Effects of addition of tilapia on the abundance of periphyton in freshwater prawn culture ponds with periphyton substrates

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Abstract

An experiment was conducted to evaluate the effect of addition of tilapia on abundance of periphyton in freshwater prawn, Macrobrachium rosenbergii (de Man) in periphyton based culture system for a period of 120 days at Fisheries Field Laboratory Complex, Bangladesh Agricultural University, Mymensingh. A large pond (83x8.9 m) was drained completely and partitioned by galvanized iron sheet into 18 small ponds of 40 m² each; of which 6 ponds were used for this experiment. The experimental ponds were divided into 2 treatments each with 3 randomly selected ponds. The absence and presence (0 and 0.5 individual m⁻²) of tilapia (*Oreochromis niloticus*) were investigated in 40 m² ponds stocked with 3 prawn juveniles (5±0.05 g) m⁻² with added substrates for periphyton development. A locally formulated and prepared feed containing 30% protein was supplied considering the body weight of prawn only. Addition of periphyton substrates significantly reduced the inorganic N-compounds (TAN, NO₂-N, and NO₃-N) in water column. Forty six genera of periphyton were identified belonging to the Bacillariophyceae (10), Chlorophyceae (21), Cyanophyceae (7), Euglenophyceae (2), Crustacea (1) and Rotifera (5) with significant difference (P<0.05) of phyto-periphyton except Euglenophyceae and without significant difference (P>0.05) of zoo-periphyton between the treatments. The abundance of periphyton biomass in terms of dry matter, ash, ash free dry matter and chlorophyll-a were significantly higher in tilapia-free ponds comparing to tilapia added ponds. Benthic organisms had no significant difference (p>0.05) between the treatments. Addition of tilapia in periphyton-based system benefited the freshwater prawn culture through (i) reducing toxic inorganic nitrogenous compounds in water (ii) reducing demand for supplemental feed (iii) using periphyton as additional natural feed and, (iv)improving survival and production of prawn and tilapia.

Keywords: Periphyton, Freshwater prawn, Tilapia

Introduction

Freshwater prawn, *Macrobrachium rosenbergii* (de Man) is indigenous to the South and South-East Asia, together with Northern Australia and the western Pacific Islands (New 1988). Through successful domestication in late 1960s (Ling, 1969), the culture of freshwater prawn has gained a great popularity worldwide, mostly in the tropical and subtropical regions, with the limited production in temperate regions (D'Abramo *et al.*, 1989). The global production of freshwater prawn has increased gradually (FAO, 1997) with the major production in East and South Asian countries like China, India, Indonesia, Bangladesh, Thailand and the Philippines.

The prawn farming area in Bangladesh is expanding very fast; the expansion rate in last 3 years is about 10% per year. At present approximately 50,000 ha of land and water bodies are used in prawn aquaculture. The freshwater prawn play a vital role in the development of socio-economic conditions of the country through increasing export trade, food production, creation of rural employment and proper utilization of natural resources (Rahman, 2000). In Bangladesh different culture systems are being practiced, such as prawn monoculture, prawn polyculture along with other fishes (mostly carps), prawn aquaculture in paddy fields and prawn aquaculture in paddy fields after paddy harvesting.

Macrobrachium culture presently practiced in Bangladesh may be categorized broadly into two culture methods- Beri (gher) culture and polyculture with carps. The cultivation of freshwater prawn in modified rice fields locally referred to as "gher" has been developed in Bangladesh (BOBP, 1990; Rosenberry, 1992; Rutherford, 1994). Periphyton is very preferable natural food for herbivorous and omnivorous fish species especially for Indian major carp (Azim et al., 2002; Keshavanath et al., 2002), for tilapia (Trewavas, 1982; Ali, 1998; Wahab et al., 1999), and for freshwater giant prawn (Cohen et al., 1983). The use of periphyton substrates in freshwater finfish and prawn production has been found promising (van Dam et al., 2002). Substrates based system can increase freshwater prawn production to a significantly higher level when compared to traditional production system (Tidwell and Bratvold, 2005). Cohen et al. (1983) reported that added substrate in ponds increased prawn production by 14% and average size

by 13%. Experiments conducted in Bangladesh showed that vertical substrate addition resulted in prawn survival of increment of 75% and production of 127% in prawn-tilapia polyculture system (Uddin, 2007). Therefore, freshwater prawn productivity can be enhanced through stimulation of suspended and attached bacteria and algae development, and by using them to improve water quality, provide additional food and improve nutrient efficiency.

Introducing substrates for periphyton development (Tidwell *et al.;* 2000, Tidwell and Bratvold, 2005; Uddin, 2007) manipulation of C:N ratio (Azim and Little, 2006; Avnimelech, 2007) and combination of both C:N ratio and periphyton substrates in freshwater prawn ponds (Asaduzzaman *et al.,* 2008) have been found promising. These techniques require installation of hard substrates and application of cheap carbohydrate resources which are available within the farmer's traditional agricultural systems. Besides substrate and carbohydrate addition, stocking of tilapia was suggested to reduce underutilization of natural foods (plankton, periphyton and microbial floc) observed in monoculture ponds (Asaduzzaman *et al.,* 2008). Tilapia in such system depends on the availability of natural foods in the form of plankton (Perschbacher and Lorio, 1993), periphyton (Azim *et al.,* 2003a; Uddin, 2007) and microbial floc (Avnimelech, 2007). In addition, tilapia driven movements increases the bottom dissolved O₂ availability leading to better mineralization and stimulating the natural food web (Jimenez-Montealegre *et al.,* 2002).

Tilapias and prawns have different food and feeding habits, but for both species, the addition of substrates resulted in extra growth and production (Tidwell *et al.*, 2000, Uddin *et al.*, 2006). This study monitored the effect of tilapia addition on prawn survival and production, pond ecology, and economic performance in presence and absence of substrates for periphyton development ponds. Thus considering above things in mind, the present study was undertaken to investigate the effects of addition of tilapia on the survival, growth and production of prawn in the presence and absence of substrates with periphyton in ponds.

