

**BIOFLOC CULTURE METHOD FOR GROWTH  
PERFORMANCE, SURVIVAL RATE AND NUTRITIONAL  
COMPOSITION OF SNAKEHEAD FISH (*Channa striata*) ON  
NATURAL EXTRACT SUPPLEMENT**

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**CERTIFICATE**

*This is to certify that the thesis entitled, “BIOFLOC CULTURE METHOD FOR GROWTH PERFORMANCE, SURVIVAL RATE AND NUTRITIONAL COMPOSITION OF SNAKEHEAD FISH (C. striata) ON NATURAL EXTRACT SUPPLEMENT” submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE (MS) IN BIOTECHNOLOGY, embodies the results of a piece of bona fide research work carried out by MD. KHAIRUL ALAM, Registration No. 19-10077 under my supervision and guidance. No part of this thesis has been submitted for any other degree or diploma.*

*I further certify that such help or source of information as has been availed of during the course of this investigation has duly been acknowledged.*

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# **DEDICATION**

**Dedicated to  
My Beloved Parents and Brother**

## List of abbreviations

Full word	Abbreviation
Percentage	%
Meter	m
Centimeter	cm
Degree Celsius	°C
And others	<i>et al.</i>
Gram	g
Potential of hydrogen	pH
Kilogram	kg
Liter	L
Milligram	mg
Milliliter	ml
Square meter	m <sup>2</sup>
Cube meter	m <sup>3</sup>
Standard deviation	SD
Standard error	SE
Gross production	GP
Net production	NP
Horse power	HP
Parts per meter	ppm
Feed conversion ratio	FCR
Food conversion efficiency	FCE
Dissolved oxygen	DO
Milligram per liter	mg/L
Gram per kilogram	g/Kg
Per kilogram	Kg <sup>-1</sup>

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# **Biofloc Culture Method for Growth Performance, Survival Rate and Nutritional Composition of Snakehead Fish (*Channa striata*) on Natural Extract Supplement**

## **ABSTRACT**

Snakehead fish (*Channa striata*) is a well known and important freshwater fish in Southeast Asian countries. The experiment was conducted at Sher-e-Bangla Agricultural University Biofloc Culture Lab to evaluate the growth performances, survival rate and nutritional composition of snakehead fish (*C. striata*). There has four treatments with three replications. Water quality parameters were monitored through temperature, dissolved oxygen, pH, ammonia, total dissolved solids, alkalinity and hardness. Maximum length increase ( $14.15 \pm 0.2$ ) cm of *C. striata* was found in treatment-4. Highest weight gain also noticed in the same treatment which was ( $105.67 \pm 6.65$ )g. Control treatment showed lowest performance among the parameter studied. Maximum specific growth rate was 2.70% in treatment-4 whereas 2.50% was minimum in treatment-1. In treatment-3 survival rate was highest, which was 75.76% and in control treatment, the lowest survival rate was recorded that was 47.88%. Feed conversion ratio was better (1.08) in treatment-4. Daily mean weight gain was noteworthy in treatment-4 which was 1.50g and lowest daily mean weight gain was recorded 1.1g in control treatment. From the analysis of proximate composition of *C. striata* 69.67% protein content was available in treatment-4 which was maximum and the lowest protein content was found 61.84% in treatment-2. Considering all the results, the optimum growth performance and nutritional composition was better in treatment-4 (Extract of Garlic and Amla) compared to other treatments. In the era of aquaculture and microbial biotechnology there has a huge potentiality to fulfill the requirement of fish protein by the culture of *C. striata* in biofloc technology.

## CHAPTER 1

### INTRODUCTION

Biofloc technology (BFT) offers benefits in improving aquaculture production that could contribute to the achievement of sustainable development goals in the area of fish biotechnology. It is based on mainly the principle of waste nutrients recycling, in particular nitrogen, into microbial biomass that can be used *in situ* by the cultured animals or can be harvested and processed into feed ingredients (Avnimelech, 2009; Kuhn *et al.* 2009). Heterotrophic microbiota is stimulated to grow by steering the (C/N) carbon nitrogen ratio in the water through the modification of the carbohydrate content in the feed or by the addition of an outer carbon source in the water (Avnimelech *et al.* 1999), so that the microbes can acclimatize the waste ammonium for new biomass production. Biofloc systems also help to prevent the introduction of disease to a farm from incoming water. Reducing water exchange is an undeniable strategy for developing farm biosecurity. Microbial biotechnology is used to assimilate the waste ammonium for new biomass production. Hence, ammonium/ammonia can be maintained at a low and non-toxic concentration by bacterial genotypic metabolism. It contributes to the improvement of the fish production. In relation to the farmer, biofloc technology could support the supply of good quality seeds by improving the reproductive performance of aquaculture animals and by enhancing the larval immunity and robustness (Ekasari *et al.* 2015; Ekasari *et al.* 2016 and Emerenciano *et al.* 2013). The microbial biotechnology is a platform for microbes and fish production in relation with biofloc mechanism.

Biofloc technology is one kind of microbial biotechnology which can be utilize for fish improvement program. This technology deals with microorganisms to ensure the security, safety, and usefulness of foods, yielding high-quality products, proper human nutrition and defense against plant and animal diseases. Nevertheless, uses of microbial biotechnology for sustainable development in aquaculture are critical because of the essential role of microorganisms in the establishment and control of ecosystem facilities, especially nutrient cycling, water quality control and disease regulation in the culture system (Timmis *et al.* 2017). Several microbial biotechnologies have been applied or are still in the developmental pipeline to increase the productivity in aquaculture by creating the



ecofriendly environment to support the growth of other aquatic organisms. These organisms are key agents of pollutant removal and recycling (Bossier and Ekasari 2017; Kabir and Aba 2019; Liu *et al.* 2019). With over seven billion people on world, the requirements for aquatic food which is increasing in such a manner that the expansion and intensification of this kind of technology with aquaculture production are highly required. Biofloc technology (BFT) is one of such novel microbial biotechnologies that has been developed with an excellent ecofriendly technology not only for higher productivity but also for sustainable development of fish production (Emerenciano *et al.* 2017; Abakari *et al.* 2020a). Flocs (aggregates) are suspended growth in ponds consists of phytoplankton, algae, bacteria, protozoans and other kinds of particulate organic matter such as feces and uneaten feed. Typical flocs are irregular by shape, have a broad distribution of particle size (1-2 mm) are fine, easily compressible, highly porous (up to more than 99% porosity) and are permeable to fluids.

Snakehead fish (*Channa striata*) is an important freshwater fish species in numerous Southeast Asian countries. *Channa striatus* locally known as “Shoal” is a commercially important fish species of the genus *Channa* and family Channidae. It contributes 4.2% of the total fish production in Bangladesh (FRSS 2008). The flesh of Shoal fish is firm, white and has the most agreeable flavor. It is one of the main food fishes in Bangladesh, China and Malaysia and cultivated in India, Pakistan and Thailand (Muntaziana 2013). The heavy dark skin is usually sold separately which is good for soup (Davison 1975). Its flesh is claimed to be rejuvenating and widely consumed for its nutritional value as well as for its beneficial effect in wound healing (Mat *et al.*, 1994, Wee 1982). Snakeheads are a peculiar group of freshwater fishes having accessory respiratory organ to utilize atmospheric air for respiration that enables them to thrive in oxygen depleted waters (Kumar *et al.*, 2012 and 2013). Snakehead can survive in adverse environments with low dissolved oxygen and high ammonia (Ng and Lim, 1990; Mollah *et al.*, 2009 and Qin *et al.*, 1997a). This fish is known for its taste, high nutrient and pharmaceutical values (Khanna, 1978).

For the cultivation of snakehead fish in biofloc system there has several benefits that influences the culture system than the traditional culture system. This culture system requires limited or zero water exchange. Higher productivity and higher biosecurity is

ensured in this culture system. By applying this kind of technology for snakehead fish culture it reduces water pollution and the danger of introduction and spread of pathogens. Cost-effective feed production is an important aspect for the production of snakehead fish in alternate culture system. Cultivation of snakehead fish in biofloc method is new in our country. Very limited information are available in this regard. Hence, production potentiality and growth performance need to study under Bangladesh condition. By cultivating snakehead fish in biofloc culture system it can be improved its nutritional quality and growth performance. By considering different benefits of biofloc culture system an attempt was made to culture the snakehead fish (*C. striata*) for study the survival rate, growth performance and other nutritional parameters in this technology. Hence, the overall objectives of the research work is given in below:

**Objectives:**

- To assess the growth performance of *Channa striata* in biofloc system using garlic and amla extract as a food ingredient
- To assess the survival rate of *C. striata* in biofloc culture method
- To evaluate the nutritional composition of *C. striata* by feeding garlic and amla extract under this culture system

## CHAPTER 2

### REVIEW OF LITERATURE

A review of literature related to the subject is an important and integral part of any research study. A critical survey of the literature on the subject will help in preparing the objectives and methodology. A review of writing along these lines helps in recognizing research holes in the subject and the requirement for the present investigation. In this part an exertion has been made to fundamentally review the writing of past research thinks about important to the examination and is introduced as follows:

Sangeeta *et al.* (2017) conducted an experiment on effect of feeding rate on growth, survival and cannibalism in striped snakehead, *C. striata* (bloch, 1793) fingerlings. A 45 days test was led to survey the growth, survival and cannibalism. In this examination, 450 striped murrel fingerlings of normal weight  $1.41 \pm 0.12$ g were arbitrarily appropriated in 15 tanks of 250L water limit. Fishes were taken care of pelleted diet having 44.1% protein with various taking care of rate 2% (T1), 4% (T2), 6% (T3), 8% (T4) and 10% (T5) of fish biomass. The weight pick up, rate gauge gain and explicit growth rate was altogether higher in T3 and T4 in contrast with T1 and T2 gatherings. The cannibalism rate was altogether higher in T1 (2%) bunch in contrast with T3, T4 and T5. Cannibalism rate was progressively decreased with increment in taking care of rate and altogether higher cannibalism ( $12.22 \pm 1.94$ ) was seen at 2% of taking care of rate (T1) in contrast with the other treatment gatherings. This examination showed that taking care of rate at 6% of biomass of striped snakehead fingerlings is ideal for better development and endurance and to limit the cannibalism. Water parameters such as: highest temperature range was (23.4-25.1 °C) in T2 and lowest range was (22.1-24.3 °C) in T1, pH was (7.8-8.5) in T5 which was highest and lowest range was (7.1-8.2) in T1, DO range was highest (6.8-7.7mg/L) in T1 and lowest range was (6.2-7.3mg/L) in T5, alkalinity range was (159-168mg/L) in T5, which was highest and (149-157mg/L) in T2, which was lowest. Proximate composition in the experimental feed (moisture 4.20%, protein 44.10%, ash 14.70% carbohydrate 26.90%). Weight gain rate was higher (1.38) in T4 and lowest rate was (0.54) in T1. Highest SGR was (1.49%) in T4 and lowest (0.71%) in T1. Percentage weight gain was

(96.26%) which was highest and lowest was (37.46%). Feed conversion ratio (FCR) was highest (3.11) in T5 and lowest (2.10) in T1. Highest food efficiency ratio (FER) was (0.48) in T1 and lowest was (0.32) in T5.

Puspaningsih *et al.* (2019) conducted a study on water quality, hematological parameters and biological performances of Snakehead fish (*C. striata*) reared in different stocking densities in a recirculating aquaculture system. Snakehead fish (*C. striata*) has been an exceptionally mainstream and significant freshwater fish species in numerous Southeast Asian nations. The reason for this examination was to assess the water quality, hematological boundaries and development exhibitions of snakehead raised at various stocking densities, in a recycling aquaculture framework. The test configuration utilized a totally randomized plan with three distinctive stocking densities as medicines: A - 2 fish L-1; B - 4 fish L-1; C - 6 fish L-1. Each treatment comprised of three replications. Snakehead fingerlings introduced a normal length of  $6.07 \pm 0.10$  cm and a normal weight of  $1.82 \pm 0.07$  g. The water quality boundaries were examined in the experiments are dissolved oxygen, pH, temperature, alkali nitrogen, nitrites, nitrates and orthophosphate. The growth exhibitions noticed were the survival rate, explicit development rate, food transformation proportion, total length, outright weight and biomass. Fish were refined in a recycling hydroponics framework (RAS), a model inherent the Research Institute for Freshwater Aquaculture and Fishery Extension, Bogor, Indonesia. The outcomes indicated that water quality boundaries were inside the ideal reach for snakehead culture. The highest survival rate was 84% in T2 and lowest was 60.67% in T3. Specific growth rate was highest (2.80%) in T2 and lowest was (2.15) in T3, FCR rate was highest (1.12) in T3 and lowest (0.80) in T2. Weight gain rate was (5.97) in T2 which was highest and lowest was (3.57) in T3, highest length increase was (3.65) in T2 and lowest was (2.97) in T3.

Saputra *et al.* (2018) conducted a study on growth performance and survival of snakehead (*C. striata*) juvenile with different stocking density reared in recirculation system. Snakehead (*C. striata*) is a nearby explicit fish animal groups and has high financial worth. The primary imperative in snakehead fish cultivating is high mortality on snakehead juvenile raising stage. This examination was led to decide the best stocking thickness on snakehead juvenile raising to accomplish ideal creation. Snakehead juveniles with a body

length of  $3.41 \pm 0.39$  cm and weight  $0.28 \pm 0.07$  g were stocked and raised for 42 days in the aquarium estimated  $40 \times 40 \times 40$  cm with a volume of 40 L. The outcome appeared that the medicines didn't influence the survival, development, and the proportion of RNA/DNA of snakehead juvenile. Survival rate of juvenile snakehead went from 92.5–94.58%. The effect of water quality estimation appeared that it was on ideal condition to help snakehead development at 3 juveniles/L stocking thickness. Moreover, distribution can be utilized to keep up water quality for ideal condition. The water temperature was kept up around (29–30°C), and the pH parameter was kept up around (5.25–6.31). The dissolved oxygen was ranged from (2.94–6.14 mg/L). The alkalinity was very high, it was from 86.43– 101.84 mg/L.

Farhana *et al.* (2016) conducted a study on commercially culture potentiality of striped snakehead fish (*C. striatus*) (Bloch, 1793) in earthen ponds of Bangladesh. An investigation was led to assess the business culture possibility of striped snakehead *C. striatus* fish in the earthen lakes in Jessore, Bangladesh. Monoculture was completed in three earthen lakes of Afil Aqua Fish Ltd. by gathering normally accessible fingerlings with a length and weight of  $11.75 \pm 0.75$  cm and  $45 \pm 10$  g, individually for a time of 180 days. Stocking density of fingerlings was 40 fish/decimal and were taken care of with incubation center began live fish fry of Bata (*Labeo bata*), Mrigal (*Cirrhinus cirrhosus*) and Silver carp (*Hypophthalmichthys molitrix*) at the rate of 1 to 3% of the total body weight and a beneficial feed mix of rice polish, mustered oil cake and fish meat at the rate of 3 to 5% of the total body weight. In situ water quality boundaries viz. water temperature, dissolved oxygen, pH were acquired from ( $26.5 \pm 5$  to  $31.5 \pm 1.5^\circ\text{C}$ ), ( $6.75 \pm 0.25$  to  $8.2 \pm 0.1$  mg/L) and ( $7.8 \pm 0.1$  to  $8.5 \pm 0.1$ ) respectively. The final weight of fish was  $850 \pm 60$  g and net production was  $32.98 \pm 0.5$  kg/decimal. The total production of remaining unutilized white fish Bata (*Labeo bata*), Mrigal (*Chirhinus cerosus*) was  $500 \pm 0.5$  kg. The survival rate and feed conversion ratio (FCR) was  $97 \pm 2\%$  and 1.56. The specific growth rates (SGR %/day) were ( $14.65 \pm 1.55$ ), ( $7.85 \pm 0.95$ ), ( $11.1 \pm 2.1$ ), ( $7.77 \pm 0.8$ ), ( $6 \pm 0.6$ ), ( $3.4 \pm 0.54$ ) and ( $4.45 \pm 0.35$ )%. Benefit Cost Ratio (BCR) was 1.85 and found essentially higher than Thai koi (*Anabus testudineus*) and Tilapia (*Oreochromis mossambicus*) from similar area and showed its way of life and monetary achievability.

War *et al.* (2011) conducted a study on growth and survival of larval snakehead *C. striata* (Bloch, 1793) fed different live feed organisms. One of the significant cultivable native finfish which merits quick consideration for business scale seed creation and cultivating is the snakehead, *C. striata*. Arrangement of appropriate live feed is the bottleneck in raising hatchlings of this fish. Culture execution of larval snakehead was inspected in the current examination by taking care of them with cladocerans (*Ceriodaphnia cornuta*, *Moina micrura* and *Daphnia carinata*) and *Artemia nauplii* as individual and blended cladoceran diet (*C. cornuta*, *M. micrura* and *D. carinata*) for about a month. Fish fed of *Artemia nauplii*, *C. cornuta* and blended cladocerans appeared higher weight gain ( $15.88 \pm 0.11$  mg), ( $9.72 \pm 0.04$  mg) and ( $10.0 \pm 0.06$  mg) separately during the 1st week. Fish fed of *C. cornuta*, *M. micrura* demonstrated better weight gain ( $12.88 \pm 0.21$  mg) and ( $11.90 \pm 0.09$  mg) individually during the second week. Fish took care of on blended cladocerans demonstrated better survival and development with less cannibalism over the most recent three weeks. Fish took care of *Artemia nauplii* demonstrated not so much development but rather more cannibalism during the most recent fourteen days. Use of cladocerans for early larval raising of *C. striata* will decrease the use for seed production. It is likewise seen that with the expansion in age and growth, the fish inclines toward enormous estimated prey than the more modest ones. Cannibalism can be reduced at different stages by providing prey of suitable size to the growing fish.

López-Elías *et al.* (2015) conducted a study on proximate composition of bioflocs in culture systems containing hybrid red tilapia fed diets with varying levels of vegetable meal inclusion. Biofloc culture systems, which depend on the advancement of microorganisms that reuse inorganic supplements and natural issue, may add to the nourishment of some cultivated species. Juvenile red tilapia refined in saltwater were taken care of pelleted diets in which 0, 33, 67, or 100% of the fish meal was substituted with a vegetable meal mix (corn, wheat, and sorghum meals). Mean volume of biofloc was 9.22 ml/L for T0, 8.67 ml/L for T10, 6.78 ml/L for T20, and 4.22 ml/L for T30, whereas the mean for the control was 5.56 ml/L. The proximate composition of the biofloc produced in the culture systems was assessed. Four experimental diets and one control diet were arbitrarily allocate to 15 test tanks. Tests of biofloc were periodically gathered to measure the total suspended solids, organic matter, and ash content and to decide the protein, lipid, and starch substance.

Toward the finish of the investigation, variables describing red tilapia production were determined. The biofloc volume, complete suspended solids, ash, and organic matter demonstrated critical contrasts among treatments, however carbohydrate (33.0–39.0%), ash (33.0-40.4%), lipid (2.6–3.5%), and protein content level was (23.7–25.4%). No huge contrasts were seen in red tilapia survival, final biomass, or feed conversion ratio.

Ekasari *et al.* (2018) conducted a study on utilization of Snakehead fish (*C. striata*) extraction by product into fish protein concentrate using acid and alkali solubilization methods. The extraction of Snakehead fish (*C. striata*) flesh for functional food produced a huge amount by product that can be utilized into fish protein concentrate (FPC). The aimed of the study is to research the nutritional value of FPC produced from snakehead fish extraction by product using acid and alkali solubilization techniques. Three arrangements of pH varieties were used for each treatment i.e 2,3,4 (acid) and 10,11,12 (base). The parameters being analyzed were yield, water content, ash content, fat content and protein contents. The results indicated that the best was acquired from treatment of pH 4 with 23.6%. The most noteworthy protein content was obtained from treatment using pH 3, with 92.03%. The best moisture content was obtained from pH 12, with 10.22%, ash content from pH 11, with 1.80%, fat content from pH 4, with 1.61%.

Muntaziana *et al.* (2013) was conducted a study on effect of selected diets on the growth and survival rate of snakehead fish fry. Twenty five days feeding experiment was conducted to assess the growth and survival rate *Channa striata* fry fed with three types of fresh foods (bloodworm, trash fish, *Acetes* shrimps). Ten fry were stocked in each aquarium. The fry fed trash fish showed a higher weight gain percentage (376.50) than those fed with *Acetes* shrimp (233.05) and bloodworm (199.08). The final average total lengths of fry fed with trash fish, bloodworm and *Acetes* shrimps were  $7.91 \pm 0.23$ ,  $7.28 \pm 0.23$ ,  $7.21 \pm 0.17$  cm respectively. The best specific growth rate (SGR) value was ( $6.24 \pm 0.17\%$ ) by fed with trash fish. The best Feed Conversion Ratio (FCR) was ( $3.63 \pm 0.27$ ) found by fed trash fish. The best dissolved oxygen (DO) level was ( $3.78 \pm 0.16$ ), pH was ( $6.52 \pm 0.02$ ), Temperature was ( $29.76 \pm 0.22$ ). The moisture% was 74.22, protein% was 78.64, lipid% was 8.26, fiber% was 2.07 and ash% content 6.78 was the best.

Yusuf *et al.* (2015) conducted a research on growth performance of catfish (*Clarias gariepinus*) in biofloc-based super intensive culture added with *Bacillus sp.* Biofloc cultured in isolated aquarium (100L) was used as source of inocula for cultivation for *C. gariepinus*. Different concentration of *Bacillus sp.*, cells was included in diets. Result showed that the survival rate of treatment C was  $89.33 \pm 2.4\%$  which was higher but not significant different compare to treatment A ( $87.33 \pm 6.59\%$ ) and B ( $86.67 \pm 9.42\%$ ). Highest SGR was found in treatment C ( $6.35 \pm 0.05\%$ ), which was not significantly different to treatment A ( $6.04 \pm 0.09\%$ ) and B ( $6.16 \pm 0.11\%$ ). Highest FCR was found in treatment C ( $0.91 \pm 0.01$ ). Protein content of biofloc from treatment C was ( $34.06 \pm 1.70$ ), A and B were ( $31.99 \pm 2.70$ ) and ( $31.81 \pm 1.51$ ) respectively. Lipid content obtained from K+ ( $14.92 \pm 2.87$ ), followed by C ( $9.17 \pm 4.53$ ), A ( $7.87 \pm 1.69$ ) and B ( $6.47 \pm 0.86$ ). The highest DO range was 5.9-7.6 mg/L, pH range was 5.74-7.95, temperature was 30.5-32.6°C and ammonia range was in between 0.26-0.27 mg/L.

Djokosetiyanto *et al.* (2017) conducted a research on survival and growth rate of striped snakehead fish (*C. striata* Bloch.) juvenile reared in acid sulfate water and rainwater medium. The point of this examination was to assess the impacts of the utilization of the various mediums on the development and endurance of the striped snakehead fish (*Channa striata*). This investigation applied a totally randomized plan with acid sulfate water medium and rainwater medium as the experimental treatments, every treatment had twelve replications. The *C. striata* juvenile with an average initial length of  $2.4 \pm 0.2$  cm and an average initial weight of  $0.21 \pm 0.05$  g raised in the aquariums measuring 30 x 25 x 35 cm (with a water volume of 25 L) with a stocking thickness of 2 people/L, for 40 days. The specimens were fed commercial feed, two times in a day (morning and evening) until to clear satiation. The water substitution was played out as much as 10% water of the all out water volume in the aquarium. The outcomes demonstrated that water essentially influenced the biometric and physiological reactions of the *C. striata* juvenile. The rainwater medium gave better outcomes appeared by the higher survival rate (73.89%) and lowest survival rate was (54.44%) in acid sulfate water, growth rate (4.40% day<sup>-1</sup>), feed efficiency (59.1%), protein intake (24.31%), energy intake (41.34%). The water temperature in the acid sulfate water medium gone from (28.42 to 30.02°C), while that in the rainwater medium went from (28.96 to 30.07°C). The dissolved oxygen level in the



acid sulfate water medium went from (5.53 to 6.10 mg/L), while that of the rainwater medium went from (5.68 to 6.25 mg/L). In the treatment the best range of pH during the investigation was 6.58-8.30 in the rainwater medium.

Putra *et al.* (2019) conducted a study on growth and survival rate of red tilapia (*Oreochromis Sp.*) cultivated in the brackish water tank under biofloc system. This investigation is expected to assess the growth and survival rate of red tilapia (*Oreochromis sp.*) developed in a biofloc framework brackish water tanks with the expansion of various carbon sources. The exploratory treatment was addition of various carbon sources to the media, in particular molasses (B), custard flour (C), white sugar (D) and without carbon sources or controls (A). Test tilapia utilized were sized  $3.71 \pm 0.11$  cm with a thickness of 300 fish/m<sup>3</sup> kept up for 40 days. Fish kept in tanks with a volume of 200 liters loaded up with sea water as much as 100: 1 with a saltness of 17 ppt, added with 10 ml/m<sup>3</sup> probiotics and carbon arrangement with a C/N proportion of 20: 1. The fish were taken care of with commercial feed (38% protein content), 3 times each day as much as 5%/body weight/day. The expansion of probiotics and carbon is done at regular intervals. The absolute weight growth (g) of treatment A, B, C and D was 2.72, 8.22, 5.18, 5.88 g respectively. Daily growth rate (%) was 3.49, 7.86, 4.80, 5.20% for treatment A, B, C and D respectively. Survival rate was 61.11% for treatment A and 92.22%, 82.22%, 84.4% for treatment B, C, D respectively. The highest feed efficiency was 119.39% and feed conversion ratio was 1.45, 0.85, 1.05, 1.08 for treatment A, B, C and D respectively.

Anand *et al.* (2018) conducted a study on effect of biofloc on water quality parameters in rohu, *Labeo rohita* (Hamilton) culture tanks. The present experiment was undertaken to investigate the effect of biofloc-based aquaculture system on water quality suitability for the growth of rohu for a period of 120 days under laboratory conditions in sets of three. Various water quality parameters were checked at weekly interval. Sugar has been given as the source of carbon and ammonium chloride as source of ammonia. Dissolved oxygen level ranged from (5.5 to 8.0 ppm) in the T1, in T2 it was (6.5 to 8.0 ppm) and in T3, (6.0 to 8.0 ppm). Ammonia ranged from (0.0 to 0.43) in T1, (0.0 to 0.5) T2 and in T3 it was (0.0 to 0.75 ppm). Alkalinity ranged from (159 to 190), (147 to 189) and (158 to 188) ppm in T1, T2 and T3 tanks respectively.

