



## Effects of probiotic and spirulina diet on growth performance of Shing, *Heteropneustes fossilis* in the laboratory condition

Krishna Chandra Roy<sup>1\*</sup>, Provati Rani Baisha<sup>2</sup>, Ulfat Jahan Lithi<sup>3</sup>, M Afzal Hossain<sup>4</sup>

<sup>1, 2, 4</sup> Department of Fisheries Management, Faculty of Fisheries Hajee Mohammad Danesh Science and Technology University (HSTU) Dinajpur, Bangladesh

<sup>3</sup> Department of Fisheries Technology, Faculty of Fisheries Hajee Mohammad Danesh Science and Technology University (HSTU) Dinajpur, Bangladesh

### Abstract

The study was conducted to understand the effects of spirulina and probiotic on the growth performance of Shing (*Heteropneustes fossilis*) at laboratory condition in Hajee Mohammad Danesh Science and Technology University. The research was designed of two treatments (T1: use of artificial diet; T2: artificial diet with spirulina and probiotic) with two replications and was performed from 1 October 2018 to 31 December 2018. Water quality parameters were regularly monitored. Fish growth was measured prior stocking and after the trial. Results indicated that water quality did not show significant ( $P > 0.05$ ) changes between the treatments during the study period. Fish growth was attributed by higher length gain (LG:  $2.35 \pm 0.20$  cm) and weight gain (WG:  $3.10 \pm 0.26$  gm) in T2 than T1 (LG:  $1.22 \pm 0.11$  cm; WG:  $2.365 \pm 0.21$  gm). Higher specific growth rate was found in T2 (0.34%) than T1 (0.28%). Low food conversion was determined in T2 (6.83) than in T1 (16.09) after the trial. This indicates the better effects of spirulina and probiotic on growth of shing in T2. However, condition factor of fish indicated good and healthy condition in both treatments. Overall, the results show that spirulina and probiotic is likely having significant role in the growth of shing in experimental condition. This study enhanced our understanding and knowledge on effects of probiotics and spirulina as supplementary diet of shing.

**Keywords:** growth, laboratory, probiotic, shing, spirulina

### 1. Introduction

Aquaculture is nowadays promisingly growing with increasing demand of animal protein in the world. Fisheries and aquaculture contribute significant role in the national economy of Bangladesh. Fisheries and aquaculture contribute 3.57 percent to the national GDP and more than one-fourth (25.30%) to the agricultural GDP during 2017-18 in Bangladesh<sup>[7]</sup>. This sector has a significant contribution in food and nutrition security through consistently providing quality animal protein. Currently, fisheries sector is providing about 60 percent of total animal protein supply to the people of Bangladesh. In the last decade, aquaculture has been extensively extended to other many common large and small indigenous fish species (SIS) as many of them are being endangered and some are extinct due to environmental degradation. Along with the SIS such as magur (*Clarias batrachus*), pabda (*Ompok pabda*), gulsha (*Mystus cavasius*) and mola (*Amblypharyngodon mola*), farming of shing (*Heteropneustes fossilis*) has been immensely attracted to the fish farmer in Bangladesh.

*Heteropneustes fossilis*, locally named Shingi or Shing is very popular and high valued fish in Bangladesh. The species is considered to be highly nourishing, palatable and tasty fish among the catfish in Bangladesh. The species is well preferred to the common people because of its less spine, less fat and high digestibility in many parts of Indian subcontinent<sup>[16]</sup>. The species contains higher iron (226 mg 100g<sup>-1</sup>) and fairly higher calcium compared to other freshwater fishes<sup>[31]</sup> and is suitable for the people whom animal fats are undesirable<sup>[29]</sup>.

