INTRODUCTION

CHAPTER 1

INTRODUCTION

Bangladesh is blessed with vast inland water bodies. Being a country of rivers and floodplains, fish plays a very important role in the daily life of many people in Bangladesh. Historically people depended mainly on natural waters for supply of fish but as a result of declining catches of wild fish due to an increased fishing pressure by the growing population as well as environmental degradation, people began to culture fish in enclosed waters. At present, aquaculture has been expanding both vertically and horizontally as pond fish culture and crustacean (shrimp and freshwater prawn) farming offer tremendous potential. A number of species is now cultured including crustaceans such as freshwater prawn (*Macrobrachium rosenbergii,* De man 1879).

Bangladesh is theorized as one of the most suitable country in the globe for freshwater prawn culture due to its climate condition. In Bangladesh 24 of freshwater prawn species are found. However, only *Macrobrachium rosenbergii* has aquaculture potentials and is commercially cultured (Akand and Hasan, 1992).

Freshwater prawn is one of freshwater species which has high potentials and market demand. Freshwater prawn is a highly valued product for international market and almost all Bangladeshi prawn are therefore exported. Statistics shows that about 70% of total freshwater prawn production is exported to foreign markets and the rest (30%) is consumed locally (Paul, 2008). During fiscal year 2009-2010, Bangladesh exported 80% shrimp and prawn products and other 20% are fish products. Besides during fiscal; year 2010-2011, Bangladesh exported 54891 MT of frozen and processed prawn and shrimp species which price was 35682 million BDT (DoF, 2012) and commonly Freshwater prawn contributes about 25-30% of total exported prawn and shrimp species (DoF, 2013).

Due to the high value and market demand of freshwater prawn large portion of population involved themselves in culture and capture from farm and wild stock. And increasing pressure and indiscriminate fishing from wild stock causes significant decline of catch from natural stock almost all-round the country.

Experiment shown that, freshwater prawn can be cultured in pond and any other confined area. Knowledge in the field of techniques and culturing methods has remarkably improved and at present the prawn has become the subject of intensive efforts to cultivate. When freshwater prawn is cultured intensively lots of problem arise because large-scale production facilities, aquatic animals are exposed to stressful conditions. Because the increased intensity of aquaculture has led to a high number of disease outbreaks with an increasing range of pathogens as a result in serious economic losses (Verma and Gupta, 2015).

Disease is the main constrain to improve the aquaculture production. To get rid of the diseases many aqua drugs (Chemicals including antibiotics) has been used so far in Bangladesh. The resistance in harmful agents (Bacterial resistance against drugs) is increasing with the intense use of different drugs. As a result the farmers are overusing these chemicals which are detrimental for human consumption. Sahul Hameed (2003) reviewed that the excessive use and misuse of antibiotics have resulted in antibiotic residuals in food and the spread of antibiotic-resistant pathogens in the aquatic environment, which threaten the safety and hygiene of food, and control of diseases in humans. Besides fishes with higher level of antibiotic residue cannot be exported to foreign countries which is harmful for the economy of the country. To reduce the use of antibiotics and other harmful chemicals, medicinal plants and extract can be used. Several hundred genera of medicinal plant are important sources for potent and powerful drugs. Plants are rich in a wide variety of secondary metabolites of phytochemical constituents such as tannins, alkaloids and flavonoids which act against different disease.

Polyphenols are kind of secondary plants metabolites which gives straw colour liquid on dilution, sweet and non-astringent flavour. This natural extract has anti-inflammatory and anti-bacterial properties. For these properties this bioactive compounds have the potential to replace antibiotics and hormones from animal feeds. Polyphenol are also rich in minerals and nutrients which play vital role in growth as well as production.

This study looked at the potentials of plant extracted polyphenols on better performance (Growth, survival rate, specific growth rate, condition factor, Food conversion ratio (FCR), proximate composition and taste) of freshwater prawn.

1.1 Objectives:

- To observe the effects of bioactive compound (Polyphenol) in prawn (Growth, survival rate, FCR, proximate composition, specific growth rate and condition factor).

1.2 Scope of the study:

The purpose of the study was to assess the potentials of plant polyphenols for better performance (Growth performance, survival rate, feed conversion ratio (FCR), proximate composition and taste, economic analysis) of farmed giant freshwater prawn. This study involved polyphenol supplement and effectiveness of polyphenol at different levels.

CHAPTER 2

REVIEW OF LITERATURE

Before conducting a research or experimental procedure it is important to have a look on the previous research activities related to the topic. Polyphenol are widely used on farm animals for their wide range of effects on growth, immune system, replacement of antibiotics etc. As a result there have a great chance on fishes. A review of the literature relevant to the present research work has been given below:

2.1 Freshwater prawn:

Macrobrachium rosenbergii is a large prawn of the family palaemonidae familiar to tropical freshwater and brackish water habitats of the indo-pacific (George, 1969). Freshwater prawn mostly found in south and south east Asia, together with Northern Australia and Western pacific islands (New, 1988). It is an important industry in many Asian country including China, India, Malaysia, Thailand, Taiwan, Bangladesh etc. However, as a primary inland cultured species freshwater prawn has commercial importance (New, 1995). It has cultural importance due to its fast growth rate, high demand in national and international market and tolerance to environment condition (Ranjit and Kurup, 2002). The farmed production of Freshwater prawn increased 2.7 times globally and 4 times during last 2 decades in Bangladesh (FAO, 2009). Freshwater prawn has commercial importance in Bangladesh and its culture practice is widely distributed in pond and rice fields while its natural distribution in rivers and also included that prawn culture bagged around 25% of the total fish and fisheries exportable products of Bangladesh. And large scale production to meet maximum profit leads aquatic animals to the stress condition which results in disease outbreak (Verma and Gupta, 2015). To get rid of the diseases farmers uses many antibiotics and chemicals. Antibiotics have harmful effect on human health and higher level of antibiotic residue cannot be exported to foreign countries. So researchers have been trying to increase the use of herbal medicine that can replace harmful antibiotics. So far some researches have shown positive result. Harikrishnan et al. (2012) reviewed that application of *Withania somnifera* extracts through supplementation of the feed

positively enhanced the innate immune system and increased survival rates in M. rosenbergii against *Aeromonus hydrophila* infection. Bhavan et al. (2010) stated that addition of fruit wastes such as orange, grape and apple peels and vegetable wastes (Rebecca and Bhavan, 2011) in giant freshwater prawn feeds increased growth and feed quality and feed efficiency when supplied at minimal levels (10–15%). Study also informed that use of microalgae and extract thereof in prawn feeds has significantly reduced mortality rate that infected with the pathogen *Aeromonus hydrophila*.

