

## EFFECTS OF ARTIFICIAL SUBSTRATE ON REARING OF *MACROBRACHIUM ROSENBERGII* POST-LARVAE IN POND NET CAGE

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### ABSTRACT

Two experiments were carried out to evaluate the effects of addition of net piece substrates in cages for nursery rearing of freshwater prawn, *Macrobrachium rosenbergii* post-larvae. In the first experiment, net piece substrates were added with an arrangement to increase the surface area of nursery cages to 50% and 75% and compared with cages without any substrate (control). In the second experiment, maintaining surface area increment at 75% in all cages, the net piece substrates were arranged in three different orientations of horizontal, vertical, and 45° angle sloping. Experiments were conducted in a pond of Faculty of Fisheries, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur, Bangladesh for a period of 45 days in June –August of 2015 and 2016. A completely randomized design was followed to assign the treatments with three replications. The 75% area increment group showed significantly higher ( $P < 0.05$ ) growth ( $1.02 \pm 0.09$  g) and survival rate ( $79.43 \pm 1.63\%$ ), compared to 50% area increment ( $0.96 \pm 0.05$  g and  $75.03 \pm 2.45\%$ ) and control group ( $0.79 \pm 0.06$  g and  $66.66 \pm 3.57\%$ ). The vertically oriented substrate group showed significantly lower growth ( $0.95 \pm 0.04$  g) and survival ( $76.33 \pm 1.15\%$ ), compared to the horizontal ( $1.06 \pm 0.05$  g and  $77.12 \pm 1.20\%$ ) and sloping ( $1.16 \pm 0.10$  g and  $79.33 \pm 0.57\%$ ) substrate group. However, there were no differences between horizontal and sloping substrate groups regarding final length, final weight, specific growth rate (SGR), and survival rate except food conversion ratio (FCR). The overall results suggested that modification of net cage nursery structure of *M. rosenbergii* might have significant effects on growth and survival of post-larvae by the manipulation of their behaviour in culture system.

**Keywords:** Net cage nursery, freshwater prawn, behavioural manipulation, growth, survival.

### Introduction

Bangladesh is a riverine country with its vast fisheries resources. Prawn and shrimp production attributes to 3.23% of the country's total gross domestic production (GDP), and the giant freshwater prawn, *Macrobrachium rosenbergii*, contributes a significant portion to it (DoF, 2018). *M. rosenbergii*, which is

indigenous to the South and Southeast Asia, together with Northern Australia and the Western Pacific Islands was successfully domesticated in 1960s (Ling, 1969). Since then prawn culture has been increased dramatically in tropical and subtropical regions due to its growing market demand and culture suitability (D'Abramo *et al.*, 1989). However, expansion

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of prawn farming in Southeast Asian countries is limited by several factors, such as the inadequate supply of quality seeds, high production cost, lack of research regarding appropriate research and extension work in technology development and dissemination (Srinivas and Venkatrayalu, 2016).

Nursing of *M. rosenbergii* is a transitional stage where post-larvae (PL) are reared at high densities to juveniles. Before stocking in grow-out ponds, nursing of PLs for 1-2 months have been identified as an essential step to increase survival rate, growth, food conversion efficiency and productivity (Alam, *et al.*, 1997; Alston, 1989; Carvalho-Filho and Mathias, 1998; Cohen, *et al.*, 1981; New and Singholka, 1985; New, 1990). Net cage nursing system is one of the techniques for nursing the PL, and it is gaining popularity due to its practicability and low cost (Beveridge, 2004; Kiyohara, 2006). For example, using a smaller mesh size of net prevents unwanted species' entry (Cuvin-Aralar *et al.*, 2013). Moreover, prawn culture in net cage facilitates easy access, management, and harvest. Besides, net clogging problem can be avoided due to the short nursery period (45-60 days), making net cage nursing of PL more viable. Several studies reported that prawn's benthic and territorialism nature results in heavy growth dependency on a waterbody's two-dimensional area (Cohen *et al.*, 1981; Tidwell *et al.*, 1998). D'Abramo *et al.* (2006) suggested that the addition of substrate in a pond can provide the prawns with the opportunity to utilize the waterbody's three-dimensional area.

