Indian J. Anim. Nutr. 2025. 42 (3): 392-402

doi: 10.56093/IJAN.v42i3.10

Indian Journal of
Animal Nutrition

Effect of Dietary Protein Substitution of Fish Meal with Insect Larvae Meal on the Growth and Digestive Functions of Koi Carp *Cyprinus carpio* var. *koi* (Linnaeus, 1758) Fry in Nursery Phase

S.Jeyaprakashsabari [1], S. Aanand[1]* and Cheryl Antony [2]

[1] Erode Bhavanisagar Centre for Sustainable Aquaculture (EBCeSA), TNJFU,Erode - 638451, Tamilnadu, India.

[2] The Director, Directorate of Sustainable Aquaculture, TNJFU, Nagapattinam - 611002, Tamilnadu, India.

* Correspondence: anand@tnfu.ac.in

ABSTRACT

An 8-week feeding trial was conducted to evaluate the effects of two insect larvae meals such as silkworm pupae (*Bombyx mori*) meal and black soldier fly (*Hermetia illucens*) larvae meal as dietary protein substitutes for fish meal on the growth performance and digestive physiology of koi carp (*Cyprinus carpio* var. *koi*) fry. For that, 15 days old induced bred koi fry (2.28/ ±/ 0.24/ cm; 0.26/ ±/ 0.14/ g) were stocked in happas (10/ ×/ 3/ ×/ 1/ m) placed within an experimental earthen pond. Nine iso-nitrogenous (351.07/ g/ kg{ ^{1}) and iso-lipidic (71.98/ g/ kg{ ^{1}) experimental diets were formulated with insect meals replacing fish meal at 20%(SWP20,BSF20), 30%(SWP30,BSF30), 40%(SWP40,BSF40), and 50%(SWP50,BSF50) inclusion levels, alongside a control diet containing only fish meal. Results showed significantly higher (P/ </ 0.05) final length and weight gain in fry fed SWP50 (3.26/ ±/ 0.01/ cm; 3.79/ ±/ 0.01/ g) and BSF50 (3.22/ ±/ 0.01/ cm; 3.56/ ±/ 0.02/ g) diets. Growth indices including feed conversion ratio, feed efficiency ratio, protein efficiency ratio and survival rate were also notably improved in these groups. Furthermore, digestive enzyme activities were significantly elevated (P/ </ 0.05) in fry fed SWP50 and BSF50 diets, indicating enhanced nutrient utilization. These findings depicts that both the insect larvae meal can effectively replace 50% of fish meal with a dietary inclusion level of 147 $g\ kg^{-1}$ in the diet of koi carp fry without compromising growth, survival and digestive function.

KEYWORDS: Digestive enzymes, Fish meal, Insect meal, Koi carp.

Article received: 27 June 2025; Article accepted: 19 July 2025

INTRODUCTION

Ornamental fishes have played a significant role in the global pet industry, with their popularity increasing steadily since the 1970s. The industry has seen an annual growth rate of 14%, and over one billion live ornamental fishes are being exported worldwide each year (Maceda-Veiga et al., 2016; Prakash et al., 2017). The ornamental fish trade is valued at approximately US$15-30 billion annually (Novak et al., 2020), with Asian countries contributing around 57% of total global exports (Dey, 2016). India, with its rich biodiversity, harbors over 374 indigenous freshwater and 700 marine ornamental fish species, along with more than 288 exotic fish species in trade (Ghosh et al., 2003; Mahapatra, 2018). Among these, koi carp (*Cyprinus carpio* var. *koi*) stands out for its vibrant colouration and aesthetic appeal, making it one of the most commercially important ornamental species in the global market (Jain et al., 2019). Moreover, the nursery phase remains a major bottleneck in the advancement of ornamental aquaculture, as it is a critical and determining stage influencing the overall success of an ornamental fish production unit. Among the various challenges faced during this stage, nutrition particularly feed quality plays a pivotal role. The performance and survival of fry are closely linked to the nutritional composition of the initial diet. Of the various nutrients, protein is most important during the nursery phase due to the high dependency of fry on protein rich diets to support their rapid growth and development.

Fish meal and fish oil, derived from wild-caught forage fish, have long been the primary protein and lipid sources in aquafeeds due to their high protein content, balanced amino acid profile, absence of anti-nutritional factors and superior digestibility (Daniel, 2018). However, the continued reliance on these resources poses sustainability concerns, with fish meal demand projected to rise by 75% from 49.7 million tons in 2015 to 87.1 million tons by 2025 (Tacon and Metian, 2015). This has prompted a global search for sustainable protein alternatives. While plant based proteins and fishery by-products show potential, insect meals have gained considerable attention due to their rapid life cycles, high productivity and favorable nutritional profiles (DeFoliart et al., 2009; Hua et al., 2019; Berggren et al., 2019). Among the various species, black soldier fly (*Hermetia illucens*) larvae meal (BSF) and silkworm pupae (*Bombyx mori*) meal (SWP) have emerged as a promising candidates to be used as an effective protein sources to replace the fish meal partially or fully in the aquafeeds (Hodar et al., 2020; Sahib et al., 2024).