Materials and Methods

Study area and pond facilities

The experiment was conducted at the Fisheries Field Complex of the Faculty of Fisheries, Bangladesh Agricultural University, Mymensingh, for a period of 120 days from 20 February to 20 June 2008 in six ponds having an area of 40 m² each. The ponds were rectangular in shape, well exposed to sun light, independent, and having water supply facilities. The water depth was maintained to a maximum of 01 m over the study period. The surrounding of all ponds was covered by 01 m height nylon net to prevent the entry of predators like birds, snakes, frogs and others.

Experimental design

The experiment was conducted under completely randomized design (CRD). Two treatments were tested each of which was replicated. The experimental design has been shown in Table 1. Table 1. Experimental Design

Properties of culture	Treatment-1 (PT ₀)	Treatment-2 (PT _{0.5})
Prawn	120 (3 juveniles m ⁻²)	120 (3 juveniles m ⁻²)
Tilapia	Nil	20 (0.5 fry m ⁻²)
C/N ratio	20	20
Periphyton substrate Arrangement	Present	Present

Pond preparation

All undesirable fish were completely eradicated by applying rotenone at a rate of 2.5 g/m³. Aquatic weeds were removed manually. The grasses of the pond dikes were also cut into small size by using scythe. After one week of rotenone application, lime (CaO) was applied at a rate of 250 kg ha⁻¹. Three days after liming, ponds were fertilized with urea and triple super phosphate (TSP) each at a rate of 25 kg ha⁻¹ and cow dung at a rate of 1,000 kg ha⁻¹.

The shelter was built by bamboo branch (locally known as *kanchi*) with date tree leaves and was installed in each pond before stocking with prawn juveniles to provide shelter for prawn. About 436 bamboo with a mean diameter of 0.05 m were posted vertically into the bottom mud of each pond, excluding a one meter wide perimeter water surface from the dike. This resulted in an additional area for periphyton development equaling about 60% (i.e. 24 m²) of the pond surface area. Ponds were not fertilized during the grow-out period. After the first fertilization and before prawn stocking, the ponds were left 10 days to allow plankton development in water column and periphyton growth on bamboo kanchi.

Stocking

Juveniles of *Macrobrachium rosenbergii* (5± 0.05g) procured from a nearby commercial hatchery were stocked at 3 juveniles m⁻² in the ponds and juveniles of *Oreochromis niloticus* (24.3± 0.24g) from the Bangladesh Fisheries Research Institute (BFRI) were stocked according to the experimental design.

Feeding

Juveniles of freshwater prawn were fed with processed palleted feed containing 30.03% crude protein daily at a rate of 10% of the body weight for the 1st month, 7% for 2nd month and 3% for the rest of the culture period. Half of the required feed for a day was supplied in the morning and rest half in the evening. Feed requirement was calculated and adjusted after monthly sampling of prawn. Locally purchased tapioca starch was used as carbohydrate source for manipulating the C/N ratio. In order to raise the C/N ratio to 20 in all the ponds, 0.9kg tapioca starch was applied for each kg of formulated feed. The preweighed tapioca starch was mixed in a beaker with pond water and uniformly distributed over the pond surface directly after the feed application at 7.00 am.

Phyto and zoo-periphyton enumeration

Plankton samples were collected monthly from each pond. A bucket contained two litres of water was used to collect 10 litres of water from five different places and depth of the pond and passed through a fine mesh (25 μ m) plankton net. The concentrated samples were transferred to a measuring cylinder and carefully made up to a standard volume of 50 ml with distilled water. Then the collected plankton samples were preserved in 10% buffered formalin in small plastic bottles each for subsequent studies.

From each of the 50 ml preserved sample, 1 ml sub-sample was examined using an S-R cell (Sedge Wick-Rafter cell S50, Microlitre) under a binocular microscope (Olympus, M-4000D, Japan) with phase contrast facilities.

One ml sub-sample from each sample was transferred to the cell and then all planktonic organisms present in 10 squares of the cell were identified and counted. Identification of plankton to the genus level was carried out using the Keys from Ward and Whipple (1959), Prescott (1962) and Belinger (1992). For each pond, mean number of plankton was recorded and expressed numerically per litre of water. Plankton density was calculated using the formula of Azim *et al.*, 2001b.

N= (P X C X 100)/L

Where,

N= The number of plankton cells or units per liter of original water

P= The number of plankton counted in ten fields

C= The volume of final concentrate of the sample

L= The volume (liters) of pond water sample.

Harvesting

Adult freshwater prawn were harvested and counted for total number separately from each plot to evaluate the survival rate. After direct counting, weight and length of each individual prawn was also taken.

Analysis of growth data

Experimental data collected during the growth trial were used to determine the growth parameters as follows:

Weight gain (g):

Weight gain = Mean final prawn weight –Mean initial prawn weight

Percent weight gain (%):

% weight gain =
$$\frac{\text{Mean final prawn weight - Mean initial prawn weight}}{\text{Mean initial weight}} \times 100$$

Average daily gain (g):

ADG (g) =
$$\frac{\text{Mean final prawn weight - Mean initial prawn weight}}{T_2 - T_1}$$

Specific growth rate (% per day):

SGR (% per day) =
$$\frac{\log_e W_2 - \log_e W_1}{T_2 - T_1} \times 100$$

Where,

 W_1 = Initial live prawn body weight (g) at time T_1 (day) W_2 = Final live prawn body weight (g) at time T_2 (day) $T_2 - T_1$ = Duration of the experiment (day).

Statistical analysis

For the statistical analysis of the data, a one-way ANOVA and DMRT were done by using the SPSS (Statistical Package for Social Science) version-11.5. Significance was assigned at the 0.05% level. Duncan's test was used to test the results of multiple ranges for comparisons of averages.

Results and Discussion

Water quality parameters

Physical parameters like transparency, temperature and chemical parameters such as pH, dissolved oxygen (DO), nitrate-nitrogen (NO $_3$ -N), phosphate-phosphorous (PO $_4$ -P), nitrite-nitrogen (NO $_2$ -N), total ammonia-nitrogen (TAN) and chlorophyll-a were measured throughout the study period. All parameters were more or less within the acceptable range for freshwater prawn culture. Water quality parameters in different treatments have been presented in Table 2.

