Efficacy of garlic supplementation in Canola meal based diets on growth performance and body composition of *Catla catla* fingerlings

Mawra Rafique[1], Hassan Usman[1], Maryam Hamid[1], Zunnorain Ahmed[2], Muhammad Imran[1], Zeeshan Khan[3], Raheela[1], Sher Hameed[3], Atija Waris[4], Saddam Hussain*[3]

[1]*Department of Zoology, Faculty of Life Sciences, Government College University Faisalabad, Faisalabad, Punjab, Pakistan*

[2] *Department of Zoology, Faculty of Sciences, University of Sargodha, Sargodha, Punjab, Pakistan*

[3]*Department of Zoology, Faculty of Basic Sciences, University of Agriculture, Dera Ismail Khan, KPK, Pakistan*

[4]*Discipline of Zoology, Institute of Molecular Biology & Bio technology, The University of Lahore, Lahore, Punjab, Pakistan*

***Corresponding Author:** Dr. Saddam Hussain

Abstract

The goal of the current research was to determine the efficacy of diets enriched with garlic powder, garlic peels and mixture in canola meal-based diets on growth performance and body composition of *Catla catla* fingerlings. Garlic powder, garlic peels and mixture was added in the diet at the levels of 0%, 1.5%, 3% and 4.5%. The 0% was considered as control group. The experiment was conducted in triplicate. Fingerlings were fed 5% of their live wet body weight. Seventy days were spent on the experiment. The data was subjected to two-way Analysis of Variance (ANOVA). The results of this research revealed that, compared to the control group, a canola meal-based diet supplemented with garlic powder, garlic peels and mixture enhanced *C. catla* growth performance and body composition, and in all diets, T 3, T 7 and T 11 levels demonstrated superior growth performance and body composition. If we compare T 3, T 7 and T 11 of these diets garlic powder, garlic peels and mixture showed better growth higher SGR, low FCR.

Keywords: *Catla catla*, Meat quality, Growth performance, Garlic powder, garlic peels and mixture

1. Introduction

Labeo rohita, frequently recognized as raho, C. catla, also known as thaila, and *Cirrhinus mrigala*, commonly referred to as mori, are species of Indian carps that are prevalent in both the public and administrative sectors in Pakistan (FAO, 2017). Catla, also known as Thaila, is a benthivorous fish that is commonly found in polyculture systems in Pakistan, where it is farmed with other species (Aslam et al., 2016). The production of Catla has increased significantly throughout the first decade of the 21st century, reaching over 2.8 million tons per year in 2012 (FAO, 2015).

The exponential growth of the global human population has resulted in significant challenges regarding nutrition, particularly in terms of food security on a global scale (FAO, 2018). The current rapid growth of aquaculture is driven by the need for high-quality fish protein (FAO, 2014). The increased expansion of aquaculture has led to a significant need for the improvement of feed quality, both for complete and supplementary feeding in tanks and ponds (Hernandez et al., 2012). *C. catla* holds significant importance among all aquaculture fishes due to its contribution to low-cost and highly nutritious protein sources for human consumption (Khan et al., 2012). It is rich in high-quality nutrients, including vitamins, a favorable amino acid profile, minerals, and fatty acids (Zhou et al., 2004, Dawood et al., 2015). Nevertheless, the exorbitant expenses, increasing need, and diminishing availability of fish meal (FM) necessitate the search for other protein sources (Hardy, 2010, FAO, 2014). These alternatives should be affordable and enhance the quality of protein (Lim et al., 2011). Several researchers have suggested using other sources of higher-quality proteins to decrease the use of FM. (Shahzad et al., 2017, Shahzad et al., 2018, Hussain et al., 2019). Plant byproducts have been widely recognized as a cost-effective and readily available alternative to fish meal in fish diets by numerous researchers over the past two decades (Wang et al., 2015, Hussain et al., 2018, Hussain et al., 2019). The study also investigated the positive impacts of plant meal on the growth-performance (GP) of fish (Hussain et al., 2011).

Garlic has the ability to enhance fish survival and provide protection against parasites, germs, and fungi. It possesses immunostimulatory properties and is a valuable medicinal herb. Garlic enhances growth and fortifies the immune system in fish aquaculture. Garlic contains organosulfur compounds, such as allicin, which are believed to have an impact. Supplementing feed with garlic extract will enhance the body's immune system. Allicin content can enhance non-specific immunity by increasing the production of cytokine genes (Setijaningsihet al., 2021).

