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EFFICACY AND SUITABILITY OF WHOLE WHEAT FLOUR ON THE GROWTH AND SURVIVAL OF ROHU (*Labeo rohita*)

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The efficacy and suitability of dietary whole wheat flour supplementation was evaluated on the growth performance and survival of rohu (*Labeo rohita*). The study was conducted in 12 experimental aquaria at the Wet Laboratory, Department of Aquaculture, Bangladesh Agricultural University, Mymensingh. A total of 180 fingerlings of mean initial weight of 1.744±00 g were stocked at the same stocking density (268 fingerlings/m^3). Four different treatments (T_1, T_2, T_3 and T_4) each with three replications were performed having different level of whole wheat flour such as 0% whole wheat flour (T_1), 5% whole wheat flour (T_2), 10% whole wheat flour (T_3) and 15% whole wheat flour (T_4). Feeds were supplied at 5% body weight twice daily in the morning at 9.00 am and in the afternoon at 5.00 pm throughout the study period. The rations were adjusted every week when new weights of the fish for various experimental aquariums were determined. At the end of the 63 days study period, the growth performance parameters such as mean final weight (g), weight gain (g), percent weight gain (%), specific growth rate (%/day) and the best feed utilization in terms of food conversion ratio were found highest at treatment 4 where fishes were fed with inclusion of 15% whole wheat flour and lowest was found at treatment 1 (fed with inclusion of 0% whole wheat flour). Water quality parameters were within the acceptable range and survival rate was found 100% in all treatment. Therefore, it can be concluded that treatment 4 (highest growth performance and survival rate of *L. rohita* with supplementation of 15% whole wheat flour containing diet) can be chosen by fish farmers for commercial culture of this species.

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INTRODUCTION

Fish is considered as second valuable agricultural product and the aquaculture sector is playing a significant role in our national economy, nutrition, employment, income generation and foreign currency earning. Bangladesh has recorded surplus fish production with an annual output of 41.34 lakh metric tons against a demand of 40.50 lakh metric tons in 2016-17 where aquaculture contributes 56.44 percent to total production. It contributes 3.61 percent to our national GDP and around one-fourth (24.41 percent) to the agricultural GDP and the country earns BDT 42876.40 million by exporting almost 68.31 thousand MT of fish and fisheries products in 2016-2017 (DoF, 2017).Bangladesh is ranked third in producing fish from inland water-bodies (10.48 lakh metric tons) and also the fifth biggest aquaculture producer (22 lakh metric tons) and 11th marine producer (1.13 lakh metric tons) in the world (FAO, 2018). Per capita fish consumption in the country reaches 62.58 grams, which is higher than their daily protein demand as per the report of the Bangladesh Bureau of Statistics (BBS, 2017). Fish also contributed to about 17 percent of animal protein and 6.7 percent of all protein consumed by global population. Moreover, fish provided more than 3.1 billion people with almost 20 percent of their average per capita intake of animal protein (DoF, 2018). Different type of probiotics and prebiotics are used to improve feed efficiency, growth performance and immune status and among them, prebiotics improve host health by stimulating the growth and activity of certain gut resident bacterial species (Gibson and Roberfroid, 1995. To increase the digestion different prebiotic feed is very important to formulate feed which facilitate smooth functions of the intestinal digestions (Pandey *et al*., 2015). Though fiber is the source for prebiotics, foods that are high in fiber are also typically high in prebiotics, should be used in feed formulation.

Whole wheat flour is a very common prebiotic food source which contains 5% prebiotic fibre by weight and is abundant in our country with a cheap rate. Rice bran gives sugar contents where whole wheat flour provides heavy fibre that enables more digestion. Considering the above circumstances, the present study was designed to evaluate the efficacy and suitability of whole wheat flour into the diets of *L. rohita* and estimate the growth performance and survival rate of *L. rohita.*

MATERIALS AND METHODS

Experimental site

The experiment was carried out in the Wet Laboratory of the Department of Aquaculture, Bangladesh Agricultural University, Mymensingh, Bangladesh between September and November 2017.