Different types of artificial substrates such as dried tree branches, bamboo pools, gravels, aquatic plants, and shells have been used and

proved to improve growth, survival rate and productivity in both indoor and outdoor prawn culture conditions (Mamun *et al.*, 2010; Tidwell *et al.*, 1998). On the other hand, the effects of the substrate's orientation in the prawn PL nursery system are not apparent. Smith and Sandifer (1975) reported that the horizontally placed substrate group showed a higher survival rate than the vertically placed substrate group. In contrast, Tidwell *et al.* (2002) and Marcus (2002) did not find any significant difference in survival rate or average individual weight of prawn between the horizontal and vertical substrate. No studies have observed the effects of addition and orientation of substrate on the net cage-prawn nursing system. We hypothesized that the increase in net cage surface area by adding substrate and substrate's orientation might significantly affect growth, survival rate, and FCR of prawn PL. With this hypothesis, two experiments were carried out to evaluate whether the substrate's addition and orientation in the net cage increase the growth, survival, and FCR of prawn PL.

## Materials and Methods

Both cage nursery experiments were conducted, for a period of 45 days in June – August covering the years of 2015 and 2016, using an existing pond of 10 decimals with an average water depth of 1.7 m that was located at the pond complex of the Faculty of Fisheries, Bangabandhu Sheikh Mujibur Rahman Agricultural University (BSMRAU), Gazipur. No extra pond management was done except cleaning aquatic weeds manually, fortnight interval liming at a rate of 1 kg dec<sup>-1</sup>, and maintaining proper water level by adding or draining.

## Design of the experiment

### *Experiment 1: Substrate addition for increasing cage surface area*

Under experiment 1, we initially tested the hypothesis that the net cage area increment using substrate can influence the growth and survival of prawn PL. The experiment consisted of three treatments, i.e. (i) no increase in surface area (control), (ii) 50% surface area increment of the net cage through the addition of substrate, and (iii) 75% surface area increment of the net cage through the addition of substrate. The treatments were assigned into a Completely Randomized Design with three replicates for each.

### *Experiment 2: Orientation of substrate addition*

We performed experiment 2 to test whether the substrate's orientation can influence the growth and survival of prawn PL. There were three treatments, i.e. (i) horizontally placed substrate,  $T_1$ ; (ii) vertically placed substrate,  $T_2$ ; and (iii) 45-degree sloping substrate,  $T_3$ . In this experiment, 75% area increment was ensured by adding a substrate for all treatment groups. The treatments were assigned into a Completely Randomized Design with three replicates for each.

### **Net cage preparation and installation**

The net cage was made of polyethylene net of 1 mm mesh size. The size of each net cage was 2.44 m × 1.83 m × 1.4 m. As substrate, 2-3 mm mesh size polyethylene net was used. In case of the 1<sup>st</sup> experiment, polyethylene nets were cut into pieces to have the desired increase in area and tied with net wall to remain the net pieces hang horizontally. Spaces were kept in between net pieces so that the applied feed

could be distributed up to the bottom. In the 2<sup>nd</sup> experiment, the net cage and the type of substrate material were similar to what we used for experiment 1. The only exception was the orientation of the substrate. Substrates were placed horizontally, vertically, and with an inclination angle of 45° according to the experimental design. Cages were installed seven days before stocking of the PL. Only bamboo poles were driven into the pond bottom and used to make the supporting frame for net cages. The upper and lower corners of each cage were fastened with bamboo poles so that the cages remained suspended about 10 cm above the pond bottom in the water column without touching the pond bottom.

