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Research Article

Effect of Stocking Density on Growth Performance of Black Tiger Shrimp (Penaeus Monodon) in Organic Aquaculture System

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Abstract

Organic shrimp aquaculture prioritizes natural feeds, sustainable practices, and avoids antibiotics and chemicals, promoting a healthier production. This study investigated the optimal stocking density for black tiger shrimp (Penaeus monodon) in organic aquaculture. Shrimp were reared at densities of 5, 7, and 9 shrimp per square meter (PL/m²) in ponds for 120 days. Water quality parameters during this period were: temperature (28.84-37.94°C), pH (6.47-8.81), dissolved oxygen (5.25-10.67 mg/L), ammonia (0.001-0.10 mg/L), and total dissolved solids (3.35-11.43 mg/L). Pond bottom soil quality included organic carbon (0.93-1.87%), organic matter (0.04-0.27%), total nitrogen (0.70-2.13 µg/g), total phosphorus (0.24-0.70 µg/g), and potassium (0.42-1.3 mEq/100 g), with nitrogen increasing at higher stocking densities. Chlorophyll a concentration ranged from 4.93 to 17.01 µg/L, and bacterial load increased with stocking density in T1 (0.21 ± 0.07×10³ CFU/mL). PCA showed higher densities led to increased ammonia from faeces and uneaten feed, raising nitrogen and Vibrio in sediments. Pearson's correlation indicated a relationship between nitrogen, phosphorus, and chlorophyll a production. The highest weight gain was in the lowest density treatment (T1: 27.54 ± 0.52 g), with the lowest in the highest density (T3: 19.08 ± 0.59 g). Survival rates ranged from 38.88% to 59.77%, with T1 having the highest survival. Specific growth rate (SGR) decreased with density, and the best feed conversion ratio was in T1 (1.50 ± 0.08). The benefit-cost ratio was highest in T1 (1.85 ± 0.03), recommending an optimal stocking density of 05 PL/m² for profitability and healthier yield.

Keywords: Black Tiger Shrimp; Stocking Density; Bottom Sediment; Vibrio and Organic Culture

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Introduction

Bangladesh is regarded as one of the world's best locations for raising shrimp due to its resources and favorable agroclimatic conditions [1]. The extensive regions of shallow water and subtropical climate offer a special environment for the production of black tiger shrimp (Penaeus monodon) [2]. Shrimp farming is a significant export industry for Bangladesh in coastal locations. From 1.52 million MT in 2001-02 to 2.51 million MT in 2020-21, the overall production of shrimp grew [3]. Early in the 1980s, this culture was being practiced along the coast to supply markets abroad and generate foreign exchange. Despite the export potential and jobs created, shrimp farming has resulted in significant environmental costs. The International Federation of Organic Agriculture Movements (IFOAM) published the first fundamental standards for organic aquaculture in 1998, making it a relatively new idea. The goal of organic aquaculture is to create fish and other aquatic products that are ethically, environmentally and socially responsible [4-6]. It has been reported by [6,7] and Sutherland, (2002) that organic aquaculture impacts the environment significantly, decreases production costs, and promotes environmental responsibility.

In the past few years, organic shrimp aquaculture has become a popular farming endeavor on Bangladesh's southwest coast, offering great profitability as a means of reducing the poverty of marginal shrimp farmers and promoting environmental sustainability [8]. In Bangladesh, organic shrimp farming is very important for a number of reasons. Sustainable agricultural techniques that reduce environmental effect are promoted by organic shrimp farming. The use of synthetic pesticides, antibiotics, and growth hormones is avoided by farmers who follow organic farming practices. However, organic aquaculture is a long-standing tradition, particularly in Asia and Bangladesh. Currently, Bangladeshi customers choose organic food over conventional food due to its distinct qualities, including safety, environmental consciousness, nutrition, and sensory qualities [9]. As a result of rising consumer demand for organic products in the worldwide market, organic shrimp production in Bangladesh has significant potential [10-12].

Various kinds of shrimp production systems, traditional, extensive and organic, with varying stocking densities and management strategies, are primarily used by farmers in Bangladesh. Optimal stocking density in aquaculture is the triggering factor for increasing survival and production. Organic shrimp stocking density is mostly influenced by the culture tank's state and the management techniques used. The key to reducing environmental risk and increasing productivity per unit is to have shrimp stocked at the optimal density. However, there is very little information available about a moderate stocking density of shrimp using synchronized culture technology that may be socially, economically, and environmentally acceptable. The data in this article suggested that a moderate organic shrimp stocking density would be better for increasing per unit production than the current level in Bangladesh's southwest coastal regions. In this context, the current experiment was carried out with the goal of enhancing organic shrimp by thoroughly employing EU Organic Aquaculture Standard Regulation (EC) No 710/2009. This study aims to optimize the stocking density of shrimp inside a modified improved culture system and to assess its impact on the culture environment.

