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Abstract

Keeping the importance and search for unconventional feed resources and/or standardizing their level of incorporation in mind, we incorporated dry-powdered water hyacinth (*Eichhornia crassipes*) meal in feeds and studied its effect on growth and digestibility in *Labeo rohita* fingerlings. Five feeds with 30 % crude protein level were formulated using *Eichhornia* meal (EM) at 0 (control), 5 (EMF1), 10 (EMF2), 15 (EMF3) or 20 % (EMF4) of the diet replacing rice bran by equal proportions. Three hundred fingerlings (7.40 ± 0.05 cm; 5.27 ± 0.12 g) were distributed into fifteen tanks (200 l capacity) and fed the experimental diets for 60 days. In the last 30 days, digestibility studies were conducted using 0.5 % chromic oxide as an external marker in feed. At 10 % inclusion of EM, the experimental fish showed the highest weight gain percent (WG%), specific growth rate (SGR), protein efficiency ratio and apparent net protein utilization with lowest feed conversion ratio. Whereas the growth performance at 15 % inclusion level was comparable with the control and further increase to 20 % level of EM showed reduced growth responses but the feed was fairly palatable to the fish. Lower digestibility was also observed in EMF4 group. It is concluded that EM can be included at 15 % level in the feed of *L. rohita* fingerlings without adversely affecting the growth, dry matter and nutrient digestibility. However, economic feasibility of this feedstuff needs to be analyzed to see whether the reduced cost of diets would compensate for the reduced performance of fish at higher inclusion levels.

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RESEARCH ARTICLE

Effect of Dietary Incorporation of Dry-Powdered Water Hyacinth (*Eichhornia crassipes*) Meal on Growth and Digestibility of *Labeo rohita* Fingerlings

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Abstract Keeping the importance and search for unconventional feed resources and/or standardizing their level of incorporation in mind, we incorporated dry-powdered water hyacinth (*Eichhornia crassipes*) meal in feeds and studied its effect on growth and digestibility in *Labeo rohita* fingerlings. Five feeds with 30 % crude protein level were formulated using *Eichhornia* meal (EM) at 0 (control), 5 (EMF1), 10 (EMF2), 15 (EMF3) or 20 % (EMF4) of the diet replacing rice bran by equal proportions. Three hundred fingerlings (7.40 ± 0.05 cm; 5.27 ± 0.12 g) were distributed into fifteen tanks (200 l capacity) and fed the experimental diets for 60 days. In the last 30 days, digestibility studies were conducted using 0.5 % chromic oxide as an external marker in feed. At 10 % inclusion of EM, the experimental fish showed the highest weight gain percent (WG%), specific growth rate (SGR), protein efficiency ratio and apparent net protein utilization with lowest feed conversion ratio. Whereas the growth performance at 15 % inclusion level was comparable with the control and further increase to 20 % level of EM showed reduced growth responses but the feed was fairly palatable to the fish. Lower digestibility was also observed in EMF4 group. It is concluded that EM can be included at 15 % level in the feed of *L. rohita* fingerlings without adversely affecting the growth, dry matter and nutrient digestibility. However, economic feasibility of this feedstuff needs to be analyzed to see whether the reduced cost of diets would compensate for the reduced performance of fish at higher inclusion levels.

Keywords *Labeo rohita Eichhornia crassipes* · Digestibility · Growth

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Introduction

Fish nutritionists and feed manufacturers are constantly searching for newer ingredients or strategies to formulate cost-effective and environment-friendly aquafeeds to meet the ever-increasing demand for quality feed as well as fish. In traditional carp culture, a mixture of rice bran and groundnut oil cake (1:1) is generally used (Mukhopadhyay and Ray 1997). However, research pertaining to nutrition in freshwater aquaculture in the past two decades has led to the development of new feed formulations for Indian carp (Mohanty et al. 1995; Ayyappan and Jena 1998; Paul et al. 1998; Mukhopadhyay and Ray 1999, 2001; Khan et al. 2004). Aquafeeds based solely or partially on plant feedstuff have been reported to be effective and less expensive (Dorsa et al. 1982; Robinson et al. 1984; Ofojekwu and Ejike 1984), and also known to have excellent amino acid profile (Jackson et al. 1982) and supported growth of carps as good as the traditional feed (Patnaik and Das 1979). In this context, use of certain aquatic weeds offers excellent scope as these nutrient-laden materials are naturally grown in large waterbodies (e.g., wetlands) without much

