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Research Paper

Nutritional efficacy of processed pearl millet (*Pennisetum glaucum*) in practical feeds for fingerling *Labeo rohita*

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Abstract

2023 has been recognized as the "International Year of Millets" by The United Nations General Assembly in acknowledgement of the importance of millets. This global initiative enhanced to increase the awareness about the nutritional benefits of millets and promote the efforts for enhancement of their production and utilization. Millets are known to get through harsh climatic circumstances and are extremely tolerant to drought. Pearl Millet (*Pennisetum glaucum*), the largest produced type of millet, is a robust cereal grain that can grow in harsh conditions. It is an excellent source of energy and rich in proteins, fiber, and minerals like iron and zinc. It also contains phytochemicals that have antioxidant properties. The experiment was conducted for 8 weeks. Experimental diets were formulated by incorporating germinated and fermented pearl millet flour at the place of wheat flour in the control diet at 7 different levels i.e., D0 (control, i.e., without pearl millet), D1 (5% dietary level), D2 (10%), D3 (15%), D4 (20%), D5 (25%), and D6 (30%). The fish were fed to apparent satiation twice in a day. *Labeo rohita*, showed improve growth performance and conversion efficiencies on the addition of pearl millet in the diet. The pearl millet incorporated feed did not show any detrimental effects on the fish survival, growth and other parameters. The highest weight gain was noticed in fish fed diet D4 at 20% inclusion of germinated and fermented pearl millet, and the lowest was in D0. The lowest FCR was in fish fed D4 diet and the highest in D0. Incorporation of processed *P. glaucum* in the diets also showed improved results in terms of antioxidant and hematological parameters. The results of the experiment demonstrated that adding pearl millet to fish diets at the rate of 20% had a positive impact on growth, haematology and antioxidant status of *L. rohita* fingerlings without compromising the health status of fish.

1. Introduction

Aquaculture is growing swiftly as a food production sector and plays crucial role ensuring global food security as well as economic development. By 2022, worldwide aquaculture production had exceeded 94 million tonnes and freshwater finfish holds the most prominent fraction (Dongyu, 2024). Current food production practices require significant changes to feed 9.8 billion people as well as minimize environmental degradation by 2025 (Oluwole et al, 2024). The ocean and associated food systems have potential to produce over six times more food than the present output. Fish and seafood contribute a crucial part in nutritional well-being and food security by supplying essential macro as well as micronutrients (SOFIA, 2022). Furthermore, worldwide fish consumption on a per capita basis has increase by twofold in past five decades (Naylor et al., 2021).

Aquaculture is one of the fast-growing food production sectors which plays vital role in meeting the growing global requirement of fish as food (SOFIA, 2022).

Feed is a major factor in determining the success of the aquaculture business, as it constitutes about 60%-70% of the operational costs. With the intensification of aquaculture, there is a growing need to develop nutrient-rich and economical feed formulations based on cost-effective, locally available feed ingredients for cultivable fish species. Also, with increasing climate change, there is a need to employ climate-smart plant-based ingredients in aquafeed. Millets are one such ingredient that offers climate resilience, efficient water usage, and nutritional benefits, making them a sustainable choice in the face of climate challenges. Millets are majorly used in livestock feed, where it has proven to be a valuable ingredient in the diet of animals like broilers, cattle, etc. These are important foods in many developing countries as the have the ability to grow under challenging climatic conditions. Millets can be considered as a substitute to traditional cereal grains due to their high protein, calcium, bioactive compounds, starch and fibre content (Banik et al., 2025). *Pennisetum glaucum*, commonly known as pearl millet, is a whole grain abundant in protein, dietary fiber, minerals, and bioactive compounds (Dias-Martins

et al., 2018; Sharma & Gujral, 2019). It is known that the germination process of the grain can increase the content of these compounds (Owheruo et al., 2019, Sarita, 2016). Processes of germination and/or fermentation employed to numerous food grains have been known to reduce the antinutritional factors, thereby improving their nutritional profile (Anaemene and Fadupin, 2022; Bilal et al., 2026).

Carp species dominate the Indian freshwater aquaculture (Rasal and Sundaray, 2020). Indian major carps (IMCs) constituting three species viz *Catla catla*, *L. rohita*, and *Cirrhinus mrigala*, are the most culturable fish species in India (Surasani et al., 2022). They contribute more than 70% of freshwater production in India (Das et al., 2022). High growth rate and good taste of rohu makes it most preferred among all species (FAO, 2022). It contributes nearly 35% of total Indian farmed fish production (Mir et al., 2017). Under suitable conditions it has potential to attain up to 1 kg within one year. Rasal and Sundaray (2020) reported that rohu is also a widely cultured species in Bangladesh, Myanmar, and Pakistan. It has high fecundity rate (approximately 2 lakh eggs per kg) and exihibit external fertilization which make it easy to breed and secure the required number of seeds for the intensive aquaculture (Rasal and Sundaray, 2020). Demand for low-cost and nutritionally balanced aquaculture feeds is also increasing with the increase in market demand of this fish (Modeel et al. 2023). Hence, present study was conducted to evaluate the potency of pearl millet as an innovative ingredient for fees of *L. rohita*.

