Aquaculture Research, 2013, **44**, 1578–1588 doi: 10.1111/j.1365-2109.2012.03165.x

# Effect of stocking density on soil, water quality and nitrogen budget in *Penaeus monodon* (Fabricius, 1798) culture under zero water exchange system

**Ramamoorthy Saraswathy, Moturi Muralidhar, Muniyandi Kailasam, Pitchaiyappan Ravichandran, Baijnatha Prasad Gupta, Kishore Kumar Krishnani, Alphis Geethanand Ponniah, Jitendra Kumar Sundaray, Akshaya Panigrahi & Arunachalam Nagavel**
Central Institute of Brackishwater Aquaculture, ICAR, Chennai, Tamilnadu, India

**Correspondence:** R Saraswathy, CIBA, 75 Santhome high road, R.A.Puram, Chennai-28, Tamil Nadu, India. E-mails: saraswati_ciba@yahoo.co.in, saraswati@ciba.res.in

## Abstract

Soil and water interactions and their influence on growth and production in different densities (SD8 & SD16) under zero water exchange were studied in two successive crops of *Penaeus monodon* in Tamil Nadu, India during 2007 and 2008. Scraping and tilling during pond preparation increased the mineralization rate than scraping only. During crop, there was no significant difference in soil organic carbon and total nitrogen between the SDs. However, as the crop progressed, organic carbon and total nitrogen content of the soil showed significant difference under both the SDs. Between the SDs, nitrate and phosphate content in water significantly differed, whereas the progress of the crop significantly increased both available and total nutrients. Mass balance of nitrogen indicated that applied feed contributed to 97.4–98.5% of input nitrogen, of which nitrogen in sediment accounted for 16.5–27.3%, nitrogen recovery in shrimp was 34.2–43.6% and the nitrogen lost through denitrification and volatilization varied from 4.7% to 34.7%. Zero water exchange system is highly efficient as nitrogen recovery is higher in shrimp and lower in discharge water. Lack of significant difference in metabolites between the SDs indicates the role of aeration and probiotics in sustaining SD16 cultures.

**Keywords:** shrimp stocking density, culture practices, soil and water quality, nitrogen budget

## Introduction

Properties of sediment and processes occurring at the bottom soil and in the soil–water interface are more critical for shrimp than for any other aquaculture species because shrimp spend most of their time at the bottom, burrow into the soil and ingest detritus from pond-bottom soil (Boyd 1989; Chein 1989). Mineralization and its rate in soil/sediment decide the availability of nutrients in water and consequently, the natural productivity and thus success of extensive and semi-intensive system of farming. Pond water and sediments interact continuously and influence the farming environment. Pond management activities such as feeding, water exchange, and aeration also affect the farming environment (Funge-Smith & Briggs 1998). Consequent upon the viral disease outbreaks, especially White Spot Syndrome Virus (WSSV) since 1995, the farmers have started adopting reduced water exchange to avoid horizontal transmission of virus into their culture system.

The nutrient dynamics of such a 'closed' system of farming is likely to be different from the 'open', high water exchange system of farming. During the recent years, a number of studies have been conducted on the nutrient budgeting under various systems of shrimp farming, in view of the environmental concerns and nutrient loading in receiving waters (Briggs & Funge-Smith 1994; Paez-Osuna, Guerrero-Galyan, Ruiz-Fernandez & Espinoza-angulo 1997; Jacksons, Preston, Peter & Burford 2003; Thakur & lin 2003; Wahab, Bergheim & Brasten 2003). Nitrogen waste outputs in various forms are generally of concern in the marine environment because of its dual role, as a nutrient and toxicant (Persson 1991). Experimental studies indicate that feed inputs during the production cycle account for the bulk of nitrogen inputs to many shrimp ponds (Briggs &

Funge-Smith 1994; Jacksons *et al.* 2003). Much of the nitrogen input is not incorporated into shrimp tissue, but enters the water column as total ammonia nitrogen (TAN), is taken up by phytoplankton and, with their death, settles on the sediment as particulate organic nitrogen. Alternatively, nitrogen input is deposited directly as uneaten feed or faeces (Burford & Williams 2001). Part of the sludge nitrogen is remineralized to enter the water column again as total ammonia nitrogen. The purpose of drying pond bottoms between crops is to reduce the moisture content of soil so that air can enter pore spaces among soil particles, which in turn enhance aerobic decomposition. This will lead to the decomposition of most of the labile organic matter remaining in the bottom soil from the previous crop and to oxidize reduced inorganic compounds (Boyd & Pippopinyo 1994). Such recycled nitrogen may account for bulk of the total ammonia nitrogen input into the water column late in the production cycle (Burford & Long more 2001).

Many studies have been conducted under water exchange systems to understand the changes in natural productivity in relation to pond environment and nitrogen budgeting (Hopkins, Hamilton, Sandfier, Browdy & Stokes 1993; Martin, Veran, Gueloget & Pham 1998; Guerrero-Galvan, Paez-Osuna, Ruiz-Fernandez & Espinoza-Angulo 1999; Tookwinas & Songsangjinda 1999; Gross, Boyd & Wood 2000; Teichert-Coddington, Martinez & Ramirez 2000; Lemonnier & Faninoz 2006; Shah, Hossain, Begum, Ahmed, Ohtomi, Rahman, Alam, Islam & Fulanda 2008). However, there are no many comprehensive studies with holistic approach to bring out the interaction between soil and water productivity with nitrogen dynamics under zero water exchange system. Hence, this study was carried out to understand the effect of stocking densities and different management methods on water and soil interactions, growth and production in commercial zero water exchange culture of *Penaeus monodon*. The nitrogen budget was also estimated and was compared with other system of farming to evaluate the level of nutrient loading in source water.