Ekasari *et al.* (2012) conducted a study on evaluation of biofloc technology application on water quality and production performance of red tilapia *Oreochromis sp.* cultured at different stocking densities. This study evaluated the impact of biofloc technology (BFT) application on water quality and production performance of red tilapia *Oreochromis sp.* at various stocking densities. Three distinctive stocking densities were applied, i.e. 25, 50, and 100 fish/ m<sup>3</sup>, and for each density there were control (without external carbon input) and BFT treatments. Mixed sex red tilapia with an initial average body weight  $77.89 \pm 3.71$ g was cultured in three m<sup>3</sup> concrete tanks for 14 weeks. Control treatments of each density tested showed more fluctuated water quality parameters all through the trial period. The most elevated total yield was observed in control treatment at the highest density treatment (43.50 kg), though the most noteworthy survival was obtained by BFT treatment at a density of 25 fish/m<sup>3</sup> ( $97.78 \pm 0.77\%$ ). Total feed used in BFT was lower than that of control treatments specifically at 50 fish/m<sup>3</sup> density. The final mean weight of BFT was 160.31g and in control treatment final mean weight was 199.16g. The final mean weight gain was 82.90g in BFT and 121.27g in control treatment. The daily mean weight gain was 0.84g in BFT and 1.22g was in control treatments. Average feed efficiency was 56.43% in BFT and 50.03% in control treatments. Survival rate in BFT was 94.78% and in control treatments it was 91%.

Sugumaran *et al.* (2018) conducted a study on growth performance, length-weight relationship of snakehead fish, *C. striata* (Bloch) fed with different diets. A ninety day feeding trial was led to study the impacts of different diets on the growth, specific growth rate of murrel *C. striata*. The temperature ranged at 28°C in all three tanks, pH ranged from (8.1 to 8.2), dissolved oxygen ranged from (6.20 to 6.87), total alkalinity ranged from (14.9 to 15.84), ammonia ranged from (0.320 to 0.340), hardness ranged from (80.01 to 81.15). The proximate analysis of diets were, the crude protein level ranged from 34.65 to 36.80, fat content ranged from 1.68 to 2.03, fiber level ranged from 8.36 to 8.49 and ash content ranged from 5.48 to 6.13. Initial average length 10.69 (cm) and final average length was 23.32 (cm), Initial average weight was 11.02 (g) and final average weight was 23.92 (g). Specific growth rate was 0.413%.

Paray *et al.* (2015) conducted a study on impact of different feeds on growth, survival and feed conversion in striped snakehead *C. striata* (Bloch 1793) larvae. Growth, survival and feed conversion ratio of striped snakehead (*C. striata*) larvae fed on different feeds during nursery rearing were assessed. Three distinct experiments were conducted using hatchlings, fry and fingerlings of *C. striata* with three replicates per treatment. Growth performance of snakehead hatchlings was analyzed by feeding them with the following diets: Diet 1 - copepod, *Thermocyclops decipiens*; Diet 2 - cladoceran, *Ceriodaphnia cornuta* and Diet 3 - *T. decipiens* and *C. carnuta* in combination, for a period of four weeks. Feeding experiment of 45 days duration conducted by feeding animal wastes (D1: chicken liver, D2: fish waste and D3 : combination of chicken liver and fish waste) to evaluate the growth and survival of *C. striata* fry showed significantly high survival rate in *C. striata* fry fed diet D3 compared to those fed D1 and D2. Among the 3 types of live feed diets, the highest increase in total length ( $30.46 \pm 0.01$  mm) and weight gain (WG) ( $231.18 \pm 0.03$  mg) was recorded in fishes fed with Diet 3 which was significantly different from Diet 1 ( $26.66 \pm 0.14$  mm and  $171.23 \pm 0.01$  mg) and Diet 2 ( $22.24 \pm 0.006$  mm and  $183.22 \pm 0.05$  mg). Diet 3 showed higher SGR ( $20.03 \pm 0.01\%$  WG per day) than Diet 2 ( $19.18 \pm 0.03\%$  WG per day). Growth rates (specific and mean) of fish in Diet 1 were  $19.42 \pm 0.03\%$  WG per day and  $70.81 \pm 1.12$  mg/g/d respectively. *C. striata* fry fed on animal byproducts over a period of 45 days are summarized as WG ( $3.82 \pm 0.04$  g) seen in D3 was significantly higher than D1 ( $3.41 \pm 0.04$  g) and D2 ( $3.22 \pm 0.02$  g). SGR was highest ( $4.78 \pm 0.09\%$  WG/d) in the fish fed with D3 followed by those fed with D1 ( $4.61 \pm 0.03\%$  WG/d) and D2 ( $4.38 \pm 0.04\%$  WG/d). MGR (mg/g/d) was better ( $35.21 \pm 0.02$ ) in D3 followed by D1 ( $34.52 \pm 0.02$ ) and D2 ( $33.59 \pm 0.01$ ) respectively. Better survival rate ( $81.34 \pm 5.54\%$ ) was noticed in D3, which was significantly different from D1 ( $76.33 \pm 4.72\%$ ) and D2 ( $76.12 \pm 2.62\%$ ).

War *et al.* (2014) conducted a research on preliminary studies on the effect of prey length on growth, survival and cannibalism of larval snakehead, *C. striata* (Bloch, 1793). Larval snakehead *C. striata* were fed with three cladocerans (*Ceriodaphnia cornuta*, *Moina micrura* and *Daphnia carinata*) and *Artemia nauplii* to record growth, survival and cannibalism. Experiments were carried out for 30 days in 40L tanks each loaded with 30L water. Fish fed with *C. cornuta* and *M. micrura* showed better length increase ( $27.72$ mm and  $26.50$ mm) respectively after 30 days. Fish fed with *D. carinata* and *M. micruna*

showed highest weight gain (15.24mg and 13.56mg) respectively after 30 days. Specific growth rate after 30 days was better by fed *D. carinata* and *M. micruna* as (30.82% and 25.20%) respectively. Mortality rate was less (12%) by fed *A. nauplii* and highest as (31.66%) by fed with *D. carinata*. High cannibalism rate was 16.32% in the whole experimental period. The survival rate was highest as 88% by fed with *A. nauplii* and lowest survival rate was 71.66% by fed with *D. carinata*.

Emerenciano *et al.* (2017) conducted a research on biofloc technology (BFT): a tool for water quality management in aquaculture. Biofloc technology (BFT) is considered the new “blue revolution” in aquaculture. Such strategy depends on in situ microorganism production which plays three major roles: (i) maintenance of water quality, by the uptake of nitrogen compounds generating *in situ* microbial protein; (ii) nutrition, enhancing culture feasibility by reducing feed conversion ratio (FCR) and a decrease of feed costs; and (iii) competition with pathogens. The aggregates (bioflocs) are a rich protein-lipid natural source of food available in situ 24 hours per day due to a complex communication between organic matter, physical substrate, and large range of microorganisms. This natural productivity plays a vital role recycling nutrients and maintaining the water quality. The ideal observed water quality parameters are- temperature ranged from (28–30°C), pH ranged from (6.8-8.0), DO level above of 4.0 mg/L is appropriate. Alkalinity more than 100 mg/L is ideal and total dissolved solids level is ideal when it less than 500 mg/L.

Ghaedi *et al.* (2019) conducted an investigation on effect of different protein levels on reproductive performance of snakehead murrel *C. striata* (Bloch 1793). In this investigation the effect of different protein levels on reproductive performance of *C. striata* was conducted. Snakehead juveniles (65.5±0.2 g) were haphazardly distributed into nine homogenous groups of 75 fish each. Three isocaloric diets differing in protein levels were prepared. The research lasted for 8 months and gonadosomatic index, absolute fecundity, egg diameter, number of mature oocytes, hatching rate, larval length, survival rate and amino acid and proximate composition of tissue, liver and ovary were observed. Growth, gonadosomatic index (GSI) and absolute fecundity increased with increment in protein

level. Protein and lipid substance of ovary was highest in fish fed 450 g/kg protein. The weight of fish was discovered significantly higher in fish fed the diet of 450 g/kg during all sampling, highest weight gain was recorded in fish fed protein at 450 g/kg diet ( $289.7 \pm 5.5$  g), trailed by fish fed protein at 400 g kg<sup>-1</sup> diet ( $250.3 \pm 4.0$  g) and at 350 g/kg diet ( $230.3 \pm 3.5$  g) at the end of the experiment.

## CHAPTER 3

### MATERIALS AND METHODS

#### 3.1 Study Area

The study was performed in the recently established "Biofloc Lab" under the Department of Aquaculture, Faculty of Fisheries, Aquaculture and Marine Science at Sher-e-Bangla Agricultural University. Freshwater snakehead fish (*Channa striata*), locally named as “shoal” was chosen as culture species in biofloc culture method for the first time in Bangladesh to assessing its culture suitability and evaluating its survival, growth and other parameters.

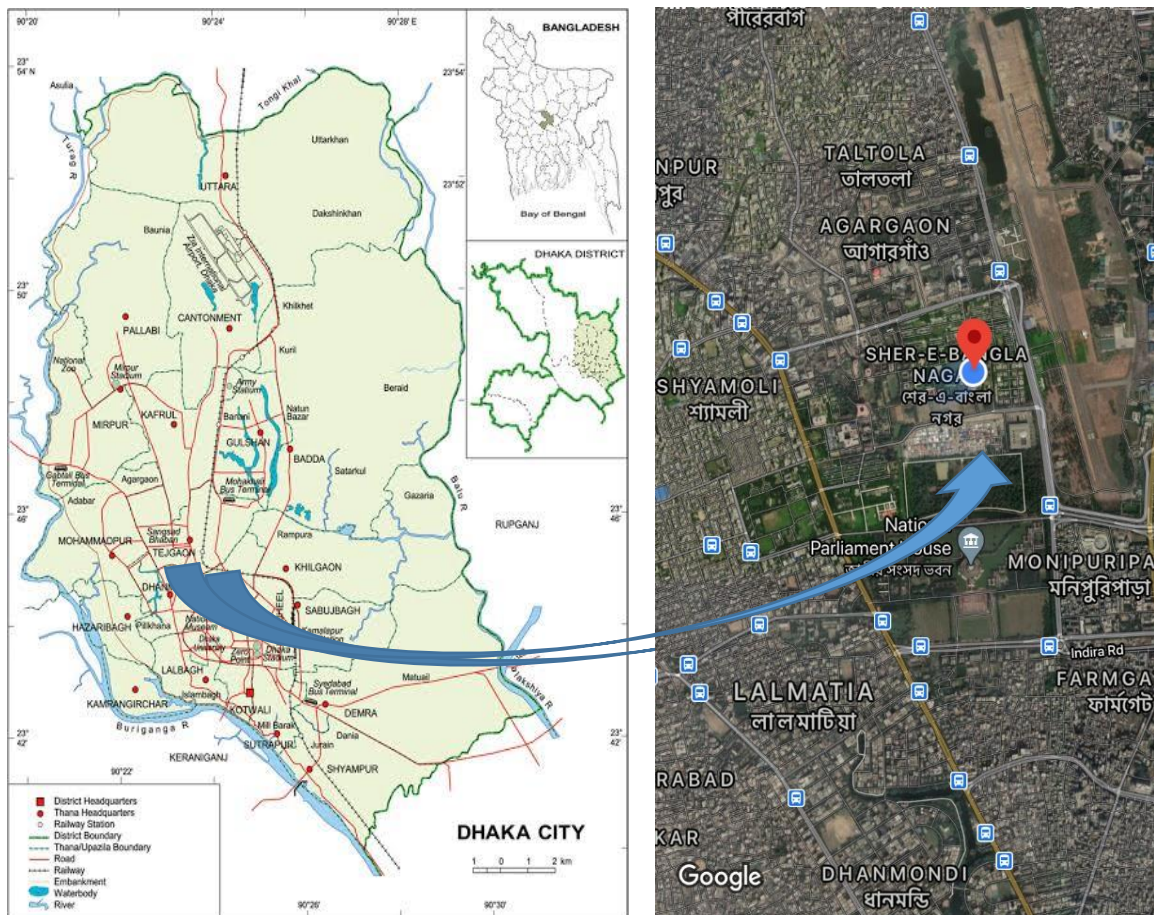


Figure 1: Study area of the experiment

### **3.2 Duration of the Study**

The time period for the study was January to December, 2020 for snakehead fish (*C. striata*) culture in biofloc system according to the supervisors schedule. The experiment was set considering the seed availability of *C. striata* and culture opportunity in that lab.

### **3.3 Experimental Design and Setup**

In this study, the experimental configuration was set by the instruction of biofloc culture system expert. There were four treatments and each of the treatments have three replications. Among of them one was control treatment and rest of three treatments where garlic (*Allium sativum*) and amla (*Emblica officinalis*) extracts were used for differentiate the growth, survival rate and other parameters of the cultured species. The experimental tanks (1000 L capacity) covered with nylon mesh mosquito net. Three hundred sixty fingerlings were equally distributed in all tanks each containing 30 fingerlings. Total volume of the water in each tank was maintained at 700 L throughout the experimental period. The aeration pipe in each tank was furnished with an air stone and water heater was also used to maintain due water temperature.

### **3.4 Pre-stocking Management**

Pre-stocking management implies some successive managerial exercises prior to stocking of fish species in tanks. Pre-stocking management focuses on proper preparation of tanks to remove the causes of poor survival, unsatisfactory growth and so forth, and also to ensure prepared accessibility of natural food in sufficient quantity and quality for the spawn/fry/fingerlings to be stocked. Pre-stocking part of the management includes the following successive measures:

#### **3.4.1 Tank Preparation**

The main prerequisite of biofloc based culture system on tank that can be made of any solid materials. In this experiment, tanks are made of iron structure that is covered by PVC coated water proof tarpaulin. Tank was treated by  $\text{KMnO}_4$  (potassium permanganate) and lime prior to pouring with water. After filling the tanks with water then proper aeration



system was given in all tanks. Salt and lime were given as 800-1000g and 5-10g per 1000 liter separately for expanding total dissolve solids and enhancing the water quality.



**Figure 2: Tank preparation for this experiment**

### **3.4.2 Floc preparation**

Floc implies the accumulation of organisms like bacteria, algae, protozoa, metazoan and so forth. In biofloc based culture, the microbial floc is utilized to change the ammonia into protein and also used as food substance for aquatic species. The microorganism (bacteria, fungus) used as a biotechnological agent for growth and development of snakehead fish culture. After preparing the water in the tank with continuing aeration for two days, then 10-20g probiotic (microbs) were added in the tank in addition with 50g raw sugar was applied for microbial development in 1000L tank. At that point a slight amount (approx. 4-5g) of commercial feed was applied for nitrogen formation which will assist to start the microbial development. In this study "Aqua Life Pro" was used for floc development. Following seven days, it was found that floc has already formed to 10ml per 1 liter which was understood by the observation in Imhoff Cone. When floc quantity is below than 40 ml/L in imhoff cone then it indicates that the floc quantity is appropriate and beneficial for fishes.





**Figure 3: Probiotic used in the experimental work**

### **3.4.3 Collection of fingerlings of shoal**

Good seed is the precondition of good result for any culture. In this experimental work fingerlings were collected from azizia hatchery of Noakhali. For getting great outcomes organoleptic tests that means healthy, good looking, disease free fingerlings were selected for collection purposes. Juvenile snakehead fish (*C. striata*) was collected for this experimental work. Various sizes of juvenile were collected ranged from 3 to 5 g.



**Figure 4: Collected fingerlings of shoal for the experiment**

#### **3.4.4 Disinfection of fingerlings**

After collecting the fingerlings from Azizia Hatchery, it ought to be disinfected. In this experiment juvenile snakehead fish (*C. striata*) was disinfected by potassium permanganate ( $\text{KMnO}_4$ ) called as potash. Around 1gm/L potash was applied in the polybag of fingerlings by which the fingerlings were carried. After applying potash in the polybag then within a minute the fingerlings were given to the experimental tanks according to the guidelines. Prior to stocking the fingerlings in the tanks, fingerlings were appropriately arranged and equally distributed. The initial weight of each fingerlings were measured and recorded.

#### **3.5 Species Stocking Density**

Juvenile snakehead fish (*C. striata*) was distributed equally in all tanks. There were total 165 pieces of juvenile *C. striata* were stocked in controlled treatments where fingerlings were equally distributed in all tanks of control treatments. Rest of the juveniles were stocked equally in garlic (*Allium sativum*) and amla (*Emblica officinalis*) extracts used treatments.

#### **3.6 Post-stocking Management**

The stage includes the exercises or maintenance from stocking to harvesting defined as post - stocking management. In case of biofloc based culture system, many activities were similar to traditional system like feeding, sampling, chemical application, harvesting and so forth. In this experiment of *C. striata* culture some different exercises were performed during the study period. All the cultural practices were done as and when necessary.

##### **3.6.1 Feeding**

Feeding is a significant issue for any sorts of culture. For getting good outcomes proper feeding should be maintained. Feeding was properly maintained on daily basis and five times feeding in two hours interval was accomplished. From the beginning of the study, feed was given at 5% rate of body weight and after that it was upto 1% rate during the study period.

### **Treatments of the Experiment:**

Four different treatments were designed to study the culture potentiality, survival rate and growth of snakehead fish. The combination of treatments are given below:

T-1 (Control) = Only commercial feed (No floc present)

T-2 (EG) = Commercial feed + Garlic extract (Floc present)

T-3 (EA) = Commercial feed + Amla extract (Floc present)

T-4 (EG & EA) = Commercial feed + 50% Garlic extract and 50% Amla extract

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

Commercial feed = “Kwality Feed” of Kwality Feed Mills Ltd. and “Agata Feed” of Agata Feed Mills Ltd. were used as commercial feed.

### **3.6.2 Use of Extracts**

In this study two types of extract were mixed with the feed for experimental purposes. Extract of Garlic (*Allium sativum*) and extract of Amla (*Emblica officinalis*) were used as feed supplement. Extract was mixed with feed (4ml/50g) before two hours of feed application in the tank. As snakehead fishes is surface feeder, feed was given by disseminating at the surface level of water.

### **3.6.3 Water Quality Parameters**

Fish perform all their bodily functions in water. Because fish are absolutely dependent upon water to inhale, feed and grow, excrete wastes, keep a salt equilibrium and reproduce. Understanding the physical and chemical characteristics of water is crucial to effective aquaculture. Water quality is the key of success or failure of an aquaculture practices. The following water parameters were studied.

#### **A) Temperature**

Temperature is a significant issue in case of biofloc based fish culture. Numerous water parameters rely upon its balance. Good temperature range assists to grow fish and other

microbial community appropriately. Temperature range was measured everyday morning during the study period. Data was collected by using digital multimeter.

## **B) pH**

pH is also an important parameter for aquatic species specially fish culture. pH defines the acid and base concentration of water. pH levels are connected with metal concentration, nutrients deficiencies and problems. In this study pH was measured everyday through using digital multimeter. For this, water was gathered in plastic mug from treatment tanks and pH sensor was used onto it. pH level was maintained between the range of 4.00-9.00 mg/L.

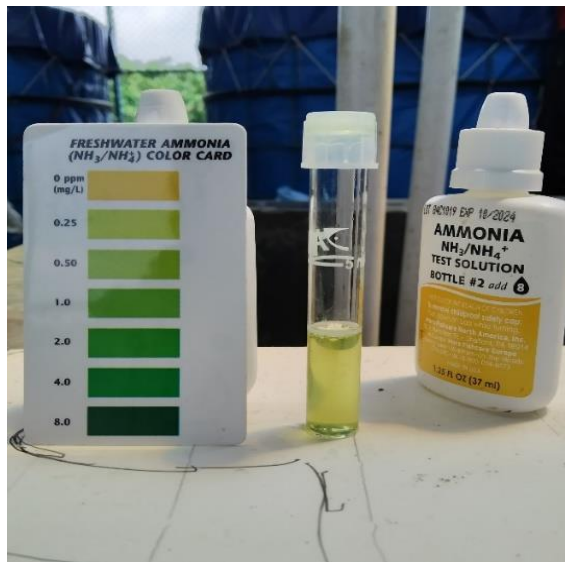


**Figure 5: Process of pH measurement**

## **C) Ammonia (NH<sub>3</sub>)**

Ammonia is the most significant harmful factor for successful fish culture. In biofloc based fish culture nitrogenous compound is an essential factor for fish survival rate. Basically, ammonia is formed by the excreta of cultured species, deposition of extra feed on the tank bottom level. The most significant thing is ammonia controlled biofloc culture because of

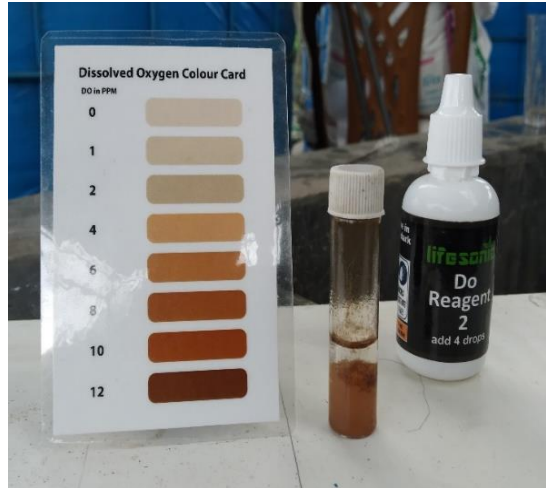
microbial accumulation in the tank water. Useful bacteria helps to change over the ammonia into protein with the help of carbohydrate sources. Furthermore, carbon source was given routinely for maintaining ammonia concentration in the water according to ammonia level. Ammonia level was measured by API's ammonia kit. Microbial biotechnological system work on conversion of ammonium into protein. Microbes metabolized the ammonia and produced protein product which was a good source of nutrient for snakehead fish.



**Figure 6: Ammonia measurement kit**

#### **D) Dissolved Oxygen (DO)**

For aquatic species dissolved oxygen is the most crucial factor for their survival. Fish inhale oxygen similarly as we do, so dissolved oxygen (DO) is a critical environmental indicator in aquaculture. Low level DO from the appropriate level is the main cause of fish mortality, and it is realized that low-oxygen conditions are their most noticeably enemy. In case of fish culture at least 5.0 ppm DO level ought to be maintained. In this experimental work an oxygen motor with 1 HP was utilized for keeping up DO level. In each tank, there were four or five air stone used for proper oxygen supply. DO level data was measured by Life Sonic's DO kit at regular routine.



**Figure 7: Dissolved oxygen (DO) measurement**

### **E) Alkalinity**

Alkalinity is the capacity of water to resist acidification without an expansion in pH. Alkalinity is the strength of a buffer solution made out of weak acids and their conjugate bases. It is estimated by titrating the solution with an acid such as HCl until its pH changes abruptly, or it reaches a known endpoint where that happens. This parameter is a proportion of the bases, bicarbonates ( $\text{HCO}_3^-$ ), carbonates ( $\text{CO}_3^{2-}$ ) and in rare instances, hydroxide ( $\text{OH}^-$ ). The appropriate range of alkalinity is between 80 – 280 mg/L. It was maintained by using proper chemical application in each treatment tanks.



**Figure 8: Alkalinity measurement of different treatments**



## **F) Hardness**

Water hardness is like alkalinity but represents different measurements. Hardness is mainly a measure of calcium and magnesium, but other ions such as aluminium, iron, manganese, strontium, zinc, and hydrogen ions are also included. The ideal total alkalinity level for most aquaculture species lies between 50-150 mg/L CaCO<sub>3</sub>, but not less than 20 mg/L. The hardness level is suitable when the level is higher than 100 mg/L. It was recorded and maintained in regular basis. The measurement of hardness was conducted by “Hanna’s Kit”.

## **G) Total Dissolved Solid (TDS)**

A total dissolved solid (TDS) is a proportion of the combined total of organic and inorganic substances contained in a liquid. This incorporates anything present in water other than the pure H<sub>2</sub>O molecules. These solids are primarily minerals, salts and organic matter that can be an overall indicator of water quality. It was also measured for each treatment.

### **3.6.4 C: N Ratio**

Carbon and nitrogen ratio (C: N) is a ratio of the mass of carbon to the mass of nitrogen in a substance. The maintenance of C/N ratio is quite prerequisite for controlling of accumulating organic nitrogen and for the production of microbial communities in the water (Asaduzzaman *et al*, 2008 and Emerenciano, 2012). The inorganic nitrogen is converted into organic nitrogen when C:N ratio is sufficient to produce bacterial cells (Aly *et al*, 2008). To minimize the artificial feed requirement, the practice of increasing C: N of higher than 10:1 by utilizing different low-cost carbon sources which are locally obtainable is common in biofloc waters (Crab, 2010). It was established that the collection of toxic inorganic components including NH<sub>4</sub><sup>+</sup> also NO<sub>2</sub><sup>-</sup> will be halted in the water when the maintenance of C/N proportion is high in the biofloc system as the ammonium consumption by the microbial community. For example, a C: N of 10:1 means there is ten units of carbon for each unit of nitrogen in the substance. In this study, carbon-nitrogen ratio was maintained by estimating the amount of carbon and nitrogen released from feed and the condition of ammonium of the water. By considering the condition of carbon and nitrogen raw sugar was applied to mitigate the actual amount.



**Figure 9: Applying carbon source in contrast of nitrogen**

### **3.6.5 Application of Vitamin-C**

Vitamin-C probably is the most significant because it is a powerful antioxidant and immune modulator for fishes. The fish body needs vitamin-C (ascorbic acid or ascorbate) to remain in proper health condition. Vitamin-C also requires in wound healing, bone and tooth development, strengthening blood vessel walls, improving immune system function, increasing absorption and use of iron and acting as an antioxidant. Vitamin-C reduces the impacts of toxic chemicals in water and prevent negative effects of water temperature fluctuations. In this experimental work, Vitamin-C “Ceevit” of Square pharmaceutical company was applied by grinding and then mixing with feed in several days for wound healing purposes.

### **3.6.6 Sampling Schedule**

Sampling is a process used in statistical investigation in which a predetermined number of observations were taken from a larger population. This tool describes the samples to take in order to evaluate a system, process, issue or problem. Sampling saves time and the data can be collected and investigated more quickly with a sample than a complete count of the entire population. In this research work sampling was done properly at 15 days of interval. Random sampling was done manually with the assistance of scoop net.



### **3.6.7 Harvesting**

Harvest means the number and weight of fish caught from each treatment and retained from a given area throughout a given period of time. Harvesting is done to analyse the different parameters of any species in an experiment after a timeframe. In this experimental work this procedure was accomplished after 4 months. This process was finished by draining 90 percent water of the tanks. By harvesting there were 40-50 pieces of snakehead fish were randomly taken to evaluate the performance of overall growth in this timeframe.

### **3.7 Growth Parameters**

Growth is an integrated physiological response enveloping external environmental conditions (food quality and quantity, temperature, water quality) and internal physiological status (health, stress, reproductive state). The growth parameters such as length, weight, size, shape were measured.

### **3.8 Evaluation of Nutritional Value**

Nutritional value refers to contents of food and the effect of constituents on body. The analysis was conducted on commercial feed that was used in the time of study, formed biofloc and the cultured species (*Channa striata*) in the final stage of experiment from a governmental organization of Bangladesh called Animal Nutrition Section, Department of Livestock Services. On a biological scale, nutritive value of food may vary for different health conditions, seasonal differences, age and sexual differences, and interspecies or more taxonomic differences.