Table 2. Means (±SE) of water quality parameters of ponds under	der two treatments
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Variables	Treatment		
(Mean±SE)	PT _{0.5} (With Tilapia)	PT ₀ (Without Tilapia)	
Temperature (°C)	30.76±0.24	30.69±0.23	
Transparency (cm)	43.69±1.26	27.15±0.55	
PH	9.59±1.52	7.89±0.06	
Surface-DO (mgL ⁻¹)	5.39±0.06	5.43±0.06	
Bottom-DO (mgL ⁻¹)	3.13±0.09	2.90±0.09	
TAN (mg L ⁻¹)	0.0368±0.0087	0.0396±0.0077	
NO2-N (mg L ⁻¹)	0.0055±0.0011	0.0072±0.0015	
NO3-N (mg L ⁻¹)	0.0427±0.0081	0.053±0.0087	
PO4-P (mg L ⁻¹)	1.19±0.2408	1.36±0.2024	
Chlorophyll-a (µg L ⁻¹)	102.9±10.19	171.80±11.88	

Temperature and pH of water were similar among the treatments. The addition of tilapia increased the bottom dissolved O_2 by 6.9% compared to the treatments without tilapia. The addition of tilapia increased the transparency and decreased the chlorophyll-a concentration of water. The chlorophyll-a concentration was always lower in tilapia added ponds ($PT_{0.5}$) compared to tilapia free ponds (PT_0) during the culture period. The mean values of TAN, NO_2 -N, NO_3 -N and PO_4 -P decreased by the addition of tilapia. All of the inorganic nitrogenous compounds (TAN, NO_2 -N, and NO_3 -N) decreased continuously during the culture periods in all treatments except for NO_3 -N in treatment without tilapia addition. The rate of decreasing of all inorganic nitrogenous compounds was higher and lower with and without addition of tilapia respectively.

Periphyton enumeration

Qualitative Study: Qualitative study of periphyton has been presented in Table 3. Periphyton population includes a complex community of microbiota (phyto-periphyton, zoo-periphyton, bacteria, fungi, larvae, inorganic and organic detritus). In the present study, only phyto-periphyton and zoo-periphyton were analyzed. There were 40 genera of phyto-periphyton under 5 groups (Bacillariophyceae, 10 genera; Chlorophyceae, 21 genera; Cyanophyceae, 07 genera and Euglenophyceae, 02 genera) and 6 genera of zoo-periphyton under 2 groups (Crustacean, 01 genera; Rotifera, 05 genera) found on bamboo *kanchi* substrates. Among the different groups, the dominant groups were Chlorophyceae, Bacillariophyceae and Cyanophyceae.

Phyto-periphyton population

Mean abundance of phyto-periphyton along with their different groups are shown in Table 3. Phyto-periphyton population of the fish ponds was composed of four major groups: Bacillariophyceae (Diatom), Chlorophyceae (Green algae), Cyanophyceae (Blue green algae) and Euglenophyceae (Euglenophytes).

Table 3. Mean abundance ± SE (x10³ cells cm⁻²) of phyto-periphyton of the ponds under two treatments each having three replicates. Values are means of 3 replicates and 5 sampling dates (N=15)

Phyto-periphyton abundance Mean±SE	Treatment PT _{0.5} (With tilapia)	Treatment PT ₀ (Without tilapia)
Bacillariophyceae	8.99±0.97 ^b	19.90±1.09 ^{a*}
Chlorophyceae	15.51±1.79 ^b	36.89±2.18 ^{a*}
Cyanophyceae	9.75±0.678 ^b	15.75±0.86 ^{a*}
Euglenophyceae	0.44±0.06 ^a	0.56±0.10 ^a
Total	34.69±3.28 ^b	73.11±3.83 ^{a*}

^{*} Mean values with the different superscripts in rows are significantly different (P<0.05)

Bacillariophyceae (Diatom): Bacillariophyceae comprised of 10 genera were observed. The mean abundance ($\times 10^3$ cells cm⁻²) of Bacillariophyceae was found to vary from 4.73 to 15.568 and 14.04 to 27.52 ($\times 10^3$ cells cm⁻²) with the mean values of 8.99±0.97 and 19.90±1.09 ($\times 10^3$ cells cm⁻²) in case of treatments PT₀ and PT_{0.5}, respectively. The abundance of treatment PT₀ was significantly higher (P< 0.05) than that of treatment PT_{0.5}. Bacillariophyceae were dominated by *Navicula*, *Nitzchia*, *Fragillaria*, *Cyclotella*, *Diatoma*, *Pinnularia*, *Surrirella* and *Synedra*. Monthly variations in the abundance of Bacillariophyceae between the treatments are shown in Table 4.

Chlorophyceae: Chlorophyceae was found to be dominat over all groups of phyto-periphyton. Among the 21 genera observed *Actinastrum, Ankistrodesmus, Chlorella, Closterium, Coelastrum, Oocystis, Pediastrum, Scenedesmus, Sphaerocystis and Ulothrix* were the dominant ones. Mean abundance ($\times10^3$ cells cm⁻²) of Chlorophyceae were 15.51 ± 1.79 and 36.90 ± 2.18 in treatments $PT_{0.5}$ and $PT_{0.5}$ respectively (Table 5). The mean abundance ($\times10^3$ cells cm⁻²) of Chlorophyceae was found to vary from 8.62 to 33.92 and 19.04 to 49.90 in case of treatments $PT_{0.5}$ and $PT_{0.5}$ respectively. The variation in abundance of $PT_{0.5}$ was significantly higher (P<0.05) than that of the treatment of $PT_{0.5}$.

Table 4. Comparison of the abundance of periphyton (diatom) (biomass scraped from bamboo kanchi in different treatments)

Variables	Treatment- PT _{0.5}	Treatment-PT ₀	Significance P
Periphytic diatom abundance (x10 ³ cells cm ⁻²)	(With Tilapia)	(Without Tilapia)	value
Cyclotella	1.06 ^b	3.11 ^a	***
Coscinodiscus	0.69 ^b	1.15 ^a	*
Diatoma	1.34 ^b	2.66 ^a	*
Fragillaria	1.49 ^a	1.34 ^b	***
Melosira	0.75 ^b	1.62 ^a	**
Navicula	0.87 ^b	2.41 ^a	***
Nitzschia	0.40	1.03	NS
Surirella	0.29	0.40	NS
Synedra	1.52 ^b	3.90 ^a	***
Tabellaria	1.58	2.13	NS
Total	8.99 ^b	19.90 ^a	***

Values are the means of 5 sampling dates, three depths, three poles and three ponds (N=135). The mean values with different superscript indicate significant difference at *P<0.05;** P< 0.01 *** P<0.001*; NS, not significant.