### **Prawn PL stocking and feeding**

In both experiments, 900 prawn PL individuals were stocked in each cage. Prawn PLs were collected from the Swarnalata Fish Hatchery, Thrishal, Mymensingh. The PLs were transported in oxygenated plastic bags to BSMRAU campus. Upon arrival, the PLs were acclimatized allowing the bags floated in the experimental pond for about half an hour. Initial length (cm) and weight (g) of 100 randomly selected PLs were recorded. The number of PLs were counted in a jar and released into each net cage following the design. A commercial prawn feed (Quality Prawn Feed) with 40% crude protein, 18% carbohydrate, 7% fat, 2% fiber, 11% ash, 3.2% calcium, and 10% moisture level was provided to the prawn PL throughout the experimental period. Feed was supplied at the rate of 100% body weight for the first seven days, and after every seven days onward the feed percentage was reduced by 10%. We fed the prawn PL twice a day, half of the ration in the morning and the rest in the evening.

### Data collection

Prawn PLs were sampled weekly and growth parameters such as length (cm) using measuring scale and weight (g) using an electronic balance of the 15 randomly taken PLs were monitored. At the end of eight weeks days of rearing period, all prawns from each cage were counted to determine survival rate. Final length (cm) and weight (g) of 25 randomly sampled prawns from each net cage were measured using a standard measuring scale and electric balance. The SGR (specific growth rate, % bw day<sup>-1</sup>), survival rate from normalized values, and feed conversion ratio (FCR) were determined following the formula below:

- i.  $SGR (\% \text{ bw day}^{-1}) = [\ln(\text{final weight}) - \ln(\text{initial weight}) \times 100] \div \text{No. of days of the experiment}$
- ii.  $\text{Survival rate } (\%) = \text{Number of fish stocked} \div \text{Number of fish harvested} \times 100$
- iii.  $\text{Feed conversion ratio (FCR)} = \text{Feed applied (dry weight)} \div \text{Live weight gain}$

Different physico-chemical parameters of water inside the net cages were recorded weekly. Water temperatures were recorded using a portable digital Celsius thermometer. Dissolved oxygen of water was measured by a portable digital dissolved oxygen (DO) meter (Model: Lutron, PDO-519) on the spot. Measurement of dissolved oxygen was done at each net cage. pH was measured on the spot using a portable digital pH meter (Model: sensION pH3 Meter Lab PHM310). Measurement of pH was done at each cage. Periphyton growth in net cages and substrate surfaces was estimated following the method described by Azim *et al.* (2001).

### Data analysis

Two-way repeated measure test was used, where treatment account for between-subject variation, and time accounts for within-subject variation, to test if water quality parameters (temperature, DO, and pH) vary among treatments and over the experimental period. To test the effect of substrate addition and orientation in the net cage on prawn final length and weight a one-way nested ANOVA ( $\alpha=0.05$ ), using “percent of area increment” as a fixed factor (experiment 1) and “net cage” as a random factor (Krzywinski *et al.*, 2014). For the second experiment, we run the same one-way nested ANOVA using “orientation of substrate” as a fixed factor. To test the effect of substrate addition and orientation on FCR, SGR, and survival rate of prawn, we used three one-way ANOVA ( $\alpha=0.05$ ). To normalize the survival rate data, we performed an arcsin square root transformation on the percentage data. Tukey’s HSD post-hoc test was performed to determine pairwise differences. Assumptions of normality and equality of variance of the data were confirmed by visual inspection of the residuals. All analyses were conducted using IBM SPSS statistics software (Version 24).

## Results

### Experiment 1

The average mean values along with the ranges of different water quality parameters those were recorded during the experimental period are presented in Table 1. There was no significant difference ( $P > 0.05$ ) in variations of each parameter among the treatments.

There was a significant effect of area increment through the substrate on final

**Table 1. Values (Mean  $\pm$  SE) and ranges of water quality parameters in different treatments of without substrate (T<sub>1</sub>), substrate with 50% area increment (T<sub>2</sub>), and substrate with 75% area increment (T<sub>3</sub>) during the experimental period**

