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**ORIGINAL RESEARCH ARTICLE** 



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# Biofloc based farming of Nile tilapia (Oreochromis niloticus) in tanks under different stocking densities

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ARTICLE HISTORY	ABSTRACT
Received: 08 January 2024 Revised received: 22 February 2024 Accepted: 14 March 2024	This experiment was conducted to optimize the stocking density of Nile tilapia ( <i>Oreochromis niloticus</i> ) in biofloc based farming system in tanks for a period of 90 days from 23 December to 22 March 2022 in Mohanpur upazilla, Rajshahi district, Bangladesh. The experiment was conducted in 5000 L cemented tanks (5m <sup>3</sup> ) under three different of stocking densities (T <sub>c</sub> : 200
Keywords	fishes/m <sup>3</sup> , T <sub>2</sub> : 150 fishes/m <sup>3</sup> and T <sub>3</sub> : 100 fishes/m <sup>3</sup> ) having three replicates each. The initial weight of fish was $11.50\pm0.35$ g and fishes were fed (34% protein content) twice a day @ 5-3 %
Benefit cost ratio Biofloc <i>Oreochromis niloticus</i> Stocking densities	body weight. Among three treatments, a significantly higher (P<0.05) average daily gain (ADG) of $1.15\pm0.07$ g with specific growth rate (SGR) of $2.44\pm0.09$ % day <sup>-1</sup> were recorded in T <sub>3</sub> treatment but a significantly higher (P<0.05) average yield of 118907.70 and 114914.20 kg/ha/3 months were obtained in T <sub>1</sub> and T <sub>2</sub> treatment, respectively. Higher average net benefit of 1231671.90 BDT/ha/3 months and BCR of 0.12 were obtained in T <sub>2</sub> treatment which is significantly higher (p<0.05) than T <sub>1</sub> and T <sub>3</sub> treatment. By an economical assessment, if the sales price is influenced by the final body weight, the reduced average harvest weight in higher stocking densities could lead to low profitability. Therefore, it is concluded that the use of intermediate stocking density, around 150 fishes/m <sup>3</sup> , has higher profitability since it produces a large proportion of harvested fish that reach high body weights, and possible high selling prices, combined with desirable biomass.

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# INTRODUCTION

Aquaculture is one of the fastest growing sectors of animal protein production in the world. Excluding algae, total world fisheries and aquaculture production showed a 45 percent growth between 2000 and 2021, reaching 182 million tons in 2021 and setting a new production record (the previous one was 179 million tons in 2018) (FAO, 2023). This represents an overall expansion of 56 million tons compared to 2000. In 2021, the total fisheries and aquaculture production (excluding algae) rose significantly (+2.7 percent) compared with 2020 after two years of relative decline contributing to the reduction of population poverty, hunger and malnutrition, generating economic growth and guaranteeing natural resources (FAO, 2023). Finfish (freshwater, diadromous and marine fish) had a share of 76 percent of the total in 2021, with a slight decline compared to 79 percent in 2000. With 38 percent of the total, marine fish were the main group of species produced in 2021, followed by freshwater fish (33 percent), mollusks (14 percent) and crustaceans (10 percent) (FAO, 2023).

Global production of tilapia in 2020 was almost 7 million tons. Currently a wide variety of tilapia species are cultivated but Nile Tilapia is the most commercialized worldwide, ranking third among farmed finfish with 8.33 percent (FAO, 2023). According