Bhavan et al. (2012) added that the active compounds of herbs, such as alkaloids, flavonoids, pigments, terpenoids, starch, steroids and essential oils have the ability to promote growth, body biochemical constituents in freshwater prawns. Besides herbs used in the culture of shrimps and prawns can improve the immunity, antimicrobial activity, appetite and provide anti-stress characteristics due to the presence of active compounds (Radhakrishnan et al., 2013).

2.2 Effect of polyphenols:

So far a number of studies have been conducted on the effect of polyphenols supplementation on human and farm animals. Polyphenols have wide range of benefits. Numerous studies in humans and rodents have shown that polyphenols are exerting anti-oxidative, anti-inflammatory, cardio-protective, cancer chemo-preventive and neuroprotective properties (Xia et al., 2010; Landete, 2012). Gessner et al. (2013a) conducted study that diets with polyphenol rich plant products from either grapes or hops exerts an anti-inflammatory effect in the small intestine and also improve food conversion ratio in piglets. Another study shows that resveratrol or quercetin exert anti-inflammatory effects and prevent development of fatty liver in rodents (Panchal et al., 2012). Polyphenols supplementation in rodents also improved the anti-oxidative status in the liver under experimental stress condition (Choi et al., 2012). Viveros et al. (2011) also observed that polyphenol-rich grape pomace extract supplemented with broilers fed diets improved in gain: feed ratio and digestibility of nutrients due to an increased absorptive surface of the intestine. Brenes et al. (2010) indicated that the positive effect of polyphenol rich grape by-products such as grape pomace and grape seed extract incorporated into feed in decreasing lipid oxidation in chicken meat. Another study has shown that the intake of polyphenol rich grape

pomace increases the antioxidant capacity of the breast and thigh meat of broiler chickens in the same way as vitamin E in experimental diets (Brenes et al., 2008). Because the antioxidant potential of GS polyphenols is 20 times higher than that of vitamin E and 50 times higher than that of vitamin C (Carpenter, et al., 2007). Supplementation of polyphenol by feeding either GME or SH exerts anti-inflammatory effects and improved feed conversion ratio (Gessner et al., 2013b; Fiesel et al., 2014).

Besides it has been shown that polyphenols pose a prebiotic activity in the intestine favouring the growth of beneficial bacteria while inhibiting the growth of pathogenic bacteria. Perumalla & Hettiarachchy (2011) indicated that polyphenol rich grapes have shown inhibitory effects against bacteria. Brenes et al. (2016) conducted study which has also shown that dietary polyphenol-rich grape products were also effective in increasing the growth of specific beneficial intestinal bacteria while competitively excluding certain pathogenic bacteria. Numerous in vitro studies have shown that the polyphenols found in grape by-products inhibit the growth of certain pathogens such as *Staphylococcus aureus*, *Escherichia coli*, *Campylobacter*, *Salmonella,* and *Helicobacter pylori* (Ganan et al., 2009). For these beneficial properties polyphenols are widely used with feed supplementation for human and farm animals. And long term supplementation of polyphenols does not lead to an accumulation of polyphenols in plasma and tissues (Bieger et al., 2008).

CHAPTER 3

MATERIALS AND METHODS

3.1 Study area:

The experiment was carried out from July 2017 to November 2017 at the roof top of professor quarters in Chittagong Veterinary and Animal Sciences University, Chittagong, Bangladesh.

3.2 Sources of polyphenol

Polyphenol were supplied by the "The Products Makers (Australia Pty Ltd.)" where they mainly provide their product named as " Polygain". Polygain contains natural Polyphenol. The polyphenol content of Polygain supplied in the trial is 30,400 mg / Kg (dosage at 0.2% - 60mg Polyphenol / kg Feed, at 0.4% - 120mg Polyphenol/ kg Feed, 0.6% - 180mg Polyphenol / kg Feed).

3.3 Experimental prawn post larvae collection:

2 cm length sized post larvae (PL) of freshwater prawn were collected from a renowned hatchery named "Halda Fisheries Ltd. situated at Potenga in Chittagong. Quality of PL was examined before stocking and only healthy and disease free PL were stocked for the experiment.

3.4 Collection of feed ingredients:

Feed ingredients and feed additives were collected from local market.

3.5 Preparation of polyphenol mixed feed

The feed which were applied in the tanks as treatment for 'freshwater prawn' was prepared in a feed mill of "Halda Fisheries Ltd. "by adding appropriate amount of Polyphenol.

3.6 Feed formulation

Quality and nutritionally balanced feed supply should be ensured for freshwater prawn culture. Because the success of any aquaculture system depends on good quality seeds and nutritionally balanced feed which offer superior growth within a stipulated period (Ahmed Ali, 1998). Ingredients added to basal diet and the feed formulation is shown in **Table 1**. And the energy provided by different feed ingredients is also showed in **Table 2**. Before application, the proximate compositions of feed were also examined and the measured percentages are included in **Table 3**.

Table 1: Feed ingredients and their inclusion level

Ingredient	Inclusion (%)
Fish Meal 40%	18.75
Fish Meal 60%	18.75
Soya bean Meal	10
Meat & Bone Meal	15.625
Rice Bran	10.625
Wheat Bran	11.25
Mustard Oil Cake	6.25
Maize	5
Wheat Flour	3.75
Total	**100**
Additives	
DCP	0.5
Pellet binder	0.5
Soybean oil	0.5

Polyphenol were added to the basal diet at a range of 0mg, 60 mg, 120 mg, 180mg per kg of feed (assigned as T_0, T_1, T_2, T_3) to prepare different experimental feed.