2. Materials and Methods

2.1 Germinated and fermented millet flour

Pearl millet grains used for the study were obtained from the local market. The grains were washed and soaked in distilled water for 12 hr at 30°C then the soaked grains were sprouted in petri-dishes lined with wet filter paper at 30°C for 24 hr using an incubator (Khetarpal and Chauhan, 1990). Natural fermentation of millet was carried out by mixing sprouted millet with distilled water at (1:2 w/v) ratio followed by homogenized to make a slurry. This slurry was incubated at a temperature of approximately 30^{O}C for 32 h. Samples were collected and transferred to aluminum dishes, and dried in a hot air oven at 70^{O}C for 3-4 h. Dried samples were subsequently ground and stored at 4^{O}C until further analysis (Elyas et al., 2002). All samples were analyzed for proximate composition (Table 1) following standard methods (AOAC, 2005).

2.2 Experimental Diets

Seven isonitrogenous (35% crude protein) and isocaloric (16.7 kJ/g gross energy) experimental diets with graded levels of germinated and fermented pearl millet flour (0, 5, 10, 15, 20, 25 and 30%) were formulated as given in Table 1. Briefly, all the ingredients were grinded and weighed carefully as per the dietary formulations. All the weighed ingredients were blended thoroughly in a Hobart electric mixer and then cooked at 80^{o}C. After that oil was added. Vitamin and mineral premixes were added after cooling the mixture. The dough was passed through a pelletizer fitted with a 2-mm die to obtain pellets, which were dried in a hot air oven at 40^{o}C to reduce the moisture content below 10%. The dried pellets thus obtained were processed to crumbles, sieved (0.20–0.25 mm), and stored at 4^{o}C till further use.

2.3 Experimental Design and Feeding Trial

Induced bred *L. rohita* were obtained from G. B. Pant University of Agriculture and Technology, Pantnagar. The fish were then transported to the wet laboratory, given a prophylactic dip in $KMnO_4$ solution (1:3000) before stocking in indoor circular aqua-blue colored, plastic-lined fish tanks (1.22 × 0.91 × 0.91 m; water volume 600 L) for 2 weeks. During this period, the fish were acclimated on control diet and reared to fingerling stage. Fingerling *L. rohita* (2.75 ± 0.05 cm) were taken from the above acclimated fish lot and stocked in triplicate groups at the rate of 20 fish per trough in 70-L circular polyvinyl troughs (water volume 55 L) fitted with a continuous water flow-through (1-1.5 L/min) system. Fish were fed test diets to apparent satiation in the form of dry crumbles (0.20-0.25 mm) twice daily. Initial and weekly weights were recorded on a top-loading balance (Precisa 120A; 0.1 mg sensitivity). Fish were deprived of feed on the day the measurements were taken. The feeding trial lasted for eight weeks. The uneaten feed was collected after each feeding and oven-dried (100°C) to calculate the feed conversion ratio (FCR). Standard methods (APHA 1992) for water quality analyses were followed. The average water temperature, dissolved oxygen, free carbon dioxide, pH, total ammonia nitrogen, nitrites, and total alkalinity, based on daily measurements, were 26.8-28.4^{O}C, 6.9-7.1 mg/L, 5.7-9.1 mg/L, 7.1-7.8, 0.29-0.33 mg/L, 0.03-0.07 mg/L, and 64.9-81.5 mg/L, respectively.

2.4 Chemical Analyses

Standard methods (AOAC, 2005) were used for proximate composition analyses of experimental diets, and initial as well as final whole-body carcass. At the beginning of the feeding trial, 10 fish were randomly sampled, euthanized in MS-222 (200 µg/mL) and pooled together. Six subsamples of a pooled sample were analyzed for initial whole-body carcass composition. At the end of the experiment, 12 fish from each replicate of dietary treatments were randomly euthanized in MS-222 (200 µg/mL) and pooled separately. Three subsamples of the pooled samples were analysed for final biochemical composition. Six another fish was randomly selected from each replicate to collect the blood and liver samples.

2.5 Blood collection

Blood samples were collected from caudal vein using a sterile syringe rinsed with ethylenediaminetetraacetic

acid (EDTA; 5mg/L) solution as an anticoagulant. Blood samples were collected from each replicate, pooled and used for haematological analysis.

2.6 Haematological analysis

Haemogloboin (Hb) was quantified colorimetrically following the method of Wong (1928). Red blood cells (RBCs) were counted under light microscope (Nikon, Japan, E-600) using Neubauer haemocytometer with Yokoyama's (1947) solution as the diluting medium. Haematocrit value (Hct) was measured by drawing fresh blood into micro-haematocrit tubes and centrifuge at 3600 rpm for about 5 min (RM 12C, Micro Centrifuge, Remi, Remi Motors, India).

2.7 Antioxidant analysis:

Superoxide dismutase (SOD) activity was determined using the method of Misra and Fridovich (1972). A tissue homogenate (0.1 mL) was added to tubes containing 0.75 mL ethanol and 0.15 mL chloroform and centrifuged at 748×g for 15 min at 4°C. To 0.5 mL of supernatant, 0.5 mL of 0.6 mM EDTA solution and 1 mL of 0.1 M carbonate-bicarbonate (pH 10.2) buffer was added. The reaction was initiated by the addition of 0.5 mL of 1.8 mM freshly prepared epinephrine and the increase in absorbance was measured at 480 nm using a spectrophotometer. SOD activity was expressed in units per milligram of protein (U/mg protein). One unit of the SOD activity was the amount of protein required to give 50% inhibition of epinephrine auto-oxidation.

