# Growth performance of black tiger shrimp (*Penaeus monodon*) in substrate based zerowater exchange system

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To evaluate the effect of periphyton based system, an on-station grow-out trial of black tiger shrimp, *Penaeus monodon* was carried out for 130 days using with (Ts) and without (Tc) submerged substrates. *P. monodon* was stocked at 8 nos. m<sup>-2</sup>, and bamboo substrates  $(1.8 \times 0.06 \text{ m})$  were fixed in treatment ponds @ 2000 numbers ha<sup>-1</sup> for development of periphyton. Provision of substrate increased the transparency (p<0.01) and reduced the turbidity (p<0.05) in treatment ponds compared to control. Similarly, comparatively lower level of total ammonia –N and nitrate–N were recorded in substrate based ponds compared to control. Periphyton biomass in terms of dry matter and ash free dry matter over substrates were  $5.9\pm0.7$  and  $3.0\pm0.4 \text{ mg cm}^{-2}$  respectively. Chlorophyll a level in water column was  $15.22\pm6.97$  and  $19.06\pm8.68 \mu g L^{-1}$  in Ts and Tc ponds respectively. At the end of the grow out trial, a higher average production of  $1640 \text{ kg ha}^{-1}$  with an average body weight (ABW)  $25.85\pm2.62 \text{ g}$  was obtained in Ts compared with control, Tc (1390 kg ha<sup>-1</sup> production and  $22.00 \pm 2.83 \text{ g}$  ABW). About, 29% improvement in feed conversion ratio ( $1.15\pm0.42$ ) noticed in substrate based ponds compared to control ponds ( $1.48\pm0.02$ ). Regression trend for length–weight analysis indicated that growth exhibited an isometric pattern with better Fulton condition factor (0.83) in tiger shrimp reared in substrate based system than in control ponds (0.82). Better growth performance of tiger shrimp in substrate based system indicates utilisation of available natural food and natural productivity as periphyton by cultured shrimps.

[Keywords: Periphyton, Substrate, Penaeus monodon, Length -weight relationship]

## Introduction

Penaeid shrimps are one of the highly soughtafter seafood commodity and cultured shrimp contribute major share of global shrimp production. The shrimp industry expanded globally due to of rising seafood demand and increased economic return. However, the expanding shrimp culture is presently facing challenges like increasing price of commercial feed, disease outbreaks and environmental degradation<sup>2</sup>. Hence, it is imperative to explore sustainable culture methods like low input high return system, which enhance the shrimp production and increase the income of smallscale farmers.

Periphyton based culture systems are one of the low input low cost systems. Periphyton is a complex mixture of autotrophic and heterotrophic microorganisms such as phytoplankton, zooplankton and heterotrophic microbes<sup>3</sup>. Submerged substrates provide sites for the development of periphyton. Substrate based aquaculture is widely practiced in finfish and prawn culture mainly in carps <sup>4, 5</sup>, tilapia <sup>6</sup>, giant freshwater prawn<sup>7</sup> and pearlspot<sup>8</sup> to enhance aquaculture productivity. Microalgae being an important dietary food source in initial stages of penaeid shrimp<sup>9</sup>, periphyton based culture provide a great scope in shrimp culture. Provision of substrate in shrimp culture systems enhances the growth of pink shrimp, *Farfantepenaeus paulensis* <sup>10</sup>, brown tiger shrimp, *Penaeus esculentus* <sup>11</sup>, black tiger shrimp, *Penaeus monodon* <sup>12, 13</sup> and western white leg shrimp, *Litopenaeus vannamei* <sup>14</sup>. Periphyton serves as quality natural food and

improves water quality through nitrification process <sup>15, 16</sup>.