Water quality parameters	Treatments		
	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>
Temperature (°C)	30.60 <sup>a</sup> $\pm$ 1.27 (28.3 – 32.3)	30.64 <sup>a</sup> $\pm$ 1.25 (28.5 – 32.5)	30.69 <sup>a</sup> $\pm$ 1.29 (28.4 – 32.5)
Dissolved Oxygen (mg L <sup>-1</sup> )	5.38 <sup>a</sup> $\pm$ 0.30 (5.0 – 5.9)	5.29 <sup>a</sup> $\pm$ 0.35 (4.7 – 5.8)	5.20 <sup>a</sup> $\pm$ 0.41 (4.5 – 5.8)
pH	7.28 <sup>a</sup> $\pm$ 0.42 (6.6 – 7.9)	7.19 <sup>a</sup> $\pm$ 0.45 (6.4 – 7.8)	7.14 <sup>a</sup> $\pm$ 0.42 (6.5 – 7.8)

prawn length (P= 0.02) and weight (P= 0.04). In addition, a significant effect of net cage on final length (P = 0.03) and final weight (P = 0.01) of prawn PL was also found. Prawn PL reared in net cage with 75% area increment and 50% area increment, showed a similar final length (P=0.37, Table 2). However, treatment with 75% and 50% area increment showed significantly higher final length (P=0.00, Table 2) than the control. Treatment with 75% area increment and 50% area increment showed 12.39% and 9.61% higher final length than the control. Prawn PL showed an increase in final weight with increasing area increment (Table 2).

Prawn PL in the 75% area increment group showed 6.25% and 29.11% higher final weight than 50% area increment and the control group, respectively (P = 0.04 and P<0.001, respectively). In addition, 50% area increment group showed significantly higher final weight (21.5%) than the control group (P = 0.008). The One-way ANOVAs revealed that increase in net cage area through addition of substrate significantly affected survival rate (P = 0.003), FCR (P = 0.008), and SGR (P = 0.01). Net cage with 75% area increment showed the highest survival rate, which was

not significantly higher than the 50% area increment group (P = 0.06). Prawn PL in 75% area increment treatment showed 19.15% higher survival rate (79.43 $\pm$ 6.05%) compared to the control group (66.66 $\pm$ 6.21%, Table 2).

However, there was no significant difference between the survival rate of PL reared in the net cage with 50% area increment (75.03  $\pm$  2.45) and no area increment (P = 0.06). Net cage with 75% and 50% area increment showed significantly higher SGR compared to net cage with no area increment (75% area increment vs. no area increment: P = 0.014, 50% area increment vs. no area increment: P = 0.03, Table 2). However, there was no significant difference between net cage with 75% area increment and 50% area increment (P = 0.82). The control group showed 7.02% and 5.30% less SGR than 75% and 50% area increment group, respectively (Table 2). FCR was 35% lower in 75% area increment group compared to the control (P = 0.007, Table 2). There was no significant difference in FCR between the control and 50% area increment group (P = 0.08). No significant difference in FCR between 75% area increment group and 50% area increment group (P = 0.15) was found.



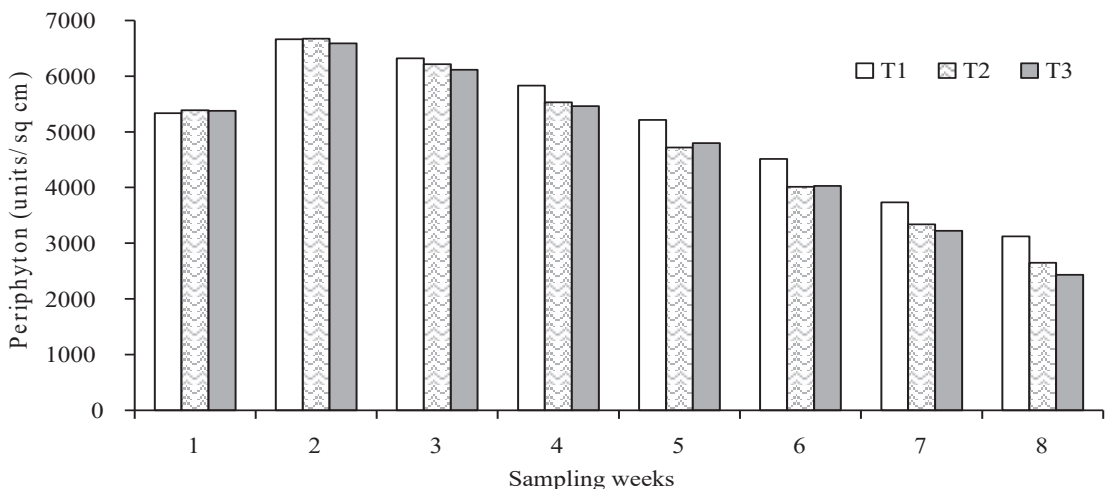
**Table 2. Growth parameters (Mean  $\pm$ SE) of *M. rosenbergii* PL in different treatments of without substrate (T<sub>1</sub>), substrate with 50% area increment (T<sub>2</sub>), and substrate with 75% area increment (T<sub>3</sub>).**

