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Comparative Evaluation of Plant-Protein Blends and Mustard Oil as Dietary Regimes on Growth Performance and Whole-Body Proximate Composition of Major Carps

Review Article

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Abstract

This study evaluated the proximate composition of *Cyprinus carpio, Labeo rohita,* and *Hypophthalmichthys molitrix* fed on plant-based diets compared with controls, along with the nutrient profile of the plant feed. A comparative experimental design was used to assess nutrient variations among fish species and diets. The experiment was conducted at Ratta Kulachi Hatchery and related laboratory facilities. Fingerlings of three species were divided into control and plant-fed groups. Moisture, ash, fiber, crude fat, crude protein, nitrogen-free extract (NFE) and energy were determined by standard methods. Plant feeding significantly alters nutrient composition across species. In *C. carpio*, plant-fed fish showed higher moisture (72.51%) but reduced fat (4.0%) compared to control (70.32% moisture, 5.0% fat). *L. rohita* on plant diet had higher protein (13.0%) but lower fat (3.90%) versus control (12.22% protein, 6.0% fat). Plant-fed *H. molitrix* showed the highest protein (16.0%) and fiber (3.33%), while its control had the lowest protein (10.0%). Ash was highest in *H. molitrix* control (4.0%) and lowest in *L. rohita* control (1.41%). NFE peaked in plant feed (65.34%) but was lowest in *L. rohita* control (6.01%). Energy ranged from 298.54 Kcal/100 g in plant feed to 114.40 Kcal/100 g in *H. molitrix* control. Plant-based diets influenced nutrient deposition differently among species, with *H. molitrix* showing superior protein utilization. Plant feed, rich in carbohydrates and energy, has potential as an aquaculture feed ingredient.

Keywords: Proximate Composition, Aquaculture Nutrition, *Cyprinus Carpio*, *Labeo Rohita*, *Hypophthalmichthys Molitrix*, Plant Feed.

Introduction

Fisheries industry is a major aspect of Pakistani economy because it is one of the major sources of income to a considerable number of people. Inland fisheries and sea fisheries, in which dams, lakes, ponds, rivers, and other water bodies are used, and commercial fish farming are also important activities in the whole country (Hassan *et al.*, 2025). Quality nutrition diet at reasonable costs contributes significantly to quality aquaculture feed in the aquaculture business to farmed fish. Dietary soybean feed is a rapidly growing product in fish feed, due to its balanced amino acid ratio and high protein content, which can satisfy the nutritional requirements of different fish species (Kumar *et al.*, 2022). A cost-effective balanced diet utilizing locally available plant by-products is essential for commercial fish culture. The most expensive source of protein in aquaculture feed is fish meal. Numerous experiments have shown significant success in replacing fishmeal with soybean meal and other soybean products, at least in part in the ration of most fish species (Abdullah *et al*., 2024). It is increasingly recognized that aquaculture is a very significant sector in improving food security and nutrition, particularly in the underdeveloped nations, which are also the poorest countries in the world. Farmed fish are provided with the required nutrients by manufactured feeds and thereby play a definitive role in modern commercial aquaculture. The food components in the food in the form of pellets or grains are effective and concentrated which facilitates the optimum growth of the fishes and effective feeding. The diet of these animals includes fishmeal and fish oil as an inseparable part (Naylor, Fang, & Fanzo, 2023). They are mixed to produce feed pellets reinforced with other nutrients like cereal grains, vitamins, minerals and vegetable proteins. It is commonly used wheat because it helps bind up the contents of fish feed. Other fish feeds are trash fish, where the fish themselves are caught and fed directly to larger aquaculture species; moist fish feed, available to a small number of fish species but difficult to store; and all-vegetable feeds such as feeds used with carp. Fishmeal, green leaf proteins and other binding agents like wheat are processed and mixed to create a modern fish substitute diet (Olabi *et al*., 2023). This study's findings indicate that including diverse plant foods and mustard oil into the diets of *Cyprinus carpio, Hypophthalmichthys molitrix,* and *Labeo rohita* markedly affected their proximate composition, encompassing moisture, crude protein, lipid, and ash levels. The fish's total nutritional quality was improved by the diets, which also demonstrated how different species responded to different dietary interventions. By clearly demonstrating how mustard oil and other plant-based compounds may be used to enhance the nutritional status and growth performance of key carp species, these findings meet the goals of the study. The research is valuable to aquaculture nutritionists and fish farmers seeking cost-effective, sustainable feed products that do not alter the proximate composition, or overall health, of cultured fish species.