Culture condition plays an important role in growth, length-weight relationships and condition factor of commercially important penaeid shrimps <sup>17</sup>. Length-weight relationships and condition factor indicate growth, rate of feeding, fatness, gonadal maturity, and general well-being of cultured shrimps<sup>18</sup> and fishes <sup>19</sup>. It varies based on culture condition, food availability, stress (overcrowding and disease), reproductive cycle etc  $^{20}$ . For example, P. monodon shows better condition factor in culture environment rich in natural productivity <sup>17</sup>. Even though, suitability of substrates in penaeid shrimp culture is documented <sup>12</sup>, <sup>14</sup>, there is a dearth of information with regard to growth performance and length-weight relationship of P. monodon in substrate based low density zerowater exchange culture system. In this context, this paper aims to compare growth, water quality parameters, periphyton biomass and length weight relationship of P. monodon cultured in zerowater exchange brackish water ponds with and without bamboo substrate.

## **Materials and Methods**

The experiment was carried out for a period of 130 days during July to November 2010 in the brackishwater farm of Kakdwip Research Centre of Central Institute of Brackishwater Aquaculture (CIBA), Kakdwip (21° 51' N and 88° 11' E), South 24 Parganas, West Bengal, India. Four earthen ponds (0.18-0.37 ha) were selected for grow out culture. Before start of the experiment, all ponds were allowed to dry until cracks developed and top soil was removed. Ponds were filled with filtered brackishwater from a nearby creek of the Muriganga river to a depth of 150 cm and kept for 5-6 days. Bleaching powder (CaOCl<sub>3</sub>) was applied (a) 600 kg ha<sup>-1</sup> to reduce risk of disease outbreak from pathogenic bacteria, virus and unwanted seed of other organisms. After 10-12 days, agricultural lime (CaCO<sub>3</sub>) was applied to all ponds at the rate of 100-200 kg ha<sup>-1</sup> based on the pond pH. Ponds were fertilized with semi decomposed cattle manure, urea and triple super phosphate (TSP) at a dose of 3000, 100 and 100 kg ha<sup>-1</sup> respectively. Bamboo poles  $(1.8 \times 0.06)$  m) were installed at a rate of 2000 number ha<sup>-1</sup> in treatment ponds. The ponds were left for 15 days to allow periphyton development over bamboo substrate. Hatchery produced black tiger shrimp, *P. monodon* (PL 20) was stocked at a density of 8 nos m<sup>-2</sup> in ponds with bamboo substrate ( $T_s$ ) and without substrate ( $T_c$ ). Crop was carried out during monsoon season as the zero water exchange system practiced relies mainly on rain to compensate evaporation and seepage loss.

Formulated shrimp feed with 38 % protein content was used during grow out period (Table 1). During the first month of the culture [0-30 days of culture (DOC)], blind feeding (1 kg feed per 100,000 postlarvae) was adopted, which is modified as per the earlier report<sup>21</sup>. Subsequently, feed quantity was adjusted based on shrimp body weights calculated from weekly sampling and assumed survival percentage from cast net sampling and check tray observation. Feed was given at 5-2% of the body weight of the shrimps from second month to final month of culture. Daily ration was distributed in two to four times per day, 40% in the morning (06:00 & 11:00 h) and 60% in the evening (18:00 & 22:00 h). The feeding regime followed was based on regular monitoring of the check tray. Yeast based probiotic preparations (fermented solution of 2 kg yeast, 30 kg molasses and 60 kg rice flour ha<sup>-1</sup>) were applied at fortnightly intervals as nutrient supplement to improve natural productivity and overall pond environment.

Water samples were collected at fortnight intervals between 09:00 and 10:00 h and analyzed immediately after return to the laboratory. Physical parameters like salinity, temperature and pH were measured using an Atago hand refractometer (Atago, Japan), thermometer and pH meter (model 10E; Deluxe) respectively. Water quality parameters like dissolved oxygen, alkalinity, total ammonia-N (TAN), nitrite-N (NO<sub>2</sub>-N), nitrate-N (NO<sub>3</sub>-N), phosphate-P (PO<sub>4</sub>-P), gross primary productivity, net primary productivity and chlorophyll a content in the water column were determined according to standard procedure<sup>22</sup>, <sup>23</sup>. For periphyton dry mater (DM) and ash free dry matter (AFDM) estimation, samples were collected from  $2 \times 2$  cm<sup>2</sup> area of bamboo substrate. Samples were dried at 105°C for dry matter estimation and ashed at 600°C for 6 h in a muffle furnace for ash free dry matter (AFDM) estimation<sup>22</sup>.