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agronomic care (Kalita et al. 2007). Aquatic and terrestrial macrophytes have been used as supplementary feeds in fish farming since the early times of freshwater fish culture (Bardach et al. 1972) and still play an important role as fish feed in extensive culture systems (Edwards 1987), as they contain substantial amounts of protein and minerals (Ray and Das 1994). Aquatic macrophytes, which often infest a waterbody and make it unsuitable for fish culture, may be converted into fish flesh through their incorporation as a feedstuff in carp diets. However, the presence of anti-nutritional factors (ANFs) within plant feedstuffs restricts their use in animal feeds (Tacon et al. 1995). Processing plant materials through a simple and cheap method like drying or fermentation might considerably decrease the ANFs and crude fibre content thereby increasing their nutritional values.

Water hyacinth (WH; *Eichhornia crassipes*) is a wild freshwater fern belonging to the Family Pontederiaceae. It forms dense mats on the water surface that block navigation and interfere with irrigation, fishing, recreation and power generation. These mats also prevent sunlight penetration and aeration of the water, leading to oxygen deficiency, competitively exclude submersed plants and reduce biological diversity. These are free-floating aquatic plants which are not accepted by cattle and Indian major carps as feed in fresh condition. There have been some studies on tilapia indicating that only low levels of WH can be incorporated into fish feeds (Edwards et al. 1985; Hutabarat et al. 1986; Klinavee et al. 1990; Soliman 2000). The relatively high fiber content of WH may limit its use in tilapia feeds (Stickney and Shumway 1974; Buddington 1980). The use of water hyacinth as a feed ingredient for other fish has been investigated. Liang and Lovell (1971) found that the addition of 5–10 % WH meal to channel catfish diets significantly improved fish growth and survival. A diet containing 20 % WH was still fairly palatable.

Growth responses of different fish species fed test diets containing different levels of WH meal have been highly variable. For example, significant reduction in growth responses were reported by Hasan et al. (1990) for *Labeo rohita* fry and Hasan and Roy (1994) for *L. rohita* fingerling when 27–30 % WH leaf meal was incorporated to replace the fishmeal protein of the control diet. Similarly, Klinavee et al. (1990) recorded significant reduction in growth responses of *Oreochromis niloticus* when fed a test diet containing 40 % WH meal. However, 50 % dietary inclusion for *Ctenopharyngodon idella* and *Cyprinus carpio* (Murthy and Devaraj 1990), 100 % inclusion for *O. mossambicus* (Dey and Sarmah 1982) and 18.5 % inclusion for *Brycon* sp. (Saint-Paul et al. 1981) recorded either similar or higher growth responses compared to control diets. Dehydrated WH has been added to the diet of channel catfish fingerlings to increase their growth (Gopal 1987). However, in some of these studies, the control diet consisted only of a rice bran-oil cake mixture, which might have caused growth retardation. Edwards et al. (1985) observed only 10–15 % reduction in SGR of *O. niloticus* when fed test diets displacing 75–100 % of a 32.5 % crude protein commercial tilapia pellet by WH meal. However, they also pointed out that the fish obtaining indirect nutrition from plankton cannot be ruled out.

Labeo rohita, non-predatory Indian major carps, are predominantly accepted in the Eastern and North Eastern parts of India both in terms of consumer preference and amenability to culture in different ecosystems. The species is primarily a herbivorous to omnivorous one and prefers to feed on plant materials (Talwar and Jhingran 1991). In this backdrop, the present study aimed to determine the effect of dietary supplementation of dry-powdered *Eichhornia crassipes* (water hyacinth) meal on growth and digestibility in *Labeo rohita* fingerlings.

Materials and Methods

Collection and Preparation of Eichhornia Meal

Eichhornia crassipes plants were manually collected in the summer from the mass of such plants existing at Charan beel, Morigaon district, Assam, India. All the plants were AQ2 washed in water to remove any extraneous matter. After removing the roots, petiole-leaf part was sun-dried for 48 h. Then these were packed in plastic bags and brought to the laboratory of ICAR-Central Inland Fisheries Research Institute, Regional Centre, Guwahati, and dried in an oven at 60 °C for 48 h. The dried plants were then ground in a grinder, sieved with a fine mesh (0.2 mm) and the powdered meal (*Eichhornia* meal, EM) was stored in plastic bags for their analysis and incorporation in the diets. The yield of EM from raw material (i.e., petiole-leaf part of water hyacinth plant) was approximately 10 %, since the moisture content of the stuff was 90 %.

