UTILISATION OF SUN-DRIED *AZOLLA CAROLINIANA* IN SUBSTITUTING GROUNDNUT OIL CAKE IN THE CONVENTIONAL CARP FEED

P. C. Das, D. P. Sinhababu, D. P. Singh and P. K. Sahu

Fish and Fisheries Unit, Central Rice Research Institute
Cuttack, Orissa, India

Sun-dried and powdered *Azolla caroliniana* substituting 20, 40, 60 and 63.6% of the protein supplement (groundnut oil cake) of the conventional diet (at 10, 29.2, 60 and 63.6% incorporation in diet, respectively) of Indian major carp was fed to *Labeo rohita* fingerlings for a period of 90 days. Substitution of groundnut oil cake up to 40% in the protein supplement resulted in 115.48±4.20% and 24.85±4.22% increment in the fish body weight and length, as against compared to 100.40±4.53% and 23.38±1.05% in the control, respectively. There was no significant \((P < 0.05)\) variation in the muscle protein and moisture contents. Feeding on a diet with increased substitution of groundnut oil cake by *Azolla* (60% or more in protein supplement) resulted in lower body weight and length increment as well as lower muscle protein content, with no significant difference in moisture content as compared to the control diet.

INTRODUCTION

Indian major carps, *viz.*, *Catla catla* (Hamilton), *Labeo rohita* (Hamilton) and *Cirrhinus mrigala* (Hamilton) are the most important freshwater carp species for aquaculture in India and neighbouring countries. In India, these species contribute almost 84% of the total freshwater aquaculture production. A number of formulated diets suiting to the herbivorous feeding habit of these species are commercially available. But, majority of the fish farmers depend on the conventional feed mixture containing groundnut oil cake (GNOC) and rice bran for feeding these species in culture ponds. Considering the high cost and often non-availability of GNOC in the market, there is a need for development of low cost diet by substituting the protein supplement with a low cost farm-based input.

*Azolla* is a heterosporous fern belong to the family Azollaceae (Salviniaceae). It contains 13-30% crude protein, 4.4-6.3% crude fat, 5.6-15.2% cellulose, 9.8-17.9% hemicellulose, 9.3-34.8% lignin and 9.7-23.8% ash on dry weight basis (Ayyappan, 2000). The rapid growth (1.5 kg fresh weight/m² water spread area per week), short doubling time (3-5 days), high contents of nitrogen and potassium as well as high decomposition rate (1.36-4.57% per day) make *Azolla* a potential bio-fertiliser for use in aquaculture ponds. It
has been used as a source of dietary protein for livestock (Nik and Motaghi, 1992; Sreemannaryana et al., 1993; Tamang and Samanta, 1993) and fish (Micha et al., 1988; El Syed, 1992; Joseph et al., 1994; Mohanty and Dash, 1995) because of its high nitrogen content. While in one hand incorporation of Azolla has demonstrated positive impact in several species (Santiago et al., 1988; Mohanty and Dash, 1995), in species like pearl spot (Etroplus suratensis) and Nile tilapia the Azolla supplemented diet resulted significantly lower growth and feed utilisation (Joseph et al., 1994; El Syed, 1992) compared to fish meal protein based control diet. Considering limited information available on the use of Azolla as a protein supplement for the Indian major carps, the present study was undertaken to evaluate efficiency of Azolla supplemented feed on the performance of rohu, L. rohita.

MATERIAL AND METHODS

The experiment was conducted in rectangular cemented tanks (2 m length x 1 m width x 1.5 m height) during July to October, 2001 using a randomised complete block design (RCBD) with five treatments and four replications for each treatment. The three feed ingredients, GNOC, dried Azolla (A. caroliniana Wild) powder and rice bran were used to prepare the different combinations of feed. The protein content of Azolla, GNOC and rice bran was 15.8, 47.7 and 7%, respectively. The conventional fish feed (GNOC and rice bran in 1:1 ratio) with a protein content of 27.4% served as the control (T1). The other treatments (T2, T3, T4 and T5) involving different combinations of the feed ingredients were formulated by keeping protein content same as that of the control. While in treatments T2, T3 and T4, Azolla substituted GNOC to the extent of 20, 40 and 60% of the protein supplement in the feed, T5 contained only Azolla and GNOC (Table 1).

