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Turmeric extract supplementation for augmenting growth and health of *Labeo gonius*: A study in the hill conditions of Meghalaya, North-east India

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Abstract

In this study, we evaluated the effects of turmeric extract (TE) supplementation on the growth and health of *Labeo gonius* (Hamilton, 1822), under Meghalaya conditions using cemented cisterns (each 1 m x 1 m x 1 m). The experimental fish, with an average size of 6.38±0.35 g/6.2±0.32 cm, were stocked at a rate of 30 fish per tank and fed feeds containing TE at concentrations of 0 (control), 2 and 4%, representing 3% of their biomass. Each feed type was assessed in replicates. We monitored water quality parameters fortnightly and assessed fish growth monthly. Fish were sampled (n=10) from each tank on the 15th, 30th and 60th days post-feeding and analysed for growth and blood parameters. The results revealed a significant increase in specific growth rate, with a remarkable 48.1% boost in the group fed with TE. Survival rates ranged from 88.4 to 90% for fish fed with TE compared to 78.4% for the control group. Fish fed with 2% TE demonstrated better condition factor (b = 2.8 to 3.1). Blood analysis showed elevated RBC and WBC counts, increased haemoglobin and total protein levels, reduced glucose and cholesterol content and glutamic oxaloacetic transaminase (GOT) and glutamic pyruvic transaminase (GPT) activity within normal ranges. Overall, optimal outcomes were observed in *L. gonius* with 2% TE supplementation in the feed. This study concludes that the inclusion of TE in the feed improves fish health and growth under the hill conditions of Meghalaya, North-east India. These findings are valuable for improving aquaculture strategies, particularly in hilly settings.

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Introduction

Aquaculture plays a crucial role in the economic and livelihood development of North-east (NE) India. However, aquaculture production in this region lags behind that of plain lands (Das, 2018). The harsh cold climate in hill ecosystems in NE India adversely affects fish growth, immunity and reproductive competence (Majhi *et al*., 2013; Das and Majhi, 2014). Particularly in hill states like Meghalaya, the annual mean water temperature rarely exceeds 20°C (Das *et al*., 2012). During the winter season (late November to February), the water temperature can rapidly drop from 22 to 6°C, causing significant stress to fish and other aquatic organisms. To promote fish immunity and growth in this environmentally challenging situation, plant-based solutions could be an ideal option.

Turmeric (*Curcuma longa*), a well-known culinary and medicinal herb in Asia, whcich contains curcumin, a bioactive compound with growth-promoting, antibacterial, immunostimulant, antioxidative and anti-inflammatory properties (Singletary, 2020). Turmeric has shown promising outcomes in aquaculture, enhancing growth rates, antioxidative status, immunity and disease resistance in various fishes (Alagawany *et al*., 2021). However, its effectiveness has not been evaluated for many fishes, particularly for fish species gaining importance in regional aquaculture, such as *Labeo gonius* (Hamilton, 1822).

L. gonius, also known as Kurio labeo, is an important fish species endemic to NE India (Jena *et al*., 2011; Das *et al*., 2021). It offers several advantages, including compatibility with other carps in composite culture setups and potential as a substitute for mrigal (Jena and Das, 2011). While numerous studies have been conducted on *L. gonius*, such as induced breeding (Behera *et al*., 2009; Das *et al*., 2010), nursery rearing (Chakraborty and Mirza, 2009), compatibility with other carps (Jena and Das, 2011; Jena *et al*., 2015; Das *et al*., 2021; Debnath and Mahanta, 2023) as well as thermal tolerance (Das *et al*., 2020), studies on its growth and immunity under varying diets and agro-ecosystems are limited.

L. gonius is slowly gaining importance as a candidate species for aquaculture in NE India; however, it suffers from low growth rate and poor survival rate, particularly in hill conditions like Meghalaya. In this study, we explored the potential of turmeric as a feed additive for improving the growth and health of the fish under the agro-climatic conditions of Meghalaya, NE India. Our hypothesis is that turmeric supplementation in the feed will enhance the growth rate, improve the condition factor and bolster immunity of *L. gonius*, leading to better survival rates and overall health in hill environments.

