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Research Article

Black soldier fly (*Hermetia illucens*) larvae meal improves growth performance, feed utilization, amino acids profile, and economic benefits of Nile tilapia (*Oreochromis niloticus,* L.)

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ABSTRACT

This study investigated the effect of replacing soybean meal (SBM) with black soldier fly larvae meal (BSFLM) on the growth performance, feed utilization, carcass body composition, and amino acids profile of Nile tilapia (*Oreochromis niloticus)*. Three isonitrogenous (30% crude protein) diets containing BSFLM in varying proportions of 0% ($BSFLM_0$), 50% ($BSFLM_{50}$), and 100% ($BSFLM_{100}$), were formulated to replace SBM. A commercial diet ($COMM_0$) sourced from the local market was used as a positive control. Male sex-reversed *O. niloticus* juveniles of a mean weight 20.88 ±0.16 g were stocked in 12 cages each at a density of 12.5 fish m^{-3}. Fish were hand-fed at 5% (28 days), 3% (54 days), and 2.5% (84 days) of the body weight twice a day (1000 hrs and 1600 hrs). Significant differences ($P < 0.05$) were found in the final body weight, body weight gain (BWG), specific growth rate (SGR), feed conversion ratio (FCR), survival rate, and condition factor (K). The best growth performance and feed utilization was recorded in fish fed on $BSFLM_{100}$. The different diets had significant effects on the body composition and amino acid profiles of the experimental fish ($P<0.05$). Fish fed on $BSFLM_{100}$ exhibited highest values for phenylalanine, threonine, Isoleucine, lysine, proline, and glutamic acid amino acids. The partial enterprise budget analysis indicated that replacing SBM with BSFLM at 50% and 100% reduced the cost of production compared to the control diet ($BSFLM_0$) and commercial diet ($COMM_0$). The study demonstrated that BSFLM is a cost-effective alternative to SBM in the diets of *Oreochromis niloticus* hence can replace soybean meal up to 100% without negative effect on growth and carcass body composition.

Keywords: Black soldier fly, *Hermetia illucens*, Nile tilapia, Soybean meal

Introduction

Aquaculture plays a crucial role in global food security and has further demonstrated potential to meet future food demand, particularly the need for protein (FAO, 2020). However, aquaculture production has been constrained by the over dependence on fish meal and fish oil as the main ingredients in fish feeds (Chang et al., 2016). Fish meal has been the major protein source in fish feeds due to its high-quality protein, well-balanced amino acid profile, high essential fatty acids, and phospholipids near to requirement levels of most cultivated aquatic organisms (Bruni et al., 2021; Luthada-Raswiswi et al., 2021). A steady decline in capture fisheries and the competition with humans and animal feed industry have resulted to rapid decrease in the global supplies of fish meal and fish oil, leading to high costs (Dee Roos et al., 2017). This in turn has prompted the search for cheaper alternative protein sources for fish feed formulation.

Soybean meal has become the key ingredient used in tilapia feeds following technological innovation in feed production and high incentives for seeking high quality and affordable alternatives for fish meal and fish oil (Chang et al., 2016). The extensive utilization of soybean meal (SBM) is attributed to its well-balanced amino acid profile and relatively high crude protein contents (Arriaga-Hernández et al., 2021) with the latter being identified as a key and most expensive dietary component in the production of fish feeds. Despite soybean's pivotal role in animal production, its use in fish feeds is constrained by the competition from the livestock feed industry, human food consumption, and industrial production of ethanol and bio-diesel (Lu et al., 2020). This has contributed to the increasing scarcity of soya beans coupled with the effects of climate change and high land-use competition.

Moreover, soybean meal is characterized by nutritional deficiencies with lysine, methionine and threonine being limited in soy-based fish diets (Malcorps et al., 2019). Great amounts of anti-nutritional components in soya have been associated with reduced efficiencies in protein and lipid utilization and digestion, mineral utilization, and anti-vitamins consequently, leading to depressed growth of fish (He et al., 2020). Locally, limited soybean processing has rendered SBM susceptible to contamination, and growth of mycotoxin producing molds mainly aflatoxins that compromises fish health and growth when used in feeds (Niyibituronsa et al., 2016).

