Growth performance of *Cirrhinus mrigala* (Ham.) fingerlings maintained on mixed feeding schedule of diets having different protein content

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ABSTRACT

Seven different mixed feeding schedules were evaluated in *Cirrhinus mrigala* fingerlings having mean body weight (BW) of 2.84g employing a low (L) protein (20.29%) diet and a high (H) protein (40.12%) diet, feeding @ 3% Bwd⁻¹ for 45 days. Regular feeding for 45 days on low protein (L) diet resulted in significantly (P<0.05) low growth and low protein efficiency ratio (PER), while feeding on 1L/3H feeding schedule resulted in good growth performance equivalent to the fish fed continuously on high protein diet. Gross protein retention (GPR), Gross energy retention (GER), Apparent nutrient digestibility (APD) and Feed conversion ratio (FCR) were similar in fingerlings fed either continuously on high protein diet or on a feeding schedule consisting of 1L/3H, while, PER values were significantly (P<0.05) high in fingerlings fed on 1L/3H diet in comparison with fish fed continuously on high protein diet. Based on total protein input, 12.36% protein can be saved without affecting growth by adopting the 1L/3H feeding schedule as compared to feeding the fish daily on high protein diet.

Introduction

Feed input is the single largest operational cost in majority of aquaculture practices, because of the high cost of protein. Usually fish growth is directly proportional to the protein content of the feeds. The optimal dietary protein required for maximal growth in farmed fishes is reported to be 50-300% higher than that of terrestrial farm animals (Cowey, 1975). These quantitative differences have been attributed to carnivorous/omnivorous feeding habits of the fishes and also to their apparent preferential use of protein over carbohydrate as a dietary protein source. The high requirement of protein by fishes and the pollution it causes have received a lot of attention (Kibria et al., 1998). To maximize nutrient utilization and minimize the solid and soluble waste load, it is therefore essential to provide optimum levels of protein to farmed fish (Cho, 1993).

Although the technique of carp culture has been standardized to some extent in India, still there is ample scope for augmenting their production through better management practices and optimization of the quantity of protein to be given through artificial feeds (Kalla et al., 2004; Singh et al., 2004). Much of the efforts in this line are directed towards reducing the protein intake without compromising growth performance and to reduce nitrogen waste in the environment. This can be achieved through the adoption of alternate (mixed) feeding schedule which has been investigated by several researchers (De Silva, 1985; Srikanth et al., 1989; Saroha et al., 2004). The studies of De Silva (1985) in *Oreochromis niloticus* and of Nandeesh et al. (1993) in Indian major carps have clearly shown...
the possibility of saving significant amount of protein input without affecting growth through the adoption of mixed feeding schedules. These studies indicated the economic viability of feeding the fish with diets of different protein content instead of feeding on a constant level of recommended protein in the diet. Therefore, the present investigation was undertaken to test the effect of mixed feeding schedule of low and high protein diets in *C. mrigala* with a view to save total feed costs and also to reduce the excretion of metabolic wastes (N-NH₄ and o-PO₄).

**Materials and methods**

Fingerlings of *C. mrigala* (mean BW 2.84 g) were obtained from the local suppliers of Hisar (Lat. 29°10’N and Long. 75°46’E). Fish were maintained in transparent glass aquaria (60X30X30 cm) kept in an air-conditioned laboratory where the temperature was maintained at 25±1°C and the lighting schedule (LD 12:12) at 12h of light (8 a.m. to 8 p.m.) alternating with 12h of darkness (8 p.m. to 8 a.m.). The average intensity of light inside the laboratory was approximately 1000 lux. Fish were acclimated in the laboratory for a minimum period of seven days prior to the initiation of experimental treatments and fed *ad libitum* on a feed containing 40% protein. The water in the aquaria was renewed daily with water which has been previously equilibrated to the desired temperature (25°C).

