

Effects of Dietary *Mentha piperita* Supplementation on Growth Performance, Feed Utilization, and Proximate Composition of *Labeo rohita* Fingerlings

Sania Ramzan^{a,1}, Saba Malik^{b,1}, Amara Akhtar^{a,*}, Hafiza Sadia^c, Sheeba Riaz^c, Namrah^a, Ahmad Ali^a, Iqra Bibi^a

^aDepartment of Zoology, Wildlife & Fisheries, University of Agriculture, Faisalabad, Pakistan

^bKey Laboratory of Water Pollution Control and Green Resource Recycling, School of the Environment, Nanjing University, Nanjing, Jiangsu 210023, China

^cState Key Laboratory of Pharmaceutical Biotechnology, School of Life Sciences, Nanjing University, Nanjing 210023, China

*Correspondence: amara.akhtar47@uaf.edu.pk

¹These authors have contributed equally to this work

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Abstract Fish is a preferred and affordable source of high-quality animal protein for humans. *Labeo rohita* is one of the most important Indian major carps cultured in polyculture systems. *Mentha piperita* (peppermint) is a medicinal plant belonging to the Lamiaceae family, with bioactive compounds that may enhance growth and feed utilization in fish. The present study investigated the effects of dietary peppermint supplementation on growth performance, feed utilization, and proximate body composition of *L. rohita*. Fingerlings (n = 10 per tank) were acclimatized for 4–6 days before the 28-day experimental trial. Four dietary treatments were prepared: T0 (control, 0 g/kg), T1 (100 g/kg), T2 (200 g/kg), and T3 (300 g/kg) peppermint supplementation. Growth performance parameters, including weight gain, fork length, total length, and specific growth rate (SGR), were measured weekly. Feed conversion ratio (FCR) and body composition (moisture, crude protein, crude fat, ash) were also evaluated. Results indicated that dietary peppermint significantly improved growth performance of *L. rohita*. Weight gain, total length, fork length, and SGR were highest in T3 (16.94 ± 1.954 g, 9.74 ± 0.492 cm, 6.79 ± 0.643 cm, 1.71 ± 0.583, respectively). FCR was lowest in T3 (best feed utilization), while moisture and crude fat were highest in T2 (75.51 ± 0.05%, 1.32 ± 0.04%), and crude protein and ash content were highest in T3 (22.26 ± 0.05%, 0.98 ± 0.04%). These results suggest that dietary peppermint supplementation, particularly at 300 g/kg, enhances growth performance, feed efficiency, and body composition of *L. rohita*. Data were expressed as mean ± SD, and statistical differences among treatments were determined using one-way ANOVA followed by Tukey's post hoc test.

Keywords: *Labeo rohita*; *Mentha piperita*; peppermint supplementation; growth performance; feed conversion ratio; specific growth rate; proximate composition; aquaculture nutrition.

Introduction

Fish plays a significant role in providing high-quality aquatic food and essential nutrients [17]. It is a rich

source of proteins, amino acids, minerals, vitamins, and polyunsaturated fatty acids (PUFAs), which contribute to human health by reducing risks of cardiovascular disorders and certain cancers [7]. Fish protein is highly digestible and contains essential

amino acids that are limited in cereals and grains, making it especially important for nutrition in developing and low-income countries [22]. Fish meat also provides omega-3 and omega-6 fatty acids, which can reduce thrombosis and atherosclerosis (22). With the growing human population, demand for fish and aquaculture products has increased globally [27]. Aquaculture is the fastest-growing food production sector and contributes substantially to fish supply in Asia, where China alone produces over 45 million tonnes annually, accounting for more than 60% of global output [9]. In Pakistan, freshwater and marine water bodies support diverse fish species, yet per capita fish consumption remains low (1.9 kg/year), highlighting the need to improve domestic aquaculture production [12; 1].