In past, shing was highly abundant in open water systems

such as floodplains, canals, *beel* and haors in Bangladesh. This species has been identified as threatened species due to over exploitation, environmental degradation of the natural habitats in Bangladesh<sup>[15]</sup>. Though the seed production of *H. fossilis* through induced breeding and culture technology has been developed in Bangladesh<sup>[3]</sup>, but now a days shing is being intensively cultured at different regions of this country. Probiotics are live non-pathogenic microorganisms that provide colonization resistance to the pathogenic microbes and thus are effective in prevention and treatment of some diseases<sup>[14]</sup>. These beneficial bacteria used to destroy pathogens in the animal industry as a better remedy and are now practiced in aquaculture to control of pathogens. Similarly, spirulina is an available microalga found all over the world. It is considered as natural sources of food in the food chain of fish<sup>[11]</sup> and used as food for humans and animals due to its higher nutritional value<sup>[4, 2, 9]</sup>. It has a great function on vitamins (especially vitamin A and B12), minerals, polyunsaturated fatty acids, carotenes and other pigments that have antioxidants activity<sup>[30]</sup>. Therefore, Spirulina could be an excellent source of protein and energy<sup>[13]</sup>.

Shing is being extensively cultured following extensive and intensive, mono or polyculture system. In these culture systems, artificial diet is being applied for their better production throughout the culture period. However, disease is also great concern and need to be carefully managed in culture of shing. Although, extensive or intensive culture of shing is led by different artificial feed, information on the use of probiotics and spirulina as supplementary feed in shing culture is very limited. Therefore, this study was focused

odfer45n a baseline study to observe the growth of shing by using probiotics and spirulina in laboratory.

**2. Methodology**

**2.1 Experimental design**

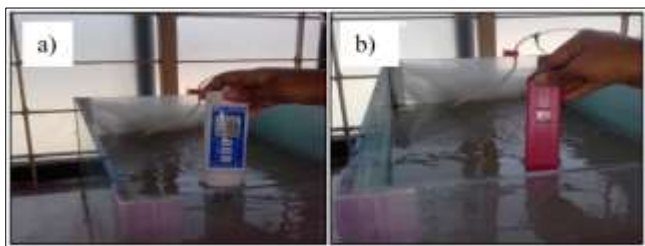
The study was carried out in the laboratory of department of Fisheries Management, Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur for a period of three months from 1 October 2018 to 31 December 2018. The experiment was designed with 2 treatments and duplicate replications. The size of aquariums were 25 cm × 45 cm × 25 cm and rectangular in shape that had no inlet and outlet (Fig. 1). Each aquarium was filled in with 20-liter tap water and aeration by using aerator and water was replaced every day.



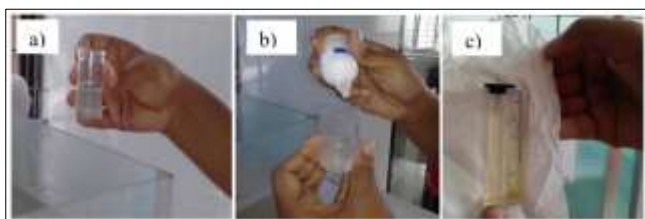
**Fig 1:** Experiment design in the laboratory

**2.2 Fish collection and management**

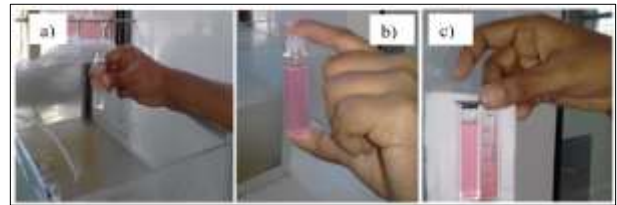
Fingerling sizes of shing were collected from a hatchery in Dinajpur district. Fish were transported in aerated holding tanks to the laboratory of HSTU, Dinajpur. The individuals were acclimated in a holding tank for two weeks. After acclimations, fish were transported to aquariums for growth trial. Water quality parameters (Do, pH, and temperature) were measured by Do meter, pH meter and Celsius thermometer (Fig. 2.) Amonia, nitrate and nitrite were monitored daily by using a multi-parameters probe. The nitrate, nitrite and ammonia levels were measured on alternative days using test kit during acclimatization and experimental period (Fig. 3, 4 and 5). The tank and aquaria were cleaned every day using siphoning system.



