Aquacult Int (2007) 15:55–66
DOI 10.1007/s10499-006-9068-8

ORIGINAL PAPER

Nitrogen dynamics in the settlement ponds of a small-scale recirculating shrimp farm (*Penaeus monodon*) in rural Thailand

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Received: 13 June 2006 / Accepted: 29 September 2006 /
Published online: 15 November 2006

Abstract Rural shrimp farmers in Thailand are being encouraged to adopt practices that will reduce the quantity and improve the quality of their effluent. A simple and cheap option for small-scale shrimp farmers is to use settlement ponds to store and remediate discharge water before being re-used. We undertook a detailed study of the settlement ponds in a small-scale commercial black tiger shrimp farm typical of rural Thailand. We found that over a 9-week period, following the harvest of one of the two farm production ponds, total nitrogen (TN) concentrations in the water column were reduced by 30%, with the greatest removal (56%) occurring during the fifth week. There was a 10% increase in dissolved organic nitrogen (DON) concentrations during the trial. Sediments were a source of total ammonia nitrogen (TAN), and the re-mineralisation rate was the highest in the first two settlement ponds. Coconut fronds added to two of the four settlement ponds to increase the surface area available for microbial activity were found to provide a site for microbial re-mineralisation of TAN, the photosynthetic uptake of TAN and oxidised nitrogen (NO_x) and nitrification. The water column was a net assimilator of TAN through autotrophic uptake. This study has shown that settlement ponds are capable of reducing water column N concentrations; however, sediment must be managed to reduce re-mineralisation during successive cropping cycles. In addition, coconut fronds were shown to improve N removal, although they should be periodically removed to maintain efficiency.

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Keywords Effluent treatment · Nutrient dynamics · Settlement ponds · Shrimp farming

Abbreviations

DON	Dissolved organic nitrogen
N	Nitrogen
NO_x	Oxidised nitrogen, i.e. nitrate/nitrite
PON	Particulate organic nitrogen
TAN	Total ammonia nitrogen
TN	Total nitrogen
TDN	Total dissolved nitrogen
TSS	Total suspended solids

Introduction

There are well-established environmental concerns in Thailand regarding the discharge of nutrient-laden shrimp farm effluents into coastal waterways (Briggs and Funge-Smith 1994; Phillips et al. 1993). In response to these concerns, the Thailand Department of Fisheries (DOF) has formulated a Code of Conduct (CoC) which outlines how shrimp farmers should attempt to reduce their impact on surrounding environments (refer to: www.thaishrimpquality.com). However, adherence to the CoC requires a concerted research effort to develop management strategies that will help farmers reduce the quantity and improve the quality of their effluent. One such strategy involves the use of semi-closed shrimp culture systems in which harvest water is stored in settlement ponds prior to its re-use for subsequent production cycles.

The use of settlement ponds as a treatment option for shrimp farmers is increasing both in Thailand and other shrimp farming countries. Settlement ponds have been shown to remove particulate material from production effluent but are less effective at removing dissolved nutrients (Chien and Liao 1995; Jackson et al. 2003b). Nutrient retention can be improved through the addition of vertical substrates to settlement ponds (Erler et al. 2004b), however the challenge is to find a cheap substrate that is suitable for small-scale farmers in rural areas.

Most of the research carried out on settlement ponds to date has involved time series monitoring and mass balance calculations to quantify the major processes. Consequently, few detailed studies been conducted on the microbial nutrient transformations occurring in the settlement ponds themselves (Erler et al. 2004a). The objective of the study presented here was to increase our understanding of how settlement ponds function and in doing so to assess their effectiveness as a treatment option for small-scale farmers in rural Thailand. We also evaluated whether the addition of additional surface area to settlement ponds increased their treatment capacity.