Table 2: Feed ingredients used in experiment and energy provided by them (kcal./kg)

Sl. No.	Ingredients Name	Protein (%)	Fat (%)	CHO (%)	Energy
1.	Fish Meal -1	7.50	2.10	0.33	63.32
2.	Fish Meal -2	11.25	2.10	0.33	84.32
3.	Soyaben Meal	3.60	1.80	3.30	50.79
4.	Meat & Bone Meal	7.81	0.70	0.33	51.76
5.	Rice Bran	1.17	1.06	4.68	35.81
6.	Wheat Bran	1.46	0.45	7.31	42.45
7.	Mastard Oil Cake	1.87	0.81	2.12	26.93
8.	Maize	0.40			2.24
9.	Wheat Flour	0.64	0.15	2.81	16.49
	Total=	35.71	9.18	21.21	374.13

Table 3: Proximate composition analysis of prepared feed

Parameter	Percentage
Moisture	14.91%
Crude protein	35.41%
Crude lipid	8.82%
Ash	22.40%

3.7 Design of experiment

The experiment was performed in cylindrical shaped plastic tank which water holding capacity was about 250 litres. Twelve tanks were taken (3 tanks for each treatment) for the experiment. A total of 600 PL were randomly allocated where for each treatment group there were 150 PL and 50 PL for each replication and distributed into four dietary treatment groups assigned as T_0, T_1, T_2, T_3 with three replication per treatment(**Table 4**). This experiment was performed in a re-circulatory system where

water was re-circulated everyday. Experiment was carried out for five months and sampling was done at 15 days intervals.

Table 4: Layout of the experiment (CRD) showing distribution of freshwater prawn in tanks and applied treatments.

Dietary treatment groups	Treatment×Replication (T_n× R_n)	No. of prawn per tank	Total no. of prawn per treatments
To (without 'Polyphenol'-Control group)	T_0R_1 (Tank no.1)	50	150
	T_0R_2 (Tank no.2)	50	
	T_0R_3 (Tank no.3)	50	
T_1 (60 mg Polyphenol' per kg feed)	T_1R_1 (Tank no.4)	50	150
	T_1R_2 (Tank no.5)	50	
	T_1R_3 (Tank no.6)	50	
T_2 (120 mg Polyphenol' per kg feed)	T_2R_1 (Tankno.7)	50	150
	T_2R_2 (Tank no.8)	50	
	T_2R_3 (Tank no.9)	50	
T_3 (180 mg Polyphenol' per kg feed)	T_3R_1 (Tankno.10)	50	150
	T_3R_2 (Tank no.11)	50	
	T_3R_3 (Tank no.12)	50	
		Total	600

3.8. Tank preparation:

Twelve tanks were prepared for the culture of prawn. Cylindrical shape plastic tank was selected which water holding capacity was about 250 litre. Plastic tank was most preferable for their longevity and easy maintenance. The upper portion of tank was cut then tank were thoroughly washed with bleaching powder to remove the unwanted and harmful chemicals. Washing process was repeated for three times to ensure that the tank were clean enough for prawn culture. Tanks were prepared with inlet, outlet, aeration and water exchange facilities. Some substrates were also added into the tank. Substrate has importance in prawn culture because use of artificial substrates to increase the amount of two dimensional space available to prawns and thereby yielding an increase in survival and production. A ground reserve tank and an overhead reserve tank were added to the set-up for water exchange facilities. Small net was used to cover the outlet of the tank that inhibits the PL escape out.

3.9 Stocking:

After collection of PL they were conditioned with the environment where they were released. Physiological condition of PL was closely examined and weak and unhealthy PL was discarded. A total of 600 PL were stocked where 150 PL for each treatment and 50 PL for each replication.

3.10 Feeding:

Polyphenol treated feed was supplied to the prawn post larvae in the tanks. Feed of particular treatment group were supplied to the particular tank and its replications. Feed was supplied two times daily, at the morning and evening. Feed was given in the feeding tray, placed into the water. Thus helped us to observe the feed intake and washed out the left over feed in the next day. As a result it was quite easy to maintain water quality from being deteriorated.

3.11 Bio-security:

The tanks were covered all the times as a result no foreign and harmful materials got contact to the tanks. Besides total experimental area were restricted for the visitors during research period. Lime was applied at regular interval to maintain the pH level. Lime also helps to control turbidity.

3.12Water quality:

Freshwater prawn is very sensible to the water quality and surrounding environment. To maintain the optimum water quality the water quality parameters such temperature, pH etc. were recorded 3 times daily. So that any kind of changes and fluctuations in water quality were noticed easily thus helped to take necessary measures.

3.13 Fortnight length gain:

The length of the experimental freshwater prawn was recorded at every 15 days intervals. And the length gain was calculated by deducting length of corresponding sampling.

Length gain = (Present body length – Immediate last body length)

3.14 Fortnight weight gain:

The weight of the experimental freshwater prawn was recorded at every 15 days intervals. And the weight gain was calculated by deducting weight of corresponding sampling.

Weight gain = (Present body weight – Immediate last body weight)

3.15 Feed conversion ratio:

The amount of feed intake per unit of weight gain is the feed conversion ratio (FCR). This was calculated by using the following formula,

$$\text{FCR} = \frac{\text{Feed intake(kg)}}{\text{Weight gain(kg)}}$$

3.16 Proximate composition:

Among the proximate composition, only protein percentage in prawn was determined by kjeldahl apparatus method.

3.17 Condition factor:

Condition factor was estimated by using the following formula

$$CF = \frac{\text{Weight of fish}}{(\text{Length of fish})^3} \times 100$$

3.18 Specific growth rate:

Specific growth rate was calculated by following formula:

$$SGR = \frac{\ln(\text{final weight}) - \ln(\text{initial weight})}{\text{duration in days}} * 100$$

3.19 Collection of samples for laboratory analysis:

After the culture period, 5 prawns were selected from each replication randomly for collection of body muscle. 15 prawns of each treatment were collected in polybags and soon after collection were preserved for lab based activities.

3.20 Statistical analysis and reporting:

All the data as weight of fish, weight gain, length of fish, length gain, data of proximate composition analysis were entered into MS excel (Microsoft office excel-2007, USA) and IBM SPSS Statistics 23 Version. Values are expressed as means ± standard deviation (SD). Data were analyzed by one-way analysis of variance (ANOVA) followed by Tukey's post hoc test to assess statistically significant differences among the control and different treated values. Statistical significance was set at $P < 0.05$.