Materials and Methods

Study area and pond preparation

The study was conducted at the pond complex of Shrimp Research Station, Bangladesh Fisheries Research Institute (BFRI) in Bagerhat from April to July of 2023. After being drained out and re-excavated to clear away the polluted layer of bottom mud, all of the chosen ponds were entirely exposed to the sun for 5-10 days to raise hydrogen sulphide oxidation capacity and get rid of other unpleasant gases. Ponds were prepared by repairing embankments and clearing weeds of various kinds. To maintain post-larval shrimp in rearing ponds for a short period of time (10-15 days), approximately 10% of the area of each treatment was surrounded by nylon mesh secured with a bamboo frame. The surrounding pond area was biosecurity walled off with blue net to keep out virus-carrying organisms. Prior to the study, ponds were given a 250 kg/ha agricultural lime (CaCO3) treatment based on the pH of the soil. Small mesh filter nets were used to fill the ponds with tidal water till a depth of 1.0 m and the water was disinfected and cleared of all animal life using chlorine at a concentration of 20 ppm. Organic fertilizers such as fermented mixes of molasses, rice bran, and yeast were administrated to the ponds in a 40:25:0.12 kg/ha ratio. After applying molasses for two to three days, 60 kg of liquid mustard oil cake was applied. The water's color changed to green after 4-5 days of fertilization.

Experimental design

The experiment had three treatments comprising of T1, T2 and T3 each with three replications having the unique farm size (607 m^2) having a stocking density of 5, 7 and 9 ind./m^2. During the experimental period, small amounts of extra feeds such wheat bran, rice bran, and mustard oil cake (20%) were frequently employed in the existing organic pond. According to the principles, regulations, and requirements set by the [13] and the International Federation of Organic Agriculture [14], a feed that contains at least 35% protein, as indicated in the table 1, formulated using organic products that were available locally. Proximate composition of the formulated feed was assessed to ensure nutrient percentage (Table 2). Shrimp were then fed three to five percent of their entire body weight twice a day, in the morning and the evening.

Water quality monitoring

Dissolved oxygen (DO), temperature, pH, salinity, total alkalinity, and ammonia contents of the pond water were measured between 9.00 and 10:00 am after seven-day intervals. Salinity was measured using a transportable refractometer (ATAGO). A common centigrade thermometer was used to measure the surface water's temperature. A digital multimeter (HQ 40d digital multimeter, HACH) was used to record the water's pH and dissolved oxygen levels. Titrimetric analysis was utilized to calculate the total alkalinity [15]. An ammonia test kit (HI 3824) was used to determine the ammonia nitrogen level.

Ingredients	Diet- (%)	Total protein (%)
Fish meal	23	16.1
Soybean meal	28	11.74
Rice bran	39.3	6.42
Wheat flour (Binder)	7	0.74
Vitamin premix	1	-
Mineral premix	1	-
Limestone	0.7	-
Total	**100**	**35**

Table 1: Percentage of ingredients used for feed formulation.

Components	Calculated value (%)
Protein	35.60 ± 1.03
Lipid	3.15 ± 0.94
Moisture	8.04 ± 0.42
Ash	12.55 ± 0.82
Fiber	8.21 ± 0.23

Table 2: Proximate composition of feed.

Note: Each component was analyzed triplicate

Bottom sediment collection

Sediment samples was collected from the top 15–100 cm of the shrimp ponds with a handheld acrylic core (50 mm diameter). Then the soil sample was dried and grind properly. Soil texture, salinity, pH, organic carbon, total nitrogen, phosphorus and potassium was analyzed in the laboratory of SRDI, Khulna.

Bacterial count

Bacterial load of the collected samples was analyzed in the laboratory of Shrimp Research Station, Bagerhat after organic feed application in treatment ponds. Thiosulfate–citrate–bile salts–sucrose (TCBS) agar media was used to identify specific bacteria. Samples of bottom sediment from the project pond was collected and the sample was streaked in TCBS agar by spread plate method. Then incubate at 37°C for overnight. Then colony was counted using digital colony counter.

Sampling of shrimp

15-20% of the shrimp is sampled every two weeks to determine biomass, control feeding amounts, and assess the physical health of the shrimp. Cast nets were used to sample shrimp. To observe the growth performance, the length and weight of 30 shrimp individuals were recorded. Length (cm) was determined using a measuring scale and weight (g) was determined using a portable balance.

Growth performance

After 120 days of cultivation, the water was drained out from the ponds and all shrimp were caught by frequent netting. To calculate the survival rate, growth, and production of every shrimp captured from each pond, these factors were individually numbered, measured, and weighted. According to Pechsiri and Yakupitiyage's equation, specific growth rate (SGR) and survival rate (%) were calculated [16]. The formulas used to determine growth performance are as follows:

$$\text{Weight gain (g)} = \text{Mean final weight (g)} - \text{Mean initial weight (g)}$$

$$\text{SGR (\%/day)} = \frac{\{\text{Ln(Final body weight)} - \text{Ln (Initial body weight)} \times 100\}}{\text{Days of culture}}$$

$$\text{Survival rate (\%)} = \left(\frac{\text{Number of shrimp harvested}}{\text{Total number of shrimp stocked}}\right) \times 100$$

$$\text{Yield of shrimp} = \text{Number of shrimp caught } \times \text{Average final weight of shrimp}$$

Feed conversion ratio (FCR) = Total weight of consumed feed / Weight gain of shrimp.