Experimental Diets

The locally available feed ingredients such as fish meal (FM), mustard oil cake (MOC), corn flour (CF), rice bran (RB), wheat flour (WF) and vitamin-mineral mixture (Minerex Forte) were used for feed formulation (Table I). *Eichhornia* meal (EM) was included at 0, 5 (EMF1), 10 (EMF2), 15 (EMF3) or 20 % (EMF4) replacing the rice bran proportionately. Weighed quantities of different ingredients were mixed (except vitamin-mineral mix) thoroughly, made into dough with appropriate amount of water, cooked in steam for 30 min and then cooled. After cooling, the dough was disintegrated and vitamin-mineral

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mix was thoroughly mixed. Pellets were prepared by a hand pelletizer through a 1 mm diameter die. Then the pellets were air dried for few hours and kept in oven for 6 h at 60 °C. After drying, the pellets were packed in airtight polythene bags, labeled and stored at room temperature (27–30 °C) until use.

Experimental Design and Feeding

Fingerlings of *L. rohita* (av. length: 7.40 ± 0.05 cm, av. weight: 5.27 ± 0.12 g) were procured from local fish seed vendors and transported in oxygen packaged condition to the wet laboratory of ICAR-Central Inland Fisheries Research Institute (CIFRI), Regional Centre, Guwahati. The stock was acclimated under aerated conditions for a period of 15 days while they were fed with a practical diet containing 30 % crude protein. Rectangular FRP tanks (covered with perforated lids) of identical size (200 l capacity) were used as experimental units for the trial. Each of the fifteen experimental tanks was stocked with twenty fingerlings following a completely randomized design (CRD) consisting of five treatments (feeds) with three replicates each. Round the clock aeration was provided to all the tanks. Chlorine-free bore well water was used as the source of water. The total volume of water in each tank was maintained at 150 l throughout the experimental period. The water quality parameters viz, temperature, pH, dissolved oxygen (DO), free carbon dioxide (CO_2), carbonate hardness, ammonia-N and nitrate–N were recorded every week following standard method (APHA AQ3 et al. 1998) to check the quality of culture water. The fingerlings were fed to visual satiation twice daily at 0700 and 1600 h. Daily feed intake was monitored and the feeding trial lasted for 60 days.

Growth and feed efficiencies

The body weight was measured at intervals of 15 days to assess the growth of fish. Before taking the body weight, the fish were starved overnight. Growth and feed efficiency parameters were calculated based on the following formulae:

$$\text{Weight gain (WG)}\,\% = 100[(\text{final weight} - \text{initial weight})/\text{initial weight}]$$

$$\text{Specific growth rate (SGR)}\,\% = 100[\{\ln(\text{final weight}) - \ln(\text{initial weight})\}/\text{experimental period}]$$

$$\text{Feed conversion ratio (FCR)} = \text{dry feed intake (g)}/\text{gain in wet weight (g)}$$

$$\text{Protein efficiency ratio (PER)} = \text{gain in wet weight (g)}/\text{protein fed (g)}$$

$$\text{Apparent net protein utilization (ANPU)} = \text{increase in whole body protein (g)}/\text{protein fed (g)} \times 100$$

$$\text{Energy retention value (ERV)} = (\text{final carcass energy} - \text{initial carcass energy})/\text{energy fed (kcal)} \times 100$$

Proximate Analysis of Tissues and Diets

Proximate composition of the whole fish was analyzed at the beginning and end of the feeding trial following the standard methods of AOAC (2005). Similarly, proximate AQ4 analysis of all the diets was determined. Briefly, moisture was determined by drying the samples at 105 °C to a constant weight. Nitrogen content of the samples was measured by Kjeltec (2200 Kjeltec auto distillation, Foss Tecator, Sweden) and crude protein (CP) was calculated by multiplying nitrogen percentage by 6.25. Ether extract (EE) was measured by Soxtec (1045 Soxtec Extraction Unit, Tecator, Sweden) using diethyl ether (boiling point, 40–60 °C) as a solvent and ash content was measured by incinerating the samples in a muffle furnace at 600 °C for 6 h. Total carbohydrate was calculated by difference, i.e. total carbohydrate% = 100 − (CP% + EE% + Ash%). The digestible energy (DE) value of experimental diets and tissue was calculated as described by Halver (1976).