Over the past decade, numerous studies have demonstrated the potential of BSF as a sustainable alternative to fish meal in aquafeeds, with no adverse effects on the growth performance. And successful inclusion of BSF has been reported in Jian carp (*Cyprinus carpio* var. *jian*) diets as both partial (Li et al., 2017) and complete replacements (Zhou et al., 2018), as well as in Amur carp with 70% replacement (Amala et al., 2018), and in mirror carp (*Cyprinus carpio* var. *specularis*) with a dietary inclusion level of 131 g/kg (Xu et al., 2020). Similarly, SWP has shown promising results as a fish meal substitute, with studies in rohu (*Labeo rohita*) (Begum et al., 1994), mirror carp (*Cyprinus carpio* var. *specularis*) (Ji et al., 2012), and Jian carp (*Cyprinus carpio* var. *jian*) (Ji et al., 2015) reporting favorable growth outcomes. Since most of the previous studies recommended partial replacement of fish meal with insect meals for optimal performance (Tran et al., 2015; Henry et al., 2015), the present study investigates the effects of dietary inclusion of BSF and SWP upto 50% replacement of fish meal on the growth and digestive performance of koi carp fry during the nursery phase under captive conditions.

MATERIALS AND METHODS

Ethics statement

The experiment was conducted at the Erode Bhavanisagar Centre for Sustainable Aquaculture (EBCeSA), Erode District, Tamil Nadu, India, in accordance with the ethical guidelines for animal experimentation established by Tamil Nadu Dr. J. Jayalalithaa Fisheries University, Nagapattinam, Tamilnadu, India.

Experimental design

The experiment followed a completely randomized design with eight dietary treatments SWP20, SWP30, SWP40, SWP50, BSF20, BSF30, BSF40, and BSF50 along with a control, each in triplicate. Experimental animals were reared in happas (10 m × 3 m × 1 m, 2 mm mesh size) installed in an earthen pond. The happas were thoroughly cleaned and sun-dried prior to installation, and the pond area was enclosed with bird-proof fencing to prevent predation.

Experimental animals

Induced-bred, 15 days old koi carp fry with an average body length of 2.28/ ±/ 0.24/ cm and average body weight of 0.26/ ±/ 0.14/ g were used as experimental animals. Following acclimatization, the fry were stocked into the experimental happas at a density of 100 fry/m² (3,000 fry per happa).

Experimental diets

Nine iso-nitrogenous and iso-lipidic experimental diets containing 35/ ±/ 0.5% crude protein and 7/ ±/ 0.5% crude lipid were formulated by replacing fish meal with varying levels of silkworm pupae meal (20%-SWP20, 30%-SWP30, 40%-SWP40, 50%-SWP50) and black soldier fly larvae meal (20%-BSF20, 30%-BSF30, 40%-BSF40, 50%-BSF50), based on the formulation by Nandeesha et al. (2002). The control diet contained only fish meal as the sole animal protein source. All the feed ingredients were grounded and homogenized using a feed pulverizer and feed mixer. The mash-type diets were then used to fed the fishes at a rate of 5% of their body weight (Paul and Giri, 2015), twice daily, for 60 days. Proximate composition of the diets including crude protein, crude lipid, ash and moisture contents were analyzed in feed analytical laboratory at Erode Bhavanisagar Centre for Sustainable Aquaculture (EBCeSA), following the standard protocols (AOAC, 2005) and the gross energy values of the experimental diets were calculated according to Henken et al. (1986). The proximate composition of all the experimental diets and the major protein sources were shown in Table 1 and Table 2.

Table 1. Estimated feed and proximate composition of all the experimental diets

Feed Ingredients	Experimental diets (g/100g)								
	Control	SWP20	SWP30	SWP40	SWP50	BSF20	BSF30	BSF40	BSF50
FM [a]	29.40	23.60	20.60	17.60	14.70	23.60	20.60	17.60	14.70
SWP [b]	-	5.80	8.80	11.80	14.70	-	-	-	-
BSF [c]	-	-	-	-	-	5.80	8.80	11.80	14.70
GNOC [d]	29.40	29.40	29.40	29.40	29.40	29.40	29.40	29.40	29.40
Rice bran [e]	24.70	24.70	24.70	24.70	24.70	24.70	24.70	24.70	24.70
Wheat flour [f]	16.40	16.40	16.40	16.40	16.40	16.40	16.40	16.40	16.40
Vit & Min [g]	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
	Proximate composition (DM %)								
Crude protein	34.95	35.25	34.87	35.07	35.15	35.38	34.83	35.14	35.33
Crude lipid	7.01	6.96	7.11	7.24	7.47	7.08	7.19	7.26	7.47
Moisture	5.60	5.32	4.90	5.54	5.22	5.46	5.28	5.56	5.06
Ash	13.59	12.66	12.87	12.79	13.01	13.21	12.94	12.95	12.76
Dry Matter	94.40	94.68	95.10	94.46	94.78	94.54	94.72	94.44	94.94
Gross Energy (MJ/kg)	1.8844	1.9015	1.8988	1.9043	1.9059	1.8952	1.8991	1.9023	1.9114