Nonetheless, studies have demonstrated that insects represent a valid substitute for the global problems associated with the expensive, unsustainable protein sources for fish feed (Huis et al., 2013). Among insect species, black soldier fly (*Hermetia illucens)* is considered as one of the most promising protein source substitute due to its wide distribution in both tropical and subtropical climate zones (Zurbrügg et al., 2018). It is also easy to culture due to the capability to convert various forms of organic wastes into biomass of high quality proteins, essential amino acids (EAA), fat, vitamins, and minerals (Gasco et al., 2019). The Black soldier fly (BSF) also reproduces easily, has a high growth rate, can control bacteria favoring low risk of zoonotic disease transmission and in addition has a prebiotic effect on the fish (Gariglio et al., 2019).

The nutritive value of *H. illucens* larvae in aquafeeds has been tested with success on several fish species including Nile tilapia (Devic et al., 2018; Agbohessou et al., 2021; Taufek, et al., 2021; Tippayadara et al., 2021), Pacific white shrimp (*Litopenaeus vannamei)*, Cummins et al., 2017), Jian carp (*Cyprinus carpio*) (Zhou et al., 2018), Atlantic salmon (*Salmo salar)* (Li et al.,2020) and Juvenile grass carp (*Ctenopharyngodon idella*) (Lu et al., 2020). However, most of the studies have focused on fish meal protein replacement with black soldier fly larvae meal (BSFLM). Thus scarcity of information on the effects of replacing SBM with BSFLM in the diets of Nile tilapia. In addition, most of the studies are focused on a specific life stage of the fish in different culture systems, especially at the larval rearing and juvenile stage hence discrepancies on the best BSFLM inclusion levels for grow out fish. This study, therefore, evaluated the utilization of defatted black soldier fly larvae (*H. illucens*) meal as SBM replacement in the diet of grow-out Nile tilapia in terms of growth performance and economic aspects during the entire production cycle.

Material and Methods

Study Area and Diet Preparation

The experiment was conducted at Kenya Marine and Fisheries Research Institute, (KMFRI) Sagana Aquaculture Centre (0º19´S and 37º12´E), Kirinyaga County, Kenya. The cultured BSFL was processed by Biobuu Limited, Kilifi, Kenya. Other feed ingredients (Table 1) were purchased from the local agrovet shop and ground separately into finer particles using hammer mill (Thomas-Wiley intermediate mill, 3348-L10 series, USA). Proximate analysis of test ingredients was conducted to guide formulation of three isonitrogenous (30 % crude protein) experimental diets. The diets were prepared by replacing SBM with BSFLM at 0% ($BSFLM_0$) (Control), 50% ($BSFLM_{50}$) and 100% ($BSFLM_{100}$). A commercial diet ($COMM_0$), sourced from local feed manufacturer in Nairobi was used as a positive control. In the production of diets: $BSFLM_0$, $BSFLM_{50}$ and $BSFLM_{100}$, the ingredients were mixed thoroughly with water to make homogenous dough and pelletized using 2-4 mm diet commercial pelletizing machine into floating pellets. The pellets were sundried, packed and stored in clean, dry and cool environment.

Proximate Analysis of the Experimental Diets and Fish Body Composition

The analysis of experimental diets, the initial and final fish body composition was carried out in triplicates following standard methods by the Association of Official Analytical Chemists (AOAC, 1995). Dry matter (DM) was determined gravimetrically by reweighing 2 grams of the sample that had been oven-dried for six hours to constant weight at 105°C. Crude protein (CP) was analyzed by Copper Catalyst Kjeldahl Method. Analysis was done by taking 1g of each sample and 2 tablets of catalyst (Kjeltabs) which were digested in 15 ml concentrated sulfuric acid at 420°C. The samples were cooled and automatically distilled in Kjeldahl equipment with 40% NaOH and ammonia gas trapped in 4% Boric acid was reverse titrated using 0.2N HCl. The nitrogen content was determined and converted to crude protein content using a nitrogen factor for the crude protein calculation of 6.25. Ash was determined by expressing the weight of 2 g of the ground sample burnt at 600°C for 3 hours in a muffle furnace as a percentage of the un-burnt sample weight. Crude fat was extracted by heating 3g of the sample in diethyl ether under reflux at 105°C for 30 min in a VELP Solvent Extraction unit. Ether extract was calculated as the difference between the original sample and the ether extract residue. Crude fibre (CF) was determined gravimetrically by chemical digestion and solubilization, and quantified by: CF (%) = dried sample (g) – ashed sample (g)/initial sample weight × 100). Nitrogen free extract (NFE) was determined by the difference between the original weight of the sample and sum of the weights of its moisture, crude protein (CP), crude fat (CF), ash and crude fibre as: 100 – (crude protein + crude fat + crude fibre + moisture + ash). Amino acids were analyzed using ion exchange liquid chromatography via continuous flow chromatography. The compounds were identified and quantified using an authentic sample mixture (amino acid standard solution (AAS 18) from Sigma-Aldrich (Chemie GmbH, Munich, Germany).