Material and methods

The study site was the Sakhorn shrimp farm in the Natub village of Songkhla province, southern Thailand. The farm consisted of four 0.2-ha production ponds surrounded by a series of four settlement ponds (ponds A–D having a combined area of 7000 m^2; see Fig. 1). Coconut fronds (Fig. 2) were suspended in two of the settlement ponds. During the harvest or emergency water exchange, the production ponds emptied into ponds/compartments A and B, which then flowed passively to ponds/compartments C and D. The farmer staggered shrimp production so that two ponds always contained shrimp that were 3 months older than those in the remaining two ponds. The farm attempted to exchange as little water as possible during the 4–6 months of the production period; therefore, retention time in the treatment system could be as long as 3 months. The settlement ponds had enough capacity to store all of the harvest water. The production ponds were filled with water from compartment D when required.

The study was divided into two components. The first monitored nutrients in the settlement ponds over a 9-week period following the harvest of two of the production ponds. The second component involved the collection and incubation of sediment cores, coconut fronds and samples of water from the settlement ponds in order to determine rates of N transformation and oxygen flux.

Nutrient monitoring

Water quality was measured nine times in each of compartments A–D over the 9-week period, with the first samples collected 2 days after the harvest of two production ponds. Water was not exchanged between the production ponds and the settlement ponds during the 9-week monitoring period. Water samples were collected and aliquots taken for PON, TDN, TAN and NO_x analysis. DON was calculated as TDN – TAN – NO_x. Total nitrogen (TN) was calculated as TDN + PON. Total phosphorous was measured in the first half of the study; however, errors in the analytical procedures did not allow us to use the results for the remainder of the study.

For the TAN, NO_x and TDN analyses, water samples were collected, immediately filtered (Whatman GF/C filters), then frozen. TAN, NO_x and TDN were analysed manually using the colorimetric methods described in Grasshoff (1983). PON samples were filtered with pre-combusted 25-mm Whatman GF/F filters which were then

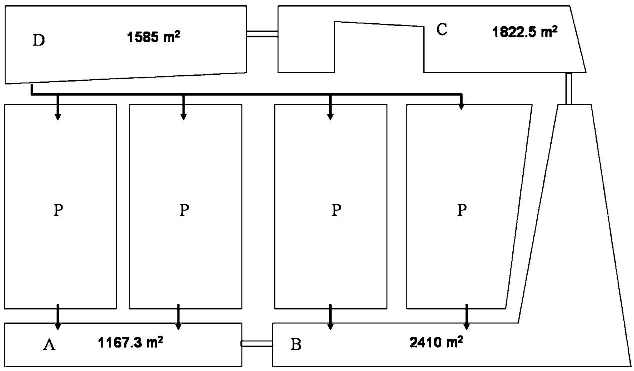


Fig. 1 Study site layout showing four production ponds (*P*) and four settlement ponds/compartments (A, B, C, D). The area of each settlement compartment is given. *Arrows* represent potential water flows

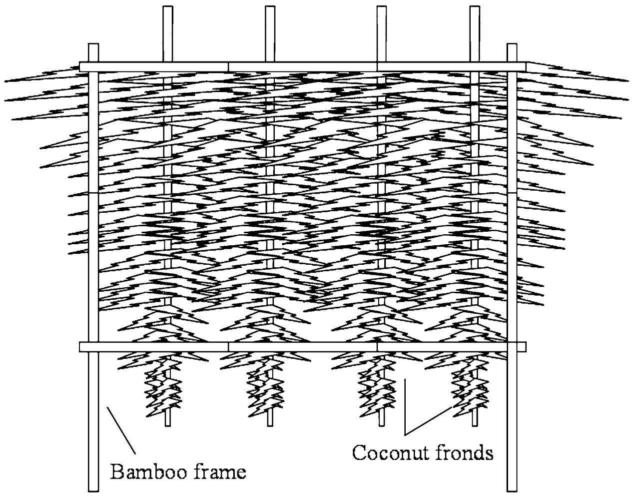


Fig. 2 Illustration of the coconut frond vertical substrate construction

dried at 90°C for 48 h, rolled into a cylinder, wrapped in aluminium foil and analysed for CN content with a LECO CHN analyser.

Total suspended solids (TSS) were collected by filtration of a known volume of sample water on pre-combusted weighed GFF filter papers according to Grasshoff (1983). These were dried and re-weighed to quantify TSS.