Growth parameters	Treatments		
	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>
Initial length (cm)	0.95 $\pm$ 0.16 <sup>a</sup>	0.95 $\pm$ 0.17 <sup>a</sup>	0.95 $\pm$ 0.15 <sup>a</sup>
Final length (cm)	4.68 $\pm$ 0.67 <sup>b</sup>	5.13 $\pm$ 0.64 <sup>a</sup>	5.26 $\pm$ 0.57 <sup>a</sup>
Initial weight (g)	0.02 $\pm$ 0.003 <sup>a</sup>	0.02 $\pm$ 0.003 <sup>a</sup>	0.02 $\pm$ 0.003 <sup>a</sup>
Final weight (g)	0.79 $\pm$ 0.06 <sup>c</sup>	0.96 $\pm$ 0.05 <sup>b</sup>	1.02 $\pm$ 0.09 <sup>a</sup>
SGR (% bw day <sup>-1</sup> )	7.54 $\pm$ 0.11 <sup>b</sup>	7.94 $\pm$ 0.13 <sup>a</sup>	8.07 $\pm$ 0.19 <sup>a</sup>
Survival rate (%)	66.66 $\pm$ 6.21 <sup>b</sup>	75.03 $\pm$ 2.45 <sup>a</sup>	79.43 $\pm$ 6.05 <sup>a</sup>
FCR	2.63 $\pm$ 0.38 <sup>b</sup>	1.98 $\pm$ 0.33 <sup>ab</sup>	1.69 $\pm$ 0.16 <sup>a</sup>

Mean values with different superscripts letters in the same row indicate significant difference at 5% level of significance.

About 19 genera of phyto-periphyton belonging to Bacillariophyceae (9), Chlorophyceae (3), Cyanophyceae (5) and Rhodophyceae (1), and five genera of zoo-periphyton belonging to Rotifera (4) and Crustacea (1) were identified as periphyton communities in the surface of net cages during the experiment (Fig. 1). The mean values periphyton (both phyto- and zoo-

periphyton) concentration in the treatments T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> were 5091  $\pm$  1233, 4815  $\pm$  1401 and 4751  $\pm$  1432 units cm<sup>-1</sup>, respectively. The average weekly variations in periphyton concentrations in different treatments are presented in Fig. 1, which showed a decreasing trend on periphyton concentration over the experimental period.



**Fig. 1. Growth of periphyton (unit cm<sup>-2</sup>) in three treatments of without substrate (T<sub>1</sub>), substrate with 50% area increment (T<sub>2</sub>), and substrate with 75% area increment (T<sub>3</sub>).**

## Experiment 2

The average mean values and their ranges of different water quality parameters during the experimental period are presented in Table 3. There was no significant difference ( $P > 0.05$ ) in variations of each parameter among the treatments.