Materials and methods

Experimental set up

The research was conducted in Meghalaya, a state located in the NE India (21°05' to 29°50'N and 85°05' to 97°05'E) from February to March 2022 (2 months). The study site situated at an elevation of 950 m above mean sea level, characterised by a typical cold to warm pre-humid climate with an annual rainfall exceeding 2000 mm. The air temperature varied between 11 and 26°C, while the water temperature ranged from 10 to 24°C. For the experiment, cemented tanks were utilised, with each tank measuring 1 cubic meter (1.0 m x 1.0 m x 1.0 m) in size. Prior to commencing the study, a meticulous cleaning and sanitisation process was carried out on the tanks. This involved employing a solution of potassium permanganate ($KMnO_4$) at a concentration of 5 mg l^{-1}. Subsequently, the tanks were filled with pond water. Following water filling, the tanks were left undisturbed for a period of 2 days to attain stability. Afterwards, fingerlings of *L. gonius*, collected from the farm of the ICAR Complex for NEH region (ICAR-RC NEH), were introduced into a hapa for acclimation over the course of a week. The stocking density was maintained at 30 fish per tank. During the stocking process, the average size of the fingerlings was determined to be approximately 6.38±0.35 g, with a length of 6.2±0.32 cm. Within 24 h of stocking, the fish were provided with experimental feeds.

Turmeric

The turmeric variety used for the preparation of turmeric extract (TE) in this experiment was "Megha Turmeric 1," which was sourced from our institute's horticulture research farm. This particular variety, known as Megha Turmeric-1, was developed by the ICAR-RC NEH, in Meghalaya through a clonal selection from the World-renowned 'Lakadong' turmeric variety, known for its exceptional quality and global recognition. It was found containing 6.4% curcumin during our analysis in the institute.

Preparation of feeds and feeding

The study involved formulating three experimental feeds with the inclusion of TE as feed additive in different concentrations: 0% (control), 2% and 4% (Table 1). The basic feed ingredients, including rice bran (RB), mustard oil cake (MOC) and fish meal (FM), were obtained from the local market (M/s The Shillong Cooperative Marketing Society Ltd., Iewduh, Shillong, Meghalaya). Initially, the basal ingredients were thoroughly mixed with water and cooked using a pressure cooker. After the mixture cooled down, vitamin-mineral mixture (1% of total feed) was added. Fresh TE, obtained by squeezing the rizhome, was then incorporated into the feeds.

The mixtures were subsequently passed through a hand pelletiser to form pellets measuring 1 mm in dia. These pellets were then dried in a hot air oven at 40°C for two days and stored in airtight containers for future use. After preparation, the feeds were subjected to proximate composition analysis using the methods outlined in AOAC (1980). The feeds exhibited a crude protein (CP) content ranging from 24.3 to 24.7% and a crude lipid content ranging from 6.5 to 6.8% (Table 1). The fish were offered the prepared feeds daily, at a rate of 3% of their body weight. Periodic aeration was provided in the tank and additionally, a 50% exchange of tank water was carried out on a weekly basis. The experimental design followed a completely randomised design (CRD), with a total of nine tanks being utilised to evaluate the three feeding treatments in triplicates.

Assessment of fish growth and survival

From each tank, ten fish (n=10) were randomly selected on 15th, 30th and 60th day after feeding to evaluate their growth. The total length (TL) of each fish was determined to the nearest centimeter (cm) using a measuring scale, while their weight was measured to the nearest 0.1 g using an analytical balance (Make: Shimadzu,

Table 1. Composition of experimental feeds

Ingredient	Control (without TE)	2% TE	4% TE
Rice bran	600	580	560
Mustard oil cake	200	200	200
Fish meal	140	140	140
Wheat flour	50	50	50
Vitamin-mineral mixture*	10	10	10
Turmeric extract (g)	0	20	40
Total (g)	1000	1000	1000
Proximate analysis (%)			
Moisture	12.2	12.5	12.5
CP	24.7	24.2	24.3
Crude lipid	6.8	6.5	6.5
Total ash	7.5	7.4	7.5
Crude fiber	48.8	49.4	49.2

Ingredients were sourced from M/s. The Shillong Cooperative Marketing Society Ltd., Shillong, Meghalaya.
*Composition of vitamin-mineral premix (quantity per 2.5 kg): Vitamin A, 5500000 IU; Vitamin D3, 1100000 IU; Vitamin B2, 2000 mg; Vitamin E, 750 mg; Vitamin K, 1000 mg; Vitamin B6, 1000 mg; Vitamin B12, 6 mcg; Calcium pantothenate, 2500 mg; Nicotinamide, 10 g; Choline chloride, 150 g; Manganese, 27,000 mg; Iodine, 1000 mg; Iron, 7500 mg; Zinc, 5000 mg; Copper, 2000 mg; Cobalt, 450 mg; L- lysine, 10 g; DL- Methionine, 10 g; Selenium, 50 ppm; Satwari, 2500 mg.