Gross return, net profit, and benefit cost ratio (BCR) economic profitability characteristics were calculated using the following formulas:

$$\text{Total production cost} = \text{Total variable cost} + \text{Total fixed cost}$$

$$\text{Gross return} = \text{Total shrimp yield (Kg)} \times \text{Price of shrimp(BDT)}$$

$$\text{Net profit (BDT)} = \text{Gross return} - \text{Total production cost}$$

$$\text{Benefit cost ratio (BCR)} = \frac{\text{Net profit}}{\text{Total production cost}}$$

Economic analysis

The total amount of money spent on shrimp production is referred to as the production cost. Cost items of shrimp farming practices were classified into two main categories, such as (a) variable costs and (b) fixed costs. Costs that directly change in response to output volume are known as variable costs. Conversely, fixed costs remained unchanged when production changed. The items of variable cost (Human labour, Shrimp fry (Post Larvae), Feeds, fertilizer, transport, construction of water supplying canal, guard shed and housing cost) and fixed costs (land use/ Lease value, interest on operating capital (OC), depreciation on firm implements and miscellaneous costs) which were engaged in the shrimp farming.

Gross return is the monetary value of the entire product. Gross returns per hectare are computed by multiplying total production by market prices. In general, net profit is referred to as entrepreneur's income. Net profit analysis is critical for determining the profitability of shrimp production. Net profit is calculated as the difference between gross returns and total production costs. The benefit-cost ratio was calculated by dividing net profit by total manufacturing expenses. It denotes return per taka invested. It is useful in determining the farm's financial efficiency. All information was gathered from the Shrimp Research Station in Bagerhat and cross-referenced with wholesale market rates. The prices are listed in Bangladesh Taka, which is the country's currency.

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Statistical data analysis

The Shapiro-Wilk and Levene tests were used to verify that the collected data were normal and that the variances were homogeneous. Data analysis was done using the Statistical Package for the Social Sciences, Version 25 (SPSS, Chicago, IL, USA). Statistical significance was established using a one-way ANOVA, and a 95% significance level was taken into account. The Duncan Multiple Range Test was used to compare the means of growth, feed efficiency, and economic profitability between the experimental groups. Next, using PAST software (Paleontological Statistics, Version 4.07), the observed water and bottom sediment parameters were subjected to factor and principal component analysis (PCA) for the variables that cause the co-variation among the data groups. To ascertain the relationship between the variables, the Pearson correlation was used.

Results

Water quality parameters

The variation in water quality is presented in Table 3. During the culture periods, all the water quality parameters were almost the same ($p > 0.05$) among the treatments. Water temperature ranged from 28.84–37.85°C during the experimental period. The average dissolved oxygen value was always > 7 mg/L. Water transparency ranged from 13-33 cm, decreasing over the culture period. The water mean pH value among different stocking ponds was 7.67 ± 0.19, respectively. Water salinity ranged from 3.38-11.85 ppt among the treatments during culture months. The alkalinity ranged from 119-190 mg/L with a mean value of 141.93 ± 2.36 mg/L among different stocking ponds. The water ammonia (NH3-N) level was always < 0.1 mg/L in culture ponds, except for a slight rise (0.021 ± 0.02 mg/L) in treatment 2 (T2), and that was not significant ($p > 0.05$) respectively. Total dissolved solids ranged from 3.3-11.43 mg/L, and conductivity ranged from 6.21-19.90 µS/cm, respectively and did not significantly vary (Table 3).

Parameters	T1	T2	T3
Water temperature (°C)	32.21 ± 0.91^{a}	32.21 ± 0.70^{a}	32.40 ± 1.03^{a}
	28.84-37.85	29.3-35.99	29.2-37.94
DO (mg/L)	8.18 ± 0.32^{a}	7.70 ± 0.42^{a}	7.63 ± 0.61^{a}
	7-9.72	6.24-10.04	5.25-10.67
Transparency (cm)	24.89 ± 1.18^{a}	21.11 ± 1.65^{a}	18.89 ± 1.63^{a}
	21-33	16-30	13-25
pH	7.51 ± 0.21^{a}	7.56 ± 0.23^{a}	7.50 ± 0.23^{a}
	6.47-8.81	6.5-8.5	6.6-8.7
Salinity (ppt)	6.86 ± 0.91^{a} 3.42-11.85	6.80 ± 0.92^{a}	7.14 ± 0.88^{a}
		3.38-10.57	3.55-10.81
Alkalinity (mg/L)	155.56 ± 7.86^{a}	154.67 ± 3.86^{a}	153.78 ± 5.20^{a}
	119-190	139-172	132-173
NH3-N (mg/L)	0.001 ± 0.00^{a}	0.021 ± 0.02^{a}	0.001 ± 0.00^{a}
	0.001-0.002	0.001-0.10	0.001-0.002
TDS (mg/L)	6.51 ± 3.33^{a}	6.95 ± 1.18^{a}	6.47 ± 1.07^{a}

	3.35-11.43	3.30-9.51	3.80-8.79
Conductivity (µS/cm)	13.79 ± 5.02^{a}	12.76 ± 1.92^{a}	11.75 ± 1.78^{a}
	6.35-19.9	6.21-16.82	7.45-15.60

Table 3: Water quality parameters (mean ± standard error) of experimental pond water at three different stocking densities (T1: 5 ind/m^2; T2: 7 ind/m^2 and T3: 9 ind/m^2).

