Research Article

Effect of black soldier fly larvae-based diet on bone mineralization, fatty acids and amino acids composition of common carp *(Cyprinus carpio)* fingerling

Ifra Sarwar[1*], Shazia Yaseen[1], Amber Sarfraz[1], Fazila Munir[2], Aqib Jahangir[1], Ansa Noreen[1], Shan Zaib[3], Muhammad Luqman[1], Yasir Nawaz[1], Muhammad Hammad Noor[4], Muhammad Sajjad Sarwar[1]

[1*]Department of Zoology, Faculty of Life Sciences, University of Okara, Okara, Pakistan
[2]Department of Zoology, Fisheries and Aquaculture, University of Okara, Okara Pakistan
[3]Department of Zoology, Abdul Wali Khan University, Mardan, Mardan 23200, Pakistan
[4]Department of Fisheries and Aquaculture, University of Veterinary and Animal Sciences, Lahore, Pakistan

***Corresponding author's email:** ifrasarwar997@gmail.com

Abstract

Black soldier fly larvae meal-based diet is an innovative method for sustainable aquaculture. Black soldier fly larvae meal has shown a promising alternative as a fish meal replacement in diets for common carp fingerlings. This study was aimed to assess the effect of dietary black soldier fly larvae on bone mineralization, fatty acids and amino acids composition of fingerlings. About three feeds were formulated where fish meal replaced by BSFL for nursing of fingerlings. Feeds were prepared for fish. After 60 days of trial, samples of muscles and bones were collected for fatty acid composition, amino acid composition and bone mineralization. The results showed that fatty acid composition in muscles was closely related with diets. Fatty acids such as cis-11, 14 Eicosadienoic (0.86±0.14) and Docosahexaenoic (1.48±0.25) showed higher significant difference. While, Docosapentanoic acid (0.82±0.12) and, Nervonic (0.305±0.07) showed lower significant difference in fish muscles. Amino acids such as serine (4.33±0.55) and aspartic acid (6.375±0.33) were more abundant and showed higher statistical difference. While, lysine (8.02±0.16) and cysteine (4.33±0.55) showed lower statistical difference. Three amino acids such as tryptophan, asparagine and glutamine were absent in common carp muscles. Mineral such as phosphorous (50.5±3.535) were more abundant and showed higher statistical difference in bones of fingerlings while the value of calcium (164±1.412) showed lower significant difference. To conclude the insect-based meal made from BSFL is a nutritionally appropriate source of protein, fat and minerals for common carp fingerlings without having negative impact on growth and feed utilization.

Keywords: Black soldier fly larvae, Fish meal, Common carp fingerlings, Amino acid composition, Bone mineralization, Fatty acid.

Article History: **Received:** 27 Mar 2025, **Revised:** 5 May 2025, **Accepted**: 20 Jun 2025, **Published:** 19 Sep 2025.

Introduction

Aquaculture is the fastest growing industry like other parts of the world [1 - 3]. Due to the rapid increase of world population, a large number of fish feed is required to hold the industry [4, 5]. Fish feed is responsible for nearly 70% of the world aquaculture productivity according to FAO [5, 6]. However, with the passage of time world population and meat consumption is suddenly increasing [7]. So, 70% more food must be produced globally to meet the growing population needs [8]. Fish meals or raw wild fish are frequently offered to cultured fish as a source of animal protein [9, 10]. Fish meals are an essential component in aquatic feeds because of their highly digested protein amino acids and also being palatable [11, 12]. However, the cost of aquaculture production increased due to the rising demand for fishmeal and unbalanced output of fishmeal [12]. Along with essential nutrients, fish meals also contain significant amounts of fatty acids [13], amino acids [14], and some of the most important minerals and vitamins [15]. Fatty acids considered as major component of fish muscle which is often found in amounts between 6 and 20 percent [16] and mostly found in mesenteric and muscle tissue, liver subcutaneous tissue as well as brain [17 -19]. The essential components of the mineralized fish skeletal system are calcium [20, 21] and phosphorus [22]. Another crucial element in the structural makeup of bone is protein [23]. With the passage of time the prices of fish meals have been increasing due to more consumption [14, 24]. Fish meals are a big challenge for the aquaculture industry as it represents about 60 to 70% cost of fish feed production [25]. The rapid pace of development and population growth is anticipated to cause a shortage of conventional feed ingredients used in aquaculture, thereby driving up demand for all types of feed resources [26]. In response, aquaculture practices focus on minimizing production costs, especially those related to nutrition while avoiding adverse impacts. Consequently, commercial fish feeds play a critical role in aquaculture, as they are specifically formulated to support optimal growth in target fish species. To ensure adequate amino acid and fatty acid profiles, these feeds commonly include fishmeal and fish oil, along with maize grain and soybean meal [27].

Insects represent the most diverse group of animals globally, with an estimated one million species [28]. Both carnivorous and omnivorous fish, as well as agricultural animals, naturally consume insects [29]. Due to their rich nutritional profile, various insect species play a crucial role in fish diets, especially during the larval and fingerling stages [30]. It has been reported that insect protein is more readily digestible than plant protein and is absorbed at rates comparable to animal protein [31]. The nutrient availability and digestibility of feed ingredients remain among the most critical factors influencing fish nutrition [32]. One of the newest methods being used is growing BSF larvae for fish feed which has the added benefit of making use of biological waste [33, 34] and offers a possible choice for the safe management of manure both human and animal [35]. The BSF larvae are incredibly nutrient-dense [36]. BSF larvae contain 30-35% fatty acids 40-45% protein 11-15% ash 0.6% p and 4.8-5.1% ca on a DM (Dry matter) basis and a variety of minerals and amino acids [35, 37]. Larvae of BSF possess large concentrations of omega-3 fatty acids which can improve fish health and yield [38, 39]. BSF larvae have been successfully introduced to domestic animals like pigs and poultry as well as a variety of fish like common carp and rainbow trout [40, 41]. Larvae of Black soldier fly can be cultivated on waste products like home-based food wastes and manure allowing wastes to be reprocessed into a useful insect protein [42, 43]. The raising BSF larvae on manure and other wastes is a value-added management technique which turns waste stuff into beneficial and marketable feed for animals [42] which also decreases harmful bacteria in swine chicken excrement [44, 45] as well as managing domestic houseflies (*Musca domestica*) in cattle facilities [46, 47]. Furthermore, BSFL culture does not need a lot of effort. Before becoming pupae BSF larvae travel from the trash pit to the collection chamber. Since adult BSF do not have functional mouth parts they do not interact with people and do not spread disease [48, 49]. The aim of this study was to provide preliminary data to estimate the impact of BSFL diet on the composition of fatty acid, amino acid and bone mineralization of common carp fingerlings.