Experimental Design

A total of six hundred sex reversed male Nile tilapia (*O. niloticus)* juveniles of 20.88 ±0.16 g mean weight were sourced from Kenya Marine and Fisheries Research Institute and acclimatized for seven days prior to the experiment. The experimental fish were randomly stocked in 12 cages each at a density of 12.5 fish/m^3 with four cages suspended in 150 m^2 earthen pond. The fish were randomly assigned to the four experimental diets containing BSFLM in varying proportions of 0% (BSFLM$_0$), 50% (BSFLM$_{50}$), 100 % BSFLM$_{100}$) and a commercial diet (COMM$_0$) in triplicates. The experimental fish were hand fed twice a day (1000 hrs and 1600 hrs) at 5 % of body weight for 28 days, 3 % of body weight for 54 days and 2.5 % of body weight for 84 days.

Table 1. Ingredients and proximate composition of the experimental diets

Ingredient inclusion (g kg^{-1})	**BSFLM$_0$**	**BSFLM$_{50}$**	**BSFLM$_{100}$**	
Wheat bran	28	28	28	
Soybean meal	30	15	0	
Black soldier fly larvae meal	0	15	30	
Maize germ	18	18	18	
Freshwater shrimp	12	12	12	
Sunflower seed cake	9	9	9	
Mono-calcium phosphate	1	1	1	
Vitamin premix	1	1	1	
Soybean oil	1	1	1	
Proximate composition (%)	**BSFLM$_0$**	**BSFLM50**	**BSFLM$_{100}$**	**COMM$_0$**
Crude protein	30.2 ±0.32	30.28 ±0.61	30.34 ±0.15	30.5 ±0.22
Crude fat	6.35 ±0.03	6.31 ±0.02	6.46 ±0.00	6.8 ±0.00
Ash (dry)	7.75 ±0.26	7.99 ±0.12	8.23 ±0.42	7.3 ±0.16
Nitrogen free extract (NFE)	42.00	45.40	45.90	48.10

*Black soldier fly treatments - BSFLM$_0$ (0%); BSFLM$_{50}$ (50%); BSFLM$_{100}$ (100%); COMM$_0$ (Commercial feed)
*Soybean treament- BSFLM$_0$ (100%); BSFLM$_{50}$ (50%); BSFLM$_{100}$ (0%); COMM$_0$ (Commercial feed)

Sampling

Fish sampling was done every 28 days. The individual length (cm) and weight (g) of the fish were determined using a measuring board and an electronic weighing scale (model EHB-3000, China) respectively. To evaluate the growth and feed efficiency the following standard formulas were used (Hopkins, 1992):

Specific growth rate (SGR, %) = 100 × [(Ln BW final (g) – Ln BW initial (g)) / days of experiment] … (1)

Body weight gain (BWG, g) = Final weight (g) - Initial weight (g)……………………..………..(2)

Feed conversion ratio (FCR) = feed provided/live weight gain (g) … (3)

Fulton's condition factor (K) = $100(W/L^3)$……………(4)

Length- weight relationship was determined by the formula=$W = aL^b$, where W = weight (g) and L = total length (cm), a is the regression slope and b is the y intercept……….(5)

Percentage Survival (%) = 100 × (final number of fish)/ (initial number of fish)……….......(6)

Water quality parameters were measured weekly at 1000 hrs using a multi-parameter water quality meter (YSI industries, Yellow Springs, OH, USA). Water samples were collected using 250 ml plastic bottles after every two weeks for ammonia, nitrites, nitrates and phosphorus analysis following the procedures by APHA (1995). The mean values of water quality parameters of the pond water were stable with minimal variations during the experiment period. The mean values were as follows: Temperature (25.31 ±0.22°C), dissolved oxygen (DO) (5.47 ±0.08 mg L^{-1}), conductivity (104.60 ±1.68 µS cm^{-1}), total dissolved solids (67.05 ±1.08 mg L^{-1}), and pH (8.35 ±0.04). Relatively low values for nutrients were recorded as follows: Phosphates (0.003 ±0.0001 mg L^{-1}), nitrites (0.001 ±0.0001 mg L^{-1}), Nitrates (0.004 ±0.0003 mg L^{-1}) and Ammonium (0.002 ±0.0001 mg L^{-1}). All the parameters were within the recommended levels for Nile tilapia.