Incubation experiments

The incubation trials were conducted during the seventh week of the study. Sediment oxygen demand (SOD) was measured by monitoring DO change over time in eight incubated sediment cores from each settlement pond. The cores were collected by first plunging a PVC pipe (40 mm × 30 cm) into the sediment and then retrieving it. A rubber stopper was placed in the sediment end of the pipe, and the water within was carefully decanted; the core was filled with autoclaved, filtered (Whatman GF/C filter) settlement pond water collected the previous week. The incubation water was spiked with $(NH_4)_2SO_4$ to reach a final concentration of 0.5 mg l^{-1} and KNO_3 to give a final concentration of 0.3 mg l^{-1}. The core was then sealed with another rubber stopper and incubated in a basket suspended in one of the settlement ponds. SOD in all cores was determined after linear regression of repeated DO measurements (5 in 24 h) in both sets of cores. At the completion of the incubation, water samples were collected for TAN and NO_X analysis. Sediment TAN and NO_x flux were calculated from before and after the measurements of core water samples.

Water column oxygen production and consumption were calculated using the electrochemical methods described by Bratvold and Browdy (1998). Briefly, DO change was monitored in light and dark BOD bottles containing filtered (120 µm) settlement pond water suspended in a temperature-controlled tank for 24 h. The tank was illuminated with four 60-W fluorescent tubes. Additional dark BOD bottles containing filtered settlement pond water and the nitrification inhibitor 2-chloro-6(trichloromethyl) pyridine were also incubated under the same conditions (Bratvold and Browdy 1998). For total oxygen production, DO was measured twice over a 24-h period in the light BOD bottles. Total water column oxygen consumption was determined after linear regression of repeated DO measurements (five in 24 h) in the dark BOD bottles with and without nitrification inhibitor. Nutrient flux in all BOD bottles was calculated as the difference in TAN or NO_x concentration over the 24-h incubation period.

The coconut frond incubations were conducted in light and dark BOD bottles filled with the spiked incubation water. Pieces of coconut leaf (roughly 5-cm lengths) from the settlement ponds were then incubated in the BOD bottles. Three control incubations, i.e., BOD bottles with the seawater mix, N-serve and pieces of dry coconut, were also performed.

Following the SOD incubations, core water was emptied and the sediment cores frozen overnight. The following day the sediments were extruded from the cores and the top 1 cm collected. The sediment samples were partially thawed, mixed and two sub-samples taken. These were dried at 110°C for 24 h, homogenised with a mortar and pestle and analysed for CN content with a LECO CHN analyser.

Following the coconut leaf incubations, the water in the bottles was carefully decanted and the pieces of leaf were then removed. The remaining organic material and water from the bottles were collected on a GF/C filter. The organic material on the coconut leaves was scraped off the surface and added to the GF/C filter. The filters were then rinsed under vacuum with freshwater and then dried at 90°C for 48 h, rolled into a cylinder, wrapped in aluminium foil and analysed for CN content with a LECO CHN analyser. The surface area of leaves from all samples was also measured.

Comparisons between parameters in the settlement compartments were made with a one-way analysis of variance. Least significant difference testing (5% confidence limits) was used if significant differences were detected in the ANOVA. The results are presented with standard deviations.

Results

Nutrient monitoring

Over the 9-week studyperiod, the entire settlement pond system reduced the concentration of TN by 30% (from 5.82 ± 2.9 to 4.0 ± 1.29 mg l^{-1}; Fig. 3), with the maximum reduction of 56% (from 5.82 ± 2.9 to 2.55 ± 0.84 mg l^{-1}) occurring during the fifth week of the trial. DON concentrations increased by 10% (from 2.35 ± 0.82 to 2.6 ± 1.15 mg l^{-1}) over the study period after an initial decrease (Fig. 4). PON and TAN concentrations decreased over the study period by 64% (from 0.94 ± 0.62 to 0.33 ± 0.13 mg l^{-1}) and 50% (from 2.09 ± 1.3 to 1.03 ± 0.33 mg l^{-1}), respectively (Fig. 4). The quality of the water generally improved both over time and as discharge water progressed through the treatment ponds (Fig. 4). The reduction in TSS was 59% (from 65.5 ± 15.1 to 26.8 ± 8.2 mg l^{-1}) and occurred during the 5th week of the study.

