>
<td>0.81±0.16a</td>
<td>0.84±0.15a</td>
<td>0.14±0.03a</td>
<td>0.31±0.07a</td>
</tr>
<tr>
<td>After</td>
<td>418.33±29.97b</td>
<td>1.19±0.18b</td>
<td>2.27±0.14b</td>
<td>0.58±0.10b</td>
<td>0.72±0.15b</td>
</tr>
</tbody>
</table>

Values are means (±S.E.) of samples (n = 6) collected from adjacent coastal waters of the shrimp ponds. Mean values in a column with the same lowercase letter are not significantly different by the independent-samples t-test (p < 0.05).
Fig. 1. The locations of sampling sites in the aquaculture shrimp ponds in the central and western parts of the Min River estuary.
**Fig. 2.** Conceptual diagram of the incubation device used for determining nutrient fluxes across the sediment-water interface (Modified according to Chen et al., 2014).

CTOS in the figure represent constant temperature oscillation incubator.
Fig. 3. Temporal variations of chlorophyll a (Chl-a) concentrations in the water column of shrimp ponds at the Min River estuary. Bars represent mean±SE (n = 9).
Fig. 4. Temporal variations of nutrient concentrations in the water column of shrimp ponds at the Min River estuary. Bars represent mean±SE (n = 36). Different lowercase letters above the bars indicate significant differences at the p<0.05 level among sampling depths at each stage. The red dashed line indicates the safe levels for ammonia and nitrite in the shrimp ponds.
Fig. 5. Temporal variations of porewater nutrient concentrations in the shrimp pond sediment at the Min River estuary. The boxes, center line, and whiskers represent the 25th–75th percentiles, median value, and 5th and 95th percentiles, respectively. The square represents the area-weighted average (n = 9). Different lowercase letters indicate significant differences (p<0.05) in mean concentrations among growth stages.
Fig. 6. Relationships between porewater NH$_4^+$-N concentrations and (a) sediment temperature and (b) TN contents from the shrimp ponds at the Min River estuary.
Fig. 7. Temporal variations of nutrient fluxes across the sediment-water interfaces in the shrimp ponds at the Min River estuary. The boxes, center line, and whiskers represent the 25th – 75th percentiles, median value, and 5th and 95th percentiles, respectively. The square represents the area-weighted average (n = 9). Different lowercase letters indicate significant differences (p<0.05) in mean fluxes among growth stages.