Experimental design

The experiment was carried out using 12 glass aquaria of 0.61 m × 0.31 m × 0.40 m installed in the Wet Laboratory of Bangladesh Agricultural University. The aquariums were aerated throughout the experimental period using aerators in order to help replenish the amount of dissolved oxygen in the water and also to produce some current for the movement of food particles in the water. The aquariums were also covered with a net of 2 mm mesh size in order to prevent the fishes in the aquariums from skipping out and also to protect the aquariums from foreign materials or predators. Four treatments were used with three replicates, where each replicates in the various treatments had labels T1R1, T1R2, T1R3 for treatment one; T2R1, T2R2, T2R3 for treatment two, T3R1, T3R2, T3R3 for treatment three and T4R1, T4R2, T4R3 for treatment four. Uneaten feed and faeces were siphoned every morning prior to feeding using a pressure tube. The fish in each of the aquarium was weighed weekly using an electronic balance.

Experimental fish

Fingerlings of *L. rohita* of similar weight range and standard length was procured from Fish Hatchery Complex, Faculty of Fisheries, Bangladesh Agricultural University, Mymensingh. The fingerlings were transported to the Wet Laboratory by using a plastic bag containing 70% water and 30% oxygen and were immediately placed in a 60 litres aquarium containing half-filled water and allowed for 10-12 minutes for exchange in temperature. A few minutes later, part of the water in the aquarium was reduced and replaced with fresh water. Five hours later, the fish were then distributed equally in six aquariums labeled A, B, C, D, E

and F for acclimatization. During the acclimatization period, each aquarium was monitored for daily fish mortality and dead fishes were removed from the aquariums. The fingerlings were fed a control diet at every 9 am and 4 pm for seven days and were then starved for 24 hours prior to the start of the experiment before evenly stocked into the 12 aquariums at 15 fingerlings per aquarium at four trials with three replicates.

Experimental diets

Locally procured feed ingredients were used to formulate the diet for the fingerlings throughout the experiment. The feed ingredients for all four diets had a constant inclusion level of: fish meal (FM) 30%, soybean meal (SBM) 20%, mustard oil cake (MOC) 12%, molasses 5.5%, vitamin premix 1%, mineral premix 1%, and chromic oxide 0.5%.Four graded levels of whole wheat flour 0%, 5%, 10% and 15% were included in the basal diet at the expense of rice bran. The ingredients used for preparation of basal experimental diets along with their proximate composition are shown in Table 1.

Table 1. Proximate composition analysis of different feed ingredients (dry basis)

Feed ingredients	Crude protein (%)	Crude Lipid (%)	Moisture (%)	Ash (%)	Crude fibre (%)	NFE
Fish meal	58.74	10.60	8.51	16.60	2.80	2.75
Rice bran	13.61	11.40	12.01	13.60	6.80	42.58
Whole wheat flour	16.13	5.60	13.14	13.40	6.95	44.78
Soybean meal	32.34	4.60	13.48	8.40	6.88	34.3

Each ingredient of the feed was ground to dust using a grinding machine. All the various components were mixed in their various ratios in order to produce the various diets needed for the experiment. A total of four diets were formulated with varying proportions at 0% (T1), 5% (T2), 10% (T3) and 15% (T4) inclusion with the whole wheat flour.The feed ingredients (finely ground and sieved) were weighed accordingly, moistened with water to form dough and pelletized using an electric meat mincer. The feeds were stored in airtight polythene bags at room temperature. The experimental diet samples were analyzed for moisture (%), ash (%), crude protein (%), crude fibre (%), lipids (%) and NFE (%). These analyses were carried out at the Fish Nutrition Laboratory of the Bangladesh Agricultural University, Mymensingh using the Association of Official Analytical Chemists official methods of analysis 15th edition, Washington DC (AOAC, 2000). Proximate composition of the three formulated feed is shown in Table 2.

Table 2. Proximate composition analysis of formulated feed (dry basis)

Diet	Moisture (%)	Crude Lipid (%)	Crude protein (%)	Ash (%)	Crude fibre (%)	NFE
Control	9.15	8.90	37.39	15.69	4.40	24.47
Diet 2	9.33	8.56	37.93	15.33	4.65	24.20
Diet 3	7.31	7.90	39.35	14.31	4.80	26.33
Diet 4	8.97	8.35	38.26	13.96	5.30	25.16

Fish feeding and aquarium management

The fish were fed at 5% of their body weight in two rations, during the morning at 9 am and the afternoon at 5 pm throughout the experiment. The amount was fixed after observing that they were not interested to have more than this percent of feed. The rations were adjusted every week when new weights of the fish for various experimental aquariums were determined. Left over feed and faeces in each tank were siphoned every morning prior to feeding and replaced with fresh water.