PHOTO GALLARY

Plate 1: Cylindrical tank

Plate 2: Tank preparation

Plate 3: Culture unit set up

Plate 4: Tank set up

Plate 5: PL stocking

Plate 6: PL stocking

Plate 7: Sample collection

Plate 8: Collected sample

Plate 9: Length measurement

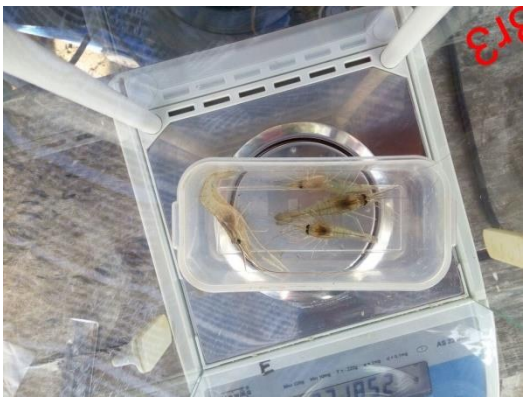

Plate 10: Weight measurement

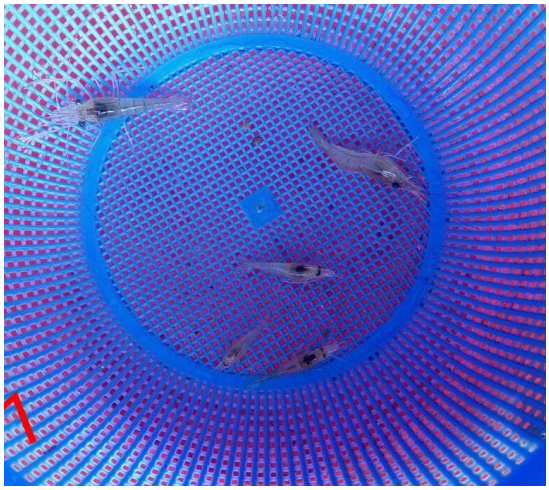

Plate 11: T_0 treated prawn

Plate 12: T_1 treated prawn

Plate 13: T_2 treated prawn

Plate 14: T_3 treated prawn

Plate 15: Comparison among T_0, T_1, T_2, T_3 treated prawn

Plate 16: Comparison among T_0, T_1, T_2, T_3 treated prawn

Plate 17: Sample preparation

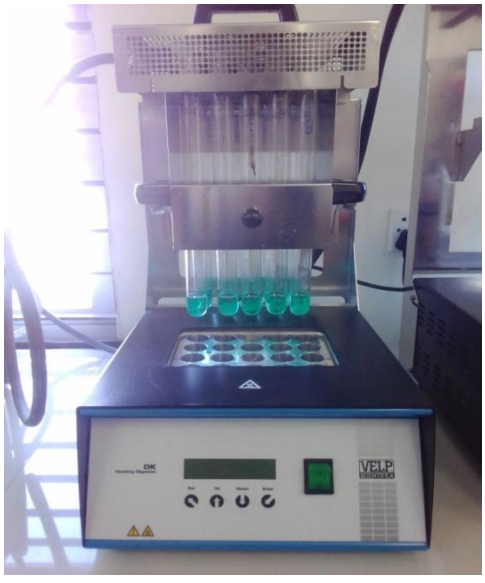

Plate 18 : Digestion unit

Plate 19: Distillation unit

Plate 20: Titration

Plate 21: Before titration titration

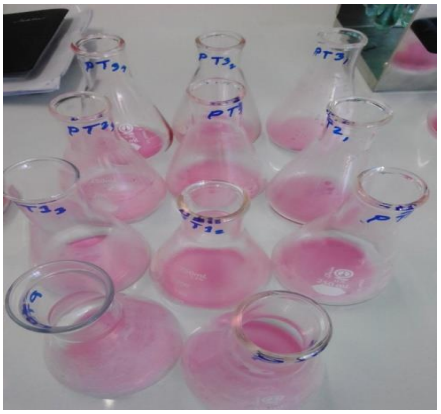

Plate 22: After

CHAPTER 4

RESULT

4.1 Effect of polyphenol on prawn length:

Length of the experimental freshwater prawn was recorded at every 15 days interval throughout the experiment period. Result shows that length gains differ effectively from the initial length to the final length taken (**Figs. 1&2**). And it's also clear that at every 15 days interval length increases from the previous sampling. At the end of the five month long experiment final result indicate that highest length 6.27 (±0.51)cm was gained in T_3 polyphenol supplemented feed treated freshwater prawn while 5.22 (±0.48)cm, 4.99 (±0.26)cm, 4.85 (±0.13)cm length were found at T_2, T_1, T_0 polyphenol supplemented feed treated prawn (**Table 5**).

Table 5: Length gain in different experimental feed treated group

Name	Initial length (cm)	Final length (cm)		Length gain (cm)
Prawn	2	T_0	4.85(±0.13)[b] (4.70- 4.95)	2.85(±0.132)[b] (2.70-2.95)
		T_1	4.99(±0.26)[b] (4.70- 5.20)	2.99(±0.257)[b] (2.70-3.20)
		T_2	5.22(±0.48)[b] (4.70- 5.65)	3.21(±0.48)[b] (2.70-3.65)
		T_3	6.27(±0.51)[a] (5.95- 6.86)	4.27(±0.51)[a] (3.95-4.86)
	Level of Significance		0.007	0.007

*Values are means ± S.D. Values are statistically significantly different (P < 0.05).