Values in each row with different superscripts are significantly different (P < 0.05).

Values in the parenthesis indicate the range of the parameters.

Bottom soil quality parameters

Culture pond bottom soil quality parameters were depicted in Table 4. Pond bottom soil salinity varied with treatment 1 compared to T2 and T3 (Table 4), T1 soil recorded with a saline soil (6.1 ± 1.2 mS/cm) respectively. Soil pH level did not significantly vary among culture ponds. Soil organic carbon was significantly varied among ponds, T3 recorded with high organic carbon (2.75 ± 0.31%) compared to T1 (1.69 ± 0.21%) and T2 (1.03 ± 0.14) respectively. Organic matter was recorded significantly higher in T3 (1.87 ± 0.11%) rather than T1 (1.16 ± 0.42%) and T2 (0.93 ± 0.26%) respectively. Among the groups, total nitrogen was recorded from 0.2-0.30%, total phosphorous ranged from 12.74-17.01 µg/g and potassium level was 0.6-0.90 mEq/100 g soil and not significantly varied, respectively.

Parameters	T1	T2	T3
Electrical conductivity (mS/cm)	6.1 ± 1.2^{a}	5.3 ± 0.98^{b}	5.6 ± 1.0^{b}
	3.42-11.85	3.38-9.73	4.01-9.09
pH	8 ± 0.56^{a}	8.1 ± 0.23^{a}	8.2 ± 0.47^{a}
	7.21-8.97	8.02-8.78	7.74-8.73
Organic carbon (%)	1.69 ± 0.21^{a}	1.03 ± 0.14^{b}	2.75 ± 0.31^{c}
	7.21-8.97	7.21-8.97	7.21-8.97
Organic matter (%)	1.16 ± 0.42^{a}	0.93 ± 0.26^{a}	1.87 ± 0.11^{b}
	0.89-1.79	0.43-1.23	0.67-2.13
Total nitrogen (%)	0.13 ± 0.03^{a}	0.11 ± 0.01^{a}	0.17 ± 0.01^{a}
	0.05-0.16	0.04-0.19	0.08-0.27
Total Phosphorus (µg/g of soil)	13.74 ± 2.43^{a}	15.07 ± 2.98^{a}	13.91 ± 3.27^{a}
	9.43-15.12	11.82-16.73	11.64-15.21
Potassium (mEq/100 g soil)	0.90 ± 0.13^{a}	0.70 ± 0.19^{a}	0.76 ± 0.24^{a}
	0.54-1.3	0.61-0.99	0.42-0.97

Table 4: Bottom soil quality parameters (mean ± standard error) of experimental pond water at three different stocking densities (T1: 5 ind/m^2; T2: 7 ind/m^2 and T3: 9 ind/m^2).

Values in each same row having different superscripts are significantly different (P < 0.05).

Values in the parenthesis indicates range of the parameters.

Chlorophyll a

The chlorophyll a value showed different pattern in different stocking density (Figure 1). The T1 ranged from 7.78-10.39 µg/L and T3 was in range of 4.93-8.42 µg/L respectively, almost same concentration during the culture period (p > 0.05). On the contrary, T2 showed 10.38-17.01 µg/L and decreasing trend with culture time (Figure 1).

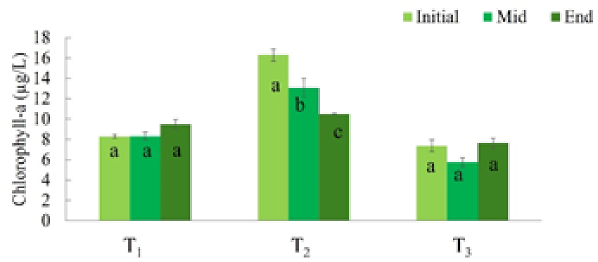

Figure1: Chlorophyll a concentration at different stocking densities. Means among treatments with different letters at a particular sampling period indicate significance (P < 0.05).

Microbial load

Bacterial load in the bottom sediment was found to be low at the initial sample and decreased to almost zero after applying lime, further increasing with the culture duration (Figure 2). The concentration of bacterial load was higher in the end sample (Figure 2). The highest vibrio load was 0.21 ± 0.07 × 103 CFU/mL in T3 compared to T2 (0.16 ± 0.05 × 103 CFU/mL) and T1 (0.07 ± 0.01 × 103 CFU/mL), respectively.