Monitoring of water quality

Physico-chemical parameters such as temperature, dissolved oxygen and pH in the various aquariums were taken once per week during the early morning periods prior to siphoning and feeding throughout the experimental period.

Determination of growth and nutrient utilization

The weekly weights of fish and feed supplied was recorded and used to compute the growth nutrient utilization parameters. Weight of fish was taken weekly using a fine mesh scoop net to capture all the fish in each aquarium, swirled for 4-5 seconds to allow water from the fish to drip out before weighing to the digital balance (AND GULF, Dubai, model: GL-300). Fingerings were released into the aquarium after recording the weight. The growth and nutrient utilization parameters such as weight gain (g), percentage weight gain (%), specific growth rate (SGR, %/day), protein efficiency ratio (PER), feed conversion ratio (FCR) and survival rate (%) were calculated.

Data analysis

The collected data were statistically analyzed by one-way ANOVA with the help of SPSS to see whether the influence of different treatments on these parameters were significant or not. The means of different treatment were compared by DMRT (Duncan, 1955) to test the significance of variation between the treatment means at $p<0.05$.

RESULTS

Growth performances of *L. rohita*

Growth parameters of *L. rohita* after feeding different diet such as Diet 1:0% whole wheat flour, Diet 2: 5% whole wheat flour, Diet 3: 10% whole wheat flour and Diet 4: 15% whole wheat flour are as follows:

Initial weight (g)

The mean initial weight of individual *L. rohita* in different treatments was 1.74±00 g (Table 3).

Final weight (g)

The mean final weight of individual *L. rohita* in different treatments varied from 3.03±0.10 g to 3.95±0.07 g. The mean final weight (g) in T_4 was significantly higher followed by T_2, T_3, and T_1, respectively (Table 3).

Weight gain (g)

The mean weight gain of individual *L. rohita* in different treatments ranged from 1.29±0.10 g to 2.21±0.07 g. The mean weight gain of experimental fish was found highest in T_4 followed by T_2, T_3 and T_1, respectively (Table 3).

Percent weight gain (%)

The percent weight gain (%) of fish in different treatments ranged from 74.28±5.93% to 127.06±4.04%. The highest percent weight gain (%) was found in treatment T_4 followed by T_2, T_3 and T_1, respectively. The percent weight gain was significantly ($p<0.05$) varied between T_1 and T_4; T_2 and T_3 but no significant variation between T_4 and T_2 (Table 3).

Specific growth rate (%/day)

The specific growth rate (%/day) ranged from 0.88±0.05% to 1.30±0.03% /day.The highest specific growth rate (1.30±0.03%/day) was found in T_4 followed by T_2, T_3, T_1, respectively. The specific growth rate was significantly ($p<0.05$) varied between T_1 and T_4; T_2 and T_3 but no significant ($P\geq0.05$) variation between T_1 and T_3; T_2 and T_4 (Table 3).

Food conversion ratio (FCR)

Mean food conversion ratio in different treatments ranged from 2.42±0.13 to 3.43±0.29. The lowest FCR was obtained in T_2 followed by T_1, T_3, and T_4, respectively. There was no significant ($P \geq 0.05$) variation in mean food conversion ratio among T_2, T_3 and T_4 but had significant variation ($p<0.05$) between T_1 and other treatments (Table 3).

Food conversion efficiency (FCE)

Mean food conversion efficiency in different treatments ranged from 0.29±0.03 to 0.41±0.03.The highest FCE was obtained in treatment T_4 followed by T_2, T_3, and T_1, respectively. There was no significant ($P \geq 0.05$) variation in mean food conversion ratio among T_2, T_3 and T_4 but had significant variation ($p<0.05$) between T_1 and others (Table 3).