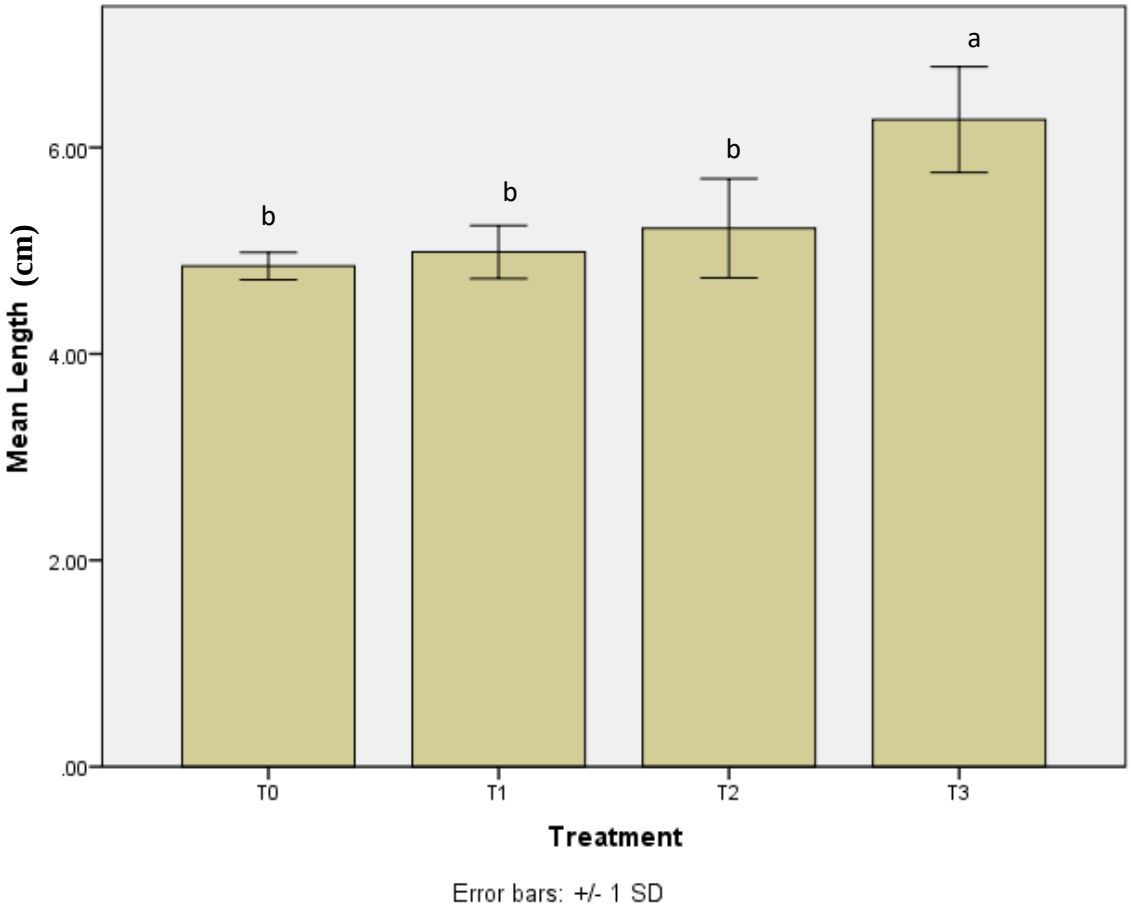

Fig 1: Effects of polyphenol on length of freshwater prawn (Means ± SD) after 5 months. The polyphenol supplemented feed groups T_1, T_2 and T_3 were compared to the control group (T_0). Values are statistically significantly different ($p < 0.05$, n=4).

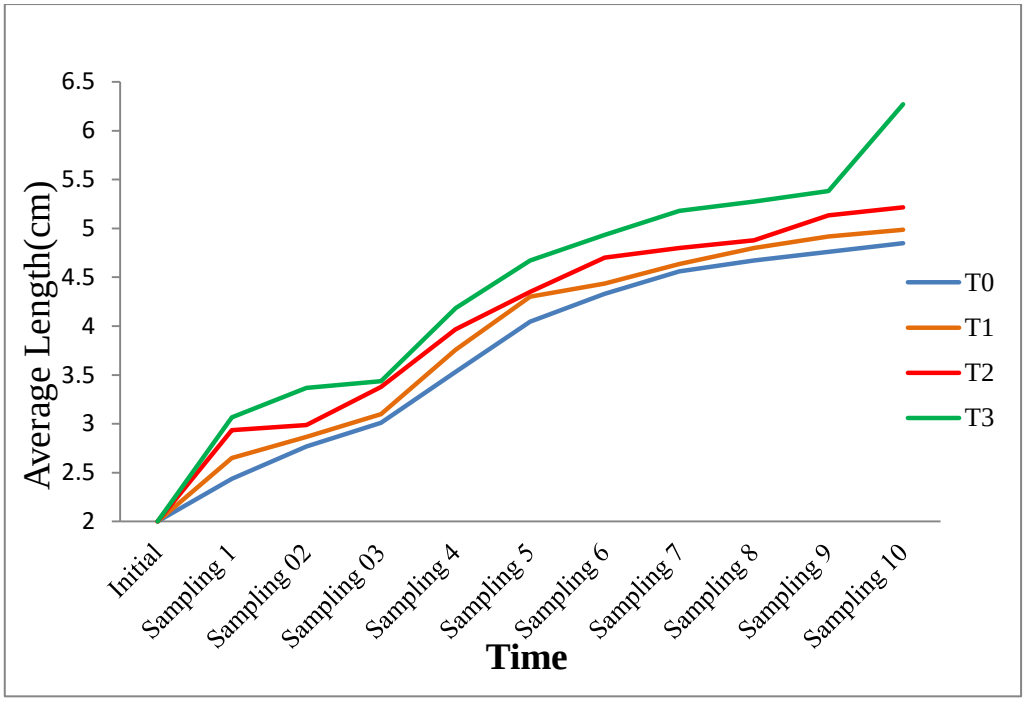

Fig 2: Graphics showing that highest length is achieved at T_3 and lowest at T_0.

4.2 Effect of polyphenol on weight:

Weight gain of the experimental prawn was also recorded at every 15 days intervals. Result indicated that at different treatment group weight gain vary significantly (**Table 6**). Result also shows that after 5 month long experiment highest average weight was found in T_3 polyphenol supplemented experimental feed treated freshwater prawn and comparatively lowest average weight gain was happened in T_0 polyphenol supplemented experimental feed treated prawn (**Figs. 3&4**). Final weight gain recorded after the culture duration is shown in **Table 6**.

Table 6: Weight gain in different experimental feed treated group

Name	Final Weight (g)	
Prawn	T_0	$0.89(\pm0.10)^b$ (0.78-0.96)
	T_1	$1.12(\pm0.19)^b$ (0.90-1.25)
	T_2	$1.16(\pm0.24)^b$ (0.89-1.34)
	T_3	$2.13(\pm0.19)^a$ (2.00-2.35)
	Level of Significance	0.00

*Values are means ± S.D. Values are statistically significantly different ($P < 0.05$).