Literature Review

According to Ahmadifar *et al.* (2021), polyphenols are natural plant compounds, and flavonoids are believed to fall under this category as well as phenolic acid, lignans, and stilbenes (C14H12). These bioactive compounds have antioxidant properties that can be of great importance to aquatic life when included in aquaculture diets. The study that was conducted by the author of the present paper has revealed countless benefits and usages of the given element in connection with aquatic life of almost all corners of the earth.

Colombo *et al.* (2023) performed a thorough analysis of alternative plant proteins in aquaculture fish diets. The authors analyzed multiple sources of plant protein, such as soybean, maize gluten meal, and pea protein concentrate, highlighting their nutritional characteristics and formulation challenges. The emphasis on managing digestibility and amino acid content in plant-based diets for fish is crucial for optimizing fish growth and feed efficiency. This study assessed the effectiveness of substituting fish meal with plant-based meals in formulated diets for Atlantic salmon (Colombo *et al*., 2023).

They studied digestible nutrients and plant protein kinds on salmon production. The study found that adding soybean and maize gluten meal in sufficient amounts improves Atlantic salmon growth and nutritional digestibility. The findings suggested aquafeed diets might use plant meal instead of fish meal (Patino, 2022).

Kuang *et al*. (2020) assert that common carp may optimize their biological characteristics when cultivated in rice fields. The ecological advantages of aquaculture and food safety have become key issues. Animal welfare significantly

influences the health of animals and the poultry industry, and it is also interconnected with human health and environmental well-being (Kuang *et al.*, 2020).

Hossain *et al*. (2024) assert that the quality and quantity of protein in fish meals are essential for promoting fish development and facilitating the rapid attainment of marketable size. Fish meal serves as a protein source in globally developed fish diets; nonetheless, it presents numerous significant issues, including escalating prices, inconsistency, adulteration, and variable quality. With the expansion of aquaculture, the demand for fish meal is increasing, the supply is inconsistent, and costs are escalating. This underscores the necessity of identifying alternative protein sources (Hossain, Small, Kumar, & Hardy, 2024).

Methods and Materials

In this study, healthy fingerling of Cyprinus carpio, Hypophthalmichthys molitrix, and Labeo rohita were captured at Ratta Kulachi Hatchery, where 20 individuals of each species were selected and put in three groups with 50 L of well-aerated water to facilitate favorable conditions of the experiment. The standard morphological keys and photographic records were used to identify and monitor species with diagnostic attributes of body shape, scale patterns, color, fin morphology and sexual dimorphism considered (Fritts *et al*., 2021), The fish was sun dried under controlled conditions of wind, humidity and temperature to reduce the moisture content and prevent the growth of microbes as per the established protocol (Nie *et al*., 2022). The experimental diet was prepared using 45 percent fish meal, 30 percent rice bran, 15 percent wheat, 10 percent mustard oil, and 2.5 percent mineral premix and fed to the test animals at 5 percent of body weight at a 60 day feeding test (Manuel, Gutierreez, & Naorbe, 2020). Four test aquaria (T1, T2, T3, and T4 as control) were used and water quality parameters, such as color, temperature, taste, odor, and turbidity, were observed according to the procedures presented by (Omer, 2019). When feeding trials were completed, fish samples were taken to the Nuclear Institute of Food and Agriculture (NIFA) where proximate composition analysis was done. The proximate analysis the estimation of moisture (AOAC 930.15), ash (AOAC 942.05), crude protein (Kjeldahl technique, AOAC 984.13), and crude fat (AOAC 920.39) using the AOAC (2003) standards as moisture and ash contents were determined by a gravimetric analysis; crude protein was calculated by dividing the nitrogen content (Nx6.25) by 4; fat content was extracted. Resultant mean values of each parameter were then estimated to accomplish comparative analysis of proximate body composition of the experimental fish species in the formulated diets.

Results and Findings

Proximate composition of various fish species and plant feed showed significant differences in nutrient contents. In *Cyprinus carpio* (control), the moisture was 70.32, ash 1.50, fiber 2.17, crude fat 5.0, crude protein 11.09 and nitrogen-free extract (NFE) 10.0 and energy 127.99 Kcal/100 g, compared to *C. carpio* fed on plant diet (slightly higher moisture 72.51), ash 3.0, fiber 3.17, crude fat 4.0, crude fat. *Labeo rohita* fed on plant feed had a moisture of 71.95, ash 1.95, fiber 2.01, fat 3.90, protein 13.0, NFE 7.12 and an energy of 125.32 Kcal/100g, whereas the control had 72.0% moisture, 1.41% ash, 3.03% fiber, higher fat (6.0) and protein 12.22, NFE 6.01. In *Hypophthalmichthys molitrix* fed on plant diet, the measured values were 71.0% moisture, 3.11% ash, 3.33% fiber, 4.54% fat, 16.0% protein, 7.0% NFE and 124.23 Kcal/100 g, as compared to the control group which had slightly lower moisture (69.50) but higher ash (4.0%), fiber (2.0), fat (5.09), protein (10 the plant feed sample recorded the lowest moisture (9.51 percent), but the highest ash (4.41 percent), fiber (8.30 percent), NFE (65.34 percent), and energy value (298.54 Kcal/100 g), fat 3.99 percent, and protein 8.34 percent.

