



Evaluation of production performance and profitability of hybrid red tilapia and genetically improved farmed tilapia (GIFT) strains in the carbon/nitrogen controlled periphyton-based (C/N-CP) on-farm prawn culture system in Bangladesh



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ARTICLE INFO

Article history:

Received 7 January 2016

Received in revised form 28 July 2016

Accepted 28 July 2016

Available online 2 August 2016

Keywords:

Hybrid red tilapia

GIFT tilapia

Freshwater prawn

C/N ratio

On-farm research

ABSTRACT

Performance of hybrid red tilapia (Mutant, *Oreochromis niloticus* × *Oreochromis mossambicus*) and GIFT tilapia strain (*Oreochromis niloticus*) in C/N-CP prawn (*Macrobrachium rosenbergii*) farming system was evaluated at the farmers' pond at Bailor union under Trishal upazilla of Mymensingh district, Bangladesh. The on-farm trial had two treatments: TR and TG (named according to the tilapia strains) with three replications. Six rectangular ponds of varying sizes (400–880 m²) were used for this experiment. Hybrid red and GIFT tilapia strains were stocked with prawn at the stocking densities of 1 tilapia fingerlings (either red or GIFT strain) and 3 prawn juveniles m⁻² in both treatments. Bamboo side shoot were posted vertically as periphyton substrate. This resulted in an additional substrate surface area of 1067 m² for periphyton development equaling 147% of the pond surface area. Considering the body weight of freshwater prawn only, feeding rates were 10% of body weight at the beginning of the study (up to 30 days), and feeding application was gradually reduced to 3% in the last month assuming 80% survival. The abundance of total benthos and periphyton as well as total periphytic biomass were significantly higher ($P < 0.05$) in TR than TG treatment and they were also differed significantly ($P < 0.05$) among different months with a decreasing trends (exception to some extent) over the experimental period. The individual harvesting weight, individual weight gain, specific growth rate, Food Conversion Ratio (FCR), survival (%), gross and net yields of prawn were similar in two treatments. In contrast, the GIFT tilapia strain showed a higher ($P < 0.05$) individual harvesting weight, individual weight gain, specific growth rate ((SGR, % bw d⁻¹), survival, gross and net yields (1935 and 1825 kg ha⁻¹, respectively) combined gross and net yields (2952 and 2784 kg ha⁻¹, respectively), and economic return (3755 US\$ with BCR 0.82) than the hybrid Red tilapia.

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1. Introduction

Freshwater giant prawn (*Macrobrachium rosenbergii*) is an important aquatic crustacean species (Akand and Hasan, 1992) and mostly distributed in the tropical and subtropical regions (Abramo and Brunson, 1996). It is a native species in the south-southeast

Asia and Asia pacific countries (Akand and Hasan, 1992), together with the northern Oceania and the western Pacific islands (New, 2002). Most of this species aquaculture production (97%) comes from four Asian countries: China, Bangladesh, Taiwan and Thailand (FAO, 2001). In 2010, the global production of all freshwater prawn groups (*Macrobrachium* spp.) was 450,158 t, of which Freshwater giant prawn production was 215,029 t (FAO, 2012). Bangladesh is considered one of the most suitable countries in the world for freshwater prawn farming because of its subtropical climate and vast water resources favoring an ideal condition for prawn production (Ahmed et al., 2008). Additional attributes of this species

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like attractive look, fine delicate flavor, good taste and growth has made it very popular (Uddin, 2007). In Bangladesh a silent revolution of freshwater prawn farming is taking place due to its high demand and export value in international markets particularly in the USA, Europe and Japan, as well as its high price in the local market (Asaduzzaman et al., 2009a,b; Ahmed et al., 2009; Ahamed et al., 2014). In 2013–2014, Bangladesh earned US\$ 515 million (BDT 41,180.8 million) by exporting 47635 t of frozen shrimp and prawn species of which about 30% was contributed solely by the freshwater prawns (DOF, 2015). Therefore, freshwater prawn farming has turned into one of the most important sectors in the national economy of Bangladesh within the last three decades (Ahmed et al., 2010; Ahamed et al., 2014). On the contrary, among all other shrimp species, brackish water penaeid shrimp (*Penaeus monodon*) merely contributed the majority (about 75%) of total exported volume (DOF, 2011). As a whole, the freshwater prawn and brackish water shrimp sectors is generating US\$380 million annually and 5.6% of the total value of exports which is the second largest export industry after readymade garments in Bangladesh (DOF, 2006).

The area of prawn culture in different categories of land and water bodies were extended from 6000 ha in 1994 (Chanda and Khondaker, 1994) to an estimated 50,000 ha (DOF, 2007) and 56248 ha in 2010 (BFEEA, 2010). More than 80% of this area is situated in the south-west greater Khulna region. It is expanding on an average of 10–20% per annum (Ahmed et al., 2010). The average annual yields of freshwater prawn from extensive ponds have been recorded at 412 kg ha⁻¹ in monoculture and 390 kg ha⁻¹ in polyculture with finfish species (Asaduzzaman et al., 2006). The typical annual yield of this species was 200–250 kg ha⁻¹ in late 1990s (Rahman, 1999). Nevertheless, this yields are very low in contrast with other adjacent prawn-producing countries (Haque et al., 2015). Therefore, increasing of the pond productivity using affordable resources and sustainable technologies is the most effective way to raise freshwater prawn production. In this perspective, high yield of freshwater prawn can be attained by intensifying aquaculture which needs a high investment, a great extent of energy, technical expertise and advanced technologies. But those are scant as well as out of reach to the majority of freshwater prawn farmers of Bangladesh. Therefore, it is necessary to develop a simple technology that can boost-up pond productivity in a sustainable way, while minimizing the uses of resources (Haque et al., 2015).

Several recent developments of freshwater prawn farming have been found promising to improve the productivity and sustainability of pond aquaculture production: (1) control of C/N ratio (Avnimelech, 2007; Hari et al., 2004); (2) development of periphyton using substrates (Tidwell and Bratvold, 2005; Azim et al., 2003a,b; Milstein et al., 2009); (3) fish driven re-suspension (Milstein et al., 2002; Ritvo et al., 2004), and (4) combination of C:N ratio, periphyton substrates and fish driven approaches in freshwater ponds (Asaduzzaman et al., 2008, 2010; Haque et al., 2015). These approaches are simple and cheap, making those also socially and economically sustainable, even for small-scale or poor farmers. Considering freshwater prawn as a key species, the combination of these approaches raised pond productivity by efficient nutrient utilization above levels obtained through applying each of these techniques independently which further enhanced environmental and economical sustainability (Asaduzzaman et al., 2008, 2009a,b, 2010; Haque et al., 2015). This combined technology has been referred to as C/N Controlled Periphyton-based (C/N-CP) system.