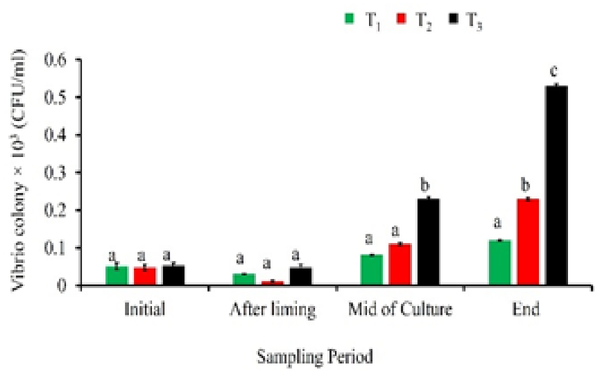

Figure 2: Vibrio load count during the culture at different stocking densities. Means among treatments with different letters at a particular sampling period indicate significance (P < 0.05).

Factor analysis

The factor loadings of the recorded water and bottom sediment parameters is depicted in Table 5. The PCA reveals that the first three principal components explain almost 78% of dataset variation. In the formation of the first principal component, explaining 40.74% of data variance, the ammonia, total nitrogen and vibrio load showed stocking density had a significant impact (Figure 3).

Effects of pond bottom soil quality on water quality

The Pearson's linear correlation coefficient between bottom soil and water quality were calculated (Table 6). Most of the concentration of water quality parameters were positively correlated with total phosphorous and organic matter. This quantity is evidence that soil quality is a crucial nutrient factor influencing water quality in culture ponds.

Media	Parameters	Factor 1	Factor 2	Factor 3
Water	Water temperature	0.035	-0.008	0.051
	Dissolved oxygen	0.014	-0.01	-0.103
	Transparency	0	-0.052	-0.233
	pH	0.017	0.006	-0.031
	Salinity	0.14	0.109	0.319
	Alkalinity	0.008	0.013	0.017
	Ammonia (NH3-N)	0.888	-0.268	0.113
	TDS	0.152	0.148	0.341
	Conductivity	0.139	0.107	0.296
	Chlorophyll-a	0.123	-0.189	-0.199
Bottom Sediment	Soil salinity	-0.016	-0.029	-0.008
	Soil pH	0.001	0.026	-0.006
	Organic carbon	-0.129	0.078	0.585
	Total nitrogen	0.288	0.37	-0.297
	Total phosphorous	0.041	-0.045	-0.028
	Soil potassium	0.006	-0.058	0.024
	Organic matter	-0.046	0.332	0.302
	Vibrio load	0.169	0.765	-0.226
	Eigenvalue	0.287	0.158	0.104
	Percentage of variance	40.746	22.454	14.773
	Percentage of cumulative variance	40.746	63.2	77.973
Possible interpretation		Stocking density influences ammonia concentration from feces and uneaten feed that lead to high nitrogen load and vibrio load in bottom sediment.	Low loading	Low loading

Table 5: Varimax rotated factor loadings from Principal Component Analysis (PCA) and possible interpretations shown as major principal components (1,2,3) of the water quality and bottom sediment.

Loadings marked in bold are considered for data interpretation.

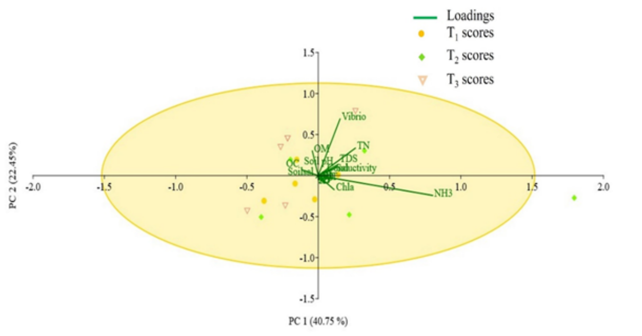

Figure 3: Principal Component Analysis (PCA) biplot of water quality and bottom sediment variables.

Parameters	Soil salinity	OC	TN	TP	OM	Vibrio load
DO	0.05	-0.15	0.1	0.45	-0.14	0.12
Transparency	0.39	-0.48*	0.3	0.25	-0.50*	-0.1
Salinity	0.05	0.05	0.19	-0.04	0.48*	0.13
NH3	-0.24	-0.18	0.3	0.68*	-0.22	0.09
TDS	-0.08	0.13	0.27	-0.17	0.42	0.17
Conductivity	0.03	0.06	0.23	-0.08	0.33	0.14
Chlorophyll a	0.09	-0.06	0.09	0.66*	-0.57*	-0.24

Table 6: Linear correlation co-efficient between bottom sediment and water quality.