Statistical comparison showed that *H. molitrix*, which was fed on plant diet, had the highest crude protein value (16.0%), and the lowest value of the protein was found in its control group (10.0%). *L. rohita* control fish had a maximum crude fat (6.0%), and the plant-fed *L. rohita* had the lowest fat (3.90%). Plant-fed *H. molitrix* contained the greatest amount of fiber (3.33%), and its control group contained the least (2.0%). Mostly the moisture was higher in plant-fed fishes, with *C. carpio* (72.51) having the highest percentage whereas *H. molitrix* control (69.50) had the lowest. The highest amount of ash was on *H. molitrix* control (4.0%), and the lowest on *L. rohita* control (1.41%). NFE values differed significantly with the highest in the case of plant feed (65.34) and the lowest in the case of *L.*

rohita control (6.01). Plant feed (298.54 Kcal/100 g), C. carpio control (127.99 Kcal/100 g) and *H. molitrix* control (114.40 Kcal/100 g) were also the highest and lowest energy values, respectively.

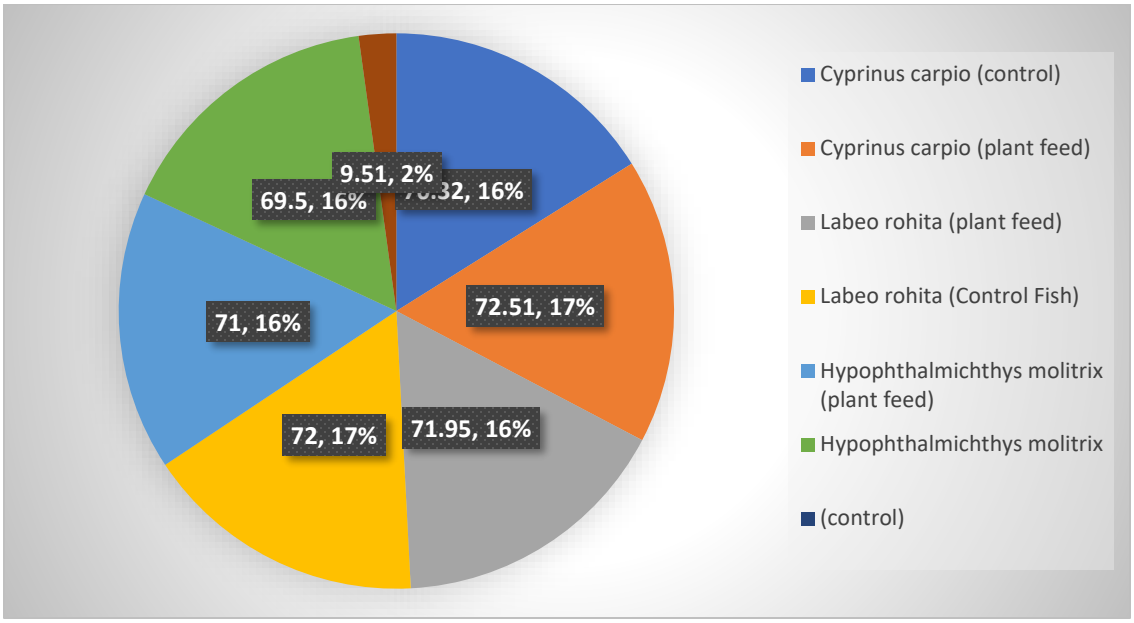
Table 1

Comparative Analysis of all Fish Nutrients

Samples	Moisture	Ash	Fiber	Crude Fat	Crude Protein	Nitrogen Free Extract (NFE)	Energy (Kcal/100 g)
Cyprinus carpio (control)	70.32	1.50	2.17	5.0	11.09	10.0	127.99
Cyprinus carpio (plant feed)	72.51	3.0	3.1	4.0	12.0	7.52	114.5
Labeo rohita (plant feed)	71.95	1.95	2.01	3.90	13.0	7.12	125.32
Labeo rohita (Control Fish)	72.0	1.41	3.03	6.0	12.22	6.01	119.03
Hypophthalmichthys molitrix (plant feed)	71.0	3.11	3.33	4.54	16.0	7.0	124.23
Hypophthalmichthys molitrix (control)	69.50	4.0	2.0	5.09	10.0	11.0	114.40
Plant Feed	09.51	4.41	8.30	3.99	8.34	65.34	298.54