Material and Methods

Procurement of fish

From the local fish seed hatchery about 20 Fingerlings of Common carp were taken. Some of had died during the experimentation, the left three were used for sixty days and kept in an aquarium and is fed a basic diet. The pH and electrical conductivity meters were used to track the parameters of the water quality.

Ethical approval

This study was conducted according to the declarations of Helsinki following the rules for animal study. The approval was obtained from the university Institutional Review Board to complete the study.

Experimental design

Experiment was conducted to examine the bone mineralization, fatty acids and amino acids composition of common carp fingerlings. In the laboratory trial were conducted in the triplet set of aquariums. First aquarium was named as controlled aquarium and other two aquariums were experimental aquarium 15 fingerlings were introduced in each aquarium. The control group provided standard fish food. In second aquarium fingerlings were provided with 20% BSF and third aquarium were provided with 40% BSF larvae-based fish food.

Research tools

Three aquariums were used to culture fish Heater generator/aerator filtration stones bed gravel lights and hood formulated feed dissection box and chemicals such as formalin chloroform NaCl as well as Charcoal were the basic requirements.

Sample collection

In order to ascertain the composition of amino acids, fatty acids, and bone mineralization samples were gathered during the final experimental trial with three fish randomly chosen from each group. Fish were anesthetized with chloroform for this purpose and a sharp blade was used to dissect the fish.

Analysis of bone mineralization

After dissection of fish samples of bone were collected from each aquarium. The bones were properly cleansed free of soft muscles and soaking for five minutes in hot (50°C) deionized water bones were ground into a fine powder prior to freeze drying placed on platinum plates and baked at 480°C for 48 hours. The materials were assayed and then digested in 6N HCl prior to minerals determination. The minerals (Ca, Mg, Zn, Mn, and Cu) were analyzed using an atomic absorption (AA) device (Perkin Elmer 300, Ueberlingen, Germany). Phosphorous was analyzed through spectrophotometry. Additionally mineral analysis of fish bone was performed using AOAC (1990) method.

Amino acid analysis

To determine the amino acid composition, muscle protein was hydrolyzed for 24 hours in an aerobic environment using 6N at 110°C. The hydrolyzed samples were neutralized with 6N NaOH and then derivatized using a kit (AccQ-FluorReagent, WAT052880, water). The derivatized samples were put into a high-performance liquid chromatography (HPLC) system with a fluorescence detector (2475, Water) and a C18 RP column. By comparing retention durations and peak regions of standards the amino acids were recognized and measured (WAT088122, Water). Minced flesh was digested for 24 hours with 5% (w/v) NaOH before being neutralized to pH 7.0 with 6NHCl for the tryptophan assay. At 530nm, tryptophan concentration was determined by spectrophotometrically.

Analysis of fatty acids

Fatty acid methyl esters (FAME) boron trifluoride in methanol were methylated and transesterified after total lipid extraction from fish muscle tissues to ascertain the fatty acid composition [50] Tissues (0.5g–1.0g) were mechanically homogenized in a 2:1 v/v ratio of chloroform to methanol in order to obtain total lipid [51]. Gas chromatography, equipped with a flame ionization detector and a highly polar fused silica cyanosiloxane column (SP -2380; 30 m length 0.25 mm inner diameter 0.20 µm film thickness) was used to separate the FAME. The temperature was intended to rise from 100°C to 230°C at a split ratio of 1:50 and a rate of 1.5°C/min using nitrogen as the carrier gas. The injector and detector had temperature settings of 250°C and 260°C respectively.

The individual FAME was identified by comparing the retention times with commercially available standards such as the 37 component FAME Mix (Supelco) and PUFA No. 3 from Menhaden Oil (Supelco) [52].

Statistical analysis

One-way ANOVA was used to analyze all the data. The statistical software package Minipad was utilized. $P< 0.05$ was used to determine significance, and all results are shown as mean ±SD.

Results

Fatty acids composition

The value of cis-11, 14 Eicosadienoic (0.86±0.14) were significantly higher in control group (To) of 0% BSFLM and lowest in experimental group (T_1) of 20% BSFLM. In Docosahexaenoic acid (1.48±0.25), the value of experimental group (T_2) of 40% BSFLM were significantly higher and lowest in T_1. While, Docosapentanoic acid (DPA) and Nervonic showed lower significant difference ($P<0.05$) in fish muscles. In this regard the value of Docosapentanoic acid (0.82±0.12) and of Nervonic (0.305±0.07) were significantly low in control group (To) of 0% BSFLM and highest in (T_2) of 40% BSFLM.

Table 1: Comparison of fatty acids content of common carp fingerlings fed on BSFL based diets (0%, 20% and 40%) for sixty days.

Parameters	To (0%)	T_1 (20%)	T_2 (30%)	F-value	P-value
Myristic	2.23±0.11	1.87±0.16	1.15±0.55	2.01	0.28
Pentadecanoic	2.745±0.28	1.76±0.82	2.395±0.36	4.79	0.116
Palmitic	28.53±1.30	31.61±0.55	28.91±0.26	8.09	0.062
Heptadecanoic	2.20±0.60	2.67±0.10	2.51±0.49	0.55	0.625

Parameters	To (0%)	T_1 (20%)	T_2 (30%)	F-value	P-value
Stearic	10.44±1.27	8.85±0.59	8.14±1.08	2.63	0.219
Palmitoleic	6.88±1.52	5.08±1.18	5.27±1.52	0.97	0.472
Oleic	18.71±0.79	20.81±0.86	21.67±1.73	3.19	0.181
Cis-11 Eicosenoic	2.34±0.37	2.37±0.72	2.63±0.18	0.21	0.822
Nervonic	0.305±0.07	0.46±0.06	0.71±0.09	12.55	0.035
Linoleic acid (LA)	14.41±0.87	11.49±0.95	13.03±0.33	7.13	0.073
Cis-11, 14 Eicosadienoic acid	0.86±0.14	0.67±0.44	0.84±0.56	0.11	0.895
Lambda-linolenic acid (GLA)	3.33±0.92	4.88±0.87	4.11±0.37	2.03	0.277
Cis-8,11, 14 Eicosatrienoic acid	1.345±0.31	1.08±0.17	1.565±0.53	0.84	0.514
Arachidonic	1.375±0.26	1.25±0.22	1.25±0.04	0.25	0.796
Eicosapentaenoic acid (EPA)	1.35±0.45	2.15±0.14	2.19±0.24	4.77	0.117
Docosapentanoic acid (DPA)	0.82±0.12	1.24±0.15	1.50±0.07	15.41	0.026
Docosahexaenoic acid (DHA)	1.405±0.04	1.39±0.10	1.48±0.25	0.16	0.855