There was a significant effect of substrate addition in the net cage on prawn's final length ( $P = 0.001$ ) and weight ( $P = 0.001$ ). However, no significant effect of net cages on final length ( $P = 0.91$ ) and final weight ( $P = 0.91$ ) of prawn PL was found. The final length of prawn PL in the net cage with the vertically placed substrate was significantly lower compared to the horizontally placed substrate (4.2%,  $P = 0.03$ ) and sloping substrate (7.2%,  $P < 0.001$ ). In contrast, there was no significant difference between a net cage with the horizontally placed substrate and sloping substrate ( $P = 0.14$ , Table 4). In case of the final weight, the net cage with sloping substrate showed 22.1% higher final weight ( $P < 0.001$ ) compared to the horizontal substrate group (Table 4). On the other hand, the horizontal substrate group showed 11.6% higher final weight than the vertical substrate group ( $P = 0.02$ ). However,

the horizontally placed substrate did not show any significant difference in final weight compared to both vertically placed substrate group ( $P = 0.06$ ) and sloping substrate group ( $P = 0.09$ ). There was a significant effect of orientation of substrate on FCR ( $P = 0.001$ ), SGR ( $P = 0.002$ ), and survival rate ( $P = 0.02$ ) of prawn PL.

Prawn PLs in vertically placed substrate treatment showed significantly higher FCR compared to the treatment with horizontal (11%,  $P = 0.02$ ) and sloping substrate (21.1%,  $P = 0.001$ ). In addition, horizontal substrate treatment showed significantly higher FCR than sloping substrate treatment (14.6%,  $P = 0.01$ , Table 4). Prawn PL in vertically placed substrate treatment showed significantly lower SGR than horizontally and sloping substrate treatments ( $P = 0.02$  and  $P = 0.001$ , respectively). However, there was no significant difference in SGR of prawn between horizontal and sloping substrate treatment ( $P = 0.06$ ). Sloping substrate and horizontally placed substrate showed 5% and 3% higher SGR than the vertically placed treatment, respectively (Table 4). Moreover, there was no significant difference

**Table 3. Values (Mean  $\pm$  SE) and ranges of water quality parameters in different treatments of horizontal ( $T_1$ ), vertical ( $T_2$ ) and sloping ( $T_3$ ) substrate during the experimental period**

Water quality parameters	Treatments		
	$T_1$	$T_2$	$T_3$
Temperature ( $^{\circ}\text{C}$ )	30.86 <sup>a</sup> $\pm$ 0.85 (29.4 – 32.4)	30.86 <sup>a</sup> $\pm$ 0.87 (29.5 – 32.4)	30.87 <sup>a</sup> $\pm$ 0.85 (29.4 – 32.3)
Dissolved Oxygen ( $\text{mg L}^{-1}$ )	5.29 <sup>a</sup> $\pm$ 0.22 (5.0 – 5.7)	5.27 <sup>a</sup> $\pm$ 0.25 (4.8 – 5.6)	5.27 <sup>a</sup> $\pm$ 0.21 (5.0 – 5.7)
pH	7.23 <sup>a</sup> $\pm$ 0.39 (6.4 – 7.8)	7.22 <sup>a</sup> $\pm$ 0.38 (6.5 – 7.7)	7.24 <sup>a</sup> $\pm$ 0.40 (6.4 – 7.7)

**Table 4. Growth parameters (Mean  $\pm$ SE) of *M. rosenbergii* PL in different treatments of horizontal ( $T_1$ ), vertical ( $T_2$ ) and sloping ( $T_3$ ) substrate**