**Experimental diets**

Two diets with low (20%) and high (40%) protein contents were formulated using processed soybean as the major protein source.

Dietary ingredients were cleaned, milled and mixed in definite proportions and a thick dough was made using lukewarm water. Wheat flour was used as binder. To each diet, 1% chromic oxide (Cr₂O₃) was added as external digestibility marker. Using a mechanical pelletizer, 0.5 mm thick pellets were obtained. The pellets were oven dried (60-62°C) and stored in air tight containers. Dietary ingredients and proximate composition of the formulated diets are given in Table 1.

Fish were individually weighed on a top pan balance and randomly distributed @ 10 fish per aquaria with two replicates for each treatment, and fed @ 3% BWd⁻¹. The following seven feeding schedules were maintained.

- **Treatment 1**: Feeding daily on low protein diet (L)
- **Treatment 2**: Feeding daily on high protein diet (H)
- **Treatment 3**: Feeding one day low protein diet alternating with one day high protein diet (1L/1H)
- **Treatment 4**: Feeding one day low, alternating with two days high protein diet (1L/2H)
- **Treatment 5**: Feeding one day low, alternating with three days high protein diet (1L/3H)
- **Treatment 6**: Feeding two days low, alternating with two days high protein diet (2L/2H)
- **Treatment 7**: Feeding two days low, alternating with three days high protein diet (2L/3H)

The amount of feed given was adjusted every 15th day after estimating the biomass of each group of fish. Fish were exposed to the diets continuously for 3h and thereafter uneaten feed was siphoned out and stored separately for drying and calculation of feed conversion ratio (FCR). The faecal matter voided by the fish was collected separately from each aquarium. The pooled faecal samples from each treatment were dried in an oven at 60°C and subsequently analyzed for digestibility estimations. On termination of experiment, the fish from all the treatments were weighed individually on a top pan balance and processed for subsequent analysis. The pH of aquaria water fluctuated between 7.25 and 7.55 and dissolved oxygen content ranged between 5-7 mg l⁻¹.
The feed ingredients, experimental diets, faecal samples, fish carcass (initial and final) were analysed following AOAC (1995). Chromic oxide levels in the diets as well as in the faecal samples were determined spectrophotometrically following the method of Furukawa and Tsukahara (1966). Dissolved oxygen and pH were monitored using F-set-3 (E. Merck, Germany).

Data analyses

Growth performance of *Cirrhinus mrigala* fingerlings on mixed feeding schedule of diets

**TABLE 1: Percentage composition of ingredients and proximate composition of experimental diets fed to *C. mrigala***

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>Diets</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>High protein</td>
</tr>
<tr>
<td>Groundnut oilcake</td>
<td>65.00</td>
</tr>
<tr>
<td>Processed full fat soybean*</td>
<td>23.50</td>
</tr>
<tr>
<td>Wheat flour</td>
<td>4.75</td>
</tr>
<tr>
<td>Rice bran</td>
<td>4.75</td>
</tr>
<tr>
<td>Soya oil</td>
<td>-</td>
</tr>
<tr>
<td>Chromic oxide (Cr₂O₃)</td>
<td>1.00</td>
</tr>
<tr>
<td>Mineral premix and amino acids (MPA)</td>
<td>1.00</td>
</tr>
</tbody>
</table>

**Proximate composition (%)**

<table>
<thead>
<tr>
<th></th>
<th>High protein</th>
<th>Low protein</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dry matter</td>
<td>93.33±0.062</td>
<td>92.40±0.040</td>
</tr>
<tr>
<td>Crude protein</td>
<td>40.12±0.105</td>
<td>20.29±0.102</td>
</tr>
<tr>
<td>Crude fat</td>
<td>8.13±0.125</td>
<td>8.12±0.017</td>
</tr>
<tr>
<td>Crude fibre</td>
<td>8.19±0.029</td>
<td>7.94±0.022</td>
</tr>
<tr>
<td>Ash</td>
<td>5.93±0.030</td>
<td>5.46±0.024</td>
</tr>
<tr>
<td>Phosphorous</td>
<td>0.56±0.008</td>
<td>0.51±0.005</td>
</tr>
<tr>
<td>NFE</td>
<td>30.95±0.106</td>
<td>50.58±0.130</td>
</tr>
<tr>
<td>Energy content (KJ g⁻¹)</td>
<td>18.00±0.003</td>
<td>16.68±0.019</td>
</tr>
</tbody>
</table>

All values are mean± S.E.of mean.