Table 1. Proximate composition of the feed used in *P. monodon* shrimp culture

Percentage (% as feed basis)
38.35
7.75
3.36
14.25
27.80
85.75
8.49

<sup>a</sup>Organic matter = 100-Ash %<sup>b</sup>Nitrogen free extract = 100-(Crude protein % + Lipid % +Crude fiber % + Ash %+Moisture

For length-weight relationship and condition factor analysis, tiger shrimps were weekly sampled, and weighed using a digital electronic balance having 0.01 g precision, and total length (TL) from rostral tip to the tip of telson was measured with precision of 0.01 cm. After 130 days of culture, shrimps were harvested and growth performances were evaluated in terms of final average body weight (g), daily growth rate (g), feed conversion ratio (FCR), total productivity (kg ha<sup>-1</sup>) and survival rate (%).

#### Statistical analysis

The data were analyzed by statistical package SPSS version 17.0 (SPSS Inc., Chicago, IL, USA). Growth performance and water quality parameters between the treatments were analyzed using Student's t-test. Significance level was determined at 95% probability level. The length–weight parameters were subjected to regression analysis, using the power function  $W=aL^b$ , where W = the dependent variable, L = the independent variable, a = the condition factor, and b = the weight increase <sup>24</sup>. The degree of association between weight and length variable was calculated by the coefficient of

determination ( $\mathbb{R}^2$ )<sup>25</sup>. Fulton's equation was used to determine sex specific condition factor following the equation a=W/L<sup>3</sup> × 100, where W= weight in g and L=length in cm<sup>26</sup>.

### **Results and Discussion**

Water quality parameters during the experimental period are presented in Table 2. The recorded water quality parameters were within the acceptable ranges for brackishwater shrimp culture<sup>27</sup>.Comparatively lower level of total ammonia nitrogen (TAN), NO<sub>3</sub>-N, NO<sub>2</sub>-N and PO<sub>4</sub>-P were observed in substrate based system while no significant difference was noticed among the experimental ponds. This may be attributed to the fact that low stocking density in the present field trial might have kept the water nutrient parameters within the favorable limit in both the treatments. Provision of substrate increased the transparency (p < 0.01)and reduced the turbidity (p<0.05) in treatment ponds compared to control. This is similar to our earlier findings where a lower turbidity level was noticed in substrate based system as substrate reduces turbidity by trapping column<sup>13</sup>. suspended particles in water Chlorophyll a content in the water column is presented in Fig 1. Average chlorophyll a levels water column were 15.22±6.97 and in 19.06 $\pm$ 8.68 µg L<sup>-1</sup> in substrate and control ponds respectively. Level of chlorophyll a in control ponds showed fluctuation throughout the culture period, and coincided with algal bloom and algal crashes. Similarly, comparatively lower gross primary productivity, 235 mg C m<sup>-3</sup> h<sup>-1</sup> was recorded in substrate based system compared to control ponds (258 mg C m<sup>-3</sup> h<sup>-1</sup>). The present findings are in line with the earlier reports where lower level of primary productivity, chlorophyll a and phytoplankton level in the water column were reported in substrate based culture ponds<sup>4</sup>,

