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Effect of prebiotic mannan oligosaccharide on growth performance, survival and proximate composition of reba carp, *Cirrhinus reba* fry

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Abstract
This study was conducted to evaluate the effect of mannan oligosaccharide (MOS) supplementation on the growth performance, survival and proximate composition of reba carp, *Cirrhinus reba* fry. Triplicate groups of reba fry (0.25±0.002 g) were fed twice per day at 10% of body weight for 12 weeks with 0 (control), 0.2 and 0.4% MOS diets. Significantly highest (P<0.05) weight gain and specific growth rate were observed in fish fed with 0.2% MOS diet. Higher survival rate (100%) where observed in fish feed with 0.2% MOS diet compared to two other feeding trails. Significantly highest hepatosomatic and viscerosomatic index and lowest intraperitoneal fat were noted when the fish fed with 0.2% MOS diet compared to those fish fed with 0.4% MOS diet. Significantly highest and lowest protein content were observed in fish fed with 0.2% MOS supplemented diet and control diet respectively, whereas the lipid and ash content was significantly highest at 0.4% MOS diet. Thus, the results of this study indicated that dietary administration of MOS at 0.2% may influence the growth performance and body composition of reba fry.

Keywords: Prebiotic, mannan oligosaccharide, growth performance, survival, proximate composition, *Cirrhinus reba*

1. Introduction
Cirrhinus reba is a commercially significant freshwater minor carp (minnows) species belongs to the family cyprinidae under the order cypriniformes [34]. The rapid expansion of aquaculture has been limited due to the unavailability of natural resources, such as land, which in turn has led to the intensification of fish farming [39]. The increasing intensification and commercialization of aquaculture systems has accelerated the outbreak of diseases that are responsible for huge fish losses [13]. Antibiotics have been used at sub-therapeutic levels to prevent disease and enhance the growth performance in fish farms. However, adverse collateral effects, such as the evolution of antibiotic-resistant bacteria, make the use of antibiotic growth promoters relatively undesirable [16]. Recurrent use of antibacterial products to control the much feared pathogens colonizing the intestinal environment involves the risk of simultaneously killing the useful intestinal micro biota necessary for optimal utilization of feed nutrients. It is therefore useful to use prebiotics to restore the beneficial intestinal microbiota and encourage its growth [53]. From the early years of the present century, prebiotics have been extensively tested for potential beneficial effects on fish growth, survival and health status [10, 25, 32, 44, 68]. Furthermore, prebiotics can improve disease resistance, intestinal villous surface area and microvillus length [30, 57, 61, 64].
Among the established prebiotics mannan oligosaccharide (MOS) is most commonly used as the dietary supplementation for fish and crustacean species [58]. MOS is a natural alternative product for anti-bacterial growth factors, which is known for connecting pathogenic microorganisms and toxins to their chemical structure [45, 49]. In this way, pathogenic bacterial growth is prevented and consequently the harmful effect of microflora metabolites is decreased. The use of MOS as a feed supplement includes its inhibitory impacts on pathogenic bacteria and can augment the immune response of the animal. As a result, the health and performance of the animal may improve if MOS is included in the diet [14]. Several study reported the effect of MOS on growth enhancement of European bass, *Dicentrachus labrax* [63]; gilthead sea bream, *Sparus aurata* [19, 33]; channel catfish, *Ictalarus punctatus* [65] and gibel carp juveniles, *Carassius auratus gibelio* [4].

In contrast to the progress made in other species, the effects of MOS on *C. reba* have received little attention. This study was initiated to evaluate the effectiveness of MOS in promoting growth performance and to determine its influence on survival and proximate composition of reba carp fry.

2. Materials and Methods

2.1 Experimental design and feeding trial

The feeding trial was conducted at the Fisheries Biology and Genetics laboratory, Hajee Mohammad Danesh Science and Technology University, Dinajpur. Reba carp fry were collected from the hatchery complex of Caritas, Bochagonj, Dinajpur, Bangladesh which were produced under the research project of NATP-CRG 488 funded by USAID and Ministry of Agriculture, government of Bangladesh. The fish were transported in oxygenated plastic bags filled with freshwater. The fish were acclimatized for two weeks prior to the initiation of the study and were fed with a commercial feed. Thereafter, the fish (average weight 0.25g) were randomly distributed into nine glass aquaria measuring (18 cm × 10 cm × 10 cm) with a stocking density of 10 fish per aquarium. Three replicate aquaria for each treatment were established. During the experimental period, the reba fry were fed the respective experimental diets based on a percentage of body weight (10%) twice a day for a period of 12 weeks. Throughout the experiment, water was supplied from a deep water tube well. Water quality parameters such as temperature, DO and pH were monitored regularly. Faeces were regularly siphoned out. Aquaria were cleaned weekly to reduce the risk of accumulation of nitrogenous waste.