### **3.9 Data Collection**

Water quality parameters were measured daily. Fish were sampled for assess the growth performance in terms of body weight (g) and total length (cm) in every 15 days of interval. Final weight of individual fish and total weight of all snakehead fish in each treatment was recorded after 120 days timeframe. Dead fish were removed when mortality occurred and counted the survived fish in every sampling period. Total number of survived fish in each treatment was calculated at the end of the experiment. The formula which were applied in this experiment are given in below:

$$\text{Feed Conversion Ratio (FCR)} = \frac{\text{Amount of feed}}{\text{Fish weight gain}}$$

$$\text{Survival rate (\%)} = \frac{\text{Final number of fish}}{\text{Initial number of fish}} \times 100$$

$$\text{Absolute length (cm/fish)} = (\text{Final body length}) - (\text{Initial body length})$$

$$\text{Weight gain (g/fish)} = (\text{final body weight}) - (\text{initial body weight})$$

$$\text{Weight gain (\%)} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}} \times 100$$

$$\text{Daily mean weight gain} = \frac{(\text{Final weight} - \text{Initial weight})}{(\text{Total day of experiment})}$$

$$\text{SGR (\%)} = \frac{\text{Ln final body weight} - \text{Ln initial body weight}}{\text{Number of days}} \times 100$$

$$\text{Net Production (kg/m}^3\text{)} = \frac{\{\text{SR (\%)} \times \text{Stocking density} \times \text{Weight gain (g)}\}}{100 \times 1000}$$

$$\text{Food Conversion Efficiency (FCE)} = \frac{(\text{Total weight of feed})}{(\text{Net production})}$$

### 3.10 Statistical Analysis

For the purpose of getting better outcome of the experimental result statistical analysis was done. All non-repeatedly measured variables (snakehead growth parameters) were analyzed by one-way ANOVA tukey's test using PAST software at  $P < 0.05$  level of significance.

## CHAPTER 4

### RESULTS AND DISCUSSION

This chapter confined the results on the investigation accomplished during the research timeframe to fulfill the mentioned objectives. Growth performance, water quality parameters and nutritional value of snakehead fish (*C. striata*) have been presented and discussed for obtaining the actual potentiality of Biofloc Technology which has been accompanied with microbial biotechnology. The experimental findings are given in below:

#### 4.1 Water Quality Parameters

Water quality parameters incorporate chemical, physical, and biological properties and can be tested or monitored based on the ideal water parameters of concern. Parameters that are daily tested or monitored for water quality include temperature, dissolved oxygen, pH, alkalinity, ammonia, hardness. When water quality is poor, it influences not only the aquatic life as well as the surrounding ecosystem too. The comprehension and understanding of water quality parameters and its interactions in BFT are crucial to the correct development and maintenance of the production cycle. In the following table water quality parameters during the experimental period were given: (Table 01)

Table 01: Water quality parameters of different treatments

Parameters	T-1 (Control)	T-2 (EG)	T-3 (EA)	T-4 (EG + EA)
Temp. (°C)	30.29±4.40	30.22±4.35	30.14±4.36	30.27±4.39
pH	7.15±0.48	7.05±0.52	7.16±0.52	7.09±0.53
NH <sub>3</sub>	0.63±0.68	0.55±0.52	0.56±0.50	0.59±0.67
DO	5.81±0.88	5.93±0.92	5.91±0.93	5.88±0.95
Alkalinity	212.06±20.16	206.18±8.57	210.88±16.42	208.53±14.77
Hardness	103.88±14.32	112.06±14.69	111.76±11.03	106.53±13.43
TDS	405.14±38.03	406.33±39.12	409.67±28.27	405.05±34.49

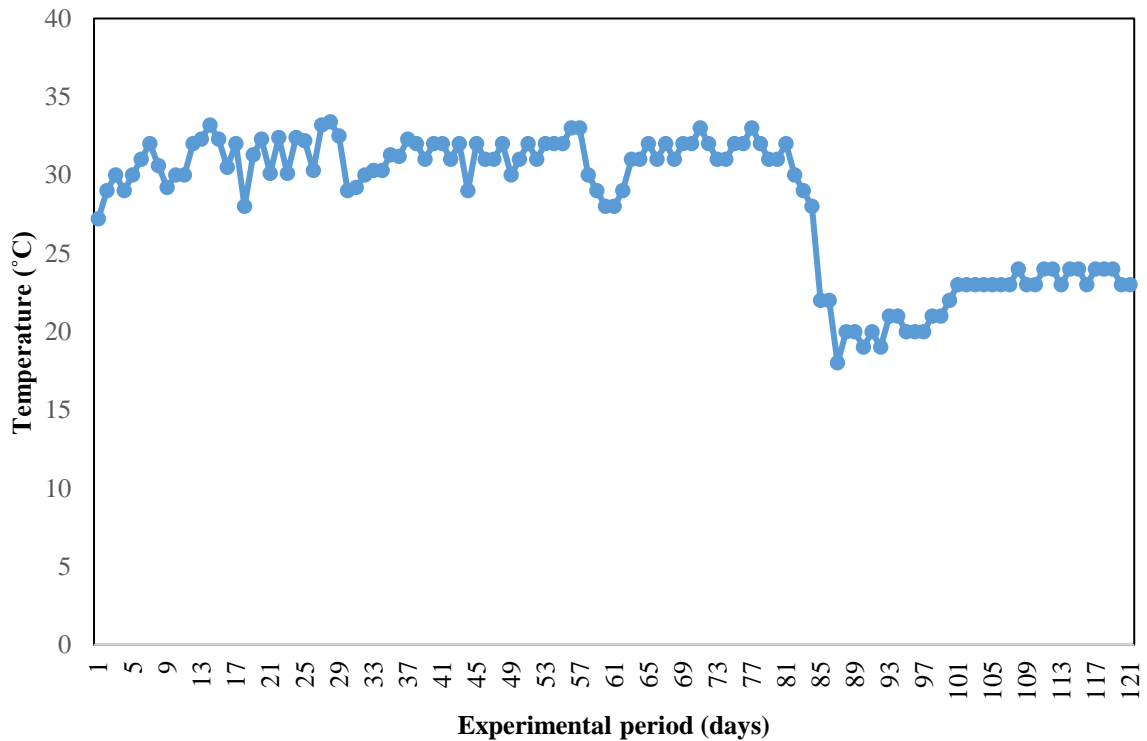
(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of both Garlic and Amla)

[Unit of pH, DO, NH<sub>3</sub>, Alkalinity, Hardness and TDS = mg/L, Temperature = °C]

### 4.1.1 Temperature

#### I. Treatment-1 (Control)

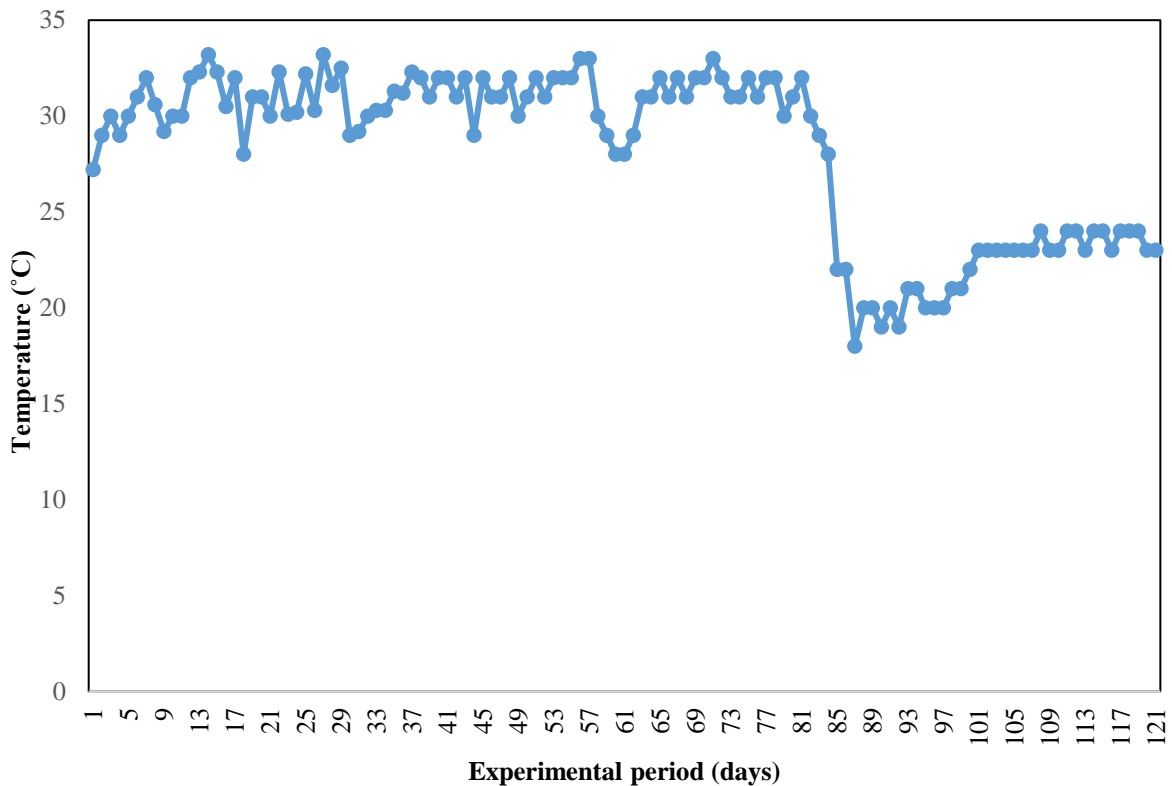
Temperature is an important factor for growth and development in case of aquatic organisms. Although aquatic organisms are poikilothermic in nature it is crucial for the growth and survival of aquatic animals. The rate of growth significantly increased with the increase of temperature. Range of temperature which is below 20°C influences the microbial growth and it also affects the appetite of aquatic species which leads them not to take proper amount of feed. Optimum range of temperature helps fish to become more active in aquatic environment. So it is crucial to maintain optimum level of temperature throughout the experimental period. In this experimental work, the average temperature range of treatment-1 (control) was  $(30.29 \pm 4.40^\circ\text{C})$ ; where highest range was  $33.29^\circ\text{C}$  and lowest range was  $18.14^\circ\text{C}$  during the experimental period. It was shown in the experiment that, the temperature gradually came down after 81 days and drastically reduced in between 81 to 89 days. (Figure 10)



**Figure 10: Temperature during the experimental period in treatment-1 (Control)**

## II. Treatment-2 ( Extract of Garlic)

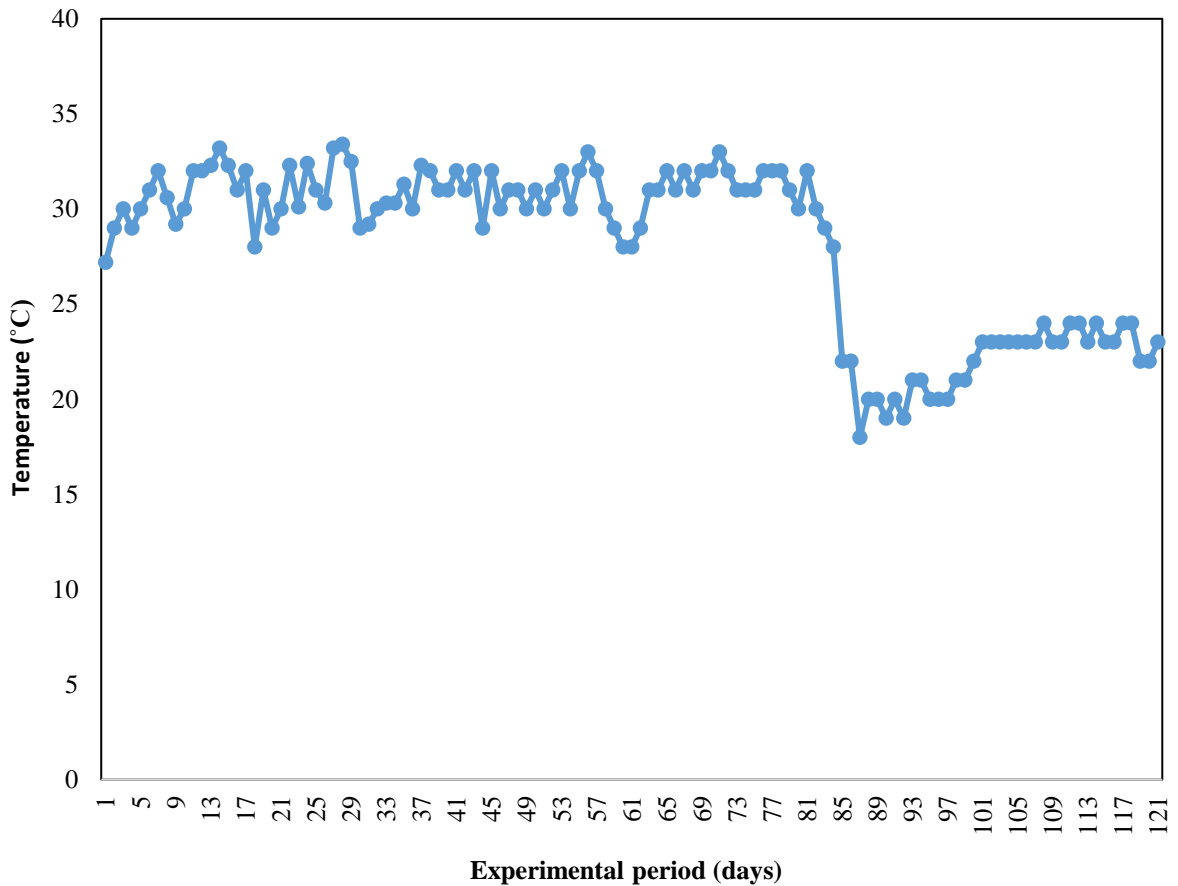
It was observed that the average temperature range was  $(30.22\pm 4.35^{\circ}\text{C})$  in treatment-2 (EG). The highest range was  $33.15^{\circ}\text{C}$  and lowest range was  $17.63^{\circ}\text{C}$  throughout the experimental duration (Figure 11). Drastic reduction of temperature was observed in this experiment as like the control treatment mentioned in figure 10.



**Figure 11: Temperature during the experimental period in treatment-2**

## III. Treatment-3 (Extract of Amla)

The average temperature range was  $(30.14\pm 4.36^{\circ}\text{C})$  in treatment-3 (EA). Where highest range was  $33.36^{\circ}\text{C}$  and lowest range was  $18.29^{\circ}\text{C}$  throughout the experimental duration (Figure 12). According to the report of Sangeeta *et al.* (2017), highest temperature range was  $(23.4-25.1^{\circ}\text{C})$  and lowest range was  $(22.1-24.3^{\circ}\text{C})$ . Muntaziana *et al.* (2013) reported that during their experimental period the temperature range was  $(29.76\pm 0.22^{\circ}\text{C})$  which helps to growth and production of their cultured species.

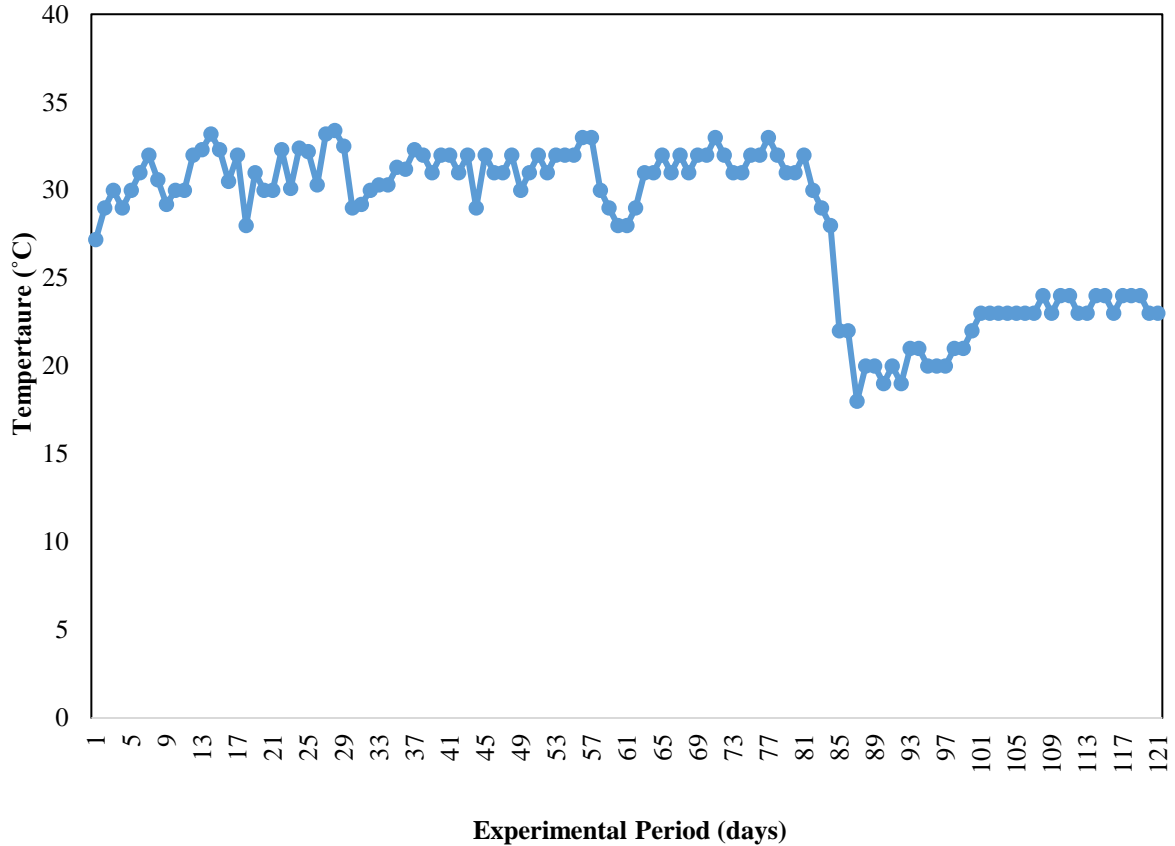


**Figure 12: Temperature during the experimental period in treatment-3**

#### **IV. Treatment-4 (Mixture of Garlic and Amla Extract)**

The average temperature range was  $(30.27 \pm 4.39^\circ\text{C})$  in treatment-4 (EG & EA). Where highest range was  $33.46^\circ\text{C}$  and lowest range was  $18.42^\circ\text{C}$  throughout the research duration (Figure 13). Temperature drastically came down after 81 days because of winter season. Due to drastic disruption of temperature fishes took less amount of feed than normal temperature period and some mortality was occurred on that time. Muslim (2007) gave a conclusion that the temperature range that could be tolerated by the *C. striata* were  $(25.5-32.7^\circ\text{C})$ . So, these study findings are fully support the actual range of temperature. Although there has some deviation of temperature during the study period but there was no

such harmful effects in those treatment tanks. So, it can be stated that the study findings are fully support the actual range of temperature which is crucial for the culture of targeted species.

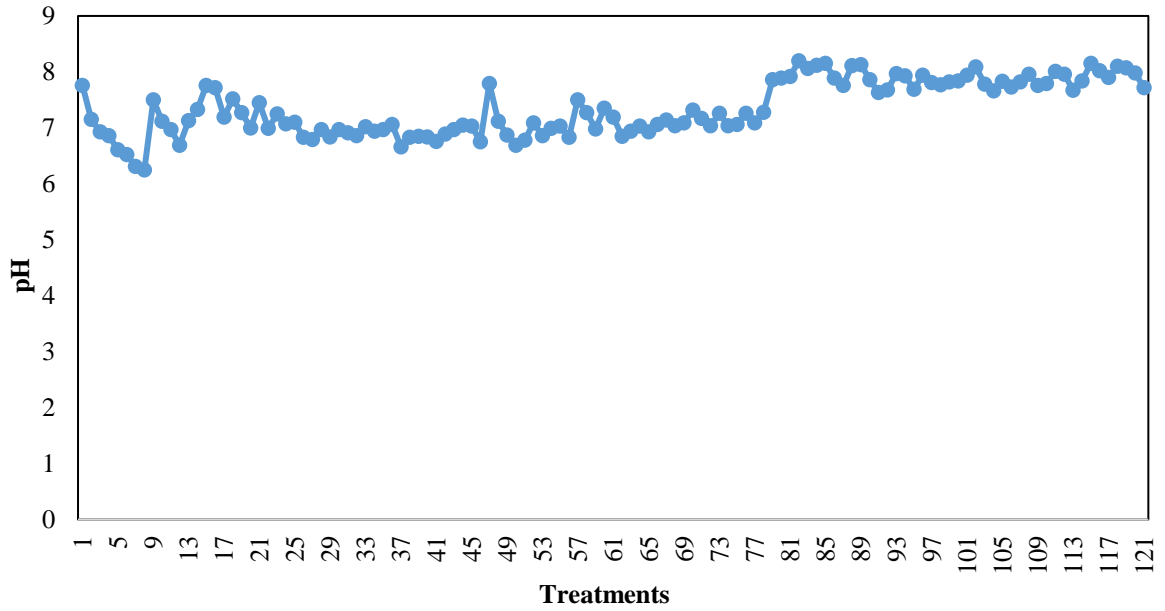


**Figure 13: Temperature during the experimental period in treatment-4**

#### 4.1.2 pH Parameter

##### i. Treatment-1 (Control)

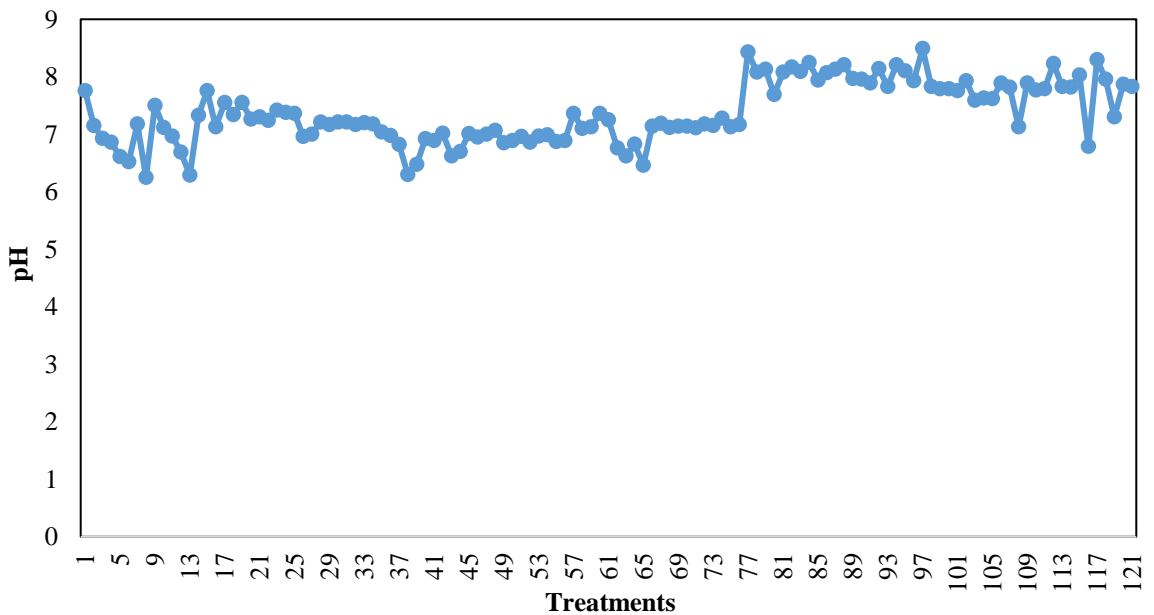
pH is a proportion of how acidic/base water is. pH denoting 'potential of hydrogen' or 'power of hydrogen' is a scale used to specify the acidity or basicity of an aqueous solution. frequent change in pH can cause stress, poor growth and even death of the cultured species in biofloc culture system. So pH is an important factor for fish growth and survival. In this research work, for treatment-1 which was control the average pH range was  $(7.15 \pm 0.48)$ ; where the highest range was 8.15 and the lowest range was 6.10 throughout the experimental duration (Figure 14).



**Figure 14: pH during the experimental period in treatment-1 (Control)**

**ii. Treatment-2 (Extract of Garlic)**

It was observed that, the average pH range was  $(7.05 \pm 0.52)$  in treatment-2 (EG). Where highest range was 8.13 and lowest range was 6.15 during the research period (Figure 15).

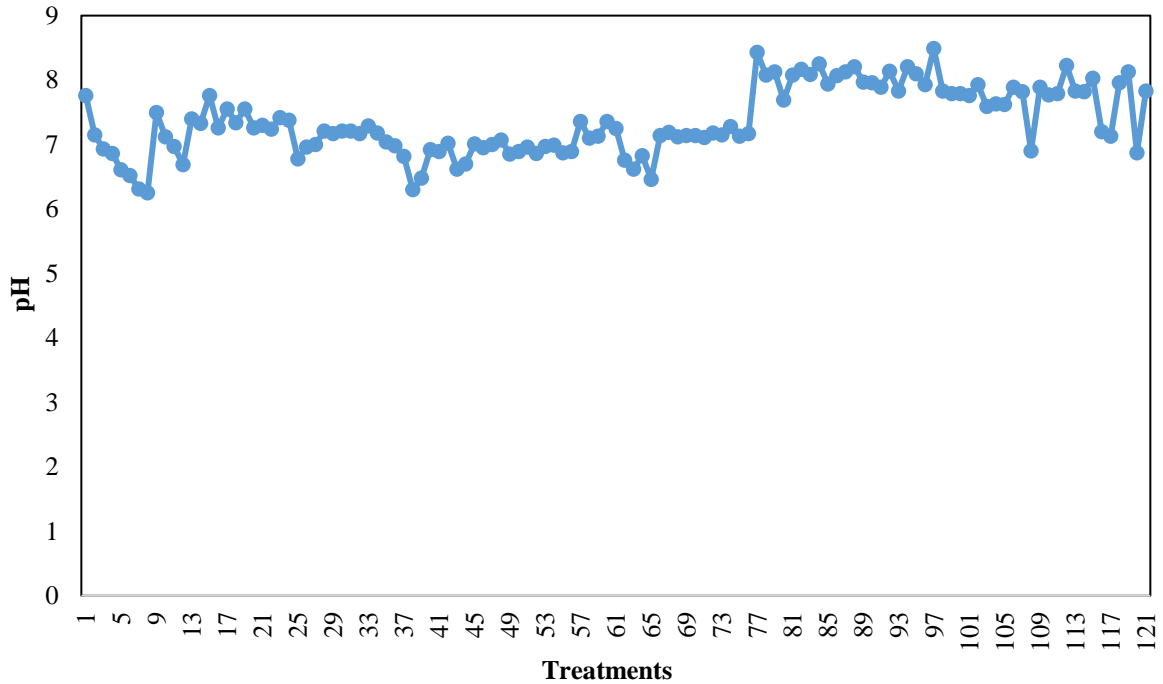


**Figure 15: pH during the experimental period in treatment-2**



### iii. Treatment-3 (Extract of Amla)

The average pH value ( $7.16 \pm 0.52$ ) was recorded in the treatment-3 (EA). Where highest range was 8.21 and lowest range was 6.05 during the experimental period (Figure 16). Lime was applied at a slight amount for maintaining the pH ratio which helps to proper growth and development of the cultured species. Sangeeta *et al.* (2017) reported that, the highest pH range was (7.8-8.5) and lowest range was (7.1-8.2) in snakehead fish culture.