Water quality parameters	T <sub>S</sub> (n= 11)	T <sub>C</sub> (n=11)
Temperature	32.57±2.26 (29–35.5)	32.59±2.29 (29–36)
pH-Morning	8.03±0.23 (7.81-8.87)	8.09±0.23 (7.75−8.5)
pH-Evening	$8.32 \pm 0.34$ (7.98–9.14)	8.37-0.28 (8-8.81)
Salinity(ppt)	13.20±3.29(9.1–18.1)	12.60±3.83 (6.8–18.1)
Alkalinity (mg/l)	135.8±19.26(108–72)	131±15.99 (104–164)
Transparency (cm)	$44.90 \pm 13.52(32 - 80)^{a}$	36.5±6.29 (25−50) <sup>b</sup>
Turbidity (ppm)	$30.5\pm9.78~(10-43)^{a}$	40.95±6.49 (32−53) <sup>b</sup>
DO (ppm)	5.93±0.33 (5.2–6.6)	5.78±0.42 (4.8–6.4)
GPP (mgC/m <sup>3</sup> /hr)	238.01±94.17 (117–487)	257.25±95.58 (140.61–485)
NPP (mgC/m <sup>3</sup> /hr)	137.68±42.56 (87.5–225)	155.04±57.06 (52.2–300)
TAN (µg/l)	109.85±24.97 (57.11–145)	117.79±36.31(69.29-157.8)
NO <sub>2</sub> -N (μg/l)	26.09±11.55 (3.35-40.4)	29.2191±19.62 (3.21–76)
NO <sub>3</sub> -N (μg/l)	$108.94 \pm 40.60$ (43.5–153.02)	111.43±49.02 (43.13–187)
$PO_{4}-P(\mu g/l)$	$50.71\pm22.94$ (10.82–84.98)	53.06±24.42 (17.32–91)

Table.2. Water quality parameters (Mean value  $\pm$  Standard Deviation) in of *P. monodon* shrimp ponds in substrate (T<sub>s</sub>) and without substrate (T<sub>c</sub>)

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Fig .1. Chlorophyll a concentration ( $\mu$ g/Liter) of pond water in control (T<sub>C</sub>) and substrate based shrimp (T<sub>S</sub>) ponds. Values are expressed as mean value ± Standard Deviation; n, number of sampling=11



Fig. 3. Average body weight of black tiger shrimp, *P.monodn* during the grow out period in control ( $T_C$ ) and substrate based shrimp ( $T_S$ ) ponds. Values are expressed as mean value  $\pm$  Standard Deviation); n,number of sampling=11



Fig .2. Periphyton biomass  $(mg/cm^2)$  developed in submerged bamboo substrate over time period. Values are expressed as mean value  $\pm$  Standard Deviation; n, numberofsampling=10

Name of species	Sample size (n)	Weight (g)	Length (cm)	a	b	$R^2$	K	
<i>P. monodon</i> male (T <sub>s</sub> )	612	3.10 - 35.50	6.80-16.70	0.0089	2.96	0.98	0.83±0.017	
<i>P.monodon</i> female $(T_s)$	588	2.80-41.10	6.70-17.40	0.0088	2.97	0.96	0.84±.022	
<i>P.monodon</i> male (Tc)	595	3.0-30.50	6.80-15.80	0.0111	2.86	0.97	$0.82 \pm 0.02$	
<i>P.monodon</i> female (T <sub>c</sub> )	574	3.40-33.00	6.90-15.90	0.0105	2.89	0.98	$0.82{\pm}\ 0.023$	

Table 3. Length weight relationship and condition factor of *P.monodon* cultured in substrate (T<sub>S</sub>) and control(Tc) based system

a, intercept; b, coefficient of regression or slope; R<sup>2</sup>, coefficient of determination, K=Fulton condition factor

According to their studies, it is assumed that competition between periphytic algae and phytoplankton in water column for light and nutrients, shading effects of periphyton substrates or movement of algae from planktonic state to the periphytic state can control the algal growth in water column. Theses suggest the fact that periphyton based system have a stabilizing influence on the phytoplankton production, and control the occurrence of algal bloom or crash by shifting less stable phytoplankton to more stable periphyton community.

Periphyton biomass in terms of dry matter (DM) and ash free dry matter (AFDM) per unit surface area were  $5.9\pm0.7$  and  $3\pm0.4$ mg/cm<sup>2</sup> respectively. Periphyton biomass found to be decreased over the time compared to initial biomass indicates constant grazing of periphyton by the shrimps (Fig 2). Periphytic algae need to be grazed constantly and kept at low biomass to maintain their high productivity<sup>6</sup>. Steadily decline in periphyton biomass during the initial period compared to later culture period may be attributed to the more preference of periphyton by the juvenile shrimp than adult *P.monodon*. Growth performance of P. monodon during the growoutcultrure period is given in Fig 3. At the end of the feeding trial, P. monodon attained higher average body weight (ABW), 25.85  $\pm 2.62$  g with a total production of 1640 $\pm 36.7$  kg ha<sup>-1</sup> in the treatment ponds compared to 22.00  $\pm 2.83$  g ABW and 1390  $\pm 2.8$  kg ha<sup>-1</sup> total production in control ponds. Similarly, about, 22.4% improvement in feed conversion ratio (FCR) was noticed in substrate based ponds  $(1.15\pm0.42)$  compared with control ponds