2.2 Diet preparation

Three experimental diets were prepared. Each contained a different dose of MOS 0 (control), 0.2% and 0.4% MOS (International Food Grade, Laboratory of USA, Purity > 90%). The concentrations of MOS were carefully chosen on the basis of previous studies [8, 63]. Fish meal and soybean meal were used as protein sources, while soybean oil and fish oil were used as lipid sources. To ensure similar energy levels in all experimental diets carbohydrate sources such as corn starch and wheat flour were used [50]. Diets were inclined by thoroughly mixing the feed ingredients (Table 1) in a food mixer for 30 minutes at a capacity of 1 kg. The feed ingredients were also mixed for another 10 minutes after adding of soybean oils and fish oil [56]. Adequate amount of water was then added to make it dough which was then extruded through a pelletizing machine to make 3 mm diameter pellets. The pellets were air dried for overnight, and then broken into smaller pieces by hand. The resultant pellets were then packed separately in plastic bags in order to store in a freezer at -20 ºC throughout the feeding trial.

Table 1: Ingredients used and proximate composition of the experimental diets containing varying levels of mannan oligosaccharide (MOS) (g kg^{-1})

Ingredients	Treatments		
	Control	0.2% MOS	0.4% MOS
Fish meal[1]	247.2	247.2	247.2
Soybean meal	280.1	280.1	280.1
Corn Starch	97.5	97.5	97.5
Wheat floor	270.0	268.0	266.0
Soybean Oil	32.60	32.60	32.60
Fish Oil	32.60	32.60	32.60
Vitamin mix[2]	20.0	20.0	20.0
Mineral mix[3]	20.0	20.0	20.0
MOS[4]	0.0	2.0	4.0

1. Danish fishmeal: crude protein, 720; crude lipid, 50.
2. Vitamin mix kg^{-1} (Rovithai Ltd 700/437 Chonburi THAILAND): Vitamin A 50 MIU, Vitamin D_3 10 MIU, Vitamin E 130g, Vitamin K_3 10g, Vitamin B_1 10g, Vitamin B_2 25g, Vitamin B_6 16g, Vitamin B_{12} 100mg, Niacin 200g, Pantothenic Acid 56g, Folic Acid 8g, Biotin 500mg, Antioxidant 0.200g and Anticake 20g.
3. Mineral mix kg^{-1}: Calcium phosphate (monobasic) 397.5 g; Calcium lactate 327 g; Ferrous sulphate 25 g; Magnesium sulphate 137 g; Potassium chloride, 50 g; Sodium chloride, 60 g; Potassium iodide, 150 mg; Copper sulphate 780 mg; Manganese oxide 800 mg; Cobalt carbonate 100 mg; Zinc oxide 1.5 g and Sodium selenite 20 mg.
4. MOS (Mannan oligosaccharide, International Food Grade, Laboratory of USA, Purity > 90%)

2.3 Water quality assessment

In the present study, water samples were collected from each aquarium. Recording on the spot data and collection of samples were made between 9.00 to 11.30 A.M. Water temperatures, pH, dissolved oxygen (DO) were recorded every 10 days interval between 9.00 am to 11.30 am. Temperature of water was taken from each aquarium by using a standard mercury thermometer. pH of water was taken from each tank by using a pH meter. The DO was measured by using a digital DO meter.

2.4 Sampling procedure

The weight of each fish was determined at the start and end of the experiment; the combined weight of the fish in each aquarium was measured fortnightly to monitor somatic growth and to adjust the amount of feed to be given. Survival of the fish was monitored throughout the experimental period. After 12 weeks, three fish from each replicate aquarium were randomly selected and analysed separately for whole body proximate composition and body indices. The selected fishes were killed by keeping them in a refrigerator. After being killed, the individual fishes were weighed immediately and kept in an oven for drying. Thereafter, the dried fishes were then grinned using a blender machine in order to determine the moisture, crude protein, crude lipid and ash content of whole fish body. The weight of the whole fish body, liver, intraperitoneal fat, and viscera were recorded to obtain the hepatosomatic index (HSI), intraperitoneal fat (IPF) and viscerosomatic index (VSI).

2.5 Growth performance, survival, feed utilization and body indices

The following growth performances, survival, feed utilization parameters and body indices were evaluated using the following equations:

2.5.1 Growth parameters, survival and feed utilization parameters

Weight gain (g)

Weight gain (g) = Final weight (g) –Initial weight (g) [62]

Specific growth rate (SGR % day^{-1})

The SGR is the momentary change in weight of fish calculated as the per cent increase in body weight per day over a given time interval and is written as [35]:

$$SGR\ (\%\ day^{-1}) = \frac{In\ w_2 - In\ w_1}{T_2 - T_1} \times 100$$

Where,
W_1 = the initial live body weight (g) at time T_1 (day).
W_2 = the final live body weight (g) at time T_2 (day)

Survival rate (%) [35]

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

Feed conversion ratio (FCR)
Feed conversion ratio is defined as the amount of dry feed intake per unit live weight gain. It is calculated as [37]:

$$FCR = \frac{Total\ dry\ feed\ intake\ (g)}{Total\ wet\ weight\ gain\ (g)}$$

To calculate FCR, the dry weight of the feed was obtained by using a correction for the analyzed moisture content of the diet. FCR is a measure of the degree of gross utilization of feed for growth.

Protein efficiency ratio (PER)
Protein efficiency ratio is defined as the gain in weight of fish per gram of crude protein fed. Protein efficiency ratio is calculated as [35]:

$$PER = \frac{Wet\ weight\ gain\ (g)}{Total\ protein\ intake\ (g)} \times 100$$

PER gives an indication of the efficiency of utilization of dietary protein.