Table 5. Effects of addition of tilapia on the abundance of periphyton (green algae) (biomass scraped from bamboo *kanchi* in different treatments)

Variables Periphytic abundance (green algae) (x10 ³ cells cm ⁻²)	Treatment PT _{0.5} (With Tilapia)	Treatment PT ₀ (Without Tilapia)	Significance P value
Actinastrum	0.49 ^b	1.08 ^a	*
Ankistrodesmus	0.41	0.46	NS
Botryococcus	0.40 ^b	1.41 ^a	*
Chaetophora	0.67 ^b	1.75 ^a	**
Chlorella	5.65 ^b	13.80 ^a	***
Gonatozygon	0.30	0.75	NS
Closterium	0.26 ^b	0.94 ^a	**
Coelastrum	0.48	0.80	NS
Draparnaldia	0.27	0.46	NS
Microspora	0.53 ^b	1.22 ^a	**
Oedogonium	0.87 ^b	1.95 ^a	**
Oocystis	0.45 ^b	1.81 ^a	***
Palmella	0.69 ^b	1.51 ^a	**
Pediastrum	0.74 ^b	1.41 ^a	**
Scenedesmus	0.80 ^b	1.82 ^a	**
Sphaerocystis	0.69 ^b	1.38 ^a	**
Stigeoclonium	0.20	0.33	NS
Tetraedon	0.33 ^b	0.71 ^a	*
Ulothrix	0.92 ^b	2.49 ^a	**
Volvox	0.20 ^b	0.49 ^a	*
Zygnema	0.17	0.22	NS
Total	15.51 ^b	36.90 ^a	***

Cyanophyceae: Cyanophyceae comprised of 7 genera and ranked second in respect of abundance. Among 7 genera, *Anabaena, Microcystis, Gomphosphaeria* and *Oscillatoria* were dominated. Average abundance ($\times 10^3$ cells cm⁻²) of Cyanophyceae was found to range from 5.98 to 17.51 and 10.29 to 22.80 with mean values of 9.75±0.68 and 15.75±0.86 ($\times 10^3$ cells cm⁻²) in case of treatments PT_{0.5} and PT₀ respectively. However, the variations in abundance of PT₀ were significantly higher (P < 0.05) than that of PT_{0.5}. Monthly variations in the abundance of Cyanophyceae between the treatments are shown in Table 6.

Table 6. Effects of addition of tilapia on the abundance of periphyton (blue green algae) (biomass
scraped from bamboo <i>kanchi</i> in different treatments)

Variables Periphytic blue green algae abundance (×10³ cells cm⁻²)	Treatment PT _{0.5} (With Tilapia)	Treatment PT ₀ (Without Tilapia)	Significance P value
(×10° cells cm°) Anabaena	1.83	1.99	NS
Anabaena			
Anacystis	0.58	0.83	NS
Aphanizomenon	2.22 ^b	3.40 ^a	*
Aphanocapsa	2.22 ^b	3.94 ^a	*
Gomphosphaeria	1.05 ^b	1.69 ^a	*
Microcystis	1.17 ^b	1.78 ^a	*
Oscillatoria	1.11 ^b	2.56 ^a	***
Total	9.75 ^b	15.75 ^a	***

Euglenophyceae: Euglenophyceae consisted of *Euglena* and *Phacus* species and between them *Euglena* was the dominant. The mean abundance ($\times 10^3$ cells cm⁻²) of Euglenophyceae was 0.44±0.06 and 0.60±0.10 in treatments PT_{0.5} and PT₀, respectively. The abundance ($\times 10^3$ cells cm⁻²) of Euglenophyceae was found to range from 0 to 0.83 in treatment PT_{0.5} and 0 to 1.39 in treatment PT_{0.}. There was no significant difference (P>0.05) between treatment PT_{0.5} and PT₀ when ANOVA was performed. Monthly variations in the abundance of Euglenophyceae among the treatments are shown in Table 7.

Table 7. Effects of addition of tilapia on the abundance of periphyton (Euglenophytes) (biomass scraped from bamboo *kanchi* in different treatments)

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Variables	Treatment PT _{0,5} (with	Treatment PT ₀ (without	Significance P
Periphytic abundance (x10 ³ cells cm ⁻²)	tilapia)	tilapia)	value
Euglena	0.25	0.30	NS
Phacus	0.19	0.30	NS
Total	0.44	0.60	NS

Total phyto-periphyton: Among the phyto-periphyton group, Chlorophyceae was the most dominant group and Euglenophyceae was the least abundant group. In treatment $PT_{0.5}$ the average abundance (x10³ cells cm⁻²) of phyto-periphyton was found to range from 23.21 to 66.99 with a mean value of 3.69±3.28, while in treatment PT_0 it varied from 45.04 to 97.99 with a mean value of 73.11±3.83. The variation in abundance of phyto-periphyton in treatment PT_0 was significantly higher (P< 0.05) than that of the treatment $PT_{0.5}$. Monthly variations in the abundance of total phyto-periphyton among the treatments have been shown in Fig. 1.

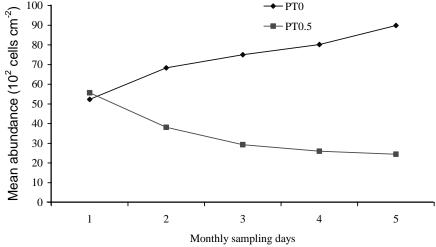


Fig. 1. Monthly variation of abundance of total phyto-periphyton in pond water under different treatments

Zoo-periphyton Population

Two major groups i.e. Crustacea and Rotifera represented zoo-periphyton population of the experimental ponds. The mean abundance of zoo-periphyton in two treatments has been shown in Table 8.

