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Growth, survival and length weight relationship of *Fenneropenaeus merguiensis* at two different stocking densities in low saline zero water exchange brackishwater ponds

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An onstation trial of banana shrimp, *Fenneropenaeus merguiensis* was conducted for 130 days in zero water exchange brackishwater ponds at two stocking densities, 10 (T_{10}) and 20 (T_{20}) nos. m^{-2}. Water parameters like total ammonia nitrogen (TAN), NO_2-N and NO_3-N did not differ significantly ($p>0.05$) among treatments while T_{20} had significantly higher ($P<0.01$) turbidity, PO_4-P and significantly lower ($P<0.01$) dissolved oxygen compared with T_{10}. At the end of grow out period, banana shrimp reached a final average body weight of 14.1±0.84 g in T_{10} and 11.0±0.14 g in T_{20} with 50.5 % higher ($p<0.05$) average productivity in T_{20} (990±7.07 kg/ha) compared with T_{10} (658±65.05 kg/ha). Regression trend for length weight analysis indicated that growth exhibited a positive allometric pattern and female showed better condition factor than male in both the treatments. Similar Fulton condition factors observed in both the treatments reflects better feed utilization and suitable culture environment even at higher stocking density. Present findings elucidate the potentials of high density culture of banana shrimp in coastal districts in India.

[Key words: Brackishwater pond, Condition factor, *Fenneropenaeus merguiensis*, Length weight relationship, Salinity, Stocking density]

Introduction

During the last two decades, white spot and other viral diseases have spread worldwide and caused large-scale mortalities and severe damage to shrimp culture industry which leads to massive economic losses [1]. In India, *Penaeus monodon* is the major cultivable shrimp species and is currently plagued with the continuous outbreaks of white spot syndrome virus since 1994[2]. Recently, intensive culture of western white leg shrimp, *Litopenaeus vannamei* started in India which is worldwide facing the disease outbreak such as taura syndrome virus, yellow head virus etc. [3 & 4]. This emphasizes the need for diversification of shrimp culture and standardization of culture practices for other suitable candidate penaeids.

Banana shrimp, *Fenneropenaeus merguiensis* ranks third among the farmed shrimp species and emerging as a good candidate species for commercial brackishwater shrimp culture due to available hatchery technology [5]. The species has traditionally been cultured in many countries of Asia-Pacific region especially in Indonesia, Australia and more than 70% of the farmed *F. merguiensis* production comes from these countries[6]. In India, it is a native species of coastal waters of West (Gujarat, Maharashtra) [7] and East coast (Orissa) [8] regions.

Stocking density plays an important role in growth and survival of cultured species. In general, optimum stocking density depends upon the type of cultured species, harvesting size of shrimp, and the number of crop per annum with the best economic return for the farmers [9]. There are various published reports about the optimal stocking density for penaeid shrimp, *F. indicus* [10], *P. monodon* [11] and *L. vannamei* [12 & 13]. However, there is a dearth of information with regards to

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grow out culture of *F.merguiensis* at different stocking densities.

Salinity is one of the most important abiotic factor affects the growth and survival of cultured banana shrimp [14]. Optimum salinity range of 15-30 ppt is found to be ideal for grow out culture of banana shrimp [15]. Growth performance of commercially important penaeid shrimps are also related with their length–weight relationship and condition factor as these varies according to culture conditions[16]. Till date no information is available with regard to length–weight relationship of *F. merguiensis* cultured at different stocking densities in low saline grow out ponds. In this context, this paper aim to compare growth performance, water quality parameters, length–weight relationship and condition factor of *F. merguiensis* cultured at different stocking densities in low saline zero water exchange brackishwater ponds.