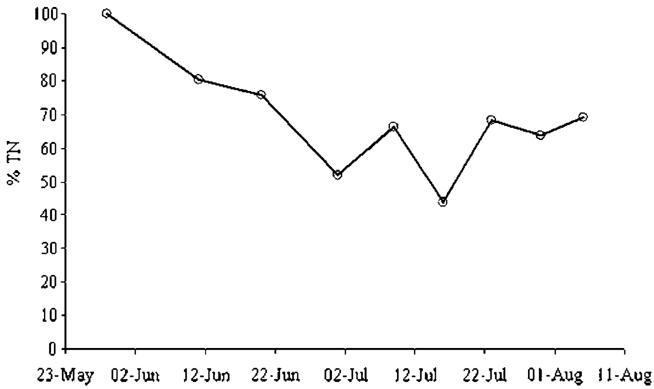


Fig. 3 Percentage of TN (averaged over the entire settlement system) remaining in the water column at the completion of the 9-week study period

The concentrations of TN, DON and TAN, averaged over the entire trial period, were significantly higher ($P < 0.05$) in compartments A and B than compartments C and D (Fig. 5). The percentage reduction in water column TN was similar in compartments A, B and C (38, 35 and 29%, respectively), while there was no change in TN concentration over the study period in compartment D. TSS loads were also significantly higher in compartment A than D over the study period (54.8 ± 13.2 relative to 27.1 ± 6.9 mg l^{-1})

There was significantly more N in the sediments ($P < 0.05$) of compartments A (20.3 ± 8.2 mg m^{-2}) and B (17.7 ± 6.3 mg m^{-2}) than in compartment D (11.1 ± 3.0 mg m^{-2}) (Fig. 6).

