

Water Quality Control in Tilapia Closed Culture System Using Filter Feeding Freshwater Clam (*Pilsbryconcha exilis compressa*)

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ABSTRACT

Evaluation of the possibility of using filter feeder freshwater clam, *Pilsbryconcha exilis compressa*, for the improvement of water quality in tilapia closed culture system was evaluated in a 2 month outdoor tank experiment. Tilapia was cultured in a closed system with and without freshwater clam for two months. Average values of turbidity, chlorophyll *a*, total particulate matter, particulate organic matter, particulate nitrogen and particulate phosphorus in tilapia-freshwater clam treatment were significantly lower ($P \leq 0.05$) than those in tilapia only treatment. However, there was no significant difference ($P \geq 0.05$) between the average values of total ammonia nitrogen from the two treatments. Average values of turbidity, chlorophyll *a*, total particulate matter, particulate organic matter, particulate nitrogen, particulate phosphorus and total ammonia nitrogen were 6.0 ± 11.6 NTU, 24.3 ± 15.8 $\mu\text{g/L}$, 7.9 ± 6.1 mg/L, 6.4 ± 5.1 mg/L, 1.419 ± 1.342 mg/L, 0.087 ± 0.058 mg/L, and 0.473 ± 0.267 mg/L, respectively, in tilapia-freshwater clam treatment. In comparison, tilapia only treatment yielded average values of 22.0 ± 11.6 NTU, 324.1 ± 222.2 $\mu\text{g/L}$, 42.5 ± 26.2 mg/L, 34.1 ± 21.1 mg/L, 5.291 ± 5.634 mg/L, 0.305 ± 0.189 mg/L and 0.599 ± 0.752 mg/L, respectively. According to the results of this experiment, *P. exilis compressa* is effective in removing particulate matter, particulate nitrogen, phytoplankton and particulate phosphorus from water in tilapia culture tanks resulting in the decrease of 72.7% of turbidity, 81.4% of total particulate matter, 81.2% of particulate organic matter, 92.5% of chlorophyll *a*, 73.2% of particulate nitrogen and 71.5% of particulate phosphorus. There was no significant difference ($P \geq 0.05$) between production rate, survival rate and growth rate of tilapia in both treatments.

Keywords: tilapia, freshwater clam, *Pilsbryconcha exilis compressa*, closed culture system

INTRODUCTION

In intensive fish culture systems, particulate organic matter consisting of

uneaten feed, fish feces, dead and living plankton and other microbes accumulate and become major causes of deterioration of water quality in culture ponds and in water

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bodies receiving pond effluents. Total estimated solid wastes output of tilapia was 331-364 kg tonne⁻¹ of feed consumed or 423-496 kg tonne⁻¹ of fish produced. Solid nitrogen waste output was 7.6-8.3 kg tonne⁻¹ of fish produced and solid phosphorus waste output was 5.6-5.9 kg tonne⁻¹ of fish produced. Dissolved N waste output was 40.9-46.2 kg tonne⁻¹ of fish produced and dissolved phosphorus waste output was 4.2-5.0 kg tonne⁻¹ of fish produced (Chowdhury *et al.*, 2013). Part of these solid wastes suspend in water while another part settle down to the bottom. In intensive culture of tilapia, disturbance by fish generate resuspension of bottom sediment. Microalgal growth stimulated by nutrients released from fish waste and uneaten feed also contributes to particulate matter concentration in water in the form of phytoplankton. Because of their potential negative effect, removal of these solids is commonly practiced as a means to manage water quality in water recirculation fish culture systems.

Filter feeding bivalves are very effective in removing suspended solids from water. According to Bayne and Newell (1983) and Bayne and Hawkins (1992), bivalves can clear seston particles greater than 3- μ diameter from water column with high efficiency. Prior to ingestion, filtered particles are sorted, less nutritious and excess particles are immediately rejected as pseudofeces, while ingested material is digested and the remains are excreted as feces. Feces and pseudofeces form biodeposits which settle down to the pond bottom.

Bivalves can utilize particulate organic wastes from aquaculture farms as feed. Excess particulate fish feed released from salmon farms was effectively captured and absorbed by blue mussels, *Mytilus edulis* (MacDonald *et al.*, 2011). Greater increases in shell height and monthly instantaneous growth rates of oysters suspended at a salmon farm were also observed (Jones and Iwama, 1991). Culturing bivalves with fish or shrimp was reported to be able to reduce nutrients and chlorophyll *a* concentration in fish ponds (Soto and Mena, 1999; Cordova and Martinez-Porchas, 2006) and reduce nutrient concentration in effluents (Sterling and Okumus, 1995 and Neori *et al.*, 2004 cited by Gifford *et al.*, 2005). Filtration by freshwater bivalves such as the Asiatic clam (*Corbicula fluminea*) and zebra mussel (*Dreissenia polymorpha*) reduced water turbidity and phytoplankton concentration and improved water quality in river estuaries and lakes (Phelps, 1994; Leach, 1993).