Table 2. Estimated proximate composition of the major protein sources

Proximate composition (DM%)	FM	SWP	BSF
Crude protein	55.25	52.50	50.15
Crude lipid	8.30	13.44	14.21
Moisture	8.21	7.11	7.02
Ash	22.50	5.65	12.56
Dry Matter	91.79	92.89	92.98
Gross Energy (MJ/kg)	1.8786	2.2677	2.1505

FM - Fish meal; SWP - Silkworm Pupae Meal; BSF - Black Soldier Fly Larvae Meal; GNOC - Ground Nut Oil Cake; Vit & Min - Vitamin and Mineral Mix

MJ/kg - Mega joules / kilogram

[a] Pearl City Fish Meal Plant, Thoothukudi, Tamilnadu

[b] Silvermine Silk Processors Private Limited, Udumalpet, Tamilnadu

[c] Eco Care Agrovet, Pondicherry

[d, e, f, g] Local market around Bhavanisagar, Tamilnadu

[g] Ingredients included per kg: Vitamin A 700000 IU, Vitamin D_3 70000 IU, Vitamin E 250 mg, Cobalt 150 mg, Copper 1200 mg, Iodine 325 mg, Iron 1500 mg, Magnesium 6000 mg, Potassium 100 mg, Sodium 5.9 mg, Manganese 1500 mg, Sulphur 0.72%, Zinc 9600 mg, DL-Methionine 1000 mg, Calcium 25.5%, Phosphorus 12.75%

Water quality parameters

Water samples were collected fortnightly from the experimental happas to analyze the physico-chemical parameters, following standard methods outlined by APHA (2005). The mean values of the water quality parameters recorded during the experimental period were as follows: Water temperature (27±0.01°C), pH (8.30±0.02), Dissolved oxygen (4.13±0.01ppm), Ammonia (0.01±0.03 ppm), Nitrite (0.01±0.04 ppm), Nitrate (0.1±0.01 ppm), Inorganic phosphate (0.79±0.05 ppm), Free CO_2 (7.33±0.01 ppm), Total hardness (110±0.01 ppm), Total alkalinity (89±0.02 ppm), Total suspended solids (0.08±0.02 ppm) and Total dissolved solids (0.70±0.14 ppm).

Bio-growth indices

Sampling was carried out to assess the growth performance of fishes fed with different experimental diets fortnightly over 60 days of experimental period. Based on the recorded data, the growth parameters such as mean length gain (MLG), mean weight gain (MWG), percentage length gain (PLG), percentage weight gain (PWG), specific growth rate (SGR), feed conversion ratio (FCR), feed efficiency ratio (FER), the protein efficiency ratio (PER) and survival rate were calculated by using the following formulae (Li et al., 2017).

$$\text{MLG} = \text{Mean final length (cm)} - \text{Mean initial length (cm)}$$

$$\text{MWG} = \text{Mean final weight (g)} - \text{Mean initial weight (g)}$$

$$\text{PLG (\%)} = \frac{\text{Final length (cm)} - \text{Initial length (cm)}}{\text{Initial length (cm)}} \times 100$$

$$\text{PWG (\%)} = \frac{\text{Final weight (g)} - \text{Initial weight (g)}}{\text{Initial weight (g)}} \times 100$$

$$\text{SGR} = \frac{\ln \text{final weight} - \ln \text{initial weight}}{\text{Experimental duration in days}} \times 100$$

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

$$\text{FER} = \frac{1}{\text{FCR}}$$

$$\text{PER} = \frac{\text{Total wet weight gain (g)}}{\text{Dry weight of protein fed (g)}}$$

$$\text{Survival rate (\%)} = \frac{\text{Total number of harvested animal}}{\text{Total number stocked}} \times 100$$

Digestive enzyme assay

At the end of the trial, samples (n=10) from each replicates were randomly collected and their intestines were dissected out for digestive enzyme analysis. A 5% tissue homogenate was then prepared using a pestle and mortar, followed by centrifugation at 5,000 rpm for 10 minutes at 4°C. The resulting supernatant was stored at –20°C for subsequent enzyme assays. Digestive enzyme activities were determined as follows: protease by the casein digestion method (Drapeau, 1976), amylase by the di-nitro-salicylic acid method (Rick and Stegbauer, 1974), and lipase by the titrimetric method (Cherry and Crandall, 1932). Additionally, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities were measured following the method of Wooten (1964).

Statistical analysis

The data collected at the end of experiment were processed and analysed by one-way ANOVA following statistical software SPSS version 20.0 at 5% significance level to test for significant differences between the mean values of various treatments and by using the Duncan Multiple Range test (SPSS Statistics for Windows, IBM. Version 20.0. Armonk, NY: IBM Corp).