Growth performance

The growth of shrimp was observed in three different stocking densities by feeding with organic feed. The length (Figure 4) and weight (Figure 5) of the stocked PL was significantly varied ($P < 0.05$) with increased time among the treatments. At the final harvest, the final length, final body weight, weight gain, average daily weight gain, and SGR were the highest at T1 compared with those in the other treatments (Table 7). The SGR showed stocking density had a high impact on the shrimp growth. Moreover, survival rate was also recorded highest 59.77% ($P < 0.05$) respectively in T1 compared to other treatments. The FCR value was highest (2.12 ± 0.06) in T3, represented lower feed utilization due to stress from high stocking density. In case of total and net productions, T1 showed significantly higher production (829 ± 5.79 and 819.89 ± 5.78kg, respectively) and was lower at T3 (742 ± 25.64 and 720.15 ± 25.63 kg, respectively), although there was no significant difference between T2 and T3. In comparison to all growth parameters stocking density of 5 ind/m^2 showed highest growth and production for 120 days culture period.

Parameters	T1	T2	T3
Initial length (cm)	2.05 ± 1.05^{a}	1.88 ± 1.5^{a}	1.28 ± 0.93^{a}
Final length (cm)	15.34 ± 0.19^{a}	13.26 ± 0.15^{b}	11.22 ± 0.17^{c}
Initial weight (g)	0.0039 ± 0.0001^{a}	0.0035 ± 0.001^{a}	0.0043 ± 0.001^{a}
Final weight (g)	27.54 ± 0.52^{a}	23.11 ± 0.30^{b}	19.08 ± 0.59^{c}
Weight gain (%)	3054.675 ± 87.73^{a}	2364.8 ± 76.70^{b}	2192.2 ± 42.23^{b}
ADWG (g)	0.25 ± 0.01^{a}	0.19 ± 0.01^{b}	0.18 ± 0.001^{b}
SGR (%)	7.69 ± 0.02^{a}	7.47 ± 0.03^{b}	7.17 ± 0.01^{c}
Survival rate (%)	59.77 ± 1.48^{a}	45.77 ± 2.15^{b}	38.88 ± 1.19^{c}
FCR	1.50 ± 0.08^{a}	1.86 ± 0.04^{b}	2.12 ± 0.06^{c}
Total production (kg/ha)	829 ± 5.79^{a}	750.75 ± 27.58^{b}	742 ± 25.64^{b}
Net production (kg/ha)	819.89 ± 5.78^{a}	738.00 ± 27.57^{b}	720.15 ± 25.63^{b}

Table 7: Growth and production performance (mean ± standard error) of shrimp (Penaeus monodon) at three different stocking densities (T1: 5 ind/m^2; T2: 7 ind/m^2 and T3: 9 ind/m^2).

Values in each same row having different superscripts are significantly different ($P < 0.05$)

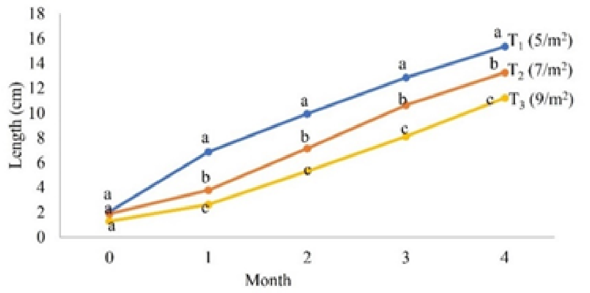

Figure 4: Growth increment (length) of shrimp (P. monodon) for 04 months at different stocking densities. Means among treatments with different letters at a particular sampling period indicate significance ($p < 0.05$).

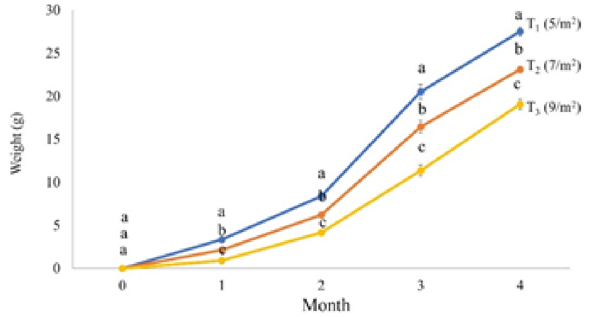

Figure 5: Growth increment (weight) of shrimp (P. monodon) for 04 months at different stocking densities. Means among treatments with different letters at a particular sampling period indicate significance ($p < 0.05$).

Economic analysis

The profitability analysis of P. monodon using organic feed stocked at different densities is described in Table 8. The minimum input value needed was lowest at the lowest density 290987.5 ± 1828.5 BDT, and highest at the highest density (348161 ± 5006 BDT) ($p < 0.05$). Therefore, the overall production cost increased over time. While considering the second scenario, we observed that in the lowest density, a higher percentage of the harvested biomass had a size of > 25 g, which was sold at 1000 BDT per kg. On the contrary, the highest density group had a maximum individual fish of < 25 g that sold at 600 BDT per kg. This resulted in a very low-profit margin with increasing density ($p < 0.05$). The lowest density (5 ind/m^2) had a maximum net profit of 540512.5 ± 4328.5 BDT, while the highest density (5 ind/m^2) group showed the lowest net profit of 134239 ± 14606 BDT. The benefit cost ratio was also higher for T1 compared to the T2 and T3 (Table 8).