## Materials and methods

### Farm details and management practices

The study was carried out in *P. monodon* ponds, each with an area of 0.62 ha and water depth of 1–1.5 m during the winter (August to December) crop of 2007 (CR_I) and 2008 (CR_II) under two different stocking densities *viz.*, SD8 (8 no. m$^{-2}$) and SD16 (16 no. m$^{-2}$). The ponds were located at Mahabalipuram, Kancheepuram District, Tamil Nadu and the water source was from Edayur back water connected with Buckingham canal. The experimental ponds were allowed to dry after the harvest during January to May. In CR_I, scraping was performed during pond preparation, whereas in CR_II, both scraping and tilling were performed. Liming was performed in all the ponds at the rate of 750–800 kg ha$^{-1}$. Stocking was performed with PL$_{18}$ of *P. monodon* at both SD8 and SD16 ponds. No water exchange and fertilization were performed during the crop periods of both the crops. Aeration and water probiotics were provided only to SD16 ponds. Aeration was provided with paddle wheel aerators (6 numbers/ha), at the rate of 6 h day$^{-1}$ from 60th day, 10 h day$^{-1}$ from 90th day and 12 h from 105th day until the harvest. The shrimps were fed commercial pellet feed throughout the crop in both the practices. Soil nutrient levels during fallow period and crop period were estimated at monthly intervals and water nutrient levels during the crop were estimated at fortnightly intervals. Culture and production details are given in Table 1.

**Table 1** Culture and production details during the crop

| | 2007 (CR_I)* | | 2008 (CR_II)* | |
|---|---|---|---|---|
| | Farm 1 | Farm 2 | Farm 1 | Farm 2 |
| Stocking density (PL ha$^{-1}$) | 80000 | 160000 | 80000 | 160000 |
| Days of culture (DOC) | 135 | 135 | 105 | 102 |
| Survival (%) | 90 | 89 | 80 | 78 |
| FCR† | 1.03 | 1.43 | 1.2 | 1.34 |
| Mean body weight (g) | 30 | 25 | 25 | 22 |
| Production (kg ha$^{-1}$) | 2000 | 3560 | 1920 | 2950 |

*Winter crop of 2007 and 2008.

†Feed conversion ratio.

### Sampling and analysis

Aquatic sediment samples were collected from the upper 5 cm layers of pond bottom using core soil sampler. This layer was selected for sampling because it is more reactive where complex chemical and microbial changes occur and has a greater influence on water quality than deeper layer (Masuda & Boyd 1994; Munsiri, Boyd & Hajek 1995; Krishnani, Rajendran, Joseph & Gupta 1997). Soil samples were collected once in a month during both fallow and crop periods and were analysed for organic carbon by wet combustion method (Walkley & Black 1934) and total nitrogen by Macrokjeldahal method (Piper 1966). Available nitrogen and phosphorus content were determined by alkaline permanganate method (Subbiah & Asija 1956) and Olsen method (Olsen, Cole and Watanabe 1954) respectively.

Water samples were collected in all the ponds at fortnightly intervals between 07:00 and 08:00 hours in 500 mL polyethylene bottles for the analysis of pH, ammonia-N, nitrite-N, nitrate-N and soluble reactive phosphorus and were analysed immediately in the laboratory. *In situ* measurements were done for temperature and salinity using thermometer and hand refractometer respectively. pH was determined as described in APHA (1989). Water nutrient parameters like nitrate-N, ammonia-N, nitrite-N and reactive P were analysed by standard methods (Strickland & Parsons 1972). Nitrogen budget was calculated for both the stocking densities based on the input (nitrogen in the form of water and feed) and output (shrimp uptake) including wastage.

Prerequisites to achieve a reduction in nitrogen waste and increased recovery in shrimp are a quantitative understanding of the nitrogen mass balance budget and a detailed understanding of the different forms of input and output nitrogen. Input nitrogen included nitrogen content in inlet water and feed pellet nitrogen (farm records for feed addition). Output nitrogen included nitrogen content in water and sediment at harvest time, shrimp nitrogen and nitrogen lost to atmosphere. The nitrogen losses through denitrification and volatilization were estimated indirectly as the difference between nitrogen inputs and outputs. The input and output were calculated as follows:

Input water nitrogen
= Nitrogen concentration in inflow water
× volume of inflow

Feed pellet nitrogen
= Nitrogen concentration in feed
× total weight of feed

Output water nitrogen
= Nitrogen concentration in outflow water
× volume of outflow

Shrimp nitrogen
= Nitrogen concentration in shrimp
× weight of harvested shrimp

### Statistical analysis

Soil and water parameter variations with days of crop and stocking densities were analysed statistically using ANOVA and Duncans Multiple Range Test (DMRT). All the statistical analyses were performed using SPSS (V) 16 software package (SPSS Inc., Chicago, IL, USA).

## Results and discussion

### Mineralization of soil nutrients during fallow period

During the fallow period, organic carbon content of the soil reduced gradually (Fig. 1) in both the crops and stocking densities (0.09–0.12%). The decrease in organic carbon content was due to microbial mineralization of organic matter present in the soil (in the form of animal excreta, dead organisms *etc.*) and release of nutrients in the available form. Metabolic products of aerobic decomposition of organic matter are carbon dioxide, water, ammonia and other nutrients like nitrogen and phosphorus (Boyd, Wood & Taworn 2002a). The main benefit of keeping the pond soil fallow is to reduce oxygen demand of bottom soil as much as possible before beginning of a new crop. The 5-month fallow period registered a 35% reduction in organic carbon content before CR_I crop, whereas the same recorded a 26% reduction before initiation of CR_II crop. The difference is mainly due to the higher initial levels observed in

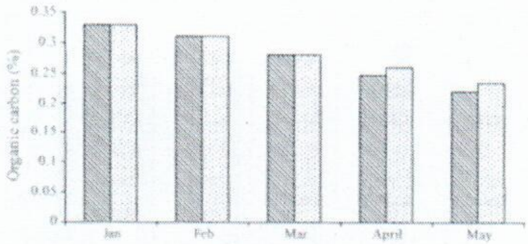

**Figure 1** Mean values of organic carbon content during fallow period.

CR_II. The pond preparation was initiated in both the crops in May with scraping of surface sediment and liming. In addition to this, tilling was performed before initiation of CR_II. After the pond preparation, in CR_I, there was no reduction in the organic carbon content of the soil. But in CR_II, where tilling was also performed, there was a further reduction of 25% of the organic carbon. In spite of higher organic carbon content at the beginning of fallow period of CR_II, there was no significant variation in organic carbon content (0.18–0.21%) between both the crops and both densities, mainly due to the enhanced mineralization in CR_II due to tilling. In wet soil, tilling probably sustains the mineralization process for a longer period and makes it more efficient. In dry soil, this could be due to the dilution effect of mixing of high organic carbon content of surface layer of soil with deeper soils of low organic carbon content (Boyd, Wood & Taworn 2002b).