Allium sativum possesses a diverse range of bioactive compounds that confer advantageous effects on animal well-being. Ajoene, alliin, and allicin are three compounds that possess various biological properties such as growth stimulant, antibacterial, antiviral, antioxidant, and antiparasitic activities. Garlic has multiple applications in aquaculture, such as its use in the form of oil, fresh mash, aqueous extract, and garlic powder. The powder form is the predominant presentation in aquaculture, typically given orally and incorporated into the feed. Garlic has been employed in aquaculture for an extended period. Its popularity has increased because to its affordability, convenience of use, minimal environmental impact, and more advantages. Consequently, its utilization could be a potent method to combat infections, enhance the well-being of organisms, and function as a substitute for antibiotics (Valenzuela et al., 2021).

Allicin (diallyl thiosulfate), which is released when the garlic clove is crushed, is one of the distinctive organosulfur compounds that are thought to be responsible for the principal biological effects of garlic, *A. sativum*. It possesses a wide range of antibacterial, antiviral, antiprotozoal, and antifungal properties. Allicin activates natural killer cells, complement, lysozyme, and antibody responses in addition to facilitating phagocytic cell function and increasing their bactericidal capabilities. Increased defense against infectious disease is linked to the stimulation of certain immune processes. Additionally, allicin can affect infections directly by preventing the formation of RNA, DNA, and proteins (Breyer et al., 2015).

2. Material and methodology

The purpose of the present study was to examine the enhancement in the overall performance of C. catla fingerlings, with specific attention to growth performance (GP) and the composition of fish carcass, when they were fed a diet enhanced with canola meal and garlic by-products. The trial was conducted in the Zoology Department of GCUF. Young fish of the species C. catla were obtained from a nearby Hatchery and were acclimated to the experimental settings for a period of two weeks. Specifically designed water tanks with a water capacity of 70 liters were utilized for the purpose of housing fingerlings. During the acclimation period, the fingerlings were given the basal diet once daily (Allan and Rowland, 1992). Water quality indicators such as pH, temperature, and dissolved oxygen (DO) were monitored on a daily basis. Oxygen was continuously supplied through a capillary system utilizing an air pump. Fingerlings of C. catla

were treated with a saline solution of 0.5% concentration to address the potential presence of microbes.

2.1. Experimental design

One control and nine experimental diets were formulated, divided into three major diet groups with diet I (GP), diet II (GPP) and diet III (Mixture). Fingerlings were distributed in tanks. Each tank contains 15 fingerlings. Total duration of the experimental period was 70 days. These test diets were then compared with control diet and with each other to assess results.

2.2. Processing of Moringa by-products and formation of feed pellets

The garlic was dried for a period of six days in a shaded area to prevent any harm to the organosulfur compounds. The cloves were extracted from the dehydrated peels. Garlic powder (GP) and garlic peel powder (GPP) were created. The composition of other feed materials, obtained from a feed mill, was analyzed for their composition according to AOAC (1995) techniques prior to formulating the diet. The results are shown in Table 1. The materials were pulverized to a particle size of 0.3 mm using a sieve and then combined at the correct concentration. Distilled water (DW) was added to create a suitable consistency dough for making pellets, as described by Lovell (1989). The pellets were placed in an oven set at a temperature of 55°C. After the diets were dried, they were stored at a temperature of 4 °C until they were ready to be used.

Table 1. ingredients composition (%) of garlic supplemented diets

Ingredients	**Control group**	**Test diet-I**	**Test diet-II**	**Test diet-III**	**Test diet-IV**	**Test diet-V**	**Test diet-VI**	**Test diet-VII**	**Test diet-VIII**	**Test diet-IX**
Garlic powder (%)	0	1.5%	3%	4.5%	0	0	0	0	0	0
Garlic peel powder (%)	0	0	0	0	1.5%	3%	4.5%	0	0	0
Mixture (%)	0	0	0	0	0	0	0	1.5%	3%	4.5%
Canola Meal	52	52	52	52	52	52	52	52	52	52
Fish meal	10	10	10	10	10	10	10	10	10	10

Wheat flour	20	18.5	17	20	18.5	17	20	18.5	17	20
Rice polish	8	8	8	8	8	8	8	8	8	8
Fish oil	7	7	7	7	7	7	7	7	7	7
Vitamin Premix	1	1	1	1	1	1	1	1	1	1
Minerals premix	1	1	1	1	1	1	1	1	1	1
Ascorbic acid	1	1	1	1	1	1	1	1	1	1