Variables	Rate (BDT)*	Treatments		
		T1	T2	T3
Guard cost (120 days)	500/day	60000 ± 0.00^a	60000 ± 0.00^a	60000 ± 0.00^a
Labor cost (pond preparation)	500/day	3000 ± 0.00^a	3000 ± 0.00^a	3000 ± 0.00^a
Lime	25/kg	2500 ± 0.00^a	2500 ± 0.00^a	2500 ± 0.00^a
Shrimp PL	0.90/pc	44971 ± 0.00^a	62959 ± 0.00^a	80948 ± 0.00^a
Organic fertilizer	35/kg	9100 ± 0.00^a	9100 ± 0.00^a	9100 ± 0.00^a
Feed cost	60/kg	81313 ± 2250^a	85392 ± 2528^a	101642 ± 5210^a
Transport cost	-	4000 ± 0.00^a	4000 ± 0.00^a	4000 ± 0.00^a
Harvest cost	5/kg	4157.5 ± 12.5^a	3850 ± 400^a	4020 ± 80^a
Fuel cost	80/L	4000 ± 0.00^a	4000 ± 0.00^a	4000 ± 0.00^a
Land lease	-	60000 ± 0.00^a	60000 ± 0.00^a	60000 ± 0.00
Miscellaneous	-	2675 ± 125^a	2851 ± 99a	3021 ± 179^a
Total variable costs	-	275716.5 ± 2112.5^a	297652 ± 2829^{ab}	332231 ± 4951^b
Total fixed costs	-	15271 ± 284^a	15157 ± 158^a	15930 ± 55^a
Total production costs	-	290987.5 ± 1828.5^a	31280 ± 12987^{ab}	348161 ± 5006^b
Gross return	-	831500 ± 2500^a	616000 ± 64000^b	482400 ± 9600^c
Net profit	-	540512.5 ± 4328.5^a	303191 ± 51013^b	134239 ± 14606^c
Benefit-cost ratio (BCR)	-	1.85 ± 0.03^a	0.96 ± 0.12^b	0.38 ± 0.05^c

Table 8: Economic analysis of P. monodon at three different stocking densities (T1: 5 ind/m2; T2: 7 ind/m2 and T3: 9 ind/m2) using organic feed.

Data are expressed as the mean ± standard error. Different superscripts are significantly different ($p < 0.05$). * 1 US $ = 110 Bangladeshi Taka (BDT).

Discussion

Organic aquaculture led to a high economic performance that improved livelihood and integrate effectively with existing farming practices [17]. Moreover, organic aquaculture conserve ecosystem, reduce non-climatic stressors and combat climatic change impacts on shrimp culture industry [18]. Therefore, the optimum density for organic culture of P. monodon is indispensable. The experiment designed to assess optimum stocking density and effects on growth, microbial load, bottom sediment quality and ultimate production in shrimp culture.

Water quality and bottom soil parameters

Water quality is crucial for shrimp culture, sudden hike of water quality parameters can result in mass mortality. Water quality parameters such as water temperature, DO, transparency, pH, salinity, alkalinity, ammonia, TDS and conductivity was observed. Our studied water temperature was 28.84-37.85° C (Table 3), the optimal level for P. monodon culture is 25-30° C [19]. The higher temperature was recorded due to the summer season. The water pH level was slightly acidic in initial stage, then treated with lime before stocking to keep it in optimum pH level (7.5-8.5) [20].

The average dissolved oxygen level was always more than 4 mg/L, and not significantly varied. Stocking density variation could not interrupt the DO, due to optimum sunlight in the culture season. [21] Mentioned that the optimum range of dissolved oxygen for shrimp and prawn is > 4 ppm which is very similar to findings of the present study. [22] Reported that the salinity should be in the range of 5-32 ppt for shrimp production, which coincides with our findings. The obnoxious ammonia was always < 1 mg/L, which indicates organic feed did not deteriorate water quality [23,24]. Water quality parameters were within acceptable limit and did not change significantly in different stocking densities.

Bottom soil releases nutrients to water and also organic matter and suspended solids deposited on bottom soil. Therefore, bottom soil quality was assessed and found almost similar result in all the ponds with an exception in organic matter. The pH value and EC value of bottom soil indicated saline soil, as the pH < 8.5 specifies saline soil [25] and EC value > 4 are indicated as saline soil [26]. A study by [27] documented similar pH and EC level in shrimp culture pond bottom soil. Organic carbon and organic matter significantly varied between different stocking density cultures. The study recorded higher percentage of organic carbon and organic matter in T3 (9 ind/m^2) and the lowest value was in T1 (5 ind/m^2) (Table 4). The high percentage value was derived from feces and uneaten feed from high stocking density [27]. The bottom soil is medium productive, as within 0.5-2 percent organic matter.