Drying of the pond bottom after the harvest led to a reduction in both total nitrogen and phosphorus content of soil. The average total nitrogen decreased from 535 to 400 mg L$^{-1}$ and average total phosphorus from 25.58 to 19.6 mg L$^{-1}$, without significant variation among both the densities and crops. In contrast, the available nitrogen and phosphorus levels increased during the mineralization process. Available nitrogen increased from 18% to 26% of total nitrogen and the available phosphorus increased from 33% to 46% of total phosphorus (Figs 2 and 3). Available nitrogen content did not vary between the densities because of similar rate of mineralization under same environment condition. Boyd *et al.* (2002a) observed that the mineralization rate in soil will increase up to the optimum moisture content and then decline if soils are dried further. Hence, drying for longer than optimum period will not be useful for mineralization. In the present case, the drying was performed for 4 months before initiating the pond preparation and the rate of mineralization during the different months was not significantly different, suggesting continued mineralization.

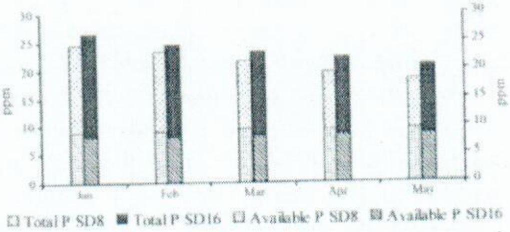

**Figure 3** Mean values of changes in phosphorus content during fallow period.

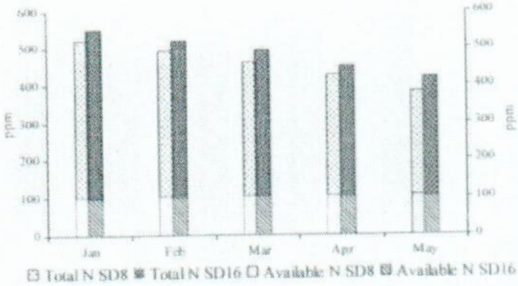

**Figure 2** Mean values of changes in nitrogen content during fallow period.

During the fallow period, when mineralization of the accumulated waste is taking place, the mean values of pH of the soil gradually reduced from 8.45 to 7.4 for both the stocking densities due to carbon-di-oxide formation as an end product of decomposition. The reduction was minimal and well within the optimal levels. However, with the application of lime during the pond preparation in the last week of May, pH was restored back to the normal range of 8.3–8.4. In the present study, the reduction was minimal and well within the optimal levels.

### Mineralization of soil nutrients during crop

Uneaten feeds, fertilizer, dead bodies of aquatics and phytoplankton deposited in the bottom soil constitute the pond sediment. Irrespective of the crop and densities, organic carbon content in the sediment increased during the crop period due to the accumulation of excess feed, metabolites, animal excreta, *etc* (Fig. 4). Significantly higher level of increase was observed during the 3rd and 4th

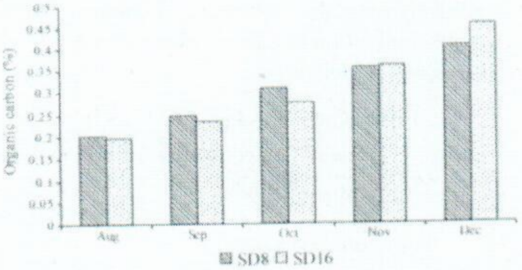

**Figure 4** Mean values of organic carbon content during crop period.

month of crop commensurate with the increased level of biomass and feed used. Such enrichment of bottom soil with organic matter in shrimp ponds has been reported by many authors (Maguire & Allen 1986; Boyd 1992; Boyd, Tanner, Madkour & Masuda 1994; Smith 1996; Martin *et al.* 1998). Furthermore, there are reports that the accumulation of organic matter is density-dependent (Briggs & Funge-Smith 1994; Smith 1996; Paez-Osuna *et al.* 1997; Martin *et al.* 1998; Islam & Alam 2008). However, in the present study, the increase was not significantly different between the densities SD8 and SD16 ($P > 0.05$). This was mainly due to high rate of decomposition and mineralization induced by aeration in SD16 ponds. Hence, proper aeration is one of the most critical management requirements in SD16 ponds not only considering the oxygen demand of the stock, but to induce the process of decomposition and mineralization.

Similarly, during crop, total nitrogen level in pond soil increased by 100 ppm in 4 months in SD8 ponds and by 140 ppm in SD16 ponds. The increase in total nitrogen content was significantly different between the months ($P < 0.05$), but not between the densities ($P > 0.05$). The expected increase in total nitrogen content in SD16 ponds with higher feeding did not occur mainly because of the maintenance of adequate dissolved oxygen levels through aeration and use of probiotics (Hopkins *et al.* 1993). Aeration increases mineralization, reduces water exchange, supplies oxygen and circulates water in the pond (Rogers 1989; Avnimelech, Mozes & Weber 1992; Howerton, Boyd & Watten 1993; Avnimelech & Ritvo 2001), thereby keeping the pond bottom free of sludge (Chang & Ouyang 1988; Losordo & Piedrahita 1991; Fast & Lannan 1992; Boyd & Dhendup 1995; Avnimelech & Ritvo 2001). It also reduces vertical stratification and increases habitat available for shrimp growth (Rogers & Fast 1988).

Organic matter accumulated in the sediment is decayed by microorganism, and forms various states of inorganic nitrogen and phosphorus. Available nitrogen level in the sediment changed during the crop period due to rate of mineralization, nutrient interaction between the soil and water and utilization of nutrients by shrimps, planktons and other grazing organisms. In the present study, around 22% of the nitrogen was present as available form in the shrimp pond soil of both SD8 and SD16 ponds and it was supported by the findings of Smith (1996), wherein he reported 77–83% of nitrogen in unavailable organic form in *P. monodon* culture with the stocking density of 25–40 $m^{-2}$ and water circulation at 10–15 $ms^{-1}$. There was no significant difference between the months with respect to available nutrients ($P > 0.05$). This is mainly due to increase in utilization of nutrients as the culture progresses for the primary productivity in the pond (Fig. 5).

Similar to total nitrogen, total phosphorus content also increased during the crop period. The level of increase of phosphorus was significantly higher during the last 2 months of crop because of higher amount of feed input. Although the total phosphorus increased by 20–27% of the initial value, the available phosphorus during the crop did not show any significant variation between the different months. In SD16, the available phosphorus was 36.8% of total phosphorus, while it was 43.6% in SD8 ponds (Fig. 6), indicating that there was a significant difference between the densities during the crop period.