Table 3. The effect of different treatments on growth performance, feed utilization and survival rate of rohu (*L. rohita*) reared in aquarium (Mean ± SD) during the study period

Variable parameters	Treatment 1	Treatment 2	Treatment 3	Treatment 4	LSD	Level of sign.
Initial weight (g)	1.74(±0.00)	1.74(±0.00)	1.74(±0.00)	1.74±(0.00)	0.00	NS
Final weight (g)	3.03(±0.10)^c	3.89(±0.09)^a	3.73(±0.19)^b	3.95(±0.07)^a	0.13	**
Weight gain (g)	1.29(±0.10)^c	2.15(±0.08)^a	1.99(±0.19)^b	2.21(±0.07)^a	0.13	**
% weight gain	74.28(±5.93)^c	123.75(±4.96)^a	114.58(±10.79)^b	127.06(±4.04)^a	7.54	**
SGR (%/day)	0.88(±0.05)b	1.28(±0.04)^a	1.21(±0.08)^a	1.30(±0.03)^a	0.06	**
FCR	3.43(±0.29)^a	2.43(±0.04)^b	2.58(±0.23)^b	2.42(±0.13)^b	0.22	**
FCE	0.29(±0.03)^b	0.41(±0.01)^a	0.39(±0.04)^a	0.41(±0.02)^a	0.03	**
PER	0.69(±0.06)^b	0.95(±0.03)^a	0.87(±0.08)^a	0.94(±0.02)^a	0.06	*
Survival rate (%)	100.00(±0.00)	100.00(±0.00)	100.00(±0.00)	100.00(±0.00)	0.00	NS

Values given in bracket are standard deviation. The values in the same row having similar letter (s) do not differ significantly otherwise differ significantly ($p<0.05$) as per Duncan Multiple Range Test (Duncun, 1955).
NS=Not significant, * significant in 5%, ** significant in 1% significance level

Protein efficiency ratio (PER)

Mean protein efficiency ratio (PER) in different treatments varied from 0.69±0.06 to 0.95±0.03.The highest protein efficiency ratio (PER) was found in T_2 followed by T_4, T_3, and T_1, respectively. There was significant ($p<0.05$) difference between T_1 and T_4; T_2 and T_3 but no significant variation between T_2 and T_4; T_3 and T_4; T_3 and T_1 (Table 3).

Survival rate (%)

The mean survival rate (%) of *L. rohita* under different treatments was 100%.There was no significant ($P \geq 0.05$) variation in survival rate of *L. rohita* in four treatments (Table 3).

Water quality parameter

The water quality parameters such as temperature, dissolved oxygen and pH of different aquariums were measured throughout the experimental period. The range of temperature, dissolved oxygen and pH were from 27.6-29.63°C, 7.98-8.20 mg/l and 8.3-9.4 (Table 4).

Table 4. Summary of water quality parameters observed during the experimental period

Parameters	Value range
Temperature (°C)	27.6 – 29.63
pH	7.98 – 8.20
Dissolved Oxygen (DO) (mg/l)	8.3 – 9.4

DISCUSSION

Effect of whole wheat flour supplementation on *L. rohita* in terms growth performance, specific growth rate and food conversion ratio including water quality parameters were discussed.

Growth performance

Weight gain (g)

In this experiment effect of whole wheat flour on the growth and survival rate of rohu (*L. rohita)* in aquarium was investigated. Compared with other treatments, growth performance of rohu were significantly ($P<0.05$) higher (2.21g) in T_4 which was provided with higher levels of whole wheat flour (15%) feed whereas lower in T_1. Ahmed *et al*., (2012) assessed the effect of commercial feed on the growth performance rohu fingerlings. Feeding rice bran (50%) he recorded highest weight gain of 85g, this is due to initial weight (395g) and culture duration. In our study, the maximum mean final weight was 3.95±0.213g in T_4, whereas, the minimum mean final weight was 3.03±0.42g in T_1. Kumari *et al.*, (2017) showed that maximum mean final weight in (*L. rohita*) was 27.79 ± 2.28 g feeding 50g/kg wheat flour with *Azolla*. In this present study less weight was gained due to lower initial weight of rohu. The mean weight gain found highest in T_4 (2.21±1.26) which had significant variation among others treatments at 5% significant level. The weight gain seems low because of stocking initial weight of fry. But percentage of weight gain was quite satisfactory. So, it can be said that 15% wheat supplements is suitable for *L. rohita* culture.