2.5.2 Determination of body indices
At the end of the present experiment, body indices of reba carp such as hematosomatic index (HSI), intraperitoneal fat (IPF) and viscera somatic index (VSI) were determined by using the following formulae:

$$Hematosomatic\ index\ (HSI\%) = \frac{Liver\ Weight\ (g)}{Body\ weight\ (g)} \times 100$$

$$Intraperitoneal\ fat\ (IPF\%) = \frac{Intraperitoneal\ fat\ weight\ (g)}{Body\ weight\ (g)} \times 100$$

$$Viscerasomatic\ index\ (VSI\%) = \frac{Viscera\ weight\ (g)}{Body\ weight\ gain\ (g)} \times 100$$

2.6 Proximate Composition Analysis
Whole body fish composition was determined by using the standard protocols as mentioned by the Association of Official Analytical Chemists [11]. The moisture content of the sample was evaluated by drying the samples in the hot air oven at 105 ℃ for 12 hours until constant weight was obtained. The moisture free samples were then used in order to determine the crude protein, lipid and ash content. The crude protein content of the fish samples was calculated indirectly by determining the total nitrogen content of the sampled fish by a Kjeldahl method using kjeldhal apparatus. The crude lipid content of the samples were determined by removing the lipid from the samples after homogenizing it in 60 ml of chloroform and methanol solution in a ratio of 2:1 [26] and thereafter the solvent was evaporated by heating in a oven at 80 °C. The moisture free samples were taken in porceline basin made crucible and weighed. Thereafter, the ash content was measured by igniting the samples in a muffle furnace at a temperature of 550 °C for 6 hours.

2.7 Data Analysis
All data were tested using one-way analysis of variance (ANOVA). Significant results (P<0.05) were further tested using one-way ANOVA followed by Duncan's Multiple Range Test [24] to identify significant difference between means. The data were expressed as average ± SE and statistical analysis was performed using SPSS version 22 and Microsoft Office Excel for window.

3. Results
3.1 Water Quality Parameters
Water temperature, pH and DO were monitored fortnightly for a period of 12 weeks, which were ranged between 23.70-24.16 °C, 8.23-8.50 and 7.26-7.43 ppm respectively (Table 2).

Table 2: Water quality parameters of reba carp, *Cirrhinus reba* fingerlings raised in the experimental aquaria

Parameters	Treatments		
	Control	0.2% MOS	0.4% MOS
Temperature (°C)	23.76±0.31	24.16±0.28	23.70±0.25
pH	8.23±0.08	8.46±0.14	8.50±0.05
DO (ppm)	7.30±0.11	7.43±0.12	7.26±0.12

3.2 Growth Performance, survival and feed utilization parameters
Growth performance, survival and feed utilization parameters of reba carp fed with varying levels of MOS diets for 12-week are presented in Table 3. Generally, growth performance, such as weight gain and SGR (%) were significantly enhanced (P<0.05) in fish fed 0.2% (1.26±0.03; 2.17±.01) and followed by control (1.18±.04; 2.06±.03) and 0.4% MOS diets (1.04±.05; 1.97±.07). Though FCR and PER did not differ significantly among the treatment but the lowest value of FCR (10.80±0.08) and highest value of PER (0.27±0.01) was found in 0.2% MOS diet. Higher survival rate (%) was found in all three treatments which ranged from 97-100%. The non significantly higher (P>0.05) survival rate (100%) was observed in 0.2% MOS supplemented diet.

Table 3: Growth performance and feed utilization parameters of reba fingerlings, *Cirrhinus reba* fed a diet containing varying levels of mannan oligosaccharide (MOS) for 12 weeks

Parameters	Treatments		
	Control	0.2% MOS	0.4% MOS
Initial Av. Wt.	0.25±0.01	0.24±0.01	0.24±0.01
Final Av. Wt.	$1.43±0.05^{ab}$	$1.50±0.04^{b}$	$1.28±0.05^{a}$
Wt. Gain	$1.18±.04^{ab}$	$1.26±0.03^{b}$	$1.04±.05^{a}$
SGR (%)	$2.06±.03^{ab}$	$2.17±.01^{b}$	$1.97±.07^{a}$
FCR	10.96±0.40	10.80±0.08	11.20±0.48
PER	0.26±0.01	0.27±0.01	0.25±0.01
Survival Rate (%)	96.66±3.33	100±.00	96.66±3.33

All values are presented as mean ± SE obtained from three replicates aquaria, (n=3); values with different superscripts in the same row indicate significant differences ($P<0.05$); SGR, specific growth rate; FCR, food conversion ratio; PER, protein efficiency ratio.

3.3 Body Indices

The effect of dietary MOS on the body indices of reba carp is shown in Table 4. Supplementation of MOS in reba carp showed a significant variation on the all body indices. Significantly lower ($P<0.05$) HSI and VSI were prominent when the reba carp fed with 0.4% MOS diets compared to 0.2% diet. While, IPF was significantly lowest ($P<0.05$) in those fishes which fed with 0.2% MOS diet over the 0.4% fed group.