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to The State of World Fisheries and Aquaculture 2020 report by the Food and Agriculture Organization, Bangladesh stands as a global leader, ranking 3rd in fish extraction from open water bodies and 5th in aquaculture production. Furthermore, the country boasts a prominent position, ranking 4th globally and 3rd in Asia, in tilapia fish production (FAO, 2023). However, growth stumbles on the limited availability of natural resources (Verdegem, 2013), requiring the development of systems to increase production and productivity with less use of water, space and energy (FAO, 2023), yet with satisfactory economic returns. In this context, intensive aquaculture systems are challenged by providing a favorable environment for high-density fish and shrimp production with little or no water exchange (Ray et al., 2010). Biofloc crops are increasingly common to meet such need (Avnimelech, 2007; Crab et al., 2007; De Schryver et al., 2008). Biofloc Technology (BFT) involves a closed system for the rearing of aquatic organisms based on nutrient recycling and conversion into microbial flakes, which serve as endogenous natural food for production animals (Azim and Little, 2008). This system is driven by the principle of nutrient recycling through high carbon: nitrogen (C: N) ratio, stimulating the growth of heterotrophic bacteria, which transform ammonia into microbial bioflocs (Burford et al., 2003; Avnimelech, 2015). Biofloc cultivation occupies smaller areas of land and water volume. However, increasing stocking density and minimum or zero water renewal result in the accumulation of feed residues, excreta and toxic inorganic compounds (Burford et al., 2003), thus compromising water quality (Avnimelech, 2007) and, hence, health of the fish. Therefore, the accumulated sludge must be drained (Emerenciano et al., 2011).

Nile tilapias are capable of absorbing suspended bio floc, being adapted to high stocking densities (Avnimelech, 2011). Tilapias are able to efficiently utilize heterotrophic bacteria and are thus suitable for cultivation in biofloc systems (Beveridge *et al.*, 1989). The produced biofloc is rich in nutrients that can be utilized as a food source by filter feeders like Nile tilapia (Avnimelech and Kochba, 2009; Azim and Little, 2008). The BFT also may act as a source of bioactive compounds that improve the defense mechanisms of aquatic animals (Crab *et al.*, 2010; Ekasari *et al.*, 2015; Long *et al.*, 2015). Combining these advantages with a specific breeding program producing a genetically improved Nile tilapia lineage well fitted to BFT, as

described by Galib et al. (2019). However, basic information, such as the suitable stocking density in the grow-out phase of Nile tilapia in BFT has yet to be defined and this is a determining factor for the economic sustainability of tilapia production in BFT (Wuertz et al., 2013; Wang et al., 2019). The determination of an optimal density should be made based on a complex assessment, which should consider the growth performance data, water quality, animal welfare, production costs, and profitability (Wuertz et al., 2013; Wang et al., 2019). Galib et al. (2019) already evaluated the effects of different stocking densities (20, 40, 60, and 80 fish·m<sup>-3</sup>) on the growth performance of Nile tilapia in the grow-out stage in a zero water exchange bio floc system and their quadratic model revealed 51.79 fish·m<sup>-3</sup> (13 Kg·m<sup>-3</sup>) as the maximum stocking density for this kind of water management on BFT. Thus, the aim of this study was to evaluate the effects of different stocking densities of Nile tilapia (Oreochromis niloticus) in grow-out stage reared in BFT. The parameters to be evaluated are water quality, growth performance, survival, production cost, gross income, and profitability of bio floc to establish optimum stocking density.

#### MATERIALS AND METHODS

#### **Experimental site**

The research was carried out for a period of three months (90 days) from December, 2021 to March, 2022 in Mohanpur upazilla, Rajshahi District, Bangladesh (Figure 1). (Latitude 24.547026° N, Longitude 88.686478° E).

#### **Experimental design**

The experiment was conducted in 5000 L cemented tanks (5m<sup>3</sup>) with a useful volume of 4000 L under three treatments of stocking density ( $T_1$ : 200 fishes/m<sup>3</sup>,  $T_2$ : 150 fishes/m<sup>3</sup> and  $T_3$ : 100 fishes/m<sup>3</sup>) each with 3 replications. After finding microbial floc formation and suitable water quality tanks were stocked with Nile tilapia (*Orechromis niloticus*) fry. The mean initial stocking weight of fish fry was (11.50±0.35g). The animals remained for three days in the experimental units with previously fertilized water before the actual start of the experiment. Throughout the experimental period, no water exchange took place, only replacement of the loss by evaporation and decantation.



Figure 1. Location of the experimental site.

