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Nutritional Evaluation and Cost Analysis of Dietary Replacement of Fish Meal with Black Soldier Fly Larvae (*Hermetia illucens)* in the Diet of GIF Tilapia

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GKR: Conducted the trial and analysis, analyzed the data and drafted the manuscript
SA, JJ, KS, RJ: Conceptualized and designed the study, and corrected the manuscript
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Key words
Fish meal, Black soldier fly larvae meal, GIF Tilapia, Growth performance, Protein source

ABSTRACT

Fish meal is one of the limiting factors in the aquaculture feed industry for the formulated diets. So, the present study endeavored to investigate the efficacy of dietary replacement of fish meal with black solider fly larvae at graded levels on feed intake, growth of GIF tilapia and economic analysis of feed. The study was undertaken in a complete randomized design (CRD) and each treatment was triplicated. Six isonitrogenous diets were formulated with black solider fly larvae to substitute fish meal (FM) protein at 0%, 20%, 40%, 60%, 80% and 100%. The GIF tilapia in each replicate were fed twice a day at a rate of 5% of their body weight and reared for a period of 90 days. GIF tilapia fed with 40% diet of black solider fly (BSF) larvae exhibited maximum growth performance and feed utilization without compromising the price of the diet. However, escalating the percentage of FM with BSF above 60% led to significant reduction in growth and feed utilization of GIF tilapia. Thus, this study suggests that GIF tilapia fed with BSF will help the aquaculturists to develop low cost feed.

INTRODUCTION

Fisheries and aquaculture remain as an important source of food, nutrition, income and livelihood for coastal fisher folks around the world. Global fish production is estimated to have reached about 185.4 million tonnes in 2022 (FAO, 2024). Aquaculture sector is steadily growing and ensures most of all fish produced to be consumed by human kind. Consumption of fish provides energy, protein and a range of essential nutrients. Eating fish is part of the cultural traditions of many peoples and fish and fishery products are a major source of food and essential nutrients for some populations. In many cases, there may be no alternative affordable food sources for many of these essential nutrients (FAO, 2014). Apart from high prices, the aquaculture industry is in competition for fish meal (FM) because FM is used for animal production and is a primary protein source in the diets of cattle, poultry and pigs (FAO, 2007).

Aquaculture has thus become more integrated into the global food system, with rapid growth in production and major transformations in feed ingredients and production technologies (Rosamond *et al*., 2021). The supplied diet must contain the necessary nutritional requirements for the fish to ensure the best growth (Ghomi *et al*., 2012). World aquaculture is the fastest growing food-producing sector in the world. Globally, aquaculture is expanding into new directions, identifying and diversifying. With increasing demand for environment friendly aquaculture, the use of growth promoters in fish nutrition is new widely accepted (Shreeja *et al*., 2018). In search for viable alternative feedstuffs to FM for aqua feeds, candidate ingredients must possess certain characteristics, including wide availability, competitive price, as well as ease of handling, shipping, storage and use in feed production (Gatlin *et al*., 2007).

The black soldier fly (BSF, *Hermetia illucens*) larvae are considered important candidate species to be used for animal feeds (Cammack and Tomberlin, 2017; Van Huis, 2013). The research and industrial-scale production of BSF larvae as feed ingredients have been intensified during the last few years (FAO, 2013; Wang and Shelomi, 2017). Its larvae can be reared on a wide range of organic

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(waste)-material, which reduce the volume of this waste by up to 50%, producing biomass with a protein content of about 42% and a fat content of up to 35 % (Sheppard *et al*., 1994). Successful and sustainable culture of finfish and shellfish depends on the use of nutritionally balanced, low-cost and eco-friendlyfeeds (Joseph and Ignatius, 2016). Dietary nutrient requirements in fish are usually estimated empirically by feeding graded levels of a specific nutrient, in a basal diet containing a different level of that nutrient and then measuring growth, feed intake, body nutrient stores or other variables (Lekshmi and Prasad, 2014). In this context, the present study was focused to estimate the effects of dietary replacement of FM with BSF larvae meal on feed intake, growth of GIF tilapia and economic analysis of feed.