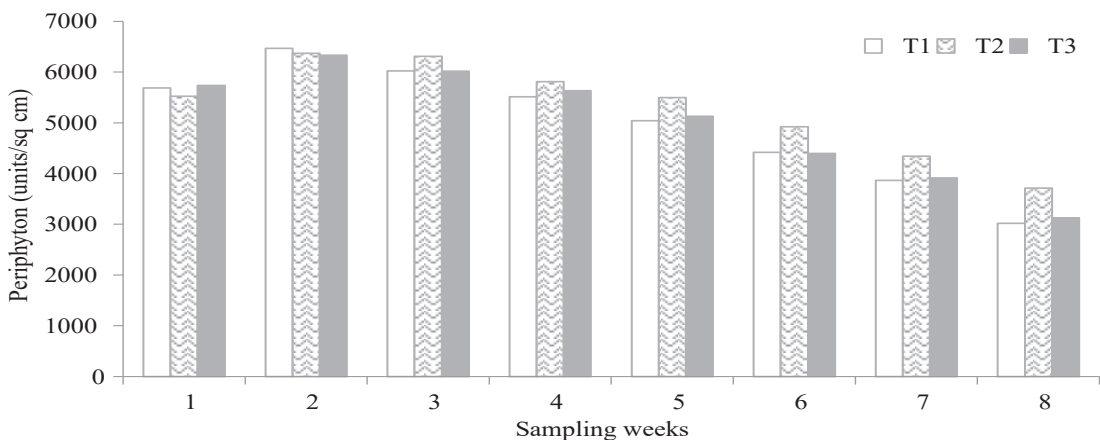
Growth parameters	Treatments		
	$T_1$	$T_2$	$T_3$
Initial length (cm)	$0.88 \pm 0.12^a$	$0.88 \pm 0.12^a$	$0.88 \pm 0.12^a$
Final length (cm)	$5.21 \pm 0.11^a$	$5.00 \pm 0.09^b$	$5.36 \pm 0.13^a$
Initial weight (g)	$0.019 \pm 0.01^a$	$0.019 \pm 0.01^a$	$0.019 \pm 0.01^a$
Final weight (g)	$1.06 \pm 0.05^a$	$0.95 \pm 0.04^b$	$1.16 \pm 0.10^a$
SGR (% bw day <sup>-1</sup> )	$8.38 \pm 0.07^a$	$8.15 \pm 0.11^b$	$8.58 \pm 0.13^a$
Survival rate (%)	$77.12 \pm 1.20^a$	$76.33 \pm 1.15^b$	$79.33 \pm 0.57^a$
FCR	$1.65 \pm 0.06^b$	$1.77 \pm 0.06^a$	$1.53 \pm 0.02^c$

Mean values with different superscripts letters in the same row indicate significant at 5% level of significance.

in the survival rate of prawn PL between horizontal and sloping substrate treatment ( $P = 0.9$ ). However, both horizontal and sloping substrate treatment showed a significantly higher survival rate of prawn PL than vertical substrate treatment ( $P = 0.03$  and  $P = 0.02$ ). Sloping substrate treatment showed 4%, and horizontal substrate treatment showed 3% higher survival rate than the vertical substrate treatment (Table 4).

About 22 genera of phyto-periphyton belonging to Bacillariophyceae (10), Chlorophyceae (9),

Cyanophyceae (4) and Rhodophyceae (1), and five genera of zoo-periphyton belonging to Rotifera (4) and Crustacea (1) were identified as periphyton communities in the surface of net cages during the experiment. The mean values periphyton concentration in the treatments  $T_1$ ,  $T_2$  and  $T_3$  were  $5016 \pm 1171$ ,  $5311 \pm 933$  and  $5034 \pm 1121$  units cm<sup>-1</sup>, respectively. The average weekly variations in periphyton concentrations in different treatments are presented in Fig. 2, which showed a decreasing trend on periphyton concentration over the experimental period.



**Fig. 2. Growth of periphyton (unit cm<sup>-2</sup>) in three treatments of substrate with horizontal ( $T_1$ ), vertical ( $T_2$ ) and 45° sloping ( $T_3$ ) orientation.**



## Discussion

We found that prawn PL showed overall better growth and survival rate with net substrate in the net cage nursery unit. In addition, the substrate placed at a 45° of sloping orientation showed better FCR.

Water quality has an immense effect on any aquatic organism. Fish and prawn have more or less the same temperature tolerances, and a temperature range between 26°C and 32°C is considered suitable for the metabolic activity of *M. rosenbergii* (Boyd and Zimmermann, 2010). Temperature ranges found in both experiments were within the acceptable range. pH is an important factor that affects survival, growth, moulting, and feeding activities of prawn. pH levels ranging from 6.2 to 7.4 has been reported suitable for prawn (Chen and Chen, 2003) 48-, 72- and 96-h LC50s (median lethal concentrations, which indicates that the pH found in the present experiments were within the acceptable limit. Although *M. rosenbergii* can survive the DO level below 2 mgL<sup>-1</sup>, a minimum of 3 mgL<sup>-1</sup> is needed to be maintained to avoid stress and optimal growth rate (Abramo, *et al.*, 2006). In our both experiments, DO concentrations were within the suitable range.