#### Tank facilities and preparation

Tanks were well constructed by concrete with inlet and outlet. After construction tanks were bleached for removal of unwanted microbes. Aerator (LP 100) and Nano bubble tube for proper aeration. At the experimental time, the physicochemical Parameter (Temperature, pH, DO, ammonia-nitrogen, nitrite-nitrogen and TDS) were checked and then raw salt, molasses and probiotics were added. To generate bio floc, at first the tanks were filled with water, and then a certain amount of nitrogenous content, such as aquatic feed, was added to provide nitrogen, and finally carbonate organic materials, such as molasses, were added to finish the process. At first Algal growth was developed, followed by froth and the appearance of brown color indicated the presence and activities of bacteria (especially heterotrophic bacteria).

#### Post stocking management

After stocking of fish fry, water probiotics (Pondcare, Sk+F, Bangladesh; containing heterotrophic bacteria of 22\*109 CFU) were used to maintain microbial floc density (20-25ml/L). To maintain the TDS at 700mg/L, raw salt was added. For the first 30 days, fish were fed starter-1 (34 percent protein content), which was used at a rate of 5% of body weight, followed by starter-2 (30 percent protein content) at a rate of 4% of body weight for the second 30 days, and grower (30 percent protein content) at a rate of 3% of body weight for the last 30 days. According to Avnimelech (1999), Molasses was also added after at a rate of 60% of feed applied to tanks to maintain an optimum C: N ratio.

#### Sampling

The water quality parameters of each tank was measured every three days interval between 9.00 am and 10.00 am. Fish were also sampled on a weekly basis to assess their growth performance. Each week, ten percent of the stocked fish were captured using a net from each tank for growth assessment, health condition and feed adjustment.

#### Measurements of physicochemical parameters of water

Every 3 days intervals, physicochemical parameters of water were measured. The water temperature was noted using a centigrade thermometer with a range of 0°C to 100°C. The temperature of the water was measured by submerging the thermometer probe 30 to 40 cm beneath the surface. The total dissolved solids (TDS), dissolved oxygen and pH was measured with the help of HACH water analysis kit (Multi meter; HQ, 40D, HACH, USA). Ammonia-nitrogen and nitrite-nitrogen was also measure by a HACH WQ testing kit.

#### **Microbial floc**

Density of floc was measured at 3 days interval by Imhoff cone. Abundance of heterotrophic bacteria (mainly *Bacillus sp.*) was determined monthly basis by plate count (agar media) method and the values were expressed in Colony Formation Unit (CFU/ ml).

#### **Estimation of yield parameters**

After 90 days of culture period, treatment-wise fishes were harvested by scoop net and hand picking. The individual length (cm) and body weight (g) of fishes (10%) and number of harvested fish were recorded for estimating different yield parameters as follows (Mithun *et al.*, 2020):

Survival rate (%) = Number of fish harvested÷ Number of fish stocked × 100

Specific growth rate (SGR, %day-1) = [ In (final weight) – In (initial weight)  $\times$  100]/ No. of days of the experiment

Final weight gain (g) = Final individual weight – Initial individual weight

Feed conversion ratio (FCR) = Feed applied (dry weight)/ Live weight gain

Net production  $(kg/m^3)$  = total biomass at harvest – total biomass at stocking

BCR = net benefit/total cost

#### **Economic analysis**

For the present study, a simple analysis of cost vs benefit has performed to find out the economics of bio floc-based tilapia farming under various treatments. The total cost (BDT/ha/3 months) was calculated using data on both fixed and variable costs. (BDT/ha/3 months) was the total return calculated from the market price of fish. The total return was deducted from the total cost to arrive at the net benefit, which was stated as (BDT/ ha/3 months).

#### **Statistical analysis**

Growth and yield parameters of fish and BCR were analyzed statistically by a one-way analysis of variance (ANOVA) using the statistical software Statistix 10. The ANOVA was followed by DMRT (Duncan Multiple Range Test) at 5% level of significance.

## **RESULTS AND DISCUSSION**

#### Water quality parameters

Water temperature: The mean values of water temperature (°C) were found to be 19.96. 19.93 and 19.90°C in  $T_1$ ,  $T_2$  and  $T_3$  treatments respectively with no significant difference (p<0.05) (Table 1). This finding agreed with Samuel *et al.* (2021) who stated that water temperature 20-25°C suitable for bio floc based fish farming. However, according to Boyd (1982), suitable range of temperature for fish culture in pond is 26-28°C.