Catalase activity was measured according to the method of Aebi (1984). One unit of catalase is defined as the amount of enzyme required to decompose 1.0 µM of hydrogen peroxide (H_2O_2) in 1 minute. The reaction was initiated by the addition of freshly prepared 1mL of 20 mM H_2O_2. The rate of decomposition of H_2O_2 was measured at 240 nm for 1 minute at 25°C. The enzyme activity was expressed as U/mg of protein.

Determination of ***lipid peroxidation*** in tissue is based on the estimation of the thiobarbituric acid reactive substances (TBARS) which is measured in the form of malondialdehyde (MDA) content. Lipid peroxidation was determined using the procedure of Utley et al. (1967), with some modifications as adopted by Fatima et al. (2000). The liver tissue was homogenized in a chilled 0.1 M KCl solution. The assay mixture contained 0.67% TBA, 10% chilled TCA and homogenate (10%) in a total volume of 3 mL. The rate of lipid peroxidation was expressed as nano-moles of MDA/g liver tissue using a molar extinction coefficient of 1.56×10^5/M/cm.

2.8 Growth Performance

Growth performance of the experimental diets was measured as a function of the weight gain by calculating the following parameters:

Absolute weight gain (g/fish) = Final body weight-Initial body weight

Protein efficiency ratio = Weight gain (g) /Protein intake (g)

Feed intake (g/fish) = Total dry feed intake (g) /Total no. of fish

Feed conversion ratio =Dry feed intake (g) /Wet weight gain (g)

Protein deposition = Protein gain (g) /Protein intake (g)

2.9 Statistical Analyses

Growth data were subjected to one-way analysis of variance (Sokal & Rohlf, 1981). Treatment means were subsequently compared by Tukey's post hoc test with a statistical significance level of $P<0.05$. Prior to analysis, data were assessed for normality as well as homogeneity using Shapiro -Wilk test and Levene's test, respectively. All the statistical analyses were conducted using Origin (version 6.1; Origin Software, San Clemente, CA, USA).

3. Results

Fish receiving diets with graded levels of pearl millet demonstrated improved growth performance and conversion efficiencies. Absolute weight gain (AWG), specific growth rate (SGR) and feed conversion ratio (FCR) were improved significantly ($P<0.05$) with increasing amount of pearl millet in the diets from D1 to D4 and then levelled off on more inclusion of pearl millet in the diets (Table 2). Antioxidant status was also improved significantly ($P<0.05$) with the inclusion of pearl millet in the diets, whereas lipid peroxidation was noted to decrease ($P<0.05$) with the incremental level of pearl millet in the diets from D1 to D4 whereas no significant ($P>0.05$) effect was noted on further inclusion of pearl millet in the diets. Significant changes ($P<0.05$) were also recorded in the hematological parameters of the fish fed experimental diets with increasing levels of pearl millet from D0 to D4 and remained static after that on further inclusion of pearl millet in the diets (Table 3 & 4). The findings of the present study indicate that the use incorporation of pearl millet in fish feeds showed a positive impact on the growth, feed conversion efficiency, antioxidant status and hematology of L. rohita fingerlings.

4. Discussion

Although wheat production has enhanced subsequently over the recent decades, its future production is expected to be challenging due to global climate change, ever-increasing world population, and water scarcity in arid and semi-arid regions (O'Leary et al., 2018). Wheat flour and wheat bran are common ingredients used in aquafeeds for their high energy and fiber content.

Table 1: Formulation and proximate composition of experimental diets

Ingredients (g/100g)	D0 (0%)	D1 (5%)	D2 (10%)	D3 (15%)	D4 (20%)	D5 (25%)	D6 (30%)
Fish Meal	10	10	10	10	10	10	10
Soybean Meal	15	15	15	15	15	15	15
Groundnut Oil Cake	40	40	40	40	40	40	40
Wheat Middlings	30	25	20	15	10	5	0
Oil	3	3	3	3	3	3	3
Pearl Millet[1]	0	5	10	15	20	25	30
Vitamin+ Mineral mix[2,3]	1	1	1	1	1	1	1
Chromium oxide	1	1	1	1	1	1	1
Total	100	100	100	100	100	100	100
Proximate analyses							
Analyzed crude protein (%)	34.81	34.64	34.59	34.92	34.85	35.02	35.18
Estimated gross energy (kJ/g)	16.83	17.77	17.92	17.85	17.81	17.68	18.05

[1]Germinated and fermented [2]Mineral mixture (g/kg of mineral premix): calcium biphosphate.H_2O 135.7; calcium lactate 326.9; ferric citrate 29.7; potassium phosphate (dibasic) 239.8; sodium phosphate.$2H_2O$ 87.2; magnesium sulphate.$7H_2O$ 132.0; sodium chloride 43.5; aluminium chloride.$6H_2O$ 0.15; potassium iodide 0.15; cuprous chloride 0.10; manganous sulphate.$6H_2O$ 0.80; cobalt chloride. $6H_2O$ 1.0; zinc sulfate.$7H_2O$ 3.0; α-cellulose 462.6; Loba Chemie, Mumbai, India; [5]Halver (2002); [3]Vitamin mixture (30 g/kg of diet; 10 g vitamin mix + 20 g α-cellulose) choline chloride 5.0; meso-inositol 2.0; L-ascorbyl-2-polyphosphate 1.0; nicotinic acid 0.75; calcium D-pantothenate 0.5; riboflavin 0.2; menadione 0.04; pyridoxine hydrochloride 0.05; thiamine hydrochloride 0.05; folic acid 0.015; biotin 0.005; alpha-tocopheryl acetate 0.40; vitamin B_{12} (cyanocobalamin) 0.0001; Loba Chemie, India