The ratio of major elements has greater influence on microbial activity and enzyme activity of soil and water, and thereby affects the rate of release of nutrients from organic manures. C:N

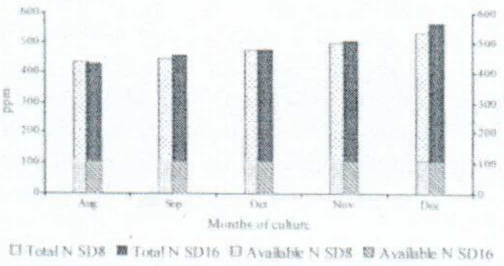

**Figure 5** Mean values of changes in nitrogen content during culture period.

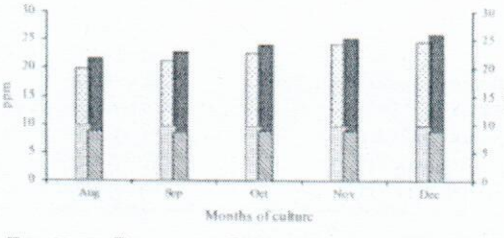

**Figure 6** Mean values of changes in phosphorus content during culture period.

ratio is highly useful in interpreting the source of organic matter and to identify excess nutrients in sediments (Mantoura 1981). It also influences the presence of phytoplankton communities. Carbon: nitrogen ratio increased during the crop period from 4.55 to 7.77 and 4.76 to 7.35 under SD8 and SD16 ponds respectively in CR_I. The same trend was observed in CR_II as well. This ratio was within the optimum range for better productivity (Gupta, Muralidhar, Joseph & Kirshnani 2001).

### Nutrients in water

The various states of inorganic nitrogen and phosphorus in the sediment are partly released into the pond water. Phytoplankton assimilate the nitrogen and phosphorus formed in the pond water, and are then consumed by the living aquatics, which include shrimp (Zhang, Tan & Ou 1989). Two equilibriums determine the variation of total nitrogen and total phosphorus concentrations in pond water: First, the equilibrium between nitrogen and phosphorus accumulation and release, secondly the equilibrium between the aquatics preying and the phytoplankton reproducing (Xia, Yang & Yan 2004).

In the present study, total nitrogen and total phosphorus content in water increased significantly with the progress of crop ($P < 0.01$). In SD16 ponds, total nitrogen ranged between 0.51 and 1.31 mg L$^{-1}$ and total phosphorus ranged from 0.085 to 0.32 mg L$^{-1}$ (Table 2). In SD8 ponds, values were comparatively less, but the same trend was observed. No significant difference was observed between the stocking densities ($P > 0.05$) with respect to total nitrogen and total phosphorus.

Ammoniacal and nitrate forms of nitrogen constitute the ready source of available nitrogen to primary fish food organisms in food chain. Among the various forms of nitrogenous nutrients, nitrate is the most important, as it is the final form, being absorbed by plankton for their growth (Begum, Hossain, Wahab & Kohinoor 2003). Nitrate concentration increased during the crop period of CR_I and it ranged from 0.074 to 0.15 and 0.075 to 0.25 mg L$^{-1}$ in SD8 and SD16 ponds respectively. There was a significant difference between the densities ($P < 0.01$) as well as between the months of crop ($P < 0.01$). The same trend was followed in the second crop also.

Another form of nitrogen, total Ammonia Nitrogen concentration in the water column, increased due to shrimp gill excretion, sediment remineralization process (Burford & Longmore 2001) and microbial processes (Burford & Glibert 1999) in the water column. Total ammonia nitrogen concentration was observed to be low initially and gradually increased as the crop progressed. The concentration varied from 0.22 to 0.89 mg L$^{-1}$ in SD8, whereas it varied from 0.27 to 0.93 mg L$^{-1}$ in SD16 ponds under CR_I (Fig. 7). The same trend was observed in CR_II upto 90 days and then reduced because of dilution of pond water with rain water (Fig. 8).

Phosphorus, although required in small quantities for aquatic biota, is the single most important element in water, as it is needed for phytoplankton growth (Hossian, Begum, Ahmed, Hoque, Karim & Wahab 2006). Phosphate-phosphorus concentration ranged from 0.142 to 0.21 mg L$^{-1}$ and

**Table 2** Mean water parameters during culture period (mean ± SE)

| | 2007 (CR_I)[*] | | 2008 (CR_II)[*] | |
|---|---|---|---|---|
| | SD8[†] | SD16[†] | SD8[†] | SD16[†] |
| pH[‡] | $7.92 \pm 0.081^{a}$<br>(7.22–8.25) | $7.75 \pm 0.078^{b}$<br>(7.04–8.13) | $8.27 \pm 0.048^{a}$<br>(8.0–8.5) | $8.22 \pm 0.043^{a}$<br>(7.97–8.44) |
| Dissolved oxygen (mg L$^{-1}$)[‡] | $6.01 \pm 0.135^{a}$ | $5.64 \pm 0.147^{b}$ | $6.03 \pm 0.078^{a}$ | $5.38 \pm 0.086^{b}$ |
| Salinity (g L$^{-1}$)[‡] | $18.5 \pm 0.3^{a}$ | $17.7 \pm 0.23^{a}$ | $19.3 \pm 0.31^{a}$ | $18.7 \pm 0.37^{a}$ |
| Temperature (°C)[‡] | $29.81 \pm 0.17^{a}$ | $30.38 \pm 0.189^{a}$ | $28.81 \pm 0.24^{a}$ | $28.82 \pm 0.24^{a}$ |
| Total N (mg L$^{-1}$)[‡] | $0.847 \pm 0.058^{a}$ | $0.91 \pm 0.055^{a}$ | $0.81 \pm 0.057^{a}$ | $0.83 \pm 0.057^{a}$ |
| Total P (mg L$^{-1}$)[‡] | $0.157 \pm 0.014^{a}$ | $0.192 \pm 0.015^{a}$ | $0.15 \pm 0.01^{a}$ | $0.169 \pm 0.016^{a}$ |

[*] Winter crop of 2007 and 2008.

[†] Stocking densities of 8 and 16 m$^{-2}$.

[‡] Values in a row with different superscript letters are significantly different ($P \leq 0.05$).

Values in parenthesis are ranges of pH.