RESULTS AND DISCUSSION

Growth performance

The effects of different experimental diets on the growth performance of koi carp fry are presented in Table 3. After the 60-day feeding trial, the highest MLG, MWG, PLG, PWG were significantly higher (P/ </ 0.05) in fish fed with SWP50 (3.26/ ±/ 0.01/ cm, 3.79/ ±/ 0.01/ g,

Table 3. Estimated bio-growth parameters of the fishes recorded at the end of the experiment

MIL – Mean Initial Length, MFL – Mean Final Length, MLG – Mean Length Gain, PLG – Percentage Length Gain, MIW – Mean Initial Weight, MFW – Mean Final

Growth Parameter	Experimental Diets								
	C	SWP20	SWP30	SWP40	SWP50	BSF20	BSF30	BSF40	BSF50
MIL (cm)	2.28±0.24	2.28±0.24	2.28±0.24	2.28±0.24	2.28±0.24	2.28±0.24	2.28±0.24	2.28±0.24	2.28±0.24
MFL (cm)	4.27±0.01 [a]	4.79±0.02 [b]	5.03±0.01 [c]	5.23±0.01 [d]	5.54±0.02 [e]	4.73±0.02 [b]	5.05±0.03 [c]	5.31±0.01 [d]	5.50±0.01 [e]
MLG (cm)	1.99±0.01 [a]	2.51±0.02 [b]	2.75±0.02 [c]	2.95±0.01 [d]	3.26±0.01 [e]	2.45±0.04 [b]	2.77±0.01 [c]	3.03±0.02 [d]	3.22±0.01 [e]
PLG (%)	87±0.01 [a]	110±0.03 [b]	121±0.02 [c]	129±0.02 [d]	142±0.01 [e]	107±0.01 [b]	121±0.02 [c]	133±0.01 [d]	141±0.02 [e]
MIW (g)	0.26±0.14	0.26±0.14	0.26±0.14	0.26±0.14	0.26±0.14	0.26±0.14	0.26±0.14	0.26±0.14	0.26±0.14
MFW (g)	1.80±0.01 [a]	2.16±0.01 [b]	2.67±0.02 [c]	2.85±0.01 [d]	4.05±0.03 [e]	2.02±0.01 [b]	2.47±0.01 [c]	2.81±0.01 [d]	3.82±0.02 [e]
MWG (g)	1.54±0.01 [a]	1.90±0.01 [b]	2.41±0.04 [c]	2.59±0.03 [d]	3.79±0.01 [e]	1.76±0.01 [b]	2.21±0.05 [c]	2.55±0.02 [d]	3.56±0.02 [e]
PWG (%)	592±0.02 [a]	731±0.01 [b]	927±0.03 [c]	996±0.01 [d]	1457±0.03 [e]	677±0.02 [f]	850±0.01 [g]	981±0.03 [d]	1369±0.02 [e]
SGR (%/day)	3.22±0.01 [a]	3.52±0.01 [b]	3.88±0.02 [c]	3.99±0.02 [d]	4.57±0.01 [e]	3.41±0.01 [b]	3.75±0.03 [c]	3.96±0.01 [d]	4.47±0.01 [e]
FCR	1.11±0.01 [a]	0.93±0.01 [b]	0.83±0.01 [c]	0.85±0.01 [c]	0.80±0.03 [c]	1.07±0.01 [d]	1.01±0.04 [d]	0.88±0.01 [c]	0.85±0.01 [c]
FER	0.90±0.01 [a]	1.07±0.01 [b]	1.20±0.01 [c]	1.17±0.01 [c]	1.25±0.02 [c]	0.93±0.01 [d]	0.99±0.04 [d]	1.13±0.01 [c]	1.17±0.01 [c]
PER	2.56±0.03 [a]	3.04±0.02 [b]	3.43±0.05 [c]	3.35±0.04 [c]	3.55±0.04 [c]	2.65±0.01 [d]	2.81±0.11 [d]	3.22±0.03 [c]	3.35±0.03 [c]
Survival rate (%)	50±0.01 [a]	60±0.03 [b]	65±0.03 [b]	68±0.01 [b]	70±0.02 [b]	65±0.01 [b]	65±0.02 [b]	68±0.03 [b]	70±0.01 [b]

Weight, MWG – Mean Weight Gain, PWG – Percentage Weight Gain, SGR – Specific Growth Rate, FCR – Feed Conversion Ratio, FER – Feed Efficiency Ratio, PER – Protein Efficiency Ratio

Values are expressed in terms of Mean±SD.

Values in the same row with different superscripts vary significantly ($p<0.05$)

142/ ±/ 0.01%, 1457/ ±/ 0.03%) and BSF50 (3.22/ ±/ 0.01/ cm, 3.56/ ±/ 0.02/ g, 141/ ±/ 0.02%, 1369/ ±/ 0.02%) diets respectively. Other growth parameters including SGR, FCR, FER, PER and survival rate were also significantly improved (P/ </ 0.05) in fish fed with SWP50 and BSF50 diets, indicating no adverse effects from replacing fish meal with SWP or BSF upto 50%. Furthermore, MLG increased linearly with higher inclusion levels of SWP and BSF, as shown by the regression equations: SWP (y/ =/ 0.0249x/ +/ 1.9954, R^2/ =/ 0.9973) and BSF (y/ =/ 0.0253x/ +/ 1.9841, R^2/ =/ 0.9954), illustrated in Figures 1 and 2, respectively.