Material and Methods

The experiment was carried out for 130 days during June to October 2010 in the brackishwater ponds at Kakdwip Research Centre, Central Institute of Brackishwater Aquaculture (CIBA), Kakdwip (21° 51′N and 88° 11′ E), South 24 Parganas, West Bengal, India. Four earthen ponds (0.1–0.17 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 brackishwater from a nearby creek of the Muriganga river to a depth of 150 cm and kept for 5-6 days. Bleaching powder ($CaOCl_3$) was applied at the rate of 600 kg ha $^{-1}$ to reduce risk of disease outbreak from pathogenic bacteria, virus and unwanted seed of other organisms. After two week, lime ($CaCO_3$) was applied to all ponds at 100-200 kg ha^{-1} based on the pond pH. Ponds were fertilized with semi decomposed cattle manure, urea and triple super phosphate (TSP) at a dose of 1500, 100 and 100 kg ha^{-1} respectively and left for 15 days to allow plankton development. Hatchery reared *F. merguiensis* (CIBA, Chennai) was stocked in duplicate ponds at the rate of 10 nos. m^{-2} (T_{10}) and 20 nos. m^{-2} (T_{20}). Crop was carried out during monsoon season as zero water exchange system relies mainly on rain to compensate evaporation and seepage loss. Uniform aeration at the rate of 1.8 Kg oxygen per KW $hour^{-1}$ was provided for 6-8 h in all the ponds using 1 HP paddlewheel aerator.

Commercially formulated shrimp feed (Bismi feed Ltd, India) with 38 % crude protein was used during the culture (Table 1). During the first month of culture [0–30 days of culture (DOC)], blind feeding was adopted in both treatments with 2 kg feed per 100,000 post larvae (PL) [17.] Subsequently, feed quantity was adjusted according to shrimp body weight based on 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 shrimp from second month to harvest. The daily feed 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). Check tray observations were monitored to keep strict feeding regime. Yeast based probiotic preparations were periodically applied as nutrient supplement, and for overall pond environment improvement. For this purpose, yeast (2 kg), molasses (30 kg) and paddy flour (60 kg) were soaked in water for 48 hours and applied at fortnightly intervals.

Water samples were collected between 09:00 and 10:00 h at fortnight intervals 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, chemical oxygen demand (COD), biochemical oxygen demand (BOD), total ammonia-N (TAN), nitrite-N (NO_2-N), nitrate–N (NO_3-N), phosphate-P (PO_4-P), primary productivity and chlorophyll a were determined according to the standard procedures [18 & 19].

Total heterotrophic bacterial load and *Vibrio* load of water was determined at monthly interval during the culture period. Water samples were collected between 07:00 and 08:00 h from 5-10 cm depth at different sites of the pond in the sterile poly-propylene bottle and pooled together.

Sample was homogenised (12000 rpm for 30 seconds) and 0.1 ml of appropriate dilutions was plated on tryptone soya agar for heterotrophic total bacterial count and on thiosulfate citrate bile salt sucrose agar (TCBS) for *Vibrio*. Plates were incubated at room temperature for 48 h and colony in the range of 30 to 300 were counted and expressed as bacterial colony forming unit (cfu).

Table 1. Proximate composition of the feed (Bismi Feed Ltd) used in banana shrimp culture

Composition	Percentage (% Dry matter)
[a]Crude protein	38
Crude fat	6
Crude fiber	3
Total ash	18
[b]Nitrogen free extract	24
Organic matter	82
Moisture	11

[a]Organic matter = 100-Ash %.

[b]Nitrogen free extract = 100- (Crude protein % + crude fat % +Crude fiber % + Ash %+Moisture)

After 130 days of culture, shrimps were harvested and final average body weight (ABW), daily growth rate (DGR), feed conversion ratio (FCR), survival, total production (kg ha^{-1}) were calculated as follows:

DGR = Average final body weight gain /days of experiment

FCR = Dry weight of feed / live weight gain

Survival = (Total number of animal survived /Total number of animal stocked) ×100

For length- weight relationship and condition factor analysis, banana shrimp were weekly sampled, and weighed using a digital electronic balance having 0.01 g precision, and total length from rostral tip to the tip of telson (TL) was measured using a ruler with precision of 0.01 cm.