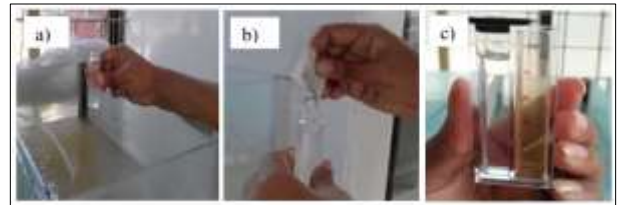
**Fig 2:** Measurement of water quality parameters a) Dissolved oxygen, b) pH of experimental aquarium



**Fig 3:** Measurement of ammonia a) Filled the beaker with water sample b) Added ammonia reagent c) Matched the color with white sheet



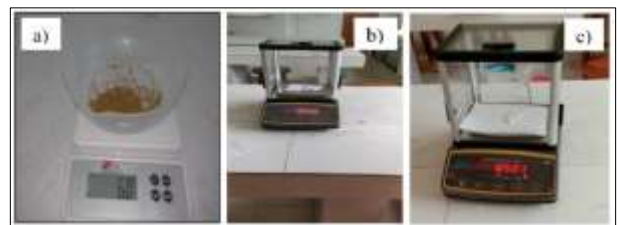
**Fig 4:** Measurement of nitrite a) Filled the glass cuvet with water sample b) Added nitrite reagent c) Matched the color with white sheet



**Fig 5:** Measurement of nitrate a) Filled the glass cuvet with water sample b) Added nitrate reagent c) Matched the color with comparator cube

**2.3 Feeding trial**

Fish were fed daily twice during the acclimation and trial period. T1 was prepared with artificial diet (Carbohydrate 30%, Protein 30 %, Fiber 6 %, Fat 7%, Ash 20%, Calcium 2.1%, Phosphorus 0.8% and Moisture 12%) without mixing any probiotic and spirulina. T2 was made by artificial feed (5% body weight) with spirulina (0.04g) and probiotic (0.2%) were mixed with adequate water (Fig. 6). Dietary feed ingredients were ground using a laboratory grinder and then blended into a homogenous doughy matter by adding water [33]. Tong way feed meal of Bangladesh Company Ltd. was used as artificial diet.



**Fig 6:** Measurement of artificial feed (a), spirulina (b) and probiotic (c)

**2.4 Sampling of fish**

During the period of experiment the fishes were caught 15 days interval by net. Growth of the fish samples were measured after 15 days interval by using length and weight of fish. Lengths of the fingerlings were measured with measuring scale and weight with weighing balance (Fig. 7). After recording length and weight, fingerlings were slowly released into the aquarium.



**Fig 7:** Measurement of weight (gm) and length (cm) of *H. fossilis*

**2.5 Growth parameters**

Growth of the fish samples were measured after 15 days interval. Lengths of the fingerlings were measured with measuring scale and weight with weighing balance from the random samples of each aquarium. The following parameters were used to evaluate the growth.

Survival rate (%) = (Number of actual fish lived/ Number of actual fish stocked) × 100

Specific growth rate (SGR, %) = [ln FW- ln IW] / (T-t) × 100

Where, ln FW = Natural log of final weight at time T, ln IW = Natural log of initial weight at time T.

Condition Factor = Mean W/  $W_{pred.}$  and  $W_{pred.} = qL^b$

Where,  $W_{pred.}$  = Predicted weight, W = body weight in gram, L = body length in cm

Feed conversion ratio (FCR) = [Feed (g) consumed by the fish]/ Weight (g) gain of the fish (W2-W1)

Where, W2 = Final weight, W1= Initial weight.

**2.6 Data analysis**

All data of growth parameters were analyzed in excel spread sheet. Water quality data were tested for normality in SPSS.

