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Black soldier fly (*Hermetia illucens, L.*) larvae meal improves growth performance, feed efficiency and economic returns of Nile tilapia (*Oreochromis niloticus, L.*) fry

Samwel Mchele Limbu[1] | **Amon Paul Shoko**[2] | **Eusebia Ernest Ulotu**[3] | **Siwema Amran Luvanga**[2] | **Fridah Mukiri Munyi**[4] | **John Obedy John**[3] | **Mary Adhiambo Opiyo**[5]

[1]Department of Aquaculture Technology, School of Aquatic Sciences and Fisheries Technology, University of Dar es Salaam, Dar es Salaam, Tanzania

[2]Tanzania Fisheries Research Institute, Dar es Salaam, Tanzania

[3]Tanzania Fisheries Research Institute, Dar es Salaam, Tanzania

[4]Kenya Marine and Fisheries Research Institute, Mombasa, Kenya

[5]Kenya Marine and Fisheries Research Institute, National Aquaculture Research Development and Training Centre (NARDTC), Sagana, Kenya

Correspondence
Samwel Mchele Limbu, Department of Aquaculture Technology, School of Aquatic Sciences and Fisheries Technology, University of Dar es Salaam, P. O. Box 60091, Dar es Salaam, Tanzania.
Email: mchelelimbu@yahoo.com

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Abstract

Insects have been proposed as potential alternative animal protein sources to replace fishmeal (FM), which is expensive and has limited availability for fish feed formulation. However, studies on the effects of replacing FM with black soldier fly (*Hermetia illucens*) larvae (BSF-L) on growth performance of Nile tilapia (*Oreochromis niloticus*) fry, water quality and economic benefits are currently limited. This study determined the effects of replacing 100% FM, 75% FM, 50% FM, 25% FM and 0% FM with cheap BSF-L meal, hereafter referred to as BSF-L0, BSF-L25, BSF-L50, BSF-L75 and BSF-L100 diets, respectively for rearing all-male Nile tilapia fry produced by YY technology. The study further determined the optimum percentage of BSF-L meal for maximum growth of Nile tilapia fry. A total of 2400 visually healthy Nile tilapia fry weighing 0.001 g were randomly stocked into 20 plastic tanks (120 fry per tank, four replicates per treatment) and fed the BSF-L diets for 12 weeks. The BSF-L75 diet increased significantly specific growth rate, total weight gain, Zihler's index of fry and nitrate in the culture water but reduced feed conversion ratio of diet and total suspended solids in the culture water ($p < 0.05$). Feeding the Nile tilapia fry with BSF-L50 diet increased significantly hepatosomatic index ($p < 0.05$). The diets with BSF-L75 and BSF-L100 reduced significantly the incidence cost by 31.97% and 28.77% ($p < 0.05$), and increased profit index by 3.97 and 3.44%, respectively. The optimum percentage of BSF-L inclusion required for maximum growth performance of Nile tilapia fry was estimated as 81% to 84% based on polynomial analysis. Taken together, feeding Nile tilapia fry with diets containing 81% to 84% BSF-L meal improved growth performance. The BSF-L75 diet enhanced feed efficiency and had no deleterious effect on the liver and intestines. Meanwhile, it improved nitrate concentration for increased natural productivity. Incorporating 75% BSF-L meal in diets for Nile tilapia fry reduced 30% of feed cost leading to 4% higher economic returns.

KEYWORDS
black soldier fly larvae meal, economic benefits, feed conversion ratio, Nile tilapia fry, sustainable aquaculture, water quality parameters

1 | INTRODUCTION

Aquaculture contributes to food security globally as a source of dietary animal protein, fatty acids and micronutrients (Devic et al., 2018). The current global human per capita fish consumption is estimated at 20.5 kg annually, of which aquaculture contributes 46% of the total production and 52% of fish consumed by humans (FAO, 2020). However, the current fish production from aquaculture has not met the demand due to challenges such as food safety, trade and markets, and governance (Subasinghe et al., 2009), diseases, climate change and the use of wild fish for feed production (Naylor et al., 2021) and increasing population (Belghit et al., 2018). Therefore, aquaculture is imperative to increase production while ensuring sustainability of the industry.