Table 4: Body indices of Reba fingerlings, *Cirrhinus reba* fed a diet containing varying levels of mannan oligosaccharide (MOS) for 12 weeks

Parameters	Treatments		
	Control	0.2% MOS	0.4% MOS
HSI (%)	1.16±.09^{ab}	1.27±0.12^{b}	0.85±0.11^{a}
IPF (%)	0.71±0.02^{ab}	0.68±0.04^{a}	0.82±0.05^{b}
VSI (%)	7.91±0.26^{ab}	8.32±0.22^{b}	7.46±0.14^{a}

All values are presented as mean±SE obtained from three replicates aquaria, (n=3); Data with different superscripts in the same row indicate significant differences ($P<0.05$); HSI, Hepatosomatic index; IPF, Intraperitoneal fat; VSI, Viscerosomatic index; MOS, Mannan oligosaccharide.

3.4 Whole Body Proximate Composition

Proximate composition of whole body of fish fed with various concentrations of MOS is shown in Table 5. Moisture content was significantly ($P<0.05$) higher in fish fed with 0.2 and 0.4% MOS treated diets compared to the control fed group. Significantly higher ($P<0.05$) body protein content was detected in fish treated with 0.2% MOS diet (16.15±0.03) compared to fish fed with 0.4% MOS (15.51±0.14) and the control group (15.39±0.13). Whereas, significantly highest ($P<0.05$) lipid and ash content /were detected in fish fed 0.4% MOS diet when compared to fish that were fed MOS diet at 0.2% and the control.

Table 5: Proximate composition of whole body of reba, *Cirrhinus reba* fingerlings, fed varying levels of mannan oligosaccharide (MOS) for 12 weeks

Treatments	Treatments			
	Moisture (%)	Protein (%)	Lipid (%)	Ash (%)
Control	77.61±0.02^{a}	15.39±0.13^{a}	0.65±0.01^{a}	3.92±0.15^{a}
0.2% MOS	78.53±0.22^{b}	16.15±0.03^{b}	0.97±0.09^{b}	3.83±0.18^{a}
0.4% MOS	78.47±0.29^{b}	15.51±0.14^{a}	1.21±0.06^{c}	4.47±0.13^{b}

Data presented as mean ± SE, (n=3). Values with different superscripts in the same column indicate significant differences ($P<0.05$).

4. Discussion

Nowadays, immunostimulants such as prebiotics have shown promise as preventive and ecologically friendly alternatives to antibiotics in aquaculture [59]. In aquaculture prebiotics are basically used for the enhancement of fish growth and disease resistance, improving commercial feasibility and sustainability of aquaculture [54]. A number of studies have revealed that the gastrointestinal (GI) tract microbiota in aquatic animals can be influenced through the application of prebiotics that can alter the conditions of the GI tack to favour certain bacterial species which may enhance growth and reduce disease susceptibility of the host organism [15, 29, 46]. Accordingly MOS, which is derivative from the cell wall of the yeast *S. cerevisiae* and is used as a dietary supplement in aquaculture [66]. MOS may also enhance health by stimulating antibody production [17].

The current research is the first time attempt to demonstrate the effectiveness of MOS on the enhancement of growth and survival of reba carp fry. In the present study, the reba carp fry responded positively to supplementation of the diet at 0.2% based on their significantly improved final average wt., WG and SGR% values compared to other treatments. Comparatively better FCR and PER were also observed in the fishes fed with 0.2% MOS supplemented.

Earlier studies have shown different response of aquatic animals to MOS supplementation; growth enhancement was observed in rainbow trout [67], green tiger prawn [30], tilapia [47], European sea bass [63], gilthead sea bream [33]; gibel carp juveniles [4], shrimp [69], white leg shrimp [7], juvenile striped catfish [8], clownfish [23]. On the contrary, no growth enhancement was found in Gulf sturgeon [51], hybrid tilapia [30], Kutum [6], giant sturgeon [40], clownfish fingerlings [52] and common carp [43]. The effect of MOS supplementation in diets has not only shown species specific trend but also it depends on the inclusion level of MOS. In the current study the lower MOS concentration at 0.2% have positive influence on the growth performance of reba carp. Similar to the present study, 0.2% MOS is sufficient to improve the growth performance of shrimp (*L. vannamei*) [69], Nile tilapia (*O. niloticus*) [1], yellow catfish (*Pelteobagrus fulvidraco*) [66] and rainbow trout (*O. mykiss*) [42].

On the other hand, 0.1% MOS exhibited better growth on common carp [5], rainbow trout [18] and clownfish [23]; 0.25% MOS exhibited better growth on *Cyprinus carpio* [37]; 0.3% MOS showed better growth in crayfish [41]; 0.4–0.41% MOS showed better growth on zebra fish (*Danio rerio*) [27]; 0.45% MOS showed better growth performance in gibel carp juveniles [4], 0.6% or 0.8% MOS had significantly higher growth performance in striped catfish (*Pangasianodon hypophthalmus*) [8]. From the above studies, it can assume that the effects of dietary MOS could differ among the aquatic animals; hence it has to be determining the inclusion level of MOS before applying it on the specific aquatic animals [69].

In addition to improving the health status of the animal, prebiotics may affect growth, feed utilization and body composition [18]. Proximate composition of whole body,

moisture, ash, protein contents of the reba carp were considerably differed ($P<0.05$) among all the experimental treatments. Similar findings also have been stated in rainbow trout [67], in common carp [5] and in Nile tilapia [1]. Contrary some previous research reported that the supplementation of MOS did not influence the body composition of Atlantic salmon [32], rainbow trout [20], common carp [12] and grey mullet [2].