In the recent years, the introduction of GIFT tilapia has added a new era for tilapia farming in Bangladesh. Its performance was found to be significantly superior to that of other tilapias in many respects (Hussain, 2009). Interest on polyculture of freshwater prawn with fish, especially tilapia, has been growing during last few years (New, 2005). It is reported that prawn/tilapia polyculture increased total pond productivity by 81% without impacting

prawn production (Tidwell et al., 2000a). Danaher et al. (2007) found higher total production, higher average weight, and more efficient feed conversion of prawn in polyculture with tilapia where tilapia was confined in cages than prawns raised in monoculture in the same ponds. In Bangladesh, the mixed culture of GIFT strain of Nile tilapia (*Oreochromis niloticus*) and freshwater prawn (*Macrobrachium rosenbergii*) have been found promising (Uddin, 2007). Asaduzzaman et al. (2009a,b, 2010) conducted several experiments on the C/N controlled periphyton-based system using monosex male tilapia as co-species with freshwater prawn. This species has been found to be compatible with freshwater prawn leading to a promising production due to its faster growth rate, more efficient utilization of natural feed and bioturbation effect. Haque et al. (2015) noted similar findings in their C/N controlled periphyton-based research.

Red tilapia, a hybrid mutant of *O. niloticus* and *O. mossambicus*, is a potential cultivable species, and has enormous demand all over the world. This is due to its attractive look, good taste, ease to culture, high adaptation capability to adverse environmental conditions, high resistance to disease, omnivorous feeding behavior, short generation time as well as faster growth (Islam et al., 2006). Despite the improved growth performance of the GIFT strain, the red tilapia (*Oreochromis* spp.) is still the dominant species (85%) farmed in Malaysia due to its preferred red coloration by local consumers with high market demand (Ng and Hanim, 2007; Teoh et al., 2011). Considering its potentials, Bangladesh Fisheries Research Institute (BFRI) introduced this tilapia hybrid in Bangladesh in 1998 from Asian Institute of Technology (AIT), Bangkok, Thailand (Hussain et al., 2004). The culture of this fish has not yet been started extensively throughout the county. Therefore, to popularize this red strain of tilapia, on-farm research works with farmers' participation are necessary. Secondly, most aquaculture techniques are tested in on-station research ponds with short culture period which might not produce similar results in on-farm ponds. The market demands and prices of freshwater prawn and fish are heavily dependent on size/weight of the fish/prawn. Therefore, longer culture period to grow culture animals up to market size is important to determine a realistic profitability of any aquaculture practice. In addition, profitability depends on stocking density of cultured species, their size at stocking and type of culture technique. Considering these aspects, the present experiment was undertaken to compare the relative production performance and profitability between red tilapia hybrid and GIFT strain of Nile tilapia in previously tested C/N controlled periphyton-based freshwater prawn system in farmers' ponds. As the technology of sex-reversed male tilapia for red hybrid had not yet been established in Bangladesh so far, and is not available throughout the country, therefore, in order to continue the resemblance, GIFT strains of Nile tilapia was taken as a replacement for the monosex male tilapia (previously tested with prawn in C/N controlled periphyton-based freshwater prawn system) to compare with Red tilapia. The effects of these two strains of tilapia on pond ecology including water quality, plankton, periphyton and benthic invertebrates were also investigated.

2. Materials and methods

2.1. Experimental design

The trial was conducted using the completely randomized block design into two treatments namely, red tilapia (treatment TR) and GIFT tilapia (treatment TG), with three replications in each treatment. In the TR treatment, freshwater prawn (*M. rosenbergii*) and hybrid red tilapia (Mutant, *O. niloticus* x *O. mossambicus*) strain and in the TG treatment freshwater prawn and mixed sex GIFT tilapia (*O. niloticus*) strain were stocked at the same density as prawn

30,000 ha⁻¹ and tilapia 10,000 ha⁻¹. Feed with maize flour for prawn and bamboo branches (*kanchi*) were provided in all treatments for maintaining C/N ratio of 20 and growing periphyton, respectively.

2.2. Experimental site and pond preparation

The experiment was carried out at the farmers' ponds in Bailor under Trishal upazilla of Mymensingh district for 152 days (15 June to 14 November 2009). Six rectangular ponds of varying sizes (400–880 m²) with an average area of 727 m² and an average depth of 1.5 m each were used for this experiment. The ponds were rain-fed and fully exposed to prevailing sunlight. The pond banks were well protected and covered with grasses. Aquatic vegetation was manually cleaned before starting the experiment. All unwanted fishes were eradicated by applying rotenone at 36.5 kg ha⁻¹. Lime (CaCO₃) was applied at 250 kg ha⁻¹ on day 1. On day 4, 15 bamboo branches per m² (mean diameter of 2.8 cm) were posted vertically into the bottom mud, excluding a 0.5 m wide free perimeter. This resulted in an average additional substrate surface area of 1067 m² for periphyton development equaling 147% of the pond surface area. On day 7, ponds were fertilized with urea and triple super phosphate (TSP) each at 50 kg ha⁻¹. Ponds were left 10 days to allow plankton development in water column and periphyton growth on substrates, and subsequently stocked with prawn and fishes.

2.3. Stocking and pond management

The juvenile of freshwater prawn (individual weight 2.47 g), fingerlings of hybrid red tilapia (individual weight 12.10 g) and GIFT tilapia strains (individual weight 11.98 g) procured from a nearby commercial hatchery were stocked in the ponds following the experimental design. After stocking, ponds were also fertilized fortnightly following the same dose as applied before stocking to maintain steady natural food abundance. A locally formulated (15% fish meal, 20% soybean meal, 20% mustard oil cake, 20% maize flour, 20% rice bran, 4% molasses and 1% vitamin-mineral premix) and prepared pellet feed (2 mm size) containing about 17% protein (dry matter basis) with a C/N ratio close to 15:1 was used. The feed was applied considering the body weight of prawn only after each sampling at a daily feeding rate of 10% body weight at the beginning the experiment (up to 30 days), and assuming 80% survival; feed application was gradually reduced to 2% body weight at the end of the culture period. Feed was distributed uniformly over the pond-surface twice daily at 07:00 and 18:00 h. Individual weights of minimum 10% of initially stocked prawn in numbers were taken at monthly interval to estimate the biomass and adjust the feeding rate. The prawns were sampled using a cast net after eliminating some bamboo side shoots, which were re-positioned after each sampling.

Locally purchased maize flour was used as the carbohydrate source for manipulating the C/N ratio. The analyzed proximate composition of feed and maize flour is given in Table 1. Approximately 0.82 kg maize flour was applied for each kg of formulated feed to raise the C/N ratio to 20:1. The pre-weighed maize flour was mixed with pond water in a beaker and evenly distributed over the pond-surface directly after the application of feed at 07:00 and 18:00 h.