Statistical analysis

The data were analyzed by statistical package SPSS version 17.0 (SPSS Inc., Chicago, IL, USA). All the data were checked for normality before analysis. Average growth rate and water quality parameters among the treatments were analysed using independent student's t-test. Probability level at α =0.05 and 0.01 was considered significant. Length- weight relationship between male and female banana shrimp were calculated using curvilinear regression methods. Length and 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 [20]. Degree of association between weight and length variable was calculated by the determination coefficient (R^2) [21]. Fulton's equation was used to determine sex specific condition factor following the equation $a=W/L^3 \times 100$, where W= weight in g and L=length in cm [22].

Results

Water quality parameters like pH, salinity, alkalinity, nitrite-N, nitrate-N, total ammonia-N (TAN) and primary productivity did not show significant variation ($P > 0.05$) among the treatment (Table 2). Temperature ranged from 29 to 35°C and salinity during the culture period reduced from 21 to 8.3 ppt (Fig.1). Group T_{20} had significantly lower ($P<0.01$) dissolved oxygen, significantly higher ($P<0.01$) turbidity and PO_4-P compared with T_{10} group. Even though there was no significant variation for gross primary productivity (GPP) among the treatment, comparatively lower GPP, 276 ± 94 mg C m^{-3} h^{-1} was noticed in T_{10} compared with T_{20} (314 ± 88 mg C m^{-3} h^{-1}). Mean values for chlorophyll a were 9.0 ± 1.2 and 11.0 ± 3.2 µg L^{-1} in T_{10} and T_{20} respectively (Fig.2). Total heterotrophic bacterial (THB) count and total *Vibrio* count (TVC) in the water sample during the culture period are presented in Table 2. THB count ranged from 2.5 ± 0.2 $\times 10^4$ to 86.0 ± 52.0 $\times$ 10^4 cfu ml^{-1} and TVC from 4.0±1.2 x 10^1 to 149.0 ± 66.0 $\times$ 10^1 cfu ml^{-1}. No significant difference ($p>0.05$) in THB or TVC load was observed among the treatments.

Average growth rate of shrimps over the time period among the treatments are presented in Fig. 3. Banana shrimp showed significant difference ($p<0.05$) with regard to final body weight, daily growth rate and total productivity among the treatments (Table 3). At the end of the production period, a mean final weight of 14.1 ± 0.84 g and 11.1 ±0.14 g were achieved in T_{10} and T_{20} respectively. However, significant increase ($p<0.05$) in average productivity by 50.5 % was observed in T_{20} (990±7.07 kg ha^{-1}) compared with T_{10} (658± 65.05 kg ha^{-1}). Similarly, significant increase ($p<0.05$) in revenue by 33.3% was observed in T_{20} compared with T_{10}. In this study, though FCR in both treatments were insignificantly different ($p>0.05$, $p=0.051$), T_{10} recorded a lower FCR (2.1±0.14) compared with T_{20} (2.7±0.11). The daily growth rate of 0.11± 0.006 and 0.08± 0.001 g day^{-1} were observed in T_{10} and T_{20} respectively.

Table 2. Water quality parameters and microbial counts (Mean value ± SD) in banana shrimp culture ponds at different stocking densities based on independent student's t-test