Determination of Diet Digestibility

For the digestibility studies, diets were formulated and prepared exactly the same way as earlier, but added 0.5 % chromic oxide (Cr_2O_3) and fed for last 30 days of the experiment. After acclimation and gut evacuation for 1 week with new feeds, faecal matter generated was collected daily. Uneaten feed and the faecal matter were siphoned out after one hour 1 feeding and then left undisturbed for one more hour with minimum aeration. Faeces were then collected by siphoning out the intact faecal pellets using a small diameter plastic pipe through a fine meshed sieve. Faeces collected were dried in an oven at 105 °C to constant weight. All the faecal matter collected from a particular tank was pooled, finely ground and stored in freezer at 4 °C till further analysis.

Determination of Chromium

Wet ashing of the feed and faecal matter samples was carried out according to AOAC (2005) method. The chromium (Cr) content of the feed and faecal matter was then estimated by

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using flame ionization atomic absorption spectrophotometer (GBC 3000, Avanta Sigma, GBC Scientific Equipment Pvt. Limited, Australia) using chromium cathode lamp.

Nutrient Measurement in Faeces

The faeces collected were analyzed for crude protein, ether extract, ash and total carbohydrate using AOAC method (2005).

Apparent Digestibility Coefficient (ADC)

The ADC of dry matter and nutrient expressed as a percentage is calculated using the formulae:

$$\mathrm{ADC\,(dry\ matter)} = 100 - 100\,(\%\ \mathrm{marker\ in\ feed}/\%\ \mathrm{marker\ in\ faeces})$$

$$\mathrm{ADC\,(nutrient)} = 100 - 100\{(\%\ \mathrm{marker\ in\ feed}/\%\ \mathrm{marker\ in\ faeces})\} \times (\%\ \mathrm{nutrient\ in\ faeces}/\%\ \mathrm{nutrient\ in\ feed})$$

Statistical Analysis

Data were analyzed by one-way analysis of variance (ANOVA) and the significant difference between the treatments was determined by Duncan's Multiple Range Test (DMRT) using SPSS (Version 14.0). Results are reported as mean ± S.E. Each tank was considered as an experimental unit for calculating growth, SGR, FCR and FER, but for all other parameters duplicate measurements from each tank were done totaling n = 6 per treatment. The level of significance employed was 0.05.

Results

The proximate composition of the feed ingredients (%dry matter basis) used for formulation of the experimental diets is given in Table 1. *Eichhornia* meal (EM) contained crude protein of 13.62 %, ether extract of 7.94 % and a higher ash content of 15.79 %. The feed formulation and proximate composition of the EM-based diets fed to the *L. rohita* fingerlings are presented in Table 2. The CP contents in the experimental diets were estimated to be near the formulated value (30 %). Ether extract (EE) did not vary significantly among different feeds, which ranged from 6.05 to 8.33 %. The ash contents displayed marked differences ($p < 0.05$) with varying levels of EM. Lowest ash content (8.71 ± 0.09) was found in control diet and increased significantly ($p < 0.05$) with increased level of supplementation reaching the highest (11.04 ± 0.07) at 20 % EM supplementation. Water temperature varied from 26.5 to 27 °C. The pH and DO ranged from 7.4 to 7.63 and 6.5–7.27 ppm, respectively. Ammonia and nitrate level varied from 0.1 to 0.13 ppm and 0.04–0.05 ppm, respectively. The carbonate hardness ranged from 249 to 256 ppm and CO_2 was not detected in any of the tanks.

Growth and feed efficiency data recorded are presented in the Table 3. The highest weight gain percent was observed in the fish fed with 10 % EM-based diet, but further increase to 15 % resulted in lower growth that was similar to the control. At 20 % inclusion, the growth of the fish was significantly lower than the control. A similar response was recorded for the specific growth rate of fish. The feed conversion ratio (FCR) in the 10 % EM-supplemented group was recorded to be the lowest (1.78) and the highest FCR (2.45) was recorded in 20 % EM-supplemented group. Protein efficiency ratio (PER) of the 5 and 15 % EM-supplemented groups was comparable with the control group, but at 20 % EM there was a reduction in PER. At 10 % level of inclusion, *L. rohita* fingerlings achieved highest PER and apparent net protein utilization (ANPU). The ERV did not vary significantly ($p > 0.05$) between the treatments.