2.4. Freshwater prawn/tilapia harvesting and estimation of yield parameters

Freshwater prawn and tilapia were harvested after draining the ponds using a shallow pump. The individual length (wooden measuring board; precision 0.1 cm) and individual weight (Denver-xp-3000, Denver Instrument Company, Arvada, CO, USA;

precision = 0.1 g) were recorded. Specific growth rate (SGR), feed conversion ratio (FCR) and net yields were calculated as follows:

$$\text{SGR}(\% \text{ bw day}^{-1}) = [(\text{Ln final weight} - \text{Ln initial weight}) \times 100] / \text{Culture periods (days)}$$

$$\text{FCR (prawn only)} = \text{Feed applied (dry weight)} / \text{Live weight gain}$$

$$\text{Net yield} = \text{Total biomass at harvest} - \text{total biomass at stocking}$$

2.5. Determination of water quality parameters

All water quality parameters viz temperature, dissolved oxygen, pH, transparency, total alkalinity, nutrients, NO₂-N, NO₃-N, NH₃-N and PO₄-P concentrations were determined according to Haque et al. (2015).

2.6. Estimation of plankton and benthic macroinvertebrates

Plankton and benthic macroinvertebrates were estimated and calculated as described by Haque et al. (2015).

2.7. Taxonomic composition and biomass of periphyton

From each pond, three *kanchi* poles were selected randomly and three 2 × 2 cm samples of periphyton were taken at each of three depths (25, 50 and 75 cm below water surface) per pole at monthly interval starting after 7 days of substrate installation. One of the three samples from three poles and three depths per pond per sampling day were taken for dry matter (DM), ash and ash free dry matter (AFDM) analysis. The other two samples were used for chlorophyll *a* determination and taxonomic identification. Periphyton biomass and autotrophic index were analyzed following APHA (1992). Periphytic algae were calculated using a binocular microscope (Olympus, M-4000D, Tokyo, Japan) as described by Azim et al. (2001).

2.8. Economic analysis

An economical analysis was performed to estimate the net return and benefit-cost ratio in both treatments. The following equation was used:

$$R = I - (\text{FC} + \text{VC} + \text{li})$$

Where, R = net return, I = income from prawn and tilapia sale, FC = fixed/common costs, VC = variable costs and li = interest on inputs. The benefit cost ratio was determined by following equation:

$$\text{Benefit cost ratio (BCR)} = \text{Total net return} / \text{Total input cost}$$

The wholesale price per kg of prawn, Red and GIFT tilapia were 400, 90 and 100 taka, respectively. The prices of inputs, prawn and fish correspond to the local (Mymensingh) wholesale market prices in June to October 2009 and are expressed in Bangladeshi taka (1US\$ = 70 BDT).

2.9. Statistical analysis

Independent samples T-Test was performed to compare water quality parameters, plankton, periphyton and benthic macroinvertebrates data, and growth and production of prawn and

Table 1
Proximate composition of the prepared feed and maize flour (dry weight basis).

Component	Moisture (%)	Protein (%)	Lipid (%)	Fiber%	Ash (%)	Carbohydrate (%)
Prepared feed	11.2	17.2	9.3	5.7	15.5	41.0
Maize flour	11.0	7.7	4.7	5.4	1.1	70.0

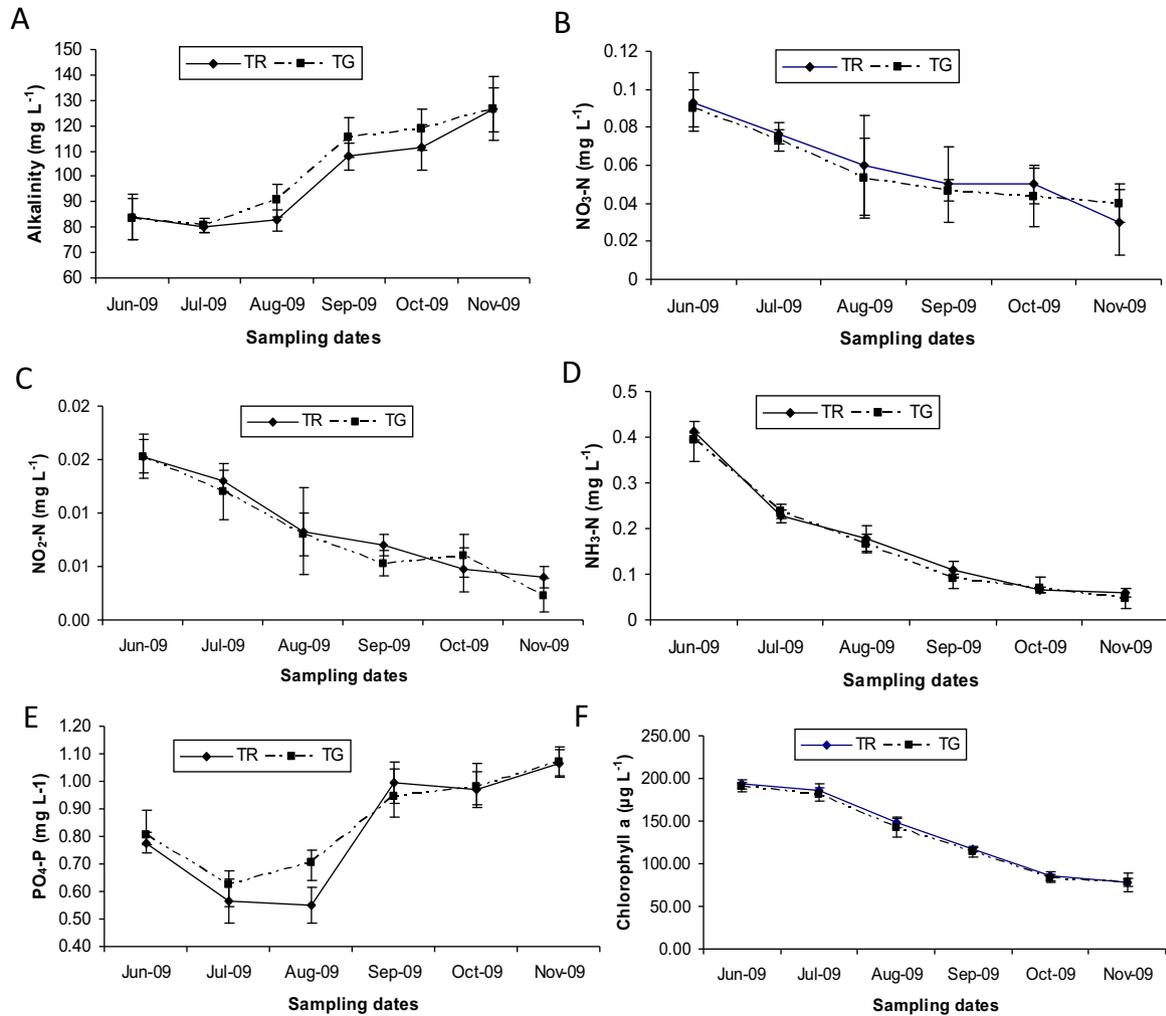


Fig. 1. Mean concentration of (A) total alkalinity, (B) nitrate, (C) nitrite, (D) ammonia, (E) phosphate and (F) chlorophyll *a* in two treatment ponds over the sampling period (mean \pm SD, N = 3).

tilapias as well as economics. Survival and percent data were analyzed using arcsine-transformed data, but percent values were reported. Temporal effects of water quality parameters, plankton, periphyton and benthic macro invertebrates data were compared in a repeated measure analysis of variance (ANOVA) where treatments as the main factors and time as the sub-factors (Gomez and Gomez, 1984). If the main effect was found significant, the ANOVA was followed by Tukey's Test. The assumptions of normal distribution and homogeneity of variances were checked before analysis. All statistical tests were carried out at a 5% level of significance using SPSS (Statistical Package for Social Science) version 16.0.