The freshwater clam, *P. exilis compressa*, is a bivalve commonly found in standing and running waters in Thailand. The clams are collected from natural beds and sold in local markets for human consumption. In this study, we evaluated the possibility of using *P. exilis compressa* to improve water quality of intensive closed culture systems of tilapia by studying the effect of freshwater clam-tilapia co-culture on particulate matter, particulate organic matter particulate nitrogen, particulate phosphorus, chlorophyll *a* concentration and other related water quality parameters and production and growth of tilapia.

MATERIALS AND METHODS

The experiment was conducted in a water recirculation culture system without water exchange using 75 x 155 x 60 cm (W x L x H) fiberglass tank. The tank was partitioned into two compartments for tilapia and bivalve by a cement board. The size of the bivalve compartment was 75 x 40 x 60 cm height while the fish compartment was 75 x 115 x 60 cm. In the fish compartment, a 90 cm long baffle was placed perpendicular to the tank partition to direct water flow. The tanks were filled with water at a depth of 50 cm. Water from the lower part of the

bivalve section was airlifted into the fish section through two 0.5 inch PVC tubes, flowing around the center baffle back to the bivalve section through a plastic screen on a rectangular hole (15 x 30 cm) on another side of the partition. The hole was cut at 10 cm above tank bottom level (Figure 1). Recirculation rate of water through the airlift system was 4 L/min. Two plastic trays (35 x 55 x 15 cm height) were placed on a PVC stand in the bivalve section. The first tray was placed 10 cm above tank bottom and the second tray was 10 cm above the first tray. Two air stones were placed on the tank bottom under the trays. Stocking rate

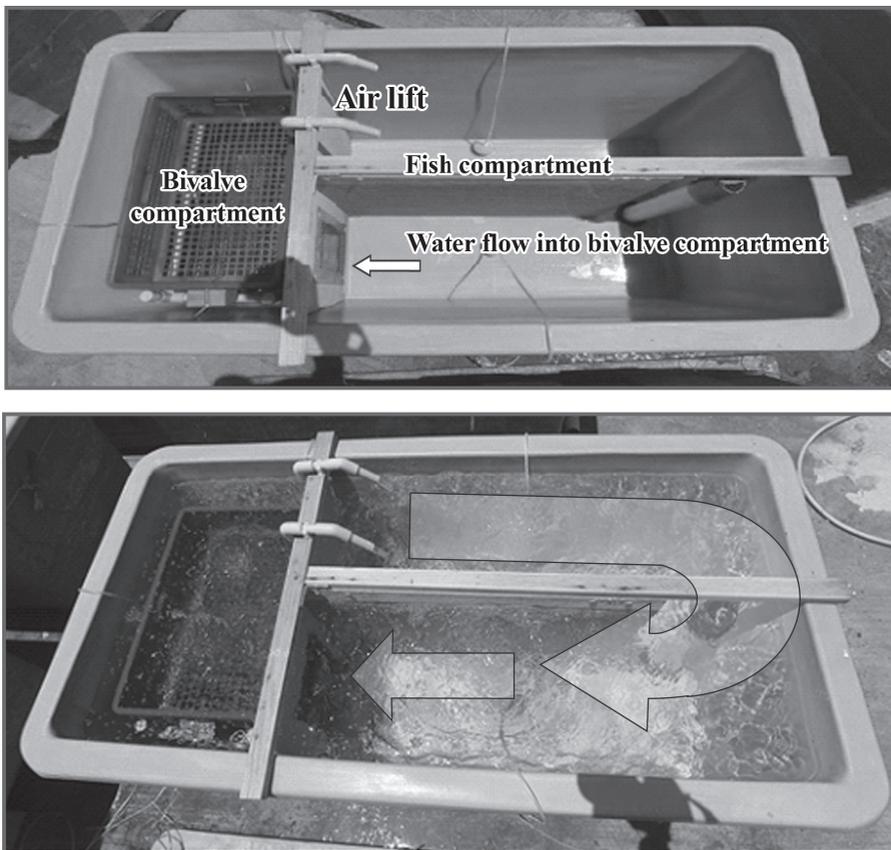


Figure 1. Experimental tanks and flow pattern of water between fish compartment and bivalve compartment.

of the bivalve was 3.0 kg/tank (1.5 kg/tray). A preliminary test of the filtering capacity of the clam was conducted to estimate appropriate stocking rate of the clam. Average weight and average number of the bivalves stocked per tank were 18.54 g and 162, respectively. Two air stones were placed in the fish compartment on both sides of the center baffle. Water used in this experiment was stored tap water mixed with water from the tilapia pond. Total volume of water in each experimental tank was 0.58 m³. Tilapia was stocked at the rate of 10 fish/tank. Initial weight of fish was between 70 and 90 g. Water volume in the fish compartment was 0.43 m³ while that in the bivalve compartment was 0.15 m³. Initial stocking rate was 0.76-0.81 kg /0.58 m³ or 1.3-1.4 kg/ m³ of total water volume for tilapia and 3.0 kg/ 0.58 m³ or 5.17 kg/ m³ of total water volume for freshwater clam.