Table 8. Mean abundance \pm SE ($\times 10^3$ cells m⁻²) of zoo-periphyton population of the ponds under two treatments having three replicates each

Periphyton abundance Mean±SE	Treatments PT _{0.5} (with tilapia)	Treatments PT ₀ (without tilapia)
Rotifers	0.72±0.05	0.70±0.06
Crustacea	0.15±0.02	0.25±0.03
Total	0.87±0.06	0.95±0.07

Rotifera: Rotifera was the most dominant zoo-periphytonic group, comprised of 5 genera namely *Asplanchna*, *Brachionus*, *Filinia*, *Lecane* and *Trichocerca*. The mean abundance ($\times 10^3$ cells cm⁻²) was found to range from 0.41 to 1.25 and 0.27 to 1.11 with the mean values of 0.72 \pm 0.05 and 0.70 \pm 0.06 in PT_{0.5} and PT₀ respectively. No significant differences (P > 0.05) were recognized PT_{0.5} and PT₀ when ANOVA was performed. Monthly variations in the abundance of Rotifers in the treatments are shown in Fig. 2.

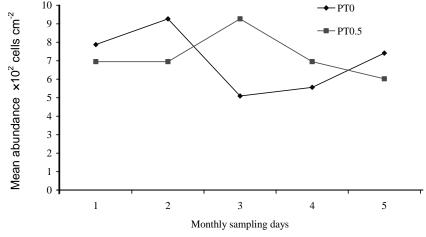


Fig. 2. Monthly variation of abundance of periphytic rotifers in pond water under different treatments

Crustacea: The abundance (x 10^2 cells cm⁻²) of Crustacea ranged from 0 to 2.78 and 0.139 to 5.56 under treatments PT_{0.5} and PT₀ respectively. Crustacean group comprised of nauplius, was dominant. The mean abundance was not significantly different (P>0.05) between the treatments. Monthly fluctuation of Crustacean in different treatments is presented in Fig. 3.

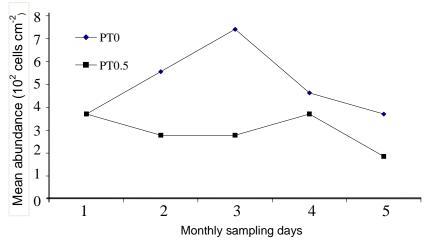


Fig. 3. Monthly variation of abundance of periphytic crustaceans in pond water under different treatments

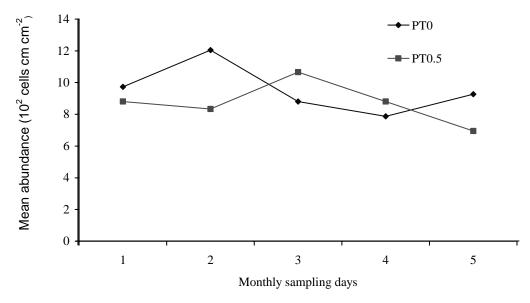


Fig. 4. Monthly variation of abundance of total zoo-periphyton in pond water under different treatments

Total zoo-periphyton: Monthly variations of total zoo-periphyton in different treatments have been shown in Fig. 4. The mean zoo-periphyton density was 0.87 ± 0.06 and 0.95 ± 0.06 (x 10^2 cells cm⁻²) in treatments PT_{0.5} and PT₀ respectively. Hence the values did not differ. Total zoo-periphyton abundance was found to range from 0.42 to 1.39 and 0.56 to 1.67 (x 10^2 cells cm⁻²) in treatments PT_{0.5} and PT₀ respectively.

Growth and yield performance of freshwater prawn

The growth of freshwater prawn in different treatments was different. The different growth performance namely length (cm) and weight (g) gain, survival rate (%), percent weight gain and specific growth rate (% per day) and survival rate are shown in Table 9.

Table 9. Effects of addition of tilapia on growth and yield parameters of freshwater prawn

Variables	Treatments P T _{0.5}	Treatments P T ₀	Signifi	cance (P	Value)
variables	(with tilapia)	(without tilapia	Р	Т	PxT
Initial Stocking weight (g)	5.0	4.9	NS	NS	NS
Final harvesting weight (g)	35.2	37.2	NS	NS	NS
Final weight gain (g)	30.2	32.3	NS	NS	NS
SGR (% bw d ⁻¹)	1.63	1.68	*	NS	NS
Food conversion ratio	2.38 ^a	2.05 ^b	**	**	NS
Survival (%)	63.6	67.8	***	NS	NS
Gross yield (kg ha ⁻¹ 120 d ⁻¹)	668 ^b	751 ^a	***	**	*
Net yield (kg ha ⁻¹ 120 d ⁻¹)	519 ^b	604 ^a	***	**	*

P = periphyton substrates; T = tilapia addition; $P \times T$ = interaction of addition of periphyton substrates and tilapia. The mean values with different superscript letters indicate significant difference at * P < 0.05; ** P < 0.01; *** P < 0.001; NS, not significant.

Water quality parameters

Water quality in lentic natural water bodies is strongly dependent on the autotrophic and heterotrophic organisms developed within the systems. In periphyton-based system, the close linkage between autotrophic and heterotrophic processes in periphyton mats speed up nutrient cycling and positively influences water quality (Milstein *et al.*, 2003). The DO concentrations were generally suitable for prawn culture, although exceptionally low bottom DO values were recorded on a few occasions in tilapia free ponds. The addition of tilapia brings some oxygen to the bottom layers by their movements (Jimenez-