However, independent sample t test was performed to compare the variation of water quality between the treatments during the study period.

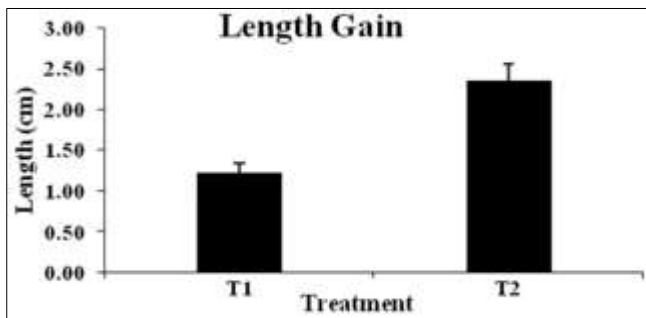
**3. Results**

**3.1 Growth performance**

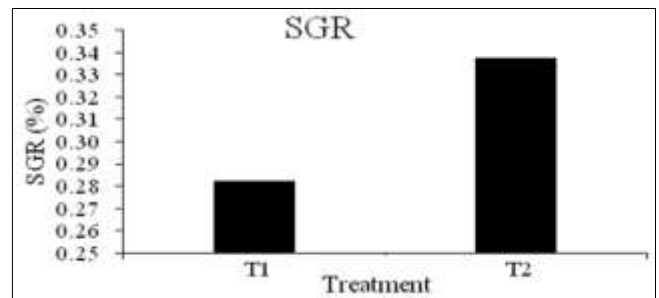
The average final length were  $10.745 \pm 0.09$  in T1 and  $11.90 \pm 0.24$  in T2 and the average length gain was  $2.35 \pm 0.20$  for T2 and  $1.22 \pm 0.11$  for T1 (Fig. 8 and 9). The average final weight was  $7.73 \pm 0.09$  in T1 and  $8.61 \pm 0.18$  in T2 (Table 1). The specific growth rate was found 0.34% (T2) and 0.28% (T1) (Fig. 10). The survival rate was 95% for T1 and 100% for T2 (Fig. 11) and FCR were 12.23 in T1 and 9.9 in T2. The slope (b), the mean body weight (BW), predicted weight ( $W_{pred.}$ ) and condition factor (CF) were calculated as 5.51, 5.2677 and 1.045, respectively for the T2 initial data. After experimental periods of T2, the slope 'b' and coefficient 'q' were obtained as 2.69451 and 0.00801 through power curve equation where the mean body weight (BW), predicted weight ( $W_{pred.}$ ) and condition factor (CF) were calculated as 8.61, 6.3343 and 1.351, respectively (Table 2).

**Table 1:** Length and weight gain status of *H. fossilis* in different treatments

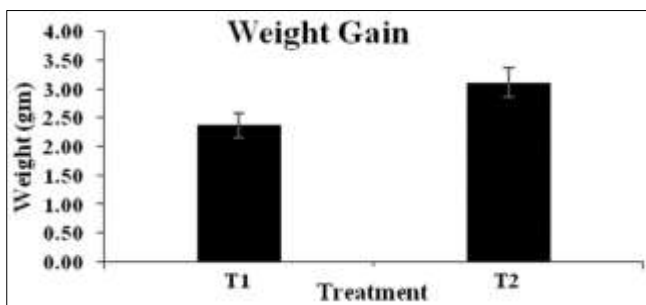
Treatment	Initial Length (cm) average ± se	Initial Weight (gm) average ± se	Final Length (cm) average ± se	Final Weight (gm) average ± se	Length Gain (cm) average ± se	Weight Gain (gm) average ± se
T1	9.52±0.10	5.365±0.20	10.745±0.09	7.73±0.09	1.22±0.11	2.365±0.21
T2	9.55±0.12	5.51±0.22	11.90±0.24	8.61±0.18	2.35±0.20	3.10±0.26