ATX224). Upon completion of the study, all fish were harvested by draining the tanks. The total number of fish was counted to calculate the survival rate. The following formulae were employed to estimate different growth attributes and the survival rate of fish.

Weight gain (g) = Final weight (g) - Initial weight (g)

Specific growth rate (SGR,%/day) = [ln (Final weight) - ln (Initial weight)] / (Day of experiment) x 100

Survival rate (%) = (No.of fish recovered) / (No.of fish stocked) x 100

Assessment of fish condition

At the end of the experiment, all fish were harvested, and their total length (TL) in millimeters and wet weight (W) in grams were measured. The statistical relationship between TL and BW was determined using the equation proposed by Le Cren (1951): $W = aL^b$, where 'a' represents the intercept and 'b' represents the slope. To calculate the values of 'a' and 'b', a linear regression analysis was performed on the natural logarithmic transformed equation: Ln(W) = Ln(a) + b Ln(L). The coefficient of determination (r^2) was estimated from the linear regression analysis and indicates the degree of association between the length and weight of the fish. The coefficient 'R' is equal to the square root of r^2. The condition of the fish was assumed based on the value of regression parameter 'b' (Pauly, 1983).

Collection of blood and serum separation

On the 15th, 30th and 60th day post-feeding with the experimental feeds, ten fish (n=10) were randomly sampled from each tank with a triangular net and then anesthetised using an MS-222 solution (0.1 ppm). Five were utilised for blood collection, while the remaining five were used for serum separation. Blood collection was done from the caudal vein of the fish employing a syringe. The collected blood was then transferred to Eppendorf tubes containing a solution of heparin, and the tubes were gently shaken. These blood samples were subsequently utilised for the analysis of red blood cell (RBC) count, white blood cell (WBC) count, and hemoglobin (Hb) levels. Regarding serum collection, after the blood was collected, it was transferred into separate Eppendorf tubes (1.5 ml each) that did not contain any heparin solution. The blood samples were allowed to clot for a duration of 3-4 h. Serum was separated from the clotted blood by subjecting the tubes to centrifugation at 3000 *g* for 5 min. The separated serum was then stored at a temperature of -20°C for subsequent analysis. The serum samples obtained were used for the analysis of various parameters, including glucose levels, total protein levels, total cholesterol levels, as well as the activity of enzymes such as glutamic oxaloacetic transaminase (GOT) and glutamic pyruvic transaminase (GPT).

Analysis of blood

To estimate RBC count and WBC count, the method described by Gupta *et al*. (2008) was followed. The haemoglobin (Hb) level was determined using the cyanmethemoglobin method, as outlined in Gupta *et al*. (2008). For the estimation of glucose level, a glucose diagnostic kit (Crest Biosystems, India) based on the GOD/POD method, developed by Trinder (1969) was employed. The serum protein level was estimated using a total protein kit utilising the Biuret method, as described by Gornall *et al*. (1949). The activity of serum GOT and GPT was estimated using diagnostic kits specifically designed for GOT and GPT. These kits followed the method developed by Reitman and Frankel (1957) and were manufactured by Crest Biosystems, India.

Analysis of water quality parameters

Water quality parameters, including temperature, dissolved oxygen (DO), total dissolved solids (TDS), pH and total ammonia, were regularly tested from each tank following standard methods (Clescer *et al*., 1998). The testing was conducted between 08:00 and 09:00 hrs. Water temperature was measured using a digital thermometer, DO levels were estimated using a DO meter (Make: Lutron PDO-519), pH was measured using a pH meter (Make: Eutech Instruments PCSTestr 35), TDS determined with a TDS meter (Make: TDS-3) and ammonia levels using ammonia testing kit (Make: API, USA). These instruments and kits were used in accordance with their respective manufacturers' instructions.

Statistical analysis

The data were analysed through statistical package for social sciences (SPSS; Version 21.0) for Windows. The statistical analysis involved one-way analysis of variance (ANOVA) and comparison of means following Duncun's multiple range test at 5% level of significance ($p \leq 0.05$).