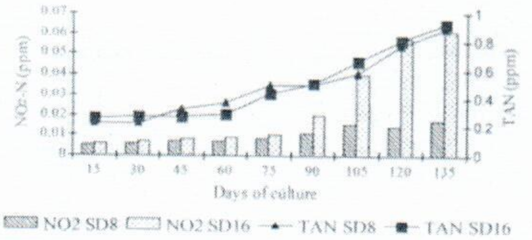

**Figure 7** Changes in nitrogen metabolites during CR_I.

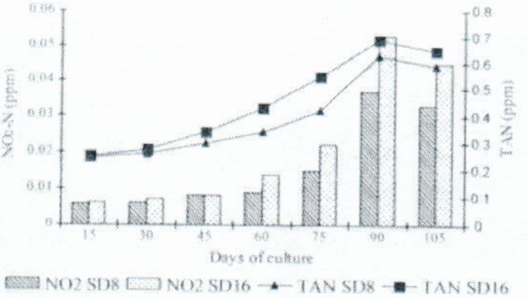

**Figure 8** Changes in nitrogen metabolites during CR_II.

0.157 to 0.37 mg $L^{-1}$ in SD8 and SD16 ponds respectively in CR_I. The availability of phosphorus in CR_II exhibited the same trend. The Phosphate-phosphorus levels were significantly lower in SD8 than SD16 ponds and it was significantly different among the different months of crop ($P < 0.05$). The trends observed in the present study are similar to findings by Shah *et al.* (2008).

Total ammonia nitrogen especially unionized-ammonia and nitrite are toxic metabolites and their toxicity is highly influenced by pH. In this study, pH was observed within the optimum range and it was comparatively low at the end of crop due to the decomposition of sediments that would liberate hydrogen ions (Praphrutham 1985). This optimum range of pH kept all the metabolites within limits. The unionized portion of total ammonia nitrogen was estimated from total ammonia nitrogen concentration, pH and temperature and it varied from 0.0047 to 0.0917 mg $L^{-1}$ and 0.0028 to 0.0603 mg $L^{-1}$ in SD8 and SD16 ponds respectively in CR_I. There was no significant difference between the stocking densities ($P > 0.05$) with respect to total ammonia nitrogen and unionized-ammonia in both the crops (Figs 7 and 8).

The concentration of nitrite nitrogen ranged from 0.005 to 0.015 mg $L^{-1}$ and 0.006 to 0.04 mg $L^{-1}$ in SD8 and SD16 ponds respectively in CR_I. Average nitrite concentrations were significantly higher ($P < 0.01$) in SD16 than SD8 in CR_I but there was no significant difference observed between the densities in CR_II ($P > 0.05$). The metabolite values were within the permissible limits for shrimp growth under both the stocking densities (Figs 7 and 8). Lack of significant increase in SD16 might be mainly due to the continuous aeration and use of probiotics. Use of probiotics regulates the micro flora to improve nutrient cycle and can create a balance between bacteria and micro algae, and maintaining a good water quality environment for the cultured animals (Li, Zhang & Yang 1997; Dalmin, Kathiresan & Purushothaman 2001).

## Nitrogen budget

Rates and magnitude of nitrogen fluxes will vary depending on the quantity and quality of nitrogen input, the types of animals under culture, rates of water exchange and a number of other environmental variables. Most sources of nitrogen entering the pond, nitrogen losses and nitrogen sinks were quantified to determine the mass balance of this nutrient and elucidate major sources of flux. Input data included nitrogen content of inlet water during filling, feed and feed quantity. Output data included nitrogen content of discharge water at harvest time, sediment nitrogen and nitrogen deposited in shrimp tissue.

The nitrogen budget in the present study was calculated as per the method used by Martin *et al.* (1998) in *Penaeus stylirostris* farming and is presented in Table 3. Nitrogen budget revealed that the total input nitrogen in both SD8 and SD16 ponds were 13.2 and 30.47 g $m^{-2}$ respectively, of which 97.4% and 98.5% respectively were contributed by feed. Briggs and Funge-Smith (1994) studied nutrient flow for a series of ponds over 2 or 3 crop cycles and revealed that 95% of the nitrogen applied to the ponds was in the form of feed and fertilizers, whereas Christopher Jackson (2003) observed that 90% of nitrogen entered the farm pond as formulated feed. Other minor contributors were input water and post larvae. Contribution from intake water is very low and it varied between 1.54% and 3.04%. Of the total nitrogen input, 43.6% and 34.2% were converted to nitrogen in shrimp tissue in SD8 and SD16 respectively, indicating that the feed conversion is higher in SD8 (FCR 1.1) as compared with SD16 ponds (FCR 1.38). This

**Table 3** Nitrogen budget in the ponds (mean values of two crops)

| No. | Parameters | ***Penaeus monodon*** **under zero water exchange** | |
|---|---|---|---|
| | Stocking density (No m$^{-2}$) | 8 | 16 |
| A | Pond area (m$^2$) | 10000 | 10000 |
| B | Harvest kg/wet weight | 1960 | 3255 |
| C | Feed pellets-N (kg ha$^{-1}$) | 127.5 | 289.6 |
| D | Shrimp harvest-N (kg ha$^{-1}$) | 57.6 | 104.2 |
| E | Feed efficiency-N (%) | 45.05 | 36.2 |
| F | Total waste-N (Kg ha$^{-1}$) | 69.9 | 185.4 |
| G | Waste-N (g kg$^{-1}$ shrimp harvested) | 35.7 | 56.7 |
| H | Total waste-N (g m$^{-2}$) | 6.99 | 18.55 |
| I | Waste in outflow water-N (g m$^{-2}$) | 1.46 | 2.93 |
| J | Waste-N in sediment + atm. (g m$^{-2}$) | 5.54 | 15.62 |
| Input-N (g m$^{-2}$)* | | | |
| K | Water-N | 0.4 (3.04) | 0.47 (1.54) |
| L | Feed pellet-N | 12.8 (97.4) | 30.0 (98.5) |
| M | Total input-N | 13.2 (100) | 30.47 (100) |
| Output-N (g m$^{-2}$)* | | | |
| N | Water-N | 1.85 (14.0) | 3.4 (11.2) |
| O | Shrimp-N | 5.76 (43.6) | 10.42 (34.2) |
| P | N in sediment | 3.6 (27.3) | 5.04 (16.5) |
| Q | N to atmosphere | 1.94 (14.7) | 10.58 (34.7) |

*Values given in brackets indicate the percentage of the compartment vs. total input/output N.