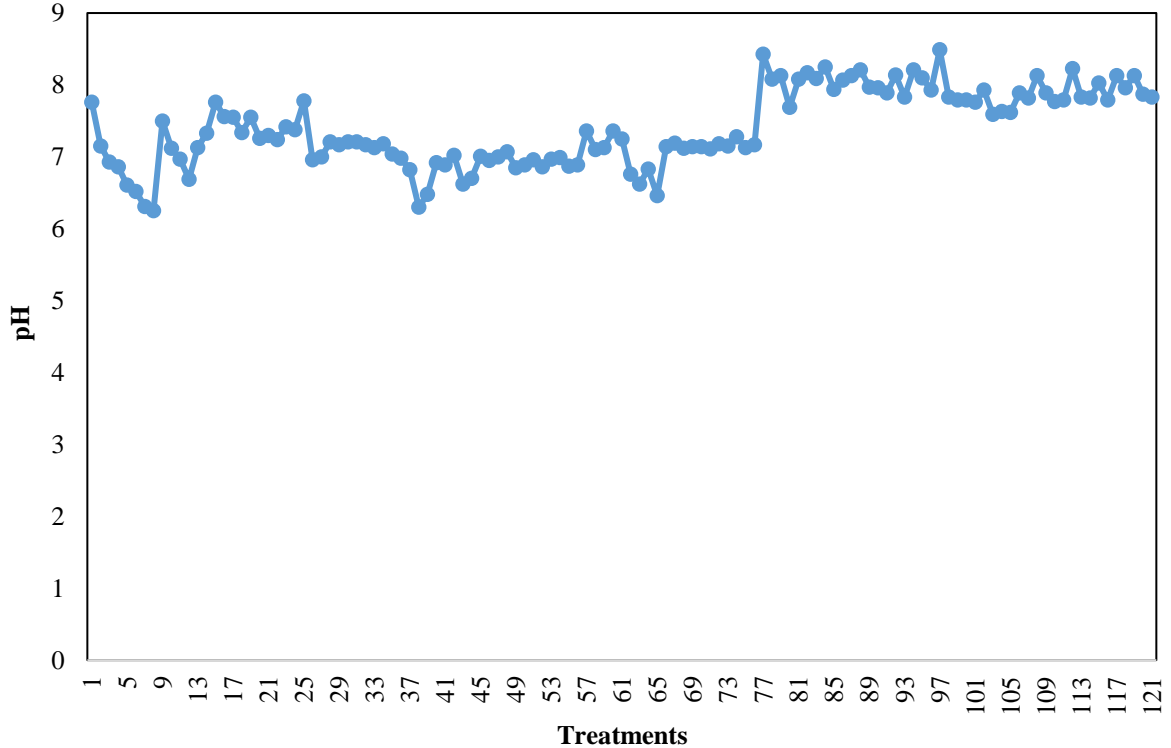


**Figure 16: pH during the experimental period in treatment-3**

### iv. Treatment-4 (Mixture of Garlic and Amla Extract)

The average value of pH in treatment-4 (EG & EA) was ( $7.09 \pm 0.52$ ). The highest value was 8.25 mg/l and the lowest value was 6.15 throughout the research duration (Figure 17). Snakehead fish *C. striata* fed with different diets having pH ranged from (8.1 to 8.2) according to the report of Sugumaran *et al.* (2018). By the experiment of Saputra *et al.* (2018) the pH parameter was around (5.25–6.31). According to Courtenay & Williams (2004), the tolerance range of pH for the life of *C. striata* was at the range of (4.25-9.4). Lower pH range tells the acidity of water that can be fatal for aquatic species. Balanced pH range helps to keep fish agile in aquatic environment. pH range of water is inversely related

to temperature but it doesn't mean that water becomes more acidic at higher range of temperature. From the pH range of all treatments it can be stated that, pH level was always in an optimum range for snakehead fish culture in the present study.

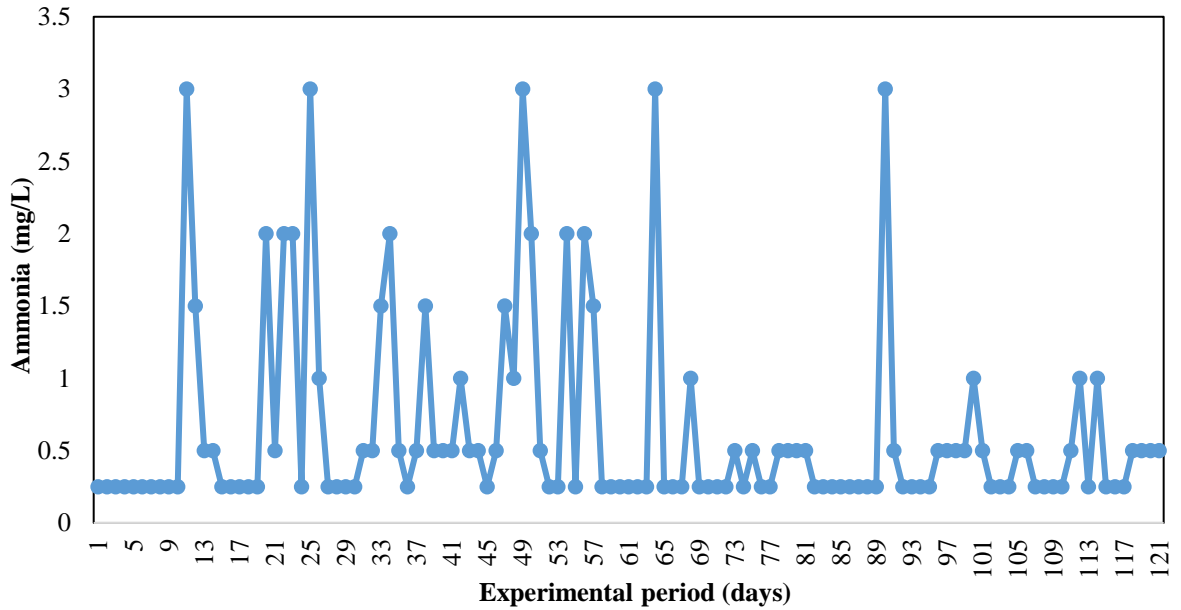


**Figure 17: pH during the experimental period in treatment-4**

#### **4.1.3 Ammonia**

##### **i. Treatment-1 (Control)**

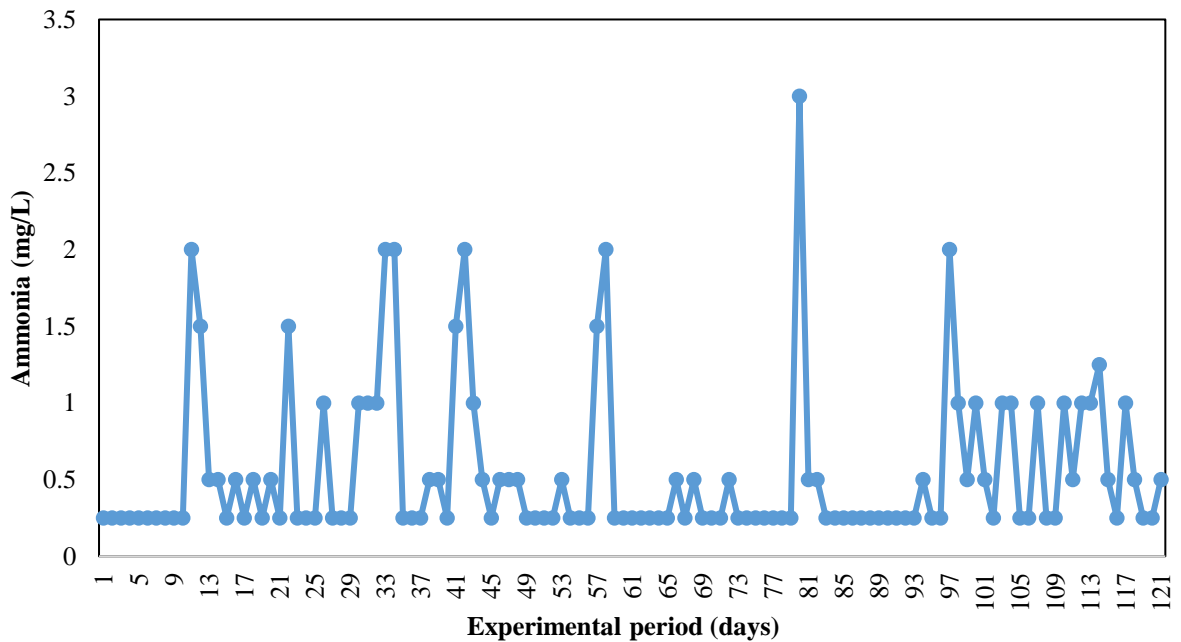
Ammonia is a compound of nitrogen and hydrogen with the equation  $\text{NH}_3$ . Ammonia ( $\text{NH}_3$ ) is created from the reform process of proteins (amino acids) contained in the feed eaten by the fish and converting them into energy. A steady double hydride, ammonia is a colorless gas with a particular characteristic of a pungent smell. In this experimental work, the average ammonia range was  $(0.63 \pm 0.68 \text{ mg/L})$  in control treatments; where the highest range was 3 mg/L and the lowest range was 0.25 mg/L throughout the experimental duration. (Figure 18)



**Figure 18: Ammonia range during the experimental period in treatment-1 (Control)**

**ii. Treatment-2 ( Extract of Garlic)**

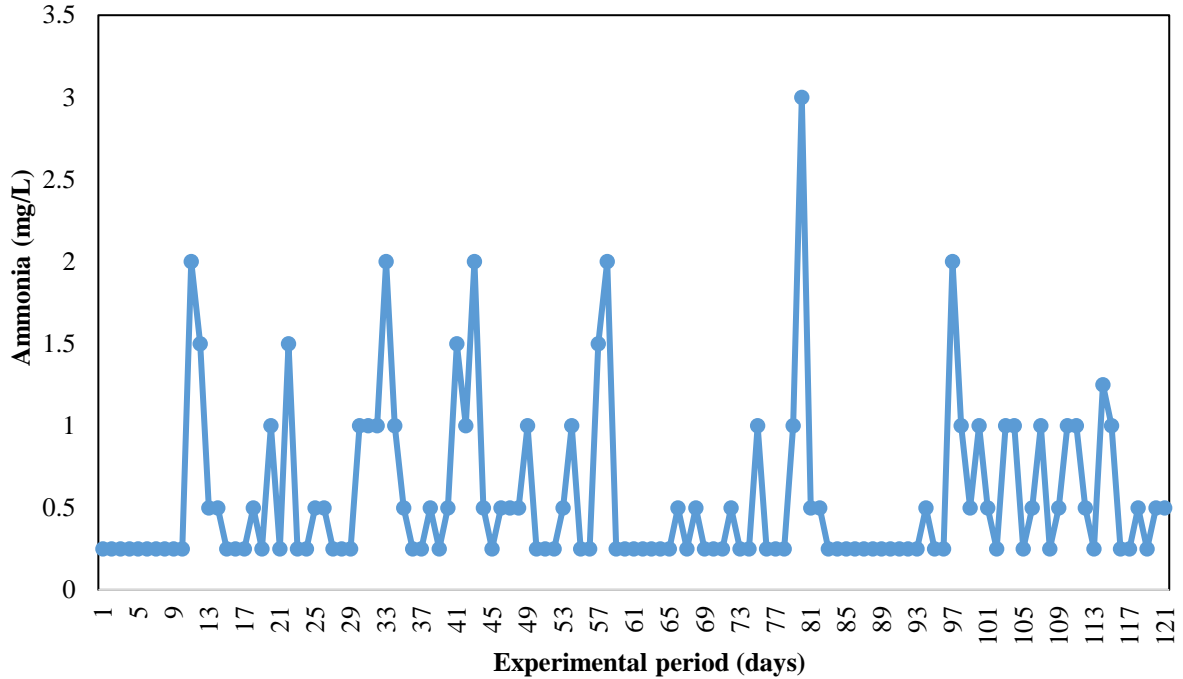
Average value of ammonia was  $(0.55 \pm 0.52)$  mg/L in treatment-2 (EG). Where the highest value was 3.10 mg/L and the lowest value was 0.25 mg/L during the experimental period. (Figure 19)



**Figure 19: Ammonia range during the experimental period in treatment-2**

### iii. Treatment-3 (Extract of Amla)

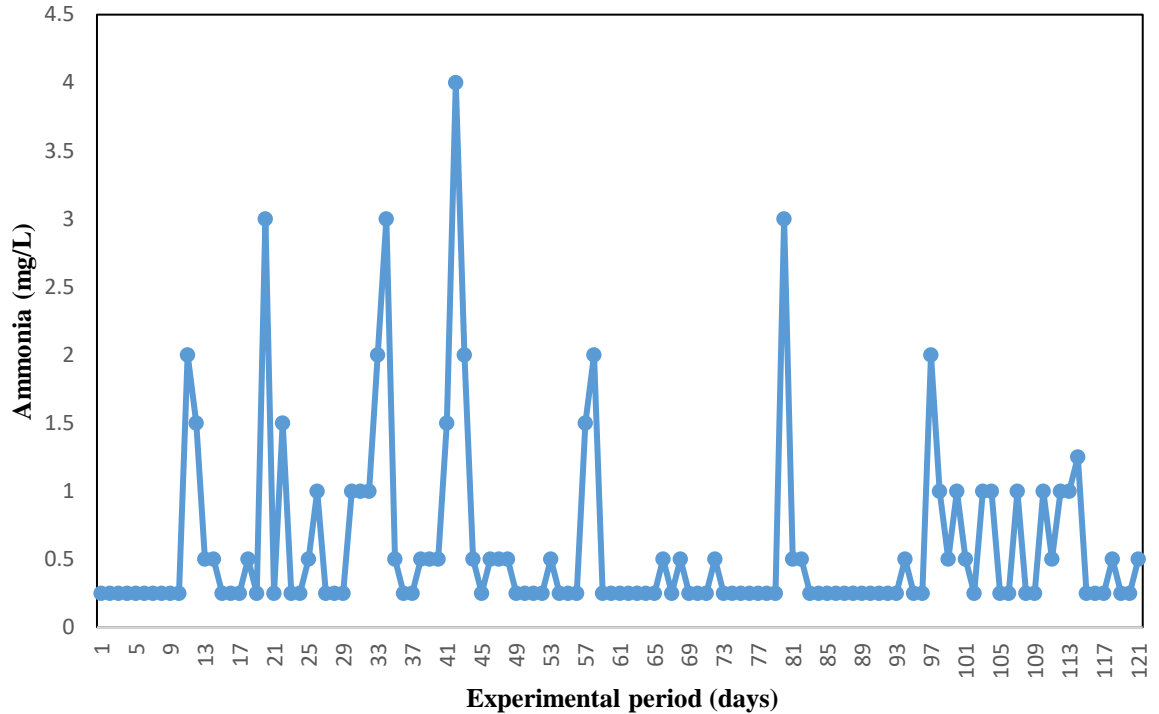
It was reported that, the average ammonia range was  $(0.56 \pm 0.50)$  mg/L in treatment-3 (EA). Where highest range was 3.05 mg/L and lowest range was 0.25 mg/L during the experimental period. (Figure 20)



**Figure 20: Ammonia range during the experimental period in treatment-3**

### iv. Treatment-4 (Mixture of Garlic and Amla Extract)

The average ammonia range in treatment-4 (EG and EA) was  $(0.59 \pm 0.67)$  mg/L. Where the highest range was 4 mg/L and the lowest range was 0.25 mg/L throughout the research duration (Figure 21). In biofloc-based intensive culture system optimum ammonia range was between  $(0.26-0.27)$  mg/L which was reported by Yusuf *et al.* (2015). Ammonia ranged from  $(0.0$  to  $0.43)$  in T1,  $(0.0$  to  $0.5)$  T2 and  $(0.0$  to  $0.75)$  mg/L in T3, which was recorded from the effect of biofloc on water quality parameters conducted by Anand *et al.* (2018). At an ammonia concentration of  $1.57$  mg/L, *C. striata* juvenile can still grow which was reported by Jianguang *et al.* 1997. Maximum time of the experimental period the ammonia range was  $(0.25-0.50)$ , Which indicates the optimum level of ammonia for any experimental work.

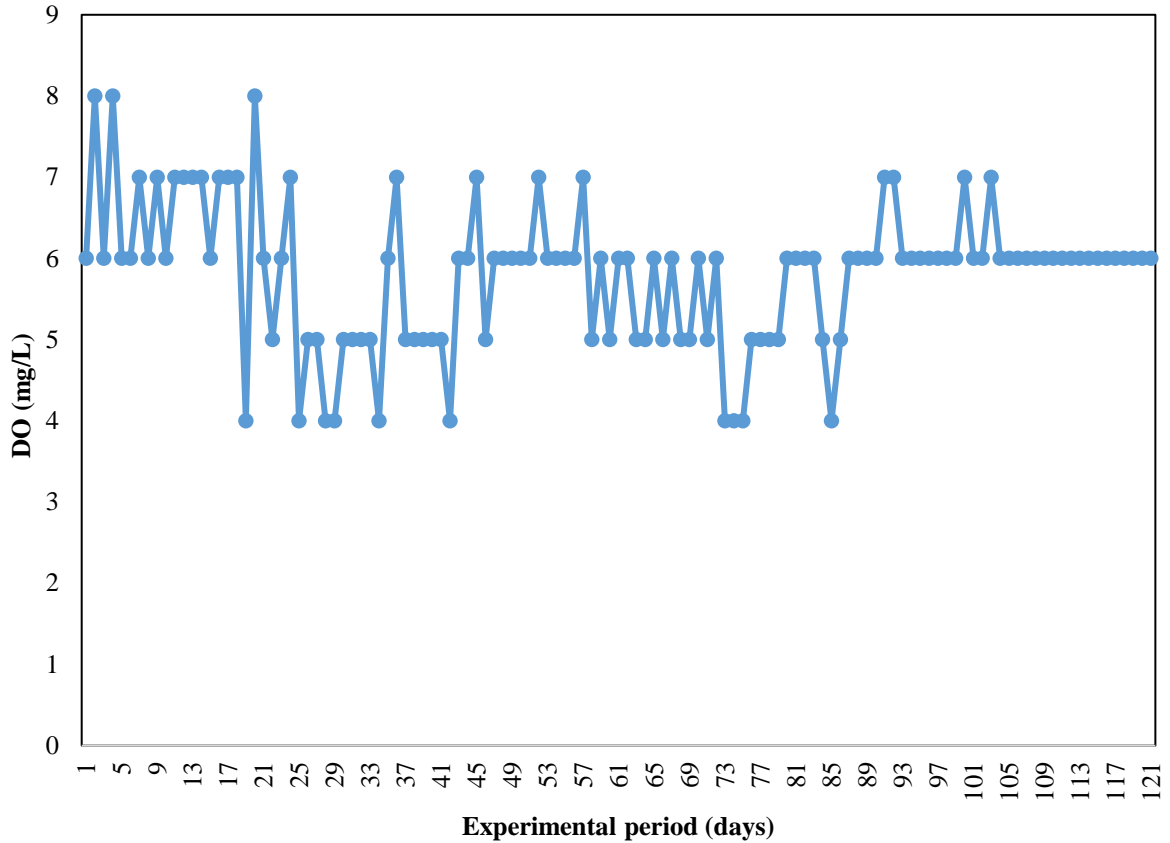


**Figure 21: Ammonia range during the experimental period in treatment-4**

#### 4.1.4 Dissolved Oxygen (DO)

##### i. Treatment-1 (Control)

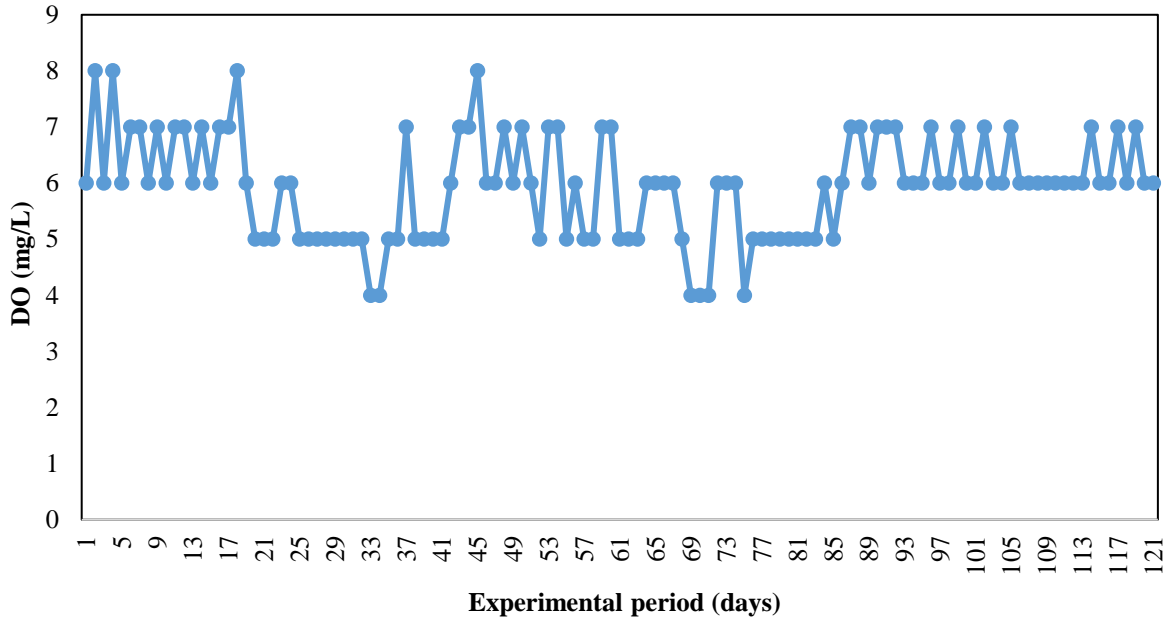
Dissolved Oxygen (DO) is the amount of gaseous oxygen (O<sub>2</sub>) that is dissolved in the water. Dissolved oxygen (DO) is an important factor for fish growth and survival. Oxygen saturation is an overall proportion of the concentration of oxygen that is dissolved in a given medium as an extent of the maximal concentration that can be dissolved in that medium. Water bodies get oxygen from the air and from oceanic plants. Running water, like that of a quick moving stream, breaks up more oxygen than the still water of a pond or lake. Oxygen enters the water by direct absorption from the atmosphere, by quick movement, or as a byproduct of plant photosynthesis. In treatment-1 (control) tanks the average DO level was noticed ( $5.81 \pm 0.88$ ) mg/L; where the highest range was 8 mg/L and the lowest range was 4 mg/L during the experimental period. (Figure 22)



**Figure 22: Range of dissolved oxygen during the experimental period in treatment-1**

**ii. Treatment-2 (Extract of Garlic)**

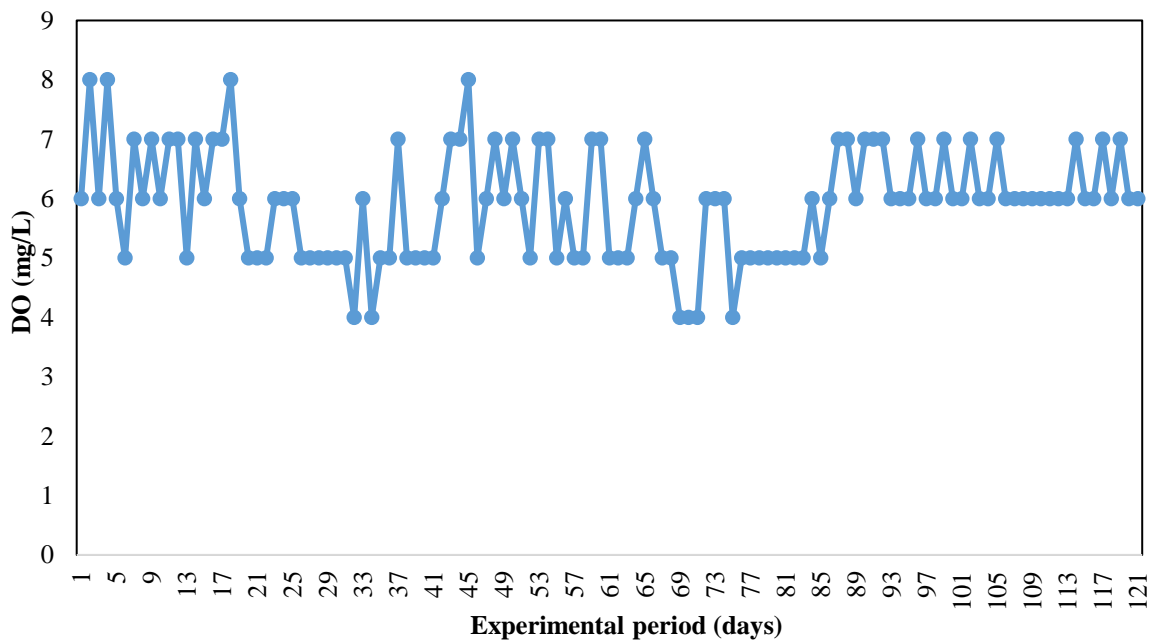
It was observed that, the average DO level in treatment-2 (EG) was  $(5.93 \pm 0.92)$  mg/l. Where the highest range was 8 mg/L and the lowest range was 4 mg/L throughout the experimental duration (Figure 23). DO is a crucial factor for snakehead fish culture because without sufficient DO level feed intake rate might be decrease and for that reason growth ratio can be hampered. Sugumaran *et al.* (2018) reported in their study that average dissolved oxygen ranged was (6.20 to 6.87 mg/L). Anand *et al.* (2018) gave a conclusion that, in treatment-1 dissolved oxygen level was from (5.5 to 8.0 ppm), in treatment-2 it was (6.5 to 8.0 ppm) and in treatment-3 dissolved oxygen range was (6.0 to 8.0 ppm).