In the present study, the whole body protein content improved significantly ($P<0.05$) in fish given the 0.2% MOS supplemented diet compared with control. In agreement with the result of this study, 0.2% MOS supplementation shown to increase body protein in Atlantic Salmon Smolts (*Salmo salar*) [21], in Nile tilapia (*O. niloticus*) [1]. Furthermore, previous study [30] reported that, body protein content increased with the increasing levels of dietary MOS for hybrid tilapia. Similar results were also observed in the case of rainbow trout [67] and Atlantic salmon [32]. Although the protein concentration in the body may be affected by dietary prebiotics MOS, but the response could be differ depending on the animal species, [30, 64, 67]. So before application, it is necessary to determine the dietary supplementation level and duration of use so that the results may not decisive [32].

For better fish production good water quality always be the first priority [38]. Generally growth, feed efficiency and feed consumption of fish are dependent on few environmental factors [28]. The water quality parameters such as temperature, pH and DO were measured in the present study throughout the experimental period. However, all of those parameters did not differ significantly among treatments.

In the present study, the water temperature (ºC) ranged from 23.70-24.16°C. This values were more or less similar to the findings of other authors as 25 to 35°C [9], 25±1ºC [30], 22.7 ± 0.7 °C [40], 25.5±1.5 ºC [3], 26.12±0.58°C [31] and 27 to 28°C [48]. In the present study pH ranged from 8.23 to 8.50. This values were more or less similar to the findings of other authors as 7.82-8.33 [30], 8.2±0.19 [47], 7.9±0.2 [3], 8.04±0.08 [31] and 8.0–8.3 [23], and it is stated that the suitable pH range for fish culture is between 6.7 and 9.5 and ideal pH level is between 7.5 and 8.5 [60], 0.03 to 9.03 is required for carp SIS polyculture [55]. During the study DO ranged from 7.26 to 7.43 ppm. More or less similar findings were reported by some authors, 7.1±0.07 [47], 7.0 ± 0.5 ppm [40] and 6.55±0.49 ppm [31]. According to DoF [22] the range of dissolved oxygen (DO) content for fish culture should be 5.0-8.0 ppm. So, it can be said that the water parameters (temperature, pH and DO) were suitable for experimental fish during study period.

5. Conclusion

In conclusion, the results observed in this study indicated that MOS is a beneficial dietary supplement, appears to have a more positive influence on the enhancement of the growth performance and feed utilization of reba carp. It is also able to improve some important body composition. The results of the present study revealed that MOS supplementation at 0.2% appears to be most effective dose in influencing the growth performance, survival, feed utilization and body composition of reba carp. Therefore, the results suggest that the inclusion of MOS at 0.2% concentration can be effectively used as a feed additive for reba carp (*C. reba*) culture.

References

1. Ahmad MH, El-Mousallamy A, Awad SMM, El-Naby ASA. Evaluation of Bio-Mos® as a feed additive on growth performance, physiological and immune responses of Nile tilapia, *O. niloticus* (L). Journal of Applied Sciences Research. 2013; 9(13):6441-6449.
2. Akbary P, Jahanbakhshi A. Growth yield, survival, carcass quality, haematological, biochemical parameters and innate immune responses in the grey mullet (*Mugil cephalus* Linneaus, 1758) fingerling induced by Immunogen® prebiotic. Journal of Applied Animal Research. 2018; 46(1):10-16.
3. Akrami R, Chitsaz H, Lakzaei F. Effect of dietary A-Max supplementation as a prebiotic on growth performance and hemato- immunological parameters of great sturgeon (*H. huso* Linnaeus, 1758) juveniles. Iranian Journal of Fisheries Science. 2018; 17(2):251-266.
4. Akrami R, Chitsaz H, Hezarjaribi A, Ziaei R. Effect of dietary mannan oligosaccharide (MOS) on growth performance and immune response of gibel carp juveniles (*Carassius auratus gibelio*). Journal of Veterinary Advances. 2012; 2(10):507-513.
5. Akrami R, Mansour MR, Chitaz H, Ziaei R. Effect of dietary mannan oligosaccharide on growth performance, survival, body composition and some hematological parameters of carp juvenile (*C. carpio*). Journal of Aquaculture Feed Science and Nutrition. 2012; 4(3):54-60.
6. Akrami R, Karimabadi A, Mohammadzadeh H, Ahmadifar E. Effect of dietary mannan oligosaccharide on growth performance, survival, body composition and salinity stress resistance in Kutum (*Rutilus frisii*) fry stage. Journal of Marine Science and Technology. 2010; 8(3, 4):47-57.
7. Aktas M, Ciger O, Genç E, Genç MA, Cavdar N. Effects of Mannan Oligosaccharide and serotonin on molting, growth, body composition and hepatopancreas histology of white leg shrimp (*Litopenaeus vannamei*) (Boone 1931). Turkish Journal of Fisheries and Aquatic Sciences. 2014; 14(1):205-211.
8. Akter MN, Sutriana A, Talpur AD, Hashim R. Dietary supplementation with mannan oligosaccharide influences growth, digestive enzymes, gut morphology, and microbiota in juvenile striped catfish, *Pangasianodon hypophthalmus*. Aquaculture International. 2016; 24(1):127-144.
9. Aminul IM. Qualities of water and soil in Aquaculture, Fish week compilation, 96. DoF Publication, Ramna, Dhaka-1000, 1996.
10. Amirkolaie AK, Karimzadeh S, Jafary AM. The effects of dietary supplement of immunogen on growth performance and visceral and hepatic somatic indices of Juvenile Rainbow Trout, (*Oncorhynchus mykiss*) (Walbaum). Asian Fisheries Science. 2013; 26:232-242.
11. AOAC. Association of Official Analytical Chemists. Official methods of analysis of AOAC International, 16th edn, vol. 1, Arlington, VA, USA. 1997, 1-3.
12. Bahrekazemi M, Jaber N. Effect of diets containing different levels of prebiotic mito on the growth factors, survival, body composition, and hematological parameters in common carp *Cyprinus carpio* fry. Journal of Marine Biology and Aquaculture. 2017; 3(1):1-6.
13. Bondad-Reantaso MG, Subasinghe RP, Arthur JR, Ogawa K, Chinabut S, Adlard R *et al*. Disease and health management in Asian aquaculture. Veterinary Parasitology. 2005; 132(3, 4):249-272.
14. Bovera F, Lestingi A, Iannaccone F, Tateo A, Nizza A.