Water quality parameters	**T_{10} (n=9)**	**T_{20} (n=9)**	**Significance**
Temperature (^oC)	33.45 ± 1.71 (29-35)	33.54 ±1.74 (29.0-35)	NS
pH	8.11 ± 0.14 (7.8-8.30)	8.09±0.14 (7.76-8.30)	NS
Salinity (ppt)	14.9 ± 3.8 (8.4-20)	14.9 ± 3.8 (8.3-21)	NS
Alkalinity (mg $CaCO_3$ L^{-1})	132.20 ± 10.34 (120-160)	133.0 ± 16.9 (96-176)	NS
Transparency (cm)	44.55 ± 17.55(22-85)	38.25 ± 14.41(23-70)	NS
Turbidity (ppm)	21.50 ± 9.69 (6-49)	31.35 ± 12.43 (8-56)	**
DO (ppm)	6.45 ± 0.28 (6.1-7.20)	5.85 ± 0.71 (5-7.80)	**
BOD (ppm)	3.76 ± 1.57 (0.6-5.70)	4.17 ± 1.20 (1.3-5.70)	NS
COD (ppm)	42 ± 10.74 (10-64)	45 ± 15.12 (20-72)	NS
GPP (mg C m^{-3} h^{-1})	276 ± 94. (117-447)	314 ± 88(160-478)	NS
NPP (mg C m^{-3} h^{-1})	151 ± 51.6(87-325)	177.5 ± 57.6(93-315)	NS
TA N (µg L^{-1})	111 ± 49.7 (12.9-186)	113 ± 36.9 (68-196)	NS
NO_2-N (µg L^{-1})	26.5 ± 20.34(1.5-73)	25.5 ± 13.39(4.6-44)	NS
NO_3-N (µg L^{-1})	102.4 ± 38.34(5.6-148)	116.5 ±53.02 (8.3-177)	NS
PO_4-P (µg/l)	27.39 ± 14.98 (5.4-60)	50.97 ± (14.6-140)	**
THC (x10^4 cfu)	25± 34.81 (4.0-20.0)	25± 18.32 (7.0-28.0)	NS
TVC (x10^1 cfu)	67± 63.2 (3.75-148.75)	47.25± 42.07 (8.75-116.25)	NS

.Ranges are in parenthesis. ** $p<0.01$; NS, not significant

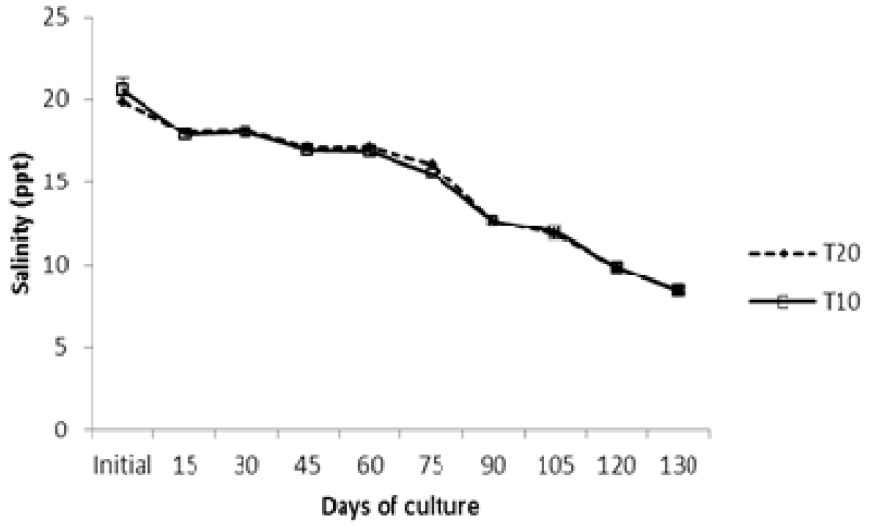


Fig .1. Salinity level of during the grow out culture period at different stocking densities (Mean ± Standard Deviation); n=10

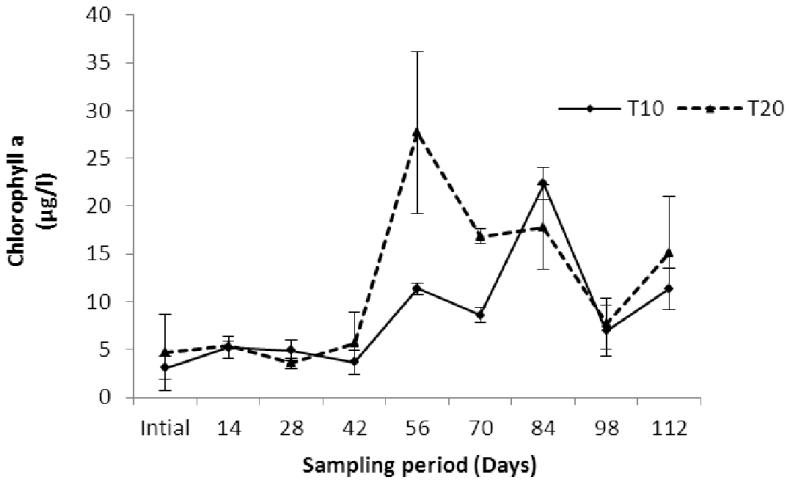


Fig .2. Chlorophyll a concentration (µg L^{-1}) of pond water at different stocking densities (Mean value ± Standard Deviation); n=9