Among freshwater species, *Labeo rohita* is one of the most commercially important Indian major carps. It is widely cultured in polyculture systems alongside catla (*Catla catla*) and mrigal (*Cirrhinus mrigala*), and is highly preferred for its nutritional quality and market value [26; 11]. Enhancing its growth performance, feed utilization, and body composition is critical for sustainable aquaculture production [19]. Intensive and semi-intensive culture systems often expose fish to stress from environmental fluctuations, high stocking densities, and disease, which can impair feed utilization, growth, and immunity [13]. While antibiotics and synthetic growth promoters have been used to enhance fish performance, their prolonged use may lead to environmental and consumer health concerns [13]. Natural feed additives, such as medicinal plants, are therefore gaining attention as safer alternatives for improving fish growth, immunity, and overall health [8; 16].

Mentha piperita L. (peppermint) is a medicinal plant from the Lamiaceae family containing biologically active compounds including menthol, menthone, menthyl acetate, and other volatile oils [18]. These compounds exhibit antioxidant, antimicrobial, appetite-stimulating, and immunostimulatory properties, making peppermint a potential natural growth promoter in aquaculture [23]. Dietary supplementation with peppermint has been shown to enhance growth performance, specific growth rate [3], feed utilization [24], and body composition while reducing mortality in fish [25]. The present study evaluated the effects of dietary *Mentha piperita* supplementation on growth performance, feed utilization, proximate body composition, and selected water quality responses in *Labeo rohita*. Specifically, the study assessed changes in weight gain, specific growth rate, feed conversion ratio, survival rate, and

the proximate composition parameters including crude protein, crude fat, moisture, and ash content of fish fed diets containing different levels of peppermint supplementation.

Methodology

Experimental Design

The feeding trial was conducted in the Fish Nutrition Laboratory, Department of Zoology, Wildlife and Fisheries, University of Agriculture, Faisalabad. Fingerlings of *Labeo rohita* were obtained from a fish hatchery at Satiana Road, Faisalabad, and acclimatized to laboratory conditions for 7 days before the start of the experiment. Four glass aquaria, each with an 80 L water-holding capacity, were used as experimental units. Each aquarium was stocked with 10 fingerlings of uniform size. The fish were randomly assigned to four dietary treatments: T0 served as the control group and received a basal diet without *Mentha piperita*, while T1, T2, and T3 received diets supplemented with *Mentha piperita* powder at 100 g/kg, 200 g/kg, and 300 g/kg, respectively. During the acclimatization period, fish were fed the basal diet once daily, and water quality parameters were maintained under suitable laboratory conditions.



Figure 1. Experimental System of fish aquarium in nutritional fish laboratory.

Feed Preparation and Feeding

Two types of diets were prepared: a basal diet without supplementation (T0) and experimental diets supplemented with *Mentha piperita* powder at 100 g/kg (T1), 200 g/kg (T2), and 300 g/kg (T3). Feed ingredients, including fish meal, sunflower meal, rice polish, minerals, vitamins, sunflower oil, and chromium oxide, were weighed, ground, and homogenized to ensure uniform consistency. The prepared diets were formed into pellets, packed in polyethylene bags, and labeled according to treatment. Fish were fed the respective diets at a rate of 6% of body weight, administered twice daily throughout the experimental period. Uneaten feed

was collected after 3 hours, dried, and used to calculate feed conversion ratios.

Growth Performance and Feed Utilization

Growth performance and feed utilization indices, including weight gain (WG), specific growth rate (SGR), feed conversion ratio (FCR), and survival rate, were measured weekly throughout the 4-week feeding trial. Fish were individually weighed to determine WG and SGR, and total feed intake was recorded to calculate FCR. Survival rate (%) was determined at the end of each week using the formula:

Weight gain % of fish was calculated as,

$$\text{Weight gain \%} = \frac{\text{Final weight} - \text{initial weight}}{\text{Initial weight}} \times 100$$

These measurements were used to evaluate the effects of dietary *Mentha piperita* supplementation on the growth performance and feed utilization of *Labeo rohita*.

Proximate Composition

At the end of the experimental period, muscle samples of *Labeo rohita* were collected to determine proximate composition, including crude protein, crude fat, moisture, and ash content. Analyses were performed following standard AOAC (2005) methods. Crude protein was determined using the Kjeldahl method, crude fat was extracted using a Soxhlet apparatus with petroleum ether, moisture content was measured by oven drying to constant weight, and ash content was determined by incineration in a muffle furnace at 550–650 °C. These analyses were used to evaluate the effects of dietary *Mentha piperita* supplementation on the body composition of the fish.