An increase in the net cage area through the addition of substrate showed a significant increase in the final length and the weight of prawn PL compared to the control group (without substrate). Different studies have also reported that the addition of substrate improved growth, survival, FCR of *M. rosenbergii* compared to those reared without substrate (Cohen and Ra'anani, 1983; Marlowe, 2006; Simpson *et al.*, 2016; Tidwell *et al.*, 1998; Tuly *et al.*, 2014). For example:

Mamun *et al.* (2010) reported that the addition of high-density polyethylene net substrate in *M. rosenbergii* nursing aquaria increased the final weight up to 32.70% compared to no substrate added aquaria. Also, an increase in growth, SGR, FCR, and survival might be attributable to less aggressive interactions and stress (Karplus *et al.*, 1992).

In the present study, substrate placement created different layers inside a net cage based on the area increment (3 layers for 75% area increment, and two layers for 50% area increment). Hence, it might have created multiple layers of feeding spots with less conspecific interactions. Nevertheless, these substrates help them seek shelter on different net cage layers, providing them with a three-dimensional area to utilize. That might be why 75% area increment group showed better growth and survival rate than any other treatments.

Periphyton that grew on the net cage's surface and the substrates might have played a vital role in the present study. Tuly *et al.* (2014) suggested that addition of substrate might help improving periphyton and natural food production for *M. rosenbergii*. In our experiments, periphyton biomass showed a decreasing trend throughout the experimental period. However, there was no significant difference ( $p > 0.05$ ) among the treatments (Fig. 1 and Fig. 2). The decreasing concentration of periphyton might due to an increased and efficient grazing on periphyton by the prawn PL. Prawn PL has an extra advantage over adult prawns, which is its smaller chelipeds. These smaller chelipeds might have helped them selectively graze on periphytic biomass. Tidwell *et al.* (1997) reported that small prawn

utilizes natural foods more efficiently than prawn of large average size. The overall FCR was better in experiment 2, and a significantly lower FCR was found in the treatment with sloping substrate (Table 4). Improvements in FCR due to substrate addition have been reported elsewhere (Tidwell *et al.*, 2000; Tuly *et al.*, 2014). It indicates that the addition of substrate in the prawn nursery unit could be beneficial. However, the proportionate increase in surface area and orientation might significantly affect improved growth, FCR and prawn PL survival rate.

The growth, FCR, SGR, and survival rate in the horizontally placed substrate group was consistent with 75% area increment treatment (Table 2). Vertically placed substrate group showed overall lower growth, SGR, and survival rate than the horizontal and sloping substrate group (Table 4). It might be because the prawn PL could not utilize vertically placed substrate efficiently for shelter or physical isolation from other individuals resulting in frequent agonistic interactions and stress. Hence, prawn PL might have shown lower growth as well as the survival rate of prawn PL. Secondly, the feed might have sunk to the bottom of the net directly, leading to increased feed wastage in vertically placed substrate treatment. Soft-shell prawns might not be efficient to utilize vertically installed substrate and are incredibly vulnerable to cannibalism and predation (Peebles, 1978; Smith and Sandifer, 1975).

However, no significant difference in growth, FCR, and survival rate of the prawn was found between horizontally or vertically placed substrate by Tidwell *et al.* (2002). In the present study, the significant differences in growth

and FCR in the sloping substrate prawn group might be that the sloping substrates facilitated prawns to feed and graze efficiently compared to horizontal and vertical oriented substrates.

### Conclusion

The results of experiment 1 showed that 75% area increment improved the growth, FCR, and survival of prawn PL. Keeping the area increment at 75%, the sloping substrate group showed similar growth performance compared with horizontal substrate group and higher FCR compared to both horizontally and vertically placed substrate. Therefore, 75% area increment with the sloping substrate may be recommended for prawn PL nursing in cages. Further studies for evaluating the appropriate stocking density of prawn PL to get maximum growth, FCR and survival rate may be carried out.

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