The regression trend indicated variation in the growth pattern for individual sexes (Fig.4 and 5). The slope or regression coefficient b ranged from 3.02 to 3.16. Intercepts (a) varied 0.005 to 0.007. The b values for both the sexes of the treatments were found to be above 3. It shows growth exhibit a positive allometric pattern, i.e. weight increases with increase in length. Effect of stocking density on Fulton's condition factor, K, for both sexes is given in Fig. 6. The condition factor in T_{10} treatment ranged from 0.69 to 0.78 in male and 0.71 to 0.81 in female while in T_{20}, it varied from 0.68 to 0.78 and 0.71 to 0.79 in male and female respectively. Female showed better condition factor, 0.75 ± 0.02 and 0.76 ± 0.03 over male 0.74 ± 0.03 and 0.75 ± 0.03 in T_{20} and T_{10} respectively.

Table 3. Yield parameters (mean ± Standard Deviation) of banana shrimp cultured at different stocking densities based on independent student's t-test

Production Factors	T_{10}	T_{20}	Significance
Average final body weight (g)	14.1 ±0.84 [a]	11.10 ± 0.14[b]	*
Daily weight gain (g)	0.11±0.006[a]	0.08±0.001[b]	*
Survival (%)	47.5±7.78[a]	45.5±0.7[a]	NS
Shrimp production (kg ha^{-1})	658.0±65.05 [a]	990.0±7.07[b]	*
Feed conversion ratio	2.1±0.14[a]	2.7±0.11[a]	NS
Total Revenue (Rs ha^{-1})	1,25,060.59±12324[a]	1,87,625±2015[b]	*

* $P < 0.05$; NS, not significant.

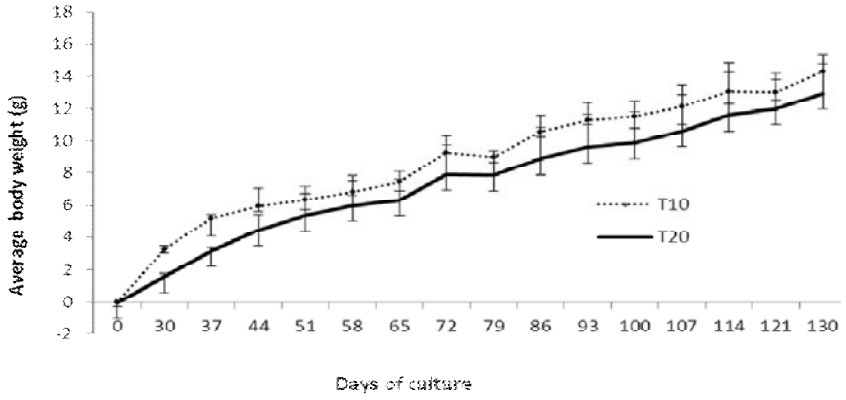
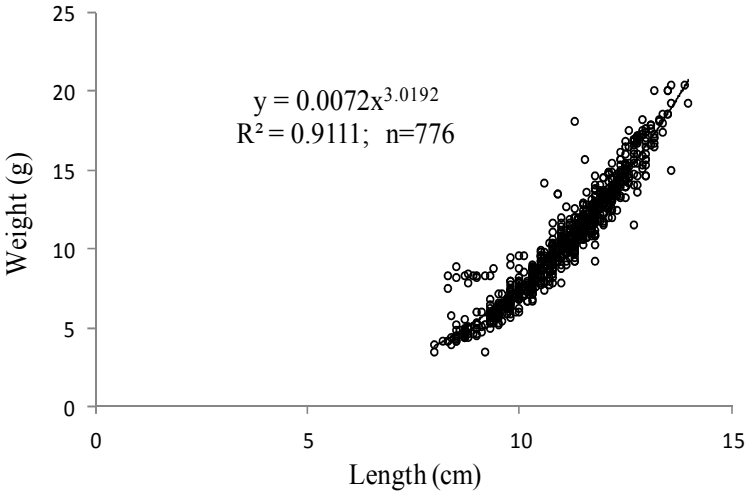