**Dissolved oxygen (DO):** The dissolved oxygen (mg/L) values ranged from 5.10 $\pm$ 0.18, 5.83 $\pm$ 0.27 and 6.04 $\pm$ 0.19 mg/L 5.83 $\pm$ 0.27 mg/L T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> treatments, respectively where a significantly (p<0.05) higher DO value was found in the treatment T<sub>3</sub> (Table 1). This finding agreed with Bhuiyan and Nessa (1998) who stated that DO concentration ranging from 5.0-7.0 mg/L is suitable for bio floc based fish farming. pH: The mean lower and higher values of pH found to be 7.64 (T<sub>3</sub>) and 7.70 (T<sub>1</sub>) respectively with no significant difference (p<0.05) (Table 1). According to Boyd (1998) the suitable range of pH is 6.5 to 8.5 for pond fish culture. Shamsuzzaman *et al.* (2017) reported pH of 6.5 to 9 as suitable for fish culture. In the present study the slightly alkaline pH range in all treatments indicate good pH condition for biological production and fish culture.

Total dissolved solids: The mean lower and higher values of total dissolved solids (TDS) obtained during the study period varied from 678.25±8.55 mg/L (T<sub>2</sub>) and 681.35±2.91 mg/L (T<sub>3</sub>), respectively with no significant difference (p<0.05) (Table 1). This finding strongly agreed with Rostika (2014) who recommended total dissolved solids (TDS) levels up to 1000mg/L can be acceptable for the bio floc based fish farming.

Ammonia-nitrogen (NH<sub>3</sub>-N): The mean value of NH3-N in this study was found to be 0.076, 0.056 and 0.035 mg/L in  $T_1$ ,  $T_2$  and  $T_3$  treatments, respectively with significant difference (p<0.05) among the treatments which is caused due to different stocking density (Table 1). Putra *et al.*, (2020) conducted study on bio floc system and found mean value of 0.03 mg/L NH<sub>3</sub>-N, which is strongly agreed with the findings of present study. Alim *et al.* (2005) who noted NH3-N ranges from 0mg/L to 1.00mg/L. Wahab *et al.* (1995) and McIntosh *et al.* (2000) noted NH<sub>3</sub>-N ranged from 0.09 to 0.99 mg/L and 0.60 to 0.29 mg/L, respectively.

**Nitrite-nitrogen (NO**<sub>2</sub>-**N**): NO<sub>2</sub>-N is an intermediate during both nitrification and de nitrification, (Chuang *et al.*, 2007; Ruiz *et al.*, 2003). The accumulation of NO<sub>2</sub>-N in intensive aquaculture is common (Wang *et al.*, 2004), due to the inhibition of free NH<sub>3</sub> during nitrification and de nitrification (Shi *et al.*, 2011). In the present study, mean value of NO<sub>2</sub>-N was varied from

 $0.003\pm0.0018$  to  $0.0018\pm0.0008$  mg/L (Table 1) and the value is lower than the mean value of NO<sub>2</sub>-N ( $0.25\pm0.12$  mg/L) reported by Putra *et al.* (2020). The concentration of NO<sub>2</sub>-N in the bio floc tanks was significantly lower than that reported in earlier studies (Asaduzzaman *et al.*, 2009 and Xu and Pan, 2012). Based on a recommendation from Wuertz *et al.* (2013), salt was added to the BFT tanks (1%, w/w-1) to compensate for the stress from NO<sub>2</sub>-N to the tilapia in the current experiment.

**Microbial floc density:** Probiotic bacteria in Bio floc system convert ammonia to nontoxic materials such as nitrate, which is favorable to phytoplankton growth. As a result, the ammonia and nitrate concentrations in the culture media remain low (Hargreaves *et al.*, 2007). Bio floc is made up of various beneficial microorganisms such as microalgae and zooplankton that are bound by organic particles, rather than bacteria (Crab, 2007). In the present experiment mean value of microbial floc density was found at the ranged from 25.0±0.00 to 26.35±1.91 ml/L (Table 1). The lower value was noted in T<sub>2</sub> treatment and the higher value was noted in T1 treatment. This finding more or less agreed with floc volume 40±5 ml/L which was reported by Scott Bowerbank Day (2015).