*Soybean seeds were hydrothermically processed in an autoclave at 15 lbs for 15 min to eliminate anti-nutritional factors (Garg et al., 2002).*

The feed ingredients, experimental diets, faecal samples, fish carcass (initial and final) were analysed following AOAC (1995). Chromic oxide levels in the diets as well as in the faecal samples were determined spectrophotometrically following the method of Furukawa and Tsukahara (1966). Dissolved oxygen and pH were monitored using F-set-3 (E. Merck, Germany).

Data analyses

- **Live weight gain (g), growth rate (per day)** in percentage body weight, percentage gain, specific growth rate (% d⁻¹), food conversion ratio (FCR), protein efficiency ratio (PER), gross protein retention (GPR) and gross energy retention (GER) were calculated according to the following equations.

  \[
  \text{Live weight gain} = W_2 - W_1 \\
  \text{Growth (% gain in body weight)} = \frac{W_2-W_1}{W_1} \times 100 \\
  \text{SGR} = \frac{\ln W_2 - \ln W_1}{t} \times 100 \\
  \text{FCR} = \frac{\text{Feed given (dry wt. g)}}{\text{Body weight gain (wet wt. g)}} \\
  \text{PER} = \frac{\text{Gain in wet weight of fish (g)}}{\text{Dry weight of protein fed (g)}} \\
  \text{GER (KJ)} = \frac{\text{Fish energy gain (KJ)}}{\text{Energy intake (KJ)}} \times 100 \\
  \text{GPR (g)} = \frac{\text{Fish protein gain (g)}}{\text{Protein intake (g)}} \times 100
  \]

Where,

- \( W_1 = \) Initial weight
- \( W_2 = \) Final weight
Apparent nutrient digestibility (APD) of the diet calculated according to Cho et al. (1982) is as follows:

\[
\text{APD} = 100 - 100 \times \left( \frac{\text{% Cr}_2\text{O}_3 \text{ in diet}}{\text{% Cr}_2\text{O}_3 \text{ in faeces}} \times \frac{\text{% nutrient in faeces}}{\text{% nutrient in diet}} \right)
\]

The energy content of the diets and fish were calculated using the average caloric conversion factors of 0.3954, 0.1715 and 0.2364 KJg\(^{-1}\) for lipid, carbohydrate and protein respectively (Henken et al., 1986).

**Results**

**Growth and survival**

The growth response of *C. mrigala* fingerlings fed on seven different dietary schedules is shown in Table 2. Live weight gain, growth (percent gain in body weight) and specific growth rate (SGR) of fish fed on seven different dietary schedules revealed a highly varied pattern. Continuous feeding on low protein diet resulted in significantly (P<0.05) low growth, while feeding on IL/3H diet resulted in good growth performance which was almost equivalent to the fingerlings fed continuously on high protein diet.

Feed and nutrient utilization efficiencies were measured in terms of FCR, PER, GPR and GER. Significant (P<0.05) variations in these parameters were also observed among different feeding schedules. FCR values were significantly (P<0.05) lowered in groups fed continuously on high protein diet or on 1L/3H feeding schedule. On the other hand, nutrient retention values (GPR, GER and APD) were significantly (P<0.05) enhanced in fingerlings maintained on a feeding schedule consisting of 1L/3H, which however, were not significantly (P>0.05) different from the fish fed continuously on high protein diet (Table 2).