 $(1.48\pm0.02)$ . This matches with the earlier reports where considerable production gain and better FCR was noticed in substrate based culture of L. vannamei<sup>14</sup> and F. paulensis<sup>10</sup>. Similarly, higher shrimp production and lower FCR was observed when P. monodon post larvae cultured in artificial substrate based systems<sup>28</sup>. It has been documented that availability of particulate organic matter like zooplankton, microalgae, bacteria and meiofauna in submerged substrate account for the shrimp growth in substrate based culture system<sup>29</sup>. Moreover, periphyton developed over submerged substrates provides an excellent grazing space for shrimp. It further confirm that shrimp being bottom dweller, grows better by grazing on periphyton than filtering suspended algae from the water column<sup>30</sup>. Presence of substrate increases the available surface area and reduces the negative effects of overcrowding which contribute to better performance of intensive culture of L. vannamei<sup>14</sup>. However, in the present study no difference in survival was noticed among the treatment. This might be due to low stocking density used in the present system that might have alleviated overcrowding problems unlike other intensive systems.

Length- weight relationship in cultured organisms indicates well-being of the organism  $^{31}$ . In regression analysis, the change of b, noted as isometric, b=3; positively allometric, b>3; and negatively allometric, b<3 indicates the rate of weight gain relative to length, and varies among different growth stages of the same species<sup>32,33</sup>. Regression trend indicated variation in the growth pattern for individual sexes (Table

3). The slope or regression coefficient b ranged from 2.86 to 2.97. Though the regression coefficient 'b' value is < 3, it is close to the isometric pattern i.e. for any unit increase in weight there was a relatively proportionate increase in length. In general, penaeid shrimp growth follows a sigmoidal pattern<sup>34</sup>. Linear function satisfactorily describes growth of *F. merguiensis*<sup>35</sup>, *P. monodon*<sup>36</sup> and *F. indicus*<sup>37</sup>. Regression coefficient (b=2.97) noticed in shrimps reared in substrate based ponds are very near to the isometric growth condition in substrate based ponds compared with control ponds (b=2.87).

This matches with the earlier report where P. monodon cultured in semi intensive ponds with good natural productivity and better ecological factors reported a higher regression coefficient, i. e 3.031 and 3.223<sup>17</sup>. The condition factor is often associated with fitness or wellbeing of an organism. Mean Fulton's condition factor, K, in treatment pond (T<sub>s</sub>) is  $0.84\pm.02$  to 0.83±0.02 in female and male while in control ponds, it was  $0.82\pm$  to 0.02 in both female and male shrimps. The co-efficient of correlation (r) for the length-weight relationship was estimated above 96%, indicate a high degree of correlation between these two parameters. Comparatively higher b value and condition factor observed for female than male in the present study agree with the earlier reports of sex-based size dimorphism in penaeids with larger sizes and faster growth rates in females compared to males for F. merguiensis  $^{35}$ , F.

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*indicus* <sup>37</sup>, *P. monodon* <sup>36</sup> and *L. vannamei* <sup>26</sup>. Better Fulton condition factor observed in substrate based treatments reflects better feed utilization, less competition for feed and more suitable culture environment in substrate based system compared to control.

## Conclusion

From the above result, it can be concluded that *P. monodon* reared in substrate based systems had higher biomass and lower FCR compared to shrimps grown in substrate free ponds. Periphytic community developed over the submerged substrate served as quality natural food for shrimps. Regression coefficient closer to isometric value and better Fulton condition factor observed in substrate based treatments reflects better feed utilization and suitable culture environment. Amount of utilization of periphyton by P. monodon at higher stocking density and utalization of periphyton at different life stages of black tiger shrimp is a subject of further research.

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