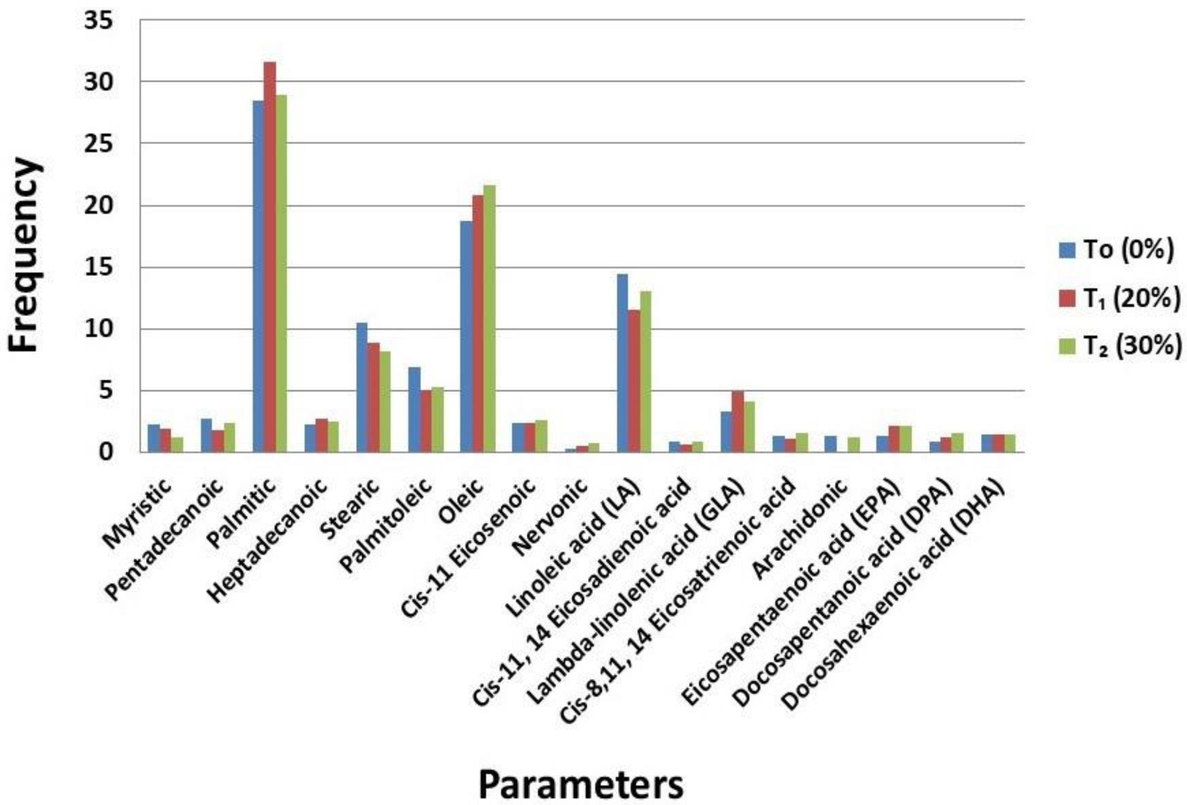

Figure 1: Graph showing Comparison of fatty acids content of common carp fingerlings fed on BSFL based diets (0%, 20% and 40%) for sixty days.

Amino acid composition

The effect of control and experimental BSFL meal-based diets on amino acids composition of common carp fingerlings are presented in Table 2. With the replacement of fish meal by BSFLM, serine and aspartic acid are more abundant and showed higher statistical difference ($P>0.05$) in the muscles of common carp fingerlings. In this regard the value of serine (4.33±0.55) is higher in experimental group (T_2) and lower in (T_1) and the value of aspartic acid (6.375±0.33) is higher in control group (To) and lower in (T_2). While the value of lysine and cysteine showed lower statistical difference ($P<0.05$). The value of lysine (8.02±0.16) and cysteine (4.33±0.55) is higher in experimental group (T_2) and lower in (T_1). The value of tyrosine amino acid is statistically significant ($P\leq0.05$). Three amino acids such as tryptophan, asparagine and glutamine are absent in common carp muscles.

Table 2: Comparison of amino acids of common carp fingerlings fed on BSFL based diets (0%, 20% and 40%) for sixty days.

Parameters	**To**	**T₁**	**T₂**	**F-value**	**P-value**
Leucine	6.32±0.63	5.58±0.46	5.26±1.62	0.54	0.629
Isoleucine	1.9±0.02	2.415±0.38	2.275±0.04	2.75	0.209
Methionine	3.715±0.27	4.24±0.52	5.05±0.13	7.44	0.069
Threonine	2.37±0.19	2.41±0.70	3.14±0.12	2.03	0.278
Lysine	5.92±0.41	7±0.16	8.02±0.16	29.3	0.011
Arginine	4.69±0.35	4.045±0.09	3.815±1.01	1.07	0.446
Histidine	4.745±0.09	4.23±0.33	4.375±0.27	2.12	0.267
Valine	4.09±0.32	3.29±0.24	4.06±0.22	5.74	0.094
Phenylalanine	3.09±0.96	2.72±0.09	2.32±0.18	0.92	0.488
Tyrosine	4.29±0.11	4.6±0.05	4.01±0.19	9.47	0.051
Cysteine	1.195±0.10	1.445±0.09	2.12±0.28	13.78	0.031
Serine	4.225±0.38	4.215±0.14	4.33±0.55	0.05	0.951
Aspartic acid	6.375±0.33	6.125±0.31	6.09±0.94	0.13	0.882
Glutamic acid	8.94±0.83	9.95±0.77	9.13±0.04	1.33	0.387
Glycine	4.355±1.03	4.335±0.06	3.575±0.24	1.04	0.455
Alanine	3.92±0.07	4.095±0.26	4.675±0.47	3.15	0.183
Proline	2.95±0.28	3.4±0.19	3.41±0.15	2.89	0.2