Bacterial load (CFU/ml): Floc density operates on the principle of increasing carbon to nitrogen ratios, through the addition of an exogenous carbon source that consequently stimulates natural heterotrophic bacterial growth in the system (Hargreaves, 2007; De Schryver *et al.*, 2008). In the present study, abundance of heterotrophic bacteria (*Bacillus sp.*) was found to be  $1.17\pm0.005 \times 10^{15}$ ,  $1.97\pm0.013 \times 10^{15}$  and  $2.68\pm0.025 \times 10^{15}$ CFU/ml in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> treatments, respectively with significant difference (p<0.05) among the treatments and that is the main thought of food producing for experimented biota (Nile tilapia) (Table 1). This thought supported by various researchers (Avnimelech, 1999; Hargreaves, 2007; Avnimelech, 2009).



Figure 2. Mean values of economic analysis of different treatments.

The heterotrophic bacteria's conversion of nitrogen to protein eventually regulates the levels of toxic nitrogenous compounds in the system. The carbon/nitrogen ratio (C/N), which is regarded as a control parameter, affects how effective heterotrophic bacteria in the BFT system are at converting toxic nitrogen compounds into bacterial biomass (Emerenciano et al., 2017; Avnimelech, 1999). To maintain optimum activity of heterotrophic bacteria, the C/N ratio should be kept within the range of 15-20:1 (Avnimelech, 2009). The control of accumulating organic nitrogen and the production of microbial communities in water are both depending on the C/N ratio being maintained (Asaduzzaman et al., 2008; Emerenciano et al., 2011). When molasses was added as a carbon source and the C/N ratio was maintained at 10:1, it was found that around 10 mg NH4+N could be completely absorbed. In bio floc waters, the practice of accelerating C: N ratio greater than 10:1 by utilizing various low -cost carbon sources that are locally available is typical to reduce synthetic feed requirement (Crab et al., 2010). Using bio floc components reduces the amount of protein in the diet, in addition to cutting feed costs (Avnimelech, 1999; Hargreaves, 2007). It was found that when the microbial population keeps the C/N ratio high in the bio floc system due to ammonium consumption, the formation of hazardous inorganic components such as  $NH_4^+$  and  $NO_2$  would be stopped within the water.

**Fish growth parameters:** In intensive aquaculture, the stocking density of a production system is an important determinant of its economic viability. Although certain species have shown a

favorable influence of stocking density on growth, it is generally accepted that stocking density is a significant component in the growth and survival of many aquatic animals (Rahman *et al.*, 2021). The volume of water or surface area per fish is related to stocking density. Increased stocking density causes increased stress, which leads to increased energy demands, resulting in a decrease in growth rate, food usage, and water quality degradation. Lower stocking density requires fish to compete less for food and space than greater stocking density. The current study presents empirical evidence on the impacts of Nile tilapia stocking density on growth and survival, finding that they were higher in bio floc tanks with lower stocking density than in bio floc tanks with higher stocking density.

Final weight (g): Bio floc technology is an environmentally friendly aquaculture approach, which has been tested for numerous aquaculture species, including tilapia, both in the field on a commercial scale (Azim and Little, 2008; Avnimelech, 2007). There is general consensus that the production performance of tilapia can be enhanced by application of bio floc technology (Avnimelech, 2007). In the present study the highest final weight of 124.50±5.66 g was found in T<sub>3</sub> treatment followed by T<sub>2</sub> (105.0±4.24g) and T<sub>1</sub> (83.90±3.68g) treatment with significant difference (P < 0.05) (Table 2). The findings of present study more or less similar with (Azim and Little, 2008; Luo, 2014; Long *et al.*, 2015) which may be due to participate of floc aggregates as an additional source of feed.

Table 1. Mean values of water quality parameters of different treatments during experimental period.