MATERIALS AND METHODS

Experimental fish and feeding trial

The experimental fish (GIF tilapia) were procured from a private fish farm at Madurai, Tamil Nadu, India. Prior to experiment, the fishes were graded to select an individual average weight of 6.50± 0.27 g and were stocked in cement tanks. Five hundred and forty fishes were distributed in 18 groups with thirty fishes in each cement tank. The cement tank had a water holding capacity of 1 ton in which the fishes were stocked for the feeding trial. Fishes were fed twice a day at a feeding frequency of 5% of fish body weight and adjusted with subsequent samplings. During the growth trial, the water quality parameters were monitored which were as follows: dissolved oxygen (DO) 5.70±0.06 mg/L, temperature 28.35±0.04°C, pH 7.75±0.21, alkalinity 123.06±1.63 mg/l, hardness 63.32±0.56 and ammonia-nil.

Experimental diets

Six different FM and BSF larvae based isonitrogenous experimental diets were formulated to contain 32% of crude protein (Table I). The experimental diets were supplemented with BSF larvae at levels of 0 (T1), 7 (T2), 14 (T3), 21 (T4), 28 (T5) and 35 (T6) to replace FM protein at 0, 20%, 40%, 60%, 80% and 100%, respectively. The experimental diet T0 observed as a control diet with 35 g/kg inclusion of FM and excluding the addition of BSF larvae. Dietary feed ingredients were finely ground, thoroughly mixed and then extruded at 60-70°C using a single screw extruder. The air tight plastic containers were utilized to store all the experimental diets at room temperature.

Growth performance

For growth analysis, the fishes were collected from all tanks using hand net and the weight (g) was measured in a weighing balance with an accuracy of 0.01g. At the end of the experimental trail, growth performances were calculated and assessed by means of weight gain (WG), specific growth rate (SGR), protein efficiency ratio (PER), thermal growth coefficient (TGC) and survival rate (SR) were calculated according to Zaid and Sogbesan (2010).

Table I. Formulation and chemical composition of the experimental diets.

Ingredients	Control	Fish meal replacement with black solider fly larvae				
		20%	40%	60%	80%	100%
Fish meal	35	28	21	14	7	0
Black solider fly larvae	0	7	14	21	28	35
Ground nut oil cake	5	8	10	14	17	20
Rice bran	20	20	20	20	20	20
Wheat flour	25	10	14	10	17	18
Corn flour	14	26	20	20	10	6
Vitamin + mineral mix	1	1	1	1	1	1
Proximate composition						
Moisture (%)	9.70	9.10	9.41	10.04	10.36	11.20
Crude protein (%)	32.16	32.41	31.94	32.05	31.91	31.93
Crude fat (%)	11.24	7.98	8.13	7.85	8.75	8.15
Crude fiber (%)	10.01	10.11	12.13	11.67	11.74	11.21
Ash (%)	13.83	20.15	17.30	18.00	15.54	16.23
CHO (%)	23.60	20.65	21.75	20.85	21.95	22.25
Gross energy (kcal/100 g)	386	385.15	388.10	397.30	393.25	393.80

Table II. Bio growth performances of GIF tilapia fed under different inclusion level of black soldier fly larvae meal.

Parameters	C1 (0 %)	T1 (20 %)	T2 (40 %)	T3 (60 %)	T4 (80 %)	T5 (100 %)
Survival(%)	91.66 ± 1.66 [a]	89.99 ± 3.33 [a]	93.33± 3.33 [a]	93.33[a]	94.99 ± 1.66 [a]	91.66 ± 5 [a]
Initial body weight (g)	6.50± 0.27 [a]	6.10 ± 0.09 [a]	6.10 ±0.01 [a]	6.27 ± 0.03 [a]	6.53± 0.08 [a]	6.13 ±0.07 [a]
Final body weight (g)	146.05±0.83[de]	142.63±0.4 [d]	154.26±0.49 [f]	148.65±0.6[e]	129.66±1.31 [b]	124.11±1.10[a]
Weight gain (WG)(g)	139.55±0.56[de]	136.52±0.30 [d]	148.16±0.48 [f]	142.38±0.6 [e]	123.13±1.23 [b]	117.98±1.03[a]
Weight gain (WG)(%)	46.05±0.83 [de]	42.63±0.4 [d]	54.26±0.49 [f]	48.65±0.67 [e]	29.66±1.31 [b]	24.11±1.10 [a]
ADG(%)	1.55±0.006 [de]	1.51±0.003 [d]	1.64±0.005 [f]	1.58±0.007 [e]	1.36±0.013 [b]	1.31±0.011 [a]
SGR(%)	3.45±0.04 [b]	3.50±0.01 [b]	3.58±0.001 [c]	3.51[bc]	3.32±0.002 [a]	3.34±0.004 [a]
FCR	1.50±0.006 [a]	1.51±0.004[ab]	1.48±0.002 [a]	1.49±0.001 [a]	1.56±0.007 [c]	1.57±0.002 [c]
FER	0.66±0.00[de]	0.66±0.002[cd]	0.67±0.00[e]	0.66±0.01[de]	0.64±0.00[ab]	0.63±0.00[a]
PER	4.36±0.01 [de]	4.26±0.009 [d]	4.63±0.015 [f]	4.44±0.02 [e]	3.84±0.03 [b]	3.68±0.03 [a]
TGC	1.58±0.04 [a]	1.83±0.01[bc]	1.82±0.03 [b]	1.98±0.03 [c]	1.80±0.03[b]	1.65±0.01 [a]