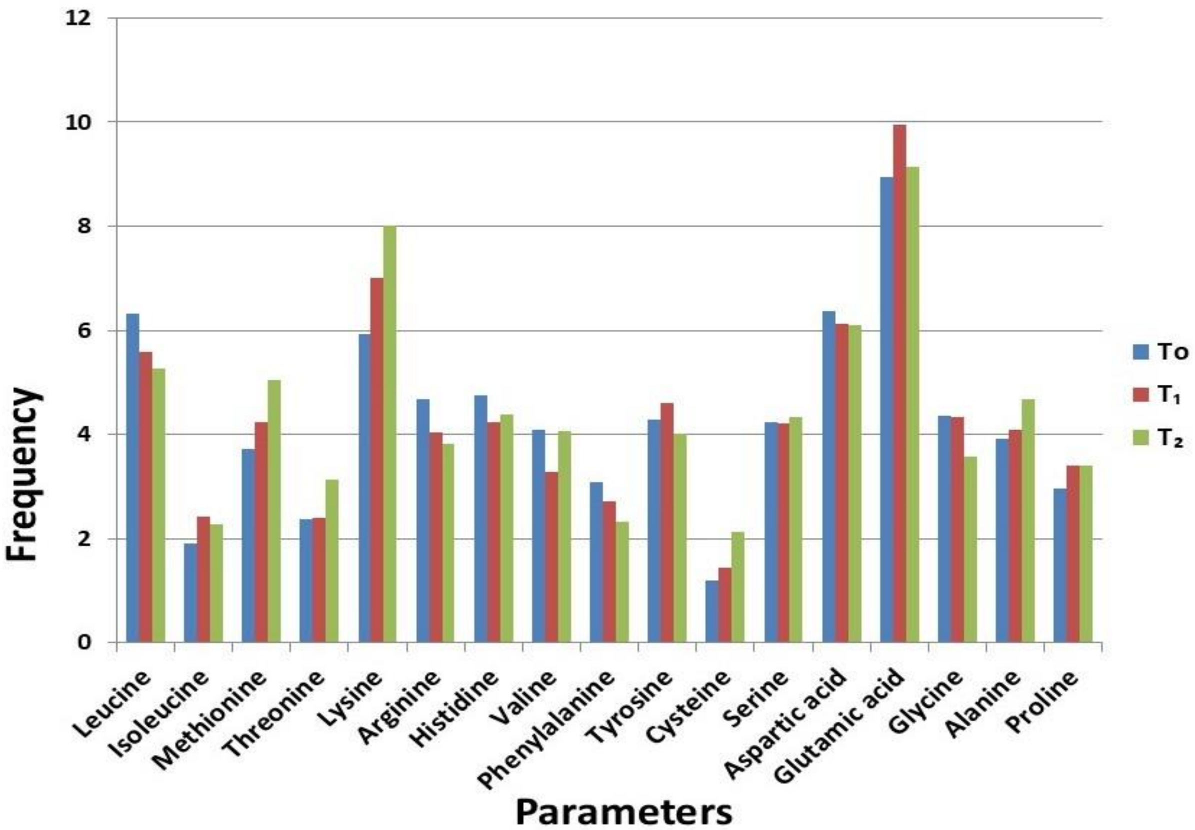

Figure 2: Graph showing comparison of amino acids of common carp fingerlings fed on BSFL based diets (0%, 20% and 40%) for sixty days.

Bone mineralization composition

The effect of control and experimental BSFL meal-based diets on minerals composition of common carp fingerlings are presented in Table 3. Minerals such as P, Ca, Mg, Cu, and Zn were present in the bones of common carp fingerlings. With the replacement of fish meal by BSFLM, phosphorous are more abundant and showed higher statistical difference ($P>0.05$) in the bones of common carp fingerlings. In this regard the value of phosphorous (50.5±3.535) were significantly higher in control group (T_o) of 0% of BSFLM and Lower in experimental group (T_2) of 40% of BSFLM. While, the value of calcium showed lower significant difference ($P<0.05$). The values of calcium (164±1.412) were statistically higher in experimental group (T_2) of 40% BSFLM and lower in control group (T_o) of 0% BSFLM.

Table 3: Comparison of bone minerals of common carp fingerlings fed on BSFL based diets (0%, 20% and 40%) for sixty days.

Parameters	To	T_1	T_2	F-value	P-value
P	50.5±3.535	50.5±0.707	48.5±0.707	0.59	0.607
Ca	158±2.828	163.5±2.121	164±1.412	4.59	0.122
Mg	4.67±0.763	5.475±0.473	5.27±0.056	1.29	0.393
Zn	137±5.65	145.5±2.121	145.5±7.778	1.49	0.355
Cu	9.475±0.219	9.72±0.19	9.71±0.084	1.22	0.409