Tissue biochemical composition of the initial fish and after rearing them for 60 days is presented in Table 4. The moisture and crude protein contents were observed to be the highest in the fish fed the 15 % EM-supplemented diets. The total lipids, ash, organic matter and digestible energy contents did not vary significantly between the groups.

Table 1 Proximate composition of feed ingredients (% dry matter basis) used in feed formulation for feeding *L. rohita* fingerlings in the experiment

Components	Mustard oil cake	Corn flour	Wheat flour	Rice bran	*Eichhornia* meal (EM)	Fish meal
Crude protein (CP)	41.0	8.3	12.0	8.1	13.62	56.77
Ether extract (EE)	10.1	4.0	1.7	12.0	7.94	6.7
Total carbohydrate (TC)	42.8	86.5	85.7	69.9	62.65	14.73
Total ash	6.1	1.2	0.6	10.0	15.79	21.8
Digestible energy[a]	426.1	415.2	406.1	420	376.54	346.3

[a] Digestible energy (kcal/100 g) = (CP% × 4) + (EE% × 9) + (TC% × 4)

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Table 2 Formulation and proximate composition (% dry matter) of the *Eichhornia* meal (EM)-based diets fed to *Labeo rohita* fingerlings for 60 days

Components	Experimental groups (% EM)					*p* value
	Control (0)	EMF1 (5)	EMF2 (10)	EMF3 (15)	EMF4 (20)	
Fish meal	15	15	15	15	15	–
Mustard oil cake	30	30	30	30	30	–
Corn flour	10	10	10	10	10	–
Rice bran	25	20	15	10	05	–
Wheat flour	19	19	19	19	19	–
Vitamin-mineral mix[1]	1	1	1	1	1	–
Eichhornia meal	0	05	10	15	20	–
Chromic oxide	0.5	0.5	0.5	0.5	0.5	–
Proximate composition (mean ± SE)						
Moisture	3.67 ± 0.50	4.50 ± 1.35	4.47 ± 0.20	2.98 ± 0.13	5.80 ± 0.67	0.053
Crude protein (CP)	29.84 ± 0.09	29.68 ± 0.65	29.69 ± 1.03	29.52 ± 0.50	30.64 ± 1.31	0.887
Ether extract (EE)	7.71 ± 0.85	8.33 ± 0.02	7.17 ± 0.66	6.87 ± 1.04	6.05 ± 0.56	0.340
Total carbohydrate (TC)	53.74 ± 0.85	52.56 ± 0.71	53.49 ± 1.85	53.55 ± 0.60	52.27 ± 1.94	0.899
Total ash	8.71 ± 0.09[a]	9.44 ± 0.08[b]	9.73 ± 0.07[c]	10.06 ± 0.06[d]	11.04 ± 0.07[e]	0.001
Digestible energy[2]	403.71 ± 4.63	403.88 ± 0.43	396.59 ± 3.03	394.14 ± 5.01	386.11 ± 2.55	0.068
Chromium (%)	0.210 ± 0.013	0.213 ± 0.018	0.211 ± 0.02	0.211 ± 0.015	0.209 ± 0.021	0.234

Different superscripts in the same row signify statistical differences ($p < 0.05$) (mean ± SE; n = 6)

[1] Vitamin-mineral mix (Minerex Forte) (quantity/1 kg): Vitamin A-20,00,000 IU; Vitamin D_3-4,00,000 IU; Vitamin E-300 IU; Vitamin B_{12}-2.4 mg; Vitamin B_2-0.8 g; Vitamin K_3-0.4 g; Calcium D panthothenate-1 g; Choline chloride-60 gm; Ca-300 g; Mn-11 g; Fe-3 g; Cu-0.8 g; Co-180 mg; Se-40 ppm; Niacinamide-4 gm; Zn-2128 mg; Tri sodium citrate as chelating agent; Approximate overages and antioxidants added

[2] Digestible energy (kcal/100 g) = (CP% × 4) + (EE% × 9) + (TC% × 4)

Data pertaining to the dry matter and nutrient digestibility of *Eichhornia* meal based diets fed to *L. rohita* fingerlings is presented in Table 5. The dry matter digestibility showed a decreasing trend with increase in EM supplementation level with the control recording the highest value. The protein digestibility was found to be highest in EMF2 group, which was similar to control and EMF1 groups. The lowest protein digestibility (77.99 ± .63) was observed in EMF4. The lipid digestibility did not vary among the supplemental levels. The carbohydrate digestibility of EM-supplemented group did not vary significantly ($p > 0.05$) up to 5 % supplemental level, but it decreased significantly ($p < 0.05$) with further increase in EM supplementation. Significantly lower energy digestibility was recorded in EMF4 compared to the control and EMF1 groups.