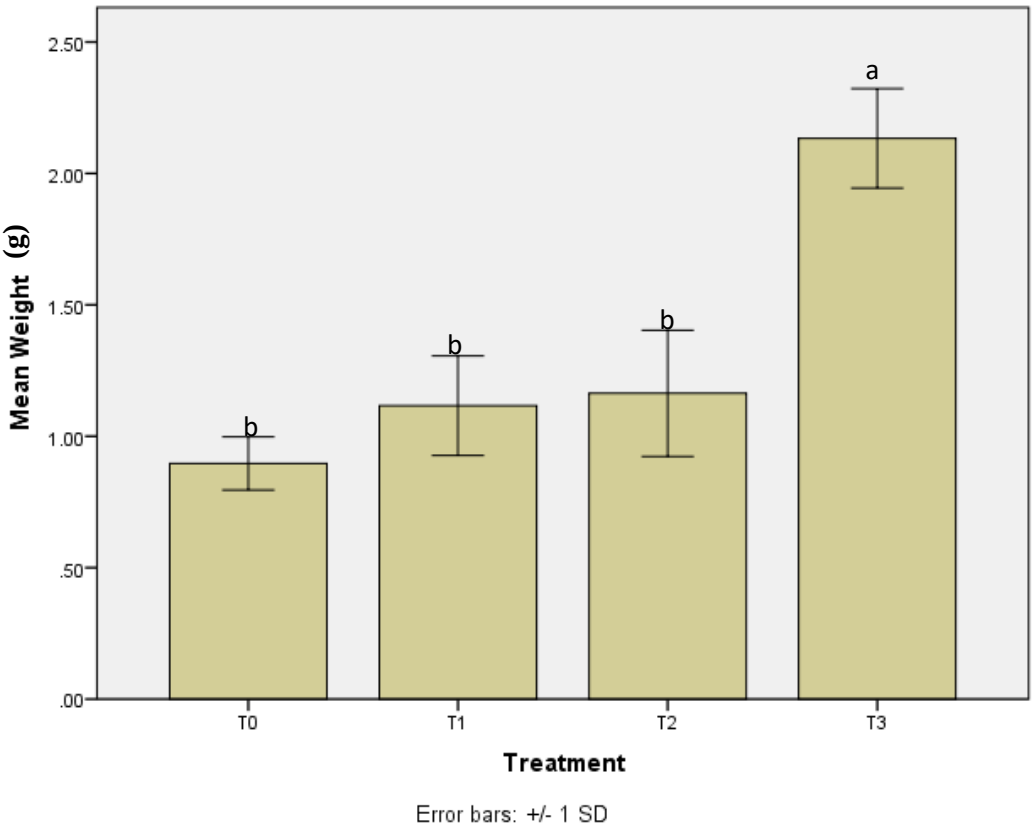

Fig 3: Effects of polyphenol on weight of freshwater prawn (Means ± SD) after 5 months. The polyphenol supplemented feed groups T_1, T_2 and T_3 were compared to the control group (T_0). Values are statistically significantly different ($p < 0.05$, n=4).

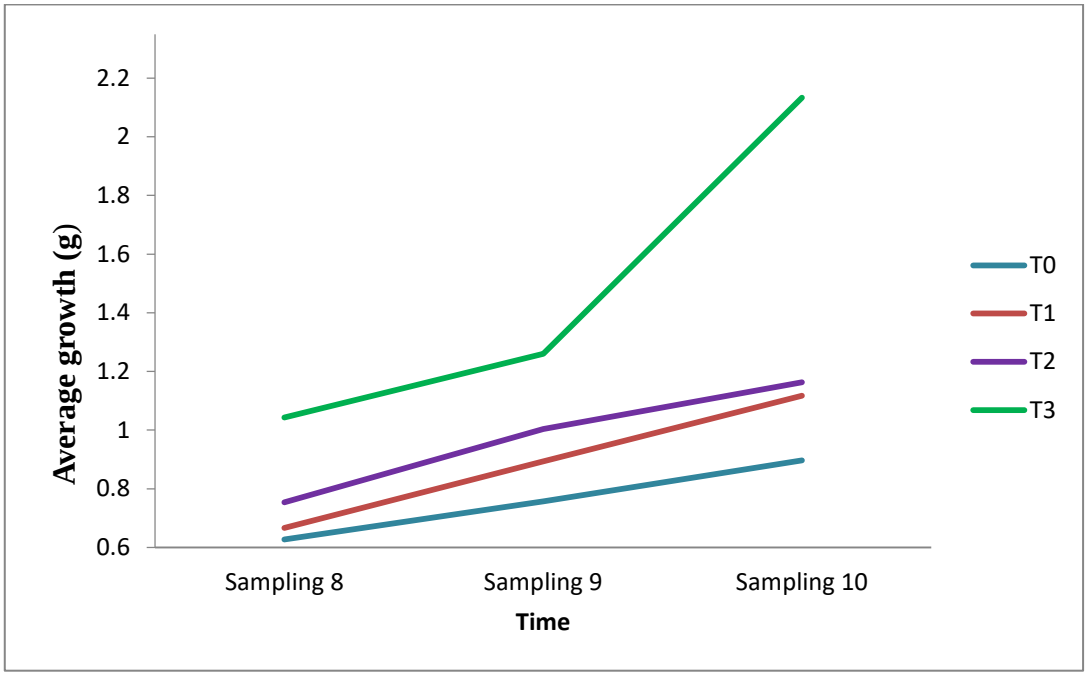

Fig 4: Average weight from 8th sampling represented in line diagram

4.3 Feed conversion ratio:

Feed conversion ratio (FCR) also varies from treatment to treatment. The experimental result shows that there was better FCR in T_3 polyphenol supplemented feed compared to other treatments. In T_3 feed supplemented treatment group the FCR value was 1.15, where 1.86, 1.85, 2 at T_2, T_1 and control respectively (**Table 7**).

Table 7: FCR value in different treatments

Species	T_0	T_1	T_2	T_3
Prawn	2	1.85	1.86	1.15

4.4 Proximate composition

Protein:

The protein percentages in experimental feed supplemented treatment groups are also examined. Experimental result shows that prawn supplemented with T_2 polyphenol mixed feed contain more crude protein (**Table 8**). Though growth performance of freshwater prawn is higher in T_3 feed supplemented group but the protein percentage is high in T_2 feed supplemented group.

Table 8: Protein percentage at different treatments

Name	T_0	T_1	T_2	T_3
Prawn	8.88%	15.54%	17.71%	14.45%

4.5 Condition factor (K):

The condition factor obtained in the present study are 0.78±0.03, 0.89±0.03, 0.81±0.06, 0.87±0.13 in T_0, T_1, T_2, T_3 supplemented treatment groups respectively (**Fig. 5**). Result shows that condition factor in different replication group were similar and were not significantly different ($p > 0.05$).