Water Quality Monitoring

Water quality parameters, including temperature, pH, dissolved oxygen, total alkalinity, and hardness, were monitored weekly in all experimental aquaria throughout the trial. Temperature and dissolved oxygen were measured using a HANNA HI-9146 electronic meter, while pH was determined with an electronic pH meter. Total alkalinity and hardness were assessed following standard APHA (1998) methods. These measurements ensured that water quality remained within suitable ranges for optimal growth of *Labeo rohita* during the feeding trial.

Statistical Analysis

All experimental data were expressed as mean \pm standard deviation (SD) and analyzed using one-way

analysis of variance (ANOVA) to evaluate the effects of dietary *Mentha piperita* supplementation. Significant differences among treatment means were determined using Tukey's post hoc test, with a significance threshold set at $P < 0.05$. Graphical representations of growth performance, feed utilization, proximate composition, and water quality parameters were generated using Python (Matplotlib and NumPy libraries).

Results

The effects of dietary *Mentha piperita* supplementation on fork length, total length gain, weight gain, and specific growth rate (SGR) of *Labeo rohita* are presented in Figure 2. Values are expressed as mean \pm standard deviation (SD) for treatments T0 (0 g/kg), T1 (100 g/kg), T2 (200 g/kg), and T3 (300 g/kg). Statistical significance among treatments was assessed using Tukey's post hoc test, where ns = non-significant, = significant, = highly significant, and = very highly significant. Fork length of *Labeo rohita* increased significantly with dietary peppermint supplementation. The lowest fork length was observed in the control group (T0: 5.43 \pm 0.743 cm), while the highest was recorded in T3 (6.79 \pm 0.643 cm). Treatments T2 (6.21 \pm 0.853 cm) and T3 showed significantly higher fork lengths compared to T0 and T1 ($P < 0.05$).

Total length gain followed a similar trend, with T3 exhibiting the highest gain (9.74 \pm 0.492 cm), whereas T0 showed the lowest (7.05 \pm 0.742 cm). Significant differences were observed between T3 and the lower supplementation levels (T0 and T1), indicating a dose-dependent effect of peppermint on growth. Weight gain also increased with peppermint supplementation. T3-fed fish showed the highest weight gain (16.94 \pm 1.954 g), followed by T2 (14.32 \pm 1.042 g), T1 (11.75 \pm 0.892 g), and T0 (9.57 \pm 0.843 g). Statistical analysis confirmed significant differences between T3 and T0 ($P < 0.05$), while T1 and T2 displayed intermediate values. Similarly, specific growth rate (SGR) increased with dietary peppermint inclusion. The control group (T0) recorded the lowest SGR (1.165 \pm 1.422), while the highest SGR was observed in T3 (1.71 \pm 0.583). Treatments T2 and T3 showed significantly higher SGR compared to T0 ($P < 0.05$). The results indicate that dietary supplementation with *Mentha piperita* significantly enhances growth performance and feed utilization of *Labeo rohita*, with the highest responses observed at 300 g/kg supplementation.

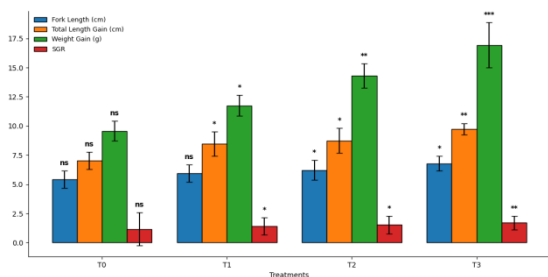


Figure 2. Effect of dietary *Mentha piperita* supplementation on fork length, total length gain, weight gain, and specific growth rate of *Labeo rohita*. Values are presented as mean \pm SD for T0, T1, T2, and T3. Asterisks indicate significant differences among treatments according to Tukey's test, where ns = non-significant, * = significant, ** = highly significant, and *** = very highly significant.