Discussion

The nutritional profile of mustard oil cake, corn flour, wheat flour, rice bran and fish meal used in the formulation of diets, in spite of differences, corresponded to values reported earlier (Tacon and Jackson 1985). The proximate analysis of *E. crassipes* plant done was that of the petiole-leaf part. The estimated crude protein (CP) and ash content of the *Eichhornia* meal was 13.62 and 15.79 %, respectively. Gohl (1981) reported a crude protein of 12.8–13.1 % of dry matter for fresh green part of water hyacinths from India. Reports from many studies showed that the ash content of whole plants varied between 17–34 % (Edwards et al. 1985; Klinavee et al. 1990; Tuan et al. 1994) while it was between 10.2 and 18.8 % for leaves (Hasan 1990; Somsueb 1995). The high content of ash in water hyacinth may be attributed to their capacity to absorb minerals from eutrophicated water in which the plants grow.

The five experimental diets fed to the fingerlings for a period of 60 days were well accepted by the fish. Inclusion of water hyacinth did not affect the crude protein, ether extract and total carbohydrate level of the feeds whereas it increased the ash content of the diets significantly. This is due to higher ash content of *Eichhornia* meal compared to rice bran.

The final weight, weight gain and specific growth rate were higher in the group fed 10 % *Eichhornia* meal, which made us to infer that *Eichhornia* meal had positive effect on growth of the experimental fish up to a dietary level of

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Table 3 Growth and feed efficiencies in *Labeo rohita* fingerlings fed diets containing different levels of *Eichhornia* meal (EM) for 60 days

Experimental groups (% EM)	Final length (cm)	Final weight (g)	WG%[1]	SGR[2]	FCR[3]	PER[4]	ANPU[5]	ERV[6]
Control (0)	10.85 ± 0.85	12.94 ± 0.15^{a}	151.73 ± 9.85^{b}	1.54 ± 0.07^{b}	2.02 ± 0.08^{b}	1.67 ± 0.07^{b}	109.84 ± 1.22^{b}	49.98 ± 3.45
EMF1 (5)	11.45 ± 0.05	13.61 ± 0.12^{a,b}	151.58^{b} ± 2.82	1.57 ± 0.02^{b,c}	1.96 ± 0.02^{a,b}	1.72 ± 0.02^{b}	106.68 ± 4.52^{b}	51.91 ± 2.31
EMF2 (10)	12.45 ± 0.55	14.96 ± 0.70^{b}	178.12 ± 7.18^{c}	1.71 ± 0.05^{c}	1.78 ± 0.05^{a}	1.90 ± 0.05^{c}	123.25 ± 1.41^{c}	56.25 ± 0.64
EMF3 (15)	10.85 ± 0.65	12.92 ± 0.74^{a}	148.80 ± 7.21^{b}	1.52 ± 0.05^{b}	2.07 ± 0.06^{b}	1.64 ± 0.05^{b}	110.73 ± 2.82^{b,c}	45.06 ± 0.97
EMF4 (20)	10.15 ± 0.15	12.05 ± 0.26^{a}	124.42 ± 2.37^{a}	1.35 ± 0.02^{a}	2.45 ± 0.03^{c}	1.33 ± 0.02^{a}	81.19 ± 5.41^{a}	44.31 ± 6.89
p value	0.058	0.050	0.018	0.017	0.002	0.003	0.003	0.255

Different superscripts in the same column signify statistical differences ($p < 0.05$) (mean ± SE; n = 30 for length and weight measurements; n = 3 for WG, SGR, survival, FCR, FER, PER, ANPU and ERV)

[1] Weight gain percent = (final wt − initial wt)/initial wt × 100

[2] Specific growth rate = {ln(final wt) − ln(initial wt)}/experimental period in days × 100