Deveryeter	Treatments				D)/alwaa
Parameter	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	F values	P values
Temperature (°C)	19.96±0.06a	19.93±0.06a	19.90±0.10a	0.600	0.579
DO (mg/L)	5.10±0.18b	5.83±0.27a	6.04±0.19a	14.957	0.005
pH	7.70±0.025a	7.65±0.025a	7.64±0.050a	2.575	0.156
TDS (mg/L)	681.28±2.85a	678.25±8.55a	681.35±2.91a	0.314	0.742
NH <sub>3</sub> -N (mg/L)	0.076±0.0035a	0.056±0.011b	0.035±0.0006c	28.312	0.001
NO <sub>2</sub> -N (mg/L)	0.003±0.0018a	0.0018±0.0008ab	0.000±0.000b	5.193	0.049
Floc (ml/L)	20.23±0.29a	20.32±0.26a	19.6±2.90a	0.162	0.854
Bacillus sp. (CFU/ml×10 <sup>15</sup> )	1.17±0.005c	1.97±0.013b	2.68±0.025a	1.141	0.380

Common letter(s) in a same row significantly different (p<0.05).

Table 2. Mean values of growth parameters of different treatments during experimental period.

Parameter —			DValues		
	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	r values	P values
Initial weight (g)	11.75±0.35a	11.85±0.77a	11.15±0.49a	1.35	0.328
Final weight (g)	83.9±3.68c	105.0±4.24b	124.5±5.66a	58.39	0.000
Weight gain (g)	72.15±4.03c	93.15±3.46b	113.35±6.15a	57.84	0.000
Survival rate (%)	92.0±1.41a	92.0±1.41a	92.5±0.14a	0.188	0.834
SGR (%, bwd <sup>-1</sup> )	1.98±0.08c	2.21±0.02b	2.44±0.09a	31.95	0.001
ADG (g)	0.73±0.04c	0.95±0.04b	1.15±0.07a	49.03	0.000
Yield (kg/ha/3months)	118907.7±8552.7a	114914.2±2863.9a	93611.1±4937.6b	15.25	0.004
FCR	1.06±0.04c	1.14±0.02b	1.18±0.01a	17.32	0.003

Common letter(s) in a same row significantly different (p<0.05).

Weight gain (g): The highest weight gain of  $113.35\pm6.15$  g was found in T<sub>3</sub> treatment followed by T<sub>2</sub> (93.15±3.46 g) and T<sub>1</sub> (72.15±4.03 g) treatments with significant difference (P < 0.05) among each other's (Table 2). Higher weight gain was attained at lower stocking densities and vice versa, which has the similarity with the findings of Islam *et al.* (1978), Kohinoor *et al.* (2008) and Nahar *et al.* (2015). The findings of this experiment are more or less similar with Hossain *et al.* (2005) who reported the weight gain of 140.60±2.84 g from over-wintered mono-sex tilapia culture for a period of 6 months feeding with a formulated diet. However, the stocking size of fry used by Hossain *et al.* (2005) was lower and culture period was higher than the present study.

Survival rate (%): The average survival rate in this study were found to be 92, 92 and 92.50 % in  $T_1$ ,  $T_2$  and  $T_3$  treatments respectively with no significant difference (p<0.05) among the treatments (Table 2). The findings of the present study were in line with Rahman and Salim, (2013), who found a survival rate of 74.33 to 93.33 % in his experiment. In a trial with formulated feeds (containing 35-45% protein), Kohinoor *et al.* (2008) found a survival rate of 75-89 %. The survival rate found in this study was higher than Dan and Little (2000) findings of 33-54 % for mono-sex overwintering fry. In general, the survival rates in this study are similar to those found by Luo, (2014), who evaluated the growth, enzyme activity, and utility of Nile tilapia grown in a recirculation system and in bio flocs and found 100 % survival rate.