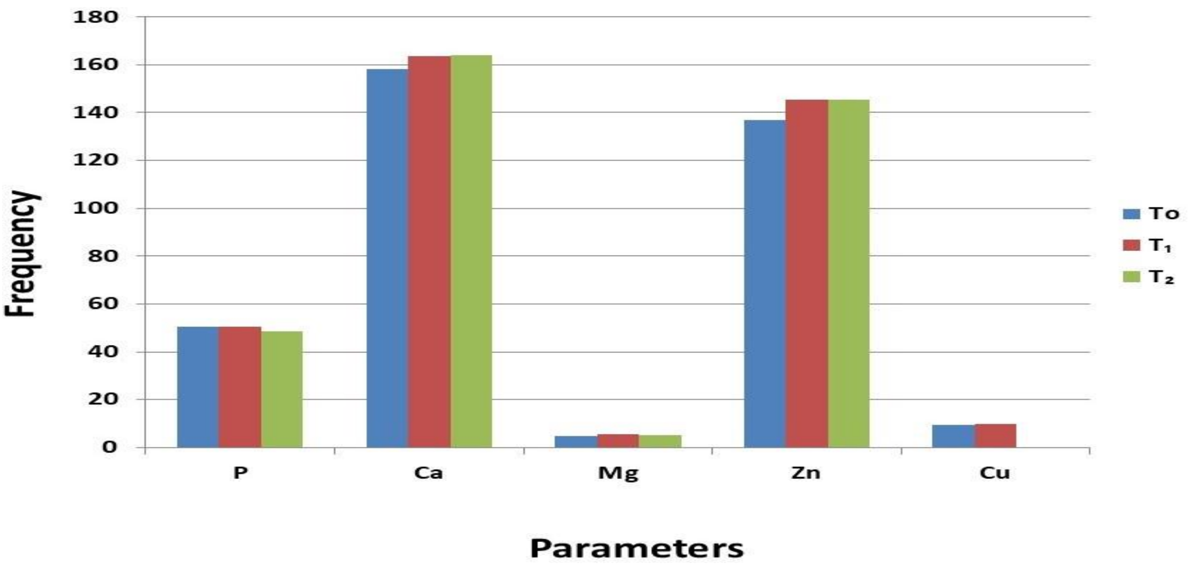

Figure 3: Graph showing comparison of bone minerals of common carp fingerlings fed on B SFL based diets (0%, 20% and 40%) for sixty days.

Discussion

In the formulation of fish feed black soldier fly larvae (BSFL) were utilized in place of fishmeal (FM) [39]. *Hermetia illucens* is one of the most promising insect species for aquaculture as a commercial enterprise [34, 53]. Because of their superior fatty acid composition amino acid profile and mineral profile of *Hermetia illucens* larvae are among the most valuable feed stocks [54, 55]. They can replace fish meals more successfully than soybean meals [37, 56]. According to the current study findings feeding black soldier fly larvae instead of dietary fish meal (FM) to common carp fingerlings improved the fatty acid content and amino acid composition of fish muscle and reduced fishmeal by up to 60% when fed to fish without affecting growth performance or feed utilization.

BSFLM may be a rich source of these essential fatty acids, as evidenced by the rise in docosahexaenoic acid (DHA) and docosapentanoic acid (DPA) in the experimental groups (T_1 and T_2). Fish growth and development depend on DHA and DPA, and deficiencies in these nutrients can result in stunted growth and health issues [57]. The experimental groups' increased DHA and DPA levels imply that BSFLM might be utilized as a wholesome and sustainable substitute for fish meal.

According to our results 17 fatty acids such as Myristic, Pentadecanoic, palmitic, Heptadecanoic, Stearic, Palmitoleic, cis-11 Eicosenoic, Nervonic, Linoleic acid (LA), cis-11, 14 Eicosadienoic, Lambda-linolenic acid (GLA), cis-8,11,14 Eicosatrienoic acid, Arachidonic, Eicosapentaenoic acid (EPA), Docosapentanoic acid (DPA) and Docosahexaenoic acid (DHA) were found in the muscles of common carp fingerlings. Cis-11, 14 Eicosadienoic and Docosahexaenoic (DHA) showed higher significant difference ($P>0.05$) in fish muscle. In this regard the value of cis-11, 14 Eicosadienoic (0.86±0.14) were significantly higher in control group (To) of 0% BSFLM and lowest in experimental group (T_1) of 20% BSFLM. In Docosahexaenoic acid (1.48±0.25), the value of experimental group (T_2) of 40% BSFLM were significantly higher and lowest in T_1. While Docosapentanoic acid (DPA) and Nervonic showed lower significant difference ($P<0.05$) in fish muscles. In this regard the value of Docosapentanoic acid (0.82±0.12) and of Nervonic (0.305±0.07) were significantly low in control group (To) of 0% BSFLM and highest in (T_2) of 40% BSFLM. The current study found that feeding common carp black soldier fly larvae greatly enhanced the fatty acid composition of the fish muscles. When BSF larvae were fed to Jian carp (*Cyprinus carpio*) similar outcomes were observed [58]. The study findings demonstrated that it is feasible to substitute BSFLM for dietary FM in the diets of Jian carp. Following nutrient enrichment fish quality would be actively affected by BSFLM.

The substitution of BSFLM for fish meals also had an impact on the fish's amino acid profile. The experimental groups' increased levels of serine and aspartic acid suggest that BSFLM can supply the fish with these vital amino acids. Fish growth and development depend on serine and aspartic acid, and deficiencies in these nutrients might result in stunted growth and health issues [59]. The experimental groups' increased serine and aspartic acid levels imply that BSFLM can be utilized as a wholesome and sustainable substitute for fish meals.

The bone mineralization results showed that the replacement of fish meal with BSFLM significantly affected the mineral composition of the fish bones. The increase in calcium in the experimental groups indicates that BSFLM can provide this essential mineral to the fish. Calcium is important for the growth and development of fish bones, and its deficiency can lead to impaired growth and health problems [22, 60, 61] The higher levels of calcium in the experimental groups suggest that BSFLM can be used as a sustainable and nutritious alternative to fish meal.

According to our results, the replacement of fish meal by BSFLM serine and aspartic acid are more abundant and showed higher statistical difference ($P>0.05$) in the muscles of common carp fingerlings. Aspartic acid in this regard the value of serine (4.33±0.55) is higher in experimental group (T_2) and lower in (T_1) and the value of aspartic acid (6.375±0.33) is higher in control group (To) and lower in (T_2). Aspartic acid and serine are non-essential amino acids and play an important role in growth and development [62, 63]. While the value of lysine and cysteine showed lower statistical difference ($P<0.05$). The value of lysine (8.02±0.16) and cysteine (4.33±0.55) is higher in experimental group (T_2) and lower in (T_1). An EAA called lysine is heavily needed for healthy growth, and a lack of it results in immunodeficiency [24, 64]. The value of tyrosine amino acid is statistically significant ($P\leq0.05$). Three amino acids such as tryptophan, asparagine and glutamine are absent in common carp muscles. In previous studies no similar results have been found about amino acid composition in common carp fingerlings fed on BSFL based diet. Furthermore, fish fed black soldier fly larvae meal had higher muscle protein content than control fish. There hasn't been any evidence of distinct protein sources changing the amount of muscle protein despite the assumption that higher levels of muscle protein are linked to higher levels of dietary protein [65]. Given the rising cost and scarcity of fish meals, BSFLM could prove to be a cost-effective and sustainable feed ingredient for fish diets. Additionally, BSFLM could serve as a high-quality source of protein for the aquaculture sector.