[3] Feed conversion ratio = feed given (dry wt)/body wt gain (wet wt)

[4] Protein efficiency ratio = body wt gain (wet wt)/crude protein fed

[5] Apparent net protein utilization = (final carcass protein − initial carcass protein)/protein fed × 100

[6] Energy retention value = (final carcass energy − initial carcass energy)/energy fed (kcal) × 100

10 %. Increasing the EM inclusion level in feed to 15 % did not deter growth of the fish. Similarly, Liang and Lovell (1971) had demonstrated that the addition of 5–10 % *Eichhornia* meal to vitamin-free channel catfish diets significantly improved fish growth and survival. Niamat and Jafri (1984) also reported the possible use of water hyacinth leaf meal as a source of cheap plant protein for fish. There may be some unknown growth promoting and/or palatability factors present in EM, which need to be estimated for verifying this assumption. However, it can be mentioned here that the dry-powdered EM smelled very pleasant to human olfactory sense. But, EM levels higher than 15 % had caused reduction in the growth and feed efficiencies. This may be due to the presence of unknown anti-nutritional factors, high fibre and/or the higher ash content. According to Gohl (1981), fresh water hyacinth contained prickly crystals (supposedly oxalate salts), which reduced its palatability. However, according to Lareo and Bressani (1982) the levels of oxalate and other anti-physiological factors present in the plant were either very low or non-existent. They reported that the level of tannins was less than 1 % of dry matter in the whole plant and only 2 % in the leaves. In the present study, dietary *Eichhornia* meal level of 10 % showed better ANPU, PER, WG and SGR, therefore it can be deduced that protein and other nutrients from EM were better utilized at 10 % supplementation in *L. rohita* fingerlings. However, a cost/benefit analysis should be conducted to evaluate the economic feasibility of this feedstuff for *L. rohita*, and whether the reduction in the cost of EM-based diets would compensate for the reduction in fish performance at higher inclusion levels. The cost of one kilogram of feed (considering the cost of ingredients only in Guwahati, Assam, India) used in the present studies were: Rs. 15.8, 15.5, 15.2, 14.9 and 14.6 for the control, EMF1, EMF2, EMF3 and EMF4 feeds, respectively. Cost of feed decreased with the incorporation of EM, because this feedstuff is available plentifully free-of-cost. Some workers have considered the economic evaluation of unconventional feed inputs for tilapia (Fagbenro 1992; El-Sayed 2003, 2008). They demonstrated that most of these feed inputs produced lower biological performance than standard (conventional) sources, but the cost/benefit analysis indicated that they were economically superior.

The biochemical composition of the fish tissues indicated poor accumulation of crude protein in the groups fed the diet with 20 % EM, while the crude protein content in the other EM supplemented groups was comparable with the control. The higher content of plant protein in this group might have reduced its efficiency for assimilation and utilization of proteins. The protein digestibility in this group was also reduced. In the present investigation, the dry matter digestibility was affected by inclusion levels of

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Table 4 Tissue biochemical composition (% dry matter) of *Labeo rohita* fingerlings fed diets containing different levels of *Eichhornia* meal (EM) for 60 days

Components	Experimental groups (% EM)					*p* value	Initial fish
	Control (0)	EMF1 (5)	EMF2 (10)	EMF3 (15)	EMF4 (20)		
Moisture	79.47 ± 0.11^{a}	79.39 ± 0.41^{a}	79.27 ± 0.06^{a}	81.03 ± 0.13^{b}	80.11 ± 0.46^{a}	0.003	81.50 ± 0.25
Crude protein (CP)	60.55 ± 0.63^{b,c}	58.34 ± 0.74^{a,b}	60.39 ± 0.40^{b,c}	61.39 ± 0.41^{c}	57.10 ± 1.54^{a}	0.017	52.33 ± 0.01
Ether extract (EE)	12.93 ± 0.83	13.51 ± 1.14	11.84 ± 0.52	9.25 ± 0.69	13.49 ± 3.06	0.308	6.59 ± 0.10
Total carbohydrate (TC)	6.41 ± 0.66^{a}	8.37 ± 0.50^{a,b}	7.94 ± 0.76^{a,b}	7.92 ± 0.81^{a,b}	9.78^{b} ± 0.44	0.035	20.75 ± 0.79
Total ash	20.11 ± 0.18	19.79 ± 0.70	19.83 ± 0.17	21.45 ± 0.06	19.63 ± 1.12	0.248	20.33 ± 0.21
Digestible energy*	384.21 ± 4.77	388.39 ± 8.37	379.88 ± 3.24	360.45 ± 3.53	388.91 ± 19.71	0.294	351.64 ± 5.81