Table 2: Effects of diets containing graded levels of *Pennisetum glaucum* on growth performance and conversion efficiencies of fingerling *Labeo rohita*

Growth Parameters	D0 (0%)	D1 (5%)	D2 (10%)	D3 (15%)	D4 (20%)	D5 (25%)	D6 (30%)
Average initial weight (g/fish)	2.78±0.04	2.72±0.03	2.77±0.05	2.74±0.04	2.79±0.05	2.71±0.06	2.76±0.03
Average final weight (g/fish)	8.73±1.43^{e}	10.26±1.61^{d}	12.28±2.19^{c}	13.83±2.71^{b,c}	18.86±3.17^{a}	18.64±3.28^{a}	19.12±2.43^{a}
Absolute weight gain (g/fish)	5.95±2.13^{e}	7.54±2.32^{d}	9.51±1.98^{c}	11.09±2.46^{b}	16.07±2.68^{a}	15.93±3.11^{a}	16.36±2.93^{a}
Specific growth rate (%/day)	2.04±0.24^{e}	2.37±0.17^{d}	2.66±0.19^{c}	2.89±0.21^{b}	3.41±0.18^{a}	3.44±0.11^{a}	3.46±0.15^{a}
Feed conversion ratio	2.97±0.45^{a}	2.91±0.68^{a,b}	2.35±0.73^{c}	2.18±0.59^{c,d}	1.67±0.48^{e}	1.63±0.35^{e}	1.62±0.32^{e}
Protein retention efficiency (%)	11.41±0.93^{d,e}	12.87±1.86^{d}	16.44±1.69^{c}	19.66±1.48^{b}	28.41±1.53^{a}	30.89±1.79^{a}	30.43±2.14^{a}
Protein gain (g/fish)	0.71±0.36^{d}	0.99±0.54^{c,d}	1.29±0.63^{c}	1.66±0.92^{b}	2.67±1.03^{a}	2.81±1.04^{a}	2.82±1.12^{a}

Mean values of 3 replicates±SEM; [2]Mean values sharing the same superscripts in the same row are insignificantly different (P>0.05).

Table 3: Effects of diets containing graded levels of *Pennisetum glaucum* on hepatic antioxidant status fingerling *Labeo rohita*

	D0 (0%)	D1 (5%)	D2 (10%)	D3 (15%)	D4 (20%)	D5 (25%)	D6 (30%)
SOD (U/mg protein)	44.83±1.27^{e}	48.42±1.19^{d}	50.79±1.37^{c}	52.17±1.25^{b}	57.25±1.24^{a}	58.32±1.67^{a}	57.84±1.84^{a}
CAT (U/mg protein)	16.23±0.29^{a}	17.27±0.38^{a}	18.65±0.51^{a}	20.55±0.65^{a}	23.87±0.61^{a}	24.37±0.68^{a,b}	24.82±0.71^{b}
MDA (nmole/g liver tissue)	19.87±0.21^{a}	19.12±0.17^{b}	17.46±0.12^{c}	15.02±0.15^{d}	11.78±0.14^{e}	11.34±0.16^{e}	11.28±0.12^{e}

Mean values of 3 replicates±SEM; [2]Mean values sharing the same superscripts in the same row are insignificantly different (P>0.05).

Table 4: Effects of diets containing graded levels of *Pennisetum glaucum* on haematological parameters of fingerling *Labeo rohita*

Haematological parameters	D0 (0%)	D1 (5%)	D2 (10%)	D3 (15%)	D4 (20%)	D5 (25%)	D6 (30%)
Haemoglobin (g/dL)	4.28±0.46^{e}	5.14±0.52^{d}	6.64±0.48^{c}	7.48±0.63^{b}	8.83±0.67^{a}	8.75±0.58^{a}	8.86±0.71^{a}
RBCs (10^6/mm^3)	1.32±0.09^{e}	1.48±0.12^{d}	1.78±0.15^{c}	2.01±0.23^{b}	2.28±0.32^{a}	2.27±0.31^{a}	2.31±0.29^{a}
Haematocrit (%)	16.54±0.93^{e}	18.43±0.81^{d}	21.11±0.96^{b,c}	22.56±1.04^{b}	28.73±1.10^{a}	27.68±1.02^{a}	28.17±1.08^{a}

Mean values of 3 replicates±SEM; [2]Mean values sharing the same superscripts in the same row are insignificantly different (P>0.05).