Table 1. Share of different ingredients in the feed mixture fed to fingerlings of Labeo rohita

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Protein supplement</th>
<th>Protein (%)</th>
<th>Share in diet (%)</th>
<th>% feed ingredients in diet</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Azolla (%)</td>
<td>GNOC (%)</td>
<td>Protein supplement</td>
<td>Basal feed</td>
</tr>
<tr>
<td>T1 (control)</td>
<td>0</td>
<td>100</td>
<td>47.7</td>
<td>7.0</td>
</tr>
<tr>
<td>T2</td>
<td>20</td>
<td>80</td>
<td>47.6</td>
<td>7.0</td>
</tr>
<tr>
<td>T3</td>
<td>40</td>
<td>60</td>
<td>35.0</td>
<td>7.0</td>
</tr>
<tr>
<td>T4</td>
<td>60</td>
<td>40</td>
<td>28.6</td>
<td>7.0</td>
</tr>
<tr>
<td>T5</td>
<td>63.6</td>
<td>36.4</td>
<td>27.4</td>
<td>7.0</td>
</tr>
</tbody>
</table>

Fingerlings of L. rohita (average body weight 7.9±0.3 g and length 95.3±3.0 mm) were collected from the rice-fish nursery ponds, treated with potassium permanganate and then released in the cemented tanks (10 fingerlings in each tank) filled with filtered
water from the reservoir pond up to one metre height. The fishes were fed at 5% of body weight with conventional feed (GNOC and rice bran) for 15 days before the start of the experiment. Body weight and length of the fingerlings were recorded at the start of the experiment. Every alternate day, around 10% water from each tank was drained out by the bottom-siphoning method and it was replaced with fresh water to maintain the water height uniformly at 1 m. Important water parameters such as temperature, dissolved oxygen, pH, temperature, total alkalinity and total hardness were measured at weekly intervals to monitor any major change in the quality. The fishes were fed with the experimental diet at 5, 4 and 3% of the body weight during 0-30, 30-60 and 60-90 days of the experiment, respectively. The fingerlings were sampled for the body weight and length measurement on 30, 60 and 90 days of the experiment. At the end of the experiment, muscle samples were collected from the fingerlings in each replication and the protein content was estimated following the micro-Kjeldahl method. The data were statistically analysed (Gomez and Gomez, 1967) and Duncan’s multiple range test was applied to find significant differences among the treatments.

RESULTS AND DISCUSSION

The data on various water parameters recorded at weekly intervals revealed that the mean water temperature (25.3±2.6°C), dissolved oxygen (5.6±0.6 mg/l), pH (8.1±0.3), free carbon dioxide (nil), total hardness (60±8 mg CaCO₃/l), total alkalinity (56±6 mg CaCO₃/l) were well within the range required for normal growth of *Labeo rohita* fingerlings.

The fingerlings fed on dried *Azolla* powder at 10 and 29.2% inclusion levels (20 and 40% substitution of GNOC, respectively) recorded similar growth as those with the control diet (Table 2). At 29.2% inclusion level, the fingerlings recorded the highest

Table 2. Growth, moisture content and muscle protein content in fingerlings of *Labeo rohita* fed with *Azolla*-based diet

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Body weight (g)</th>
<th>Body length (mm)</th>
<th>*Muscle protein on dry weight basis (%)</th>
<th>Moisture (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Initial</td>
<td>Final</td>
<td>Increment (%)</td>
<td>Initial</td>
</tr>
<tr>
<td><strong>T₁</strong></td>
<td>8.0±0.2</td>
<td>16.0±1.0</td>
<td>100.4±4.5</td>
<td>95.4±4.7</td>
</tr>
<tr>
<td><strong>T₂</strong></td>
<td>8.0±0.3</td>
<td>15.4±0.5</td>
<td>94.2±5.0</td>
<td>97.0±0.8</td>
</tr>
<tr>
<td><strong>T₃</strong></td>
<td>8.2±0.4</td>
<td>17.6±0.7</td>
<td>115.5±4.2</td>
<td>96.5±3.2</td>
</tr>
<tr>
<td><strong>T₄</strong></td>
<td>7.6±0.1</td>
<td>11.5±1.4</td>
<td>51.9±8.7</td>
<td>93.0±0.2</td>
</tr>
<tr>
<td><strong>T₅</strong></td>
<td>7.9±0.2</td>
<td>11.3±0.5</td>
<td>44.2±2.2</td>
<td>94.6±2.0</td>
</tr>
</tbody>
</table>