Specific growth rate (SGR %/ day)

In present study, specific growth rate varied from 0.88 ± 0.05 to 1.30 ± 0.03. SGR value was highest in T_4 where the highest whole wheat flour level (15%) and lower in T_1 (control) where fish reared without whole wheat flour. Tian *et al.* (2012) found the highest value of SGR of grass carp feeding 33% wheat and recommended that not more 33% whole wheat flour is suitable for normal growth rate *C. idella*. So in case of *L. rohita* 15% whole wheat flour supplementation is efficient for growth.

Food conversion ratio (FCR) and Food conversion efficiency (FCE)

In present study, the lowest FCR value was recorded in T_4 and the highest FCR value was recorded in T_1 where 0% and 15% whole wheat flour were supplemented respectively.. Ahmed *et al*. (2012) reported that the food conversion ratio (4.12 ± 0.4) feeding with rice bran (50%).Tian *et al.* (2012) recoded highest food conversion efficiency and lowest feed conversion ratio of grass carp feeding 33% wheat. Bishnoi *et al*. (2017) recorded highest FCR value (1.76±0.03) of *L. rohita* feeding with aloevera supplementation. The food conversion efficiency T_1, T_2, T_3 and T_4 was 0.29±0.03, 0.41±0.01, 0.39 ±0.04 and 0.41±0.02, respectively. So highest food conversion efficiency and lowest feed conversion ratio was found in T_4 and recommended for *L. rohita*.

Protein efficiency ratio (PER)

The lowest protein efficiency ratio (0.69 ± 0.06) was observed in T_1 (control) and the highest (0.95 ± 0.03) in T_2. Tian *et al.* (2012) found the highest value of PER (0.80) of grass carp feeding 33% wheat which is more or less similar to the present study.

Survival rate (%)

The survival rate was found 100% in every treatment. There were no significant variations among the four treatments. The highest survival rate might be the cumulative result of good water quality parameters due to weekly water exchange, quality feed use, and proper maintenance during culture. This result of 100% survival in four treatments indicated that wheat supplementation has significant effect on survival rate of *L. rohita.* Whole wheat flour has high nutritional value as compared to rice bran because it contains more protein and fibres than rice. Rice has heavy carbohydrates whereas wheat has heavy fibres. Fibre gives more digestion and rice bran gives sugar content. Similarly, whole wheat flour that includes bran also is healthier and easier to digest. Whole wheat flour has carbohydrates which takes time to digest as compared to rice bran because rice bran contains starch that is quicker to digest. In the present study replacement of rice bran with whole wheat flour indicates significant effect on the growth performance of *L. rohita.* In T_4, 15% rice bran is replaced with 15% whole wheat flour showed the highest growth performance and survival rate of *L. rohita*. So it can be concluded that 15% whole wheat flour is suitable for the growth of *L. rohita.*

Water quality parameters

The water quality parameters play an important role for maintaining healthy environment for aquatic organisms. The water temperature monitored during the study period in the experimental tanks varied from 27.6 to29.63°C. Kausar and Salim (2006) investigated the effect of different water temperature ranges on growth performance and concluded that water temperature ranging from 24-26°C seemed to be the most effective for rearing of *L. rohita*. From the above statement, water temperature in aquarium was similar of pond temperature. The dissolved oxygen content from present experimental study ranged from 8.3 to 9.4mg/l. Hossain (2009) and Alam (2009) measured the dissolved oxygen in ponds of Agro-3 arm, Trishal, Mymensingh ranged from 5.5 to 6.5 mg/l. Hossain (2009) and Alam (2009) measured the pH value in ponds of Agro-3 Farm, Trishal, Mymensingh ranged from 7.54 to 8.3 and 7.72 to 8.03, respectively. In this experiment pH value was recorded 7.98 to 8.20 which was similar to other study.

CONCLUSION

The result of the present study proves significant role of wheat flour supplementation on the growth performance of *L. rohita* and the optimum inclusion level was 15%. Replacing 15% rice bran with 15% whole wheat flour was quite satisfactory because of its nutritional content and path of its digestion. So it can be concluded that 15% whole wheat flour has paramount importance in enhancing the production of *L. rohita* and recommended to incorporate with the feed.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

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