**Figure 23: Range of dissolved oxygen during the experimental period in treatment-2**

**iii. Treatment-3 (Extract of Amla)**

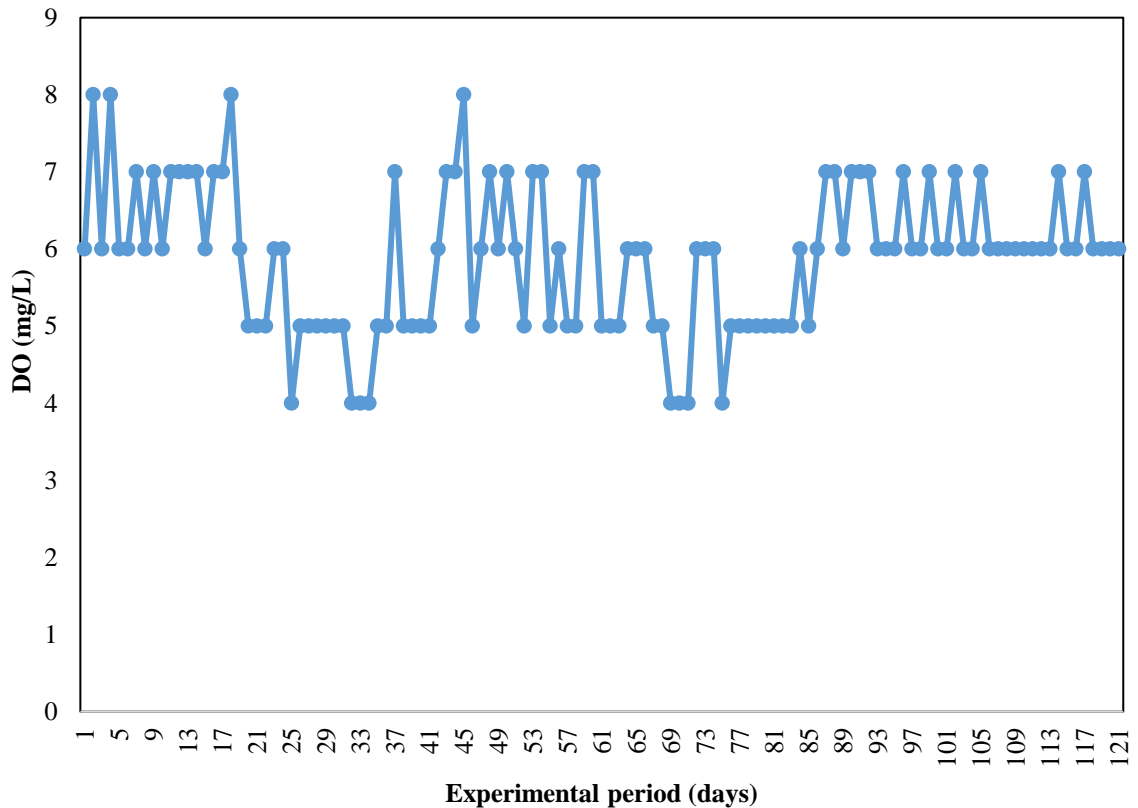
It was reported that, the average DO level in treatment-3 (EA) was  $(5.91 \pm 0.93)$  mg/L. Where the highest range was 8 mg/L and the lowest range was 4 mg/L during the experimental period. (Figure 24)



**Figure 24: Range of dissolved oxygen during the experimental period in treatment-3**

#### iv. Treatment-4 (Mixture of Garlic & Amla Extract)

In treatment-4 (EG and EA) the average DO level was found  $(5.88 \pm 0.95)$  mg/L. Where the highest range was 8 mg/L and the lowest range was 4 mg/L throughout the research duration (Figure 25). Throughout the experimental period DO level fluctuated from (4.0 to 8.0 mg/L) in all treatments. The dissolved oxygen (DO) range was (5.9-7.6 mg/L); which was found from the experiment of Yusuf *et al.* (2015). Courtenay and Williams (2004) gave a conclusion that, *C. striata* can survive in the low dissolved oxygen level that is less than 5 mg/L. This fish is categorized as a fish which has an ability to take oxygen directly from the air. When the level of DO is in a stable position it is said that fish species are in a favorable environment for their proper growth and development. The dissolved oxygen level in these treatments medium during the experiment were in the optimal range. So, the findings of this study fully support the appropriate range of dissolved oxygen level for the culture of *C. striata*.



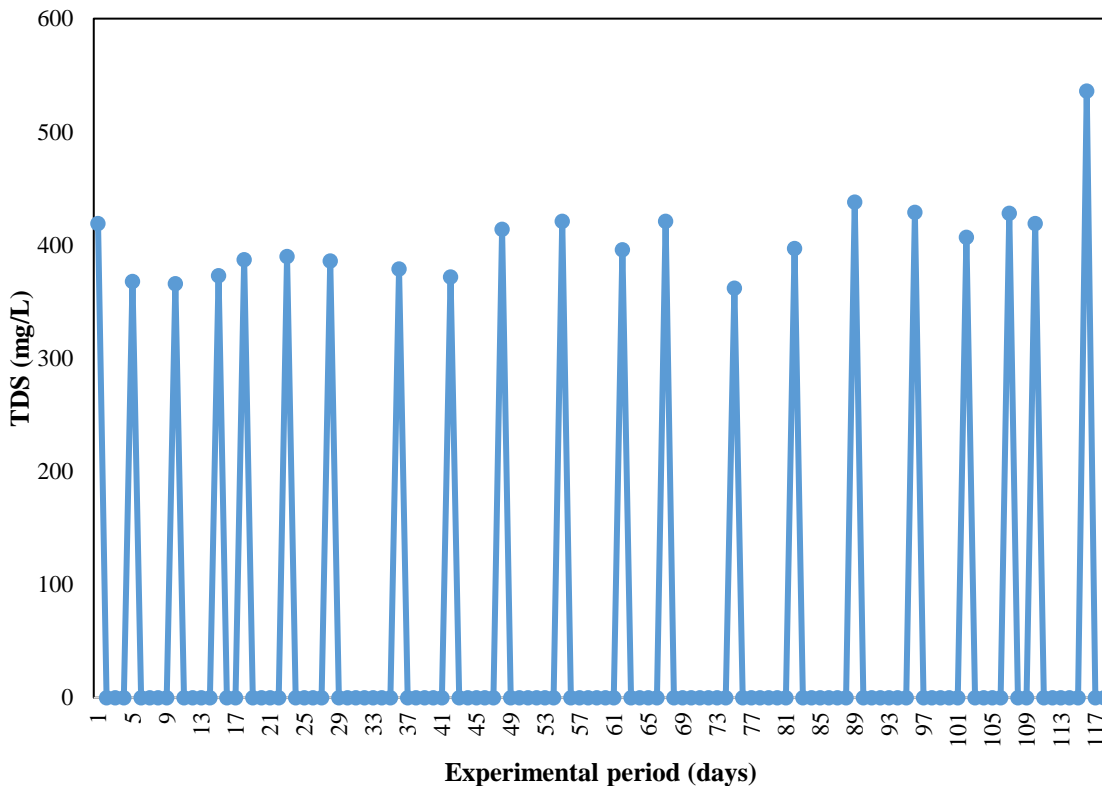
**Figure 25: Range of dissolved oxygen during the experimental period in treatment-4**



#### 4.1.5 Total Dissolved Solids (TDS)

##### i. Treatment-1 (Control)

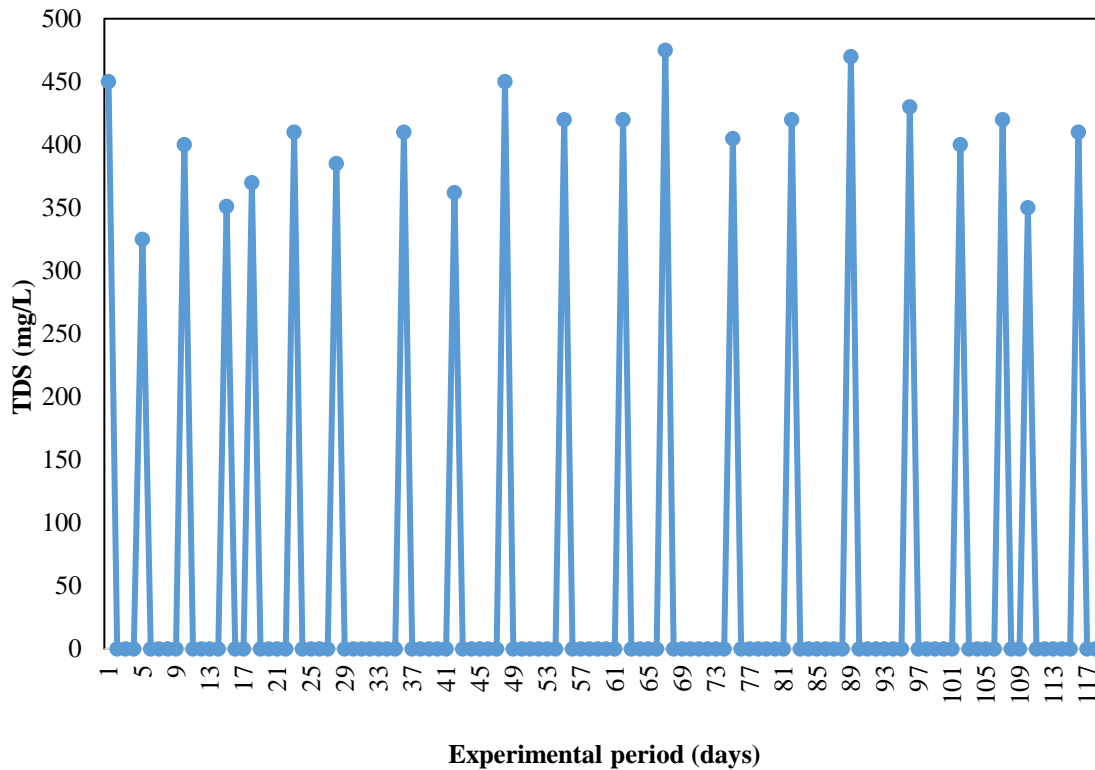
Total dissolved solids is a proportion of the dissolved combined content of all inorganic and organic substances present in a liquid on molecular, ionized or micro-granular suspended form. Generally, the functional definition is that the solids should be small enough to endure filtration through a filter with 2-micrometer (nominal size, or smaller) pores. Despite the fact that TDS is not generally considered a primary pollutant, it is utilized as an indication of the presence of a broad cluster of chemical contaminants. In this experimental work, the average total dissolved solids (TDS) was  $(405.14 \pm 38.03)$  mg/L in treatment-1 (control), where the highest range was 536 mg/L and the lowest range was 366 mg/L throughout the experimental duration. (Figure 26)



**Figure 26: Range of TDS during the experimental period in treatment-1 (Control)**

## ii. Treatment-2 (Extract of Garlic)

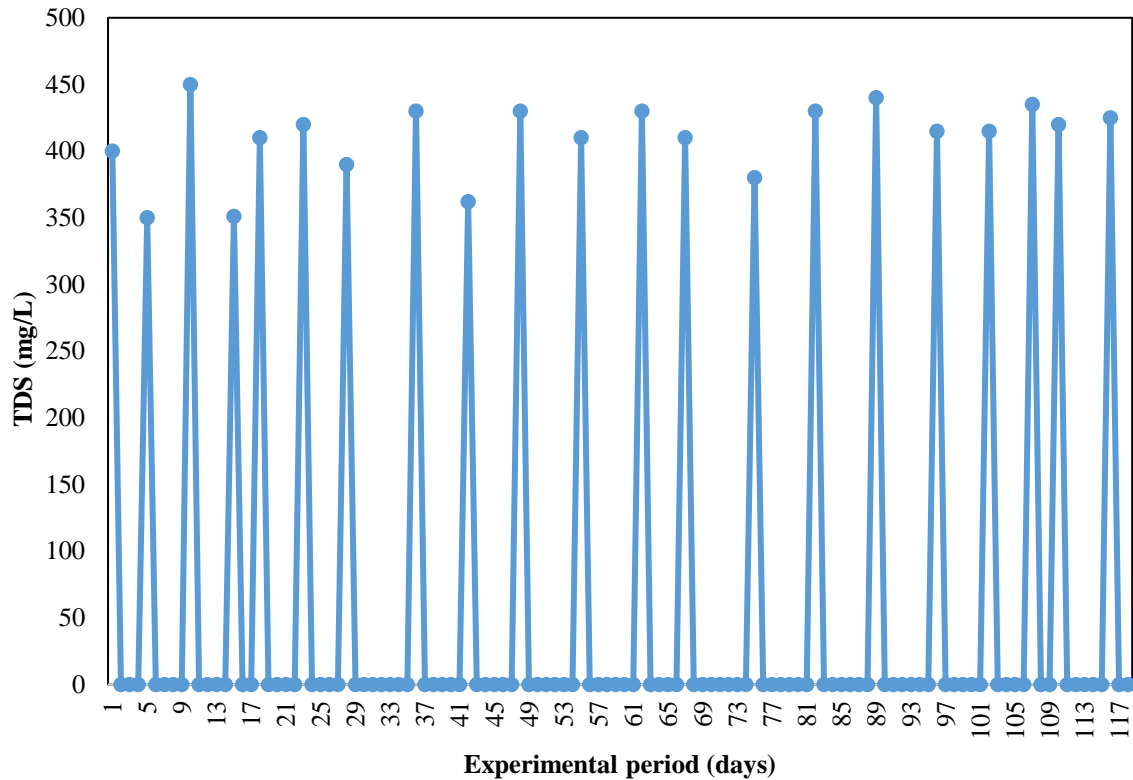
It was observed that, the average total dissolved solids (TDS) in treatment-2 (EG) was  $(406.33 \pm 39.12)$  mg/L, where the highest range was 475 mg/L and the lowest range was 330 mg/L during the experimental period. (Figure 27)



**Figure 27: Range of TDS during the experimental period in treatment-2**

## iii. Treatment-3 (Extract of Amla)

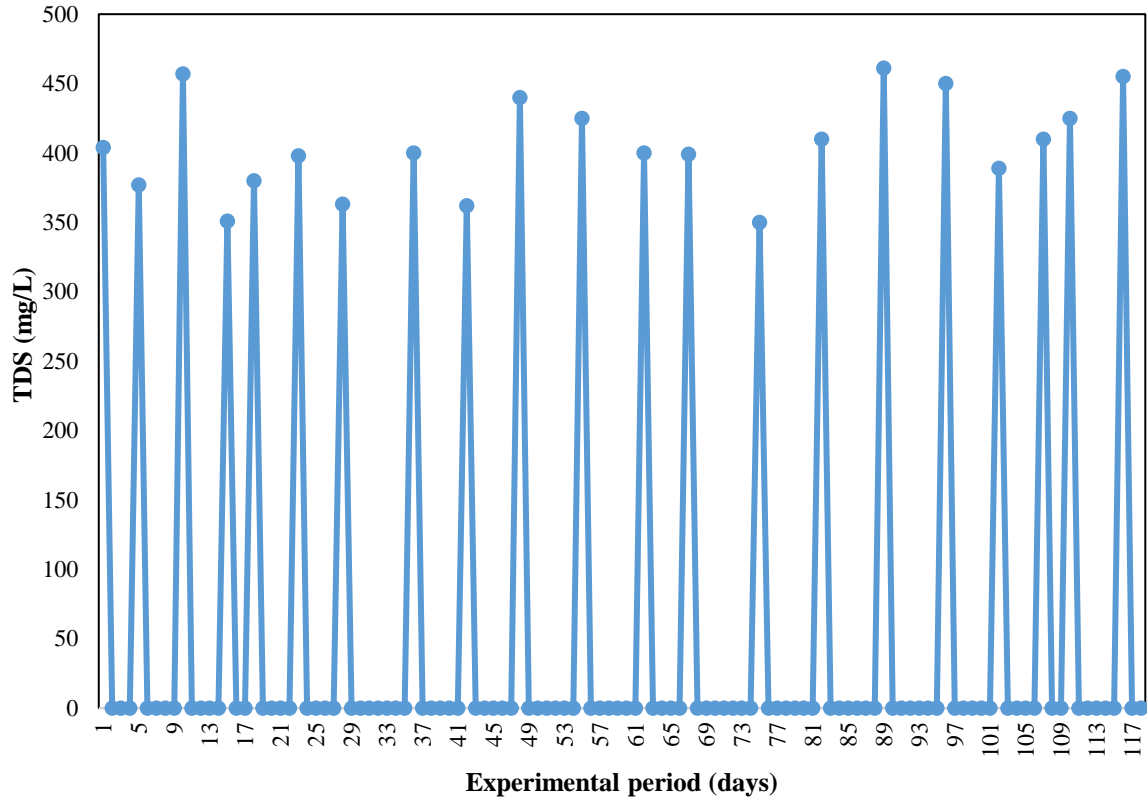
The average total dissolved solids (TDS) was  $(409.67 \pm 28.27)$  mg/L in treatment-3 (EA), where the highest range was 450 mg/L and the lowest range was 350 mg/L throughout the research duration. High levels of TDS which is above 500 mg/L implies it is unsuitable for utilization and causes several diseases. On the contrary, the utilization of low TDS water such as below 300 mg/L, normally occurring or received from a treatment process, does not bring about harmful impacts to the cultured species. (Figure 28)



**Figure 28: Range of TDS during the experimental period in treatment-3**

**iv. Treatment-4 (Mixture of Garlic and Amla Extract)**

The average TDS value in treatment-4 (EG and EA) was  $(405.05 \pm 34.49)$  mg/L. The highest range was 455 mg/L and the lowest range was 350 mg/L during the experimental period (Figure 29). Total dissolved solids level accepted as an ideal range when it is less than 500 mg/L, which was reported by Emerenciano *et al.* (2017). Throughout the experimental period, total dissolved solids level was below 500 mg/L, which denoted as an optimum level of TDS for the process of biofloc culture system. Balanced total dissolved solids level helps to keep fishes active in aquatic environment. Despite the fact that TDS values are fluctuated frequently but not more than 500 mg/L, which was a good sign of water quality. By comparing the findings of total dissolved solids level with the previous study, the range of TDS in this experiment was suitable for the growth and development of the cultured species.

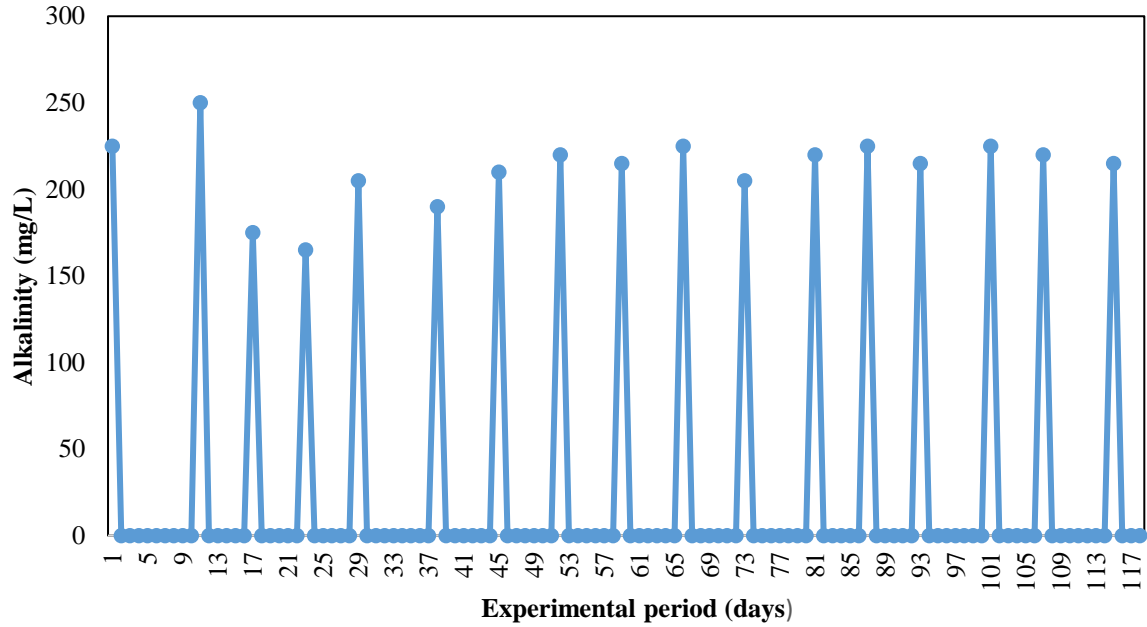


**Figure 29: Range of TDS during the experimental period in treatment-4**

#### 4.1.6 Alkalinity

##### i. Treatment-1 (Control)

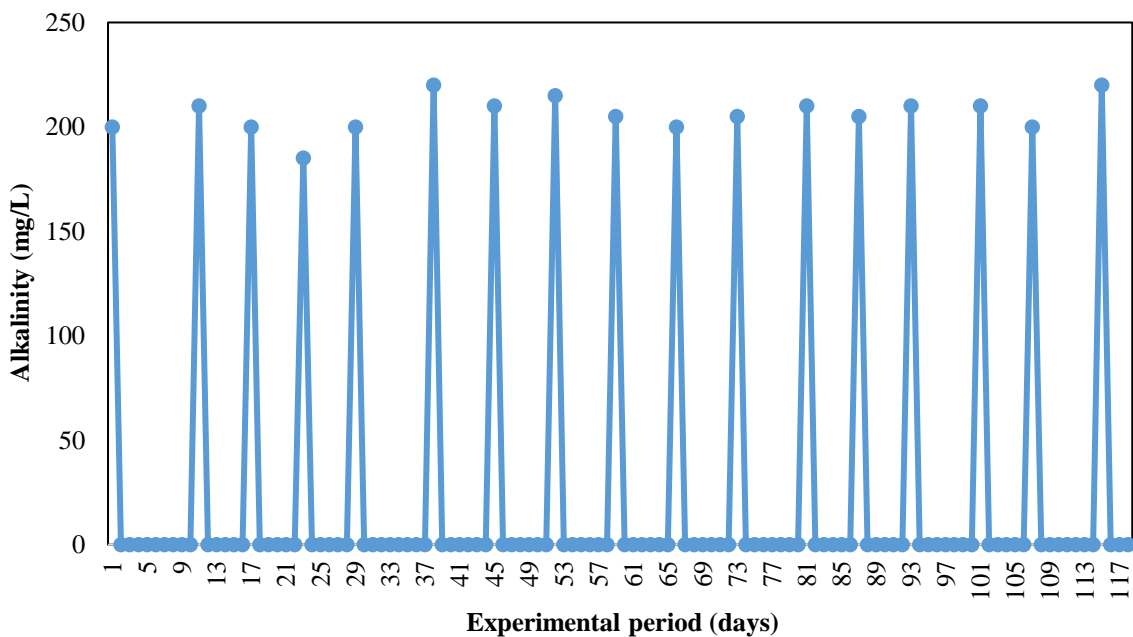
Alkalinity is the limit of water to resist acidification. Alkalinity is the strength of a buffer solution comprises of weak acids and their conjugate bases. Measuring alkalinity is significant in determining the water's capacity to neutralize acidic pollution. It is probably one of the best measures of the sensitivity of the water to acid inputs. In this experimental work, the average alkalinity range in treatment-1 (Control) was  $(212.06 \pm 20.16)$  mg/L. The highest range was 250 mg/L and the lowest range was 165 mg/L throughout the experimental duration (Figure 30). Saputra *et al.* (2018) reported a result that, the alkalinity range was from (86.43 - 101.84 mg/L) during their research period.



**Figure 30: Alkalinity during the experimental period in treatment-1 (Control)**

**ii. Treatment-2 (Extract of Garlic)**

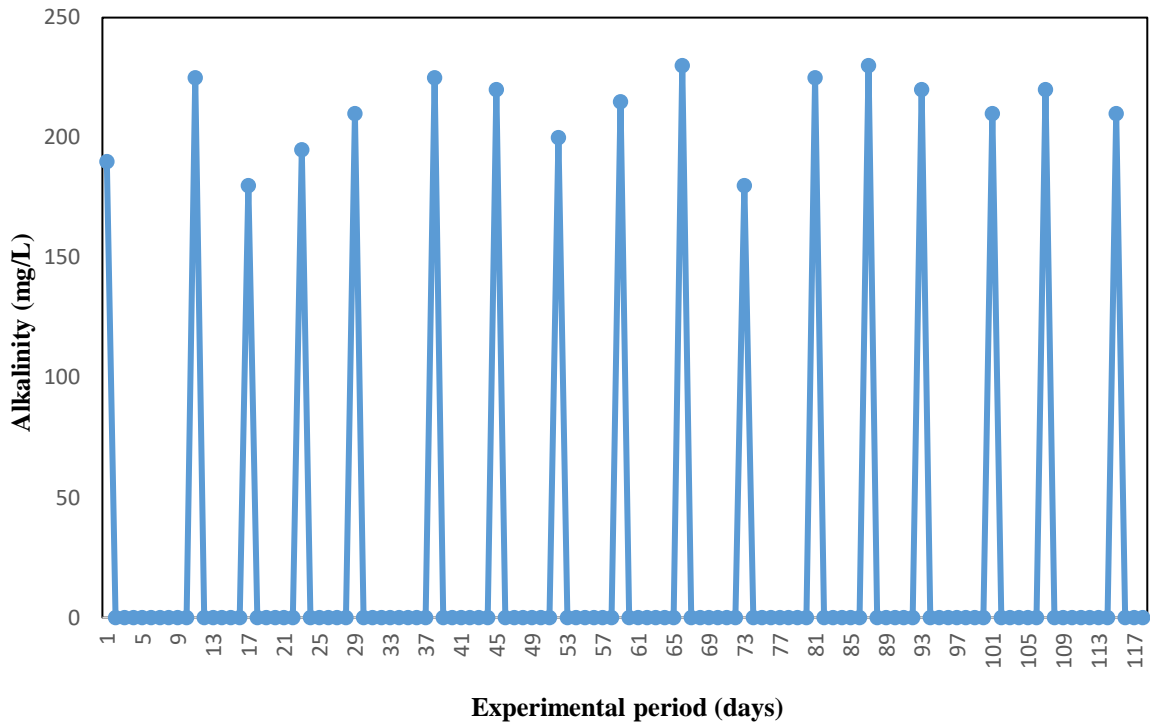
It was observed that, the average alkalinity range was  $(206.18 \pm 8.57)$  mg/L in treatment-2 (EG), where the highest range was 220 mg/L and the lowest range was 185 mg/L during the experimental period. (Figure 31)



**Figure 31: Alkalinity during the experimental period in treatment-2**

### iii. Treatment-3 (Extract of Amla)

The average alkalinity range was  $(210.88 \pm 16.42)$  mg/L in treatment-3 (EA). The highest range was 230 mg/L and the lowest range was 170 mg/L throughout the experimental duration (Figure 32). Alkalinity measures the aggregate amount of base present in the water and indicates the ability of water to resist large level of pH changes. That's why it is significant to measure the level of alkalinity of water after few days interval.