Enterprise Budget Analysis

Partial enterprise budget analysis was used to evaluate the economic implications of substituting SBM with BSFLM in the diets of Nile tilapia. Variable costs included the cost of labor, feeds, and fingerlings (Table 2). The cost of formulated diets ($BSFLM_0$, $BSFLM_{50}$, and $BSFLM_{100}$) was calculated based on the cost of the ingredients in each diet and the cost of feed production while the price of the commercial diet was based on feed company retail prices. Labor costs were based on the prevailing market prices within the experiment site. The US dollar exchange rate against Kenya shillings was pegged at KES 107.15. The study assumed that all other costs of production were constant for all dietary treatments and thus not considered. Fish harvested per treatment was sold at USD 3.73 kg^{-1}, which is the market price of Nile tilapia. Further, to gauge the profitability of the alternative the break-even price was calculated by dividing total costs per unit price of fish (Musa et al.,2021).

Data Analysis

The data was cleaned and subjected to the Shapiro–Wilk test of normality. Statistical analyses were performed using MS excel and Statistical Package for the Social Sciences (SPSS version 23). Mean comparisons were done by the analysis of variance (one-way ANOVA) followed by Tukey HSD post hoc test to determine the pairwise differences among the diets. Significant differences were considered at $P < 0.05$. Percentage data were arcsine transformed prior to analysis to normalize data.

Table 2. Partial enterprise budget input expenditures

Item	Units	Quantity	Unit cost (USD)	Total(USD)
Nile tilapia fingerlings	Pcs/cage	50	0.09	4.5
$BSFLM_0$	Kg/cage	12.57	1.06	13.32
$BSFLM_{50}$	Kg/cage	12.68	0.95	12.05
$BSFLM_{100}$	Kg/cage	13.74	0.86	11.82
$COMM_0$	Kg/cage	13.31	0.94	12.51
Labor costs	USD/cage/Month	6	0.4	2.4
Cost of packaging fish	USD/Kg	-	0.009	-
Cost of transporting fish	USD/Kg	-	0.027	-
Tax on sale of fish	USd/Diet	-	0.47	-
Total yield $BSFLM_0$	Kg	17.77		
Total yield $BSFLM_{50}$	Kg	16.08		
Total yield $BSFLM_{100}$	Kg	20.54		
Total yield $COMM_0$	Kg	20.81		

Results and Discussion

Growth Performance and Nutrient Utilization

The BSFLM replacement level had a significant effect on the growth of the fish ($P < 0.05$). $BSFLM_{100}$ recorded the highest weight gain followed by $COMM_0$, $BSFLM_{50}$ and finally $BSFLM_0$ (Table 3). Fish body weight increased by 89 % in fish fed on $BSFLM_{100}$ and $COMM_0$, while an increase of 87% was observed in fish fed on $BSFLM_0$ and $BSFLM_{50}$. Fish fed on $BSFLM_{100}$ had significantly higher BWG and SGR ($P<0.05$) and the lowest FCR compared to those fed on $BSFLM_0$ and $BSFLM_{50}$. Fish fed on $BSFLM_0$ presented significantly lower BWG, SGR and the highest FCR. All the diets recorded a positive correlation co-efficient (R^2) that was not significantly different among the diets ($P > 0.05$). The values of condition factor recorded for all the fish ranged between 1.86 and 1.89. There were significant differences ($P<0.05$) in Fulton's condition factor (K) between the $BSFLM_0$, $BSFLM_{50}$, $BSFLM_{100}$, and $COMM_0$ with control diet ($COMM_0$) having the highest condition factor. Survival rates were relatively higher in all the treatments with significant variations ($P<0.05$) among the diets. The highest survival rates were recorded in diet $BSFLM_0$ (95.33%) while the lowest in $BSFLM_{100}$ (92.67 %).

Body Composition Analysis

Significant difference ($P<0.05$) was recorded for crude protein, crude fat, and Nitrogen free extracts (NFE) in the body composition of the fish (Table 4). However, no significant differences ($P>0.05$) was recorded in ash content in fish in all the treatments. Fish fed on $BSFLM_{100}$ exhibited significantly ($P<0.05$) higher values for phenylalanine, threonine, isoleucine, lysine, histidine and leucine, while for non-essential amino acids: glutamic acid, cysteine, proline were higher in fish fed in $BSFLM_{100}$ as compared to control diets ($COMM_0$ and $BSFLM_0$) (Table 5). Fish fed on $COMM_0$ registered highest mean values for arginine, glycine, methionine, tyrosine, tryptophan and tyrosine although, they were statistically similar to fish fed on $BSFLM_{100}$. The lowest values for glycine, alanine, lysine, cysteine, glutamic acid, methionine, and isoleucine were registered in fish fed on $BSFLM_{50}$ while arginine, histidine, tryptophan, leucine threonine, proline were lowest in fish fed on $BSFLM_0$ (control).