Values are expressed as Mean ± SE. Values with different superscripts differ significantly ($P < 0.05$) for each parameter. One-way ANOVA was used by following Duncan multiple range test for post hoc analysis. WG, weight gain; SGR, specific growth rate; PER, protein efficiency ratio; SR, survival rate; FCR, feed conversion ratio; TGC, Thermal growth coefficient.

Economic analysis

Cost analysis for different test diets were performed for 1000 g of feed. Comparison of weight gain and cost was studied to estimate the feasibility for aquaculture enterprises.

Statistical analysis

All the data were tested for normality (Shapiro-Wilk test) and homogeneity of variance (Levene's test). The data were expressed as Mean±SEM. Statistical test were performed using SPSS software version 20.0 at 5% level of significance, following Student's t-test.

RESULTS AND DISCUSSION

Growth performance and feed utilization:

Growth performance and feed utilization of GIF tilapia after 90 days trial are shown in Table II. Significant difference was observed in WG, FCR, PER, SGR and TGC of fishes fed with BSFLM supplemented diets and control diet. The mean WG of the fishes fed with BSFLM at 40 % was 154.26 ± 0.49 g which is higher than that of the fishes fed with control feed (146.05 ± 0.83 g). The mean WG of fishes fed with other concentrations like 80 and 100 % showed decreasing trend. When SGR of the fishes fed with BSFLM are considered, 40% yielded the best SGR (3.58 ± 0.001) rather than the control and the other treatments.

According to Henry *et al*. (2015) and Barroso *et al*. (2014), insect meals such as the BSFM have comparable nutritional qualities to FM. It has been demonstrated that freshwater crustacean diets can substitute invertebrate protein sources for FM (Riddick, 2014). Thus, it is advised to use BSLM in place of FM. Guerreiro *et al*. (2021) found that the concentration of 100 g/kg BSLM had no negative effects on *Argyrosomus regius* when it was substituted for the dietary inclusion levels of BSLM at 100, 200, and 300 g/ kg with 17%, 35%, and 52% of FM. These results are similar to that of present results which showed that the 40 % inclusion of BSLM in the diets of GIFT showed higher growth performance. But according to Zhou *et al*. (2018), 20% substitution of BSLM for fish meal resulted in inferior growth performance in Jian carp because BSLM contains chitin. According to Newton *et al*. (2005) feeding 100% larvae did not provide enough dry matter or protein intake for channel catfish grown in tanks to allow sufficient growth. A comparison between menhaden FM (menhaden fish meal comes from the dried tissue of menhaden species) and BSFLM showed that the latter could be advantageous as a replacement for FM.

Muin *et al*. (2017) found that switching from FM to BSLM may be successful up to 50% inclusion levels without experiencing any negative consequences. The total replacement of FM by BSLM was studied over a 59-day period using juvenile Nile tilapia. This result was similar to our results which showed 40% inclusion showed higher growth performance and beyond that showed lower body weight gain and higher FCR.As indicated by Kroeckel *et al*. (2012) growth and feed intake decreased in juvenile turbot (*Psetta maxima*) as BSFLM increasingly substituted (16.5-75.6%) for FM in the diet. They recorded a significant reduction in feed intake, coupled with lower protein digestibility and growth performance at higher BSFM levels (330 g/kg fishmeal) in Turbot (*Psetta maximus*). They also suggested higher rates reduced intake and nutrient availability, with a further reduction in growth rate, possibly because of the presence of chitin.