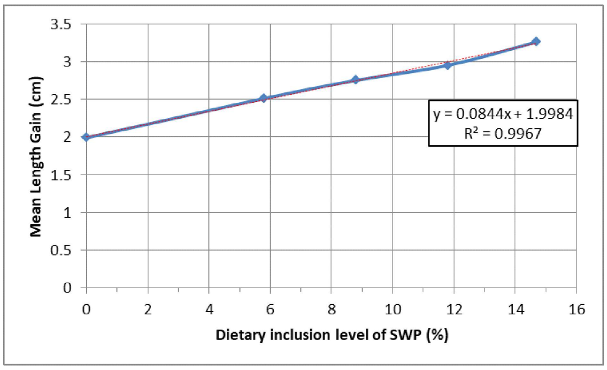

Figure 1. Linear regression of MLG for the SWP included diets

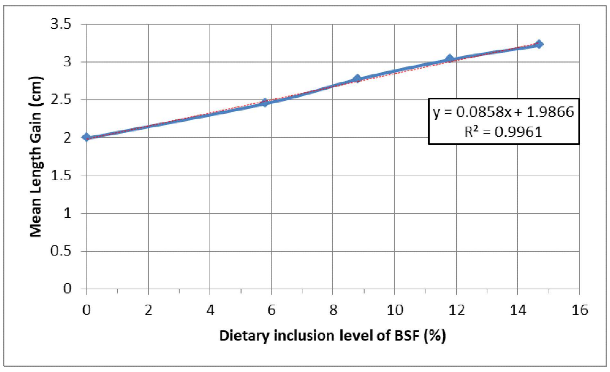

Figure 2. Linear regression of MLG for the BSF included diets

Digestive enzymes

Digestive enzyme activities such as protease, amylase, and lipase were significantly higher (P/ </ 0.05) in fish fed with SWP50 (17.88/ ±/ 0.02, 12.72/ ±/ 0.05, and 22.89/ ±/ 0.01/ µM/ mg{ ^{1} protein) and BSF50 (17.01/ ±/ 0.05, 12.99/ ±/ 0.01, and 22.97/ ±/ 0.01/ µM/ mg{ ^{1} protein) diets respectively. These results indicate efficient protein utilization in both SWP50 and BSF50 diet groups. Similarly, AST and ALT activities were significantly elevated (P/ </ 0.05) in SWP50 (12.87/ ±/ 0.04 and 13.88/ ±/ 0.02/ nM/ mg{ ^{1}/ min{ ^{1}) and BSF50 (12.86/ ±/ 0.01 and 13.69/ ±/ 0.01/ nM/ mg{ ^{1}/ min{ ^{1}) diet fed fishes respectively. The digestive enzyme activity data across treatments are summarized in Table 4.

The findings of the present study depicts that the inclusion level of 147 g Kg^{-1} of SWP and BSF in the diet of koi carp fry significantly outperformed other treatments and the control in terms of growth and digestive function. Among the two best performed treatments, SWP50 showed superior performance, with 1.2% and 0.7% higher MLG and PLG respectively, and 6.4% higher MWG and PWG compared to BSF50. This suggests that 147 g Kg^{-1} inclusion level of SWP yields the most favorable growth outcomes. However, BSF50 also proved effective, achieving a 62% greater length gain than the control group, which indicates that it can also be used as an sustainable fish meal substitute. Additionally, growth parameters (MLG and MWG) showed a progressive increase with higher inclusion levels of both SWP and BSF, highlighting their suitability in koi fry diets. A similar study in common carp (*Cyprinus carpio*) also reported improved final body length when fish meal was replaced with 30% of soldier fly (*Ptecticus tenebrifer*) and mealworm (*Tenebrio molitor*) meals (Mamuad et al., 2021). Further, in the present study, the SGR, FCR, FER and PER were significantly better in SWP50 and BSF50 diets. These results are consistent with earlier findings in rainbow trout (*Oncorhynchus mykiss*) where BSF inclusion at 40% (Renna et al., 2017) and SWP at 50% (Dheke, 2013) resulted in improved FCR and SGR without compromising survival.

In addition to improved growth, the present study showed a corresponding increase in digestive enzyme activities with higher dietary inclusion levels of SWP and BSF in koi fry diets. This trend contrasts with findings in Jian carp (*Cyprinus carpio* var. *jian*), where feeding defatted BSF upto 100% resulted in no significant differences (P/ >/ 0.05) in amylase and lipase activities across treatments (Li et al., 2017). Among the enzymes studied in the present study, protease and lipase activities were notably higher than amylase, likely due to the greater reliance of early stage fish on protein rich diets over carbohydrates. The elevated enzyme levels observed in this study suggest enhanced digestive capacity and efficient nutrient utilization in fish fed insect based

Table 4. Estimated digestive enzyme activities of the fishes fed with different experimental diets