As a consequence of lowered wheat production, the aquafeed industry would face a scarcity of quality energy sources. Successful use of substitutes of traditional ingredients which are economically and environmentally feasible, even in small proportions, is desirable as it can contribute in reducing the feed cost as well as overall production cost (Amaya et al., 2007). Plant sources are the most appropriate alternatives among the many carbohydrate/energy sources available for animal feeds. The inclusion of millets in fish diets has been accomplished in many cultured fish (Boscolo et al., 2010; Olaniyi & Falaye, 2013; Taiwo et al., 2017; Bulani & Sharma, 2023; Nigam et al., 2023; Sharma et al., 2023). Among the plant energy sources, cereals are the most promising species. However, the use of cereals in aquafeeds is limited due to the presence of a variety of anti-nutritional factors and low bioavailability of certain nutrients (El Mekawy et al., 2013). High nutritive value of cereals such as their protein and energy content make them highly valuable component of feeds for ruminants and monogastric animals. Amongst the cereals, millets have been extensively used as a chief energy providing source in feeds of aquaculture and poultry (Hidalgo et al., 2004; Gulia et al., 2007; Khalil et al., 2023). However, presence of antinutritional factors limits their use. Several processing methods including soaking, boiling, fermentation and germination have been used widely to reduce the level of antinutrional factors, thereby improving the nutritional quality of complementary foods (Agostini et al., 2010). Using combination instead of single process can be more effective in eliminating antinutrients (Inyang and Zakari, 2008). During fermentation and germination, phytates are degraded by activated phytases (Temesgen, 2013).

Germination in millets activates enzymes such as α-amylase, protease, and lipase. These enzymes help in breaking down complex molecules like starch, proteins, and lipids into simpler forms (Azeez et al., 2022). During germination, nutrients stored in the seed, such as carbohydrates, proteins, and fats, become more readily available. This enhances the nutritional value of the pearl millet (Singh et al., 2015). Germination process substantially reduces the phytic acid and tannins (antinutrients) which are known to show inhibitory effects on availability and absorption of the nutrients. This process makes the nutrients in pearl millet more bioavailable and digestible (Bheemaiah Balyatanda et al, 2024.). Germination can increase the content of indispensable amino acids and improve the overall protein quality of pearl millet. This is particularly beneficial for human and animal consumption (Rahman et al, 2023). Activities of phenolic compounds and

flavonoids which are helpful in reducing the oxidative stress and enhancing the disease resistance are increased by germination process. (Yang et al., 2025). Germination significantly enhances the nutritional quality and biological activity of pearl millet, making it more suitable for consumption and improving its potential as a feed crop. Malting has been reported to enhance the nutritional quality of grains in terms of digestibility, energy density, vitamin and protein content (Preetika, et al., 2004). During germination process, amylase acts on starch molecules and results into its degradation to oligosaccharide and disaccharide. Germinated flour has lower water holding capacity and high energy density which makes it more suitable to be used in infant foods, weaning foods and enteral foods. Similarly, malted millet flour can also be incorporated in the manufacturing of various food items (Shobana et al., 2013). Process of germination has been reported to activate the enzyme which results into the disintegration of carbohydrates, proteins and lipids into simple sugars, amino acids and fatty acids, respectively.

Fermentation causes the total polyphenols to decrease. Activates polyphenol oxidase may be the reason for reduction of polyphenols (Dhankher and Chauhan, 1987). Fermentation can extend the shelf life of flour by converting sugars into lactic acid, which creates an acidic environment by lowering pH. Thus inhibits the growth of harmful bacteria. Fermentation at 28°C for three days causes reduction in the bulk density of pearl millet flour (Chinma et al., 2021). Phytochemical leaching results in improved lightness in pearl millet flour due to fermentation. It also reduces the concentration of antinutrients such as tannins, phytates, and trypsin inhibitors boosting nutritional value. Fermentation produces several benefits, including improved flavors, colors, shelf-life, and functional properties, as well as improved acceptance of products. In another study conducted by Tewe et al. (1999) fermentation resulted into significantly reduced phytate level in millet thereby increasing the bioavailability of minerals. It is also consistent with the results documented by Abdallah et al. (1998) on phytate and mineral content of pearl millet.

Growth reflects the metabolic interactions and adjustments sustained by the nutritional status. Optimum growth rate and feed conversion efficiency were exhibited by *L. rohita* fingerlings fed 20% pearl millet diets. This can be attributed to the fact that at this level, the fermented millet was best utilized by the fish in order to enhance weight gain hence they attained the highest specific growth rate recorded among all the groups receiving experimental diets. Channel catfish showed equivalent weight gain and feed efficiency when either maize or peal millet was fed at 30% of total diet. In this experiment, the inclusion of pearl millet in the feed of *L. rohita* at the expense of wheat middings was found to be feasible up to 20%. Burtle and Newton (1995) reported equivalent weight gain and feed efficiency in channel catfish when either maize or pearl millet was fed at 30% of the total diet. The adverse effects of incorporating plant-based feed ingredients at higher levels in the diets have been extensively reported on several fish species (Walker et al., 2010; Gomes et al., 2004; Wang et al, 2012; Torstensen et al., 2008; Sanz et al., 1994; Yousif et al., 2019; Barnes et al., 2014; Refstie et al., 2001; Opstvedt et al., 2003; Mundheim et al., 2004).

Hematological parameters can be influenced by several factors such as diet composition, environmental conditions, and physiological status. In the present study, inclusion of pearl millet in the diets of *L. rohita* significantly affected hematological parameters. The improved values of these parameters with increasing levels of pearl millet in the diet can be associated with the balanced nutritional profile of pearl millet. Improved Hb content with increasing amount of pearl millet in the diets can be associated with the high iron content of this ingredient as reported by researchers (Patni and Agarwal, 2017; Aher et al., 2025). There are several reports available in which hematological parameters reflected association with the use of millets in the diets.