The proximate composition of *Labeo rohita* fed diets supplemented with different levels of *Mentha piperita* is presented in Figure 3. Dietary peppermint supplementation influenced the body composition of fish, including crude protein, crude fat, moisture, and ash content. Crude protein content increased progressively with higher levels of peppermint supplementation. The lowest protein was observed in the control group (T0: 19.21 \pm 0.02%), while the highest was recorded in T3 (22.26 \pm 0.05%). Statistically significant differences were observed between T3 and T0 ($P < 0.05$), whereas intermediate levels (T1: 20.28 \pm 0.05%, T2: 21.29 \pm 0.05%) showed moderate increases.

Crude fat content was highest in T2 (1.32 \pm 0.04%) and lowest in T1 (1.23 \pm 0.04%), with T0 and T3 showing intermediate values. Moisture content increased slightly with supplementation, reaching a maximum in T2 (75.51 \pm 0.05%) and was lowest in T0 (70.64 \pm 0.05%). Ash content followed a similar trend, with the highest ash observed in T3 (0.98 \pm 0.04%) and the lowest in T0 (0.86 \pm 0.06%). Significant differences among treatments were indicated by Tukey's post hoc test, with asterisks marking statistically significant variations. Overall, the results demonstrate that dietary inclusion of *Mentha piperita* positively affects the proximate composition of *Labeo rohita*, enhancing protein and fat contents while modulating moisture and ash levels.

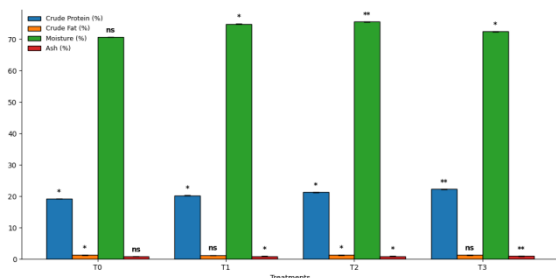


Figure 3. Proximate composition of *Labeo rohita* fed diets supplemented with *Mentha piperita* at 0 g/kg (T0), 100 g/kg (T1), 200 g/kg (T2), and 300 g/kg (T3). Values are presented as mean \pm SD. Asterisks indicate statistically significant differences among treatments according to Tukey's post hoc test ($P < 0.05$; $P < 0.01$).

Water quality parameters, including temperature, pH, and dissolved oxygen, were monitored throughout the experimental period and are presented in Figure 4. Temperature remained within a suitable range across all treatments, with mean values ranging from 28.11 \pm 0.97 $^{\circ}$ C in T0 to 30.32 \pm 0.97 $^{\circ}$ C in T2. The highest mean temperature was recorded in T2, while the lowest was observed in the control group. The pH values also varied among treatments, with mean values ranging from 8.92 \pm 0.92 in T0 to 9.77 \pm 0.75 in T2. The highest pH was recorded in T2, followed by T3, T1, and T0. Dissolved oxygen values ranged from 7.57 \pm 0.61 mg/L in T0 to 8.26 \pm 0.65 mg/L in T1. Overall, the recorded water quality parameters remained within acceptable ranges for the culture of *Labeo rohita* and supported normal growth during the feeding trial.

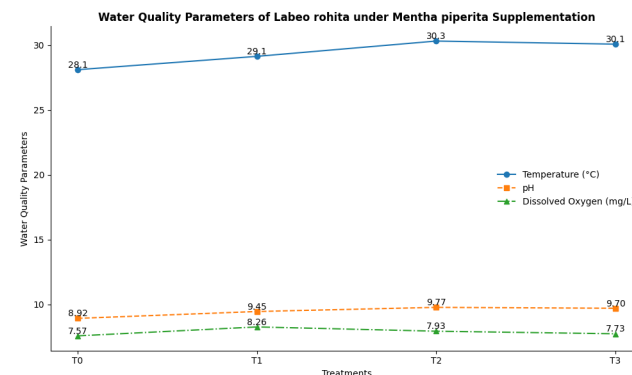


Figure 4. Water quality parameters recorded during the feeding trial of *Labeo rohita* fed diets supplemented with *Mentha piperita*. Values represent mean \pm SD of temperature, pH, and dissolved oxygen across treatments T0, T1, T2, and T3.