Results and discussion

Water quality

Water quality is a critical factor in successful fish farming, directly influencing fish health, growth and overall well-being. It is essential to regularly monitor and manage key water parameters to support fish in maintaining normal physiological functions (Majhi *et al*., 2006; Das *et al*., 2018). In this study, the mean values of water temperature, DO, pH, TDS and total ammonia (Table 2) were consistent with previous aquaculture studies conducted in the hilly regions of Meghalaya, North-east India (Majhi and Das, 2014; Das *et al*., 2021; Debnath and Mahanta, 2023).

Water temperature recorded in the study indicates sub-optimal conditions for fish rearing, reflecting the typical cold climate of the region where annual mean temperatures rarely exceed 20°C (Das *et al*., 2012). The temperature data presented in this study represents the mean of February to March, which falls within late winter and early spring, showing relatively warmer water temperatures compared to the annual averages reported in earlier studies.

Originally planned for December-January, the experiment was postponed to February due to extreme weather conditions, which

Table 2. Water quality parameters in the treatment tanks

Feeding	Temperature (^{0}C)	DO (ppm)	pH	TDS (ppm)	Ammonia-N (ppm)
Control	21.25±0.25	7.25±1.25	8±0	86±22	0.75±0.25
2% TE	21.25±0.05	7.85±0.85	8±0	85.5±29.5	0.67±0.23
4% TE	21.15±0.05	7.85±1.55	7.5±0.5	78±6	0.67±0.25

could have induced additional stress factors and confounding variables for the fish. Future studies should explore the potential benefits of turmeric extract under more challenging temperature conditions, including those below 20°C, to fully assess its efficacy in mitigating the impact of cold stress on fish growth and immunity in hill ecosystems.

Fish growth and survival

The feeds in our study had crude protein (CP) content ranging from 24.2 to 24.7% and we used a feeding rate of 3%. We found that including turmeric extract (TE) at 2 and 4% in the diet positively affected the growth and survival of *L. gonius* in the production system, where the mean water temperature ranged from 21.15 to 22.05°C. Over the 60-day rearing period, fish fed the control diet without TE reached an average weight of 11.4 g, while those receiving diets with TE attained average weights of 15.3 and 15.5 g, respectively, representing a substantial 34.2 to 36% increase (Fig. 1). Similarly, the specific growth rate (SGR) of fish fed the control diet without TE was 1.03% day^{-1}, whereas those fed diets with TE achieved significantly higher SGRs of 1.52 and 1.53% day^{-1}, showcasing a remarkable 47.5 to 48.5% improvement (Fig. 2).

In comparison to our study, Debnath and Mahanta (2023) reported an SGR of 1.03% day^{-1} in *L. gonius* co-cultured with *L. rohita* in cemented tanks under Meghalaya conditions (water temperature 19.5 to 19.8°C), by feeding with a conventional feed containing 22.2% crude protein (CP) at 2% of the fish's body weight and 1.52 to 1.53% day (Fig. 2) by feeding with a pellet feed containing 22.4% CP at 2 to 6% of the fish's body weight over a two-month period. Jena *et al*. (2015) conducted a year-round study on *L. gonius* in a major carp culture system. At a 10% species composition and a feed containing 28% CP, the SGR of *L. gonius* was 1.21% day^{-1}. At a 20% species composition with the same feed, the SGR was slightly lower at 1.18% day^{-1}. This study was carried out at water temperatures ranging from 25.9-33.3°C, which are higher than in our study. In another study by Jena and Das (2011), *L. gonius* was reared in a year-round major carp culture system with feed comprising rice polish and groundnut oil cake (1:1). The reported SGRs for *L. gonius* were 0.91-0.92% day^{-1}, with water temperatures varying between 15 and 35°C, dropping to 15-22°C during the winter months.

In another study, Jena *et al*. (2011) reported higher SGRs of 3.47 to 4.66% per day in a co-rearing system of major and medium carp fry, at water temperatures of 24.7-28.1°C. This higher growth rate was attributed to a higher feeding rate of 6 to 8%. Azim *et al*. (2001) observed an SGR of 2.54% per day in *L. gonius* over a four-month period at water temperatures of 30.3-30.4°C. This higher growth rate might be due to the effects of periphyton grown on bamboo substrates used in the trial.