The experiment consisted of three replications. Three similar tanks were also set up for control with the same stocking rate of tilapia without bivalve in plastic baskets. Fish were fed twice daily to satiation with 30% protein commercial pelleted feed. Experimental period was 8 weeks. Water samples were collected once a week from the fish compartment to measure dissolved oxygen (DO), pH, turbidity, total particulate matter (TPM), particulate organic matter (POM), particulate nitrogen (PN), particulate phosphorus (PP), chlorophyll *a*, and total ammonia nitrogen (TAN). Experimental animals were counted and weighed at the beginning and the end of the experiment to calculate survival rate and growth rate.

TPM was analyzed by filtering the samples through GF/C glass fiber filter, dried in an oven at 103-105°C overnight then weighed (APHA *et al.*, 2005). Then the samples were transferred to a muffle furnace and ignited at 450°C for 4 h, cooled in a dessicator and weighed to obtain POM fraction which was equal to the weight lost after 4 h in muffle furnace. PN was analyzed using Kjeltac 1035 analyzer unit, Tecator: Digestion block, Foss, Model 2520. PP was analyzed spectrophotometrically after percholic acid digestion. Chlorophyll *a* content was analyzed using spectrophotometric determination method after extracting with acetone (APHA *et al.*, 2005). Turbidity was measured by Turbidimeter HACH 2100Q. DO was measured by YSI Dissolved oxygen meter. pH of water was measured by YSI pH meter. TAN was measured by Phenate Method (APHA *et al.*, 2005). Data were analyzed for mean and standard deviation and compared by t-test.

RESULTS AND DISCUSSION

According to the results of this experiment, the introduction of freshwater clam, *P. exilis compressa* into closed culture system of tilapia can improve water quality in the culture system by reducing water turbidity, phytoplankton, total particulate matter, particulate organic matter, particulate nitrogen and particulate phosphorus (Table 1, Figure 2). Average values of turbidity, chlorophyll *a*, total particulate matter, particulate organic matter, particulate nitrogen and particulate phosphorus in

Table 1. Comparison of average values of turbidity, TPM, POM, chlorophyll *a*, TAN, PN, PP, DO and pH of water in tanks culturing tilapia and tilapia + freshwater clam (mean±S.D.).

Water quality parameter	Initial	Tilapia	tilapia - bivalve	% decrease
Turbidity (NTU)	3.1	22.0±11.6 ^a	6.0±4.3 ^b	72.7
TPM (mg/L)	5.6	42.5±26.2 ^a	7.9±6.1 ^b	81.4
POM (mg/L)	5.1	34.1±21.1 ^a	6.4±5.1 ^b	81.2
Chlorophyll <i>a</i> (µg/L)	11.8	324.1±222.2 ^a	24.3±15.8 ^b	92.5
TAN (mg/L)	0.022	0.599±0.752 ^a	0.473±0.267 ^a	-
PN (mg/L)	0.199	5.291±5.634 ^a	1.419±1.342 ^b	73.2
PP (mg/L)	0.011	0.305±0.189 ^a	0.087±0.058 ^b	71.5
DO (mg/L)	8.0	7.1±0.64 ^a	7.1±0.60 ^a	-
pH	8.2	8.3±0.30 ^a	8.0±0.17 ^b	-

Average values denoted by different superscript in each parameter are statistically significant ($P \leq 0.05$).

tanks with tilapia-freshwater clam were significantly lower ($P \leq 0.05$) than those in tanks with tilapia only. Average values of turbidity, chlorophyll *a*, total particulate matter, particulate organic matter, particulate nitrogen and particulate phosphorus in treatment with tilapia and freshwater clam were 6.0 NTU, 24.3 µg/L, 7.9 mg/L, 6.4 mg/L, 1.419 mg/L and 0.087 mg/L, respectively, compared to average values of 22.0 NTU, 324.1 µg/L, 42.5 mg/L, 34.1 mg/L, 5.291 mg/L and 0.305 mg/L of turbidity, chlorophyll *a*, total particulate matter, particulate organic matter, particulate nitrogen and particulate phosphorus, respectively, in treatment with tilapia only (Table 1). In tilapia only treatment, weekly average values were between 5.2 and 31.0 NTU for turbidity, 42.1 and 658.6 µg/L for chlorophyll *a*, 8.1 and 75.8 mg/L for total particulate matter, 8.1 and 60.1 mg/L for particulate organic matter, 0.223 and 18.445 mg/L for particulate nitrogen and 0.049 and 0.631 mg/L for particulate phosphorus (Figure 2). Weekly average

values of turbidity, chlorophyll *a*, total particulate matter and particulate organic matter steadily increased from week 1 to week 4. Then average values of turbidity and chlorophyll *a* continuously decreased from week 5 to week 8 while average values of total particulate matter and particulate organic matter fluctuated a little bit from week 5 to week 6 and then steadily decreased to week 8. Average values of particulate nitrogen increased from week 1 to week 5 then continuously decreased from week 6 to week 8. Average values of particulate phosphorus decreased a little bit from week 1 to week 2 then continuously increased to week 4 before steadily decreased from week 5 to week 8 (Figure 2).