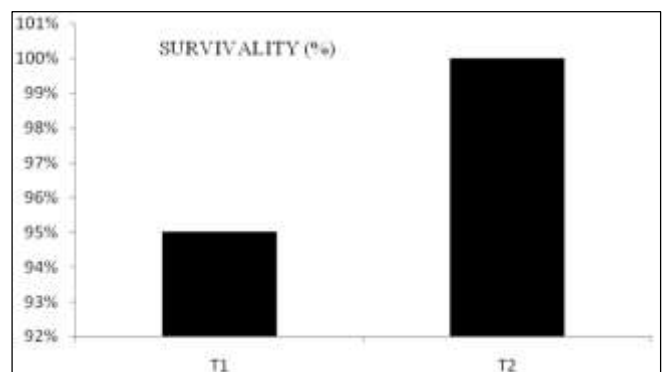
**Fig 8:** Average length gain (LG) by *H. fossilis* in different treatments



**Fig 10:** Specific growth rate of *H. fossilis* in both treatments



**Fig 9:** Average weight gain (WG) by *H. fossilis* in different treatments



**Fig 11:** Survival rate of *H. fossilis* in different treatments

**Table 2:** Condition factor (CF) of *H. fossilis* in T1 and T2

Treatment	Length-Weight	Number	Mean Length (cm)	Mean Weight (g)	Intercept, a	Slope, b	Coefficient, q	$W_{pred.}$	CF	Comments
T1	Initial	40	9.52	5.365	-4.96801	2.93394	0.00695	5.1671	1.038	Ideal
	Final	40	10.745	7.73	-2.99371	2.05422	0.05009	6.5777	1.175	Fatty
T2	Initial	40	9.55	5.51	-3.68473	2.36933	0.0251	5.2677	1.045	Ideal
	Final	40	11.9	8.61	-4.82775	2.69451	0.00801	6.3343	1.359	Fatty

Here,  $W_{pred.}$  = Predicted weight, CF= Condition Factor, Ideal condition = 0.95-1.05 ≈ 1.0; Fatty condition = >1.05; Not ideal = < 0.95

### 3.2 Water quality parameters

The water quality parameters such as temperature, pH, dissolved oxygen, ammonia, nitrite and nitrate were monitored regularly. The average temperature of T1 and T2 were 23.43°C and 23.46°C and the average pH of water were found 8.197 and 8.119 in treatment 1 and treatment 2 and dissolved oxygen were found 6.969 ppm and 6.806 ppm in treatment 1 and treatment 2 in an average. There were no significant difference ( $P > 0.05$ ) of temperature, pH and dissolved oxygen in water for T1 and T2 (Table 3). Ammonia, nitrite, nitrate were found 0.5mg/L, 1mg/L and 10mg/L in an average for both treatments.

**Table 3:** Results of water quality parameters measured during the study period

Water quality	Levene's Test for Equality of Variances		t-test for Equality of Means		
	F	P	t	Df	Sig. (2-tailed)
DO	0.46	0.50	0.60	70.00	0.55
pH	0.04	0.84	1.20	70.00	0.24
Temp	0.00	0.95	0.11	70.00	0.91