Figure 1

Comparative Analysis of all Fish Nutrients



Discussion

The comparative analysis of nutrient composition reveals clear differences between control and plant-fed fish samples, reflecting the influence of diet on proximate composition. In Gulfam and Rohu fish, plant-based feed slightly increased moisture and ash content but reduced crude fat, crude protein, and overall energy levels compared to their respective controls, indicating a dilution effect or lower caloric density of plant feed. In Silver fish, however, plant feed markedly improved crude protein levels (15.02% compared to 9.39% in control), while maintaining moderate energy content, suggesting that this species may utilize plant-based nutrients more efficiently for protein synthesis. The nitrogen-free extract (NFE) levels fluctuated among species, reflecting differences in carbohydrate metabolism and feed assimilation. The plant feed itself, characterized by very low moisture but extremely high NFE and energy, appears nutrient-dense in carbohydrates but relatively low in crude protein compared to fish muscle tissue. Overall, our results point to the significance of species-specific feed formulation in aquaculture techniques by indicating that plant-based diets can affect body composition differently across species, decreasing fat and energy in some instances while boosting protein in others.

The kind and the quantity of food consumed by fish determines its growth and other related attributes. Unless we possess an appropriate array of familiar and extensively tried feed components, formulating and establishing quality and economically accountable feed is not only hard but unattainable. The main goal of these trials was to identify a variety of substances available in bulk on the market, aimed at providing convenience to farmers and manufacturers. These substances could be utilized effectively to improve fish health and boost fish production at a minimal cost (Iqbal & Iqbal, 2014). The Rohu species exhibits a remarkable response to the investigative diet concerning the protein content, measured at 12.60%. Conversely, the protein level of the silver carp shows a significant increase of 15.02, indicating that research food has a substantial effect on this species. Conversely, the Gulfam species exhibited a nutritional response that differed slightly from that of its counterparts, which had a significantly lower protein level of 11.75.

Conclusion

This comprehensive 60-day research supports the addition of the following ingredients to these three fish foods. Rohu (Labeo rorhita) and Silver carp (Hypophthalmichthys molitrix) were shown to provide beneficial outcomes when experimental food was utilized in place of fish food. These food components have been included into fish feed, according to the proximate analysis of fish and food. The patterns of development reveal how well the fish's diet was adjusted to promote health and growth. The complicated connections between food and Rohu and Silver Carp's biological processes necessitate a diet adapted to their dietary needs for rapid growth. This recommended swap of half of fish meal for plant meal fits Rohu and Silver Carp development needs and improves their growth and wellbeing. Knowing how food content and growth effect feed composition can help you correct Gulfam, Silver Carp, and Rohu feed recipes.

Recommendations

According to the findings of this research, it can be suggested that aquaculture players should pay attention to the inclusion of specific plant meals and mustard oil in carp feed to enhance the nutritional value and save money without impairing the condition of fish health or development. The next step in future studies is to maximize the inclusion of these ingredients, determine their long-term impacts on fish physiology, and determine how economical they can be when produced in commercial farming. Additionally, studies on digestibility, amino acid profiles, and the influence of plant-based diets on fish immune responses will further strengthen the development of sustainable and nutritionally balanced feed formulations for *Cyprinus carpio*, *Hypophthalmichthys molitrix*, and *Labeo rohita*.

Future Scope

To evaluate the growth performance, reproductive health, and immunological responses of fish exposed to different plant-based feed formulations, future research should focus on prolonged feeding studies. To enhance digestibility and nutritional digestion, research may also look at the use of natural supplements like probiotics or enzymes, as well as alternative plant components like oilseeds or legumes. Aquaculture operators will also be able to make informed decisions with the help of economic studies that examine the cost-effectiveness of plant-based diets in comparison to conventional foods. To understand more about the metabolic pathways that various animals use to consume plant-based nutrients, molecular or biochemical approaches may also be employed. Scientists would have a better grasp of how to provide sustainable food as a result.

Declarations

Ethical Approval and Consent to Participate: This study strictly adhered to the Declaration of Helsinki and relevant national and institutional ethical guidelines. Informed consent was not required, as secondary data available on websites was obtained for analysis. All procedures performed in this study were by the ethical standards of the Helsinki Declaration.

Consent for Publication: Here, we, the authors, give our consent for publication.

Availability of Data and Materials: Data will be provided upon written request from the corresponding author.

Competing Interest: The writers of this article affirm that they are free from any conflicts of interest, whether financial or otherwise, that may have affected their independence in completing this work.

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