Table 3. Growth performance parameters of *O. niloticus* fed on diets with different levels of defatted black soldier fly larvae meal and commercial diet

Parameter	$BSFLM_0$	$BSFLM_{50}$	$BSFLM_{100}$	$COMM_0$
Initial length (cm)	10.57 ±0.07	10.54 ±0.06	10.57 ± 0.06	10.56 ± 0.05
Initial weight (g)	20.83 ±0.33	20.86 ±0.33	20.85 ±0.33	20.88 ±0.30
Final length (cm)	21.04 ±0.13^a	21.35 ±0.13^a	21.67 ±0.12^b	21.53 ±0.17^b
Final weight (g)	166.19 ±3.18^a	169.03 ±2.91^a	193.35 ±3.69^b	188.56 ±5.46^b
SGR (% day $^{-1}$)	1.23 ±0.02^a	1.24 ±0.01^a	1.32 ±0.013^b	1.29 ±0.02^b
BWG (g)	145.36 ±3.22^a	148.17 ±2.94^a	172.40 ±3.66^b	167.68 ±5.46^b
FCR	1.87 ±0.04^a	1.86 ±0.046^b	1.79 ±0.03^a	1.82 ±0.05^a
Condition factor (K)	1.86 ±0.01^a	1.86 ±0.01^a	1.88 ±0.01^{ab}	1.89 ±0.01^b
R^2	0.98^a	0.98^a	0.98^b	0.98^b
Survival (%)	95.33 ±0.10^a	94.67 ±0.10^b	92.67 ±0.20^c	93.33 ±0.26^d

* Means within the same row with different superscript letters are significantly different at $P < 0.05$, n=30

Table 4. Body carcass composition of Nile tilapia fed on diets with different levels of defatted black soldier fly larvae meal and commercial diet

Parameter (%)	**$BSFLM_0$**	**$BSFLM_{50}$**	**$BSFLM_{100}$**	**$COMM_0$**
Ash	7.0 ±0.59	7.2 ±0.39	6.8 ±0.15	6.9 ±0.49
Crude protein	72.4 ±0.81^a	73.8 ±0.46^a	75.3 ±0.96^b	75.7 ±1.03^b
Crude fat	3.3 ±0.01^a	2.9 ±0.29^b	5.0 ±0.31^c	8.3 ±0.26^d
NFE	5.5^a	6.5^b	2.3^c	1.13^d

** Means within the same row with different superscript letters are significantly different at $P < 0.05$; n=3; NFE - Nitrogen free extract

Table 5. Amino acids profile of *O. niloticus* fed on diets with different levels of defatted black soldier fly larvae meal and commercial diet

Amino acids (%)	$BSFLM_0$	$BSFLM_{50}$	$BSFLM_{100}$	$COMM_0$
Essential amino acids				
Phenylalanine	18.17 ±2.30^a	17.70 ±2.90^b	19.79 ±5.40^c	19.44 ±4.00^d
Threonine	3.87 ±0.10^a	5.37±0.70^b	5.78 ±1.60^c	5.25 ±1.10^d
Arginine	0.31 ±0.10^a	0.45 ±0.30^a	0.43 ±0.50^b	0.51 ±0.60^b
Valine	6.13 ±0.60^a	4.33 ±1.10^b	4.64 ±1.00^c	4.2 ±1.00^d
Isoleucine	2.72 ±0.40^a	2.43 ±0.50^b	2.94 ±0.80^c	2.82 ±0.60^d
Methionine	2.25 ±0.20^a	1.29 ±0.10^b	1.46 ±0.70^c	1.98 ±1.10^d
Tryptophan	12.98 ±0.40^a	20.71 ±1.90^b	13.02 ±2.70^c	15.06 ±5.40^d
Lysine	11.88 ±1.20^a	14.52 ±1.30^b	15.73 ±3.60^c	15.3 ±3.70^d
Histidine	1.36 ±0.10^a	1.24 ±0.50^a	1.43 ±0.30^b	1.47 ±0.30^b
Leucine	0.93 ±0.10^a	0.91 ±0.40^a	0.99 ±0.40^b	0.75 ±0.30^b
Non-essential amino acids				
Alanine	2.87 ±0.20^a	2.73 ±1.10^a	2.59 ±0.70^b	2.58 ±0.80^b
Tyrosine	1.16 ±0.10^a	1.21 ±0.60^a	0.84 ±0.20^a	0.93 ±0.20^a
Glycine	23.60 ±.60^a	17.47 ±1.00^b	20.32 ±8.70^c	21.9 ±4.10^d
Glutamic acid	3.41 ±0.30^a	2.88 ±1.10^b	3.65 ±0.60^c	2.25 ±0.70^d
Cysteine	2.72 ±0.30^a	2.12 ±0.70^a	2.33 ±0.90^b	1.92 ±0.40^c
Proline	5.59 ±0.60^a	3.57 ±0.60^b	4.29 ±0.90^c	3.72 ±1.20^d