Table III. Economic analysis of black soldier fly larvae meal (BSLM) incorporated experimental diets and control diets.

Variables	C1 (Control)	T1 (20 %)	T2 (40 %)	T3 (60 %)	T4 (80 %)	T5 (100 %)
Seed cost (Rs)	180	180	180	180	180	180
Feed cost (Rs)	1041.85	1023	1088.95	1051.55	956.03	921.42
Total cost (Rs)	1221.85	1203	1268.95	1231.55	1136.03	1101.42
Total feed consumed (kg)	12.63	12.40	13.20	12.75	11.59	11.17
Total production (kg)	8.76	8.55	9.25	8.91	7.77	7.44
Selling price (Rs / kg)	150	150	150	150	150	150
Total amount realized (Rs)	1314.49	1283.67	1388.38	1337.89	1166.98	1117.03
Net income (Rs)	92.63	80.66	119.42	106.33	30.94	15.61
Cost of production / kg (Rs)	139.42	140.57	129.97	138.07	146.02	147.90

Diets comprising BSLM (0%, 25%, 50%, 75%, and 100%) meal added to *D. rerio* larvae showed no effect on zebra fish growth (Zarantoniello *et al*., 2020). Up to 50% of defatted BSLM fed to *C. carpio* significantly boosted growth without having any adverse effects, however 75% of the fish group receiving BSLM showed intestinal damage and problems related to nutritional stress (Li *et al*., 2017). Weththasinghe *et al*. (2021) observed a linear decrease in protein and lipid digestibility, protein efficiency ratio and lipid retention in extruded diets for Atlantic salmon as the level of dietary *Hermetia illucens* meal increases. Thus the results of this research are in coherent with those of previous studies, suggesting an optimum fish meal replacement level of 20–50% using insect meal (Wing-Keong *et al*., 2001; St Hilaire *et al*., 2007b; Cummins *et al*., 2017) in different aquaculture species. Similar to our findings, the previously mentioned studies also reported the limitations in replacing fish meal beyond 50% of diet using insect meal.

Economic analysis

Table III shows the cost analysis of test diets. In this study the cost of diets BSF 20%, BSF 40%, BSF 60%, BSF 80%, BSF 100% and control are Rs.140.57, Rs.129.97, Rs.138.07, Rs.146.02, Rs.147.90 and Rs.139.4, respectively.

In the present study the cost of BSF 100 % is higher (Rs.147.90) when compared to BSF 80% (Rs.146.02). Considering weight gain into account BSF 40% shows higher growth in which its cost is Rs.129.97. Due to high demand and less availability of FM, an alternative protein source like BSFLM can be effectively used in fish feed preparation. Practically, in field conditions farmer seeks for a cost effective feed with higher growth of animal. In such manner, this study suggests replacing FM at 40% with BSFLM without compromising the price of the diet when compared to zero replacement of BSFLM. Thus, this study will help to develop low cost aquaculture enterprise.

CONCLUSION

Many studies have reported using black solider fly meal in different forms as a feed additive in the animal and aquaculture industry. However, the results of the present study suggested that the dietary inclusion of 40% of black solider fly larvae could enhance feed intake and growth performance of GIF tilapia. With regard to cost factor the BSFLM is promising feed ingredient without affecting the profit. The aquaculture sector can lower production costs without sacrificing the quality of fish by substituting black soldier fly larvae feed.

DECLARATIONS

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IRB approval

The study was approved by Institutional Review Board of Tamil Nadu Dr. J. Jayalalithaa Fisheries University, Tamil Nadu, India.

Ethical approval

After the approval of the statutory authorities of the Tamil Nadu Dr. J. Jayalalithaa Fisheries University, Nagapattinam, Tamil Nadu, India, the research work was carried out in adherence with the current animal welfare laws in India. The care and treatment of the experimental animal was carried out by guidelines of the CPCSEA (Committee for the Purpose of Control and Supervision of Experiments on Animals), Ministry of Environment and Forests (Animal Welfare Division, Govt. of India).

Data availability statement

The data that support the findings of this study are available within the article.

Statement of conflict of interest

The authors declare that they have no conflict of interest.

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