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Comparative growth performance of rohu (*Labeo rohita*) fed with low cost feeds using locally available animal proteins in Purulia district of West Bengal

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Abstract

The present study was done to formulate a low-cost fish feed using locally available animal protein wastes. To evaluate the efficacy of three different animal waste added feeds (containing fish meal, snail meat and silk worm pupae respectively) against conventional feed (containing rice bran, mustard oil cake and ground nut oil cake used as control) for fingerlings of IMC, Rohu, Labeo rohita, Cyprinidae. Twelve aquariums (50 L capacity) filled with ground water (P^H7-8) were used for the experiment for 3 months during July - September, 2019. The feeds were formulated on the basis of crude protein requirement of Rohu. Proximate composition of the above mentioned ingredients and formulated feeds were analyzed in respect to moisture, ash, crude fibre, fat and crude protein content. Fish survival was 100 % in all the treatments. Evaluation of the diets was carried out on the basis of weight gain, percent weight gain, food conversion ratio, condition factor, specific growth rate. It was observed that crude protein level among different animal wastes, fish meal recorded highest (52.84%) followed by silk warm pupae (52.79%) and lowest in snail meal (43.33%).The specific growth rate of rohu was significantly highest in T_4 followed by T_2 and T_3 and lowest in T_1.There was significant difference in weight gain, percent weight gain, FCR and K. No significant difference in length gain. and significant difference in specific growth rate was observed when fish fed with silk worm pupae or snail meal or fish meal. The study suggests that these low- cost feeds can be applied for increasing fish production in West Bengal, Purulia in particular.

Keywords: rohu, animal wastes, formulated feed, proximate composition, growth performance

INTRODUCTION

For the improvement of fisheries and to achieve maximum yields from aquaculture it is necessary to provide artificial feed. For commercial culture of fish, the formulation of low cost balanced diet using raw materials, locally available animal byproducts are needed. Recently fish meal has become the most extensive protein ingredient in aquaculture feeds. Several animal protein sources were evaluated to formulate the diets for fish such as poultry by-product meal, meat and bone meal, blood meal, snail, silk worm pupae, shrimp meal, earthworm meal, maggot meal and other invertebrate meal [1, 4, 5, 6, 7, 8, 9, 10, 11, 13, 14,15,16,17]. Main tendency is to total replacement of fish meal with alternative protein source [4, 5, 9, 14]. In Purulia, nearly 81.23% farmers practicing traditional extensive type farming, poly culture is prevalent, 75.32% of them do not use any kind of supplementary feed and perform multiple-stocking multiple harvesting [2].The major objective of this study was replacement of fish meal with locally available animal wastes without reducing the nutritional quality of feed.

MATERIALS AND METHODS

Experimental site

The study was carried out in Regional Research Sub-Station (RRSS), Bidhan Chandra Krishi Viswavidyalaya, Raghunathgpur, Purulia, West Bengal. The feeding trial was carried out in twelve 50 x 50 x 50 cm glass (L x W x H) aquariums each and replicated three times for 90 days. All analyses were taken place at the Laboratory of Animal Nutrition Department, West Bengal University of Animal and Fishery Science, Belgachia, Kolkata.

Aquarium preparation

The aquariums were filled up with tube well water to a depth of 30l. In each day 25 % of water was changed, while complete changing of water was done in every 5 day. The water level was retained up to studied period (90 days) with regular additions of tube well water to compensate evaporation losses.

Fingerlings and stocking

The spawn of rohu (*Labeo rohita*) were transported from Ramsagar carp hatchery in oxygenated bags and stocked in tanks of RRSS, BCKV, Raghunathpur, Purulia farm. Fingerlings of rohu with an average weight of 12-13 g size were collected from farm pond,

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acclimated to laboratory conditions for 14 days, during which they were fed with control feed. Fishes were randomly stocked in 20 fish aquarium^{-1}. All fishes were weighted at the time of stocking and data was recorded for final comparison purposes. Aquariums were provided with continuous aeration through All Extreme AE-8300 150 PSI 12V portable Air compressor Tyre Inflator.