The different compartments of nitrogen were calculated as follows:

C = N concentration in feed × total weight of feed; D = N concentration in shrimp × weight of shrimp harvested; E = (D/C) × 100; F = C–D; H = F/A; I = N–K; J = H–I; K = (N concentration in inflow water × volume of inflow)/A; L = C/A; M = K+L; N = N concentration in outflow water × volume of outflow; O = D/A; Q = M – (N+O+P).

finding is similar to the observation by Xia *et al.* (2004), wherein he determined that 32.94% of total nitrogen was in shrimp tissue under *P. vannammei* with the stocking density of 50 m$^{-2}$. The nitrogen in waste water was 11.2% of the total input nitrogen in SD16 and 14% in SD8.

When calculating nitrogen budgets, unaccounted loss is assumed to be nitrogen lost to the atmosphere through volatilization of ammonia and denitrification that are often not measured directly. Therefore, in most studies including the present one, these factors are estimated indirectly as the difference between the nitrogen inputs and outputs. Unaccounted loss in the present study was 14.7–35.8% of the total input nitrogen. However, widely varying results have been obtained in other studies. Most estimated less than 15%; Martin *et al.* (1998) and Briggs and Funge-Smith (1994) estimated about 10% in low stocking density ponds. Much higher losses up to 57% (Boyd 1985) and 66% (Paez-Osuna, Guerrero-Galvan & Ruiz-Fernandez 1999) have also been reported.

Denitrification is the reduction of nitrate to gaseous nitrogen and has been shown to be important path way for nitrogen loss in marine ecosystems (Seitzinger, Nixon & Pilson 1984). In addition to adequate nitrate, a source of organic carbon and anaerobic conditions are required for denitrification. Hargreaves (1998) concluded that low nitrification rate was the major limitation for denitrification in pond-based aquaculture. Volatilization does not account for significant loss of nitrogen in typical marine aquaculture ponds with TAN less than 1 mg L$^{-1}$ and in the pH range of 7.5–8.5 (Kochba, Diab & Avnimelech 1994; Lorenzen, Struve & Cowan 1997). In the present study, substantial nitrate was present in SD8 (14%) and SD16 ponds (18.7%); therefore, potential nitrogen lost through denitrification and volatilization did exist. It is supported by Daniels and Boyd (1989) and they reported 55% of nitrogen lost through denitrification and volatilization, in brackishwater fish ponds, when it had very low exchange rate (about 10% over 5 months) and 10–30% of nitrate present. Our finding is in agreement with these studies, although the extent of unaccounted nitrogen as a function of multi factorial attributes varies from experiment to experiment.

Between the stocking densities, unaccounted loss was aided by aeration under SD16 ponds (Reeves 1972). The increase in nitrogen lost to atmosphere

**Table 4** Comparison of nitrogen budget studies

| | **Semi intensive farming** | | | | | **Intensive shrimp farming** | |
|---|---|---|---|---|---|---|---|
| | **Martin *et al.* (1998)** | | ***Penaeus monodon* (present study)** | | **Paez-Osuna *et al.* (1999)** | **Funge-Smith and Briggs (1998) Open type system** | **Briggs and Funge-Smith (1994)** |
| System | 10% water exchange/day | | Zero water exchange | | 4% water exchange/day | 5–10% water exchange/day | Na |
| Species | *P. stylirostris* | | *P. monodon* | | *P. vannamei* and *P. stylirostris* | Shrimp farms | Marine shrimp ponds |
| Stocking density (no. $m^{-2}$) | 7 | 15 | 8 | 16 | Na | Na | Na |
| Discharge water N (%) | 34.3 | 26.6 | 14.0 | 11.2 | 11.2 | 27.0 | 35.0 |
| Shrimp harvest N (%) | 21.5 | 25.0 | 43.6 | 34.2 | 27.0 | 18.0 | 24.0 |
| N in sediment (%) | 20.1 | 16.1 | 27.3 | 16.5 | na | 24.0 | 31.0 |
| N in atmosphere (%) | 24.1 | 32.4 | 14.7 | 34.7 | 65.7 | 30.0 | na |

Na, not available.

aided by aeration in SD16 ponds explains the comparatively lower nitrogen content in discharge water. Hopkins *et al.* (1993) reported that unaccounted loss is much higher in zero water exchange ponds than in water exchange ponds and it was 13–46% of nitrogen added as feed.

It is also possible that some unaccounted nitrogen was sequestered in the sediment through accumulation of organic sludge (Jackson 2003; Briggs & Funge-Smith 1994). In this study, there was no attempt to measure increase in sediment volume and to estimate the amount of nitrogen accumulating in the sediment, yet sediment accumulation of nitrogen in shrimp farms is generally substantial.

A comparison of nitrogen budgets of semi intensive and intensive systems of farming with present study on zero water exchange system is given in Table 4. It showed that nitrogen recovery in shrimp is higher in the case of the zero water exchange system for *P. monodon* farming than in the case of farming with water exchange. Similarly, nitrogen in discharge water was the lowest in the zero water exchange system. Hence, this system seems to be more efficient and environmentally safer than the other systems of farming. Within this system, SD8 ponds resulted in a better nitrogen recovery in shrimp and higher accumulation of nitrogen in sediment, whereas in SD16, accumulation in sediment is less and leads to more nitrogen to atmosphere. The higher accumulation of nitrogen in sediment can be reduced by better management practices during pond preparation and crop period. The nitrogen in discharge water was more or less same in both the densities (14% in SD8 and 11.2% in SD16) and it may be mainly due to the aeration provided only in SD16 crop and low input used in SD8 crop.

## Conclusion

The present study indicated that zero water exchange system of *P. monodon* shrimp farming is more efficient and environmentally safe due to high nitrogen recovery in shrimp as well as lower nitrogen content in discharge water compared with other systems of farming. Increased stocking density (SD16) with aeration and use of probiotics resulted in similar metabolite load as that was observed in SD8 ponds without aeration, in spite of the higher level of feed used and yield obtained. Similarly, pond preparation method of tilling improved the mineralization of organic matter in soil.

## References

APHA (American Public Health Association), American Water Works Association, Water Pollution Control Association (1989) *Standard Methods for the Examination of Water and Waste Water* (17th edn). American Public Health Association, Washington, DC, USA.

Avnimelech Y. & Ritvo G. (2001) Aeration, mixing and sludge control in shrimp ponds. *Global Aquaculture Alliance Advocate* **4**, 51–53.