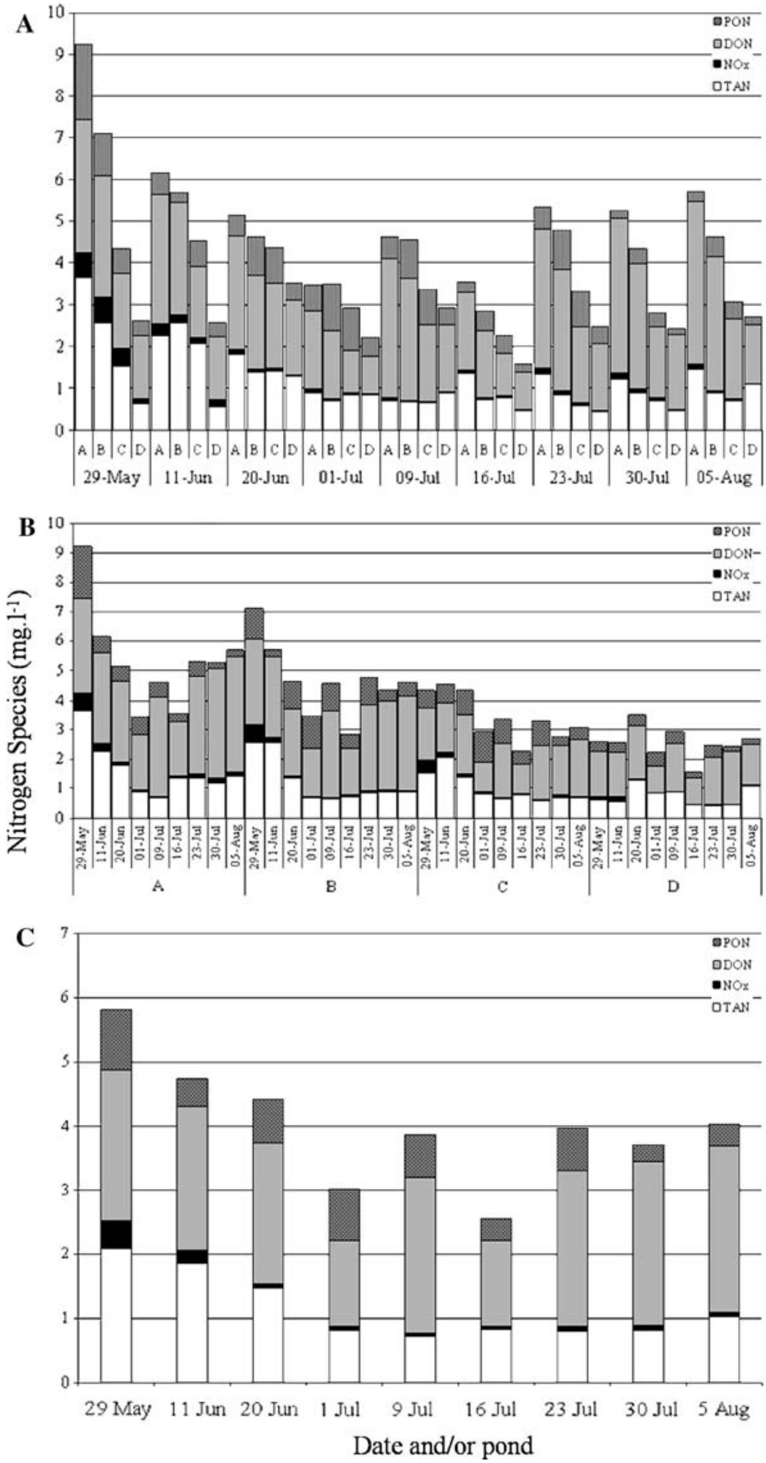
Incubation experiments

There was a net uptake of DO in all settlement ponds (Table 1). The level of SOD appeared to be higher in compartment A than in the other compartments, however the incubation was not sensitive enough to detect a significant difference ($P > 0.05$). TAN was released from all sediments (Table 1) and was significantly higher ($P < 0.05$) in compartments A and B (108 ± 26 and 120 ± 39 mg N m^{-2} day^{-1}) than in compartment D (65 ± 25 mg N m^{-2} day^{-1}). There was an overall uptake of NO_x in all sediments (Table 1).

The coconut leaf substrates in compartments A and C had a net uptake of DO (Table 1). Both nitrification and photosynthesis (presented in units of oxygen production/uptake) were found to occur on the fronds in compartment C (Table 1), while photosynthesis but not nitrification was observed in compartment A (Table 1). There was an overall uptake of TAN and NO_x from the incubation water to the coconut leaf substrates (Table 1).

In the water column there was a net uptake of DO, but photosynthesis was still active in water from all compartments (Table 1). There was a decreasing trend in the

Fig. 4 Concentrations of nitrogen species in the water column of the settlement system over the study period. (**a**) Concentration in each compartment at each sampling date, (**b**) change in concentration in each compartment over time, (**c**) average concentration over entire study in each compartment ▶


A
B
C
PON
DON
NOx
TAN
Nitrogen Species (mg.l-1)
29-May
11-Jun
20-Jun
01-Jul
09-Jul
16-Jul
23-Jul
30-Jul
05-Aug
29 May
11 Jun
20 Jun
1 Jul
9 Jul
16 Jul
23 Jul
30 Jul
5 Aug
Date and/or pond

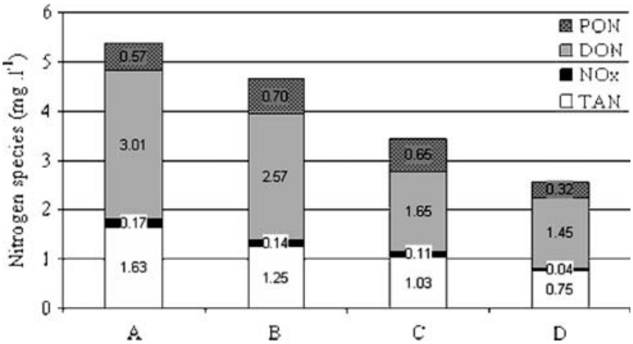


Fig. 5 Concentrations of nitrogen species in the water column of the individual settlement compartments averaged over the entire trial period