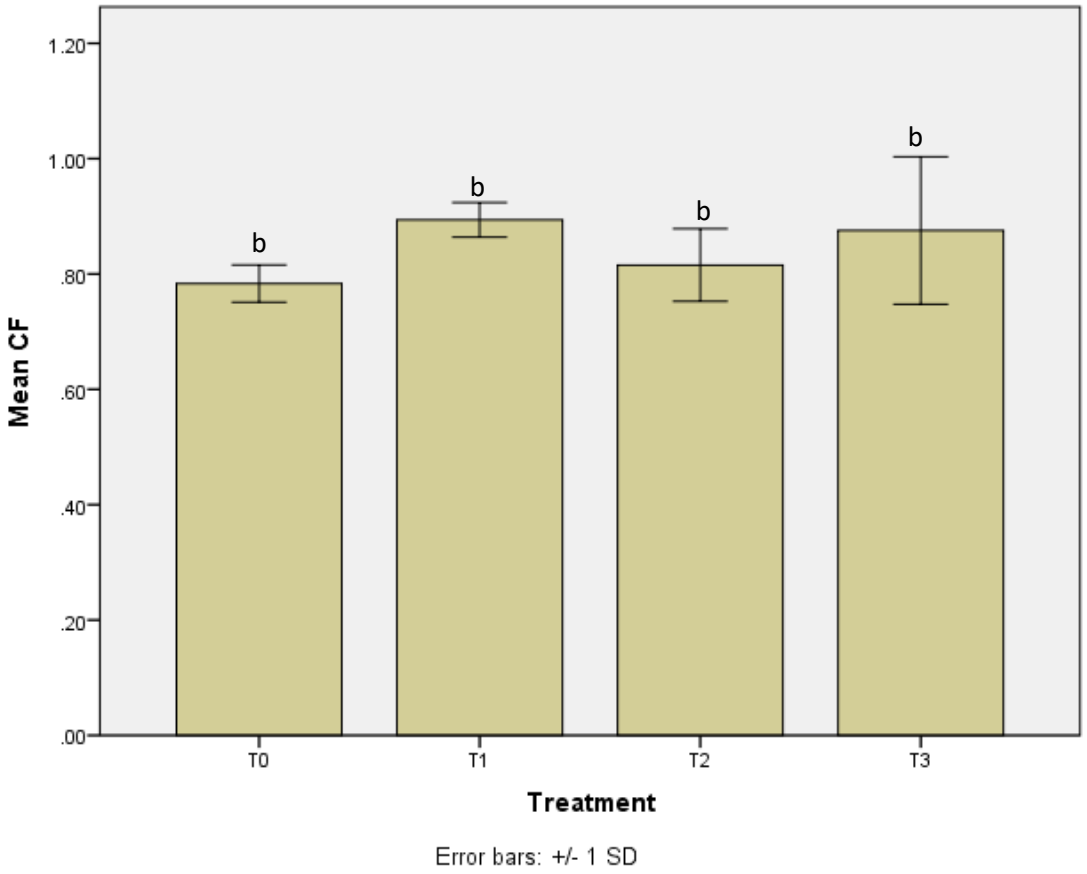

Fig 5: Condition factor represented in diagram. Values are not statistically significantly different ($p > 0.05$, n=4).

4.6 Specific growth rate:

Specific growth rate found in T_0, T_1, T_2, T_3 treatment groups are 0.26±0.14, 0.35±0.05, 0.31±1.4, 0.49±0.22 respectively (**Fig 6**). Result shows that specific growth rate in different replication group were similar and were not significantly different ($p > 0.05$).

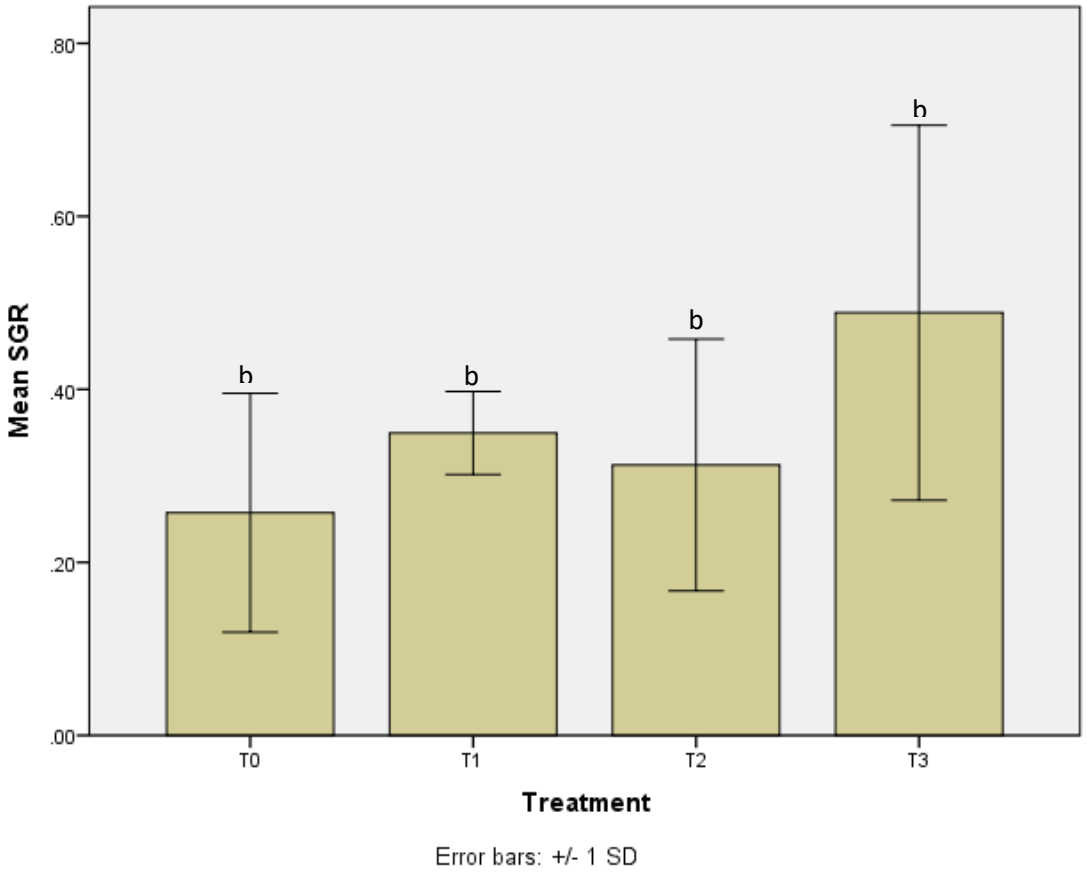

Fig 6: Specific Growth Rate represented in diagram. Values are statistically significantly different ($p < 0.05$, n=4).