3. Results

3.1. Water quality parameters

Water quality parameters (Table 2) did not vary significantly between the treatments. Temporal variations were

observed (Fig. 1), with increasing alkalinity and phosphate and decreasing nitrogenous species and chlorophyll *a* throughout the culture period. The highest mean temperature (31.17 °C) was recorded on the last week of June in the TG treatment whereas the lowest mean temperature (26.62 °C) was recorded on the 2nd week of November in the TR treatment. The highest mean transparency (53 cm) was found in the 2nd week of November in TG and the lowest (27 cm) was in the 2nd week of June in TR. The water pH ranged from 7.08 to 7.88 throughout the study. The highest mean dissolved oxygen concentration (7.27 mg L⁻¹) was recorded in the 2nd week of September and the lowest (3.43 mg L⁻¹) was recorded in the 1st week of August in TR. The highest total alkalinity was observed in the 2nd week of November in TG and the lowest was observed in the 2nd week of June in TR.

No inorganic nitrogenous compounds (NH₃-N, NO₃-N and NO₂-N) were affected by the treatment. However, the temporal effects for all nitrogenous compounds were observed with the decreasing trends towards the end of the study (Fig. 1). There

Table 2
Mean (\pm SD) values of recorded water quality parameters.

Parameters	N	Treatments		Level of significance at 5%
		TR	TG	
Temperature ($^{\circ}$ C)	66	29.30 \pm 1.16	29.35 \pm 1.12	NS
Transparency (cm)	66	40.42 \pm 6.74	41.59 \pm 6.78	NS
pH range	66	7.08–7.88	7.26–7.78	NS
Dissolved oxygen (mg L^{-1})	66	5.26 \pm 0.94	5.19 \pm 0.96	NS
Total Alkalinity	18	98.72 \pm 18.87	102.56 \pm 19.97	NS
Total $\text{NH}_3\text{-N}$ (mg L^{-1})	18	0.060 \pm 0.025	0.058 \pm 0.021	NS
$\text{NO}_3\text{-N}$ (mg L^{-1})	18	0.009 \pm 0.005	0.008 \pm 0.005	NS
$\text{NO}_2\text{-N}$ (mg L^{-1})	18	0.18 \pm 0.13	0.17 \pm 0.12	NS
$\text{PO}_4\text{-P}$ (mg L^{-1})	18	0.82 \pm 0.22	0.86 \pm 0.17	NS
Chlorophyll <i>a</i> ($\mu\text{g L}^{-1}$)	18	134.87 \pm 46.89	131.10 \pm 45.76	NS

N = number of samples; NS = Means are not significantly different ($P > 0.05$) based on Tukey's test. TR, red tilapia; TG, GIFT tilapia.

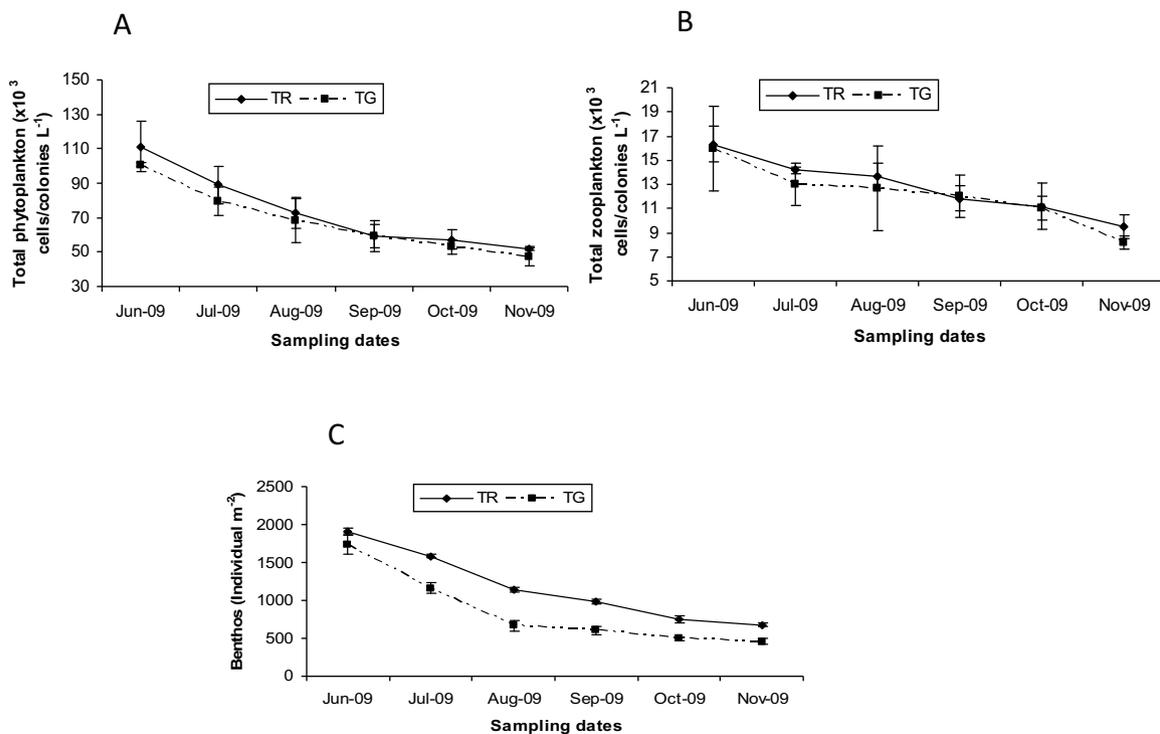


Fig. 2. Abundance of (A) phytoplankton, (B) zooplankton and (C) benthos in two treatment ponds over the sampling periods (mean \pm SD, N = 3).

were no treatment effects for $\text{PO}_4\text{-P}$, but a temporal effect was observed with the increasing trends throughout the study period. A temporal effect was on the chlorophyll *a* with the decreasing trend over the period (Fig. 1).