Discussion

The present study evaluated the effects of dietary *Mentha piperita* supplementation on growth performance, feed utilization, proximate body composition, and water quality parameters of *Labeo rohita*. Fish are an important source of high-quality protein, essential fatty acids, and micronutrients, and improvement in aquaculture nutrition is necessary to enhance fish growth and production [5]. The use of medicinal plants as natural feed additives has gained attention because they may improve growth, nutrient utilization, antioxidant activity, and general health

without the negative effects associated with synthetic chemicals or antibiotics [6].

In the present study, dietary supplementation with *M. piperita* improved the growth performance of *L. rohita*. The highest weight gain was recorded in T3 (16.94 ± 1.954 g), followed by T2 (14.32 ± 1.042 g), T1 (11.75 ± 0.892 g), and T0 (9.57 ± 0.843 g). The control group showed the lowest weight gain, indicating that peppermint supplementation had a positive effect on fish growth. This improvement may be attributed to the presence of bioactive compounds in peppermint, including menthol, menthone, and menthyl acetate, which may enhance appetite, digestion, nutrient absorption, and metabolic activity [23]. Similar findings were reported by [24], who observed improved growth performance, specific growth rate, and feed utilization in fish fed diets supplemented with *M. piperita*. [25] also reported that herbal supplementation improved weight gain and specific growth rate in fish.

Feed conversion ratio is an important indicator of feed utilization efficiency because it reflects how effectively dietary nutrients are converted into fish biomass [15]. In the present study, FCR improved in peppermint-fed groups compared with the control, with the lowest FCR observed in T3 followed by T2, indicating better feed utilization at higher *M. piperita* supplementation levels. This improvement may be associated with the phytochemical compounds present in peppermint, which can enhance digestive enzyme activity, nutrient availability, gut function, and feed assimilation. Similar findings were reported by [14], who observed improved FCR and growth performance in fish fed diets containing *M. piperita*. [4] also reported that dietary peppermint essential oil improved growth performance, feed conversion, digestive enzyme activity, and oxidative status in juvenile Nile tilapia, supporting the role of peppermint in improving nutrient utilization. Similarly, [21] observed improved growth, survival, and haemato-biochemical responses in common carp fingerlings supplemented with peppermint. In the present study, SGR also increased with dietary peppermint inclusion, with the highest value recorded in T3 and the lowest in T0. This suggests that peppermint supplementation enhanced growth rate, likely through improved feed intake, digestion, and nutrient assimilation. The 100% survival rate observed in all treatments further indicates that the diets were safe and that the culture conditions remained favorable throughout the trial.

Dietary *M. piperita* supplementation also influenced the proximate composition of *L. rohita*. Crude protein content increased with higher supplementation levels

and was highest in T3 ($22.26 \pm 0.05\%$) compared with T0 ($19.21 \pm 0.02\%$). The increase in body protein may indicate improved protein deposition and more efficient nutrient utilization in fish fed peppermint-supplemented diets. This response may be associated with the bioactive compounds of peppermint, which can enhance digestive efficiency, nutrient absorption, metabolic activity, and physiological condition. Similar observations were reported by [24] and [25], who found increased protein content and improved growth responses in fish fed herbal-supplemented diets. In addition, [20] reported that dietary peppermint powder improved growth performance and physiological responses in Caspian roach, while [2] observed that *M. piperita* supplementation enhanced growth, immunity, and disease resistance in Nile tilapia. [10] also reported improved body composition and hematological responses in fish fed plant-based dietary supplementation, supporting the role of medicinal plants in improving nutrient deposition. Crude fat content was highest in T2 ($1.32 \pm 0.04\%$) and lowest in T1 ($1.23 \pm 0.04\%$). The variation in lipid content may be related to differences in nutrient metabolism, lipid utilization, and energy storage among treatments. Herbal additives may influence lipid digestion and utilization by stimulating digestive secretions and improving metabolic activity. Moisture content was also affected by dietary treatment, with the highest value recorded in T2 ($75.51 \pm 0.05\%$) and the lowest in T0 ($70.64 \pm 0.05\%$). Ash content was highest in T3 ($0.98 \pm 0.04\%$) and lowest in T0 ($0.86 \pm 0.06\%$), indicating that peppermint supplementation may have influenced mineral deposition in fish tissues. These results suggest that *M. piperita* supplementation can modify the body composition of *L. rohita*, particularly protein and mineral content, and may improve the nutritional quality of fish under controlled culture conditions.