* Means within the same row with different superscript letters are significantly different at $P < 0.05$; n=3

Partial Enterprise Budget Analysis

The mean yield was higher in the fish fed on $COMM_0$ and $BSFL_{100}$ followed $BSFLM_0$ and $BSFLM_{50}$ (Table 6). All the experimental diets registered significantly positive net return above total costs ($P<0.05$). The total cost and variable cost decreased gradually as SBM was replaced with BSFLM with lowest values exhibited by $BSFLM_{100}$. Break-even price were significantly different with lowest values (USD 2.36 ±0.01) being recorded in $BSFLM_{100}$ ($P<0.05$).

The present study was conducted to evaluate the effect of replacing soybean meal (SBM) with black soldier fly larvae meal (BSFLM) on the growth performance, feed utilization, carcass body composition and amino acid profile of Nile tilapia (*Oreochromis niloticus*). All the diets registered gradual increase in the mean weight. This can be attributed to the similarity in the initial crude protein and fat content of the extruded experimental diets coupled with the well-balanced and formulated diets. Fish fed on $BSFLM_{100}$ registered statistically similar final weight to control diet $COMM_0$ though significantly higher than diets $BSFLM_0$ and $BSFLM_{50}$. Similar findings have been reported in juvenile grass carp (*Cyprinus carpio*) by Lu et al., (2020) but contrary to (Dietz & Liebert, 2018). in Nile tilapia that registered lowest weight with highest soybean concentrate replacement with BSFLM.

Fish body weight increased by 89% in fish fed on $BSFLM_{100}$ and $COMM_0$ while 87% in fish fed on $BSFLM_0$ and $BSFLM_{50}$. Previous studies on Nile tilapia, have reported contrasting results to this study, for instance, a lower body weight increase of 30% (Tippayadara et al., 2021), 64% (Devic et al., 2018), 73% (Muin et al., 2017) and higher body weight gain of 96% (Rana, 2009) have been reported in diets containing 100% BSFLM. The differences in the increase of the body weight between the present and previous trials may have been mainly associated with varying fish development stages and study periods of between 32 and 96 days compared to the present study that lasted for a period of 168 days.

Table 6. Partial enterprise budget for the dietary treatments with different levels of defatted black soldier fly larvae meal and commercial diet

Parameters	Units	Treatments			
		$BSFLM_0$	$BSFLM_{50}$	$BSFLM_{100}$	$COMM_0$
Total yield	Kg	17.77 ±0.01[a]	16.08 ±0.01[b]	20.54 ±0.01[c]	20.81 ±0.02[d]
Gross revenue	USD	66.28 ±0.01[a]	59.98 ±0.01[b]	76.61±0.01[c]	77.6 ±0.35[d]
Variable costs	USD	59.5 ±0.12[a]	57.42 ±0.01[b]	47.93 ±0.00[c]	58.97 ±0.02[d]
Fixed costs	USD	0.47 ± 0.02	0.47 ±0.00	0.47 ±0.00	0.47 ±0.00
Total costs	USD	59.97 ±0.01[a]	57.88 ±0.01[b]	48.4 ±0.23[c]	59.44 ±0.01[d]
Net return above total costs	USD	6.32 ±0.01[a]	2.09 ±0.00[b]	28.21 ±0.01[c]	18.16 ±0.02[d]
Unit selling price	USD	3.73 ±0.01	3.73 ±0.02	3.73 ±0.00	3.73 ±0.01
Breakeven price (total costs)	USD	3.37 ±0.01[a]	3.6 ±0.01[b]	2.36 ±0.01[c]	2.86 ±0.01[d]
Breakeven yield (total costs)	Kg	16.08 ±0.02[a]	15.52 ±0.01[b]	12.98 ±0.01[c]	15.96 ±0.01[b]

* Values are means ± S.E. Values with the same superscripts in a row are not significantly different