Oxidative stress is caused due to the unevenness between the manufacture and elimination of reactive oxygen species (ROS), which can change the structure and the function of cellular components (Barbacariu et al., 2024). Organisms including fish own an antioxidant defense system that constitutes both enzymatic and non-enzymatic components and that can also be obtained from diet. In order to avoid excessive levels of oxidative damage due to increase in reactive oxygen species production, organisms can increase their synthesis of endogenous antioxidant enzymes to combat the action of reactive oxygen species production or it can be boosted by the dietary components (Wesołowski et al., 2023). Fish with promising antioxidant capacities demonstrate lower oxidative stress, improved immune response, and high adaptability to climatic changes (Birnie-Gauvin et al., 2017). In the present study, the inclusion of pearl millet in the diets of *L. rohita* reflected improved activities of antioxidant enzymes such as SOD and CAT. Malondialdehyde is the final product of lipid peroxidation and serves as a marker for oxidative stress (Cordiano et al., 2023). MDA level of *L. rohita* was found to decrease with increasing dietary pearl millet up to the 20 % inclusion, suggesting that pearl millet may improve the antioxidant capacity of this fish. Previous research on grey mullet and common carp have also demonstrated the antioxidant effects of millets in the diet of fish due to the presence of phenolic compounds in the ingredients.

5. Conclusion

Findings of this study indicate that adding processed pearl millet to fish diets at a rate of 20% had a positive impact on growth, antioxidant status and hematology of *L. rohita* fingerlings. Thereby, facilitating the development of cost-effective artificial feeds for the intensive culture of this fish.

6. Statements and Declarations

Ethical Approval: Not Applicable

Conflict of Interest: The authors declare that they have no conflict of interest.

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References

Abdalla, A. A., El Tinay, A. H., Mohamed, B. E., & Abdalla, A. H. (1998). Effect of traditional processes on phytate and mineral content of pearl millet. *Food chemistry*, *63*(1), 79-84.

Aebi, H. (1984). Catalase in vitro. *Methods Enzymol* 105, 121-126.

Agostini, JDS; Nogueira, RB; Ida, EI (2010). Lowering of phytic acid content by enhancement of phytase and acid phosphatase activities during sunflower germination. *Braz. Arch. Bio. Technol.* 53:975-980.

Aher, R. R., Bhunia, R. K., Kaur, S., Palakolanu, S. R., Taleon, V., Sharma, K. K., ... & Bhatnagar-Mathur, P. (2025). Exploring rancidity in pearl millet flour: A lipidomic and biochemical approach. *Journal of Cereal Science*, 104320.

Amaya, E. A., Davis, D. A., & Rouse, D. B. (2007). Replacement of fish meal in practical diets for the Pacific white shrimp (*Litopenaeus vannamei*) reared under pond conditions. *Aquaculture*, *262*(2-4), 393-401.

American Public Health Association. (1992). *Standard Methods for the Examination of Water and Wastewater* (18th ed.). Washington DC: American Public Health Association.

Anaemene, D., & Fadupin, G. (2022). Anti-nutrient reduction and nutrient retention capacity of fermentation, germination and combined germination-fermentation in legume processing. *Applied Food Research*, *2*(1), 100059.

AOAC. (2005). Official methods of analysis of AOAC International; Horwitz W, eds. Gaithersburg, MD, Washington, DC, USA: AOAC International.

Azeez, S. O., Chinma, C. E., Bassey, S. O., Eze, U. R., Makinde, A. F., Sakariyah, A. A., ... & Adebo, O. A. (2022). Impact of germination alone or in combination with solid-state fermentation on the physicochemical, antioxidant, in vitro digestibility, functional and thermal properties of brown finger millet flours. *Lwt*, *154*, 112734.

Banik, A., Kumar, S., Dubey, M. K., Brahmchari, R. K., Nayak, S. K., & Srivastava, P. P. (2025). Foxtail millet bran as a functional feed ingredient: Effects on growth, feed efficiency, and nutrient digestibility in grass carp (*Ctenopharyngodon idella*). *Animal Feed Science and Technology*, 116563.

Barbacariu, C. A., Dumitru, G., Rimbu, C. M., Horhogea, C. E., Dîrvariu, L., Todirașcu-Ciornea, E., ... & Burducea, M. (2024). Inclusion of Sorghum in *Cyprinus carpio* L. Diet: Effects on Growth, Flesh Quality, Microbiota, and Oxidative Status. *Animals*, *14*(11), 1549.

Bheemaiah Balyatanda, S., Gowda, N. N., Subbiah, J., Chakraborty, S., Prasad, P. V., & Siliveru, K. (2024). Physiochemical, bio, thermal, and non-thermal processing of major and minor millets: A comprehensive review on antinutritional and antioxidant properties. *Foods*, *13*(22), 3684.

Bilal, M., Abideen, Z. U., Ankutse, P., Sattar, D. E. S., & Ramadan, M. F. (2026). Advancement in legume processing: exploring germination and fermentation techniques for enhanced nutritional and functional properties. *Journal of Food Measurement and Characterization*, 1-25.

Burtle G.J. and Newton G.L. (1995). Catfish performance on pearl millet grain. Proceedings of 1st National grain pearl millet Symposium Univ. Georgia, Tiffon. In I.D Teare(ed.) 116-118.