**Proximate carcass composition**

Final body composition of fish changed significantly (P<0.05) in comparison with the initial composition. Protein and energy contents increased, while those of moisture and ash decreased with increase in the high protein ratio (1L/1H, 1L/2H, 1L/3H). Significantly (P<0.05) high protein and energy levels were seen in fish fed continuously on high protein diet as compared with the fish maintained on other feeding schedules including 1L/3H. Fat deposition was significantly higher (P<0.05) in groups fed on high protein as well as 1L/3 diet compared to the other groups. Carcass phosphorous contents remained significantly (P<0.05) high in all the treatments except in the group fed on low protein diet (Table 3).

**Discussion**

The results of the present study on growth performance and nutrient utilization have revealed that it is economically beneficial and biologically more productive to feed *C. mrigala* one day on low, followed by three days on high (1L/3H) protein diets instead of feeding the fish continuously only on high protein (HP) diet. It appears that this fish does not require the same protein input every day like common carp (Srikanth et al., 1989) and nile tilapia (De Silva, 1985).

GPR, GER, APD and FCR values were similar in fingerlings fed either continuously on high protein diet or on a feeding schedule consisting of 1L/3H. PER values were significantly high in fish fed on 1L/3H diet in comparison with the fingerlings fed continuously on high protein diet. Nandeesha et al. (1993) evaluated mixed feeding schedule on growth performance of *Catla catla* and *Labeo rohita*. Their studies have revealed that feeding for one or two days on low protein (15.8%) diet followed by three days on high protein (31.8%) diet was the best schedule for catla. On the other hand, feeding rohu, alternatively on low and high protein diets (1L/1H) or one or two days on low protein diet, followed by three days on high protein diet were found to be the most suitable feeding schedules. Saroha et al. (2004) in *Channa punctatus* and Garg et al. (unpublished) in *Heteropneustes*...
TABLE 3: Influence of various feeding regimes on proximate carcass composition (% wet weight basis) in C. mrigala fingerlings

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control</th>
<th>LP</th>
<th>HP</th>
<th>1L/1H</th>
<th>1L/2H</th>
<th>1L/3H</th>
<th>2L/2H</th>
<th>2L/3H</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture</td>
<td>76.40±0.029</td>
<td>75.46±0.062</td>
<td>73.07±0.031</td>
<td>74.32±0.020</td>
<td>73.84±0.028</td>
<td>73.45±0.031</td>
<td>74.28±0.020</td>
<td>74.09±0.031</td>
</tr>
<tr>
<td>Crude protein</td>
<td>11.71±0.110</td>
<td>12.02±0.020</td>
<td>14.44±0.012</td>
<td>13.18±0.028</td>
<td>13.57±0.034</td>
<td>13.92±0.023</td>
<td>13.18±0.028</td>
<td>13.56±0.014</td>
</tr>
<tr>
<td>Crude fat</td>
<td>3.98±0.011</td>
<td>4.70±0.005</td>
<td>5.10±0.040</td>
<td>4.87±0.031</td>
<td>4.98±0.031</td>
<td>5.06±0.020</td>
<td>4.88±0.028</td>
<td>4.94±0.037</td>
</tr>
<tr>
<td>Ash</td>
<td>3.32±0.026</td>
<td>3.25±0.028</td>
<td>2.73±0.023</td>
<td>2.04±0.023</td>
<td>2.93±0.057</td>
<td>2.89±0.057</td>
<td>3.05±0.023</td>
<td>2.96±0.034</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>0.45±0.001</td>
<td>0.51±0.005</td>
<td>0.56±0.008</td>
<td>0.54±0.005</td>
<td>0.55±0.005</td>
<td>0.56±0.005</td>
<td>0.55±0.005</td>
<td>0.55±0.005</td>
</tr>
<tr>
<td>Gross energy (kJ/g)</td>
<td>5.12±0.004</td>
<td>5.48±0.012</td>
<td>6.22±0.018</td>
<td>5.82±0.016</td>
<td>5.98±0.024</td>
<td>6.09±0.015</td>
<td>5.83±0.007</td>
<td>5.91±0.002</td>
</tr>
</tbody>
</table>

All values are mean ± S.E. of mean. Means with the same letter/s in the same row are not significantly (P>0.05) different.