In tilapia-freshwater clam treatment, weekly average values were between 1.4 and 13.2 NTU for turbidity, 7.5 and 53.4 µg/L for chlorophyll *a*, 1.6 and 19.2 mg/L for total particulate matter, 1.4 and 14.7 mg/L for particulate organic matter, 0.105 and

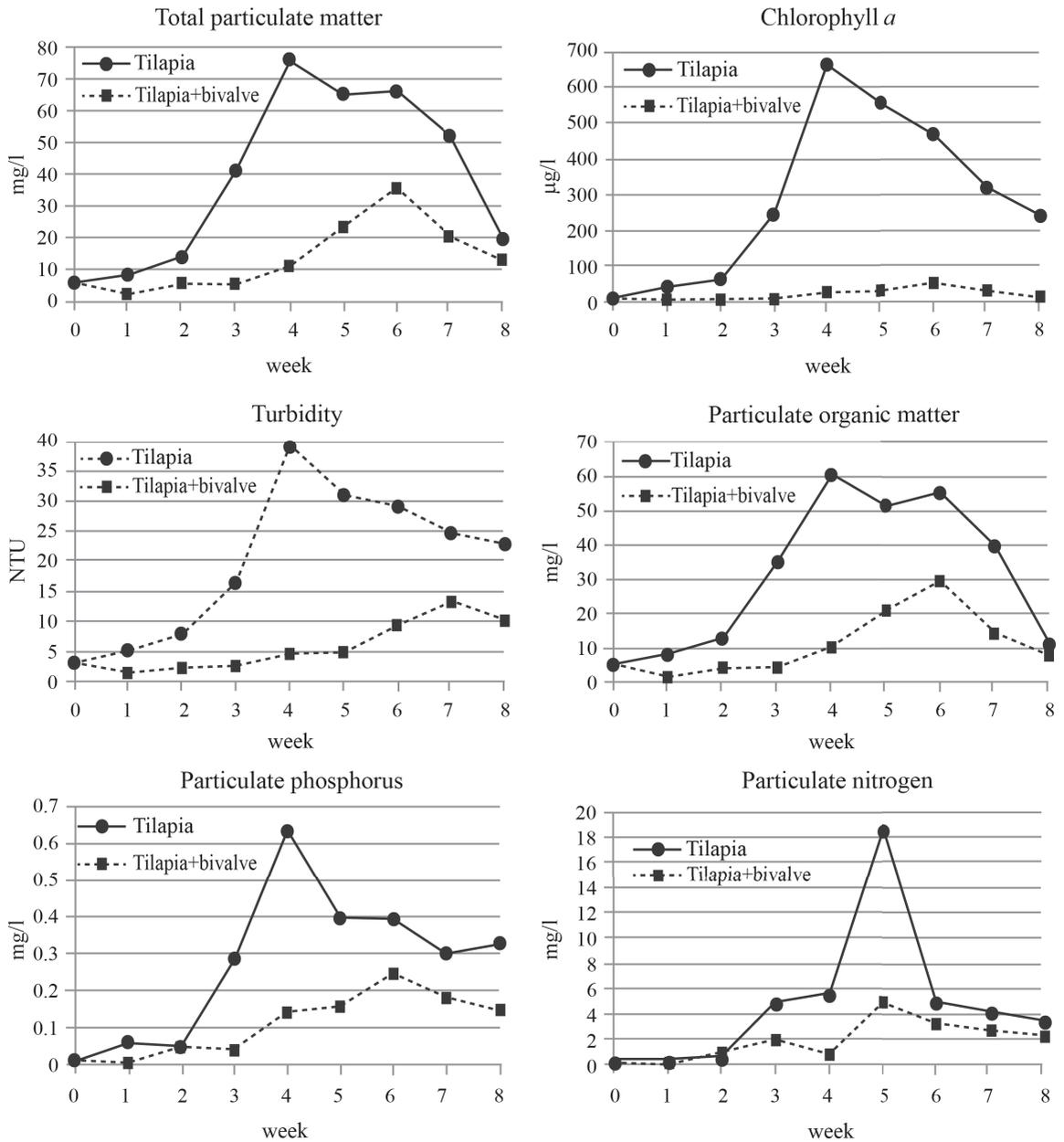


Figure 2. Average values of total particulate matter, chlorophyll *a*, turbidity, particulate organic matter, particulate phosphorus and particulate nitrogen in tanks culturing tilapia and tilapia-freshwater clam.