Avnimelech Y., Mozes N. & Weber B. (1992) Effects of aeration and mixing on nitrogen and organic matter transformations in simulated fish ponds. *Aquacultural Engineering* **11**, 157–169.

Begum M., Hossain M.Y., Wahab M.A. & Kohinoor A.H.M. (2003) Effects of iso-phosphorus fertilizers on water quality and biological productivity in fish pond. *Journal of Aquaculture in the Tropics* **18**, 1–2.

Boyd C.E. (1985) Chemical budgets for channel ponds. *Transaction of the American Fisheries Society* **114**, 291–298.

Boyd C.E. (1989) *Water Quality Management and Aeration in Shrimp Farming Fisheries and Allied Aquacultures Department Series No: 2*. Auburn University, Alabama, USA.

Boyd C.E. (1992) Shrimp pond bottom soil and sediment management. In: *Proceedings of Special Session on Shrimp Farming* (ed. by J.Wyban), pp. 166–181. World Aquaculture Society, Baton Rouge, Louisiana, USA.

Boyd C.E. & Dhendup T. (1995) Quality of potential effluents from the hypolimnia of watershed ponds used in aquaculture. *The Progressive Fish-Culturist* **57**, 59–63.

Boyd C.E. & Pippopinyo S. (1994) Factors affecting respiration and dry bottom soils. *Aquaculture* **120**, 283–293.

Boyd C.E., Tanner M.E., Madkour M. & Masuda K. (1994) Chemical characteristics of bottom soils from freshwater and brackishwater aquaculture ponds. *Journal of World Aquaculture Society* **25**, 517–534.

Boyd C.E., Wood C.W. & Taworn T. (2002a) Pond soil characteristics and dynamics of soil organic matter and nutrients. In: *Nineteenth Annual Technical Report Pond Dynamics/Aquaculture Collaborative Research Support Programme* (ed. by K.Mcelwee, K.Lewis, M.Nidiffer & P.Buitrgo), pp. 1–10. Oregon State University, Corvallis, Oregon.

Boyd C.E., Wood C.W. & Taworn T. (2002b) *Aquaculture Pond Bottom Soil Quality Management*. Oregon State University, Corvallis, Oregon, USA.

Briggs M.R.P. & Funge-Smith S.J. (1994) A nutrient budget of some intensive marine ponds in Thailand. *Aquaculture and Fisheries Management* **25**, 789–811.

Burford M.A. & Glibert P.M. (1999) Short term nitrogen uptake and regeneration in early and late growth phase shrimp ponds. *Aquaculture Research* **30**, 215–227.

Burford M.A. & Longmore A.R. (2001) High ammonium production from sediments in hypereutrophic aquaculture ponds. *Marine Ecology Progress Series* **224**, 187–195.

Burford M.A. & Williams K.C. (2001) The fate of nitrogenous waste from shrimp feeding. *Aquaculture* **198**, 79–93.

Chang W.Y. & Ouyang H. (1988) Dynamics of dissolved oxygen and vertical circulation in fish ponds. *Aquaculture* **74**, 263–276.

Chein Y.H. (1989) Study on the sediment chemistry of tiger prawn, kuruma prawn and red tailed prawn ponds in I-Lan Hsien. *Council of Agriculture Fisheries Series* **16**, 257–275.

Dalmin G., Kathiresan K. & Purushothaman A. (2001) Effect of probiotics on bacterial population and health status of shrimp in culture pond ecosystem. *Indian Journal of Experimental Biology* **39**, 939–942.

Daniels H.V. & Boyd C.E. (1989) Chemical budgets for polyethylene-linked, brackish water ponds. *Journal of the World Aquaculture Society* **20**, 53–60.

Fast A.W. & Lannan J.E. (1992) Pond dynamic processes. In: *Developments in Aquaculture and Fisheries Science* (ed. by A.W.Fast & L.J.Lester), pp. 431–456. Elsevier, New York.

Funge-Smith S.J. & Briggs M.R.P. (1998) Nutrient budget in shrimp ponds: implications for sustainability. *Aquaculture* **164**, 117–133.

Gross A., Boyd C.E. & Wood C.W. (2000) Nitrogen transformations and balance in channel catfish ponds. *Aquacultural Engineering* **24**, 1–14.

Guerrero-Galvan S.R., Paez-Osuna F., Ruiz-Fernandez A.C. & Espinoza-Angulo R. (1999) Seasonal variation in the water quality and chlorophyll a of semi-intensive shrimp ponds in a subtropical environment. *Hydrobiologia* **391**, 33–45.

Gupta B.P., Muralidhar M., Joseph K.O. & Kirshnani K.K. (2001) Effect of salinity level and manure dose on the release of nutrients from brackish water soil. *Aquaculture* **2**, 191–200.

Hargreaves J.A. (1998) Nitrogen biochemistry of aquaculture ponds. *Aquaculture* **166**, 181–212.

Hopkins J.S., Hamilton R.D., Sandfier P.A., Browdy C.L. & Stokes A.D. (1993) Effect of water exchange on production water quality effluent characteristic and nitrogen budgets of intensive shrimp ponds. *Journal of the World Aquaculture Society* **24**, 304–320.

Hossian M.Y., Begum M., Ahmed Z.F. Hoque M.A., Karim M.A. & Wahab M.A. (2006) A study of the effect of Iso-phosphorus fertilizers on plankton production in fish ponds. *South Pacific Studies* **26**, 101–110.

Howerton R.D., Boyd C.E. & Watten B.J. (1993) Design and performance of a horizontal, axial-flow water circulator. *Journal of Applied Aquaculture* **3**, 163–183.

Islam M.L. & Alam M.J. (2008) Optimization of stocking rates of tiger shrimp under modified improved culture system. *Journal of Bangladesh Agricultural University* **6**, 341–348.

Jackson C.J. (2003) Managing the development of sustainable shrimp farming in Australia. The role of sedimentation ponds in treatment of farm discharge water. *Aquaculture* **226**, 23–34.

Jacksons C., Preston N., Peter J.T. & Burford M. (2003) Nitrogen budget and effluent nitrogen components at an intensive shrimp population. *Aquaculture Engineering* **4**, 85–92.

Kochba M., Diab S. & Avnimelech Y. (1994) Modeling of nitrogen transformation in intensive aerated fish ponds. *Aquaculture* **120**, 95–104.