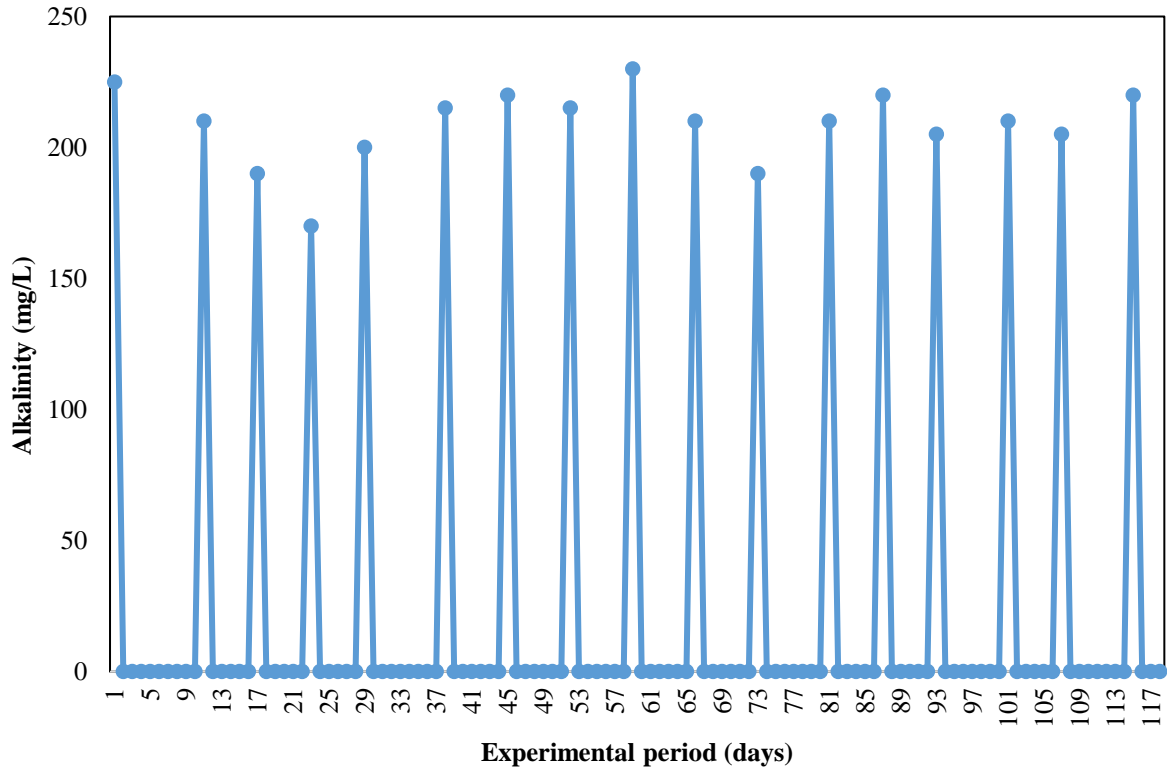


**Figure 32: Alkalinity during the experimental period in treatment-3**

### iv. Treatment-4 (Mixture of Garlic and Amla Extract)

In treatment-4 (EG and EA) the average alkalinity range was  $(208.53 \pm 14.77)$  mg/L, where the highest range was 240 mg/L and the lowest range was 165 mg/L during the experimental period (Figure 33). Anand *et al.* (2018) gave a conclusion that, the alkalinity ranged from (159 to 190 mg/L), (147 to 189mg/L) and (158 to 188 mg/L) in their experimental which was in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> tanks respectively. Emerenciano *et al.* (2017) reported that, the alkalinity level more than 100 mg/L is ideal for any water quality as well as for the culture of any aquatic

species. In the entire duration of this experiment, the alkalinity level varies from (160 to 250) mg/L in all treatments. By considering the alkalinity ranges with the aforementioned results it can state that the alkalinity level was favorable for the culture of *C. striata* in biofloc system.

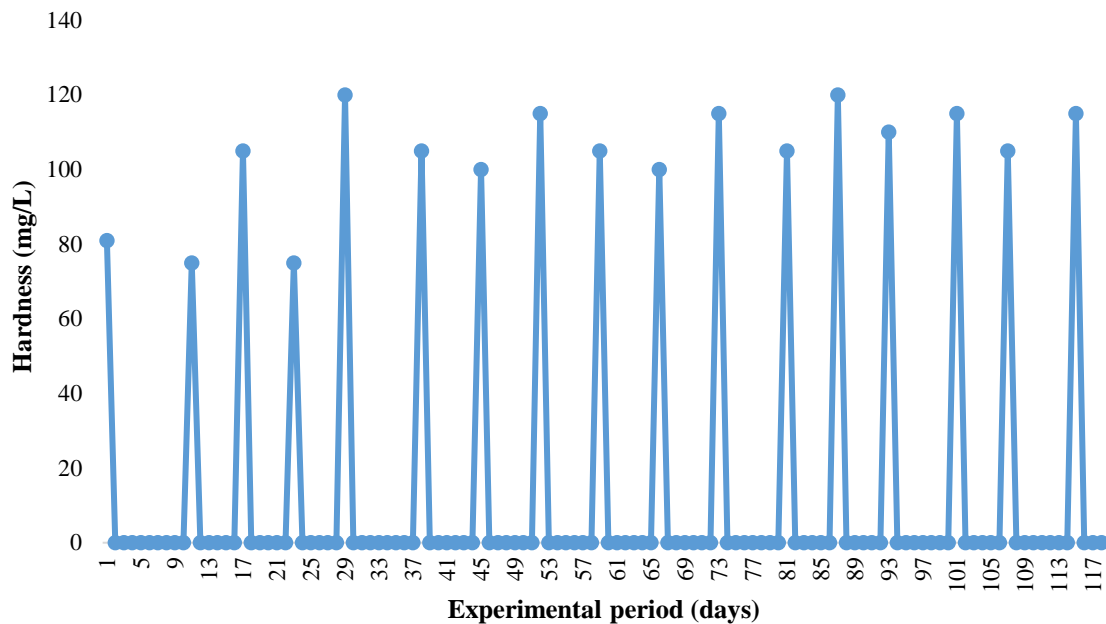


**Figure 33: Alkalinity during the experimental period in treatment-4**

#### 4.1.7 Hardness

##### i. Treatment-1 (Control)

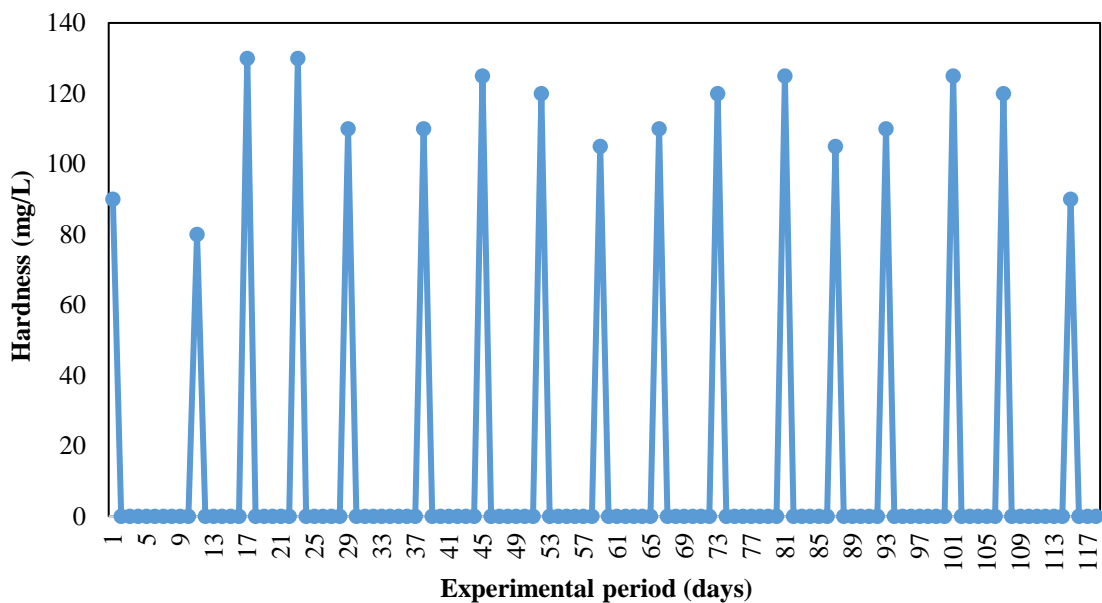
Hardness of water described as "hard" which contains high amounts of normally occurring dissolved calcium and magnesium. Absolute hardness is the amount of the calcium and magnesium concentrations, both expressed as calcium carbonate, in milligrams per liter (mg/L). Hardness occurs because of the presence of multivalent metal ions which come from minerals dissolved in the water. The average hardness range during this experiment was  $(103.88 \pm 14.32)$  mg/L in treatment-1 (control); where the highest range was 120 mg/L and the lowest range was 78 mg/L throughout the experimental duration. (Figure 34)



**Figure 34: Hardness during the experimental period in treatment-1 (Control)**

**ii. Treatment-2 (Extract of Garlic)**

The average hardness level during this experiment was  $(112.06 \pm 14.69)$  mg/L in Treatment-2 (EG); where the highest range was 130 mg/L and the lowest range was 80 mg/L during the experimental period. (Figure 35)

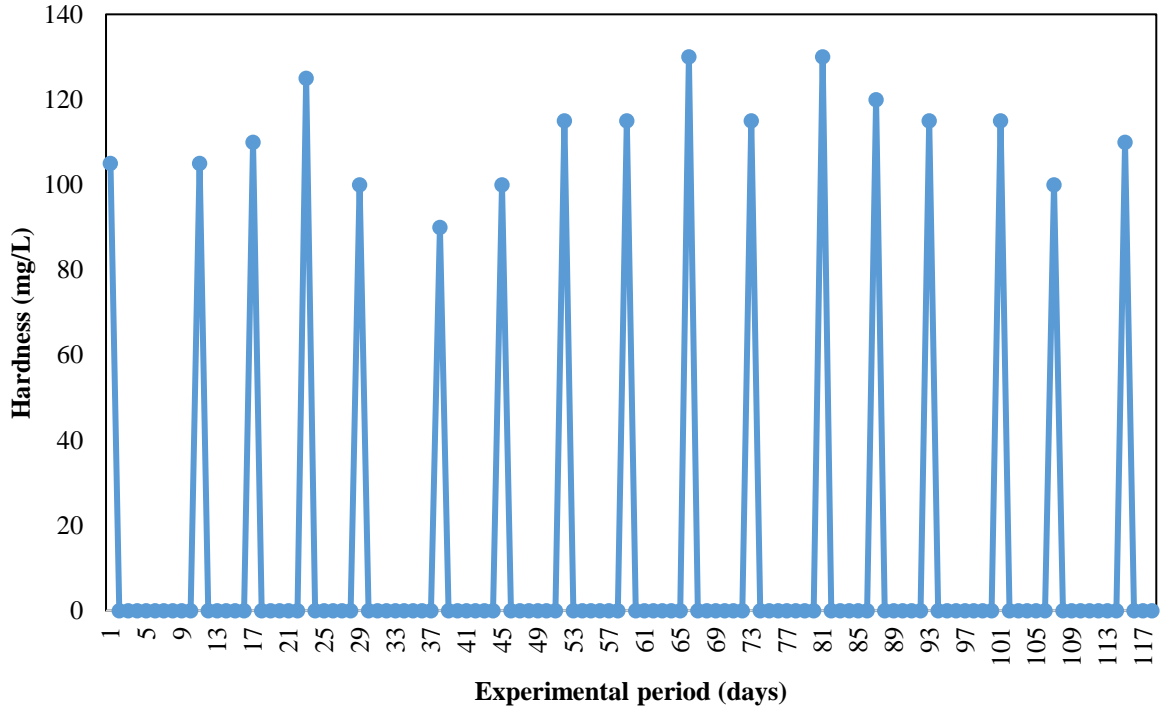


**Figure 35: Hardness during the experimental period in treatment-2**



### iii. Treatment-3 (Extract of Amla)

The average hardness range during this experimental period was found  $(111.76 \pm 11.03)$  mg/L in Treatment-3 (EA); where the highest range was 125 mg/L and the lowest range was 90 mg/L throughout the experimental duration. (Figure 36)

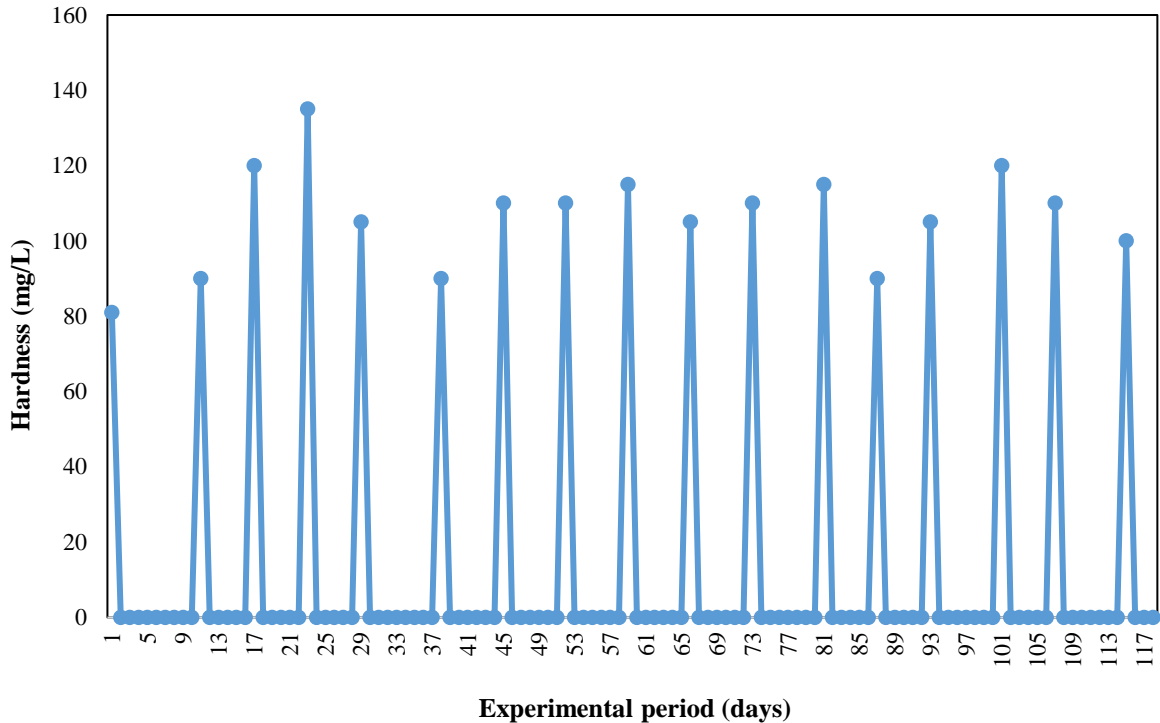


**Figure 36: Hardness during the experimental period in treatment-3**

### iv. Treatment-4 (Mixture of Garlic and Amla Extract)

In treatment-4 (EG and EA) the average hardness level was  $(106.53 \pm 13.43)$  mg/L; where the highest range was 137 mg/L and the lowest range was 81 mg/L throughout the experimental duration (Figure 37). The hardness level varies from (75 to 137) mg/L in all treatments throughout the experimental period. Sugumaran *et al.* (2018) gave a conclusion that, the impact of different diets on growth performance, length-weight relationship of *C. striata* about a ninety days feeding trial had a hardness level ranged from (80.01 to 81.15 mg/L). Generally the harder the water, the lower the harmfulness of other metals to aquatic life. Maximum time of this research work, hardness level was more than 100 mg/L during

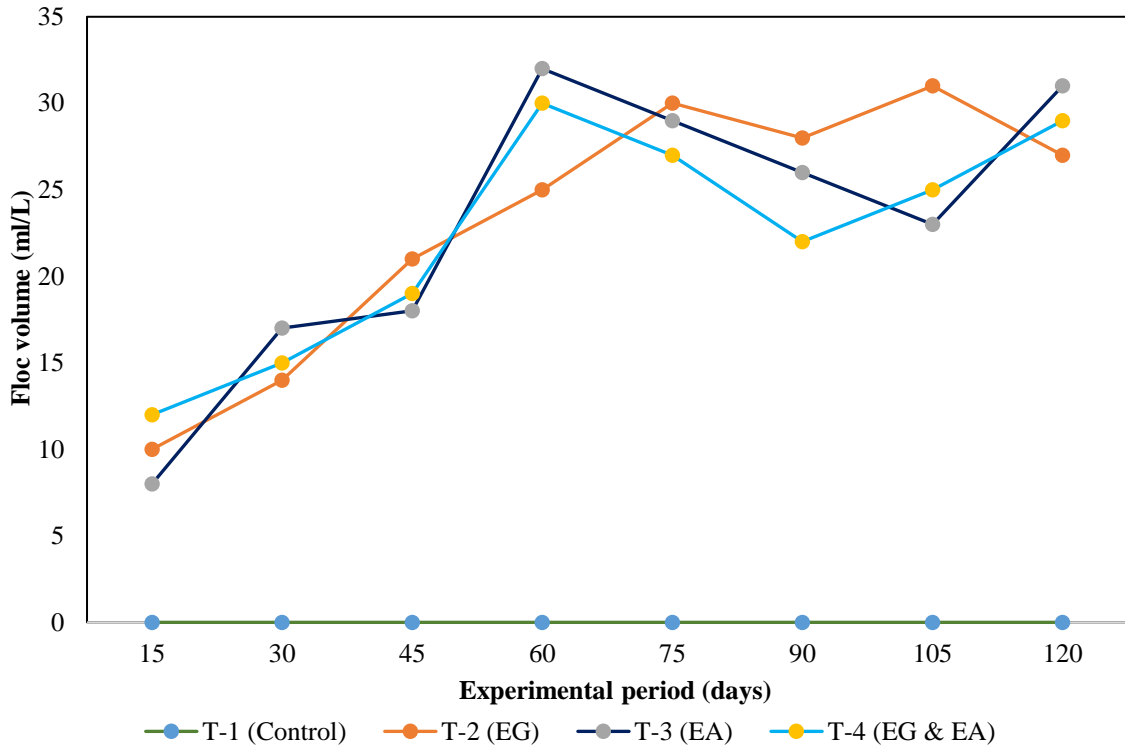
the experimental period in all treatments. So, considering the previous research work it is explicit that, the hardness level was optimal throughout the experimental duration.



**Figure 37: Hardness during the experimental period in treatment-4**

#### 4.1.8 Volume of Floc

Flocs are suspended growth in aquatic body consists of phytoplankton, algae, bacteria, protozoans and other kinds of particulate organic matter such as feces and uneaten feed. In T-2 (EG) volume of floc was ranged from (10 to 30) ml/L and in T-3 (EA), T-4 (EG and EA) the volume of floc ranged was from (8 to 32) ml/L and (12 to 28) ml/L respectively. The highest floc volume in T-2, T-3, T-4 was 30, 32 and 28 ml/L respectively (Figure 38). According to the study of López-Elías *et al.* (2015) mean volume of floc during their research period was 9.22 ml/l for T0, 8.67 ml/L for T10, 6.78 ml/l for T20, and 4.22 ml/L for T30, whereas the mean volume for the control treatment was 5.56 ml/L. Requirement of floc volume varies from species to species.



**Figure 38: Floc volume in all treatment tanks during the experimental period**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

## 4.2 Morphological Parameters of Snakehead Fish

Phenotypic parameter such as length, weight are most important for snakehead cultivation in biofloc method. In this experimental work, data were collected in every 15 days interval for assessing the progress of snakehead fish growth and probability of disease infestation. Treatment based observations are given in below:

### 4.2.1 Mean Length of Snakehead Fish

Organisms usually increase in size (length, weight) during development. The basic factors that influence the growth of fish are the quantity of food available, the number of fish utilizing same food source, temperature, oxygen and other water quality parameters besides the size, age and sexual maturity of the fish. Mean length of snakehead fish were recorded after every 15 days of interval. The length of snakehead fish (*C. striata*) during the experimental period in each treatments are given in the following table: (Table 2)

Table 2: Mean length of cultured snakehead fish (*C. striata*) reared in different treatments

Sampling phase	Sampling date	Mean length (cm)/Days			
		T-1 (Control)	T-2 (EG)	T-3 (EA)	T-4 (EG & EA)
Initial	23.08.20	6.71±0.49	6.71±0.49	6.71±0.49	6.71±0.49
1 <sup>st</sup>	07.09.20	8.57±0.79	9.79±0.73	9.68±0.82	9.86±0.69
2 <sup>nd</sup>	22.09.20	10.86±0.69	11.76±0.61	12.29±0.49	11.92±0.57
3 <sup>rd</sup>	07.10.20	11.14±0.69	13.14±0.89	12.87±0.93	12.72±0.94
4 <sup>th</sup>	22.10.20	12.14±0.69	13.75±0.73	13.47±0.76	13.86±0.69
5 <sup>th</sup>	07.11.20	14.43±0.79	15.36±1.12	15.86±1.07	15.15±1.16
6 <sup>th</sup>	22.11.20	15.57±0.98	17.09±0.83	16.94±0.88	17.43±0.79
7 <sup>th</sup>	07.12.20	17.57±0.79	18.83±0.78	19.29±0.72	19.86±0.69
8 <sup>th</sup>	22.12.20	18.86±0.69	20.41±0.72	20.23±0.75	20.86±0.69

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

When fishes were stocked for culture at the initial stage of this experiment the mean length was (6.71±0.49) cm. In first two weeks all treatments showed better results but after that period mean length was started getting better in treatment-2. But after five sampling period treatment-4 successively gave better length than other treatments (Table 2). After first three sampling, mean length rate was slightly decreased in treatment-1 and the length value was not satisfactory than other treatments where extracts were applied with feed. The comparison of mean length of several treatments that was collected on eight sampling schedule in every fifteen days of interval. In this experiment it was clearly seen that treatment-4 gave better length than control and other treatments. From stocking day containing 6.71±0.49 cm of *C. striata* juvenile reached into 20.86±0.69 cm which was the highest length value among all treatments. The mean length was better in those treatments where extracts were used. After final sampling the highest mean length was observed in treatment-4 comparing all sampling data. Sagumaran *et al.* (2018) reported that, initial average length of *C. striata* was 10.69 (cm) and final average length was 23.32 (cm) during their research period. Muntaziana *et al.* (2013) gave a conclusion that, the final average

lengths of fry (*C. striata*) fed with trash fish, bloodworm and *Acetes* shrimps were  $7.91 \pm 0.23$ ,  $7.28 \pm 0.23$ ,  $7.21 \pm 0.17$  cm respectively and the experiment was conducted for about twenty five days.



Plate 01: Juvenile stage at stocking



Plate 02: The highest mean length after 15 days in treatment-4



Plate 03: The highest mean length after 30 days in treatment-3



Plate 04: The highest mean length after 45 days in treatment-2





Plate 05: The highest mean length after 60 days in treatment-4



Plate 06: The highest mean length after 75 days in treatment-3



Plate 07: The highest mean length after 90 days in treatment-4



Plate 08: The highest mean length after 105 days in treatment-4



Plate 09: The highest mean length after 120 days in treatment-4

#### 4.2.2 Mean Weight of Snakehead Fish

The following mean weight was recorded on the basis of random sampling in every 15 days of interval during the experimental period. Total eight sampling was done during the experimental timeframe.

Table 3: Mean weight of cultured snakehead fish (*C. striata*) reared in different experimental treatments

Sampling phase	Sampling date	Mean weight (g)/Days			
		T-1 (Control)	T-2 (EG)	T-3 (EA)	T-4 (EG & EA)
Initial	23.08.20	4.29±0.49	4.21±0.42	4.32±0.55	4.43±0.53
1 <sup>st</sup>	07.09.20	10.71±1.60	11.02±1.39	10.97±1.42	11.14±1.35
2 <sup>nd</sup>	22.09.20	15.52±3.02	21.76±2.14	20.58±1.69	23.43±2.44
3 <sup>rd</sup>	07.10.20	23.57±2.37	32.43±3.64	31.27±3.69	34.71±3.49
4 <sup>th</sup>	22.10.20	40.43±7.14	50.75±12.96	53.46±12.38	58.29±11.48
5 <sup>th</sup>	07.11.20	51.52±6.84	65.82±6.42	67.57±6.33	70.71±6.32

6 <sup>th</sup>	22.11.20	62.37±3.35	82.65±10.71	78.92±10.74	84.57±10.63
7 <sup>th</sup>	07.12.20	71.29±4.57	94.17±12.67	89.63±13.69	96.71±12.59
8 <sup>th</sup>	22.12.20	86.14±5.67	102.49±7.33	98.78±7.44	110.10±7.18

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

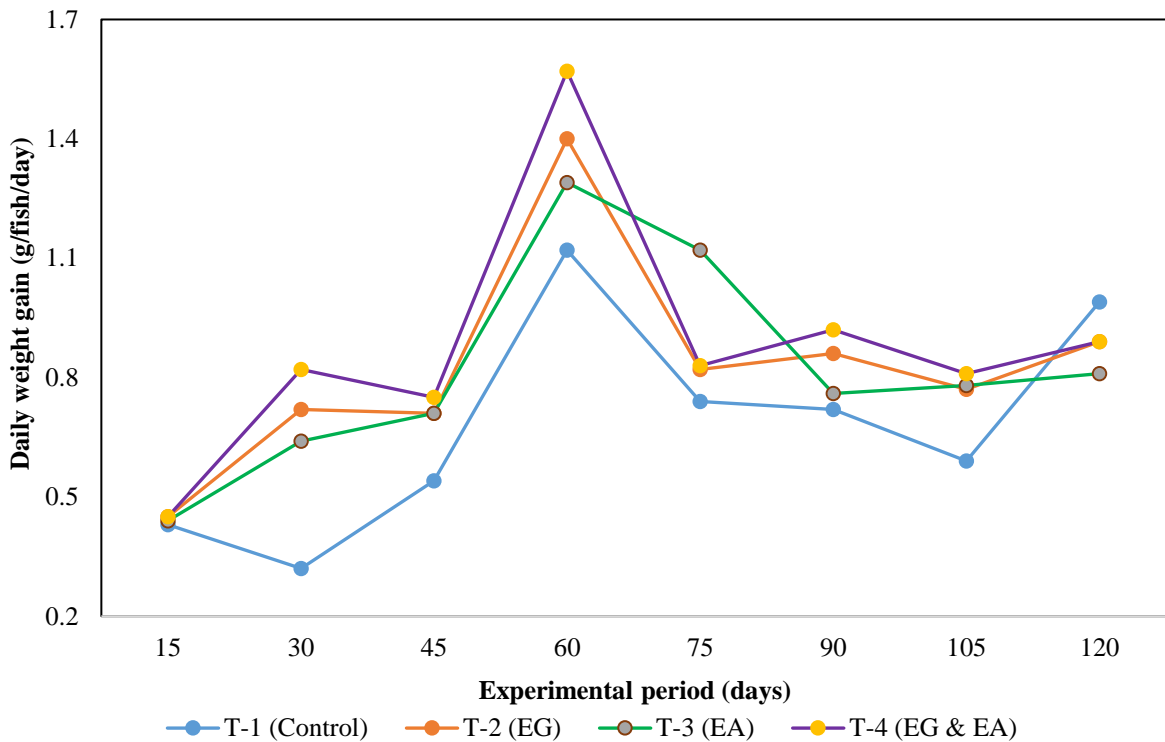
At the beginning of the experiment apparently mean 4 gm snakehead fry was stocked for culture. In first two weeks treatment-1 gave better results which was almost identical with other treatments. But after that mean weight was started getting better in those treatments where extracts were used with feed. Weight rate was slightly lower in treatment-2 and treatment-3 than treatment-4. Minimum weight rate can be caused due to adverse environmental condition, less rate of feed intake and so on. The minimum weight was seen in control treatments where stocking weight was 4.29±0.49 g and final sampling weight was 86.14±5.67 g obtained after 120 days (Table 3). According to the study of Sagumaran *et al.* (2018) initial average weight was 11.02 (g) and final average weight was 23.92 (g) where the experimental period was about ninety days. By applying the extracts of garlic (*Allium sativum*) and amla (*Embllica officinalis*) the tendency of appetite for feed increased and that's why the cultured species took feed as much as double quantity than the control treatments. By comparing the results of weight in this study it is clear that treatment-4 gave better results than control and other treatments as well. From the initial day of treatment-4 containing 4.43±0.53 g of *C. striata* juvenile reached into 110.10±7.18 g in 8<sup>th</sup> phase (Table 3). The rate of weight in treatment-4 was almost identical at the beginning of the study but as much as the day increased it breaks all records comparing with other treatments.