Use of dietary mannan oligosaccharides during rabbit fattening period: Effects on growth performance, feed nutrient digestibility, carcass traits, and meat quality. Journal of Animal Science. 2012; 90(11):3858-3866.
15. Burr G, Gatlin D, Ricke S. Microbial ecology of the gastrointestinal tract of fish and the potential application of prebiotics and probiotics in finfish aquaculture. Journal of World Aquaculture Society. 2005; 36(4):425-436.
16. Cabello FC. Heavy use of prophylactic antibiotics in aquaculture: A growing problem for human and animal health and for the environment. Environmental Microbiology. 2006; 8(7):1137-1144.
17. Cetin N, Guclu BK, Cetin E. The effects of probiotic and mannanoligosaccharide on some haematological and immunological parameters in turkeys. Journal of Veterinary Medicine Series A. 2005; 52(6):263-267.
18. Denji KA, Mansour MR, Akrami R, Ghobadi S, Jafarpour SA, Mirbeygi SK. Effect of dietary prebiotic mannan oligosaccharide (MOS) on growth performance, intestinal microflora, body composition, haematological and blood serum biochemical parameters of rainbow trout (*Oncorhynchus mykiss*) juveniles. Journal of Fisheries and Aquatic Science. 2015; 10(4):255-265.
19. Dimitroglou A, Merrfield DL, Spring P, Sweetman J, Moate R, Davies SJ. Effects of mannan oligosaccharide (MOS) supplementation on growth performance feed utilization, intestinal histology and gut microbiota of gilthead sea bream (*Sparus aurata*). Aquaculture. 2010; 300(1, 4):182-188.
20. Dimitroglou A, Merrifield DL, Moate R, Davies SJ, Spring P, Sweetman J *et al*. Dietary mannan oligosaccharide supplementation modulates intestinal microbial ecology and improves gut morphology of rainbow trout, *O. mykiss* (Walbaum). Journal of Animal Science. 2009; 87(10):3226-3234.
21. Dimitroglou A, Reynolds P, Ravnoy B, Johnsen F, Sweetman J, Johansen J *et al*. The effect of mannan oligosaccharide supplementation on Atlantic salmon smolts (*Salmo salar* L.) fed diets with high levels of plant proteins. Journal Aquaculture Research and Development. 2011; S1(011): 1-6.
22. DoF. Matshapakkha Sankalan (Bengali) Department of Fisheries, Ministry of Fisheries and Livestock, Government of the people's Republic of Bangladesh. 1996, 81.
23. Do-Huu H, Nguyen TK. Influence of dietary mannan oligosaccharides supplementation on fingerling clownfish, *Amphiprion ocellaris* (Cuvier, 1830). Journal of Coastal Life Medicine. 2017; 5(8):325-329.
24. Duncan DB. Multiple range and multiple (F) tests. Biometrics. 1955; 11(1):1-42.
25. Ebrahimi G, Ouraji H, Khalesi MK, Sudagar M, Barari A, Zarei D *et al*. Effects of a prebiotic, Immunogen, on feed utilization, body composition, immunity and resistance to *Aeromonas hydrophila* infection in the common carp *Cyprinus carpio* (Linnaeus) fingerlings. Journal of Animal Physiology and Animal Nutrition. 2012; 96(4):591-599.
26. Folch J, Lees M, Stanley GS. A simple method for the isolation and purification of total lipids from animal tissues. Journal of Biological Chemistry. 1957; 226(1):497-509.