The effect of control and experimental BSFL meal-based diets on bone minerals composition of common carp fingerlings are summarized in Table 3. Minerals such as P, Ca, Mg, Cu, and Zn were present in the bones of common carp fingerlings. Phosphorus (P) is an important mineral for fish because it is necessary for growth and the mineralization of bone [66, 67]. With the replacement of fish meal by BSFLM phosphorous are more abundant and showed higher statistical difference ($P>0.05$) in the bones of common carp fingerlings. In this regard the values of phosphorous (50.5±3.535) were significantly higher in control group (T_o) of 0% of BSFLM and Lower in experimental group (T_2) of 40% of BSFLM. While the value of calcium showed lower significant difference ($P<0.05$). The values of calcium (164±1.412) were statistically higher in experimental group (T_2) of 40% BSFLM and lower in control group (T_o) of 0% BSFLM.

As with our findings rising P levels cause an increase in the mineral concentrations in the bones which is consistent with the findings of Chavez-Sanchez, Martinez-Palacios [68] who demonstrated a strong correlation between P levels in the diet and bone mineralization.

The study finding that the ideal P requirement for growth was lower than the requirement for mineralization was consistent with that of Chavez-Sanchez, Martinez-Palacios [69], and Satoh, Hernández [70] who stated that mineralization in American cichlid continued to increase with dietary P levels above that required for the maximum growth of the fish. However, in sturgeon [71 - 73] and goldfish [74 - 77] an inverse response was observed contrary to our study [71].

Conclusion

The findings of the current study conclusively show that fishmeal (FM) to BSFL meal substitution improved the growth rate fatty acid composition amino acid composition, and bone mineralization of common carp. Using insects as fish feed is challenging, but this application may increase the expansion in the aquaculture sector. Meal of larvae of black soldier flies could be very interesting. Therefore, we draw the conclusion that the insect-based meal made from black soldier fly larvae (BSFL) is a nutritionally appropriate source of protein, fat and minerals for common carp fingerlings.

Acknowledgments

Authors are thankful to the university department for the completion of work.

Author's contribution

All authors contributed equally to the manuscript.

References

1. Bostock J, McAndrew B, Richards R, Jauncey K, Telfer T, Lorenzen K, Little D, Ross L, Handisyde N, Gatward I, Corner R. Aquaculture: global status and trends. Philosophical Transactions of the Royal Society B: Biological Sciences. 2010 Sep 27;365(1554): 2897-2912
2. Ahmad A, Abdullah SRS, Hasan HA, Othman AR, Ismail NI. Aquaculture industry: Supply and demand, best practices, effluent and its current issues and treatment technology. Journal of Environmental Management. 2021; 287:112271.
3. Subasinghe R, Soto D, Jia J. Global aquaculture and its role in sustainable development. Reviews in aquaculture. 2009;1(1):2-9.
4. Sadiqa A, Parveen S, Riaz HF, Ayoub U, Shaheen A, Rasheed S, Zahra M, Nawaz Y, Zafar J, Khan MS. Toxic Impact Of Detergent (Bright) On Growth, External Morphology, Behaviour, Liver Toxicology, Haematological Parameters And Histopathology Of Nile Tilapia (Oreochromis Niloticus). Journal of Survey in Fisheries Sciences. 2024;11(4):45-54.
5. Tacon AG, Hasan MR, Metian M. Demand and supply of feed ingredients for farmed fish and crustaceans: trends and prospects. FAO Fisheries and Aquaculture technical paper. 2011 (564):I.
6. Tacon AG, Metian M. Global overview on the use of fish meal and fish oil in

industrially compounded aquafeeds: Trends and future prospects. Aquaculture. 2008;285(1-4):146-158.
7. Smil V. Should we eat meat?: Evolution and consequences of modern carnivory: John Wiley & Sons; 2013.
8. Fróna D, Szenderák J, Harangi-Rákos M. The challenge of feeding the world. Sustainability. 2019;11(20):5816.
9. Pauly D, Christensen V. Primary production required to sustain global fisheries. Nature. 1995;374(6519):255-257.
10. Shepherd C, Jackson A. Global fishmeal and fish-oil supply: inputs, outputs and marketsa. Wiley Online Library; 2013. p. 1046-1066.
11. Ebrahimi GH, Ouraji H, Khalesi MK, Sudagar M, Barari A, Zarei Dangesaraki M, Jani Khalili KH. Effects of a prebiotic, Immunogen®, on feed utilization, body composition, immunity and resistance to Aeromonas hydrophila infection in the common carp Cyprinus carpio (Linnaeus) fingerlings. Journal of animal physiology and animal nutrition. 2012 Aug;96(4):591-599.
12. Naylor RL, Hardy RW, Buschmann AH, Bush SR, Cao L, Klinger DH, Little DC, Lubchenco J, Shumway SE, Troell M. A 20-year retrospective review of global aquaculture. Nature. 2021 Mar 25;591(7851):551-563.
13. Zinn KE, Hernot DC, Fastinger ND, Karr-Lilienthal LK, Bechtel PJ, Swanson KS, Fahey Jr GC. Fish protein substrates can substitute effectively for poultry by-product meal when incorporated in high-quality senior dog diets. Journal of animal physiology and animal nutrition. 2009 ;93(4):447-455.
14. Cho J, Kim I. Fish meal–nutritive value. Journal of Animal Physiology and Animal Nutrition. 2011;95(6):685-692.
15. Balami S, Sharma A, Karn R. Significance of nutritional value of fish for human health. Malaysian Journal of Halal Research. 2019;2(2):32-34.
16. Tocher DR. Metabolism and functions of lipids and fatty acids in teleost fish. Reviews in fisheries science. 2003;11(2):107-184.
17. Moradi Y, Bakar J, Motalebi A, Syed Muhamad S, Che Man Y. A review on fish lipid: composition and changes during cooking methods. Journal of Aquatic Food Product Technology. 2011;20(4):379-390.
18. Sargent J, Tacon A. Development of farmed fish: a nutritionally necessary alternative to meat. proceedings of the Nutrition Society. 1999;58(2):377-383.
19. Henderson RJ, Tocher DR. The lipid composition and biochemistry of freshwater fish. Progress in lipid research. 1987;26(4):281-347.
20. Hossain M, Yoshimatsu T. Dietary calcium requirement in fishes. Aquaculture nutrition. 2014;20(1):1-11.
21. Larsen T, Thilsted SH, Kongsbak K, Hansen M. Whole small fish as a rich calcium source. British Journal of Nutrition. 2000;83(2):191-196.
22. Lall SP, Kaushik SJ. Nutrition and metabolism of minerals in fish. Animals. 2021;11(09):2711.
23. Gutasi A. Benefit and drawbacks of fish meal substitution in aquaculture diets. 2021.
24. Tacon AG, Metian M. Feed matters: satisfying the feed demand of aquaculture. Reviews in Fisheries Science & Aquaculture. 2015;23(1):1-10.
25. Niklitschek EJ, Soto D, Lafon A, Molinet C, Toledo P. Southward expansion of the C hilean salmon industry in the P atagonian F jords: main environmental challenges. Reviews in Aquaculture. 2013;5(3): 172-195.
26. Sicuro BJRiA. The future of caviar production on the light of social changes: a new dawn for caviar? 2019;11(1):204-219.
27. Council NR, Earth Do, Studies L, Research IfLA. Guidance for the description of animal research in