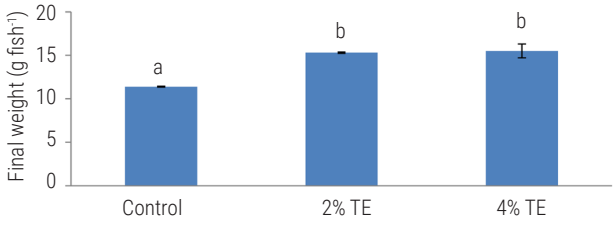

Fig. 1. Final weight in turmeric extract (TE)-fed *L. gonius*

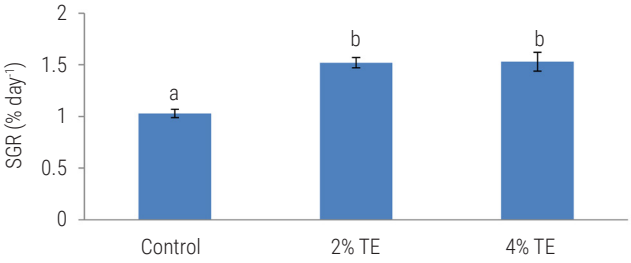

Fig. 2. SGR in turmeric extract (TE)-fed *L. gonius*

Overall, the study demonstrated the beneficial effects of TE on *L. gonius* under Meghalaya conditions. The observed growth enhancement can be attributed to the positive effects of TE on various physiological processes. Curcumin, the primary bioactive compound in turmeric, has been shown to enhance the activity of digestive enzymes in fish intestine (Fernandez-Lazaro *et al*., 2020), facilitating efficient digestion and nutrient absorption, which contributes to overall growth and health. Additionally, the appealing flavour of curcumin may have increased feed palatability, leading to heightened feed intake (Alagawany *et al*., 2021), further supporting the observed growth enhancement. These findings align with previous studies that have demonstrated the growth-promoting effects of turmeric and curcumin in various fish species. For example, Sahu *et al*. (2008) reported enhanced growth and immunity in *L. rohita* fed turmeric-supplemented diets, while Mahmoud *et al*. (2017) observed improved growth performance in *Oreochromis niloticus* fed diets containing curcumin. Similarly, Giri *et al*. (2019) and Ashry *et al*. (2021) reported positive impacts on growth and overall health in *Cyprinus carpio* and *Sparus aurata*, respectively, when turmeric or curcumin was included in their diets.

In addition to promoting growth, the inclusion of TE significantly improved the survival rate of *L. gonius* in our study. Fish fed diets containing TE exhibited a survival rate ranging from 88.4 to 90%, compared to 78.4% for those fed the control diet (Fig. 3). This finding is particularly notable when compared to previous studies on *L. gonius* reared in the hill regions of Meghalaya. Das *et al*. (2021) reported a survival rate of 78 to 83% for *L. gonius* co-cultured with *C. carpio* and fed a diet containing rice polish and mustard oil cake, while Das *et al*. (2020) reported survival rates of 29.5 to 76.8% for *L. gonius* seed rearing using a similar diet. Jena *et al*. (2015) reported 69.2 to 70.8% survival in *L. gonius* with 10 to 20% levels of inclusion in a year-round major carp-based culture system. Jena and Das (2011) reported survival rates of 75.5 to 79.8% in *L. gonius* in a year-round major carp-based culture system. The enhanced survival observed in our study suggests that turmeric extract may have bolstered the immune system and overall health of *L. gonius*, potentially mitigating the adverse effects of cold stress in hill environments.

Fish condition

The condition factor serves as an indirect measure of a fish's overall health and well-being (Froese *et al*., 2006), with a higher value typically indicating better health. In our study, fish fed without turmeric extract had a 'b' value of 1.86, while those fed diets containing 2 and 4% turmeric extract displayed 'b' values ranging from 2.8 to 3.1. Notably, fish nourished with the 2% TE