Montealegre et al., 2002), thus increasing the bottom dissolved oxygen. Periphyton lowered the PO₄-P of the overlying water which was also reported by the Hansson (1990) and Bratvold and Browdy (2001). Langis et al. (1988) and Ramesh et al. (1999) reported that the bacterial biofilm (periphyton) reduced toxic nitrogenous compounds through promotion of nitrification. In substrate-based ponds, nitrifying bacteria develop on the substrates which are located in the water column where more oxygen is available than at the sediment-water interface. In addition, periphytic algal community contributes to the processing of the nitrogenous wastes in ponds (Shilo and Rimon, 1982; Diab and Shilo, 1988). Thompson et al. (2002) reported that the attached diatoms and periphytic filamentous Cyanobacteria were responsible for the largest uptake of ammonium from the water in intensive shrimp culture ponds. The very low nitrogenous compounds in all treatments compared to other studies of freshwater prawn farming (Wahab et al., 2008; Kunda et al., 2008) and decreasing trends over time were due to the high C:N ratio of 20 and the addition of substrates for periphyton development. In the present study, tapioca starch was used as carbohydrate source for maintaining C:N ratio at 20. Increasing nutrient inputs caused lower NO₂-N concentration in the water column and over the time, which can be attributed to low availability of TAN as substrate for nitrification (Avnimelech, 1999; Hari et al., 2004). Thus the lower level of nitrogenous compounds (NH₃-N; NO₂-N; NO₃-N) over time could be attributed due to the addition of carbonaceous substrates that lead to increased microbial biomass, which immobilized TAN (Asaduzzaman et al., 2006; Asaduzzaman et al, 2008; Hari et al., 2004) and uptake of the nitrogenous compounds by periphyton. Addition of tilapia decreased the total nitrogen in the sediment possibly due to increased denitrification in response to fish driven oxygenation events (Torres-Berristain et al., 2006). In addition, another cause might be due to the re-suspension of pond bottom which release nutrients to the water column and tilapia harvested more phytoplankton in the water column keeping the algae more young stage which needed more nutrients as well.

Periphyton production

The major natural food types in ponds are phytoplankton, zooplankton, microbial floc, periphyton and benthic macroinvertebrate. The amounts of these natural food in ponds are influenced by management factors such as species combination, stocking density and ratio, and nutrient input quality and quantity (Milstein, 1993; Diana *et al.*, 1997).

The periphyton community constitutes a major component of aquatic biological systems (Biggs, 1987). Perithyton includes both the phyto-perithyton and zoo-periphyton and sometime aquatic insects (Wetzel, 1983; Biggs, 1987). In the present study, phyto-periphyton and zoo-periphyton were only recorded as periphyton. The periphyton community was composed of Bacillariophyceae, Chlorophyceae, Cyanophyceae, Euglenophyceae, Crustacea and Rotifera. A total of 40 genera of periphyton were identified in the treatments PT_{0.5} and PT₀. The most dominant genera were *Cyclotella*, *Navicula*, *Synedra*, *Chaetophora*, *Chlorella*, *Pediastrum*, *Scenedesmus*, *Oscillatoria*, *Ulothrix*, *Tabellaria*, *Gopmhosphaeria*, and *Microcystis* colonizing the bamboo substrates in large numbers. Islam (1996), Haque (1996), Kawser (1998) and Ali (1998) observed similar pattern of findings on natural substrates in BAU campus ponds. Wahab *et al.* (1999) reported 53 genera of periphyton collected from scrap of bamboo in fishponds in Bangladesh among which 12 genera rarely occurred. Huchette *et al.* (2000) identified about 32 species of diatom as periphyton along with other microorganisms of both animal and plant kingdoms growing on artificial substrates in tilapia cages.

The mean abundance of periphyton in treatment PT_0 was very high, indicating that bamboo is a good substrate for periphyton growth. Eminson and Cattaneo *et al.* (1978) stated that the hard substrates such as bamboo poles are the most suitable substrate for periphyton growth.

The addition of tilapia decreased the phyto-periphyton and biomass per unit surface area, indicating the preference of tilapia towards periphyton as food. Tilapias are omnivores capable of feeding on benthic and attached algal and detrital aggregates (i.e.periphyton) (Dempster *et al.*, 1993; Azim *et al.*, 2003a). There is also evidence that Nile tilapia grows better grazing on periphyton than filtering suspended algae from water column (Hem and Avit, 1994; Guiral *et al.*, 1995; Huchette *et al.*, 2000; Azim *et al.*, 2003b). The abundance of periphytic zooplankton was similar in all treatments, indicating that the zooplankton communities were less preferable for the tilapias or escaping predation during grazing.

Growth and yield performance of prawn

The growth and net yield of freshwater prawn were significantly higher with no tilapia than with tilapia, indicating that inter-specific competition between tilapia and prawn decreased the net yield of prawn. Although Uddin (2007) reported that tilapia addition might affect prawn survival during molting but the similar survival revealed that addition of substrates might have minimized the territoriality effect of tilapia on prawn. The FCR calculated based on prawn biomass increased significantly with the addition of tilapia because part of the feed was eaten by the tilapia whereas, substrates decreased FCR value by 13%. Uddin (2007) reported that FCR was 13% lower in fed-periphyton-based ponds compared to substrate free fed ponds. In case of tilapia, substrate addition increased the gross and net yield, indicating that substrates provide additional natural food for tilapia (Uddin, 2007).

Conclusion

The addition of periphyton substrates increased survival rate of prawn. Final weight and weight gain were significantly higher (P<0.05) in treatment PT_0 than those in treatment $PT_{0.5}$. Specific growth rate was significantly higher in PT_0 than in $PT_{0.5}$. The addition of tilapia decreased the gross and net yield of prawn. Tilapia had significant effects on FCR of freshwater prawn, substrates decreased FCR by 13.5% whereas addition of tilapia increased 16% gross yield.