Enzymes	Experimental Diets								
	C	SWP20	SWP30	SWP40	SWP50	BSF20	BSF30	BSF40	BSF50
Protease (µmole/mg protein)	14.80± 0.04 ^{a}	15.28± 0.01 ^{a}	16.58± 0.02 ^{a}	17.39± 0.01 ^{a}	17.88± 0.02 ^{a}	16.12± 0.03 ^{a}	16.56± 0.02 ^{a}	16.90± 0.01 ^{a}	17.01± 0.05 ^{a}
Amylase (µmole/mg protein)	8.76± 0.02 ^{a}	10.62± 0.01 ^{b}	11.75± 0.02 ^{c}	12.65± 0.01 ^{c}	12.72± 0.05 ^{c}	11.18± 0.01 ^{b}	12.70± 0.03 ^{c}	12.93± 0.02 ^{c}	12.99± 0.01 ^{c}
Lipase (µmole/mg protein)	18.76± 0.01 ^{a}	20.26± 0.05 ^{b}	21.83± 0.02 ^{c}	22.50± 0.01 ^{c}	22.89± 0.01 ^{c}	20.85± 0.03 ^{b}	22.13± 0.02 ^{c}	22.88± 0.01 ^{c}	22.97± 0.01 ^{c}
AST (nanomoles/mg/min)	8.60± 0.03 ^{a}	10.91± 0.01 ^{b}	11.25± 0.02 ^{c}	12.56± 0.05 ^{c}	12.87± 0.04 ^{c}	10.93± 0.03 ^{b}	12.93± 0.02 ^{c}	12.64± 0.01 ^{c}	12.86± 0.01 ^{c}
ALT (nano moles/mg/min)	9.71± 0.03 ^{a}	11.52± 0.04 ^{b}	12.78± 0.01 ^{c}	13.51± 0.01 ^{c}	13.88± 0.02 ^{c}	12.35± 0.03 ^{b}	12.81± 0.01 ^{c}	13.50± 0.02 ^{c}	13.69± 0.01 ^{c}

Values are expressed in terms of Mean±SD.
Values in the same row with different superscripts vary significantly ($p<0.05$).

diets. Similar findings were reported by Farhoudi et al. (2013), who noted increased protease activity in common carp larvae as an adaptation to protein rich diets. The elevated amylase and lipase levels were attributed to the development of the exocrine pancreas during the growth. Moreover, Rani (2012) emphasized that increased digestive enzyme activities such as protease, amylase, and cellulose support higher nutrient digestibility and retention.

Amino acids serve as essential building blocks of proteins and contribute approximately 14% to 85% of the energy requirements in teleost fish (Ballantyne, 2001). Aspartate aminotransferase (AST) activity is considered a key indicator of amino acid metabolism in fish (Jurss and Bastrop, 1995). In the present study, elevated AST and ALT activity levels observed in SWP50 and BSF50 diet groups may reflect their efficient utilization of amino acids in the experimental diets. Similar findings were reported in Jian carp, where increased AST activity in the hepatopancreas and muscle was linked to active amino acid catabolism (Jiang et al., 2015). Taken together, these findings indicate that the enhanced enzyme activities observed in the present study contributed to improved feed utilization, as reflected in the superior growth performance of fish fed SWP50 and BSF50 diets compared to the control. In addition, an economic evaluation of all experimental diets was conducted using the standard method described by Ardra et al. (2024). The results indicated that SWP50 and BSF50 diets not only enhanced biological performance but also achieved lower production costs and more favorable economic conversion ratios. The economic conversion ratio of all the experimental diets were shown in Table 5.

Table 5. Economical evaluation of all the experimental diets included with graded levels of SWP and BSF as a replacement for fish meal

Experimental diets	Feed cost per kg		Economic Conversion Ratio	
	INR	US$	INR	US$
Control	78	0.93	86.58	1.04
SWP20	72	0.86	66.96	0.80
SWP30	69	0.83	57.27	0.69
SWP40	66	0.79	56.1	0.67
SWP50	63	0.75	50.4	0.60
BSF20	76	0.91	81.32	0.97
BSF30	75	0.90	75.75	0.91
BSF40	74	0.89	65.12	0.78
BSF50	73	0.87	62.05	0.74

ECR, Economic Conversion ratio = FC R × Feed cost

From the above research findings, the results supports the utility of SWP and BSF as effective, eco-friendly alternatives to replace fish meal in aquafeeds. Both insect meals offer a sustainable solution for reducing the aquaculture industry's reliance on wild-sourced fish meal. Additionally, their ability to convert organic waste into high-quality protein makes them environmentally beneficial. Moreover, the enhanced digestive enzyme activities were observed in fishes fed with SWP and BSF included diets, which suggest that these protein sources are efficiently metabolized and utilized by the fishes. Overall, insect larvae meals offer a promising path forward for the development of nutritionally effective and sustainable aquafeeds. In brief both SWP and BSF can be used as a efficient feed ingredient in the nursery rearing of koi carp at a dietary inclusion level of 147 g kg^{-1} with better growth performance and survival rate.

CONCLUSION

The findings of the present study states that insect larvae meals can serve as effective alternative protein sources in aquafeeds, replacing fish meal without compromising growth and survival in koi carp fry. Further, both of the insect meals such as silkworm pupae meal and black soldier fly larvae meal were successfully replaced 50% of fish meal with a dietary inclusion level of 147 g kg^{-1} in the diet of koi carp fry during the nursery phase, resulting in improved growth performance and yield. Moreover, insect meals are more cost-effective than fish meal, which in turn will reduce the overall production costs by lowering feed expenses typically accounting for nearly 60% of the total operating cost in aquaculture.