27. Forsatkar MN, Nematollahi MA, Rafiee G, Farahmand H, Martínez-Rodríguez G. Effects of prebiotic mannan oligosaccharide on the growth, survival, and anxiety-like behaviors of zebrafish (*Danio rerio*). Journal of Applied Aquaculture. 2017; 29(2):183-196.
28. Fry FEJ. The effect of environmental factors on the physiology of fish. Fish physiology. 1971; 6:1-98.
29. Gatlin III DM. Nutrition and fish health. In "Fish Nutrition" (J. Halver and RW Hardy, eds.). Academic Press, San Diego, CA, USA. 2003, 671-702.
30. Genc MA, Yilmaz E, Genc E, Aktas M. Effects of dietary mannan oligosaccharides (MOS) on growth, body composition, and intestine and liver histology of the Hybrid Tilapia (*O. niloticus*, *O. aureus*). The Israeli Journal of Aquaculture. 2007; 59(1):10-16.
31. Gharaei A, Rayeni MF, Ghaffari M, Akrami R, Ahmadifar E. Influence of dietary prebiotic mixture (α - mune) on growth performance, haematology and innate immunity of Beluga sturgeon (*H. huso*) juvenile. International Journal of Aquatic Biology. 2016; 4(4):277-284.
32. Grisdale-Helland B, Helland SJ, Gatlin DM. The effects of dietary supplementation with mannanoligosaccharide, fructooligosaccharide or galactooligosaccharide on the growth and feed utilization of Atlantic salmon (*Salmo salar*). Aquaculture. 2008; 283(1-4):163-167.
33. Gultepe N, Salnur S, Hossu B, Hisar O. Dietary supplementation with Mannan oligosaccharides (MOS) from Bio-Mos enhances growth parameters and digestive capacity of gilthead sea bream (*Sparus aurata*). Aquaculture Nutrition. 2011; 17(5):482-487.
34. Gupta S. Some observations on the biology of *Cirrhinus reba* (Cuvier). Journal of Fish Biology. 1975; 7(1):71-76.
35. Helland SJ, Grisdale B, Nerland S. A simple method for the measurement of daily feed intake of groups of fish in tanks. Aquaculture. 1996; 139(1, 2):157-163.
36. Hevroy EM, Espe M, Waagbo R, Sandness K, Rund M, Hemer GI. Nutrition utilization in Atlantic salmon (*Salmo salar*) fed increased level of fish protein hydrolysate during a period of fast growth. Aquaculture Nutrition. 2005; 11(4):301-313.
37. Hunt AO, Yilmaz FO, Berkoz M. An investigation of effects of mannan oligosaccharide on growth, body composition, and lysozyme activity of carp (*Cyprinus carpio*). Aquaculture Studies. 2016; 16(1):027-035.
38. Jhingran VG. Fish and fishes of India (3rd ed.). Hindustan Publication of Corporation, New Delhi. 1991, 727.
39. Klinger D, Naylor R. Searching for Solutions in Aquaculture: Chartinga Sustainable Course. Annual Review of Environment and Resources. 2012; 37:247-276.
40. Mansour MR, Akrami R, Ghobadi SH, Denji KA, Ezatrahimi N, Gharaei A. Effect of dietary mannan oligosaccharide (MOS) on growth performance, survival, body composition, and some hematological parameters in giant sturgeon juvenile (*Huso huso* Linnaeus, 1754). Fish Physiology and Biochemistry. 2012; 38(3):829-835.
41. Mazlum Y, Yilmaz E, Genc MA, Guner O. A preliminary study on the use of mannan oligosaccharides (MOS) in fresh water crayfish, *Astacus leptodactylus eschscholtz*, 1823 juvenile diets. Aquaculture International. 2011; 19:111-119.
42. Minguez C, Webster PG, Villa M. Effect of a prebiotic supplementation of mannan oligosaccharide on growth