$P < 0.05$  significant

### 4. Discussion

To improve the quality and sustainability of aquaculture production various alternative methods have been developed [21]. Among these methods, probiotics have been shown to have an important role in aquaculture [32]. In addition, spirulina can be used as crucial component for animal feeding [17, 28], and may be an excellent source of useful nutrients for humans [12]. In the present study, better growth performance was observed in *Heteropneustes fossilis* maintained on the supplementary feed (artificial feed with probiotics and spirulina) than the artificial one. Similar findings were reported that growth in sea bass juvenile was better in the treated groups, used as a probiotic than the control [6]. Besides, growth performance and the mean survival rate was higher (96%) than that of the control group (87%) for African catfish (*Clarias gariepinus*) that was fed with probiotic [26]. During this study, higher survivability was observed in supplementary feed. It may be due to the immune performance of fish because non-specific immune responses could be enhanced by the use of spirulina with normal diet [8]. It was also recorded that length and weight gain was high (2.35±0.20 and 3.10±0.26), feed conversion ratio (9.9) was low, specific growth rate (0.34%) was high and fish health condition was comparatively better in the use of spirulina and probiotic. In previous study it was found that supplemented feed with probiotic improved the feed conversion ratio and growth rates for *Clarias gariepinus* [1]. It was also observed that higher survival, SGR, FCR values, final weight and daily weight gain [24] were obtained in probiotic treatments in *Oreochromis niloticus* [20] in aquaculture. Our results also agree with the authors [25, 5, 34], where they found better growth performance and FCR by using probiotics in common carp. It may be due to high ability of food absorption, better nutrient digestibility and increase of enzyme activities [10]. In this study it was not proved but the observation was same. In case of water quality parameters, insignificant ( $P < 0.05$ ) differences were noticed in this experiment. The temperature (25.1-20.3 for T1 and 25.2-20.4 for T2), average pH (ranged from 8.197±0.27 for T1 and 8.119±0.28 for T2) and dissolved oxygen (6.969 ppm for T1 and 6.806 ppm for T2) were recorded in different treatments during this study. Previous

studies measured temperature ranged from 24.60-30 (T1), 23.88-29.84 (T2) and 24.50-30.30 (T3) [18] on the growth and production performance of indigenous stinging catfish, *Heteropneustes fossilis*; and where, temperature ranged from 24.0- 29.5 in the experiment of reproductive biology of *Ompok bimaculatus* in the lotic waterbodies of Tripura [22]. It was also found that pH ranged from 8.10±0.43 (T1), 8.28±0.25 (T2) and 8.13±0.19 (T3) in the experiment of growth, survival and production of *H. fossilis* fingerlings under nursery ponds [23]. These findings were more or less similar to the present study. Our results also agree with other authors [27, 19], whom reported dissolved oxygen 0.8 to 7.85 ppm in carp polyculture system with silver carp; and dissolved oxygen 2 to 7.4 ppm in the development of culture technology of small indigenous fish.

However, the exchange of freshwater daily in both treatment aquarium, the fish excretes did not get any opportunity to create pollution in the experiment environment. As a result, there was an observation of insignificant differences in water quality parameters between the supplementary and artificial diet. Finally, further studies could be done to evaluate the probiotic and spirulina effects on water quality parameters with this species.

### 5. Conclusion

This study clearly demonstrated that the use of supplementary diet (probiotic, spirulina premix with artificial diet) has significant role in promoting the growth of *H. fossilis*. So, it can be concluded that spirulina and probiotic were potential feed ingredients and their uses played an effective role in *H. fossilis* culture. In this experiment, the probiotic was used only for the development for the water condition but not for the fish gut and was performed in laboratory condition. For the feasibility test, the trial could be further performed in field and further study was recommended for microbial assay to determine the gut biota to identify the better biological performance in growth of this species.

### 5. Acknowledgment

The authors would like to express their gratitude and thank to the Institute of Research and Technology, HSTU for funding this research. They also extend thanks to the Hatchery owner and laboratory technician for helping in the laboratory.

### References

1. Al-Dohail MA, Hashim R, Aliyu-Paiko M. Effects of the probiotic, *Lactobacillus acidophilus*, on the growth performance, haematology parameters and immunoglobulin concentration in African Catfish (*Clarias gariepinus*, Burchell 1822) fingerling. *Aquaculture Research*. 2009; 40(14):1642-52.
2. Andrade DLR, Castro BT, Mejía CJ, Mejía CG, Castillo MSGCV. La importancia de Spirulina en la alimentación acuícola. *Contactos. Journal of Biotecnología y Aplicaciones Industriales*. 2005; 57:13-16.
3. Azadi MA, Siddique MS. Fecundity of catfish (in Bangladesh) *Heteropneustes fossilis* (Bloch). *Bangladesh Journal of Zoology*. 1986; 14:33-39.
4. Belay A, Kato T, Ota Y. *Spirulina (Arthrospira)*: potential application as an animal feed supplement. *Journal of Applied Phycology*. 1996; 8:303-311.
5. Bogut I, Milakovic Z, Bukvic Z, Brkic S, Zimmer R.