4.167 mg/L for particulate nitrogen and 0.004 and 0.173 mg/L for particulate phosphorus (Figure 2). Average values of these water quality parameters slightly decreased from initial values to the end of the first week. Then average values of these parameters except particulate nitrogen exhibited an increasing trend from week 1 to week 6, then continuously decreased to week 8. Average values of particulate nitrogen increased from week 1 to week 3, dropped down a little bit in week 4, increased again in week 5, and then steadily decreased to week 8 (Figure 2).

The increase of turbidity, total particulate matter, particulate organic matter, particulate nitrogen and particulate phosphorus to maximum values in week 4 and week 5 in tilapia only treatment likely resulted from the growth of phytoplankton which peaked during that period as indicated by the chlorophyll *a* content. The decrease in turbidity, particulate matter, particulate organic matter, chlorophyll *a*, particulate nitrogen and particulate phosphorus in both treatments in the last two weeks likely resulted from the heavy rain which caused the overflow of water from the culture tanks during that period.

Results of this study indicated that freshwater clam could reduce turbidity, particulate matter, particulate organic matter, chlorophyll *a*, particulate nitrogen and particulate phosphorus in water in intensive tilapia closed culture systems by 72.7, 81.4, 81.2, 92.5, 73.2 and 71.5%, respectively. Sterling and Okumus (1995) and Neori, *et al.* (2004) cited by Gifford, *et al.* (2005) reported that culturing oyster or mussel with

salmon could reduce nutrient concentration in the effluent. Soto and Mena (1999) reported significant decreases of chlorophyll *a* and total phosphorus in the closed culture system of juvenile salmon with freshwater mussel (*Diplodon chilensis*) in outdoor tanks compared to controls without bivalves. Chlorophyll *a* concentration in tanks with mussel was reduced by two orders of magnitude (from ~ 300 to 3µg/L) compared to tanks without mussels. In addition, total phosphorus was reduced by about one order of magnitude after days 18 to 39. Significantly, the decrease in chlorophyll *a* was also reported by Cordova and Martinez-Porchas (2006) in earthen pond polyculture of Pacific white shrimp, *Litopenaeus vannamei*, giant oyster, *Crassostrea gigas*, and black clam, *Chione fluctifraga*. The decrease in total particulate matter, particulate organic matter, particulate nitrogen and particulate phosphorus in the tilapia-freshwater clam treatment indicated the possibility of using freshwater clams to improve water quality in intensive tilapia culture systems by reducing particulate organic matter and particulate nutrients from the water column. Filtering suspended particles by bivalves was also reported to have a positive effect concerning aquatic animal disease control. According to Tendencia (2007), the polyculture of green mussels (*Perna viridis*), brown mussel (*Perna Indica*), or oyster (*Crassostrea* sp.) with shrimp could be used to control shrimp disease caused by luminous bacteria.

There was no statistically significant difference ($P \geq 0.05$) between overall average values of total ammonia nitrogen in tilapia only treatment and tilapia-freshwater clam

treatment. Average values of total ammonia were 0.599 ± 0.752 mg/L in tilapia only treatment and 0.473 ± 0.267 mg/L in tilapia+freshwater clam treatment (Table 1). Weekly average values (from week 1 to week 8) were between 0.031 and 1.918 mg/L for of total ammonia nitrogen in tilapia only treatment and between 0.092 and 0.790 mg/L in tilapia+freshwater clam treatment (Figure 3).

Martinez-Cordova and Martinez-Porchas (2006) reported that polyculture of Pacific white shrimp, *Litopenaeus vannamei*,

giant oyster, *Crassostrea gigas*, and black clam, *Chione fluctifraga* in earthen ponds resulted in a significant decrease in total ammonium nitrogen. Soto and Mena (1999) also reported that freshwater mussel, *D. chilensis*, reduced concentration of ammonia about one order of magnitude after from days 18 to 39 in closed culture system of salmon in an outdoor tank experiment. In contrast, the result in this present study indicated that the introduction of freshwater clam (*P. exilis compressa*) into closed culture system of tilapia did not result in the