3.2. Abundance of plankton and benthos

The plankton population did not vary significantly between the treatments (Table 3) but significant variations on different sampling dates (temporal effects) were observed on the abundance of plankton. Phytoplankton represented about 85% of the total plankton in number in both treatments. The plankton communities consisted of four major groups of phytoplankton and two groups of zooplankton in both treatments. Thirty nine genera of phytoplankton belonging to Chlorophyceae (16), Bacillariophyceae (12), Cyanophyceae (9), and Euglenophyceae (2) were found. Chlorophyceae followed by Bacillariophyceae was the most dominant group in number of genera in each treatment. The dominant genera were *Chlorella*, *Oocystis*, *Pediastrum*, *Chaetophora* and *Scenedesmus* (Chlorophyceae); *Fragillaria*, *Cyclotella*, *Navicula*, *Coscinodiscus*,

Tabellaria and *Melosira* (Bacillariophyceae); *Microcystis*, *Anabaena*, *Gleocapsa*, *Oscillatoria* and *Gomphosphaeria* (Cyanophyceae); and *Euglena* and *Phacus* (Euglenophyceae). Sixteen genera of zooplankton including seven of Rotifera and nine of Crustacea were also identified. The abundance of total phytoplankton and total zooplankton decreased gradually in both treatments throughout the experimental period (Fig. 2).

The benthic invertebrates were divided into Chironomidae, Oligochaeta, Mollusca and unidentified organisms. Chironomidae followed by Oligochaeta was the most dominant group among benthic invertebrates in both treatments. The mean abundance of benthic invertebrates with their major groups, except Chironomidae varied significantly between the treatments (Table 3) with higher numbers recorded in red tilapia ponds. A significant decreasing trend was also observed for the number of total benthic invertebrates and their major groups with few exceptions during the experimental period (Fig. 2).

Table 3
Abundance of plankton and benthic invertebrates (mean \pm SD) with different groups identified in two treatments.

Variables	Treatments			Level of significance at 5%
	N	TR	TG	
Plankton ($\times 10^3$ cells or colonies l^{-1})				
Bacillariophyceae	18	15.08 \pm 5.97	13.47 \pm 3.65	NS
Chlorophyceae	18	30.69 \pm 10.78	28.19 \pm 11.43	NS
Cyanophyceae	18	22.44 \pm 5.32	21.53 \pm 6.12	NS
Euglenophyceae	18	5.36 \pm 3.03	4.89 \pm 2.79	NS
Total phytoplankton		73.58 \pm 22.77	68.08 \pm 19.51	NS
Rotifera	18	6.58 \pm 1.4	6.28 \pm 1.84	NS
Crustacea	18	6.19 \pm 1.58	5.86 \pm 1.94	NS
Total Zooplankton	18	12.78 \pm 2.5	12.14 \pm 3.1	NS
Total plankton	18	86.36 \pm 24.96	80.22 \pm 21.63	NS
Benthic invertebrates (Individual m^{-2})				
Chironomidae	18	526.75 \pm 227.20	395.06 \pm 266.86	NS
Oligochaeta	18	330.86 \pm 117.97 ^a	246.09 \pm 121.55 ^b	*
Mollusca	18	235.39 \pm 97.07 ^a	164.61 \pm 72.91 ^b	*
Un-identified	18	81.48 \pm 33.03 ^a	49.38 \pm 34.09 ^b	*
Total benthic invertebrates	18	1,174.49 \pm 456.62 ^a	855.14 \pm 474.10 ^b	*

NS = Values are not significantly (different based on Independent-Samples T-test).

* Values with different superscript letters in the same row indicate significant difference ($P < 0.05$).

Table 4
Abundance and biomass of periphyton (mean \pm SD) assessed in two treatments.

Variables	Treatments		Level of significance at 5%
	TR	TG	
Periphytic abundance ($\times 10^3$ cells or colonies cm^{-2})			
Bacillariophyceae	21.15 \pm 3.64 ^a	17.69 \pm 2.54 ^b	*
Chlorophyceae	19.71 \pm 3.44	18.60 \pm 3.88	NS
Cyanophyceae	7.86 \pm 0.56 ^a	6.26 \pm 1.09 ^b	*
Euglenophyceae	1.16 \pm 0.61	1.08 \pm 0.61	NS
Total phyto-periphyton	49.88 \pm 5.76 ^a	43.73 \pm 5.57 ^b	*
Rotifera	2.07 \pm 0.29 ^a	1.85 \pm 0.27 ^b	*
Crustacea	0.41 \pm 0.27	0.34 \pm 0.29	NS
Total zoo-periphyton	2.48 \pm 0.51	2.19 \pm 0.45	NS
Total periphyton	52.36 \pm 6.14 ^a	46.60 \pm 5.66 ^b	*
Quantitative biomass			
Dry matter ($mg\ cm^{-2}$)	2.76 \pm 0.46 ^a	1.85 \pm 0.88 ^b	*
Ash ($mg\ cm^{-2}$)	1.13 \pm 0.28 ^a	0.80 \pm 0.40 ^b	*
Ash free dry matter ($mg\ cm^{-2}$)	1.64 \pm 0.48 ^a	1.05 \pm 0.63 ^b	*
Chlorophyll- <i>a</i> ($\mu g\ cm^{-2}$)	11.57 \pm 3.30 ^a	8.73 \pm 4.31 ^b	*
Autotrophic index	145.57 \pm 43.22	120.22 \pm 44.45	NS

NS, Values are not significantly different.

* Values with different superscript letters in the same row indicate a significant difference ($P < 0.05$) based on Independent-Sample T-Test.

3.3. Periphyton composition and biomass

Forty nine genera of algal-periphyton belonging to Chlorophyceae (22), Bacillariophyceae (13), Cyanophyceae (10), and Euglenophyceae (2), and eight genera of zoo-periphyton belonging to Rotifera (7) and Crustacea (1) were identified as periphytic communities in both treatments during the experiment (Table 4). Chlorophyceae was dominant in species number and Euglenophyceae was the least abundant group of periphytic algae in both treatments. Rotifera was found to be a more abundant zoo-periphytonic group than crustacean in both treatments. *Oedogonium*, *Palmella*, *Botriococcus*, *Chaetophora*, *Sphaerocystes*, *Gonatozygon* and *Chlorella* (Chlorophyceae); *Synedra*, *Cyclotella*, *Diatoma*, *Navicula*, *Coscinodiscus* and *Fragillaria* (Bacillariophyceae); *Oscillatoria*, *Aphanocapsa*, *Anacystis* and *Microcystis* (Cyanophyceae); *Euglena* and *Phacus* (Euglenophyceae); *Karatella*, *Brachionus* and *Asplanchna* (Rotifera) and larval Nauplius (Crustacea) were the dominant genera in both treatments. The total abundance of periphyton population with Bacillariophyceae, Cyanophyceae and Rotifera groups varied significantly between the treatments (Table 4). Their abundance were also affected significantly with higher values in TR than TG by different sampling dates (temporal effect) and a decreasing trend was observed for their numbers

over the experimental period. The periphytic compositions differed from plankton composition. *Drapamaldia*, *Oedogonium*, *Palmella*, *Sphaerocystis* and *Stigeoclonium* were the different periphytic genera which were not common in the plankton communities.