Fig. 3. Average body weight (Mean ±SD) of *F. merguiensis* at two different stocking densities; n=16

a)



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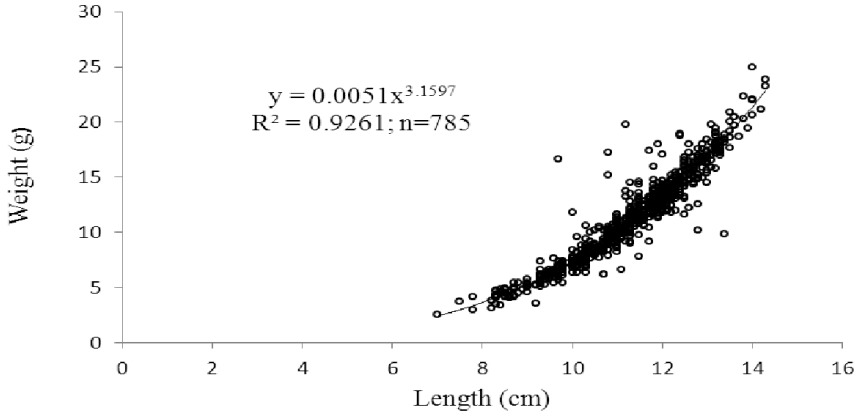
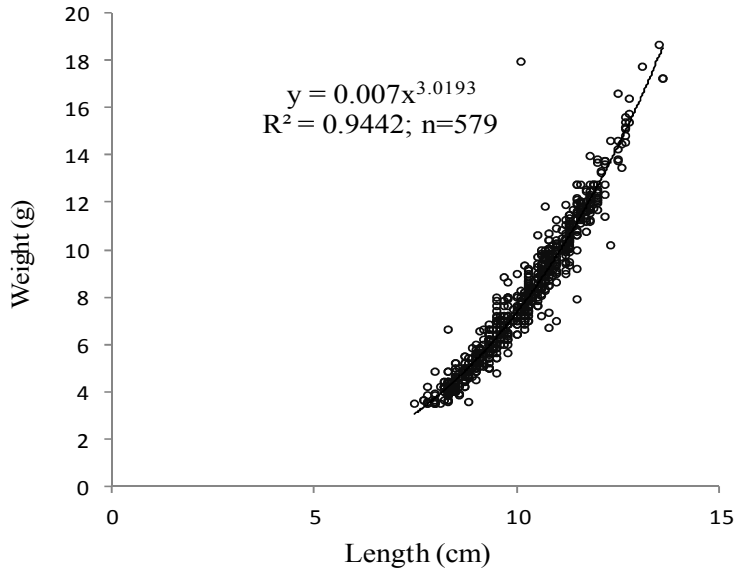


Fig 4. Length-weight measurement of *F. merguiensis* in T_{10} treatment during grows out period. X axis- length in cm and Y axis - weight in gram a) Male b) Female

a)