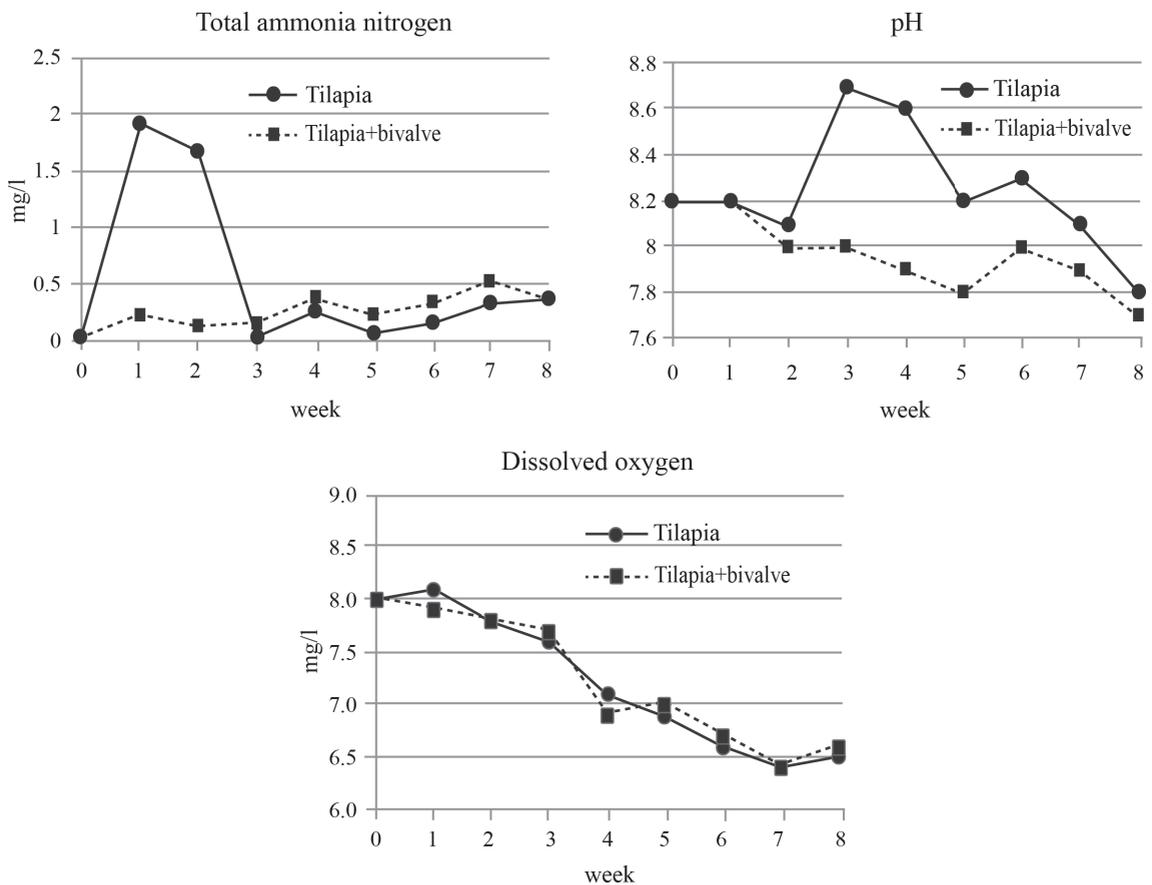


Figure 3. Average values of total ammonia nitrogen, pH and dissolved oxygen of water in tanks culturing tilapia and tilapia-freshwater clam.

decrease of overall average total ammonia nitrogen concentration in the water. However, in the first two weeks, average values of total ammonia nitrogen in tilapia-freshwater clam treatment were significantly lower ($P \leq 0.05$) than those in the tilapia only treatment (Figure 3). A high concentration of total ammonia in tilapia only treatment in the first two weeks was likely a result of the combined effects of the accumulation of particulate organic matter which decomposed and released ammonia and the low density of phytoplankton to absorb the ammonia. The average concentration of particulate organic matter at the end of the first week was 8.1 mg/L in tilapia only treatment compared to the average concentration of 1.4 mg/L in tilapia+freshwater clam treatment (Figure 2). The Average concentration chlorophyll *a* at the end of first week in tilapia only treatment was 42.1 $\mu\text{g/L}$ compared to 3.0 $\mu\text{g/L}$ of average concentration of chlorophyll *a* in tilapia+freshwater clam treatment. The dense algal bloom in tilapia only treatment beginning from the third week until the end of the experiment in which chlorophyll *a* content rose to a concentration level of 240.7-658.6 $\mu\text{g/L}$ resulted in steeply decreasing total ammonia nitrogen to the level of 0.031-0.360 mg/L (Figures 2 and 3).

There was no statistically significant difference ($P \geq 0.05$) between overall average of dissolved oxygen in tilapia only treatment and tilapia+freshwater clam treatment. Average values of dissolved oxygen was 7.1 \pm 0.64 mg/L in tilapia only treatment and 7.1 \pm 0.60 mg/L in tilapia+freshwater clam treatment (Table 1). Weekly average values from week 1 to week 8 of dissolved oxygen were between 6.4 and 8.1 mg/L for tilapia

only treatment and between 6.4 and 8.0 mg/L in tilapia+freshwater clam treatment (Figure 3).