2.3 Growth Study

At the conclusion of the trial, fish in every tank were mass weighed to calculate final weight. The growth performance of *C. catla* was assessed using common equations.

$$\text{Weight gain \%} = \frac{\text{(Final weight – Initial weight)}}{\text{Initial weight}} \times 100$$

$$\text{SGR (\%/day)} = \frac{\text{(ln. final wt. of fish – ln. initial wt. of fish)}}{\text{Trial day}} \times 100$$

$$\text{FCR} = \frac{\text{Total dry feed intake (g)}}{\text{Wet weight gain (g)}}$$

2.4. Chemical analysis of feed and carcass

Subsequently, a feeding study lasting 90 days was conducted to assess the moisture contents and carcass of the fish. This was done by subjecting the samples to oven drying at a temperature of 105°C for approximately 12 hours. The analysis of EE, or crude fat, was conducted using the Soxhlet system, while the analysis of CP, or crude protein, was conducted using the Micro Kjeldahl Apparatus, both following approved techniques. The analysis of crude fibers (CF) was conducted using the loss on ignition method, which involved digesting dried defatted samples with a solution containing 1.25% H2SO4 and 1.25% NaOH.

2.5. Statistical Analysis

For estimation data of growth and body composition was subjected into two way analysis of variance (ANOVA) (Steel et al., 1996). By turkey's honesty significant difference test, the

difference among means was matched and considered significant at $p<0.05$ (snedecor & Ccochran, 1991). The data was analyzed by using software SPSS.

3. Results

The results of *C. catla* fingerlings' growth parameters on canola based diet supplemented with garlic powder (0%, 1.5%, 3% and 4.5%) are shown in (table 4.1). Significant improvements in growth performance of *C. catla* fingerling were seen at the T_3 diet level when canola was supplemented with 3% garlic powder. Significant improvements in *C. catla* fingerlings growth were seen at the T_6 diet level when canola was supplemented with 1.5% garlic peel powder. Significant improvements in *C. catla* fingerlings growth were seen at the T_{11} diet level when canola was supplemented with 3% mixture. At this level and all other levels given in the table, there was a significantly ($p<0.05$) enhanced weight gain percentage of (243.08±1.78), lowest FCR (1.30±0.02) was noted, while SGR (1.37±0.00) was highest.

Table 2. Growth performances of *C. catla* fingerlings fed on garlic powder, peel powder and mixture supplemented diet

Treatments	Supplementary Diets	Supplement level	Initial weight (g)	Final weight (g)	Weight Gain (g)	Weight Gain (%)
T_1	Diet-I GP	0	6.13±0.02^{a}	13.13±0.02^{cd}	6.99±0.01^{d}	114.07±0.54^{c}
T_2		1.5%	6.13±0.01^{a}	17.48±0.01^{b}	11.35±0.01^{b}	185.20±0.50^{d}
T_3		3%	6.13±0.03^{a}	19.48±0.15^{a}	13.34±0.18^{a}	217.50±4.00^{b}
T_4		4.5%	6.12±0.01^{a}	14.03±0.01^{c}	7.91±0.05^{d}	129.17±0.32^{a}
T_5	Diet-II GPP	0	6.13±0.02^{a}	10.1±0.08^{d}	4.03±0.10^{d}	65.71±1.92^{d}

T_6		1.5%	6.13±0.01[a]	18.07±0.03[a]	11.94±0.05[b]	194.57±1.82[b]
T_7		3%	6.13±0.03[a]	16.23±0.03[b]	10.10±0.04[b]	164.87±0.92[b]
T_8		4.5%	6.12±0.01[a]	13.13±0.02[cd]	7.00±0.03[d]	114.43±0.22[d]
T_9	Diet-III Mixture	0	6.13±0.02[a]	14.64±0.03[c]	8.51±0.05[c]	138.75±1.28[c]
T_{10}		1.5%	6.13±0.01[a]	18.98±0.01[a]	12.85±0.02[a]	209.68±0.54[a]
T_{11}		3%	6.14±0.03[a]	21.05±0.01[a]	14.92±0.04[a]	243.08±1.78[a]
T_{12}		4.5%	6.12±0.02[a]	16.70±0.06[b]	10.58±0.06[b]	172.73±0.96[b]