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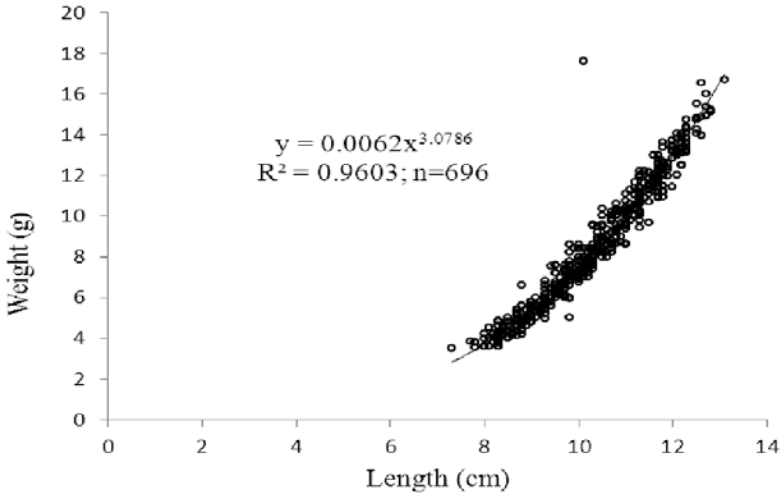
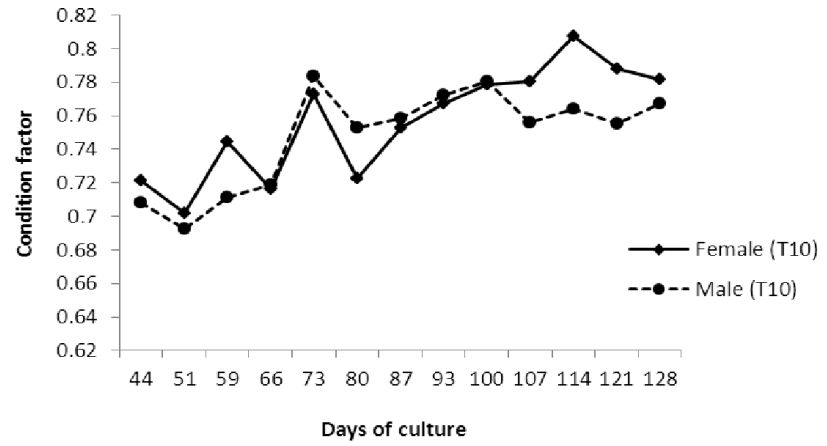


Fig. 5. Length-weight measurement of *F. merguiensis* in T_{20} treatment during grow out period, X axis- length in cm and Y axis - weight in gram a) Male b) Female

a)



b)

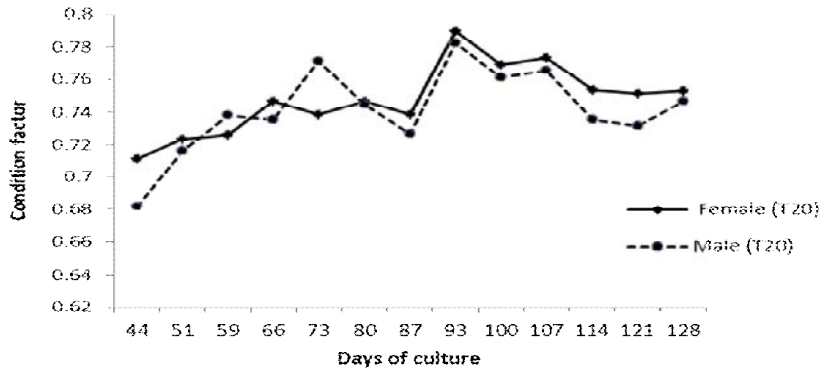


Fig. 6. Condition factor of *F. merguiensis* in T_{10} and T_{20} treatment during grow out period

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Discussion

The recorded water quality parameters and the level of TAN, NO_2-N in both the treatments were within the acceptable ranges for brackishwater shrimp culture[23, 24]. Chlorophyll-a concentration in both the treatments increased insignificantly with the progress of culture and was in similar range of fertilization based *L. vannamei* ponds[9]. However, it was much lower than the value (above 200 µg L^{-1}) reported for highly intensive shrimp ponds[25]. In zero water exchange culture ponds, nutrients keep on accumulate over the culture period and support good natural productivity [26].This suggest the better natural productivity in the present systems. Recorded gross and net primary productivity in the present study was within the ranges for brackishwater culture ponds [27].