Chinma, C.E., Abu, J.O., Asikwe, B.N., Sunday, T., & Adebo, O.A. (2021). Effect of germination on the physicochemical, nutritional, functional, thermal properties and in vitro digestibility of Bambara groundnut flours. *Lwt*, *140*, 110749.

Cordiano, R., Di Gioacchino, M., Mangifesta, R., Panzera, C., Gangemi, S., & Minciullo, P. L. (2023). Malondialdehyde as a potential oxidative stress marker for allergy-oriented diseases: an update. *Molecules*, *28*(16), 5979.

Das, B. K., Parida, P. K., DebRoy, P., & Roy, A. (2022). A situational analysis of small-scale inland open water fisheries in India: from vulnerability to viability. *Work Paper V2V*, *2028*.

Degani, G. and Viola, S. (1987). The protein sparing effect of carbohydrate sources in the diets of eels, *Anguilla anguilla*. Aquaculture 64, 283–291.

Dhankher, N., & Chauhan, B. M. (1987). Effect of temperature and fermentation time on phytic acid and polyphenol content of rabadi—a fermented pearl millet food. *Journal of Food Science*, *52*(3), 828-829.

Dias-Martins, A.M.; Pessanha, K.L.F.; Pacheco, S.; Rodrigues, J.A.S.; Carvalho, C.W.P. Potential Use of Pearl Millet (*Pennisetum glaucum* (L.) R. Br.) in Brazil: Food Security, Processing, Health Benefits and Nutritional Products. Food Res. Int. 2018, 109, 175–186.

Dongyu, Q. (2024). 2024 the state of world fisheries and aquaculture-blue transformation in action. *The State of World Fisheries and Aquaculture*, R1-232.

El Mekawy, A., Diels, L., De Wever, H., & Pant, D. (2013). Valorization of cereal based biorefinery byproducts: reality and expectations. *Environmental science & technology*, *47*(16), 9014–9027. https://doi.org/10.1021/es402395g

Elyas, S. H., El Tinay, A. H., Yousif, N. E., & Elsheikh, E. A. (2002). Effect of natural fermentation on nutritive value and in vitro protein digestibility of pearl millet. *Food chemistry*, *78*(1), 75-79.

Falaye, A.E., Jauncey, K. and Tewe, O.O. (1999). The growth performance of Tilapia (*Oreochromis niloticus*) fingerlings fed varying levels of cocoa Husk Diets J. Aqua. Trop. 14 (1).

FAO, F. (2022). The state of world fisheries and aquaculture 2022. Towards blue transformation.

Gulia, S.K., Wilson, J.P., Carter, J. and Singh, B.P. Progress in grain pearl millet research and market development Issues in new crops and new uses 2007, 196-203.

Hidalgo, M. A., Davis, A. J., Dale, N. M., & Dozier III, W. A. (2004). Use of whole pearl millet in broiler diets. *Journal of Applied Poultry Research*, *13*(2), 229-234.

Inyang, C. U., & Zakari, U. M. (2008). Effect of Germination and fermentation of Pearl millet on proximate, chemical

and sensory properties of instant" Fura"-a Nigerian cereal food. *Pakistan journal of Nutrition*, *7*(1), 9-12.

Khalil, M. A., Tarsha, H. A., & Kussaibati, R. J. (2023). Effects of replacing maize by proso millet on performance of broiler chickens. *Journal of World's Poultry Research*, *13*(2), 261-267.

Khetarpaul, N., & Chauhan, B. M. (1990). Fermentation of pearl millet flour with yeasts and lactobacilli: in vitro digestibility and utilisation of fermented flour for weaning mixtures. *Plant Foods for Human Nutrition*, *40*(3), 167-173.

Lim C., Dominy W. (1990). Evaluation soybean meal as a replacement for marine animal protein in diet for shrimps (*Penaeus vannamei*). Aquaculture 86:401-407.

Mir, I. N., Sahu, N. P., Pal, A. K., & Makesh, M. (2017). Synergistic effect of l-methionine and fucoidan rich extract in eliciting growth and non-specific immune response of *Labeo rohita* fingerlings against *Aeromonas hydrophila*. *Aquaculture*, *479*, 396-403.

Misra, H., Fridovich, I. (1972). The role of superoxide anion in the autooxidation of epinephrine and a simple assay for superoxide dismutase. *J Biol Chem* 247, 3170.

Modeel, S., Joshi, B. D., Yadav, S., Bharti, M., & Negi, R. K. (2023). Mitochondrial DNA reveals shallow population genetic structure in economically important Cyprinid fish *Labeo rohita* (Hamilton, 1822) from South and Southeast Asia. *Molecular Biology Reports*, *50*(6), 4759-4767.

Naylor, R. L., Hardy, R. W., Buschmann, A. H., Bush, S. R., Cao, L., Klinger, D. H., ... & Troell, M. (2021). A 20-year retrospective review of global aquaculture. *Nature*, *591*(7851), 551-563.

O'Leary GJ, Aggarwal PK, Calderini DF, Connor DJ, Craufurd P, Eigenbrode SD, Han X, Hatfield JL. Challenges and Responses to Ongoing and Projected Climate Change for Dryland Cereal Production Systems throughout the World. *Agronomy*. 2018; 8(4):34.

Obun C.O. (2008). Performance, Digestibility and Carcass and Organ Weights of Finisher Broiler Chicks Fed Graded Levels of Fermented Locust Bean (*Parkia biglobosa*) Seed Meal. Asian J. Poult. Sci., 2: 17-23.