**Specific Growth Rate (SGR):** The values of specific growth rate were found to be ranged from  $1.98\pm0.08$ ,  $2.21\pm0.02$  and  $2.44\pm0.09$  % bwd<sup>-1</sup> in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> treatments, respectively with significant difference (p<0.05) among the treatments (Table 2). The higher value of SGR of  $2.44\pm0.09$  % bwd<sup>-1</sup> was found in the treatment T<sub>3</sub> whereas lowest value ( $1.98\pm0.08$  % bwd<sup>-1</sup>) was found in the treatment. The result of the study strongly agreed with Rahman and Salim, (2013) who found SGR varied from 1.64 to 1.82 % bwd<sup>-1</sup>. Jannat *et al.* (2012) observed SGR varied from 3.14 to 3.21% bwd<sup>-1</sup> from higher to lower stocking density. The present result is much better than the result of those (1.40-1.81 % bwd<sup>-1</sup>) reported by Dan and Little (2000) for mono-sex GIFT tilapia fry.

Average daily growth (ADG): The survival rate in this study were found to be  $0.73\pm0.04$ ,  $0.95\pm0.04$  and  $1.15\pm0.07g$  in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> treatments, respectively with significant difference (p<0.05) among the treatments (Table 2). The lower ADG value ( $0.73\pm0.04g$ ) was found in treatment T<sub>1</sub> whereas the higher value ( $1.15\pm0.07g$ ) was found in treatment T<sub>3</sub>. This may be due to stocking density. Higher ADG was observed at lower fish density while lower ADG was observed at higher fish density. In this present study, average daily growth more or less similar with Crab *et al.* (2009) who found an average daily growth rate of 0.27-0.29 g for the hybrid tilapia in a 50 days of culture periods in bio floc technology in winter season.

Yield (kg/ha/3 months): The biomass of Nile tilapia (Oreochromis

niloticus) culture could produce an equivalent of 155 ton/ha/ crop in an intensive BFT (Avnimelech, 2007). The yield values (kg/ha/3months) were found to be 118907.7±8552.7, 114914.2±2863.9 and 93611.1±4937.6 kg/ha/3 months in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> treatments, respectively with significant difference (p<0.05) where  $T_1$  and  $T_2$  significantly different from  $T_3$  treatment (Table 2). The present result is higher than Nahar et al. (2015) who obtained tilapia production varied from 2882.5±117 to 3802.88±139 kg/ha/6months. The total tilapia production (kg/ha/6 months) reported in this study is identical to that obtained for overwintering tilapia fry by Hossain et al. (2005) and Dan and Little (2000). In this study, higher stocking density resulted higher production, while lower fish density resulted in higher growth. Al-Harbi and Siddigui (2000) reported a similar non-linear relationship between fish growth and fish yield in tilapia cultured in fiberglass tanks. The present study backs up the findings of Khan (2008) and Alim et al. (2005), who found that higher stocking densities provided better results than lower stocking density.

Feed Conversion Ratio (FCR): Findings indicate that food conversion ratio significantly varied from  $1.06\pm0.04$  to  $1.18\pm0.01$ . Lowest FCR was noted in treatment T<sub>1</sub> and highest FCR was noted in treatment T<sub>3</sub> and moderate value of FCR ( $1.14\pm0.02$ ) was noted in treatment T<sub>2</sub> (Table 2). Chakraborty (2005), Rahman and Salim (2013), Torafdar (2013), Islam *et al.* (2019) and Mustafa *et al.* (2010) found the values of FCR about 2.02-2.04, 1.63-2.90,1.7, 2.66-3.00 and 2.46-3.78, respectively while Hossain *et al.* (2005) found the values of FCR of 1.64±0.02 for mono-sex GIFT tilapia fed on formulated diet at over-wintered culture.