LP=Low protein diet, HP=High protein diet

TABLE 2: Influence of various feeding regimes on growth, digestibility and nutrient retention in C. mrigala fingerlings

<table>
<thead>
<tr>
<th>Parameters</th>
<th>LP</th>
<th>HP</th>
<th>1L/1H</th>
<th>1L/2H</th>
<th>1L/3H</th>
<th>2L/2H</th>
<th>2L/3H</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial live weight (g)</td>
<td>2.94</td>
<td>2.92</td>
<td>2.76</td>
<td>2.89</td>
<td>2.90</td>
<td>2.79</td>
<td>2.66</td>
</tr>
<tr>
<td>Final live weight (g)</td>
<td>3.88</td>
<td>5.31</td>
<td>4.43</td>
<td>4.74</td>
<td>5.18</td>
<td>4.54</td>
<td>4.43</td>
</tr>
<tr>
<td>Live weight gain (g)</td>
<td>0.94±0.022</td>
<td>2.38±0.071</td>
<td>1.66±0.030</td>
<td>1.85±0.016</td>
<td>2.28±0.090</td>
<td>1.74±0.030</td>
<td>1.76±0.035</td>
</tr>
<tr>
<td>Growth (percent gain in body weight)</td>
<td>31.96±0.118</td>
<td>81.51±0.500</td>
<td>60.14±0.372</td>
<td>64.21±1.316</td>
<td>78.60±3.151</td>
<td>62.68±1.675</td>
<td>66.19±1.303</td>
</tr>
<tr>
<td>Specific growth rate (SGR % d⁻¹)</td>
<td>0.62±0.000</td>
<td>1.32±0.006</td>
<td>1.04±0.005</td>
<td>1.10±0.017</td>
<td>1.28±0.040</td>
<td>1.08±0.022</td>
<td>1.12±0.017</td>
</tr>
<tr>
<td>Feed conversion ratio (FCR)</td>
<td>3.70±0.005</td>
<td>1.44±0.002</td>
<td>1.91±0.005</td>
<td>1.81±0.047</td>
<td>1.48±0.058</td>
<td>1.86±0.046</td>
<td>1.72±0.034</td>
</tr>
<tr>
<td>Protein efficiency ratio (PER)</td>
<td>1.32±0.008</td>
<td>1.73±0.004</td>
<td>1.74±0.004</td>
<td>1.64±0.044</td>
<td>1.91±0.073</td>
<td>1.78±0.044</td>
<td>1.80±0.035</td>
</tr>
<tr>
<td>Gross protein retention efficiency (GPR)</td>
<td>17.29±0.658</td>
<td>30.80±0.303</td>
<td>27.19±0.742</td>
<td>27.06±0.742</td>
<td>32.10±1.024</td>
<td>27.66±0.724</td>
<td>29.23±0.510</td>
</tr>
<tr>
<td>Gross energy retention efficiency (GER)</td>
<td>10.64±0.110</td>
<td>29.17±0.166</td>
<td>21.19±0.144</td>
<td>22.90±0.547</td>
<td>27.95±0.024</td>
<td>21.60±0.438</td>
<td>23.64±0.060</td>
</tr>
<tr>
<td>Apparent protein digestibility (APD)</td>
<td>87.25±0.651</td>
<td>89.44±0.251</td>
<td>88.34±0.281</td>
<td>88.04±0.141</td>
<td>88.89±0.106</td>
<td>87.50±0.061</td>
<td>88.16±0.482</td>
</tr>
</tbody>
</table>

All values are mean ± S.E. of mean. Means with the same letter/s in the same row are not significantly (P>0.05) different.