Feed ingredients and preparation of feed

The percentage of ingredients in each diet is shown in Table 2. The ingredients were finely ground separately steam cooked at 140^{0}C and passed through a 5mm extruder die to prepare pellets at RRSS farm. The dry pellets were mashed and reduced in size (2 mm) to meet the fish's requirement. The mass was then bagged and stored at room temperature (25-37^{0}C) until feed. Three artificial supplementary feeds were formulated using fish meal (T_2), dried snail meat (T_3) and dried silk worm pupae(T_4) respectively and control diet (T_1) contained rice bran, mustard oil cake and ground nut oil cake. All the diets having 34-36 % protein level (Table-2) based on several studies regarding the recommended ranges for protein level of IMC 30-45 % [3]. The feeds were supplied twice daily at 6.00-7.00 and 18.00-19.00 at 4 % body weight. Dirt, uneaten feed, droppings and other particles were siphoned out from tanks daily.

Fish growth studies

At the end of each 2 week period, a random sample of 3 fishes were taken from each aquarium by a drag net and weighted and measured. Data were recorded and fish were put back into aquariums. At the end of the experiment growth parameters including net weight gain (NWG), percent weight gain (PWG), Feed conversion Ratio (FCR), condition factor (K), specific growth rate (SGR %) were calculated according to the following formulae:

Food utilization parameter

1. **Net weight gain (NWG)** = Mean final weight (g)- Mean initial weight (g)
2. **Percent weight gain (PWG)** = Final weight (g)- Initial weight (g)x100/ Initial weight (g)
3. ***Food conversion ratio (FCR)*** was calculated by**:**

$$FCR = \frac{\text{feed intake(g)}}{\text{Wet weight gain(g)}}$$

4. ***Condition factor (K)*** was calculated as:

$$K = \frac{W}{L_3} \times 100$$

W= Weight of fish

L= Standard length of fish

5. **Specific growth rate (SGR %)**= ln (Final wet body weight)- ln (initial wet body weight)x 100/ Number of days (Hopkins,1992)

Physio-chemical parameters

Water temperature, pH, dissolved oxygen (DO), free carbon dioxide, Nitrite were measured on a bi-weekly basis between 9.00 and 10.00 a.m. following the standard methods, APHA(1998).

Statistical analysis

Data were analyzed by one-way analysis of variance (ANOVA). Statistical significance was determined at 5% ($P<0.05$).

RESULTS

Table 1: Proximate composition (dry matter) of ingredients

Ingredients (g kg-^{1})	FM	SM	SWP	GNOC	MOC	RB
Moisture	2.87	3.37	2.75	7.31	7.96	7.13
Ash	19.72	26.54	5.91	7.81	7.42	15.21
Crude Fibre	7.28	0.19	8.51	3.82	8.14	35.94
Fat	8.24	3.18	22.28	7.0	8.34	6.78
Crude protein	52.84	43.33	52.79	45.59	35.43	10.14

FM= Fish meal, SM= Snail meat, SWP= Silk warm pupae, GNOC =Ground nut oil cake, MOC=mustard oil cake, RB=Rice Bran

Chemical compositions of experimental diets

The result of proximate analysis of experimental ingredients is presented in Table 1. Table 2 shows percentage of ingredients in diets and proximate composition of experimental diets having a higher concentration of crude protein in feed containing silk worm pupae.

Growth performance and nutrient utilization

At the expiration of the experiment, growth evaluation indices were used to assess response of *L. rohita* to fish meal, snail meal, silk worm pupae supplemented diets and the result is presented in table 3. There were no significant differences ($p>0.05$) in the mean initial stocking weight of *L. rohita* in all treatments.

Table 2: Percentage of ingredients in diets (%)

Ingredients	T_1 (Control)	T_2	T_3	T_4
GNOC	23.12	23.79	26.20	22.50
MOC	25.30	23.79	26.20	22.50
FM	-	23.79	-	-
SM	-	-	26.20	-
SWP	-	-	-	22.50
Binder	1.0	1.0	1.0	1.0
Vit/Min premix*	1.0	1.0	1.0	1.0
Veg oil	1 ml	1 ml	1 ml	1 ml
Salt	1	1	1	1
Total	100	100	100	100
Proximate composition (dry matter) of experimental diet				
Crude Protein	35.00	35.64	34.94	36.07
Moisture	26.59	25.50	22.44	23.61
Oil	14.76	14.28	16.06	16.23
Ash	23.65	24.58	26.56	24.09

Vit/Min premix*: Agrimin Forte powder composition /Kg: Vitamin A= 700000 I.U., Vitamin D3= 70000 I.U., Vitamin E= 250 mg,br>Cobalt: 150 mg, Copper: 1200 mg, Iodine: 325 mg, Iron: 1500 mg, Magnesium: 6000 mg, Potassium: 100 mg, Sodium: 5.9 mg, Manganese: 1500 mg, Sulphur: 0.72%, Zinc: 9600 mg, Calcium 25.5%, Phosphorus: 12.75%, DL-Methionine: 1000 mg;