REFERENCES

Amala, P.P., Khan, U., Ballal, B. and Hassan, M.A. 2018. Black soldier fly *(Hermetia illucens)* pre-pupae meal as a fish meal replacement in the diet for Amur carp *(Cyprinus carpio)*. Journal of the Inland Fisheries Society of India. 50(2): 33-38.

AOAC. 2005. Official Methods of Analysis.18th Edn. Association of Official Analytical Chemists. Washington, D.C, USA.

APHA. 2005. Standard Methods of Water and Wastewater. 21st Edn. American Public Health Association. Washington, D.C, USA.

Ardra, M., Pradhan, C., Das, S. and Pillai, D. 2024. The effect of fish meal replacement with organic acid fermented black soldier fly (*Hermetia illucens*) larvae meal on growth, nutrient utilization, metabolic enzyme activity, antioxidant status and immunity in Pangasius (*Pangasianodon hypophthalmus*). Aquaculture. 591: 741114.

Ballantyne, J.S. 2001. Amino Acid Metabolism. In: Fish Physiology. Eds. Hoar, W.S., Randall, D.J. and Farrell, A.P. Academic Press, New York, p 77-107, ISSN: 1546-5098.

Begum, N.N., Chakraborty, S.C., Zaher, M., Abdul, M.M. and Gupta, M.V. 1994. Replacement of fishmeal by low cost animal protein as a quality fish feed ingredient for Indian major carp, *Labeo rohita*, fingerlings. Journal of the Science of Food and Agriculture. 64(2): 191-197.

Berggren, A., Jansson, A. and Low, M. 2019. Approaching ecological sustainability in the emerging insects-as-food industry. Trends in Ecology and Evolution. 34(2): 132-138.

Cherry, I.S. and Crandall Jr, L.A. 1932. The specificity of pancreatic lipase: its appearance in the blood after pancreatic injury. American Journal of Physiology-Legacy Content. 100(2): 266-273.

Daniel, N. 2018. A review on replacing fish meal in aqua feeds using plant protein sources. International Journal of Fisheries and Aquatic Studies. 6(2): 164-179.

DeFoliart, G., Dunkel, F.U. and Gracer, D. 2009. Toward a Recipe File and Manuals on "How to Collect" Edible Wild Insects in North America. The Food Insects Newsletter. 4(3): 1-9.

Dey, V.K. 2016. The global trade in ornamental fish. INFOFISH International. 20: 164.

Dheke, S. 2013. Growth response of rainbow trout *(Oncorhynchus mykiss)* on substitution of shrimp meal by different protein sources. M.Sc. Theses, Institute of Science and Technology, Tribhuvan University, Kathmandu, Nepal.

Drapeau, G.R. 1976. Protease from *Staphyloccus aureus*. In: Methods in Enzymology. Elsevier, Amsterdam, p 469-475, ISSN: 0076-6879.

Farhoudi, A., Abedian, K.A., Nazari, R.M. and Makhdoomi, C.H. 2013. Changes of digestive enzymes activity in common carp (*Cyprinus carpio*) during larval ontogeny. Iranian Journal of Fisheries Sciences. 12(2): 320-334.

Ghosh, A., Mahapatra, B.K. and Datta, N.C. 2003. Ornamental fish farming - Successful small scale aqua business in India. Aquaculture Asia. 8(3): 14-16.

Henken, A.M., Lucas, H., Tijssen, P. and Machiels, M. 1986. A comparison between methods used to determine the energy content of feed, fish and faeces samples. Aquaculture. 58(3-4): 195-201.

Henry, M., Gasco, L., Piccolo, G. and Fountoulaki, E. 2015. Review on the use of insects in the diet of farmed fish: past and future. Animal Feed Science and Technology. 203: 1-22.

Hodar, A.R., Vasava, R.J., Mahavadiya, D.R. and Joshi, N.H. 2020. Fish meal and fish oil replacement for aqua feed formulation by using alternative sources: A review. Journal of Experimental Zoology India. 23(1): 13-21.

Hua, K., Cobcroft, J.M., Cole, A., Condon, K., Jerry, D.R., Mangott, A., Praeger, C., Vucko, M.J., Zeng, C., Zenger, K. and Strugnell, J.M. 2019. The future of aquatic protein: implications for protein sources in aquaculture diets. One Earth. 1(3): 316-329.

Jain, A., Kaur, V.I. and Hollyappa, S.A. 2019. Effect of dietary supplementation of carrot meal on survival, growth and pigmentation of freshwater ornamental fish, koi carp, *Cyprinus carpio* (L.). Indian Journal of Animal Nutrition. 36(4): 405-413.

Ji, H., Cheng, X., Li, J., Zhang, J. and Liu, C. 2012. Effect of dietary replacement of fish meal protein with silkworm pupae on the growth performance, body composition, and health status of *Cyprinus carpio* var. *specularis* fingerlings. Journal of Fisheries of China. 36(10): 1599-1611.