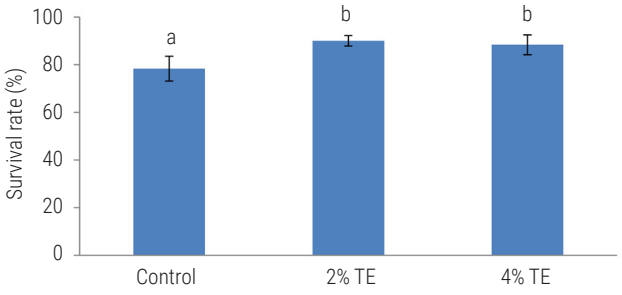

Fig. 3. Survival rate (%) in turmeric extract (TE)-fed *L. gonius*

included diet showed a 'b' value exceeding 3.0, indicating isometric growth and an ideal condition. Although we were unable to make direct comparisons with other studies, the higher condition factor observed in our study suggests that the inclusion of TE in the diet improved the overall condition of *L. gonius*. This improvement may be attributed to the enhanced nutrient bioavailability and assimilation facilitated by turmeric extract. Previous studies have demonstrated that curcumin enhances the activity of digestive enzymes, leading to more efficient nutrient absorption and energy utilisation in fish physiology (Ashry *et al*., 2021). Furthermore, an increased condition factor indirectly indicates greater energy reserves in the fish, enhancing their resilience to stressors such as infections (Chellappa *et al*., 1995).

Haematological parameters

The haematological parameters showed significant improvements in fish fed diets containing turmeric extract (TE) throughout all sampling days. Fish fed the control diet without TE had an average RBC count of 0.82 x 10^6 mm^{-3}, while those fed diets with 2 and 4% TE displayed RBC counts ranging from 1.16 to 1.23 x 10^6 mm^{-3} (Fig. 4). Similarly, the average WBC count was 25.4 x 10^3 mm^{-3} in fish fed the control diet, whereas those fed diets with TE exhibited WBC counts ranging from 27.6 to 31.6 x 10^3 mm^{-3} (Fig. 5). Haemoglobin levels followed a similar trend, with fish fed the control diet showing an average of 7.8 g dl^{-1} and those fed diets with TE displaying levels ranging from 8.6 to 9.3 g dl^{-1} (Fig. 6).

The significant increase in RBC and Hb levels in TE-fed fish highlights the immune-boosting properties of TE (Sahu *et al*., 2008; Mahmoud *et al*., 2017). Although direct challenge studies were not conducted in this research due to limited resources, the observed improvements in RBC and Hb levels suggest an enhancement in the

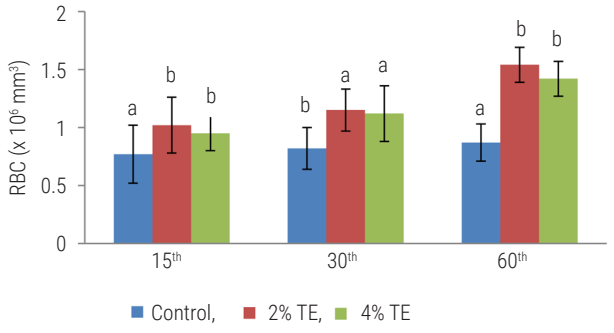

Fig. 4. RBC levels in TE-fed *L. gonius*

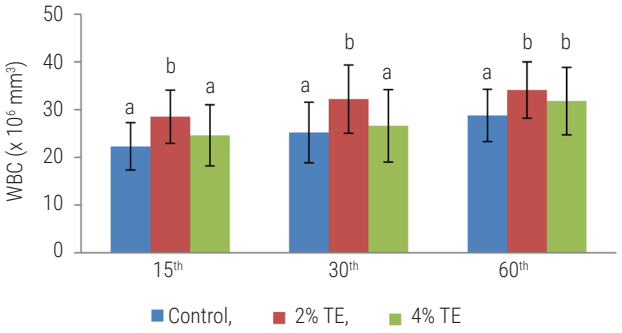

Fig. 5. WBC levels in TE-fed *L. gonius*

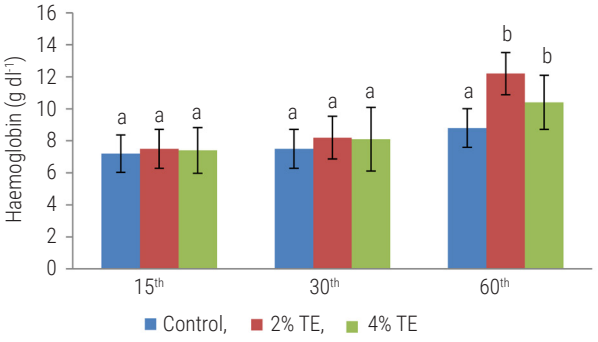

Fig. 6. Hb levels in TE-fed *L. gonius*

stress tolerance capacity and overall health of the fish after feeding them with TE (Rehulka, 2000). Similarly, the observed increase in WBC count in turmeric-fed fish suggests a strengthening of their immune system, potentially aiding in their survival and overall health in the challenging environments (Menezes *et al*., 2006).