Table 3: Growth response and feed efficiency of Rohu, *Labeo rohita* fingerling in fed experimental diets

Parameters	T1 (Control)	T_2	T_3	T_4
Mean initial weight (g)	13.17^{a}±0.70	12.87^{a}± 4.01	12.67^{a} ± 1.81	13.01^{a}± 3.46
Mean Final weight (g)	14.49^{a} ± 0.71	17.31^{b}±5.45	16.84^{b} ± 3.19	18.49^{b} ± 3.10
Mean weight gain (g)	1.32^{a}± 0. 73	4.43^{c} ± 5.15	4.18^{b} ± 3.49	5.43^{c} ±2.93
% weight gain	10.22^{a}±5.91	47.80^{b}± 77.72	35.95^{ab}± 34.36	41.56^{b}±22.58
Mean initial length (mm)	11.74^{a}± 0.86	11.74 ^{a}± 1.23	12.11^{ab} ± 1.0	12.69^{b} ± 0.91
Mean final length (mm)	12.70^{a}±0.49	13.14^{ab}±1.56	13.27^{b}±0.80	14.05^{c} ±0.71
Mean length gain (mm)	0.95^{a}±0.95	1.24^{a}±0.81	1.16^{a}±1.14	1.36^{a}±0.79
FCR[1]	1.90^{b}±3.44	1.23^{b}±1.06	0.67^{a}±0.80	1.34^{b}±1.91
K[2]	0.71^{b} ±0.07	0.76^{b} ±0.38	0.72^{b} ±0.11	0.67^{a} ±0.13
Specific Growth Rate (SGR % day-1)	0.31^{a} ±0.77	1.65^{b}±1.37	1.58^{b}±1.39	1.89^{b} ±0.18
Survival rate (%)	100± 0.00	100± 0.00	100± 0.00	100± 0.00

FCR[1] = Food conversion ratio, K[2] = condition factor
Means in a given column with the same superscript letter were not significantly different at p<0.05.

Table 4: Water quality parameters in the experimental aquarium fed with different experimental feed

Parameters	T1 (Control)	T_2	T_3	T_4
Water temperature (^{0}c)	27.1	27.1	27.1	27.1
pH	8.0 ±0.01	7.9 ± 0.12	7.7 ± 0.01	7.8 ± 0.02
Dissolved oxygen	8.01 ± 1.11	8.5 ±0.11	8.0 ± 0.12	8.2 ± 0.14
Free Carbon di oxide (mg l^{-1})	7.90 ±0.05	7.1 ±0.05	7.2 ± 0.01	7.4 ± 0.09
Nitrite	0.02 ± 0.03	0.04 ±0.01	0.05 ± 0.02	0.03 ± 0.04

Also, no mortality was recorded during the experimental period and fishes accepted diets administered as they were observed to be actively fed. Results showed that mean weight gain (5.43±2.93), length gain (1.36±0.79) and SGR (1.89±0.18) of fish fed with diet T_4 was found to be significantly higher and lowest (1.32±0.73), (0.95±0.95) and (0.31±0.77) in T_1 respectively. Percent weight gain (47.80±77.72) was significantly higher in T_2 and lower (10.22±5.91) in T_1. FCR (1.90±3.44) and K (0.71±0.07) were obtained significantly higher in T_1 and lowest (0.67±0.80) in T_3 and (0.67±0.13) in T_4 respectively. The rate of fish survival ranged 100% ($p>0.05$) in all the treatment groups.

Water quality parameters

The values of water quality parameters measured are shown in Table 4. Temperature 27.1^{0}C, pH ranged from 7.7-8, dissolved oxygen from 8.0 to 9.01 mg l^{-1}, free CO_2 ranged from 7.1-8.9 while nitrite concentration was between 0.02-0.05.

DISCUSSION

This investigation indicated that progressive weight gain was reported in all treatments throughout the duration of experiment. That was the indication that the rohu responded positively to all the diets. Our initial hypothesis was that feed with replaced fishmeal will have significantly lower fish growth parameters than control feed. But in experiment, fish growth parameters in all groups with replaced fishmeal showed better result than in control group (T_1).