Ji, H., Zhang, J.L., Huang, J.Q., Cheng, X.F. and Liu, C. 2015. Effect of replacement of dietary fish meal with silkworm pupae meal on growth performance, body composition, intestinal protease activity and health status in juvenile Jian carp *(Cyprinus carpio* var.

jian). Aquaculture Research. 46(5): 1209-1221.

Jiang Jun., Lin Feng., Ling Tang., Yang Liu., Weidan Jiang. and Xiaoqiu Zhou. 2015. Growth rate, body composition, digestive enzymes and transaminase activities, and plasma ammonia concentration of different weight Jian carp (*Cyprinus carpio* var. *jian*). Animal Nutrition. 1(4): 373-377.

Jurss, K. and Bastrop, R. 1995. Amino Acid Metabolism in Fish. In: Biochemistry and Molecular Biology of Fishes. Eds. Hochachka, P.W. and Mommsen, T.P. Elsevier, Amsterdam, p 159-189, ISSN: 1873-0140.

Li, S., Ji, H., Zhang, B., Zhou, J. and Yu, H. 2017. 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. 477: 62-70.

Maceda-Veiga, A., Dominguez-Dominguez, O., Escribano-Alacid, J. and Lyons, J. 2016. The aquarium hobby: can sinners become saints in freshwater fish conservation. Fish and Fisheries. 17(3): 860-874.

Mahapatra, B.K. 2018. Ornamental fishery resources in India: Diversified option for livelihood improvement. In: National seminar on recent trends in fishery and ecological science. Eds. Mahapatra, B.K. and Patra, S.B. 19, May, 2018. p 33-44.

Mamuad, L., Lee, SH., Jeong, C.D., Ramos, S., Miguel, M., Son, A.R., Kim, S.H., Cho, Y.I. and Lee, S.S. 2021. Ornamental fish, *Cyprinus carpio*, fed with fish meal replacement *Ptecticus tenebrifer* and *Tenebrio molitor*. Aquaculture Research. 52(3): 980-990.

Nandeesha, M.C., Gangadhara, B. and Manissery, J.K. 2002. Further studies on the use of mixed feeding schedules with plant and animal based diets for common carp *Cyprinus carpio* (Linnaeus). Aquaculture Research. 33(14): 1157-1162.

Novak, J., Kalous, L. and Patoka, J. 2020. Modern ornamental aquaculture in Europe: early history of freshwater fish imports. Reviews in Aquaculture. 12(4): 2042-2060.

Paul, B.N. and Giri, S.S. 2015. Fresh water aquaculture nutrition research in India. Indian Journal of Animal Nutrition. 32(2): 113-125.

Prakash, S., Ajithkumar, T.T., Raghavan, R., Rhyne, A., Tlusty, M.F. and Subramaniam, T. 2017. Marine aquarium trade in India: Challenges and opportunities for conservation and policy. Marine Policy. 77: 120-129.

Rani, S. 2012. Effect of replacement of fish meal in commercial aquafeed by soybean on growth and metabolism of most valuable freshwater cultivated fish species. University Grants Commission, F.No. 41-123/2012(SR), New Delhi, India.

Renna, M., Schiavone, A., Gai, F., Dabbou, S., Lussiana, C., Malfatto, V. and DeMarco, M. 2017. Evaluation of the suitability of a partially defatted black soldier fly *(Hermetia illucens)* larvae meal as ingredient for rainbow trout *(Oncorhynchus mykiss* Walbaum*)* diets. Journal of animal science and biotechnology. 8(1): 1-13.

Rick, W. and Stegbauer, H.P. 1974. Amylase Measurement of Reducing Groups. In: Methods of Enzymatic Analysis. Eds. Bergmeyer, H.V. Academic Press, New York, p 885-889, ISBN: 978-0-12-091302-2.

Sahib, Q.S., Ahmed, H.A., Beigh, Y.A., Shah, S.M., Ganai, A.M., Farooq, J. and Sheikh, G.G. 2024. Silkworm Pupae Meal: A Potential Unconventional Protein Source for Animal Feeding. Indian Journal of Animal Nutrition. 41(1): 11-28.

Tacon, A.G.J. and Metian, M. 2015. Feed matters: satisfying the feed demand of aquaculture. Reviews in Fisheries Science & Aquaculture. 23(1): 1-10.

Tran, G., Heuze, V. and Makkar, H. 2015. Insects in fish diets. Animal Frontiers. 5(2): 37-44.

Wooten, I.D.P. 1964. Microanalysis in Medical Biochemistry. Churchill Ltd, Gloucester Place, London.

Xu, X., Ji, H., Yu, H. and Zhou, J. 2020. Influence of dietary black soldier fly *(Hermetia illucens* Linnaeus*)* pulp on growth performance, antioxidant capacity and intestinal health of juvenile mirror carp *(Cyprinus carpio* var. *specularis)*. Aquaculture Nutrition. 26(2): 432-443.

Zhou, J.S., Liu, S.S., Ji, H. and Yu, H.B. 2018. Effect of replacing dietary fish meal with black soldier fly larvae meal on growth and fatty acid composition of Jian carp *(Cyprinus carpio* var. *jian)*. Aquaculture Nutrition. 24(1): 424-433.