Oluwole, O., Ibidapo, O., Arowosola, T., Raji, F., Zandonadi, R. P., Alasqah, I., ... & Raposo, A. (2023). Sustainable transformation agenda for enhanced global food and nutrition security: a narrative review. *Frontiers in nutrition*, *10*, 1226538.

Owheruo, J. O., Ifesan, B. O., & Kolawole, A. O. (2019). Physicochemical properties of malted finger millet (Eleusine coracana) and pearl millet (Pennisetum glaucum). *Food science & nutrition*, *7*(2), 476-482.

Patni, D., & Agrawal, M. (2017). Wonder millet-pearl millet, nutrient composition and potential health benefits-A review. *International Journal of Innovative Research and Review*, *5*(1), 6-14.

Preetika, A., Padmini, G., & Shobha, U. (2004). Nutrient dense mixes for enteral feeding in India. *Nutrition & Food Science*, *34*(6), 277-281.

Rahman, S., Mohammed, S., Dubey, P. K., & Kumar, S. (2023). A comprehensive review on the effect of germination on the physiochemical properties of wheat, millet, and legumes. *Journal of Food Chemistry & Nanotechnology*, *9*(S1), S323-S334.

Rasal, K. D., & Sundaray, J. K. (2020). Status of genetic and genomic approaches for delineating biological information and improving aquaculture production of farmed rohu, *Labeo rohita* (Ham, 1822). *Reviews in Aquaculture*, *12*(4), 2466-2480.

Rasal, K. D., & Sundaray, J. K. (2020). Status of genetic and genomic approaches for delineating biological information and improving aquaculture production of farmed rohu, *Labeo rohita* (Ham, 1822). *Reviews in Aquaculture*, *12*(4), 2466-2480.

Sarita, E. S., & Singh, E. (2016). Potential of millets: nutrients composition and health benefits. *Journal of Scientific and Innovative Research*, *5*(2), 46-50.

Sehgal, S., Kawatra, A. & Sing, G. (2003). Recent technologies in Pearl Millet and Sorghum processing and food product development. *CC Technical Paper no.34 Proceedings of the Expert Meeting ICRISAT Patancheru, Andhra Pradesh, India*.

Sharma, B., & Gujral, H. S. (2019). Influence of nutritional and antinutritional components on dough rheology and in vitro protein & starch digestibility of minor millets. *Food Chemistry*, *299*, 125115.

Sharma, R., & Sharma, B. (2023). Effect of Sorghum millet (Jowar) on different water quality parameters for *Oreochromis niloticus* (Nile tilapia) (Linnaeus 1758) fingerlings. *Pharm Innov J*, *12*, 4008-4012.

Shobana, S., Krishnaswamy, K., Sudha, V., Malleshi, N. G., Anjana, R. M., Palaniappan, L., & Mohan, V. (2013). Finger millet (Ragi, *Eleusine coracana* L.): a review of its nutritional properties, processing, and plausible health benefits. *Advances in food and nutrition research*, *69*, 1-39.

Singh, A. k., Rehal, J., Kaur, A., & Jyot, G. (2015). Enhancement of Attributes of Cereals by Germination and Fermentation: A Review. *Critical Reviews in Food Science and Nutrition*, *55*(11), 1575–1589. https://doi.org/10.1080/10408398.2012.706661

Sokal, R. R., & Rohlf, F. J. (1981). Taxonomic congruence in the Leptopodomorpha re-examined. *Systematic Zoology*, *30*(3), 309-325.

Surasani, V. K. R., Raju, C. V., Sofi, F. R., & Shafiq, U. (2022). Utilization of protein isolates from rohu (*Labeo rohita*) processing waste through incorporation into fish sausages; quality evaluation of the resultant paste and end product. *Journal of the Science of Food and Agriculture*, *102*(3), 1263-1270.

Taylor, J.R. Millet Pearl: Overview. In Encyclopedia of Food Grains: Second Edition; Colin Wrigley: Brisbane, Australia, 2015; ISBN 9780123947864.

Tewe, O.O., Losel, D.M. & Abu O.A. (1999). Solid state fermentation of sweet potato using two monoculture fungi: Changes in protei fatty acids and mineral composition. Tropical Journal of Animal Science, 5: 219-224.

Utley, H.C., Bernheim, F., Hachslien, P. (1967). Effects of sulfhydryl reagent on peroxidation in microsome. *Arch Biochem Biophys* 260, 521-531.

Wesołowski, R., Mila-Kierzenkowska, C., Pawłowska, M., Szewczyk-Golec, K., Saletnik, Ł., Sutkowy, P., & Woźniak, A. (2023). The influence of winter swimming on oxidative stress indicators in the blood of healthy males. *Metabolites*, *13*(2), 143.

Wong, S.Y. (1928). Colorimetric determination of iron and haemoglobin in blood. *J Biol Chem* 77, 409–412.

Yang, J., Zhang, Y., Jia, J., Wang, C.,& Fu, Y. (2025). Flavonoid-lignin crosstalk: engineering metabolic flux for optimised plant growth and stress resilience. *Plant, Cell & Environment*, *48*(11), 8141-8160.

Yokoyama, H.O. (1947). Studies on the origin, development and seasonal variations in the blood cells of the perch, *Perca flavescens*. (Ph.D. Thesis) University of Wisconsin, Madison Wisconsin 144 pp.