The DM, Ash, AFDM and Chlorophyll *a* contents varied significantly between treatments (Table 4) as well as among different sampling months during the experimental period. A decreasing trend was observed for the DM, AFDM (except September in TR) and chlorophyll *a* content (except October in TG) over the culture period. The Autotrophic Index (AI) values found were 74.98–221.53 and 71.11–235.96 in TR and TG, respectively.

3.4. Growth and yield parameters of freshwater prawn and tilapia

Individual harvesting weight, individual weight gain, specific growth rate, FCR, gross and net yields of prawn did not vary significantly between the treatments, while the same parameters of GIFT tilapia in TG treatment were significantly higher than red tilapia in TR treatment (Table 5). In addition, a significantly higher combined gross and net yield of prawn and tilapia were observed in TG (15.63 and 8.94%, respectively) than TR treatment (Table 5).

Table 5
Growth and production performance (Mean \pm SD) of prawn and tilapia in two treatments during a 152-day culture period.

Variables	Treatments		Significance level at 5%
	TR	TG	
Prawn			
Individual stocking weight (g)	2.47 \pm 0	2.47 \pm 0	NS
Individual harvesting weight(g)	40.53 \pm 2.51	42.93 \pm 1.35	NS
Individual weight gain (g)	38.06 \pm 2.51	40.46 \pm 1.35	NS
Specific growth rate (% bw d ⁻¹)	1.84 \pm 0.04	1.88 \pm 0.02	NS
Feed conversion ratio	2.34 \pm 0.09	2.27 \pm 0.06	NS
Survival (%)	75.86 \pm 2.33	78.97 \pm 0.76	NS
Gross yield (kg ha ⁻¹ 152 d ⁻¹)	922.97 \pm 74.81	1,017.10 \pm 28.84	NS
Net yield (kg ha ⁻¹ 152 d ⁻¹)	866.76 \pm 73.58	958.58 \pm 28.93	NS
Tilapia			
Individual stocking weight (g)	12.10 \pm 0.00	11.98 \pm 0.00	NS
Individual harvesting weight(g)	200.33 \pm 5.03 ^b	211.33 \pm 3.51 ^a	*
Individual weight gain (g)	188.23 \pm 5.03 ^b	199.35 \pm 3.51 ^a	*
Specific growth rate (% bw d ⁻¹)	2.89 \pm 0.017 ^b	2.93 \pm 0.01 ^a	*
Survival (%)	81.36 \pm 3.24 ^b	91.57 \pm 4.41 ^a	*
Gross yield (kg ha ⁻¹ 152 d ⁻¹)	1,630.32 \pm 87.61 ^b	1,935.26 \pm 102.25 ^a	*
Net yield (kg ha ⁻¹ 152 d ⁻¹)	1,531.87 \pm 84.07 ^b	1,825.56 \pm 97.26 ^a	*
Combined effects			
Gross yield (kg ha ⁻¹ 152 d ⁻¹)	2,553.29 \pm 144.79 ^b	2,952.36 \pm 130.92 ^a	*
Net yield (kg ha ⁻¹ 152 d ⁻¹)	2,398.63 \pm 139.18 ^b	2,784.14 \pm 126.02 ^a	*
Contribution to yield (%)			
Prawn gross yield	36.13 \pm 1.53	34.46 \pm 0.57	NS
Prawn net yield	36.11 \pm 1.64	34.45 \pm 0.53	NS
Tilapia gross yield	63.86 \pm 1.53	65.53 \pm 0.57	NS
Tilapia net yield	63.88 \pm 1.64	65.55 \pm 0.54	NS

NS, Values are not significantly different ($P > 0.05$).

bw, body weight; d, day.

*Values with different superscript letters in the same row indicate a significant difference based on independent samples T-Test.

Table 6
Comparison of economics (mean \pm SD, N = 3) between two treatments during the culture period (152 days) calculated on the basis of 1 ha pond.

Variables	Amount	Price rate (BDT)*	Treatments	
			TR	TG
Fixed/Common cost				
Land rental cost	1 ha	20,000 ha ⁻¹ y ⁻¹	10,000	10,000
Rotenone	12.5 kg	250 kg ⁻¹	3000	3000
Lime	250 kg	10 kg ⁻¹	2500	2500
Urea	50 kg	10 kg ⁻¹	500	500
TSP	50 kg	25 kg ⁻¹	1250	1250
Substrates (reuse-5 times)	150000 P	1 piece ⁻¹	150,000	150,000
Fuel cost	500 units	4 unit ⁻¹	2000	2000
Labor	50 man-day	120 man-day ⁻¹	6000	6000
Prawn juveniles	30000 ha ⁻¹	4 juvenile ⁻¹	120,000	120,000
Tilapia juveniles	10,000 ha ⁻¹	2 juvenile ⁻¹	20,000	20,000
Subtotal			315,250	315,250
Variable cost				
Feed		25 kg ⁻¹	3,822.18 \pm 669.13	3,942.35 \pm 1452.22
Maize		15 kg ⁻¹	2,063.46 \pm 357.75	2,223.21 \pm 818.96
Subtotal			5,885.64 \pm 1005.07	6,165.57 \pm 2271.18
Total				
Bank Interest on costs (5 months)		12% annually	16,056.78 \pm 50.25	16,070.78 \pm 113.56
Total inputs			337,192.4 \pm 1055.32	337,486.4 \pm 2384.74
Financial returns				
Prawn sale		400 kg ⁻¹	369,188.83 \pm 29926.39	406,841.12 \pm 11537.10
Red/GIFT tilapia sale		90/100 kg ⁻¹	146,728.75 \pm 7885.13 ^b	193,525.75 \pm 10225.20 ^a
Total returns (BDT)			515,917.58 \pm 35,137.67 ^b	600,366.87 \pm 21,719.38 ^a
Total net returns (BDT)			178,725.15 \pm 34,880.63 ^b	262,880.52 \pm 23,571.73 ^a
Benefit cost ratio (BCR)			0.55 \pm 0.10 ^b	0.82 \pm 0.07 ^a

Φ Mean values with different superscript letters in the same row indicate a significant difference based on independent samples T-Test.

TSP, triple super phosphate.

*BDT, Bangladesh Taka (70 BDT = 1US\$).

3.5. Economics

Although there was no significant difference in the total expenditure between two treatments, a significantly higher ($P < 0.05$)

gross earning as well as net profit with the BCR of 0.82 was incurred from TG than TR with BCR = 0.55 (Table 6). Substrates were the most expensive inputs (about 45% of total costs) in both treatments followed by prawn juveniles (35.59%). Feed cost was 3.14% higher in

TG than TR. A significantly higher earnings was obtained from GIFT tilapia sale in TG than red tilapia sale in TR treatment. Net return in TG treatment was US\$ 3755 as compared to US\$ 2553 in TR treatment. However, 68–72% of total income was obtained from prawn sale, while the rest came from tilapia.