Growth rate was higher in group fed the diets with SWP and lower in group fed the diets with Snail meal or no animal protein (T_3 and T_1 respectively). The results proved that total replacement of fishmeal with SWP produced higher growth performance to those fed with single fishmeal.

In recent years, focus has been shifted from the use of fish meal to other animal proteins as protein source in fish diets. Several animal byproducts have been used in the field of aquaculture such as seafood processing byproducts (Malaweera *et al*., 2013), garden snail, *Limicolaria spp* (Ovie *et al*., 2010) in diet of *Clarias gariepinus*, silk worm pupae (*Bombyx Mori*) in coldwater aquaculture (Asimi *et al*., 2017; Raja *et al*., 2019), silkworm pupae in carp cultre (Hertrampf *et al*., 2000), silkworm pupae in *C. gariepinus* fingerlings (Kurbanov *et al*., 2015).

Previous work on the snail meat and silk worm pupae had been reported to contain 66.76% crude protein in raw garden snail meat (Ovie *et al*., 2010: Sogbesan *et al*., 2006), 55.1 % crude protein in raw silk worm pupae (Hertrampf *et al*., 2000), 50-70% crude protein silk worm pupae (Sampathkumar *et al*., 2019). In this study when raw silk worm pupae were examined, crude protein of 52% in silk worm pupae was within recommended range used by other researchers. There had not been any previous report on snail, *Bellamya bengalensis* (crude protein of 43.33%). It was also observed in this study that *L. rohita* fed silk worm pupae supplemented diets had better weight gain than the other treatments. There was no significant difference in specific growth rate when fed with silk worm pupae or fish meal or snail meat. Kurbanov *et al*. (2015) reported higher growth rate and feed utilization parameters when fingerlings of *Clarias gariepinus* fed the diets with mixed fishmeal and silkworm pupae protein (SPP) (the highest was 50:50) and lower in those fed 100 % of SPP or fishmeal. This finding is contrary to that of Ovie *et al*. (2010) who reported no significant difference in mean weight gain, FCR, SGR, gross FCR and K when fish fed the varying levels of garden snail. The fish fed 75% garden snail had a lower growth as a result of mortality. Similarly, Rathore and Yusufzai (2018) reported higher weight gain, protein efficiency ratio and feed efficiency when use shrimp head meal (50%) in the diet of nile tilapia fry. Furthermore, this result also agrees with that of Sogbesan *et al*. (2006) who used 25% garden snail meat meal in the diet of *C. gariepinus* fingerlings.

Similar observation has been reported by Longvah *et al*. (2015) on defatted eri silk worm (*Simia ricinii*) meal (75%) with 44% total essential amino acids making it an ideal candidate for preparing protein concentrate isolates with enhanced protein quality that can be used in animal nutrition. Engmann *et al*. (2015) revealed that snail (*Achinata achinata*) meat is rich in protein 82.96% and low in both total charbohydrate (3.26%) and fat (3.98%). Similar observation by Nandeesha *et al*. (2000) concluded silk worm meal replaced part, or all of the fish meal in common carp (*Cyprinus carpio*) diets, similar performed same (growth and feed conversion). Similar observation by Rangachayulu *et al*., 2003 and Hwoan *et al*., 2010 recorded a combination of soybean meal and silkworm pupae meal replaced fish meal resulting in slightly higher survival rate and better growth performance in Juvenile abaloners (*Haliotis discus hannai Ino*). Asimi *et al*. (2017) revealed keeping in view the nutritional value of silkworm pupae and its profuse availability in the state of Kasmir, cost effective feed could be formulated and feeding trials in cold water aquaculture could be conducted.

The investigation has shown that SWP was founded to be available alternative to fish meal replacement in rohu fingerling feeds. SWP is very promising for Purulia because that district has developed silk breeding and majority of women involves in silk thread extraction. Silkworm pupa for itself is sub-product which can be

utilized. Our investigations showed that this protein can be used in aquafeed, and such utilization will increases rationality of silkworm breeding and provide aquafeeds production with high quality source of animal protein.

CONCLUSION

This study has shown that incorporating silkworm pupae or snail meat in the diet of *L. rohita* led to higher growth and better nutrient utilization than the control. From the result of our investigation, we can conclude that silkworm pupa protein and snail meat can be alternative source of replacement for fish meal in rohu, *Labeo rohita* feed and up to 25-30 % replacement of fish meal with silkworm pupae or snail meat protein advised.

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