scientific publications. 2011.
28. Chapman AD. Numbers of living species in Australia and the world. 2009.
29. Henry M, Gasco L, Piccolo G, Fountoulaki EJAFS, Technology. Review on the use of insects in the diet of farmed fish: past and future. 2015;203:1-22.
30. Payne CL, Scarborough P, Rayner M, Nonaka KJTiFS, Technology. A systematic review of nutrient composition data available for twelve commercially available edible insects, and comparison with reference values. 2016;47:69-77.
31. Traksele L, Speiciene V, Smicius R, Alencikiene G, Salaseviciene A, Garmiene G, Zigmantaite V, Grigaleviciute R, Kucinskas A. Investigation of in vitro and in vivo digestibility of black soldier fly (Hermetia illucens L.) larvae protein. Journal of Functional Foods. 2021 Apr 1;79:104402.
32. Sándor ZJ, Banjac V, Vidosavljević S, Káldy J, Egessa R, Lengyel-Kónya É, Tömösközi-Farkas R, Zalán Z, Adányi N, Libisch B, Biró J. Apparent digestibility coefficients of black soldier fly (Hermetia illucens), yellow mealworm (Tenebrio molitor), and blue bottle fly (Calliphora vicina) insects for juvenile African catfish hybrids (Clarias gariepinus× Heterobranchus longifilis). Aquaculture Nutrition. 2022;2022(1):4717014.
33. Diener S, Zurbrügg C, Tockner K. Conversion of organic material by black soldier fly larvae: establishing optimal feeding rates. Waste Management & Research. 2009;27(6):603-610.
34. Barragan-Fonseca KB, Dicke M, van Loon JJ. Nutritional value of the black soldier fly (Hermetia illucens L.) and its suitability as animal feed–a review. Journal of Insects as Food and Feed. 2017;3(2):105-120.
35. Zhuang P, McBride MB, Xia H, Li N, Li Z. Health risk from heavy metals via consumption of food crops in the vicinity of Dabaoshan mine, South China. Science of the total environment. 2009;407(5):1551-1561.
36. Sandrock C, Leupi S, Wohlfahrt J, Kaya C, Heuel M, Terranova M, Blanckenhorn WU, Windisch W, Kreuzer M, Leiber F. Genotype-by-diet interactions for larval performance and body composition traits in the black soldier fly, Hermetia illucens. Insects. 2022 Apr 30;13(5):424.
37. Sánchez M. Insect meal as renewable source of food for animal feeding: a review. Sánchez, M. J Cleaner Prod. 2016;6:112-126.
38. St-Hilaire S, Sheppard C, Tomberlin JK, Irving S, Newton L, McGuire MA, Mosley EE, Hardy RW, Sealey W. Fly prepupae as a feedstuff for rainbow trout, Oncorhynchus mykiss. Journal of the world aquaculture society. 2007 Mar;38(1):59-67.
39. Ramos-Elorduy J. Energy supplied by edible insects from Mexico and their nutritional and ecological importance. Ecology of food and nutrition. 2008;47(3):280-297.
40. Nairuti RN, Musyoka SN, Yegon MJ, Opiyo MA. Utilization of black soldier fly (Hermetia illucens Linnaeus) larvae as a protein source for fish feed–a review. Aquaculture Studies. 2021;22(2).
41. Bondari K, Sheppard D. Soldier fly, Hermetia illucens L., larvae as feed for channel catfish, Ictalurus punctatus (Rafinesque), and blue tilapia, Oreochromis aureus (Steindachner). Aquaculture Research. 1987;18(3):209-220.
42. Sheppard DC, Newton GL, Thompson SA, Savage S. A value added manure management system using the black soldier fly. Bioresource technology. 1994;50(3):275-279.
43. Diener S, Studt Solano NM, Roa Gutiérrez F, Zurbrügg C, Tockner K. Biological treatment of municipal