Economic analysis: The mean total cost varied from 11960042.2, 11009727.7 and 9464285.7 BDT/ha/3 months in  $T_1$ ,  $T_2$  and  $T_3$  treatments, respectively with significant difference (p<0.05) where  $T_1$  and  $T_2$  significantly different from T3 treatment (Figure 2). The average total return varied from 12185013.0, 12241399.9 and 10297217.8 BDT/ha/3 months in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> treatments, respectively with significant difference (p<0.05) where T1 and T2 significantly different from  $T_3$  treatment. Average net benefit varied from 224971.1, 1231671.9 and 832932.2 BDT/ha/3 months in  $T_1$ ,  $T_2$  and  $T_3$  treatments respectively with significant difference (p<0.05) where T2 significantly different from T1 and T3 treatment (Figure 2). Average highest benefit cost ratio (BCR) varied from 0.02, 0.12 and 0.09 in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> treatments, respectively with significant difference (p<0.05) where  $T_2$  significantly different from  $T_1$ and  $T_3$  treatment (Figure 2). Overall findings indicated that no significant difference was found for the mean values of water quality parameter. Water quality parameters were found within suitable range for all the treatments. Significantly higher yield (p<0.05) was found with treatment  $T_1$  (3.47% higher than treatment T<sub>2</sub> and 27.02% higher than treatment T<sub>3</sub>). Significantly, highest total cost was found with treatment T<sub>1</sub> (8.63% higher than treatment  $T_2$  and 26.37% higher than treatment  $T_3$ ). Significantly, highest total income was found with treatment  $T_2$  (0.46% higher than treatment  $T_1$  and 18.90% higher than treatment  $T_3$ ). Significantly, highest value of net profit was found with treatment T2 (447.48% higher than treatment  $T_1$  and 49.07% higher than treatment T3). Finally, highest CBR was found with treatment  $T_2$  (500% higher than treatment  $T_1$  and 33.34% higher than treatment T3). Khan (2008) and Alim *et al.* (2005) obtained the best production and net profit from higher stocking densities. Rahman and Salim, (2013) reported the highest net benefit of 443458 BDT /ha with a stocking density of 50.000/ha. Nahar *et al.* (2015) cultured the mono-sex GIFT tilapia in bio floc-based farming and reported net profit of 245563.7 BDT/ha/6months.

In spite of having higher yield in treatment T1, economic benefit was found highest in treatment T<sub>2</sub>. This might be due to the lowest final weight obtained with higher stocking density  $(T_1)$ . The final weight in treatment T1 resulted comparatively lower return, net benefit and BCR than T<sub>2</sub>. Assumption almost agreed with Ray et al. (2010) who cultured tilapia and reported that lowest growth, lower final weight and net benefit with higher stocking density. Findings also indicated that comparatively lower BCR was obtained with almost all the treatments. Samad et al. (2010) reported BCR of 1.55 to 2.07 and 1.60-2.03 and Bob -Manuel and Erondu (2010), respectively while culturing tilapia farming in ponds. Lower economical profit in bio floc based tilapia farming in this study might be due to the higher involvement of feed cost (52-59% of the total cost) with the treatments. Assumption almost agreed with Ekasari et al. (2015) who reported higher feed cost (58.34% of total cost) involvement in bio floc -based tilapia farming. Assumption agreed with Emerenciano et al. (2017) who recommended less use of feed in bio floc-based fish farming system since, fishes are fed with floc. Therefore, further research is necessary to decrease the feed cost and thereby to promote the low cost bio floc based fish farming in tanks.

#### Conclusion

Aquaculture need to be developed in a suitable and sustainable way to protect the environment and the natural resources. In this context, bio floc technology (BFT) has been an alternative to achieve sustainability in the production of aquatic organisms by improving water quality through maintaining carbon and nitrogen in the system. The results of the present study suggest that Nile tilapia culture can be performed in bio floc system; however, water quality parameters were slightly affected by higher stocking density, mainly in T<sub>1</sub> treatment with 200 nos/m<sup>3</sup> stocking densities. Besides, high stocking density resulted in a low growth rate, reducing the profitability of BFT if worse selling prices for lighter fish are considered. Among the three different stocking densities, 150 nos/m<sup>3</sup> seems to bring better results in animal and economic performance for the production system, regardless of selling prices.

# Author's contribution

Conceptualization: MHK; Methodology, MHK; Software, MHK; Validation, MHK and MAH; Formal analysis, MHK; Investigation, MHK and MAH; Resources, MHK; Data curation, MHK; Writing—original draft preparation, MHM; Writing—review and editing, MHM and MHK; Visualization, SKD and MAH; Supervision, MAH; Project administration, MHK and MAH. All authors have read and agreed to the published version of the manuscript

Conflict of interest: The authors declare no conflict of interest.

Ethical approval: Not applicable.

**Data availability:** The data that support the findings of this study are available on request from the corresponding author.

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