Results of body composition of *C. catla* fingerlings fed on GP supplemented canola based diets at different levels (0, 1.5, 3, and 4.5%) is presented in tables 4.5 and 4.6, respectively. The body of *C. catla* fingerlings showed the highest CP at 3% feeding level of GP supplemented canola based diets (17.54±0.02). CP at 3% level GP-based was significantly (p<0.05) different from all other 1.5 and 4.5% of GP supplemented canola based and also clearly different from 0% (control) level of diet in *C. catla* fingerlings. Additionally, it was noted that *C. catla* fingerlings had minimal CP levels at the highest GP level of 4.5%. At 3% feeding level of GPP supplemented with canola based diets, *C. catla* fingerlings had the highest CP (18.23±0.01). In *C. catla* fingerlings, the quantity of crude protein at 3% GPP supplemented with canola based was significantly (p<0.05) different from all other 1.5 and 4.5% of GPP supplemented with canola based and also noticeably different from 0% (control) level of diet. Additionally, it was observed that at the maximum GPP dosage of 4.5%, the crude protein levels in *C. catla* fingerlings were quite low. When 3% of GPP supplemented with canola based meals were added, maximum CF was observed.

Table 3. Body composition of *C.catla* fingerlings fed on garlic powder, peel powder and mixture supplemented diets

Treatments	Supplementary Diets	Supplement levels	Protein	Fat	Ash	Moisture
T_1	Diet-I GP	0	12.58±0.01^{d}	5.47±0.02^{a}	3.83±0.02^{a}	78.11±0.01^{a}
T_2		1.5%	16.96±0.03^{b}	3.19±0.05^{d}	3.15±0.50^{c}	76.68±0.56^{c}
T_3		3%	17.93±0.01^{a}	3.54±0.04^{c}	3.05±0.03^{c}	75.47±0.08^{d}
T_4		4.5%	14.98±0.01^{c}	4.02±0.01^{b}	3.53±0.00^{b}	77.46±0.02^{b}
T_5	Diet-II GPP	0	12.38±0.01^{d}	5.57±0.02^{a}	4.03±0.02^{a}	78.01±0.01^{a}
T_6		1.5%	16.83±0.04^{b}	3.42±0.01^{c}	2.77±0.03^{c}	76.96±0.05^{c}
T_7		3%	17.54±0.02^{a}	3.97±0.01^{c}	2.98±0.01^{c}	75.50±0.03^{d}
T_8		4.5%	14.58±0.01^{c}	4.32±0.01^{b}	3.73±0.04^{a}	77.36±0.04^{b}
T_9	Diet-III Mixture	0	12.74±0.01^{d}	5.27±0.02^{a}	3.93±0.02^{a}	78.056±0.02^{a}
T_{10}		1.5%	17.22±0.01^{a}	3.01±0.01^{d}	2.92±0.27^{c}	76.84±0.29^{c}
T_{11}		3%	18.23±0.01^{a}	3.17±0.01^{d}	3.12±0.095^{b}	75.47±0.08^{d}
T_{12}		4.5%	15.08±0.01^{c}	4.12±0.01^{b}	3.36±0.05^{b}	77.16±0.05^{b}

4. Discussion

Effect of fish feed fortification with garlic powder, garlic peel powder and mixture on growth performance and carcass composition of *C. catla* fingerlings fed on canola meal based diets

In order to meet the demands of growing aquaculture industry, importance has been given on the formulation of commercial diets using affordable and important nutritionally balanced ingredients. To overcome the higher prices of fish feed, plant derived ingredients could be used which will be helpful in increasing fish production and reducing the cost of feed. Several researches supported the fact that plant ingredients can be incorporated into fish feed partially or completely. Fishes fed with plant feed showed both positive and negative results. One of the earliest known medicinal herbs is garlic (*A. sativum*). Plants and their extracts can serve as immune system boosters, offering a greener and more natural option to combat infectious diseases, as opposed to relying on medicines and synthetic compounds. Prior research has shown that plants and their derivatives possess immunostimulant and health-promoting qualities. They can also act as growth enhancers, appetite stimulators, antimicrobials, anti-inflammatory agents, anti-tumoral

agents, and improve the efficiency of nutrition utilization and physiological condition. (Valenzuela et al., 2021).

Outcomes of present research evaluated that 3% garlic powder supplementation level of diet is an ideal level for better growth performance. The maximum weight gain of fish was observed in group fed with diet-III at 3% level when compared with control and other 2 groups. The second superlative results were obtained when fingerlings were given diet-I at same level (3%). The significant increase were observed in fish fed with 2.0% GPE, African catfish, *C. gariepinus* fingerlings (Gabriel et al., 2019). The effects of dietary GP on *P. leptodactylus* growth rate, physiological function, innate immunity, and antioxidant capacity, especially at 2% (Rezaei et al., 2022). The results of Chowdhury et al. (2021) study indicates that combining turmeric with either garlic or garlic in an equal ratio at a 1% inclusion level may help to promote development, digestion, and metabolic functions. The findings indicated that adding garlic powder to the feed had a significant impact on both absolute growth and daily growth rate, with the addition of 1% showing the highest values for weight gain (17.75±0.48 g) and daily growth rate (0.42±0.01 g/day) (Setijaningsih et al., 2021). This is because of effects to allicin at higher dosages is known to be harmful to fish, resulting in slow growth and even death in fish.