#### 4.2.3 Daily Mean Weight Gain

Daily mean weight gain was counted in the interval of every sampling period, where mean weight gain was higher in the middle part of the experimental period. In T-1 (control) the highest daily mean weight gain rate was 1.1g where it was decreased after that day but after 105 days weight gain rate was slightly higher. After completing 60 days, the average daily weight gain rate was seen in better condition in T-4 and T-2 and the rate was 1.50g, 1.38g respectively (Figure 39). The daily mean weight gain was 0.84g in BFT and 1.22g in



control treatments which was according to the study of Ekasari *et al.* (2012). Daily weight gain fluctuation is normal. In most cases weight was fluctuated for various reasons such as environmental factors, disease infestation possibilities and feeding requirements. Weight gain progress depends on feeding rate in case of optimum range of water quality factors and fish health conditions. Proper feeding rate also give better outcome. Although the fluctuation of weight gain it is clear that T-4 gave excellent result compared to other treatments.



**Figure 39: Daily mean weight gain of cultured snakehead fish (*C. striata*)**

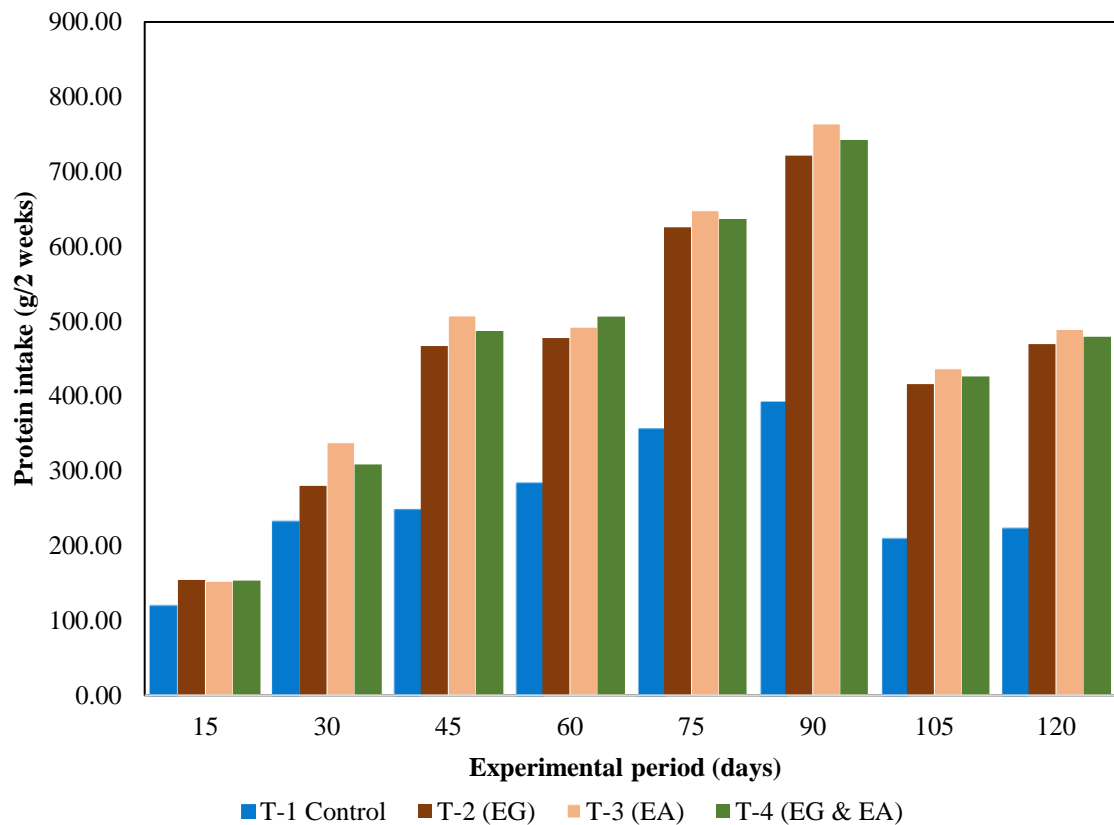
(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

#### 4.2.4 Weekly Nutrition Intake

##### i. Protein intake

Nutrition from feed are prerequisite for proper growth rate of any particular species. In snakehead fish culture, feed with better protein content helps to grow quickly and viable survival rate. In biofloc technology, less amount of feed is required. But it doesn't hamper on nutritional requirements because fish take protein, lipid by consuming floccules. In this

experiment, the highest average protein intake rate was  $(720.10 \pm 12.18)$  g/sampling which was in treatment-3 tanks (Figure 40). Rate of protein intake were almost same in every sampling period in those tanks where extracts of garlic and amla were applied. According to the figure: 40 it can be state that, among all sampling stages protein intake rate was higher after 90 days of sampling period. Ghaedi *et al.* (2019) reported that, highest weight gain was recorded in fish fed protein at 450 g/kg diet which was  $(289.7 \pm 5.5)$  g, trailed by fish fed protein at 400 g/kg diet and that was  $(250.3 \pm 4.0)$  g also at 350 g/kg diet protein intake rate was  $(230.3 \pm 3.5)$  g at the end of the experiment. In a study by Desilva and Radampolak (1990) on the effect of varying protein levels on growth and fecundity in *Oreochromis niloticus*, the optimal protein level for growth of both male and female was 300 g/kg. However, the greatest percentage of spawned females occurred among those fed 250 or 300 g/kg protein.

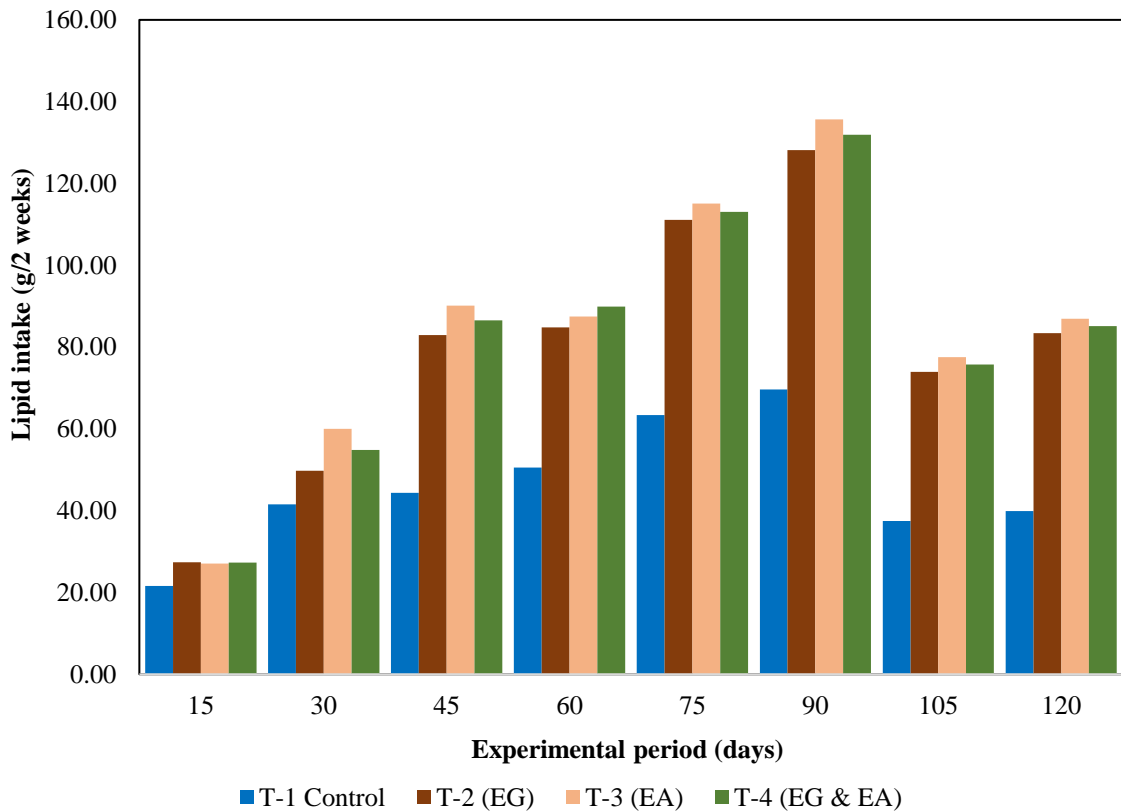


**Figure 40: Protein intake during the experimental period**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

## ii. Lipid intake

Lipids are the major source of metabolic energy for growth and reproduction of fish which was stated by Henderson *et al.* 1984; Sargent *et al.* 1989. Lipid contains cholesterol and phospholipids which are needed for fishes in all stages of the development. In this experimental work, average highest (62.85±2.02)g lipid intake occurred in treatment-1 (control) and in treatment-2, treatment-3 and treatment-4 highest average lipid intake rate was (126.74±2.41), (136.37±2.32), (130.64±2.38)g respectively. Like protein the lipid intake rate was highest in those treatments where garlic and amla extracts were applied. By applying these extracts with feed the appetite for taking feed was increased day by day. For that reason fishes consumed more feed and as a result lipid intake rate was highest in treatment-2, treatment-3 and treatment-4 than control treatment. (Figure 41)



**Figure 41: Lipid intake during the experimental period**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

### 4.3 Growth Parameters of Snakehead Fish (*C. striata*)

Growth is a fundamental biological response including external ecological and internal physiological status. In case of fish survival rate, weight gain, length increase, feed conversion ratio, protein intake, lipid intake, specific growth rate etc. are considered as growth parameters. In this experimental work, the data which were recorded from snakehead fish (*C. striata*) culture are given in below: (Table: 4)

Table 4: Growth parameters (Mean) of snakehead fish (*C. striata*) in biofloc technology

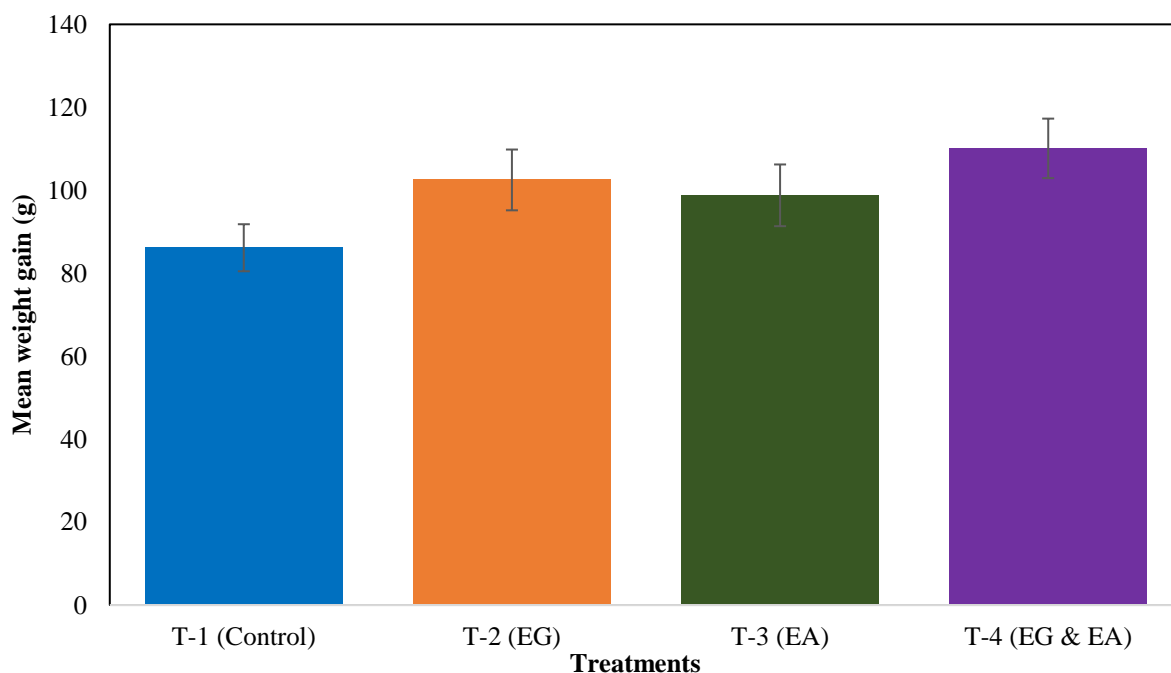
Parameters	T-1 (Control)	T-2 (EG)	T-3 (EA)	T-4 (EG & EA)
Initial Weight (g)	4.29±0.49	4.21±0.42	4.32±0.55	4.43±0.53
Final Weight (g)	86.14±5.67	102.49±7.33	98.78±7.44	110.10±7.18
Mean Weight Gain (g)	81.85±5.18	98.28±6.91	94.46±7.46	105.67±6.65
No. of Initial Fish	165	165	165	165
No. of Fish Survived	71	108	125	118
Total Weight Gain (g/m <sup>3</sup> )	5427.6±15.6	10614.3±13.6	11807.5±9.3	12204.6±8.7
Weight Gain (%)	1907.9±14.9	2334.4±12.6	2186.6±16.7	2385.3±15.3
Initial Length (cm)	6.71±0.49	6.71±0.49	6.71±0.49	6.71±0.49
Final Length (cm)	18.86±0.69	20.41±0.72	20.23±0.75	20.86±0.69
Mean Length Increase (cm)	12.15±0.2	13.70±0.23	13.52±0.26	14.15±0.2
Survival Rate (%)	47.88±1.9	65.45±2.3	75.76±0.8	71.52±1.4
Specific Growth Rate (%)	2.50±0.2	2.66±0.1	2.61±0.2	2.70±0.2
Feed Conversion Ratio	1.36±0.1	1.13±0.1	1.17±0.1	1.08±0.1
Gross Production (kg/m <sup>3</sup> /crop)	6.1±0.7	11.7±0.9	12.5±0.4	13.2±0.2
Gross Production (kg/m <sup>3</sup> /year)	18.3±0.4	35.1±0.8	37.5±0.5	39.6±0.6
Net Production (kg/m <sup>3</sup> /crop)	5.9±0.03	9.72±0.02	11.15±0.06	11.38±0.04

Net Production (kg/m <sup>3</sup> /year)	11.8±0.07	19.44±0.04	22.3±0.13	22.76±0.08
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(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

### 4.3.1 Final Weight

This experiment was started with juvenile *C. striata* consisting the initial weight were (4.29±0.49), (4.21±0.42), (4.32±0.55), (4.43±0.53)g and after accomplishing the experiment (after 120 days) final mean weight of individual snakehead fish were (86.14±5.67), (102.49±7.33), (98.78±7.44) and (110.10±7.18)g in treatment-1, treatment-2, treatment-3 and treatment-4 respectively (Figure 42). Final mean weight was significantly higher in treatment-4 and there has a significant differences ( $P < 0.05$ ) in final mean weight among the treatments. Ekasari *et al.* (2012) reported that, the final mean weight of BFT was 160.31g and in control treatment the mean final weight was 199.16 g which is according to the study of red tilapia *Oreochromis sp.* cultured at different stocking densities in biofloc technology for 14 weeks. The result of this present study showed that, final mean weight was better where extracts were used than the control treatments.

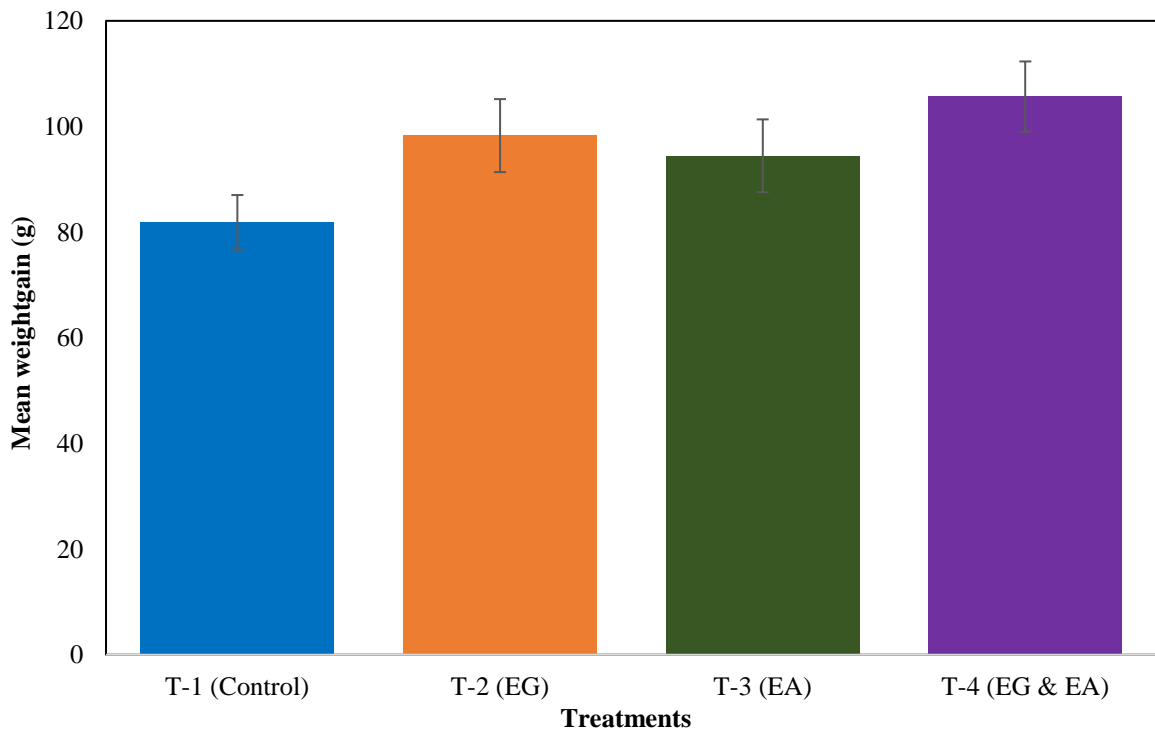


**Figure 42: Final mean weight of cultured snakehead fish (*Channa striata*)**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

### 4.3.2 Mean Weight Gain

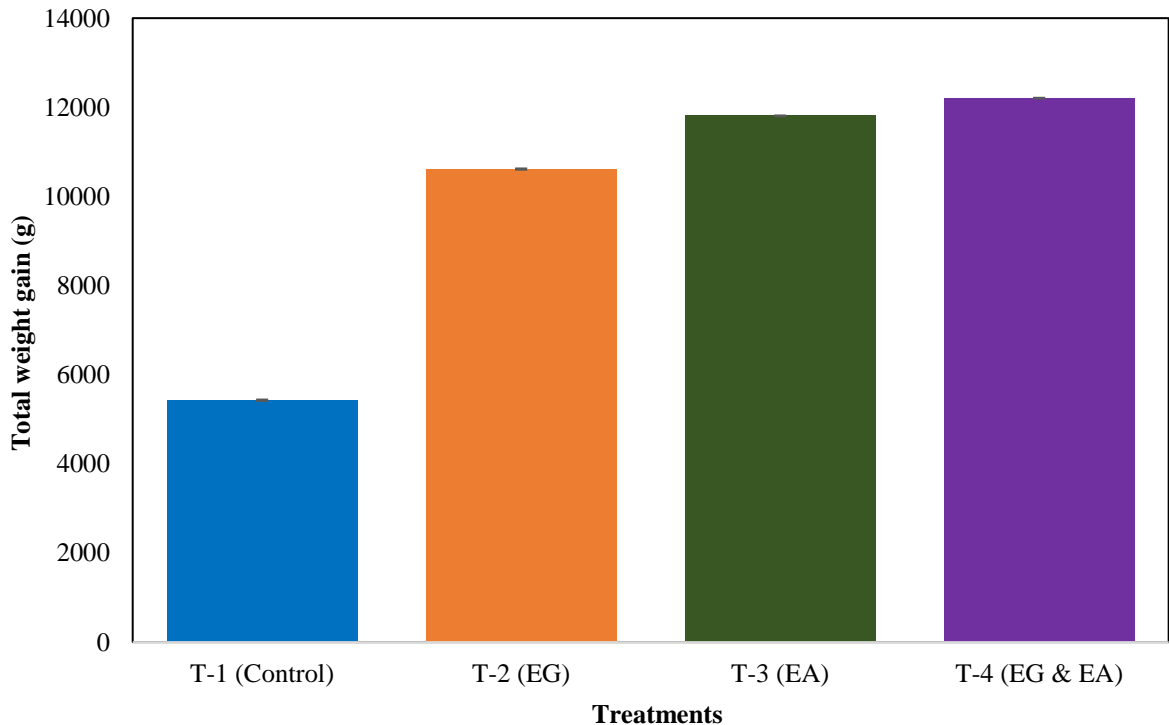
The mean weight gain of individual fish in treatment-1 (control) was  $(81.85 \pm 5.18)$ g and in treatment-2, treatment-3 and treatment-4 mean weight gain were  $(98.28 \pm 6.91)$ ,  $(94.46 \pm 7.46)$  and  $(105.67 \pm 6.65)$ g respectively during 120 days of the experimental period (Figure 43). The mean weight gain was significantly better in treatment-4 and there has a significant differences ( $P < 0.05$ ) among control and other treatments. In case of other treatments the outcome was satisfactory where extracts were used than control treatment. War et al. (2014) gave a conclusion that fish fed with *D. carinata* and *M. micruna* showed highest weight gain (15.24 mg and 13.56 mg) respectively after 30 days of experiment. The highest weight gain ( $231.18 \pm 0.03$  mg) was recorded in fishes fed with Diet-3 (*T. decipiens* and *C. carnuta*) which was significantly different from Diet-1 (copepod, *Thermocyclops decipiens*) ( $171.23 \pm 0.01$  mg) and Diet-2 (cladoceran, *Ceriodaphnia cornuta*) ( $183.22 \pm 0.05$  mg) in combination, for a period of four weeks; which was accordance by the report of Paray et al. (2015).



**Figure 43: Mean final weight gain of cultured snakehead fish (*Channa striata*)**  
(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

### 4.3.3 Total Weight Gain

In control treatments, the total weight gain was (5427.6±15.6)g, whereas in treatment-2 total weight gain was (10614.3±13.6)g and in treatment-3, treatment-4 it was (11807.5±9.3)g and (12204.6±8.7)g respectively (Figure 44). From the figure it is clear that treatment-4 gave excellent outcome among other treatments and by analyzing the result of these treatments a significant differences ( $P < 0.05$ ) of total weight gain was found. In those treatments where extracts were used the total weight gain rate was almost double than the control treatments. Which indicates that, the growth rate was better where mixture of garlic (*Allium sativum*) and amla (*Embllica officinalis*) extracts were applied with the feed. By using these extracts cultured species were not infected in any kind of disease and they took feed in a high quantity that was almost double than the cultured species in control treatments. Much feed intake rate and better metabolism capacity were noticeable in those treatments where extracts were used and those factors were influenced them (*C. striata*) to grow in a significant rate than control treatments.

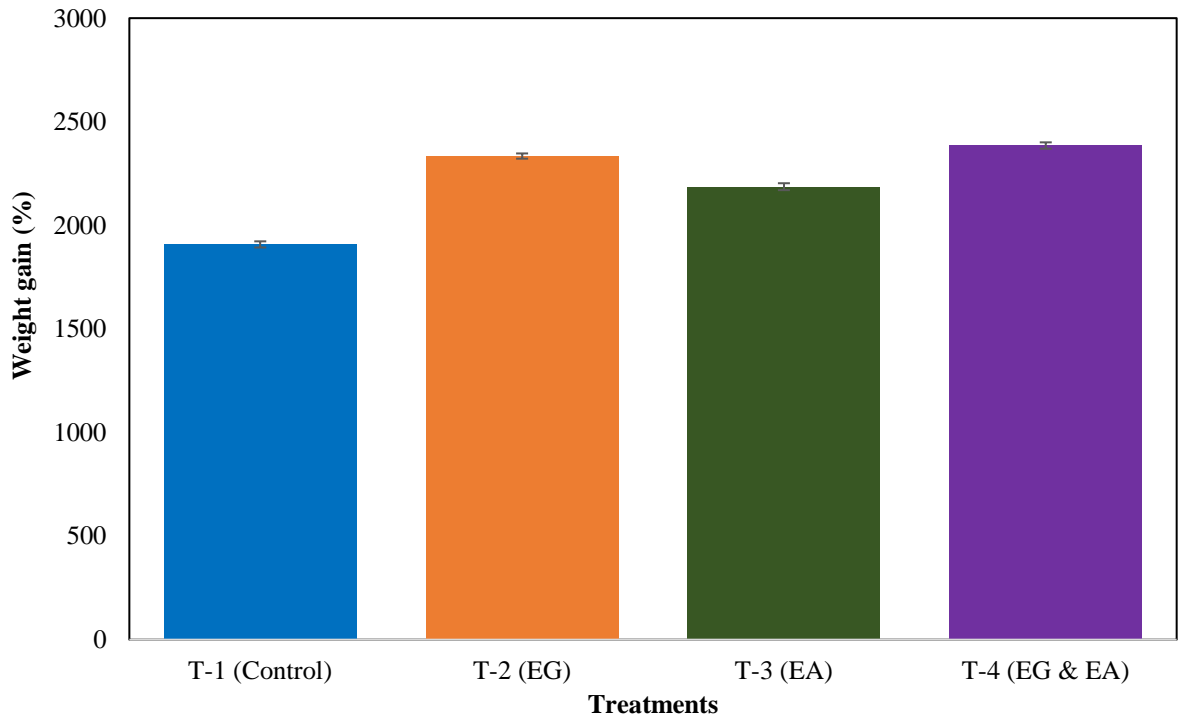


**Figure 44: Total weight gain of cultured snakehead fish (*Channa striata*)**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

#### 4.3.4 Percentage Weight Gain

In this study, the percentage weight gain of snakehead fish cultured in treatment-1 (Control) was (1907.9±14.9%) and (2334.4±12.6%), (2186.6±16.7%), (2385.3±15.3%) were found in treatment-2, treatment-3 and treatment-4 respectively (Figure 45). Best weight gain percentage was in treatment-4 (EG and EA) and least percentage was in treatment-1 (Control). Applying both extracts of garlic and amla in treatment-4 gave best weight gain percentage than control and other treatments. The percentage weight gain rate was almost similar in those treatments where extracts were used. There has a significant differences ( $P < 0.05$ ) of percentage weight gain among all treatments. Muntaziana *et al.* (2013) was conducted a study to assess the growth and survival rate *C. striata* fry fed with three types of fresh foods (bloodworm, trash fish, *Acetes* shrimps) for about twenty five days feeding experiment where highest weight gain percentage was (376.50%). So, from the value it is clearly understandable that the percentage weight gain was excellent where extracts were used compared to control treatments.