References

- Ali, M.H. 1998. The potential of periphyton-based monoculture of a major carp, calbaush *Labeo calbasu* (Hamilton). M. S. dissertation, Department of Fisheries Management, Bangladesh Agricultural University, Mymensingh, Bangladesh. 85 pp.
- Asaduzzaman, M., Salam, M.A., Wahab, M.A., Kunda, M. and Rahman, M.B. 2006. Effects of control of C/N ratio by low-cost carbohydrate addition on water quality and pond ecology in freshwater prawn *Macrobrachium rosenbergii* post-larvae nursing system. Bangladesh Journal of Fisheries Research, 10 (2):82-92.
- Asaduzzaman, M., Wahab, M.A., Verdegem, M.C.J., Azim, M.E., Haque, S. and Salam, M.A. 2008. C/N ratio control and substrate addition for periphyton development jointly enhance freshwater prawn *Macrobrachium rosenbergii* production in ponds, *Aquaculture*, 280: 117-123.
- Avnimelech, Y. 1999. C/N ratio as a control element in aquaculture systems. Aquaculture, 176: 227-235.
- Avnimelech, Y. 2007. Feeding with microbial flocs by tilapia in minimal discharge bio-flocs technology ponds. Aquaculture, 264: 140-147.
- Azim, M.E. 2002. 'Periphyton-Based Aquaculture'. Published in weekly OBSERVER Magazine, Bangladesh. 32: 14-16 pp.
- Azim, M.E. and Little, D.C. 2006. Intensifying aquaculture production through new approaches to manipulating natural food. CAB Reviews: in Agriculture, Veterinary Science, Nutrition and Natural Resources 1, No. 062, 23 pp.
- Azim, M.E., Wahab, M.A., van Dam, A.A., Beveridge, M.C.M., Milstein, A. and Verdegem, M.C.J. 2001b. Optimization of fertilization rate for maximizing periphyton production on artificial substrates and the implications for periphyton-based aquaculture. *Aquacult. Res.*, 32: 749-760.
- Azim, M.E., Verdegem, M.C.J., Mantingh, I., Van Dam, A.A. and Beveridge, M.C.M. 2003a. Ingestion and utilization of periphyton grown on artificial substrates by Nile Tilapia, *Oreochromis niloticus* L. *Aquacult. Res.*, 34: 85-92.
- Azim, M.E., Verdegem, M.C.J., Singh, M., Van Dam, A.A. and Beveridge, M.C.M. 2003b. The effects of periphyton substrate and fish stocking density on water quality, phytoplankton, periphyton and fish growth. *Aquacult. Res.*, 34: 685-695.
- Belinger, E.G. 1992. A Key to Common Algae. The Institute of Water and Environmental Management, London, UK.
- Biggs, B.J.F. 1987. Periphyton, Aquatic Biology and Hydroelectric Power Development in New Zealand. Henriques, P.R. (ed), 192-206 pp.
- BOBP. 1990. Shrimp seed collectors of Bangladesh. Bay of Bengal Programme (BOBP), Madras, India.
- Bratvold, D. and Browdy, C.L. 2001. Effect of sand sediment and vertical surfaces (AquaMats TM) on production, water quality and microbial ecology in an intensive *Liptopenaeus vannameil* culture system. *Aquaculture,* 195: 81-94.
- Cohen, D., Ra'anan, Z. and Barnes, A. 1983. Production of the freshwater prawn *Macrobrachium rosenbergii* in Israel. Integration into fish polyculture systems. *Aquaculture*, 31: 67-76.
- D'Abramo, L.R., Heinen, J.M., Robbinette, H.R. and Collins, J.S. 1989. Production of freshwater prawn *Macrobrachium rosenbergii* stocked as juveniles at different densities in temperate zone ponds. *J. World Aquaculture Soc.*, 20: 81-89.
- Dempster, P.W., Beveridge, M.C.M. and Baird, D.J. 1993. Herbivory in the tilapia *Oreochromis niloticus*: A comparison of feeding rates of phytoplankton and periphyton. *J. Fish Biol.*, 43: 385-392.
- Diab, S. and Shilo, M. 1988. Effect of light on the activity and survival of *Nitrosomonas* sp. and *Nitrobactor* sp. isolated from fish pond. *Bamidgeh*, 40: 50–60.
- Diana, J. S., Szyper, J.P., Batterson, T.R., Boyd, C.E. and Piedrahita, R.H. 1997. Water quality in ponds. In: Egna, H.S., Boyd, C.E. (Eds.), Dynamics of pond aquaculture. CRC press LLC, 53-72 pp.
- Eminson, D. and Cattaneo, G. 1978. A laboratory experiment to examine effects of nutrient enrichment on macrophyte growth. Verh. Int. Ver. Limnol., 28: 82-87.
- FAO. 1997. Aquaculture development. FAO Technical Guidelines for Responsible Fisheries, Food and Agricultural Organization of the United Nations, Rome.
- Guiral, D., Gourbault, N. and Helleouet, M.N. 1995. Sediment nature and meiobenthos of an artificial reef (Acadja) used for extensive aquaculture. *Oceanol. Acta.*, 18: 543-555.