4. Discussion

4.1. Water quality parameters

A number of water quality parameters such as, all physical, chemical and biological factors influencing the beneficial use of water were recorded in experimental ponds. Most of the water quality parameters were within the acceptable range for prawn and tilapia throughout the experiment (Boyd and Zimmermann, 2000; New, 2002). The water temperature was within the ideal range for freshwater prawn culture (Boyd and Zimmermann, 2000; Kunda et al., 2008; Rahman et al., 2010; Haque et al., 2015). Water transparency, roughly indicating the presence or absence of natural food particles of fish as well as productivity of a waterbody were found to fluctuate between 27 and 53 cm in both treatments. These exceeded the upper limit of the recommended range (15–40 cm) as suggested by Boyd (1992), but were more or less comparable to the values of Bangladeshi ponds (Asaduzzaman et al., 2009a; Rahman et al., 2010; Haque et al., 2015). The measured dissolved oxygen concentration and pH were within the suitable range for freshwater prawn and tropical fish culture (Boyd and Zimmermann, 2000; New, 2002; Kunda et al., 2008; Asaduzzaman et al., 2008, 2009a,b, 2010; Rahman et al., 2010). The occasional low DO concentration (3.43 mg L^{-1} in the 1st week of August in TR) was not at a stress level for freshwater prawn as well as tilapia. Freshwater prawn become stressed at a dissolved oxygen level below 2 mg L^{-1} (Rogers and Fast, 1988), and when it declines below 1 mg L^{-1} , prawn become exhausted with serious physiological effects leading to suffocation (Boyd and Zimmermann, 2000). On the contrary, tilapia are generally more tolerant to low dissolved oxygen concentration, even to 0.1 mg L^{-1} (Magid and Babiker, 1975) but optimum growth is attained at concentrations $>3 \text{ mg L}^{-1}$ (Ross, 2000).

The observed mean values of total alkalinity with the ranges in all treatments were very similar to that as suggested by Boyd (1990) and are also comparable to the findings of other authors in this region (Kunda et al., 2008; Asaduzzaman et al., 2010; Rahman et al., 2010). The observed increase of total alkalinity over time may be due to the application of increasing amounts of maize flour to maintain high C/N ratio. This practice increase dissolved organic carbon in the water, and organic anions add alkalinity to the water (Wetzel and Likens, 1991).

The observed ranges of $\text{PO}_4\text{-P}$ concentrations in both treatments were more or less similar to Bangladeshi ponds (Asaduzzaman et al., 2008, 2009a,b, 2010; Rahman et al., 2010). An increasing trend of $\text{PO}_4\text{-P}$ with few exceptions in treatment TR over time might be attributed to inducing nutrient release by the tilapia driven re-suspension with increasing body size. Tilapia release nutrients from the accumulated organic matter of the sediment into the water phase through the mud-water exchange mechanism. This enhances the overlying water $\text{PO}_4\text{-P}$ concentration (Saha and Jana, 2003; Asaduzzaman et al., 2010). The mean values of chlorophyll *a* in both treatments were comparable to the findings of some authors in the same region (Kunda et al., 2008; Asaduzzaman et al., 2008, 2009a,b, 2010; Rahman et al., 2010). A decreasing trend of Chlorophyll *a* overtime might be due to the low concentrations of inorganic nitrogen which limit the algal biomass (MacLean et al., 1994). Other possibility is consumption by the growing biomass of fish.

4.2. Abundance of plankton and benthic invertebrates, periphyton composition and biomass production

Phytoplankton, zooplankton and benthos are the key natural food items in any aquatic environment influencing fish production. Their abundance in the culture system is governed by a number of management factors, such as fish species combination, stocking density and ratio, quality and quantity of nutrient inputs (Diana et al., 1997). The species composition of plankton identified in the present experiment was representative of the prawn farming ponds and rice fields in Bangladesh (Uddin, 2007; Kunda et al., 2008; Wahab et al., 2008; Asaduzzaman et al., 2009b, 2010; Rahman et al., 2010). Periphyton is immensely important in an aquatic system due to providing community structure and primary productivity that supports a wide range of aquatic organisms (Azim et al., 2005). It comprises bacteria, fungi, protozoa, phytoplankton, zooplankton, benthic organisms and a range of other invertebrates and their larvae (Azim et al., 2001). In the present study, phyto-periphyton and zoo-periphyton were only recorded as periphyton. The periphyton abundance was similar as found in prawn/fish ponds (Azim et al., 2004; Uddin, 2007; Asaduzzaman et al., 2009b). The mean values of periphyton biomass in terms of DM, Ash, AFDM and Ash content were comparable to the values reported by some authors in this region (Azim et al., 2001; Uddin et al., 2006; Asaduzzaman et al., 2008, 2009a,b, 2010; Haque et al., 2015). The significant temporal effects as well as the decreasing trends of total phytoplankton, total phyto-periphyton (except higher on August and September in TG and TR, respectively), periphytic biomass with an exception to some extent in both treatments over time might be due to increased grazing pressure by increased biomass of both tilapias (Haque et al., 2015). Tilapia affected phytoplankton directly by grazing and indirectly by nutrient re-suspension. The direct effect was more pronounced than the indirect one, indicating that tilapia had a higher grazing pressure on phytoplankton (Asaduzzaman et al., 2009b) over the time. It is reported that tilapias are omnivorous capable of feeding on benthic and attached (periphyton) algal and detrital aggregates (Bowen, 1982; Azim et al., 2003b,c). The more or less similar pattern of trends of periphytic biomass were observed by Asaduzzaman et al. (2008, 2009a,b, 2010) and Haque et al. (2015) in tilapia added treatments of CN-CP prawn culture ponds. It is needed to graze periphytic algae constantly and kept at low biomass to maintain their high productivity (Huchette et al., 2000).

The lower mean values and higher decreasing trends of phytoplankton, benthos, total phyto-periphyton with bacillariophyceae and cyanophyceae and periphytic biomass in TG might be an indication that the grazing pressure was considerably higher in GIFT tilapia than red tilapia. This might be reflected as a significantly higher individual growth rate, wet gain, specific growth rate, gross yield as well as net yield of GIFT tilapia than Red Tilapia. Ng and Hanim (2007) found that GIFT tilapia was more efficient in utilizing dietary protein compared to red hybrid tilapia.

The observed significant temporal effects as well as the decreasing trends of total zooplankton, benthos and total zoo-periphyton over time might be attributed to: (a) increased grazing pressure by increased biomass of prawn or (b) partially by increased tilapia biomass due to less preference on zooplankton over phytoplankton or (c) entirely by tilapia biomass as a major component of their diet. Freshwater prawn preferred to forage on animals like trichopterans, chironomids, oligochaetes, nematodes, gastropods and zooplankton in ponds (Tidwell et al., 1997). Uddin et al. (2007) reported that electivity indices of tilapia were negative for all zooplankton and positive for all phytoplankton. It is also claimed that green, brown (diatoms) and blue green algae as well as numerous species of rotifers, cladocerans and copepods are major components of the tilapia diet (Cuvin-Aralar, 2003). Tilapia fry and juvenile

stages feed on zooplankton and then shift to filter feeding at the later stages (Bowen, 1982).