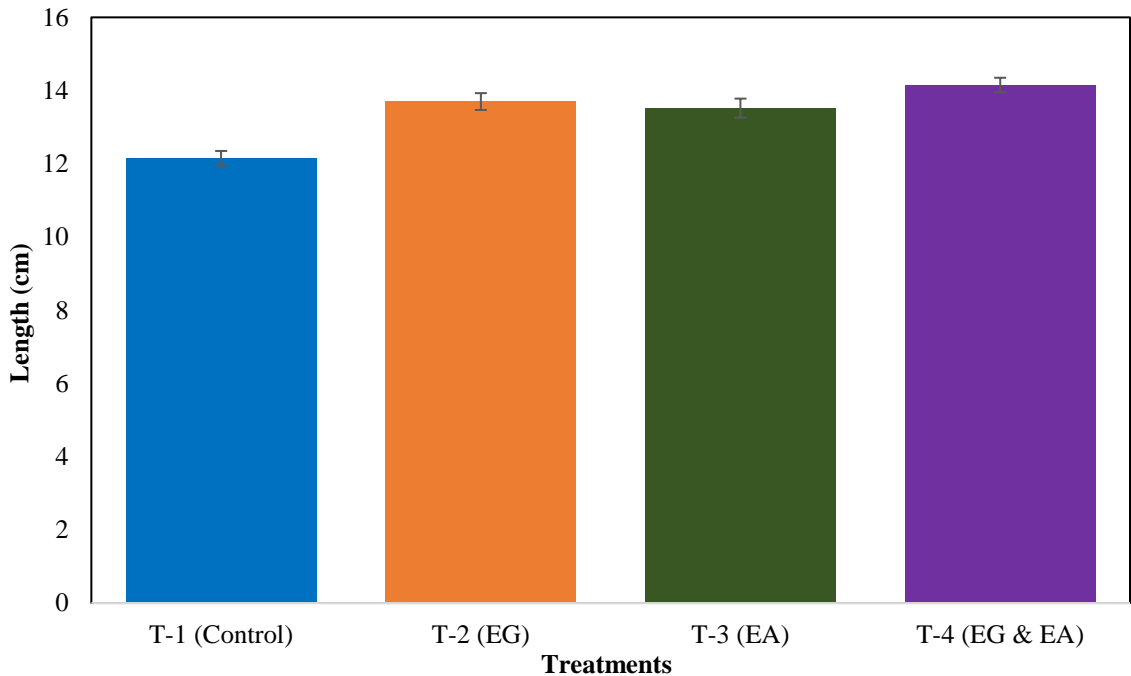
**Figure 45: Percentage weight gain of cultured snakehead fish (*C. striata*)**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)



### 4.3.5 Mean Length Increase

The mean length increased in treatment-1 (control) was (12.15±0.2) cm, on the other hand in treatment-2, treatment-3 and treatment-4 the length were (13.70±0.23), (13.52±0.26) and (14.15±0.2) cm respectively (Figure 46). Mean length increased of *C. striata* showed no significant differences ( $P < 0.05$ ) among those treatments which were used for the experimental purpose. Fish fed with *C. cornuta* and *M. micrura* showed better length increased (27.72 mm and 26.50 mm) respectively after 30 days of experiment which is accordance to the report of War *et al.* (2014). Paray *et al.* (2015) reported that, among three types of live feed diets (Diet 1 - copepod, *Thermocyclops decipiens*; Diet 2 - cladoceran, *Ceriodaphnia cornuta* and Diet 3 - T. *decipiens* and *C. carnuta*) fed by *C. striata* the highest increased total length was (30.46±0.01 mm) and that was recorded by fed Diet-3, which was significantly different from Diet-1 (26.66±0.14 mm) and Diet-2 (22.24±0.006 mm). Increased of length was almost identical where extracts were used and among them highest length increased was recorded from treatment-4 where both garlic and amla extracts were used than control and other treatments.

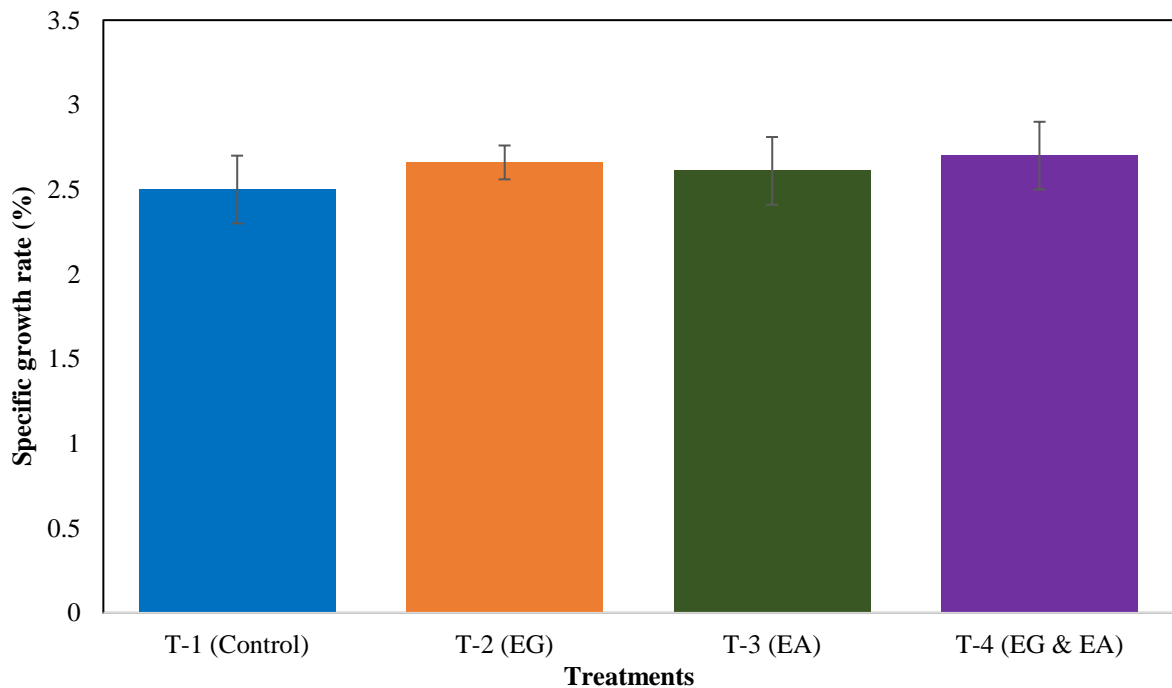


**Figure 46: Length increase of cultured snakehead fish (*Channa striata*)**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

#### 4.3.6 Specific Growth Rate

Specific growth rate was ( $2.50\pm 0.2\%$ ) in treatment-1 (control) whereas in treatment-2, treatment-3 and treatment-4 the specific growth rate were ( $2.66\pm 0.1\%$ ), ( $2.61\pm 0.2\%$ ) and ( $2.70\pm 0.2\%$ ) respectively (Figure 47). Among the treatments the specific growth rate was significantly higher than control and other treatments ( $P < 0.05$ ). From the present study, comparing all treatments the specific growth rate was highest in treatment-4 where both extracts of garlic and amla were applied. Puspaningsih *et al.* (2019) conducted a study on water quality, hematological parameters and biological performances of Snakehead fish (*C. striata*) reared in different stocking densities where they reported that among three types of treatment specific growth rate was highest (2.80%) in treatment-2 and lowest (2.15%) in treatment-3. Muntaziana *et al.* (2013) showed that the best specific growth rate (SGR) value was (6.24%) which found from the study about on effect of selected diets on the growth and survival rate of snakehead fish (*C. striata*) fry. So the findings of this experiment is fully supportable if we compare the results with the aforementioned studies.

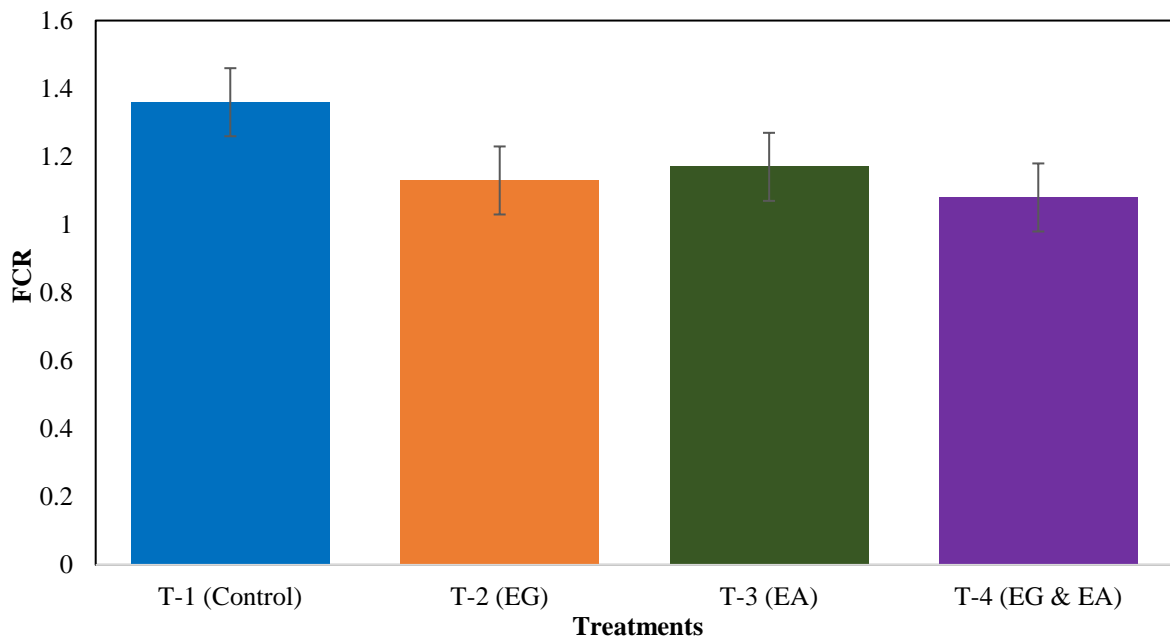


**Figure 47: Specific growth rate of cultured snakehead fish (*Channa striata*)**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

#### 4.4 Feed Conversion Ratio (FCR)

Feed conversion ratio is a ratio measuring of the efficiency with which the bodies of cultured species convert applied feed into the desired output. In control treatment of the present work the FCR was  $(1.36\pm 0.1)$  and in experimental treatments where extracts of garlic (*Allium sativum*) and amla (*Emblica officinalis*) were used the FCR were  $(1.13\pm 0.1)$ ,  $(1.17\pm 0.1)$  and  $(1.08\pm 0.1)$  in treatment-2, treatment-3 and treatment-4 respectively (Figure 48). Among all treatments treatment-4 gave significantly remarkable rate than control and other treatments ( $P < 0.05$ ). In the experimental tanks where flocs were available the cultured species (*C. striata*) took their desired nutrients from biofloc as well as beneficial microorganisms and that's why feed conversion ratio was better than control treatment. Yusuf *et al.* (2015) suggested that the highest FCR  $(0.91\pm 0.01)$  which was found by them in treatment-C. Putra *et al.* (2019) reported FCR 1.45, 0.85, 1.05 and 1.08 for treatment A, B, C and D respectively. In the study of Sangeeta *et al.* (2017), highest FCR was found (3.11) and lowest (2.10). The value of FCR of present experiment is better than aforementioned studies and that will encourage the farmers as well as in commercial level for biofloc based culture system.

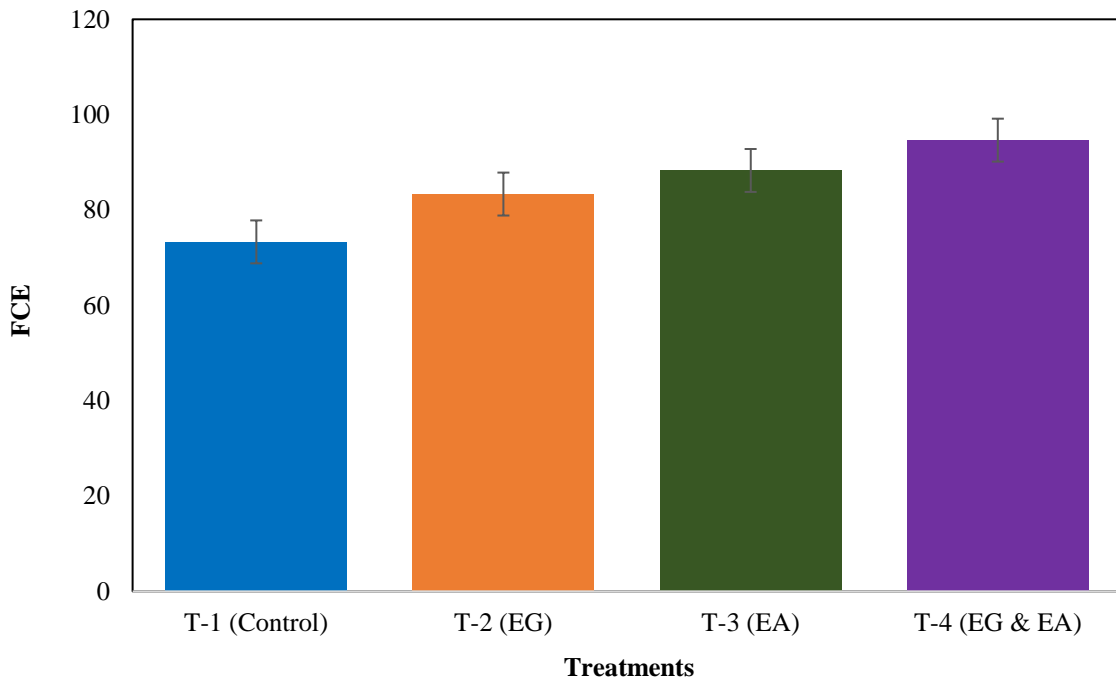


**Figure 48: Feed conversion ratio (FCR) of the experimental treatments**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

#### 4.5 Food Conversion Efficiency (FCE)

Food conversion efficiency means the capacity of any organisms such as fish to convert the feed which they consume to its body flesh or weight. It is the effectiveness of a feed is normally estimated by the sum necessary to produce a unit weight of fish. In this present research, food conversion efficiency was better in treatment-4 which was 94.65 and lowest FCE was 73.31 in treatment-1 which was control. Among all treatments FCE percentage in treatment-2 and treatment-3 were 83.34 and 88.29 which was almost nearby between each other (Figure 49). By considering the results of all treatments best outcome of food conversion efficiency was found in treatment-4 where garlic and amla extracts were applied in an identical manner.



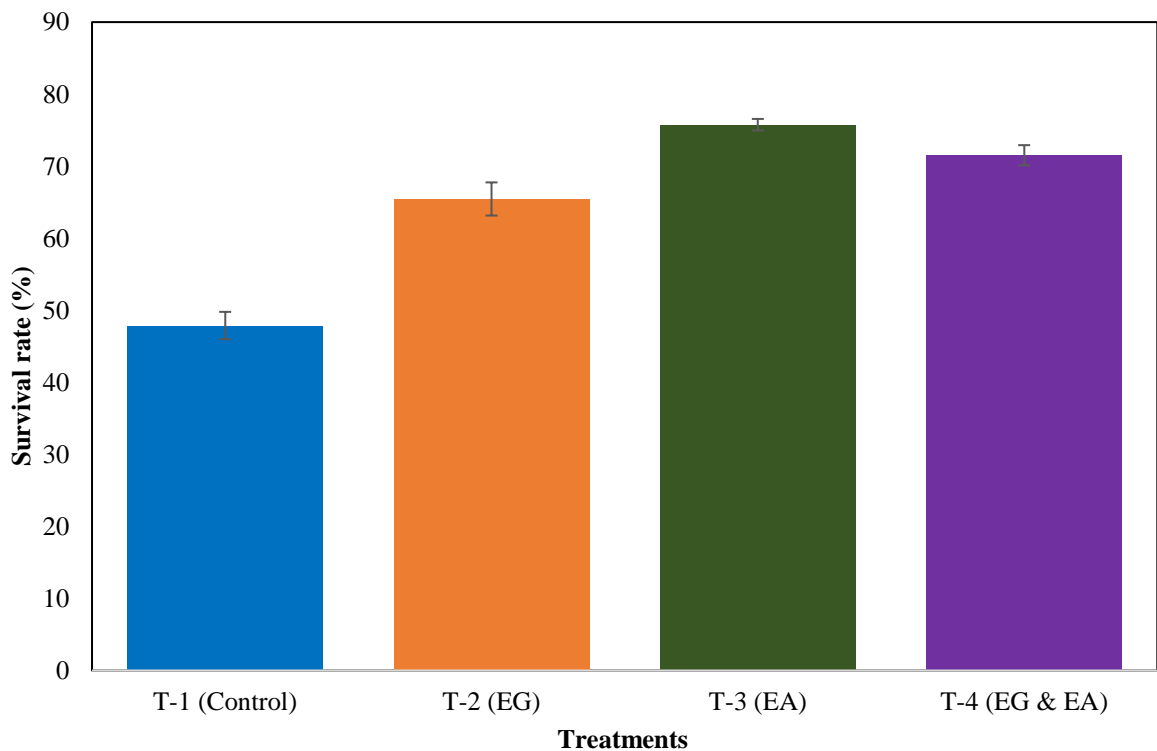
**Figure 49: Feed conversion efficiency (FCR) of the experimental treatments**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

#### 4.6 Survival Rate

Survival rate is a crucial factor for fish culture as well as in biofloc system. It indicates the live organism's percentage after accomplishing the research work duration. In this present work, the survival rate in treatment-1 (control) was  $(47.88 \pm 1.9)\%$  and in treatment-2,

treatment-3 and treatment-4 survival rate were  $(65.45 \pm 2.3)$ ,  $(75.76 \pm 0.8)$  and  $(71.52 \pm 1.4)\%$  respectively (Figure 50). Survival rate was significantly better ( $P < 0.05$ ) in treatment-3 as well as other treatments where extracts were applied than control treatments. In control treatments the survival rate was remarkably low because of the absence of biofloc and that's why water parameters level was fluctuated in a huge level which was dangerous. Putra *et al.* (2019) found that, survival rate of their experiment was 61.11% for treatment A and 92.22%, 82.22% and 84.4% for treatment B, C and D respectively. The highest survival rate was 84% in treatment-2 and lowest 60.67% in treatment-3 which was reported by Puspaningsih *et al.* (2019). The higher survival rate was (73.89%) and lowest survival rate was (54.44%) which was from the research work of Djokosetiyanto *et al.* (2017). Compared with aforementioned studies the survival rate was better in those treatments where extracts were applied which helps to develop the immunity level of *C. striata* than control treatments.



**Figure 50: Survival rate of cultured snakehead fish (*Channa striata*)**

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla)

## 4.7 Nutritional Value

Nutritional value denotes contents of food and the impact of constituents on body. It incorporates protein, fat, carbohydrates, fiber, ash, additives, enzymes, vitamins etc. Nutritional value as part of food quality is the evaluation of well-balanced ratio of the essential nutrients in terms of food or diet concerning the nutrient requirements of their consumers. The nutritional value of applied feed, microbial floc and cultured species (*C. striata*) are given in below:

### 4.7.1 Proximate Composition of Feed

The feed we applied during the experimental period contained 35% crude protein, 9.89% ash, 6.51% crude fiber, 42.85% carbohydrate and 5.33% fat. Moisture content was 11.01% and 88.99% dry matter content were available in the feed which we applied to the cultured species (Table 5). By comparing the analysis of proximate composition of feed with the aforementioned research it can be state that the nutritional value of feed was optimal to provide proper nutrition to the cultured species (*C. striata*). The proximate analysis of diets where crude protein level ranged from 34.65 to 36.80% got best results in growth performance as well as fat content ranged from 1.68 to 2.03%, fiber level ranged from 8.36 to 8.49% and ash content ranged from 5.48 to 6.13% containing feed should be provided which was reported by Sugumaran *et al.* (2018).

Table 5: Proximate composition of applied feed

Parameters	Nutritional composition
Moisture	11.01
Dry Matter	88.99
Crude Protein	35
Total Ash	9.89
Acid Insoluble Ash	0.42
Crude Fiber	6.51
Crude Fat	5.33
Carbohydrate	42.85

#### 4.7.2 Proximate Composition of Floc

Microbial floc is an essential component in biofloc culture system. In this present study, protein content level of floc was 28.34%, ash content was 6.77%, 47.1% carbohydrate, fiber and fat content was 8.18% and 9.61% respectively (Table 6). Ash, lipid and protein content level of floc were (33.0-40.4%), (2.6–3.5%) and (23.7–25.4%) respectively which were according to the report of López-Elías *et al.* (2015). The findings have similarities with the final statement of Yusuf *et al.* (2015) who stated that, protein content of floc from treatment C was (34.06±1.70), A and B were (31.99±2.70) and (31.81±1.51) respectively. To fulfill the demand of daily nutritional requirements and to enhance the growth the percentage of crude protein is crucial. By comparing the results of previous studies with present experiment it is clear that the result of proximate composition of floc was satisfactory.

Table 6: Proximate composition of microbial floc

Parameters	Nutritional composition
Moisture	78.21
Dry Matter	21.79
Crude Protein	28.34
Total Ash	6.77
Acid Insoluble Ash	Nil
Crude Fiber	8.18
Crude Fat	9.61
Carbohydrate	47.1

#### 4.7.3 Nutritional Composition of Cultured Snakehead Fish

Nutritional composition are detailed sets of information on the nutritionally important components of foods and provide values for energy and nutrients including protein, carbohydrates, fat, ash and other important food components such as fiber. These

components are essential in our daily diets. The chemical composition of any edible organism is very crucial. Prior to developing and implementing effective intervention projects to improve nutrition at the population level, it is imperative to know the nutritional situation of the target group. The nutritional composition of the fish varied from species to species and even within the same species. The nutritional composition of cultured species (*C. striata*) is given in below:

Table 7: Proximate composition (% on DM basis) of *C. striata* cultured in biofloc technology (Mean)

Parameters	T-1 (Control)	T-2 (EG)	T-3 (EA)	T-4 (EG & EA)
Moisture	72.05±1.92	74.31±3.2	72.57±0.56	73.35±0.7
Dry matter	27.95±1.92	25.69±3.2	27.43±0.56	26.65±0.7
Crude Protein	64.76±1.29	61.84±1.49	65.37±1.13	69.67±0.68
Total Ash	10.77±0.35	9.94±0.43	11.76±0.25	9.05±0.37
AIA	Nil	Nil	Nil	Nil
Crude Fiber	0.39±0.02	0.61±0.06	0.53±0.08	0.59±0.03
Crude Fat	17.41±0.25	15.58±0.33	17.18±0.27	18.15±0.22
Carbohydrate	6.67±0.75	12.03±1.2	5.16±0.78	2.54±3.64

(EG= Extract of Garlic, EA= Extract of Amla, EG & EA= Extract of Garlic and Amla, DM= Dry Matter and AIA= Acid Insoluble Ash)

The proximate analysis of cultured snakehead fish helps to understand the difference of nutritional value from biofloc and traditional culture system. In this present study, the highest crude protein was found 69.67±0.68% in treatment-4 and lowest was 61.84±1.49% in treatment-2. Highest ash, fiber, fat were 11.76±0.25% in treatment-3, 0.61±0.06% in treatment-2 and 18.15±0.22% found in treatment-4 respectively. On the other hand, lowest ash, fiber, fat content were 9.05±0.37% in treatment-4, 0.39±0.02% in treatment-1 and 15.58±0.33% in treatment-2. The highest carbohydrate content was 12.03±1.2% in



treatment-2 and lowest value was  $2.54 \pm 3.64\%$  in treatment-4 (Table 7). Protein content was found of *C. striata* which was 78.64% according to the study of Muntaziana *et al.* (2013). From the study of Sugumaran *et al.* (2018) the crude protein level ranged from 34.65 to 36.80%. According to the research of Sangeeta *et al.* (2017) the ash content was found which was about 14.70%. Ash content ranged from 5.48 to 6.13% which was from the experimental work of Sugumaran *et al.* (2018). Muntaziana *et al.* (2013) reported that, the fiber content of their cultured snakehead fish (*C. striata*) was 2.07%. According to the research work of Muntaziana *et al.* (2013) lipid content was about 8.26%. Sugumaran *et al.* (2018) reported (1.68 to 2.03%) fat content from their experimental species. According to the previous research works it is evident that the nutritional composition of cultured species (*C. striata*) was optimum.

## CHAPTER 5

### SUMMARY AND CONCLUSION

As the population of the world is increasing day by day the need for fish protein is also increasing correspondingly. Vertical production of fish is necessary for densely populated country like Bangladesh. Biofloc is a new technology for the vertical production of fish and which has the possibility and potentiality. The experimental work was conducted on “Biofloc culture method for growth performance, survival rate and nutritional composition of snakehead fish (*Channa striata*) on natural extract supplement”. Snakehead fish become a vital component of global aquaculture both in terms of quantity and value. Due to numerous cultivable attributes such as high market demand, fast growth rate and its compatibility to survive in an adverse water condition. To achieve the sustainable development goals biofloc technology (BFT) offers benefits in improving aquaculture production by using microbial biotechnology. This technology may result in higher productivity with less impact to the environment. The species (*C. striata*) was selected for this experiment because it has been well known and demandable fish species in our country. Hence, the investigation was undertaken to evaluate the potentiality of culture (*C. striata*) in biofloc technology to sustain and increase the production ability by using less property of lands.

There were four treatments used in this experiment with three replications. Maintaining the water quality parameters is a prerequisite for this kind of study like Biofloc Technology (BFT). Parameters of water quality that are monitored on a daily basis include temperature, dissolved oxygen, pH, alkalinity, ammonia and so on. For evaluating the growth performance and nutritional composition in this experiment we used garlic (*Allium sativum*) and amla (*Emblica officinalis*) extracts which were derived from the raw products by using a biotechnological process which increased the immunity level of cultured species. In biofloc based culture, the volume of microbial floc requirement varies from species to species. No floc was observed in the control treatment whereas 10 to 30 ml/L microbial floc was observed among the rest of the treatments.

The length and weight performance of cultured species (*C. striata*) were noteworthy in the treatments where extracts were used as compared to control treatments. The nutritional

composition of *C. striata* was significantly better in extracts used treatments where highest crude protein percentage (69.67%) was found in treatment-4 (EG and EA). Likewise other nutritional compositions were better in experimental treatments than the control treatments. Where garlic (*Allium sativum*) and amla (*Emblica officinalis*) extracts were applied the total weight gain in those treatments were double than the control treatments. The best specific growth rate (2.70%) was found in treatment-4 (EG and EA). Feed conversion ratio helps to estimate the quantity of feed that will be required in the growing process. The highest FCR was estimated  $1.08 \pm 0.1$  in treatment-4 and lowest was  $1.36 \pm 0.1$  in treatment-1. The highest protein intake was found  $(720.10 \pm 12.15)$ g/sampling and lipid intake rate was highest  $(136.37 \pm 2.32)$ g in treatment-3 (EA).

The Biofloc Technology (BFT) is a framework that maximizes aquaculture productivity by using microbial biotechnology to increase the adequacy and utilization of fish feeds, where toxic materials such as nitrogen components are treated and converted to a useful product, like a protein for using as supplementary feeds to the fish. Moreover, the microorganisms in Biofloc Technology (BFT) play a significant role in biosecurity by inhibiting the pathogenic microorganisms growth. From the results of this experiment, application of garlic (*Allium sativum*) and amla (*Emblica officinalis*) extracts in treatment-4 provided the best outcome in case of growth performance and nutritional value.

### **Recommendation**

It can be state that, snakehead fish can be cultured in this advanced culture system with the help of microbial biotechnology. The result of the experiment can play a crucial role in developing a sustainable aquaculture and an instruction for upcoming researcher and a recommendation for farmer who are interested in this technology in Bangladesh as well as all over the world. So this technique needs to be developed by more researches to keep up and maintain this systems in the future.

## CHAPTER 6

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