The values with same superscript in a column do not differ significantly \((p > 0.05)\)
increment in both body weight (115.5%) and length (24.9%). Mohanty and Dash (1995) reported 168.2% weight gain and good diet utilisation in rohu fry fed with \textit{A. caroliniana} at 60% inclusion level, comparing diets with 30, 40, 50 and 60% \textit{Azolla} incorporation. Santiago \textit{et al.} (1988) observed increased growth and improved food conversion ratio in Nile tilapia (\textit{Oreochromis niloticus}) with increasing levels of dried \textit{A. pinnata} in the diet with no influence on survival rate. In the present experiment also, no significant variation in fish mortality was observed in any of the treatments, irrespective of the inclusion level of \textit{Azolla}.

The growth of the fingerlings decreased significantly when the level of \textit{Azolla} in the diet was increased to 56.8 and 63.6% in treatments $T_4$ and $T_5$, respectively, differing the findings of Mohanty and Dash (1995) in fry of the same fish. Possibly, the workers had used another strain of \textit{A. caroliniana} with a different nutrient composition. With increasing dietary levels of \textit{Azolla}, Almazan \textit{et al.} (1986) reported lower growth performance and poor food conversion ratio in Nile tilapia (\textit{O. niloticus}) fed with fresh \textit{Azolla} or dried \textit{Azolla} in pellet and powder form. Similar results were reported by Joseph \textit{et al.} (1994) in \textit{E. suratensis} fed with increasing dietary levels of \textit{Azolla} (above 25%) and Fasakin and Balogun (1998) in \textit{Clarias gariepinus} fingerlings. Replacement of GNOC with \textit{Azolla} above 40% in protein supplement (29.2% \textit{Azolla} in diet) in the present study gave similar result of lower growth than the control, suggesting that the level of replacement of the protein supplement should be carefully fixed. A reduction in the fish growth at higher levels of \textit{Azolla} supplementation could be due to its poor digestibility and higher lignin content (Ayyappan, 2000).

Comparison of the muscle protein content on dry weight basis revealed no significant differences ($P > 0.05$) among treatments $T_1$ (control), $T_2$ and $T_3$, whereas treatments $T_4$ and $T_5$ showed significantly lower protein content. Treatment $T_3$ recorded the highest muscle protein content of 82.6±0.8%. Micha \textit{et al.} (1988) reported that \textit{Azolla} incorporation in the diet decreased growth rate, increased the water and ash content and drastically reduced lipid content of the body tissues in \textit{O. niloticus} and \textit{Tilapia rendali}, while crude protein content was not affected significantly. In the present study, crude protein content marginally increased with \textit{Azolla} at 29.2% inclusion level and then decreased significantly when level of \textit{Azolla} was increased to 56.8 and 63.6%. Reduction of muscle protein content in \textit{E. suratensis} fed with a diet containing more than 25% \textit{Azolla} has been reported earlier (Joseph \textit{et al.}, 1994).

The moisture content of the fingerlings in different treatments with varying levels of \textit{Azolla} did not differ significantly. This observation supports the findings of Micha \textit{et al.} (1988) in \textit{O. niloticus} but contradicts the findings of Joseph \textit{et al.} (1994) in \textit{E. suratensis}, whose moisture content increased with the use of \textit{Azolla}. 
It is concluded that *Azolla* can effectively substitute the protein supplement (GNOC) and rice bran in the conventional diet of Indian major carps by about 30%, without any adverse effects on their survival, growth rate and muscle protein content.

**ACKNOWLEDGEMENTS**

The authors are thankful to the Director, Central Rice Research Institute, Cuttack for providing facilities for conducting the experiment.

**REFERENCES**