In general, increasing shrimp density has negative effect on shrimp growth[12]. The present findings agree in general with the previous reports about the negative correlation between body weights and stocking densities in *L.vannamei*[9], *F. indicus*[10] and *F. merguiensis* [35]. Comparatively, higher FCR observed in T_{20} against T_{10} support that

increases in stocking density reduces feed conversion efficiency [28& 29]. Feed conversion value of 1.5-2.5 for artificial diets (by dry weight) is acceptable in penaeid shrimp culture[30]. Protein content in the commercial feed used in the study was within the acceptable range as it is reported that optimum protein requirement of banana shrimp is 34-42% [31].

Banana shrimp exhibited daily growth rate, 0.11 ± 0.006 and 0.08 ± 0.001 g day^{-1} in T_{10} and T_{20} respectively. In an eight week indoor growth trial of juvenile banana shrimp at different salinities, a daily growth rate of 0.02 g day^{-1} at 15 ppt, 0.1 to 0.12 g day^{-1} at 20 to 30 ppt with the highest growth rate of 0.21 g day^{-1} at 40 ppt was observed [14]. However, monoculture of tiger shrimp in tide-fed ponds attained an average growth rate of 0.27 g day^{-1} [23]. Slow growth of banana shrimp in the present study might be due to the result of decrease in salinity from 21 to 8.3 ppt during the culture period due to monsoon. Adult banana shrimps osmoregulate well between 15–40 ppt, with isosmotic point at 27 ppt [32& 33]. Apart from salinity, temperature also acts a crucial factor which control banana shrimp growth [15& 34]. However, in a tropical country like India, temperature may not have played a decisive role in growth as it was within the acceptable range for growth (29-35^{0}C).

There was no significant difference in survival among the treatments. The findings are in consonance with the earlier report which recorded an insignificant difference in survival rate of *L. vannamei* stocked at different stocking densities[12]. Low survival observed in the present study can be attributed to low salinity noticed during grow out period. It was reported that salinity plays an important role in survival of banana shrimp [34], and survival rate was reported to be very low below 15 ppt[14]. Similarly, a higher survival up to 69.36% and 85.99 % was noticed in high saline experimental ponds with a salinity level of 12-45 and 21-45 ppt in 130 days culture at 20 and 10 nos. sq. m [35]. However, productivity around 990 kg ha^{-1} which is 50.5% higher compared to T_{10} indicates the potentials of high density culture of banana shrimp in low saline systems.

Length-weight relationship in cultured organisms indicates rate of feeding and wellbeing of the organism[36]. 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 [37, 38]. In general, penaeid shrimp growth follows a sigmoidal pattern[16]. Linear function satisfactorily describes growth over size ranges of juvenile *F. merguiensis* [8& 39] and *P. monodon* [40]. In the present study, b value greater than 3 indicates positive allometric growth with more increase in weight than length. This results matches with the previous findings which state b value in the range of 3.03 and 3.22 for *P. monodon* [41]. Stocking density affects length- weight relationship of the cultured shrimps and b value less than 3 indicate crowded conditions or feeding problems in culture system [12& 42]. Even though density depended relationship was not observed in the present study, positive allometric growth shows that cultured animals were in better conditions in both the treatment without much stress. 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* [7], *F. indicus* [43], *P. monodon* [40]and *L. vannamei* [22]. Similar Fulton condition factor observed in both the treatments reflects better feed utilization, less competition for feed and more suitable culture environment even at higher stocking density.

Conclusion

From the above result, it can be concluded that increasing stocking density of *F. merguiensis* result significantly higher production with no significant difference in survival. The findings also suggest that a final body weight of 11-15g is attainable in banana shrimp culture in low saline environment during the monsoon months. Alternatively, it supports the potential of the banana shrimp culture in low saline coastal districts during summer periods when water salinity is above 15 ppt. A crop rotation of *P. monodon* with *F. merguiensis* may also be an option as this would reduce pressure on the wild broodstock of the former, and may also reduce the chances of recurrence of disease prevalence in *P. monodon* culture systems. Physiological impact of salinity on growth, productivity and food consumption pattern of banana shrimp in grow out culture is a matter of further research.

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