organic waste using black soldier fly larvae. Waste and Biomass Valorization. 2011;2:357-363.
44. Zheng L, Crippen TL, Singh B, Tarone AM, Dowd S, Yu Z, Wood TK, Tomberlin JK. A survey of bacterial diversity from successive life stages of black soldier fly (Diptera: Stratiomyidae) by using 16S rDNA pyrosequencing. Journal of medical entomology. 2013 May 1;50(3):647-658.
45. Erickson MC, Islam M, Sheppard C, Liao J, Doyle MP. Reduction of Escherichia coli O157: H7 and Salmonella enterica serovar Enteritidis in chicken manure by larvae of the black soldier fly. Journal of food protection. 2004;67(4):685-690.
46. Bradley SW, Sheppard D. House fly oviposition inhibition by larvae ofHermetia illucens, the black soldier fly. Journal of chemical ecology. 1984;10(6):853-859.
47. Liu C, Wang C, Yao H. Comprehensive resource utilization of waste using the black soldier fly (Hermetia illucens (L.))(Diptera: Stratiomyidae). Animals. 2019;9(6):349.
48. Panikkar P, Parakkandi J, Khan F, Das BK, Udayakumar A, Eregowda VM, Yandigeri M. Use of black soldier fly (Hermetia illucens) prepupae reared on organic waste as feed or as an ingredient in a pellet-feed formulation for Nile tilapia (Oreochromis niloticus). Environmental Science and Pollution Research. 2022 Oct;29(48):72968-72978.
49. Sheppard C. House fly and lesser fly control utilizing the black soldier fly in manure management systems for caged laying hens. Environmental entomology. 1983;12(5):1439-1442.
50. Yanniotis S, Kotseridis G, Orfanidou A, Petraki A. Effect of ethanol, dry extract and glycerol on the viscosity of wine. Journal of Food Engineering. 2007; 81(2):399-403.
51. Folch J, Lees M, Sloane Stanley GH. A simple method for the isolation and purification of total lipids from animal tissues. J biol Chem. 1957;226(1):497-509.
52. Sargent J. Fish oils and human diet. British Journal of Nutrition. 1997;78(1):S5-S13.
53. Huis AV, Itterbeeck JV, Klunder H, Mertens E, Halloran A, Muir G, Vantomme P. Edible insects: future prospects for food and feed security. 2013.
54. Abd El-Hack ME, Shafi ME, Alghamdi WY, Abdelnour SA, Shehata AM, Noreldin AE, Ashour EA, Swelum AA, Al-Sagan AA, Alkhateeb M, Taha AE. Black soldier fly (Hermetia illucens) meal as a promising feed ingredient for poultry: A comprehensive review. Agriculture. 2020 Aug 6;10(8):339.
55. Makkar HP, Tran G, Heuzé V, Ankers P. State-of-the-art on use of insects as animal feed. Animal feed science and technology. 2014;197:1-33.
56. Tran G, Heuzé V, Makkar H. Insects in fish diets. Animal frontiers. 2015;5(2):37-44.
57. Sargent JR, Tocher DR, Bell JG. The lipids. Fish nutrition. 2003:181-257.
58. Li S, Ji H, Zhang B, Zhou J, Yu H. Defatted black soldier fly (Hermetia illucens) larvae meal in diets for juvenile Jian carp (Cyprinus carpio var. Jian): Growth performance, antioxidant enzyme activities, digestive enzyme activities, intestine and hepatopancreas histological structure. Aquaculture. 2017;477:62-70.
59. Wilson RP. Amino acids and proteins. Fish nutrition: Elsevier; 2003. p. 143-179.
60. Lall S, Kaushik S. Nutrition and Metabolism of Minerals in Fish. Animals 2021, 11, 2711. s Note: MDPI stays neutral with regard to jurisdictional claims in published …; 2021.
61. Lall SP, Lewis-McCrea LM. Role of nutrients in skeletal metabolism and pathology in fish—An overview.

Aquaculture. 2007;267(1-4):3-19.
62. Fuso A, Barbi S, Macavei LI, Luparelli AV, Maistrello L, Montorsi M, Sforza S, Caligiani A. Effect of the rearing substrate on total protein and amino acid composition in black soldier fly. Foods. 2021 Jul 30;10(8):1773.
63. Krogdahl Å, Penn M, Thorsen J, Refstie S, Bakke AM. Important antinutrients in plant feedstuffs for aquaculture: an update on recent findings regarding responses in salmonids. Aquaculture research. 2010;41(3):333-344.
64. Schiavone A, De Marco M, Martínez S, Dabbou S, Renna M, Madrid J, Hernandez F, Rotolo L, Costa P, Gai F, Gasco L. Nutritional value of a partially defatted and a highly defatted black soldier fly larvae (Hermetia illucens L.) meal for broiler chickens: apparent nutrient digestibility, apparent metabolizable energy and apparent ileal amino acid digestibility. Journal of animal science and biotechnology. 2017 Dec;8:1-9.
65. Bureau DP, Kaushik SJ, Cho CY. Bioenergetics. Fish nutrition. 2003:1-59.
66. Borlongan IG, Satoh S. Dietary phosphorus requirement of juvenile milkfish, Chanos chanos (Forsskal). Aquaculture Research. 2001;32:26-32.
67. 67. Council NR. Nutrient requirements of fish and shrimp: National Academies Press; 2011.
68. Chavez-Sanchez C, Martinez-Palacios CA, Martinez-Perez G, Ross LG. Phosphorus and calcium requirements in the diet of the American cichlid Cichlasoma urophthalmus (Gunther). Aquaculture Nutrition. 2000;6(1):1-10.
69. Chavez-Sanchez C, Martinez-Palacios CA, Martinez-Perez G, Ross LG. Phosphorus and calcium requirements in the diet of the American cichlid Cichlasoma urophthalmus (Günther). Aquaculture Nutrition. 2000;6(1):1-9.
70. Satoh S, Hernández A, Tokoro T, Morishita Y, Kiron V, Watanabe T. Comparison of phosphorus retention efficiency between rainbow trout (Oncorhynchus mykiss) fed a commercial diet and a low fish meal based diet. Aquaculture. 2003;224(1-4):271-282.
71. Xu Q, Xu H, Wang C, Zheng Q, Sun D. Studies on dietary phosphorus requirement of juvenile Siberian sturgeon Acipenser baerii. Journal of Applied Ichthyology. 2011;27(2):709-714.
72. Jin J, Wang C, Tang Q, Xie C, Dai Z. Dietary phosphorus affected growth performance, body composition, antioxidant status and total P discharge of young hybrid sturgeon (♀ Huso huso×♂ Acipenser schrenckii) in winter months. Journal of Applied Ichthyology. 2012;28(5):697-703.
73. Guo Z, Li L, Liu T, Wang Y, Li Z, Zhang P, Liu H. Dietary citric acid improves growth performance, feed utilization, phosphorus utilization, lateral scute hardness, and duodenal microstructure in juvenile hybrid sturgeon (Acipenser baerii♀× A. schrenckii♂). Aquaculture Reports. 2023 Dec 1;33:101833.
74. Braga WF, Araújo JG, Martins GP, Oliveira SL, Guimarães IG. Dietary total phosphorus supplementation in goldfish diets. Latin American Journal of Aquatic Research. 2016;44(1):129-136.
75. Sales J, Janssens GP. Nutrient requirements of ornamental fish. Aquatic Living Resources. 2003; 16(6):533-540.
76. Souto CN, Lemos MVAd, Martins GP, Araújo JG, Lopes KLdAM, Guimarães IG. Protein to energy ratios in goldfish (Carassius auratus) diets. Ciência e Agrotecnologia. 2013;37:550-558.
77. Roslan MNAM, Estim A, Venmathi Maran BA, Mustafa S. Effects of aquatic plants on nutrient concentration in water and growth performance of fantail goldfish in an aquaculture system. Sustainability. 2021;13(20): 11236.