Krishnani K.K., Rajendran K.V., Joseph K.O. & Gupta B.P. (1997) Soil and water characteristics of shrimp ponds affected with viral disease. *Journal of Inland Fisheries Society of India* **29**, 37–42.

Lemonnier H. & Faninoz S. (2006) Effect of water exchange on effluent and sediment characteristics and on partial nitrogen budget in semi-intensive shrimp ponds in New Caledonia. *Aquaculture Research* **37**, 938–948.

Li Z., Zhang Q. & Yang H. (1997) The effect of the probiotics to the shrimp ponds. *Aquaculture of China* **5**, 30–31.

Lorenzen K., Struve J. & Cowan V.J. (1997) Impact of farming intensity and water management on nitrogen dynamics in intensive pond culture: a mathematical model applied to Thai commercial shrimp pond. *Aquaculture Research* **28**, 493–508.

Losordo T.M. & Piedrahita R.H. (1991) Modelling temperature variation and thermal stratification in shallow aquaculture ponds. *Ecological Modelling* **54**, 89–226.

Maguire G.B. & Allen G. (1986) Pilot scale studies into New South Wales prawn farming. *Australian Fisheries* **45**, 26–32.

Mantoura R.F.C. (1981) Organo-metallic interactions in natural waters. In: *Marine Organic Chemistry* (ed. by E.K.Duursma & R.Dawson), pp. 179–223. Elsevier, Amsterdam.

Martin J.L.M., Veran Y., Gueloget O. & Pham D. (1998) Shrimp rearing: stocking density, growth impact on sediment, waste out put and their relationships studied through the nitrogen budget in rearing ponds. *Aquaculture* **164**, 135–149.

Masuda K. & Boyd C.E. (1994) Phosphorus fractions in soil and water of aquaculture ponds built on clay utensils at auburn, Alabama. *Journal of the World Aquaculture Society* **25**, 379–395.

Munsiri P., Boyd C.E. & Hajek B.F. (1995) Physical and chemical characteristics of bottom soil profiles in ponds at Auburn, Alabama, and a proposed system for describing pond soil horizons. *Journal of the World Aquaculture Society* **26**, 346–377.

Olsen S.R., Cole C.L. & Watanabe F.S. (1954) Estimation of available phosphorus in soil by extraction with sodium bicarbonate. United States Department of Agriculture Circular, 939.

Paez-Osuna F., Guerrero-Galyan S.R., Ruiz-Fernandez A.C. & Espinoza-angulo R. (1997) Fluxes and mass balances of nutrients in a semi intensive shrimp farm in north western Mexico. *Marine Pollution Bulletin* **34**, 290–297.

Paez-Osuna F., Guerrero-Galvan S.R. & Ruiz-Fernandez C.A. (1999) Discharge of nutrients from shrimp farming to coastal waters of the Gulf of California. *Marine Pollution Bulletin* **38**, 585–592.

Persson G. (1991) Eutrophication resulting from salmonid fish culture in fish and salt waters: Scandinavian experience. In: *Proceedings of the First International Symposium on Nutritional Strategies in Management of Aquaculture Waste* (ed. by C.B.Cowey & C.Y.Cho), pp. 163–185. Fish Nutrition Research Laboratory, University of Guelph, Canada.

Piper C.S. (1966) *Soil and Plant Analysis*. Hans Publishers, Bombay, 368pp.

Praphrutham P. (1985) *Soil Chemistry*. Soil Science Department, Faculty of Agriculture, Kasetsart University, Bangkok, 502pp.

Reeves J.G. (1972) Nitrogen removal: a literature review. *Journal of the Water Pollution Control Federation* **44**, 1895–1908.

Rogers G.L. (1989) Aeration and circulation for effective aquaculture pond management. *Aquacultural Engineering* **8**, 349–355.

Rogers G.L. & Fast A.W. (1988) Potential benefits of low energy water circulation in Hawaiian prawn ponds. *Aquacultural Engineering* **7**, 155–165.

Seitzinger S.P., Nixon S.W. & Pilson M.E.Q. (1984) Denitrification and nitrous oxide production in a coastal marine ecosystem. *Limnology and Oceanography* **29**, 73–83.

Shah M.M.R., Hossain M.Y., Begum M., Ahmed Z.F., Ohtomi J., Rahman M.M., Alam M.J., Islam A.A. & Fulanda B. (2008) Seasonal variations of phytoplanktonic community structure and production in relations to environmental factors of the southwest coastal waters of Bangladesh. *Journal of Fisheries and Aquatic Science* **3**, 102–113.

Smith P.T. (1996) Physical and chemical characteristic of sediments from prawn forms and mangrove habitats on the Clarence River, Australia. *Aquaculture* **146**, 47–83.

Strickland J.D.H. & Parsons T.R. (1972) *A Practical Hand Book of Seawater Analysis*. Bulletin, 167. Fisheries Research Board of Canada, Ottawa, 310pp.

Subbiah B. & Asija G.L. (1956) A rapid procedure for estimation of available nitrogen in soils. *Current Science* **25**, 259–260.

Teichert-Coddington D.R., Martinez D. & Ramirez E. (2000) Partial nutrient budgets for semi-intensive shrimp farms in Honduras. *Aquaculture* **190**, 139–154.

Thakur D.P. & Lin C.K. (2003) Water quality and nutrient budget in closed culture system. *Aquaculture Engineering* **27**, 159–176.

Tookwinas S. & Songsangjinda P. (1999) Water quality and phytoplankton communities in intensive shrimp culture ponds in Kung Krabaen bay, Easter Thailand. *Journal of the World Aquaculture Society* **30**, 36–45.

Wahab M.A., Bergheim A. & Brasten B. (2003) Water quality and partial mass budget in extensive shrimp ponds in Bangladesh. *Aquaculture* **218**, 413–423.

Walkley A. & Black C.A. (1934) An estimation of the Degtijareff method for determining soil organic matter and a proposed modification of the chromic acid titration method. *Soil Science* **37**, 93–101.

Xia L.Z., Yang L.Z. & Yan M.C. (2004) Nitrogen and phosphorus cycling in shrimp ponds and the measures for sustainable management. *Environmental Geochemistry and Health* **26**, 245–251.

Zhang Y.Z., Tan Y.J. & Ou Y.H. (1989) Ecology of aquacultural ponds. In: *China Pond Aquaculture* (ed. by Y.Z.Zhang), pp. 41–119, China Science Press, Beijing.