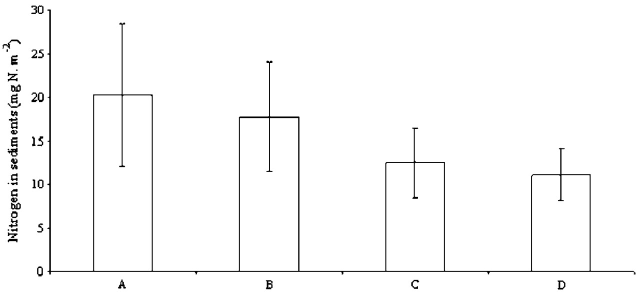


Fig. 6 Concentrations of TN (±standard deviation) in the sediments from each treatment compartment

rate of photosynthesis from compartment A to D; however no significant differences were detected. There was a small net uptake of TAN in the water column from each compartment (Table 1).

Discussion

Shrimp farmers in Thailand are facing increased pressure to reduce the quantity of nutrients being discharged into the environment. In rural Thailand the majority of farms are small-scale (1–2 ha) operations, with the farmers having little understanding of the complex nutrient cycling that occurs in their ponds. In an effort to assist farmers to better manage their coastal resources while meeting environmental regulations regarding the discharge of nutrient-laden shrimp farm effluents into coastal waterways, the Thailand DOF has developed a CoC for shrimp farmers. One objective of the CoC is to encourage farmers to re-cycle production water through settlement ponds. However, only a few detailed studies have been carried out that describe what is actually happening in the settlement ponds.

In this study we found that the quality of the discharged production pond water, in terms of TN concentration, improved over the 9-week trial period; this was particularly so during the first 5 weeks after discharge. Jackson et al. (2003b) showed that the major removal pathway for discharged nutrients in treatment ponds is

Table 1 Flux of oxygen and nutrients in and out of the sediments, the coconut fronds and the water column during the seventh week of the study

Compartments	Sediment				
	SOD (g DO m^{-2} day^{-1})	TAN flux (mg N m^{-2} day^{-1})	NO_x flux (mg N m^{-2} day^{-1})		
A	–0.83 ± 0.25	108 ± 26	–19.3 ± 7.1		
B	–0.67 ± 0.21	120 ± 39	–16.2 ± 5.4		
C	–0.67 ± 0.21	93 ± 28	–11.0 ± 4.2		
D	–0.53 ± 0.18	65 ± 25	–19.9 ± 7.3		
	Coconut leaf DO flux (g DO m^{-2} day^{-1})	Photosynthesis (g DO m^{-2} day^{-1})	Nitrification (g DO m^{-2} day^{-1})	TAN flux (mg N m^{-2} day^{-1})	NO_x flux (mg N m^{-2} day^{-1})
A	–0.05 ± 0.02	0.03 ± 0.01	0	–5.11 ± 1.31	–2.18 ± 0.70
C	–0.08 ± 0.03	0.05 ± 0.02	0.02 ± 0.01	–1.30 ± 0.42	–1.96 ± 0.63
	Water column DO flux (g DO l^{-2} day^{-1})	Photosynthesis (mg DO l^{-2} day^{-1})	TAN flux (mg N l^{-2} day^{-1})	NO_x flux (mg N l^{-2} day^{-1})	
A	11 ± 3.8	0.27 ± 0.08	–0.47 ± 0.15	–0.28 ± 0.08	
B	21 ± 7.9	0.20 ± 0.07	–1.21 ± 0.33	–0.30 ± 0.10	
C	2 ± 0.9	0.15 ± 0.04	–0.49 ± 0.15	–0.17 ± 0.06	
D	60 ± 25	0.11 ± 0.03	–0.34 ± 0.10	–0.13 ± 0.04	

[a] All values are the means of multiple measurements ± the standard deviation

through the settlement of particulate material. This was observed in our study for PON, which showed the largest change in concentration during the 9-week trial. TSS concentrations were also significantly reduced over the study period. However, the removal of particulate material from the water column is only a temporary benefit.