- Hansson, L.A. 1990. Quantifying the impact of periphytic algae on nutrient availability for phytoplankton. *Freshwat. Biol.*, 24: 265-273.
- Haque, M.M. 1996. Effects of periphyton on pond fish culture. M. S. dissertation, Department of Fisheries Biology and Limnology, Bangladesh Agricultural University, Mymensingh. 87 pp.
- Hari, B., Kurup, M.B., Varghese, J.T., Sharma, J.W. and Verdegem, M.C.J. 2004. Effects of carbohydrate addition on production in extensive shrimp culture systems. *Aquaculture*, 241: 179-194.
- Hem, S. and Avit, J.L.B. 1994. First results on "acadjas enclos" as an extensive aquaculture system (West Africa). *Bull. Mar. Sci.*, 55: 1038-1049.
- Huchette, S.M.H., Beveridge, M.C.M., Baird, M.C.M. and Ireland, M. 2000. The impact of grazing by tilapias (*Oreochromis niloticus* L.) on periphyton communities growing on artificial substrate in cages. *Aquaculture*, 186: 45-60.
- Islam, A.T.M.F. 1996. The use of bamboo substrates to promote periphyton growth as feed for Nile tilapia (*Oreochromis niloticus*) in small seasonal ponds. M.S. dissertation, Department of Fisheries Biology and Limnology, Bangladesh Agricultural University, Mymensingh. 56 pp.
- Jiménez-Montealegre, R., Verdegem, M., Zamora, J.E. and Verreth, J. 2002. Organic matter sedimentation and resuspension in tilapia (*Oreochromis niloticus*) ponds during a production cycle. *Aquacult. Eng.*, 26: 1-12.
- Kawser, M.R. 1998. Effect of periphyton on production of major carps. M. S. thesis, Department of Fisheries Management, Bangladesh Agricultural University, Mymensingh. 82 pp.
- Keshavanath, P., Gangadhar, B., Ramesh, T.J., van Dam, A.A., Beveridge, M.C.M. and Verdegem, M.C.J. 2002. The effect of periphyton and supplemental feeding on the production of the indigenous carps *Tor khudree* and *Labeo fimbriatus*. *Aquaculture*, 213: 207–218.
- Kunda, M., Azim, M.E., Wahab, M.A., Dewan, S., Roos, N. and Thilsted, S.H.. 2008. Potential of mixed culture of freshwater prawn (*Macrobrachium rosenbergii*) and self-recruiting small species mola (*Amblypharyngodon mola*) in rotational rice-fish/prawn culture systems in Bangladesh. *Aquacult. Res.*, 39: 506-517.
- Langis, R., Proulx, D., de la Noue, J. and Couture, P. 1988. Effects of bacterial biofilm on intensive *Daphnia* culture. *Aquacult. Eng.*, 7: 21-38.
- Ling, S.W. 1969. Methods of rearing and culturing Macrobrachium rosenbergii. FAO Fish. Rep. 57, pp.589-606.
- Milstein, A. 1993. Water quality and freshwater fish culture intensification: the Israeli example. Aquacult. Fish. Manage., 24: 715-724.
- Milstein, A., Azim, M.E., Wahab, M.A. and Verdegem, M.C.J. 2003. The effects of periphyton, fish and fertilizer dose on biological processes affecting water quality in earthen fish ponds. *Env. Biol. Fish.*, 68: 247-260.
- New, M.B. 1988. Freshwater prawns: status of global aquaculture, 1987. NACA Technical Manual No. 6. A World Food Day Publication of the Network of Aquaculture Centres in Asia. Bangkok, Thailand. 58p.
- New, M.B. 2002. Farming freshwater prawns: a manual for the culture of the giant river prawn (*Macrobrachium rosenbergii*). FAO Fisheries Technical Paper no. 428, FAO, Rome, Italy, 212 pp.
- Perschbacher, P.W. and Lorio, W.J. 1993. Filtration rates of catfish pond phytoplankton by Nile tilapia *Oreochromis niloticus*. *J. World Aqua. Soc.*, 24(3): 434-437.
- Prescott, G.W. 1962. Algae of the Western Great Lakes Area. Wm. C. Brown Co., Dubuque, Iowa, 977 pp.
- Rahman, M.M. 2000. Effect of addition of calbaush (*Labeo calbasu*) in the periphyton based aquaculture system of rohu and catla. Department of Fisheries Management, Bangladesh Agricultural University, Mymensingh. 82 pp.
- Ramesh, M.R., Shankar, K.M., Mohan, C.V. and Varghese, T.J. 1999. Composition of three plants substrates for enhancing carp growth through bacterial biofilm. *Aquacult. Eng.*, 19: 119-131.
- Rosenberry, B. 1992. The post-larvae fishermen of Bangladesh. World Shrimp Farming, 17(3): 7-9.
- Rutherford, S. 1994. CARE and Gher: Financing the small Fry, Investigation of How Freshwater Prawn cultivation is financed. Bangladesh Aquaculture and Fisheries Resources Unit (BAFRU), Dhaka, Bangladesh, 75 pp.
- Shilo, M., Rimon, A. 1982. Ammonia transformation in intensive fish ponds. Bangladesh, 34: 101-114.
- Thompson, F.B., Abreu, P.C. and Wasielesky, W. 2002. Importance of biofilm for water quality and nourishment in intensive shrimp culture. *Aquaculture*, 203: 263-278.
- Tidwell, J.H. and Bratvold, D. 2005. Utility of added substrates in shrimp culture. In: Azim M.E., Verdegem, M.C.J., van Dam, A.A., Beveridge, M.C.M. (Eds.), Periphyton: Ecology, Exploitation and Management. CABI Publishing, Wallingford, UK, 247-268 pp.
- Tidwell, J.H., Coyle, S., Van Arnum, A. and Weibel, C. 2000. Production response of freshwater prawns *Macrobrachium rosenbergii* to increasing amounts of artificial substrate in ponds. *J. World Aquacult. Soc.*, 31(3):452-458.
- Torres-Berristain, B., Verdegem, M.C.J., Kerepeczki, E. and Verreth, J. 2006. Decomposition of high protein aquaculture feed under variable oxic conditions. *Water Research*, 40: 1341-1350.
- Trewavas, E. 1982. Tilapia: taxonomy and speciation (in Africa). In: R. S. V. Pullin; R. H. L. McConnel (eds.). International Center for Living Aquatic Resource Management, 3-13 pp.
- Uddin, M.S. 2007. Mixed culture of tilapia (*Oreochromis niloticus*) and freshwater prawn (*Macrobrachium rosenbergii*) in periphyton-based ponds. Ph.D. Thesis. Fish Culture and Fisheries Group, Wageningen University, Netherlands, 208 pp.
- Uddin, M.S., Azim, M.E., Wahab, M.A. and Verdegem, M.C.J. 2006. The potential of mixed culture of genetically improved farmed tilapia (GIFT, *Oreochromis niloticus*) and freshwater prawn (*Macrobrachium rosenbergii*) in periphyton-based systems. *Aquacult. Res.*, 37: 241-247.
- Van Dam, A.A., Beveridge, M.C.M., Azim, M.E. and Verdegem, M.C.J. 2002. The potential of fish production based on periphyton. *Rev. Fish Biol. Fish.*, 12: 1-31.
- Wahab, M.A., Kunda, M., Azim, M.E., Dewan, S. and Thilsted, S.H. 2008. Evaluation of freshwater prawn-small fish culture concurrently with rice in Bangladesh. *Aquacult. Res.*, 1-9.
- Wahab, M.A., Azim, M.E., Ali, M.H., Beveridge, M.C.M. and Khan, S. 1999. The potential of periphyton based culture of the native major carp kalbaush, *Labeo calbasu* (Hamilton). *Aquacult. Res.*, 30: 409-419.
- Ward, H.B. and Whipple, G.C. 1959. Freshwater Biology. John Wiley and Sons Inc, New York
- Wetzel, R.G. 1983. Limnology (2nd ed.) Saunders College Publishing, New York. 767 pp.