- Influence of probiotic *Streptococcus faecium* M74 on growth and content of intestinal microflora in carp *Cyprinus carpio*. Czech Journal of Animal Science. 1998; 43:231-235.
6. Carnevali O, Vivo L, Sulpizio R, Gioacchini GI, Olivotto I, Silvi S, *et al.* Growth improvement by probiotic in European sea bass juveniles (*Dicentrarchus labrax*, L.), with particular attention to IGF-1, myostatin and cortisol gene expression. *Aquaculture*. 2006; 258:430-438.
  7. DoF (Department of Fisheries). Yearbook of Fisheries Statistics of Bangladesh, 2017-18. Fisheries Resources Survey System (FRSS), Department of Fisheries. Ministry of Fisheries, Dhaka, Bangladesh, 2019, 35-129.
  8. Duncan PL, Klesius PH. Effects of feeding Spirulina on specific and non-specific immune responses of channel catfish. *Journal of Aquatic Animal Health*. 1996; 8:308-313.
  9. El-Hindawy MM, Abd-Razic MA, Gaber HA, Zenhom MM. Effect of various level of dietary algae *Scenedesmus sp.* on physiological performance and digestibility of Nile tilapia fingerlings. 1st Scientific Conference of the Egyptian Aquaculture Society. Sharm El-Sheikh-Sinai, Egypt, 2006, 137-149.
  10. Fuller R. Probiotic in man and animals. *Journal of Applied Bacteriology*. 1989; 66:365-378.
  11. Giwojna P. Arrow crabs: Housebreaking the narrow snouted bristle horn. *Journal of Freshwater and Marine Aquarium*, 1987, 10:96.
  12. Glombitza KW, Koch M. Secondary metabolites of pharmaceutical potential. In Cresswell RC, Rees TAV, Shah M (eds), *Algal and Cyanobacterial Biotechnology*. Longman Scientific & Technical, Harlow, 1989, 161-238.
  13. Harel M, Koven W, Lein L, Bar Y, Beherens P, Stubblefield, Zohar Y, *et al.* Advanced DHA, EPA and ArA enrichment materials for marine aquaculture using single cell heterotrophs. *Aquaculture*. 2002; 213:347-362.
  14. Hossain MI, Kamal MM, Mannan MM and Buiya MAB. Effects of Probiotics on growth and survival of shrimp (*Penaeus monodon*) in coastal pond at Khulna, Bangladesh. *Journal of Scientific Research*. 2013; 5(2):363-370.
  15. IUCN. Red book of threatened fishes of Bangladesh. IUCN-The World Conservation Union, 2016, pp. 1-116.
  16. Khan MN, Islam AKMS and Hussain MG. Marginal analysis of culture of stinging catfish (*Heteropneustes fossilis*): Effect of different stocking density in earthen ponds. *Pakistan Journal of Biological Science*. 2003; 6(7):666-670.
  17. Kim SS, Rahimneja S, Kim KW, Lee KJ. Partial replacement of fish meal with Spirulina pacifica in diets for parrot fish (*Oplegnathus fasciatus*). *Turkish Journal of Fisheries and Aquatic Sciences*. 2013; 13(2):58.
  18. Kohinoor AHM, Khan MM, Yeasmine S, Mandol P, Islam MS. "Effects of stocking density on growth and production performance of indigenous stinging catfish, *Heteropneustes fossilis* (Bloch)". *International Journal of Agriculture Research Innovation and Technology*. 2012; 2:9-14.
  19. Kohinoor AHM. Development of culture technology of three small indigenous fish mola (*Amblypharyngodon mola*), punti (*Puntius sophore*) and chela (*Chela cachius*) with notes on some aspects of their biology. Ph.D. dissertation, Dept. of Fisheries Management, Bangladesh Agricultural University, Mymensingh, 2000, 363 p.