Overall average of water pH in tilapia only treatment was 8.3 \pm 0.30 which was significantly higher ($P \leq 0.05$) than the average value of 8.0 \pm 0.17 in tilapia+freshwater clam treatment (Table 1). Weekly average values from week 1 to week 8 of water pH were between 7.8 and 8.7 in tilapia only treatment and between 7.7 and 8.2 in tilapia and freshwater clam treatment (Figure 3).

Filtration of particulate matter by freshwater clam removed large quantities of phytoplankton and other particulate matter (uneaten feed, fish feces and dead algal cells) from the water column resulting in the reduction of chlorophyll *a*, particulate organic matter, particulate nitrogen, particulate phosphorus and turbidity of water. In tilapia-freshwater clam treatment, filtration of particulate matter by the bivalve kept the weekly average total particulate matter and chlorophyll *a* content less than 19.2 mg/l and 53.4 $\mu\text{g/l}$, respectively (Figure 2). Large amounts of feces and pseudofeces produced by the freshwater clam were found accumulated at the bottom of the bivalve compartment. In the tilapia only treatment in which no freshwater clam was put into the bivalve baskets, most of the particulate matter also settled down in the bivalve compartment. Bottom sediment disturbance by tilapia generated resuspension of settleable particulate matter which was carried out from the fish compartment with water that flow into the bivalve compartment resulting in small amounts of sediment accumulating in the fish compartments of both treatments.

There were no significant differences in growth performance of fish in both treatments (Table 2). The average total weight at harvest, size at harvest, increase in bodyweight, survival rate and feed conversion ratio of tilapia in the two treatments were not significantly different ($P \geq 0.05$). In the tilapia only treatment, average total weight of fish at harvest was 2726.7 ± 64.3 g per tank which was equivalent to a production rate of 4.70 kg/m^3 . The average body weight of fish at harvest was 272.7 ± 6.4 g. The average increase in body weight was 196.5 ± 7.4 g/fish/8 weeks and the average feed conversion ratio was 1.28 ± 0.04 . In the tilapia+freshwater clam treatment, the average total weight of fish at harvest was 2686.7 ± 167.7 g per tank which was equivalent to a production rate of 4.63 kg/m^3 . The average size of fish at harvest was 268.7 ± 16.8 g. The average increase in body weight was 193.0 ± 20.2 g/fish/8 weeks and the average feed conversion ratio was 1.30 ± 0.06 . Survival rate of experimental fish was 100% in both treatments.

The production rate in the tilapia only treatment indicated that at least 4.70 kg/m^3 of fish can be produced in closed culture systems without water exchange. However, it has to be noted that part of the culture tank was partitioned and acted as a settling area for settleable particulate matter. Higher production rate of tilapia using this closed culture system may also be expected by co-culture of freshwater bivalve with tilapia using higher stocking rate of fish. However, more investigations are needed in order to obtain proper stocking density of bivalve and fish for optimum production.

Average survival rate of freshwater clam was $65.0 \pm 23.3\%$. High mortality of the clam probably resulted from toxic metabolites generated from the decomposition of large amounts of organic sediment accumulating underneath the bivalve trays. It is likely that this is the main cause of slow growth rate and high mortality of the freshwater clam. The average growth rate of the clam was 0.68 ± 0.24 g/individual/8 weeks. Periodic

Table 2. Survival rate, weight gain, final body weight and feed conversion ratio of tilapia cultured with and without freshwater clam (mean \pm S.D.).

Treatment	Tilapia	tilapia - bivalve
Initial number	10	10
Initial total weight (g)	761.7 ± 53.9	756.7 ± 40.4
Average initial weight (g/fish)	76.2 ± 5.4	75.7 ± 4.0
Survival rate (%)	100	100
Final total weight (g/tank)	2726.7 ± 64.3^a	2686.7 ± 167.7^a
Final average weight (g/fish)	272.7 ± 6.4^a	268.7 ± 16.8^a
Average weight gain (g/fish/8 week)	196.5 ± 7.4^a	193.0 ± 20.2^a
Average weight gain (%)	259.1 ± 25.4^a	256.4 ± 39.7^a
Feed conversion ratio	1.28 ± 0.04^a	1.30 ± 0.06^a

Average values denoted by different superscript in each parameter are statistically significant ($P < 0.05$).

removal of sediments from the bivalve compartment should be an effective means to improve environmental conditions, survival rate and growth rate of the clam. Stocking rate of the clam at the rate of 3 kg per 0.58 m³ of the bivalve compartment may be too high and could also have generated negative effects on the bivalve themselves (Table 3). Proper stocking rate of the clam should improve survival rate and growth of the bivalve. Jones and Iwama (1991) reported greater increases in shell height and monthly instantaneous growth rates of oysters suspended at the salmon farm.