The lowest BWG and SGR were recorded in fish fed on $BSFLM_0$ and $BSFLM_{50}$ with 100 and 50% soybean inclusion levels respectively. Similar results were reported by Lu et al. (2020) in juvenile grass carp but contrary to Dietz & Liebert (2018) in Nile tilapia fed on BSFL meal based diets. Previous studies have also reported reduced feeding and growth as a result of higher levels of dietary soybean proteins and recommended different optimal inclusion levels of 25% (Sharda et al., 2017 & Abdel-Warith et al., 2020), 4% (Amesa et al., 2018), 4.5% (Godoy et al., 2019) in Nile tilapia. Soybean meal has been reported to have growth-inhibitory effect which has been linked to anti-nutritional factors (Lu et al., 2020) that lower nutrient digestibility, nutrient bio-availability and palatability due to excessive degrees of non-soluble fibre and starch (Daniel, 2018). Similarly, the depressed growth in fish fed on diets with high inclusions (50 and 100 %) of SBM could be attributed to lack of phytase enzyme in tilapia to hydrolyze phytates in soybean that causes unavailability of amino acids.

Better BWG and FCR exhibited in fish fed on $BSFLM_{100}$ could be due to better amino acid profile with higher levels of glycine, alanine, isoleucine, leucine and phenylalanine compared to control $BSFLM_0$ (El-Hack et al., 2020). Contrary to the findings of this study, lower SGR and BWG in diets containing high inclusion levels of BSFLM have been reported by Devic et al., (2018) in Nile tilapia (*Oreochromis niloticus*) and Lu et al., (2020) in grass carp (*Ctenopharyngodon idellus*). The diminishing performance was linked to imbalances in amino acids and inadequate chitinase activity in diets with higher BSFLM. Similar growth patterns of the fish fed on diets with 100% BSFLM and the commercial diet indicated that the diets had similar nutritional values to support the growth of fish.

Fish fed on $BSFLM_{100}$ registered lowest FCR compared with other diets. The FCR values in the present study were lower than those reported by Muin et al., (2017) and Tippayadara et al., (2021) when BSFLM was included in the diets of tilapia. A lower FCR serves as an indicator of better feed utilization (Devic et al., 2018). The low FCR in fish fed on $BSFLM_{100}$ is in line with Xiao et al., (2018) findings in yellow catfish (*Pelteobagrus fulvidraco)* when conventional protein sources were replaced at 100% with BSFLM. Low FCR in fish fed $BSFLM_{100}$ may have been a factor of utilization of defatted BSFLM. Observations by Basto et al., (2020) indicated that defatting insect meals improved digestibility and utilization of nutrients though the chitin content remained similar between defatted and non-defatted insect meals.

The high values of FCR recorded in fish fed on $BSFLM_0$ and $BSFLM_{50}$ indicates that high content of SBM negatively influenced feed utilization. This could be due to the unavailability of macro and micronutrients in SBM that compromises feed utilization. High levels of soybean in fish diets could have resulted to anti-nutritional factors specifically, tannin which is ascribed to inhibition of digestive enzymes, phytic acid chelation which diminishes bioavailability of calcium, phosphorus, magnesium, manganese, zinc and iron (He et al., 2020). Gossypol in soya bean meal have also been reported to diminish digestibility and feed consumption by lowering the bioavailability of lysine. Soybean processing by thermal heating is a contributing factor in loss of some amino acids that influence protein, carbohydrate and moisture on milliard reactions (Ghosh & Ray, 2017). Length - weight relationship (r≥0.98, b=3) of the fish of the current study indicated isometric growths in all the fish fed on the experimental diets that entailed an increase in weight in every unit increase in length (Daliri, 2012). This suggests good physiological conditions of fish in relation to its welfare associated with optimal physico-chemical and biological qualities of water in fish ponds.

The survival rates from the present study were above 92% in all the dietary treatments. The survival rates agree with results in Nile tilapia (Tippayadara et al., 2021), yellow catfish (*Pelteobagrus fulvidraco*) (Xiao et al., 2018) and juvenile grass carp (*Ctenopharyngodon idellus*) (Lu et al., 2020) fed on black soldier fly larvae meal. As reflected in the condition factor, acclimatization of fish prior to stocking, consistent and appropriate feeding regimes in an optimal environment in favor of fish physiological processes played a major role in high survival rates.