Jackson et al. (2003b) also showed that the removal of particulate material eventually leads to the mineralisation of organic N and the release of dissolved N to the water column. In our study we observed that DON concentrations increased in the water column. DON is the most dominant form of dissolved N in shrimp farming (Burford and Williams 2001; Jackson et al. 2003a) and is the first product of PON breakdown by microbes (Fenchel et al. 1998). As DON is eventually broken down by microbes in an oxygen-consuming process that releases TAN (Hargreaves 1998), increasing concentrations of DON will ultimately lead to high TAN production and low DO in the settlement ponds. In this study we found the sediments in the settlement ponds to be anaerobic below a depth of 2–3 cm. However, because settlement ponds are not dried and tilled at the end of each crop, there is a build-up of organic material from previous production cycles.

We also observed that the initial reduction in TAN concentrations was followed by an increase, most likely as organic matter became mineralised in the sediments. The incubation trials clearly demonstrated that TAN was being released into the water column from the sediments. Furthermore, the release of TAN was greatest in the first settlement compartments where the level of sediment N was the highest.

The net reduction in TN shows that the settlement system in our study was effective at improving water quality, although we also demonstrated that the settlement of particulate material is followed by the release of dissolved N into the water column. Settlement ponds are therefore limited in their capacity to remediate effluent and merely recycle nutrients rather than remove them. In order to improve the nutrient removal capacity of settlement ponds farmers have a number of options. The simplest is to periodically remove the settlement pond sludge or to dry ponds between crops. Although this may initially be a costly exercise, farmers can avoid potential crop losses by maintaining good water quality. We found that the settlement ponds were capable of holding and treating the water following the harvest of two of the four production ponds. However without regular removal of sludge, water quality in the settlement ponds will most likely diminish over time.

Another option is to trap nutrients on substrates that can be easily removed and disposed of. In this study we attempted to use coconut fronds as a mechanism to absorb nutrients. The principle pathways are the uptake of TAN and NO_x by benthic autotrophs and the uptake of DON for bacterial growth. We found that coconut fronds were effective at assimilating TAN, with the most likely pathway being photosynthetic uptake. We also found that nitrification occurred. This process transforms TAN to NO_x and does not represent a removal of N but rather reduces the toxicity of the water for re-use and facilitates denitrification. Overall the net result of the microbial processes on the coconut fronds was a net uptake of both TAN and NO_x. This is encouraging as it demonstrates that increasing the available surface area can facilitate the uptake of N. However, like sediments, coconut fronds have a limited capacity to absorb nutrients and therefore must be periodically removed. Coconut fronds also do not assimilate nutrients for growth, hence a living substrate, such as mangroves, could be considered.

The use of artificial wetlands, including those containing mangrove plants, to remove nutrients has been investigated by a number of workers (Rivera-Monroy et al. 1999; Sansanayuth et al. 1996; Thimdee et al. 2003; Tilley et al. 2000). While these systems can effectively reduce nutrients, they can be unpredictable (Gautier et al. 2001) and require large tracts of land [estimated 22 ha of mangrove wetland to treat each hectare of shrimp pond (Robertson and Phillips 1995)]. In Thailand, some farmers share a wetland to scrub nutrients; however the effectiveness of this practice has not been quantified. An additional benefit of mangrove or wetland systems is the reduction in potential pathogens and viruses (Cameron et al. 2003; Decamp and Warren 2000; Gersberg, et al. 1987). This is particularly important when farmers are continuously re-using settlement pond water.

In summary, we have detailed the nitrogen transformations occurring in the settlement ponds of a small-scale, semi-closed shrimp farm in rural Thailand. The settlement ponds were found to assist in the removal of N from the effluent of two of the four production ponds. However, the sediment was a site of active TAN remineralisation, and we suggest that periodic removal of settled sediments would prolong the life of the settlement ponds. We also found that the addition of coconut fronds to the settlement ponds, as a means of increasing the surface area for microbial growth and nutrient uptake, can be a useful technique for remediating production effluent. Again, however, we suggest that fronds must be periodically replaced to maintain efficiency. We also encourage further investigation into the use of artificial wetlands for subsequent treatment of effluent from shrimp farms.

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