CHAPTER 5

DISCUSSION

The present study was conducted to know the potentials of plant polyphenols for better performance of farmed giant prawn. A number of effects have been documented in this present study on some parameters such as length gain, weight gain, FCR, condition factor, specific growth rate, protein concentration.

5.1 Effect on length and length gain:

From the present study it was found that the initial body length of freshwater prawn PL of all treatment groups were indifferent. But when different polyphenol supplemented feed were fed to the freshwater prawn PL, the length gain differed among different treatment group. In every sampling it was found that the length gain differed in different groups. From the final sampling it was found that the highest length gain happened in T_3 polyphenol supplemented treatment group while 2^{nd}, 3^{rd} , and lowest length gain found in T_2, T_1 and control respectively. The highest body length gain in T_3 polyphenol supplemented feed indicated that higher polyphenol supplementation result in higher body length gain. And if the amount of polyphenol were decreased then the body length gain were also decreased also. In control feed treatment group body length gain was lowest as there was no addition of polyphenols

From this study it is observed that polyphenols have great effect on body length gain as well as growth. This length gain happened because polyphenols may have attractant properties which influence the feed intake of freshwater prawn. It also support the study conducted on broilers that polyphenols supplemented feed fed improved the gain: feed ratio and digestibility of nutrients due to an increased absorptive surface of the intestine (Viveros et al., 2011). Polyphenols are also rich in vitamin and minerals which have direct effect on improvement of body length gain.

It was a comparative study where growth among different treatments group were considered rather than overall production.

5.2 Effect on weight and weight gain:

Present study has also shown that polyphenol has effect on weight gain of freshwater prawn. In final sampling it was found that the highest weight gain happened in T_3 polyphenol supplemented treatment group while 2nd, 3rd and lowest weight gain found in T_2, T_1 and control treatment group respectively. Highest weight gain in T_3 treatment group indicated that the increasing amount of polyphenols supplemented feed leads to the increase in weight gain. In this comparative study it was found that feed without polyphenol show lower weight and weight gain than the fish fed with polyphenol supplementation ($p<0.05$) which undoubtedly indicate that polyphenol has great effects on prawn.

Besides, polyphenol seems to be acting as an attractant. Use of polyphenol removes the fishy odour of fish meal used in formulating feed and creates a fresh smell in it. Prawns were attracted to the feed, fighting and splashing to consume the feed containing polyphenol than the control. That enhanced feed consumption which ultimately increase the survival rate and also responsible for reduction of left over feed. Thus enhance feed intake and growth rate. Lower feed residues also help to maintain water quality.

5.3 Effect on feed conversion ratio (FCR):

From the present study we have also found polyphenol has wide range of effect on feed conversion ratio (FCR). Study has shown that higher level of polyphenol reduces FCR value. Which is also similar to the observations of Viveros et al., (2011) that polyphenol-rich grape pomace extract supplemented with broilers fed diets improved in gain: feed ratio and digestibility of nutrients due to an increased absorptive surface of the intestine. As the level of polyphenol supplementation increases then the FCR value reduce. The present result also support the findings that diets with polyphenol rich plant products from either grapes or hops exerts an anti-inflammatory effect in the small intestine and also improve food conversion ratio in piglets (Gessner et al., 2013a). Supplementation of polyphenol by feeding either GME or SH exerts anti-inflammatory effects and improved feed conversion ratio (Fiesel et al., 2014). Effects on FCR value greatly influence the economic outcome of any culture practice. From this point of view, polyphenol has great potentials in freshwater prawn culture.

5.4 Effect on protein concentration:

The percentage of protein was estimated to be higher in polyphenol treated fishes than the control group. This may happen because polyphenol is rich in anti-oxidants. It inhibits the activation of nuclear factor kappa B (NF-κB) and also able to induce antioxidative and cytoprotective effects by inducing nuclear factor (Tangney and Rasmussen, 2013). Beside polyphenol act as anti-inflammatory both *in vitro* and *in vivo.* Polyphenols supplementation also has potential benefit for athletes which is helpful to exercise performance or oxidative damage (Kathryn & Myburgh, 2014).

5.5 Effect on condition factor and specific growth rate:

Information on condition factor (K) can be effective to culture system management because they provide information to the producer of the specific condition under which organisms are developing (Araneda et al., 2009). It reflects recent physical and biological condition, and fluctuates by interaction among feeding conditions, parasitic infections and physiological factors (Le Cren, 1951) and it indicates the changes in food reserves and therefore an indicator of the general fish condition. The result observed in the present study indicated that condition factor in different replication group are not significantly different ($p>0.05$) and the prawns were in good condition.

The specific growth rate (% SGR per day) of freshwater prawns of the present study ranged from 0.49±0.22 to 0.26±0.14 in T_3 to T_0 respectively. Though feed containing higher level of polyphenol (180mg per kg feed) showed highest growth rate compared to other treatment groups but the specific growth rate ware not significant ($p>0.05$).

CHAPTER 6

CONCLUSION

Freshwater prawn is one of the most commercially important fishery product which contribute greatly to our national economy. It also contributes to the nation protein requirement by fulfilling protein demand. Now-a-days huge numbers of population are involved in freshwater prawn farming. Disease occurrence is a common issue and a matter of headache for the poor farmers in freshwater prawn because of their highly sensitivity to water quality and other environmental stress. Use of high doses of antibiotic is a common phenomenon which is very harmful for the consumer as well as export market.

The results of this experiment are supporting consumer protection by describing the way of preventing diseases in freshwater prawn with the use of polyphenols. The research is also showing the potential role of polyphenols in the growth performance, immune response of prawn, safety and disease free prawn as well as improved proximate composition of the fishes. Lots of research can be conducted on the effects of polyphenols in different fishes as well as effect of polyphenol in fish muscle. This type of research work will be a new dimension for improving fisheries industry in Bangladesh.

CHAPTER 7

RECOMMENDATIONS

According to this research work, the following recommendations may be done:

- Farmer may use polyphenol in freshwater prawn feed because it lower FCR value and increased growth performance.
- Polyphenols may also be used in improving the total production and food safety.
- As it is a pilot study, further studies may be conducted on similar or different field to make a concrete remark on effect of polyphenol.
- Use of polyphenol can be added to the national policy level.

CHAPTER 8

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