The body carcass composition of fish was significantly influenced by different dietary treatments. There were significant differences in crude protein, crude fibre and total lipids content of the carcass. This may have been associated with the changes in the synthesis, deposition rate in muscle and/or different growth rate (Abdel-Warith et al., 2020). Fish fed on different diets differed significantly in crude protein content (Dietz & Liebert, 2018). Fish fed on control diet and $BSFLM_0$ had lower crude protein levels compared to $BSFLM_{100}$ and $COMM_0$. This could be due to difference in absorption kinetics of free and protein-bound amino acids that create asynchrony in post absorptive availability of individual amino acids. Cummins et al., (2017) found reduced protein and lipid levels in Pacific white shrimp (*Litopenaeus vannamei*) associated with deficiencies in essential amino acid (EAA) as well as increasing imbalances of essential and non-essential amino acids. This is contrary to the findings by Devic et al., (2018) in Nile tilapia where there were no significant differences in protein levels with increasing BSFLM supplementation. Low lipid content in fish fed on $BSFLM_0$ and $BSFLM_{50}$ is similar to the findings in rainbow trout (Sealey et al., 2011).

Protein quality is linked to amino acids content. High quality proteins are readily digestible and contain the dietary essential amino acids (EAA) in quantities that correspond to human requirements. In the current study amino acids concentration in the fish fillet increased with increasing dietary BSFLM. Fish fed on $BSFLM_{100}$ and $COMM_0$ had enhanced and satisfactory amino acid profile compared to fish fed on $BSFLM_0$ and $BSFLM_{50}$. Fish fed on $BSFLM_{100}$ had significantly higher values for six out of the ten identified essential amino acids associated with amino acid balance in the feed coupled with better physiological utilization after digestion, absorption and oxidation (Wu et al., 2014). This is also reflected in low FCR. The amino acids profile of the fish fed on $BSFLM_{100}$ and the fish fed on $COMM_0$ were comparable which means the quality of the feed with 100% BSFM led to the same effect on the amino acid profile of the fish fed on commercial diet ($COMM_0$).

Feed constitutes a major expense in aquaculture species hence profitability is a factor of cost minimization. In the current study feed costs were 46.33% ($BSFLM_{100}$), 48.37% ($BSFLM_{50}$,) 50.79% ($BSFLM_0$) and 53.04% ($COMM_0$) of the total variable costs. Replacing SBM with BSFLM at 50% and 100 % reduced the cost by 8.8% and 12 % respectively compared to control diet $COMM_0$. This was contributed by lower BSFLM price of 0.80 USD/kg against SBM of 0.90 USD/kg at the time of the experiment. Although $COMM_0$ had the highest total yield and gross revenue attributed to good growth performance, the diet registered lower net return above total costs in comparison to $BSFLM_{100}$. Similar results of cost of feed reduction and increased net returns have been reported by Abdel-Tawwab et al., (2020) in European sea bass (*Dicentrarchus labrax)*, Wachira et al., (2021) in Nile tilapia and Rawski et al., (2021) in Siberian sturgeon, when conventional sources of proteins were replaced with BSFLM. In addition, non-utilization of basal ingredients, mainly SBM and oil reduced the costs of formulating $BSFLM_{100}$ hence contributing to its cost effectiveness. Lower net return above total costs values were recorded in fish fed diet $BSFLM_0$ and $BSFLM_{50}$ mainly contributed by high amounts of soybean and fish meal included in these diets to supplement on limiting macro and micro elements. This is also compounded by low utilization of the feeds that contributed to low yields. As evidenced by low values for break-even price (total costs) and break even yield, replacing SBM with 100% BSFLM is economically viable.

Conclusion

This study demonstrated that fish fed on $BSFLM_{100}$ exhibited better growth performance in terms of body weight gain, specific growth rate and feed conversion ratio compared to the commercial diet. Similarly, amino acids deposition in fish fed on $BSFLM_{100}$ was significantly higher than $COMM_0$ confirming enhanced and comparative nutritive effect of $BSFLM_{100}$ and $COMM_0$. The study also confirms BSFLM as a cost effective alternative source of protein to the SBM in the diets of Nile tilapia.

Compliance with Ethical Standard

Conflict of interests: The authors declare that for this article they have no actual, potential, or perceived conflict of interests.

Ethics committee approval: The experiment was conducted following the standard operating procedures (SOPs) of the Kenya Marine and Fisheries Research Institute (KMFRI) guidelines for handling animals registered with the National Commission for Science, Technology and Innovation (NACOSTI) registration number NACOSTI/2016/05/001. The SOPs comply with the Prevention of cruelty to animals Act 1962, CAP 360 (Revised 2012) of the laws of Kenya and the EU regulation (EC Directive 86/609/EEC).

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