Enzymatic parameters

The activity levels of GOT and GPT are commonly used indicators of physiological stress and liver health in animals (Udayakumar *et al*., 2009). In our study, we observed a significant decrease in the activity of GOT and GPT in fish fed diets containing turmeric extract (TE) throughout the sampling period. The average activity of GOT in fish fed the control diet without TE was 61 IU l^{-1}, whereas those fed diets with 2 and 4% TE exhibited GOT activity levels ranging from 46.2 to 51.1 IU l^{-1} (Fig. 7). Similarly, the mean activity of GPT in fish fed the control diet was 16.3 IU l^{-1}, while those fed diets with TE showed levels ranging from 14.1 to 14.7 IU l^{-1} (Fig. 8).

These findings suggest that the inclusion of TE in the diet helped maintain normal health conditions for the fish. The observed decrease in the activity of these enzymes in TE-fed fish indicated amelioration in physiological stress and liver injury, potentially contributing to their overall well-being (Udayakumar *et al*., 2009).

Biochemical parameters

The biochemical parameters assessed in this study, including glucose, total protein and total cholesterol levels, showed significant improvements in fish fed diets TE. Fish fed the control diet without TE had an average glucose level of 91.2 mg dl^{-1}, while those fed

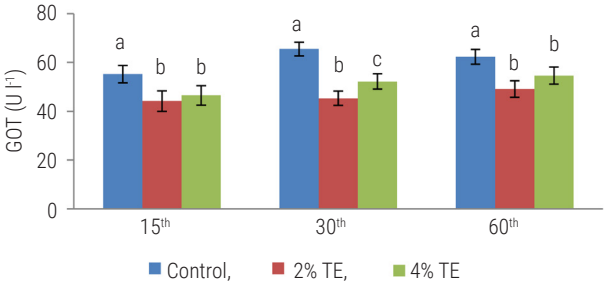

Fig. 7. GOT activity in TE-fed fish

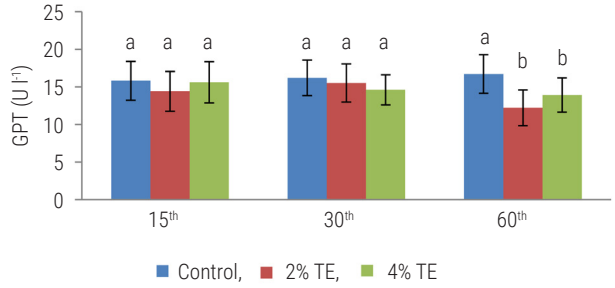

Fig. 8. GPT activity in TE-fed fish

diets with 2 and 4% TE exhibited glucose levels ranging from 77.5 to 80.3 mg dl^{-1} (Fig. 9). Total protein levels were estimated to be 7.4 g dl^{-1} in fish fed the control diet, while those fed diets with TE showed levels of approximately 8.5 g dl^{-1} (Fig. 10). The total cholesterol levels were 174.9 mg dl^{-1} in fish fed the control diet, compared to 152.1-160.7 mg dl^{-1} in turmeric-fed fish (Fig. 11).

These findings are consistent with previous studies that have reported improvements in biochemical parameters in fish supplemented with herbal diets (Yogeshwari *et al*., 2015). The observed improvements in biochemical attributes also suggest an enhancement of the fish's non-specific immune system, increased resistance to pathogenic infections and improved osmotic pressure for better distribution of body fluids and essential compounds, such as steroid hormones, haemin and fatty acids, between intravascular compartments and body tissues (Asadi *et al*., 2012).

It is essential to note that while excessive turmeric supplementation can have adverse effects on animals, including the potential to disrupt glucose metabolism (Chowdhury *et al.,* 2021), in the present study, no significant fluctuations were observed in blood glucose