LP=Low protein diet, HP=High protein diet

Growth performance of Cirrhinus mrigala (Ham) fingerlings on mixed feeding schedule of diets
fossilis obtained high growth when fed on a feeding schedule consisting of IL/3H diets. However, no well defined patterns in growth and digestibility parameters were observed in O. niloticus (De Silva, 1985) and in common carp (Srikanth et al., 1989; Nandeesh, 1990).

FCR and PER remained low in fish fed continuously on high protein diet as compared to those groups maintained on mixed feeding schedules in the present study. These findings are similar to those reported in tilapia (De Silva, 1985); C. catla and L. rohita (Srikanth et al., 1989; Nandeesh et al., 1993); H. fossilis (Garg et al., unpublished) and C. punctatus (Saroha et al., 2004). The optimum protein requirements of both fry and fingerlings of C. mrigala for high survival and growth have been investigated to be around 40% both under laboratory and field conditions (Kalla et al., 2003; 2004; Singh et al., 2004). The high growth and low FCR recorded in fish fed continuously on high (40%) protein diet in the present study is in agreement to these studies. Kalla et al. (2003) observed no improvement in growth parameters with further increase in protein levels; rather a slight decrease in growth values was observed in C. mrigala.

Fish carcass analyzed at the beginning and at the end of the experiment indicated a decline in moisture and ash contents, while an increase in protein accumulation in all the treatments was observed. The fish fed continuously on low protein diet had low carcass protein and high ash contents, while fingerlings fed on high protein diets had low ash contents and slightly higher values of carcass protein than fingerlings fed on 1L/3H feeding schedule. Srikanth et al. (1989) also observed high deposition of protein in common carp. Carcass moisture and lipid appeared to be inversely related and these results appear to be in agreement to those reported by Ray and Das (1992) in L. rohita and Shearer (1994) in trout. Slight variations in carcass phosphorus levels were observed, however, these values were within the normal range.

In the absence of sufficient/optimal protein contents in the diets, fish use the energy of lipids and carbohydrates. Lipids are well utilized as energy sources but only restricted amounts can be used because a large supply of dietary fat affects carcass composition, which are undesirable owing to high processing losses, storage problems and consumer acceptability and also brings about technical problems during diet preparation. Further, addition of substantial quantity of dietary lipid results in excess fat deposition and growth reduction (Torrbee et al., 1993). Since the fat content in the diets were fixed at about 8% and the crude fat content of the carcass varied between 4.7-5.1 in mrigal, these values are considered to be not high enough to degrade the quality of fish flesh.

The differential response in growth performance of fish fed on diets may be attributed to the differences in protein levels. Alternate administration of high/low protein diets has influenced the growth performance of fish independently of the mean dietary protein input, which may be attributed to a rhythm in certain basic metabolic activities (De Silva, 1985). Similar response of fish fed on high protein diet or 1L/3H schedule may indicate that the fingerlings might have used the necessary nutrients and energy from the carbohydrates of the diets which were about 20% high in low protein diet. According to Flange et al. (1978), the increase in carbohydrate content of the diet actually reduces the activity of the proteolytic enzymes. Results of the present study revealed that digestibility of the protein is affected by carbohydrate content of the diets.

Since growth performance was not significantly different in fish fed either continuously on high protein diet or on a feeding schedule consisting of 1L/3H, it is possible to save 12.36% protein by adopting 1L/3H feeding schedule. De Silva (1985), Srikanth et al. (1989), Nandeesh (1990) also found 1L/3H schedule to be the best for farmed fish.
The studies have revealed the possibility of saving protein by adopting mixed feeding schedule without compromising growth and nutrient retention. Although the present study provides some indication on the growth pattern, the findings have to be verified by large scale field trials prior to adoption in fish culture ponds.

Acknowledgements

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