Total nitrogen and total phosphorous was not significantly varied. Soil nitrogen contributed to the phytoplankton production and chlorophyll a concentration due to high salinity in water (10-20 ppt) transform soil nitrogen to available nitrogen [28]. Similar studies that studied on shrimp pond soil available phosphorus content, that was recorded between 0.22 and 4.11 mg/100g in India [29], which coincides with our study. The present study recorded potassium level is high of optimum level (40 mg/L) which have a little benefit. [30] Documented average 193 mg/kg potassium in shrimp pond of Thailand.

The chlorophyll a concentration >10 µg L-1 is recommended by [30] for shrimp culture pond. The study recorded a slight lower concentration in T1 and T3 at initial stage, whereas T2 was recorded within the recommended level. During the experiment, controlled pond fertilization and shrimp feed on those chlorophyll a render the extreme condition and the chlorophyll a level was unchanged. On the other hand, as T2 was recorded with a high concentration therefore fertilization was reduced, and stocked PL thrive on plankton that reduced the concentration with the culture time. This finding also similar with [31].

Higher stocking density has been linked to higher diseases prevalence, although no disease history was found in the study. The study focused on Vibrio due to the mass mortality of shrimp by Vibrio harveyi [32] and high load in bottom sediment as it depends on the amount of uneaten feed and excreta [33]. High stocking density leads to high organic load and higher percentage of microbial load (Figure 2), that indicates Vibrio count of sediment influenced by stocking density. [34] Reported similar increasing pattern of vibrio load in shrimp and culture water. The Vibrio load was in the maximum threshold value (103 CFU/mL – 104 CFU/mL) [35] and in similar range reported by [33,36].

As described in the figure, PC1 explains the positive loading of parameters for ammonia, total nitrogen and vibrio load and negative loading was very low for other parameters. Higher the stocking density need more feed, that leads to more uneaten feed that deposited in the bottom, decomposed and increase the ammonia in water and total nitrogen percentage in the bottom sediment. The Pearson's correlation showed water ammonia exhibited positive relation with total phosphate (r = 0.68) of sediment, this phosphate releases in water and influenced the chlorophyll a production (r = 0.66) (Table); as a stable phytoplankton community is a great concern for a healthy farming of shrimp [37].

Effect on growth and feed utilization

One of the key zoo-technical elements that directly affects the survival, growth, production, and profit of prawns is the stocking density [38]. Prior research has indicated an adverse correlation between shrimp growth and survival and stocking density [34,39,40]. The average survival rate during the culture period was 38.8% - 59.7%, which coincides with [34] and supported increased stocking density reduced the survival rate [41-43]. Reductions in survival of cultured animals at high stocking density could be related to increased competition for space and feed [44].

In the present study higher FCR value (2.12) was observed in the treatment T3 than T1 (1.5%) and T2 (1.8%), where recommended FCR value < 2 [20]. A study on stocking density of P monodon recommended 6 ind/m^2 as found FCR value 1.8 [34]. It can be stated that growth was impacted by a higher stocking density that increased total biomass, which in turn reduced food conversion efficiency with a higher FCR value [41].

In spite of these, individual weight gain by the shrimp of treatment T1 was highest and followed by T2 and T3 (Table 3); due to the lower density that increased the feed efficiency more space that ensure stress free environment. Similarly, values of 7.69, 7.47 and 7.17 for specific growth rate (SGR) found in treatments T1, T2 and T3 might also be due to the effect of stocking density. Similar studies recorded the same pattern of negative co-relation between stocking density and growth in P. monodon [40,45]. The highest total production was 829 kg/ha in T1 followed by T2 and T3. According to [46], stocking density varies from 0.75 to 1.5 shrimp ind/m^2 and the total production was 385.43kg/ha in well managed traditional shrimp farming of Bangladesh. Compare to the above, organic shrimp farming with 5 ind/m^2 can be considered as more promising as the total production is more than two times higher [47].

Profitability analysis

The cost of feed and seed will probably go up when stocking intensity rises. Consequently, the total production cost was relatively higher as the stocking increased. The production biomass of the system, particularly in the grow-out culture system, is more important in determining the net profit and BCR. Up to recently, research has mostly concentrated on surveys on organic shrimp farming and profit analysis. Here, we aimed to produce a more comprehensive economic growth status that included the effects of various stocking densities. According to the benefit-cost ratio and net profit, larger-sized shrimp had a greater market value and higher growth at lower stocking density (5 ind/m^2), which translated into higher profit margins.

Conclusion

In this current investigation, we present compelling evidence indicating that stocking density and feed exert analogous influences on shrimp growth, water quality, bottom sediment quality, and microbial load. Our study systematically elucidates the comprehensive landscape of organic shrimp farming technology, ultimately identifying an optimal stocking density crucial for achieving profitable economic growth and effective water management. Specifically, our findings strongly suggest that a low density of 5 individuals per square meter (5ind/m^2) emerges as the ideal condition, promoting superior water quality, growth, and health status within the context of traditional organic shrimp culture methods. This research underscores the significant sustainability of organic shrimp farming, offering valuable insights from ecological, fish health, and economic standpoints.

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