The higher values of phyto-periphyton on August and September might be due to less grazing pressure of tilapia in those months. The more or less similar patterns of trends for phyto-periphyton community were also observed by Azim et al. (2004) and Uddin (2007). The similar abundance of periphytic zooplankton in both treatments indicated that the zooplankton communities were less preferable for the tilapias or escaped predation (Asaduzzaman et al., 2009b). The significant lower values of total periphyton in TG than TR were influenced by the total phyto-periphyton indicating that photosynthetic organisms or algae were the major community of periphyton, which were more or less reflected to the range of Autotrophic Index (APHA, 1992).

The mean values of autotrophic index with ranges were comparable to the findings of other authors in this region (Azim et al., 2001; Asaduzzaman et al., 2008, 2009a,b, 2010). The low range of AI values in both treatments might be continuous grazing pressure of periphyton mass (Huchette et al., 2000) by both tilapias. It is stated that AI values between 100 and 200 are considered as algae dominating periphytic matter (APHA, 1992). The more/less observed similar ranges of AI values in this experiment indicated that the periphytic biofilms were algae dominating one.

4.3. Growth and yield parameters of prawn and tilapia

Growth and yield parameters including individual harvesting weight, individual weight gain, specific growth rate, FCR, survival (%), gross and net yields of prawn were similar in both treatments. That may be due to the fact that the factor was only the tilapia strain in both treatments in CN-CP prawn farming system. Nevertheless a comparatively higher production of gross as well as net yields of prawn (10.2 and 10.6%, respectively) in GIFT tilapia added treatment might be due to relatively higher consumption of natural food (zooplankton and benthos) directly or indirectly. The indirect effect might be greater suspension of sediment by GIFT tilapia than red tilapia over the time facilitating further feeding of freshwater prawn. It is reported that prawn in their habitat prefer to forage on animals like trichopterans, chironomids, oligochaetes, nematodes, gastropods and zooplankton in their natural habitats (Tidwell et al., 1997). Prawn predation on benthos especially chironomids may be facilitated due to digging and sieving of sediments by tilapia (Asaduzzaman et al., 2010). It is reported that approximately 2.5 higher yields were obtained from stirred ponds compared to unstirred ones (Costa-Pierce and Pullin, 1989). However, the gross and net productions of freshwater prawn in both treatments were promising with higher values compared to the findings in the prawn farming ponds and rice fields in Bangladesh (Asaduzzaman et al., 2006; Uddin, 2007; Kunda et al., 2008; Asaduzzaman et al., 2009a,b, 2010; Rahman et al., 2010). The reasons of higher production might be as to higher stocking density (3 m^{-2}) with a longer period of time (152 days) and large PL as well as improved culture technique.

The observed significantly higher gross and net yields of GIFT tilapia in TG treatment than red tilapia in TR were influenced by their significantly higher individual harvesting weight, individual weight gain, specific growth rate as well as survival. That might be attributed to a significantly higher grazing of periphyton and benthos by GIFT tilapia than red tilapia over the time. It is reported that growth performance of GIFT tilapia is superior to other tilapias in many aspects (Hussain, 2009). The varying growth of tilapia may be influenced by social and behavioral interactions due to competition for food (Mair, 2002a,b). The higher survival of GIFT tilapia might be due to consumption higher amount of zooplankton and benthos at young stage than red tilapia or might be the difference in genetic factor. Mamaril (2001) reported that an abundance of zooplanktons

and benthos when eaten by young tilapia positively influenced survival in cage-reared fishes, especially tilapias. It is also reported that heavy mortalities of red tilapia during the fry and fingerling rearing appears to be related to genetic factors (Siddiqui and Al-Harbi, 1995). However, the production of tilapia in both treatments was higher to that found by Asaduzzaman et al. (2009a,b, 2010) and Haque et al. (2015) in their C/N controlled periphyton-based prawn culture research, which might be attributed to the difference in species composition, species density, sex, individual stocking weight, feed management, culture area, duration, and so on. The combined gross as well as net yield was satisfactory and those were significantly higher in GIFT tilapia added treatment than the treatment stocked with red tilapia, and might be reflected by the significantly higher growth of GIFT tilapia than red tilapia. That indicated that natural foods in the form of periphyton, benthos as well as plankton compensated the demand of supplementary feed were higher by GIFT tilapia than red tilapia. Both tilapias are regularly observed grazing on substrates for periphyton. Uddin et al. (2006) provided artificial feed considering the biomass of freshwater prawn only in periphyton-based prawn-tilapia ponds so that tilapia can be benefited by consuming natural food grown on there. The combined gross as well as net yield of prawn and tilapia in both treatments was higher than that of Asaduzzaman et al. (2009a,b) and Haque et al. (2015) and might be due to the difference in stocking density, stocking weight, sex, season, culture area, duration, and so on.

4.4. Economics

The estimated net profit in the best-performed treatment (TG) was 3755 US\$ per hectare with a BCR of 0.82 in a 152-day culture period. The net profit with BCR was higher than that of Asaduzzaman et al. (2009a,b, 2010) in their C/N controlled periphyton-based research ponds. That might be attributed to the difference in species composition, species density, sex, individual stocking weight, feed management, culture area, duration, and so on. Although the net freshwater prawn production (36% of the total net production) was less than the GIFT tilapia production (64%) in terms of biomass, its higher sale price contributed about 65% of the total benefit in that treatment confirming that freshwater prawn as the prime species as well as cash crop, and the tilapia might be considered as the secondary/bonus species. In most polyculture systems, there is a target species and some minor species as well. A bonus to the yield of the target species is usually obtained by the yield of those minor species (Garcia-Perez et al., 2000). As the prime species, prawn has a higher international market, the GIFT tilapia was rather of bonus production for household consumption as well as selling in the local market.

5. Conclusion

It may be concluded that GIFT tilapia grazed more and grew better than red tilapia. Therefore, it is a more convenient species than red tilapia in CN-CP based prawn farming system contributing the overall higher production and economic return. Their higher survival and production are influenced by their better utilization efficiency of natural food, especially benthos and periphyton. As the experiment was done with mixed-sex GIFT tilapia and prawn in CN-CP based system, therefore, a further better net benefit could be expected from the result obtained from this experiment by conducting the research through stocking of prawn with sex-reversed male tilapia of GIFT strain in the CN-CP based prawn farming system.

Acknowledgements

This research was financially supported by the Ministry of Science and Technology, Bangladesh. The authors are grateful to the farmers for their kind assistance during sampling and feeding of fishes and prawns. All water quality analyses were performed at the Water Quality and Pond Dynamics laboratory of Bangladesh Agricultural University, Mymensingh, Bangladesh.

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