  20. Lara-Flores M, Olvera-Novoa MA, Guzmán n-Mendez BE and Lopez-Madrid W. Use of the bacteria *Streptococcus faecium* and *Lactobacillus acidophilus* and the yeast *Saccharomyces cerevisiae* as growth promoters in Nile tilapia (*Oreochromis niloticus*). *Aquaculture*. 2003; 216:193-201.
  21. Li J, Tan B, Mai K, Ai Q, Zhang W, Xu W, *et al.* Comparative study between probiotic bacterium *Arthrobacter XE-7* and chloramphenicol on protection of *Penaeus chinensis* post-larvae from pathogenic vibrios. *Aquaculture*. 2006; 253:140-147.
  22. Malla S, Banik S. Reproductive biology of an endangered catfish, *Ompok bimaculatus* (Bloch, 1794) in the lotic waterbodies of Tripura, North-East India. *International Journal of Fisheries and Aquatic Studies*. 2015; 2(4):251-260.
  23. Monir MS, Rahman S. Effect of stocking density on growth, survival and production of shing (*Heteropneustes fossilis*) fingerlings under nursery ponds in Northern region of Bangladesh. *International Journal of Fisheries and Aquatic Studies*. 2015; 2(3):81-86.
  24. Moriarty DJW. Disease control in shrimp aquaculture with probiotic bacteria. In Proceedings of the 8th International Symposium on Microbial Ecology; Atlantic Canada Society for Microbial Ecology; Halifax, NS, Canada, 1999, pp. 237-243.
  25. Noh SH, Han K, Won TH, Choi YJ. Effect of antibiotics, enzyme, yeast culture and probiotics on the growth performance of Israeli carp. *Korean Journal of Animal Science*. 1994; 36:480-486.
  26. Nwanna LC, Tope-Jegede OH, Jegede T. Use of probiotic as growth promoter in African catfish *Clarias gariepinus* (Burchell, 1822) juveniles. *American journal of Biotechnology and molecular sciences*. 2017; 6(1):17-24.
  27. Paul S. Comparison between carp polyculture system with silver carp (*Hypophthalmichthys molitrix*) and with small indigenous fish mola (*Amblypharyngodon mola*). MS Thesis, Department of Fisheries Management, Bangladesh Agricultural University, Mymensingh, 1998, 85p.
  28. Qureshi MA, Kidd MT, Ali RA. *Spirulina platensis* extract enhances chicken macrophage functions after in vitro exposure. *Journal of Nutritional Immunology*. 1989; 3(4):35-45.
  29. Rahman MA, Gheyasuddin H, Rasid MH, Choudhury MFZ. Proximate composition and native quality of freshwater Zeol fishes of Bangladesh. *Bangladesh Journal of Fisheries*. 1982; 2-5:34-43.
  30. Reddy AS, Reddy VS, Golovkin MA. Calmodulin binding protein from Arabidopsis is induced by ethylene and contains a DNA-binding motif. *Biochem. Biophys. Res. Commun*. 2000; 279:762-769.
  31. Saha KC, Guha BC. Nutritional investigation on Bengal fish. *Indian Journal of Medical Research*. 1939; 26:921-927.
  32. Skjermo J, Vadstein O. Techniques for microbial control in the intensive rearing of marine larvae. *Aquaculture*. 1999; 177:333-343.
  33. Teimouri M, Amirkolaie AK, Yeganeh S. Effect of

- Spirulina platensis* meal as a feed supplement on growth performance and pigmentation of rainbow trout (*Oncorhynchus mykiss*). World Journal of Fish and Marine Sciences. 2013; 5(2):194-202.
34. Yanbo W, Zirong X. Effect of probiotics for common carp (*Cyprinus carpio*) based on growth performance and digestive enzyme activities. Animal Feed Science and Technology. 2006; 127:283-292.