In this study the lowest feed intake was observed at higher garlic powder levels which mean that highest levels of garlic powder in feed affects the palatability of fish and reduces the fish feed intake. The maximum feed intake was recorded at 3% while lowest feed intake was observed when fed at 4.5%. The fingerlings at 3% combination diet showed highest feed intake, indicating that lower and best value of FCR is also recorded at this level. The outcomes of present investigation are nearly similar to the outcomes of Patel et al., 2022 which revealed that FCE and PER were increased with increasing level of GP up to GP 2% then slightly decreased in GP 3%. While in case of FCR inverse trends of decreased FCR with increasing level of GP up to 2% was observed then slightly increased. Similarly, pattern in *O. niloticus* was observed by Shalaby et al. (2006). In accordance to present study significant in *O. mykiss* (Adinehet al., 2020) fed with dietary garlic.

The findings from the analysis of whole-body composition revealed a substantial rise in the levels of body crude protein and ash content when the diet consisted of 3%. Conversely, the levels of moisture and crude fat were higher across all groups. This finding was consistent with the study conducted by Nyadjeu et al., 2021, which demonstrated that garlic has the ability to

enhance crude protein levels and reduce crude fat levels. Among the several treatments applied to African catfish, T3 exhibited the most significant effects. The combination diet with the lowest quantity of garlic resulted in the highest crude protein and crude fat content, indicating that lower doses of garlic yield superior outcomes. Deng et al. (2011) found that the higher levels of fat and protein in fish bodies may be the cause of the enhanced flavonoid content in plant extracts, which in turn enhances the intake, absorption, and processing of nutrients. Furthermore, Liang et al. (2012) found that an elevation in garlic consumption resulted in an augmentation of both fat and protein levels in the body of grass carp. An increase in fat and moisture content was seen in A. satium at a concentration of 1.5%, whereas there was a decrease in ash and fiber content. These findings align with the results reported by Zhong et al. (2012).

The study investigated the effects of varying amounts of garlic powder, garlic peels powder, and a mixture on growth parameters and the overall body proximate composition of garlic powder. In this study, fingerlings that were given a meal containing a 3% concentration of garlic powder showed the highest levels of crude protein and moisture. These levels were similar to the findings of Akbary et al.'s 2016 examination of the carcass composition of brown trout. The inclusion of 30 grams per kilogram of garlic in the diet resulted in a notable augmentation of body protein. The diet containing 10 g/kg garlic had increased moisture content, while the diet containing 20 g/kg garlic had higher ash content. Nevertheless, there were no notable disparities seen in body fat. In contrast to our findings, Shalaby et al. (2006) reported that the fish species (A. ruthensis) had the lowest fat and maximum protein content in their complete body when fed a diet with 30 g of garlic powder per kilogram. Farahi et al. (2010) found that including 1%, 2%, and 3% garlic into the meals of rainbow trout resulted in notable variations in the levels of crude protein, fat, and ash content in the fish's body, as compared to the control group. The increased ash content in the bodies of fish in the group that was fed a diet supplemented with 1.5% garlic can be explained by the continuous availability of food, which allows aquatic organisms to absorb minerals and nutrients (Samadi, 2012).

Conclusions

The effects of garlic powder, garlic peel and mixture supplementation on growth performance and evaluation of C. catla fingerlings meat quality enhancement by body composition were observed. Garlic powder, garlic peel and mixture were given to the fingerlings in amounts of 0%, 1.5%, 3%, and 4.5%, respectively. 0% was used as the control group. When fingerlings were

given the T 3 test diet (a canola meal based diet supplemented with garlic powder, garlic peel and mixture), C. catla had the greatest growth metrics, including weight increase, weight gain%, SGR and minimum FCR, and if we compare these (garlic powder, garlic peel and mixture) diet-III the T 11 level of garlic powder and mixture best growth performance and highest body composition and T 6 level of garlic peel powder had best growth performance and highest body composition.

Conflict of Interest

All the authors declare that they have no any conflict of interest.

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