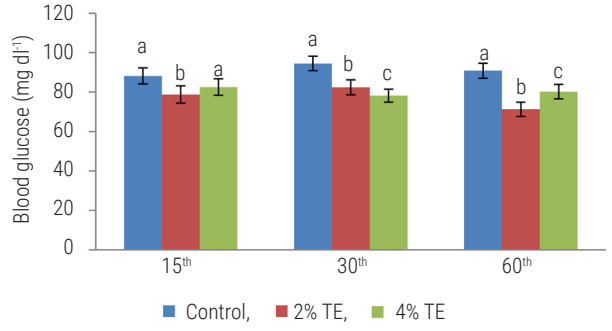

Fig. 9. Glucose level in TE-fed fish

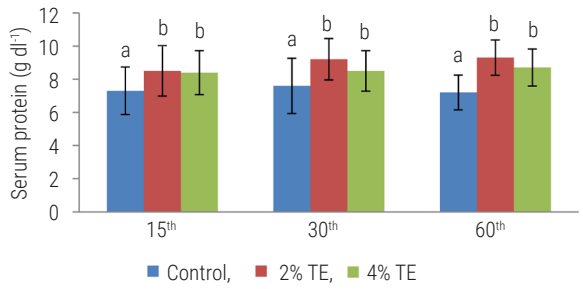

Fig. 10. Total protein level in TE-fed fish

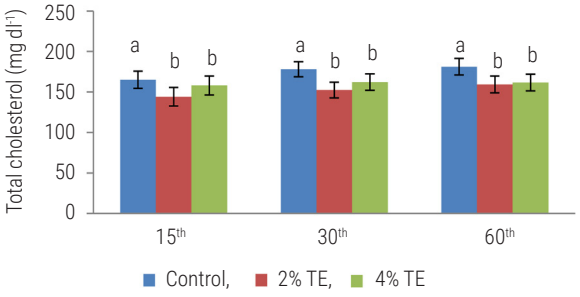

Fig. 11. Total cholesterol level in TE-fed fish

levels across any of the treatments with TE. The glucose levels remained within the typical range (40 to 90 mg dl^{-1}) observed in fish (Malini *et al*., 2018). This suggests that the doses of TE used in this study, specifically 2 and 4% in the feed, were within the safe threshold for fish.

Optimal inclusion level and feasibility

Based on the observed growth, survival, biochemical and haematological responses in this study, the inclusion of 2% TE in the fish diet provided optimal outcomes for *L. gonius*. The 2% inclusion level proved more effective than the 4% level in enhancing various parameters, including haematological, enzymatic and biochemical indices.

Regarding the feasibility of using 2% TE as a feed additive, it is important to consider the potential cost implications. While the inclusion of turmeric extract at a 2% level may seem relatively high compared to the small quantities typically used in human diets, it is within the range of turmeric inclusion levels (50 mg to 40 g kg^{-1}) that have been explored in previous studies on various fish species without any detrimental effects (Sahu *et al*., 2008; Mahmoud *et al*., 2017; Giri *et al*., 2019; Ashry *et al*., 2021). The optimal dosage may vary depending on factors such as fish species, feeding habits, fish size, duration of the study and farming conditions.

We acknowledge that our experimental feeds' fish meal content (14%) exceeds typical commercial levels for freshwater species. This decision ensured adequate nutrition for *L. gonius*, whose dietary requirements under varying microclimates are not well-established. Our inclusion rate, while high, aligns with other carp studies: Mondol *et al.* (2011) used 20-40% for *L. bata* and Chiu *et al.* (2013) reported 0-30% for carps. However, we recognise commercial feeds aim

for lower rates to optimise cost-effectiveness and environmental sustainability. Future research should explore reducing fish meal levels while maintaining optimal growth and health outcomes in *L. gonius*. Strategies could include partial replacement with plant-based protein sources or supplementation with essential amino acids. These approaches would align more closely with sustainable aquaculture feed formulation trends while potentially preserving the beneficial effects observed in this study.

In summary, the application of 2% turmeric extract, derived from a locally developed variety (Megha Turmeric-1) by the ICAR Complex for NEH region in Meghalaya, has proven to be advantageous in enhancing the growth, condition and health of *L. gonius* reared in the hill environment of Meghalaya. However, a comprehensive cost-benefit analysis is recommended to evaluate the overall economic viability of incorporating this feed additive into commercial aquaculture operations. This analysis should take into account the potential increase in feed costs associated with the addition of turmeric extract, as well as the potential benefits in terms of improved growth, survival and overall health of the fish. It is important to be cautious when utilising turmeric and similar products, which are valuable food resources for humans. Further research is necessary to ascertain the specific advantages and optimal utilisation of turmeric extract in aquaculture systems, taking into consideration factors such as fish species, farming conditions, duration of the study and potential cost implications.

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