Water quality parameters, including temperature, pH, and dissolved oxygen, remained within suitable ranges during the experimental period [28]. Maintenance of appropriate water quality is important because poor water conditions can reduce feed intake, growth, and survival [14]. The 100% survival observed in all treatments further confirms that the experimental conditions were favorable and that peppermint supplementation did not produce any harmful effect on fish survival. Overall, the findings of the present study indicate that dietary *M. piperita* supplementation improved growth performance, feed utilization, and proximate body composition of *L. rohita*. Among the tested treatments, T3 showed the

most favorable response for weight gain, SGR, FCR, crude protein, and ash content, while T2 showed the highest crude fat and moisture contents. Therefore, peppermint may be considered a promising natural feed additive for improving growth and nutritional quality of *L. rohita* under controlled culture conditions.

Conclusions

The present study was conducted to evaluate the effect of dietary *Mentha piperita* supplementation on growth performance, feed utilization, proximate body composition, and water quality parameters of *Labeo rohita*. Growth performance was assessed through weight gain, fork length, total length gain, specific growth rate, and feed conversion ratio. The highest weight gain was recorded in T3 (16.94 ± 1.954 g), while the lowest was observed in T0 (9.57 ± 0.843 g). Fork length was also highest in T3 (6.79 ± 0.643 cm) and lowest in T0 (5.43 ± 0.743 cm). Total length gain showed a similar trend, with the highest value in T3 (9.74 ± 0.492 cm) and the lowest value in T0 (7.05 ± 0.742 cm). Specific growth rate was maximum in T3 (1.71 ± 0.583) and minimum in T0 (1.165 ± 1.422). Feed conversion ratio improved with peppermint supplementation, with the lowest FCR observed in T3, indicating better feed utilization. Proximate body composition was also influenced by dietary *M. piperita*. Crude protein content was highest in T3 ($22.26 \pm 0.05\%$) and lowest in T0 ($19.21 \pm 0.02\%$). Crude fat content was maximum in T2 ($1.32 \pm 0.04\%$) and minimum in T1 ($1.23 \pm 0.04\%$). Moisture content was highest in T2 ($75.51 \pm 0.05\%$) and lowest in T0 ($70.64 \pm 0.05\%$). Ash content was maximum in T3 ($0.98 \pm 0.04\%$) and minimum in T0 ($0.86 \pm 0.06\%$). Water quality parameters, including temperature, dissolved oxygen, and pH, were maintained within suitable ranges throughout the experimental period. Survival rate remained 100% in all treatments, indicating that the experimental conditions were favorable and that dietary *M. piperita* supplementation had no harmful effect on fish survival. Overall, the results suggest that dietary *Mentha piperita* supplementation improved growth performance, feed utilization, and proximate body composition of *Labeo rohita*. Among the tested treatments, T3 showed the most favorable response for weight gain, fork length, total length gain, specific growth rate, crude protein, and ash content.

Author Contributions

Sania Ramzan: Writing – original draft, Visualization, Methodology, Investigation, Formal analysis, Data

curation, Conceptualization. Saba Malik: Writing – review & editing, Visualization, Formal analysis, Conceptualization. Amara Akhtar: Writing – review & editing, Validation, Supervision, Resources, Project administration, Funding acquisition. Hafiza Sadia: Writing – review & editing, Conceptualization. Sheeba Riaz: Writing – review & editing, Conceptualization. Namrah: Writing – review & editing, Conceptualization. Ahmad Ali: Writing – review & editing, Conceptualization. Iqra Bibi: Writing – review & editing, Conceptualization.

Conflicts of Interest

The author(s) declare(s) that there is no conflict of interest regarding the publication of this paper.

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Data Availability

Data will be available on request.

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