The result of this experiment indicated that freshwater clam-tilapia co-culture can be used as a means to reduce particulate nutrients and particulate organic matter including phytoplankton in intensive culture systems of tilapia even though higher production of fish was not obtained from co-culture of freshwater clam with tilapia at stocking rate using in this study. The reduction of pollutants in culture water resulting from the filtration ability of freshwater clams definitely results in better environmental conditions in culture tanks and less pollutant (organic matter, phytoplankton and nutrients) loading in fish culture effluents.

Table 3. Survival rate, weight gain and final body weight of freshwater clam cultured with tilapia (mean±S.D.).

Initial number	162±7.0
Initial total weight (g)	3000
Average initial weight (g/individual)	18.54±0.78
Survival rate (%)	65.0±23.3
Final total weight (g/tank)	2016.7±700.6
Final average weight (g/clam)	19.22±1.02
Average weight gain (g/individual/8 weeks)	0.68±0.24
Average weight gain (%)	3.6±1.2

CONCLUSION

According to the results of this study, freshwater clam, *P. exilis compressa*, can be used effectively to improve water quality in intensive tilapia culture systems. Filtration by freshwater clams can significantly reduce total particulate matter, particulate organic matter, phytoplankton, particulate nitrogen and particulate phosphorus in culture water resulting in the reduction of pollutant loading in fish farm effluents and better environmental conditions in fish culture system. However, more studies are

needed to obtain proper stocking density of bivalve and fish in order to obtain optimum production as well as farm scale investigation. Finally, hatchery techniques to produce juvenile freshwater clam also need to be developed.

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Authors must display good knowledge of the primary scientific literature. Authors must also prepare manuscripts according to the journal's standards and instructions in order to facilitate prompt review and processing of papers.

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- Abstract - summarizes the study in not more than 200 words, on a page by itself
- Introduction - lays out the problem addressed, current level of knowledge, the aims of the study, and the hypotheses tested
- Materials and methods - includes all crucial information to allow replication of the study
- Results - gives concise summary of data in Tables and Figures
- Discussion - places the study in the larger context of fisheries science and literature
- Conclusion - encapsulates the scientific contribution of the study
- Acknowledgement - credits contributors of thoughts, expertise and funds
- References - must substantially include the peer-reviewed primary literature

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3. Leave a triple space before and after all headings. Use capital and lower case letters, never all capitals. Avoid footnotes, addenda or appendices; if they are really important, incorporate them briefly in the text. Underline only the words to be italicized. Define acronyms or unfamiliar abbreviations at first mention in the text. Do not give any acronym in parenthesis if it is not used later again in the text.

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Example: Asian sea bass (*Lates calcarifer*, Centropomidae).

5. Place a (leading) zero before the decimal in numbers less than 1. Give dates in the form 10 January 1994. Spell out numbers less than 10 unless they stand beside standard units of measure (eight fish and 8 kg). Do not spell out numbers larger than 10 unless they are used to start a sentence.

6. Use metric units or the International System of Units (with base units meter, gram, second, liter, mole, joule, etc.). Common units such as day, tons, hectare, watts, horsepower, °C and ppt salinity may be acceptable. Use abbreviations of units only beside numerals (e.g., 5 m); otherwise spell out units (e.g., only meters away). Do not use plural forms or periods for abbreviations of units. Use superscripts and subscripts

instead of the bar (/) for compound units; for example, $2 \text{ t} \cdot \text{ha}^{-1} \cdot \text{year}^{-1}$ instead of 2/t/ha/year, $10 \text{ g} \cdot \text{m}^{-2}$ instead of 10 g/m².

7. In designing Tables and Figures, bear in mind the journal's page (17.8 cm x 25.3 cm or 7" x 10") and any reduction needed. Table headings, Figure explanations and other labels must be understandable without reference to the text. Number Tables and Figures consecutively, one per page. Tables must have horizontal lines only at the top and bottom and no vertical lines at all. Leave spaces to indicate groupings of data. Figures must be neat and simple line drawings, computer-generated graphics, or good-quality black and white photographs. Labels or lettering on Figures must be of a size readable after reduction (up to 60%). Send electronic images (.jpg or .tif format) at first submission and the originals only with the revised manuscript if necessary.

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| (proceedings) | Ronquillo, I.A. 1974. A review of the roundscad fishery of the Philippines. Proceedings Indo-Pacific Fisheries Council 15 (3):351-375. |
| (book chapter) | Smith, I. and K.C. Chong. 1984. Southeast Asian milkfish culture: economic status and prospects. In: Advances in milkfish biology and culture (ed. J.V. Juario, R.P. Ferraris and L.V. Benitez), pp.1-20. Island Publishing House, Manila. |
| (journal article)* | Widodo, J. 1991. Maturity and spawning of shortfin scad (<i>Decapterus macrosoma</i>) (Carangidae) of the Java Sea. Asian Fisheries Science 4:245-252.
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