Different superscripts in the same column signify statistical differences ($p < 0.05$; mean ± SE; n = 6)

* Digestible energy (kcal/100 g) = (CP% × 4) + (EE% × 9) + (TC% × 4)

Table 5 Apparent dry matter digestibility[1] (%) and nutrient digestibility[2] (%) of *Eichhornia* meal (EM)-based diets fed to *Labeo rohita* fingerlings for 60 days

EM inclusion (%)	Dry matter digestibility	Protein digestibility	Lipid digestibility	Carbohydrate digestibility
Control (0)	79.04 ± 1.45^{c}	83.41 ± 1.20^{b,c}	82.43 ± 2.82	81.09 ± 1.03^{b}
EMF1 (5)	77.49 ± 1.59^{b,c}	82.37 ± 1.56^{b,c}	82.46 ± 1.62	80.79 ± 1.00^{b}
EMF2 (10)	76.62 ± 0.81^{a,b,c}	84.96 ± 0.10^{c}	82.80 ± 0.10	74.44 ± 1.86^{a}
EMF3 (15)	74.69 ± 1.06^{a,b}	80.39 ± 0.06^{a,b}	80.96 ± 3.33	76.39 ± 0.08^{a}
EMF4 (20)	72.96 ± 0.65^{a}	77.99 ± 0.63^{a}	80.19 ± 1.83	75.59 ± 1.25^{a}
p-value	0.037	0.018	0.894	0.031

Different superscripts in the same row signify statistical differences ($p < 0.05$; mean ± SE; n = 6)

[1] Apparent dry mater digestibility (%) = 100 − 100 (% marker in feed/% marker in faeces)

[2] Apparent nutrient digestibility (%) = 100 − 100 {(% marker in feed/% marker in faeces) × (% nutrient in faeces/% nutrient in feed)

macrophyte meal. This may be due to the higher amount of indigestible ash and fibre present in the feed at higher macrophyte meal level. Dry matter digestibility was higher at 10 % inclusion and it was reduced significantly from 15 % inclusion level onwards compared to control. In a study on rohu using water hyacinth, percentage dry matter digestibilities reported were 65 and 78 % when incorporated at 60 and 30 % levels, while for catla it varied between 48 and 74 % at incorporation levels of 45 and 15 %, respectively (Nandeesha et al. 1991; Hasan and Roy 1994). Studies in grass carp showed digestion of 50–60 % when water hyacinth was used in the feed (Riechert and Trede 1977). In contrast to these results, Ray and Das (1994) reported much higher protein digestibility value (94 %) of water hyacinth leaf meal for rohu fry (3.6 g). Apparent digestibility of water hyacinth was reported to vary between species and the level of incorporation (Hasan and Roy 1994; Murthy and Devaraj 1990).

Eichhornia meal inclusion at higher levels decreased the protein and carbohydrate digestibilities. Eichhornia meal inclusion up to 15 % level did not affect the protein digestibility, but carbohydrate digestibility reduced at 10 % inclusion level onwards. The reduced nutrient digestibility of the macrophyte meal-based diet was attributed to increasing ash and fibre contents of the diet which increased with the increasing level of macrophyte meal (De Silva and Perera 1983).

Conclusion

From the foregoing discussion, it is concluded that *Eichhornia* meal can be included at 15 % level in the feed of *L. rohita* fingerlings without adversely affecting the growth, dry matter and nutrient digestibility of the feed. Though further EM inclusion at 20 % showed poorer growth performance than the control, the feed was still visibly palatable to the fish. Ash, fibre and anti-nutritional factors might be adversely affecting inclusion of higher levels of the macrophyte meal in fish feeds. Though the former two inherent characters of the EM-supplemented diets will be difficult to deal with, but anti-nutritional factors (once characterized) may be suitably dealt with by exogenous supplementation of dietary enzymes. From the present

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study, it is advocated to conduct a cost/benefit analysis to evaluate the economic feasibility of this feedstuff for *L. rohita*, and whether the reduction in the cost of EM-based diets would compensate for the reduction in fish performance at higher inclusion level.

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