traits and mortality of rainbow trout (*O. mykiss*). Aquaculture, Aquarium, Conservation and Legislation. 2016; 9(6):1260-1264.
43. Moghaddam PM, Keyvanshokooh S, Nejad SZ, Salati AP, Zanoosi HP. Effects of mannan oligosaccharide supplementation on growth, some immune responses and gut lactic acid bacteria of common carp (*C. Carpio*) fingerlings. Veterinary Research Forum. 2015; 6(3):239-244.
44. Mussatto SI, Mancilha IM. Non-digestible oligosaccharides: A review. Carbohydrate Polymers. 2007; 68(3):587-597.
45. Newman K. Mannan-oligosaccharides: Natural polymers with significant impact on the gastrointestinal microflora and the immune system. Biotechnology in the feed industry. 1994; 10:167-174.
46. Nicolas JL, Gatesoupe FJ, Froueli S, Bachere E, Gueguen Y. What alternatives to antibiotics are conceivable for aquaculture. Productions Animals. 2007; 20:253-258.
47. Ozluer-Hunt A, Berköz M, Ozkan F, Yalin S, Ercen Z, Erdogan E *et al*. Effect of mannan oligosaccharide on growth, body composition, and antioxidant enzyme activity of tilapia (*Oreochromis niloticus*). The Israeli Journal of Aquaculture-Bamidgeh IIC. 2011; 63(2):619-627.
48. Pasala S, Pemmineti NJ, Mohammad SA, Kothuru P. Effect of water probiotic in water quality maintenance and growth of rohu (*L. rohita*) fingerlings. European Journal of Pharmaceutical and Medical Research. 2018; 5(1):280-286.
49. Patterson JA, Burkholder KM. Application of prebiotics and probiotics in poultry production. Poultry Science. 2003; 82(4):627-631.
50. Phume P. Optimal protein-lipid level and replacement of fish meal with plant protein sources in feeds formulated for (*P. hypophthalmus*, Sauvage, 1878). PhD thesis, submitted to the School of Biological Sciences, Universiti Sains Malaysia, Penang, Malaysia. 2011; 1-166.
51. Pryor GS, Royes JB, Chapman FA, Miles RD. Mannan oligosaccharides in fish nutrition: Effects of dietary supplementation on growth and gastrointestinal villi structure in Gulf of Mexico sturgeon. North American Journal of Aquaculture. 2003; 65(2): 106–111.
52. Ramanadevi V, Muthazhagan K, Ajithkumar T, Thangaraj M. The efficacy of dietary yeast mannan oligosaccharide on growth and survival rate in *Amphiprion ocellaris* fingerlings. European Journal of Biotechnology and Bioscience. 2013; 1(2): 12-15.
53. Rehulka J, Minarik B, Cink D, Zalak J. Prebiotic effect of fructo–oligosaccharides on growth and physiological state of rainbow trout, *Oncorhynchus mykiss* (Walbaum). Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis. 2014; 59(5): 227-236.
54. Ringo E, Olsen RE, Gifstad TO, Dalmo RA, Amlund H, Hemre GI *et al.* Prebiotics in aquaculture. Aquaculture Nutrition. 2010; 16(2): 117-136.
55. Roy NC, Kohinoor AHM, Wahab MA, Thilsted SH. Evaluation of performance of Carp-SIS polyculture technology in the rural farmer's pond. Asian Fisheries Science. 2002; 15: 41-50.
56. Salaghi Z, Imanpuor M, Taghizadeh V. Effect of different levels of probiotic primalac on growth performance and survival rate of persian sturgeon (*Acipenser persicus*). Global Veterinaria. 2013; 11(2):238-242.
57. Salze G, McLean E, Schwarz MH, Craig SR. Dietary mannan oligosaccharide enhances salinity tolerance and gut development of larval cobia. Aquaculture. 2008; 274:148–152.
58. Sang HM, Fotedar R. Effects of mannan oligosaccharide dietary supplementation on performances of the tropical spiny lobsters juvenile (*Panulirus ornatus*, Fabricius 1798). Fish and Shellfish Immunology. 2010; 28(3): 483-489.
59. Sang HM, Ky LT, Fotedar R. Dietary supplementation of mannan oligosaccharide improves the immune responses and survival of marron, *Cherax tenuimanus* (Smith, 1912) when challenged with different stressors. Fish and Shellfish Immunology. 2009; 27(2): 341–348.
60. Santhosh B, Singh NP. Guidelines for water quality management for fish culture in Tripura. ICAR Research Complex for NEH Region. Tripura Center. 2007; Publication no 29.
61. Staykov Y, Spring EP, Denev ES, Sweetman EJ. Effect of a mannan oligosaccharide on the growth performance and immune status of rainbow trout (*Oncorhynchus mykiss*). Aquaculture International. 2007; 15(2):153-161.
62. Tacon AGJ. Standard methods for the nutrition and feeding of famed fish and shrimp. Redmond, Washington: Argent Laboratories Press. 1990, 454.
63. Torrecillas S, Makol A, Caballero MJ, Montero D, Gines R. Improved feed utilazition, intestinal mucus production and immune parameters in sea bass (*Dicentrarchus labrax*) feed manan- oligosaccarides. Aquaculture Nutrition. 2011; 17(2):223-233.
64. Torrecillas S, Makol A, Caballero MJ, Montero D, Robaina L, Real F *et al.* Immune stimulation and improved infection resistance in European sea bass (*Dicentrarchus labrax*) fed mannan oligosaccharides. Fish Shellfish Immunology. 2007; 23(5):969-981.
65. Welker TL, Lim C, Yildirim-Aksoy M, Klesius PH. Effect of short-term feeding duration of diets containing commercial whole-cell yeast or yeast subcomponents on immune function and disease resistance in channel catfish, *Ictalurus punctatus*. Journal of Animal Physiology and Animal Nutrition. 2011; 96(2):159-171.
66. Wu ZX, Yu YM, Chen X, Liu, H, Yuan JF, Shi Y *et al*. Effect of prebiotic konjac mannan oligosaccharide on growth performances, intestinal micro flora, and digestive enzyme activities in yellow catfish, *Pelteobagrus fulvidraco*. Fish Physiology and Biochemistry. 2014; 40(3):763-771.
67. Yilmaz E. Genc MA, Genc E. Effects of dietary mannan oligosaccharides on growth, body composition, and intestine and liver histology of rainbow trout, *Oncorhynchus mykiss*. Israeli Journal of Aquaculture-Bamidgeh. 2007; 59(3):182-158.
68. Yousefian M, Amiri MS. A review of the use of prebiotic in aquaculture for fish and shrimp. African Journal of Biotechnology. 2009; 8(25):7313-7318.
69. Zhang J, Liu Y, Tian L, Yang H, Liang G, Xu D. Effects of dietary mannan oligosaccharide on growth performance, gut morphology and stress tolerance of juvenile Pacific white shrimp, *L vannamei*. Fish and Shellfish Immunology. 2012; 33(4):1027-32.