One of the most important aspects required to increase aquaculture production while ensuring sustainability of the industry is the formulation of good quality and affordable feeds, which guarantee fast growth and survival rate of cultured fish without causing environmental pollution. Under current production economics, feed is the most expensive input in fish production frequently amounting to more than 50% of the total variable costs depending on culture system intensity (Limbu & Jumanne, 2014). Historically, fishmeal (FM) has been used as an important high-quality animal protein ingredient for aquaculture feeds production (Abdel-Tawwab et al., 2020; Xiao et al., 2018). However, FM is continuously becoming expensive due to direct competition for human consumption and other uses (Xiao et al., 2018) and has limited availability because of dwindling catches from wild fish stocks (Pauly et al., 2002). Accordingly, the continued use of FM threatens aquaculture industry sustainability from both economic and ecological points of view (Tacon & Metian, 2008). In fact, fisheries have rarely been 'sustainable' (Pauly et al., 2002) and thus cannot be relied upon for supply of FM for aquaculture feed production. Therefore, studies geared towards finding alternative dietary protein sources to replace FM in aquaculture feed industry are required to ensure increased production and sustainability of the industry.

Insects have been proposed and are used as potential ingredients for feed production in poultry industry (Allegretti et al., 2018; Onsongo et al., 2018) and aquaculture industry (Barroso et al., 2014; Henry et al., 2015). Insects possess the required nutritional quality (Kouřimská & Adámková, 2016; Nowak et al., 2016) and are accepted by fish consumers (Verbeke et al., 2015; House, 2016). Black soldier fly (*Hermetia illucens*) larvae (BSF-L) is one of the development stages with the highest potential to replace FM as a low-cost nutrient-rich alternative protein ingredient for fish feeds production (Caimi et al., 2021). BSF-L has an ability to convert organic waste into food, generating value and closing nutrient loops as it reduces organic pollution and their subsequent management costs as compared to other insects (Chia et al., 2019; Shelomi, 2020; van Huis & Oonincx, 2017; Wang & Shelomi, 2017). Therefore, BSF-L offers a possibility of replacing the expensive and limited supply of FM in fish feeds for increased production and sustainable aquaculture industry to meet the growing global demand for fish products.

In fact, BSF-L meal/diet has been used successfully to replace FM in diets for various fish species such as adult rainbow trout (*Oncorhynchus mykiss*) (Elia et al., 2018; Renna et al., 2017), juvenile rainbow trout (St-Hilaire et al., 2007), Atlantic salmon (*Salmo salar*) (Belghit et al., 2018), Jian carp (*Cyprinus carpio* var. Jian) (Li et al., 2017; Zhou et al., 2018), European seabass (*Dicentrarchus labrax*) (Magalhães et al., 2017), Siberian sturgeon (*Acipenser baerii*) (Caimi et al., 2020), and Nile tilapia (*Oreochromis niloticus*) (Kishawy et al., 2022), postsmolt Atlantic salmon (Belghit et al., 2019; Lock et al., 2016), and fingerlings of Nile tilapia (Devic et al., 2018) and European seabass (*Dicentrarchus labrax*) (Abdel-Tawwab et al., 2020). Despite these results, replacing FM with BSF-L meal reduced growth of juvenile turbot (*Psetta maxima*) (Kroeckel et al., 2012) and sub adult channel catfish (*Ictalurus punctatus*), in which 100% BSF-L reduced feed utilization (Bondari & Sheppard, 1987).

Nile tilapia is the third most important and economic farmed fish around the world (FAO, 2020). Fish fry is an important and indispensable stage needed for stocking culture systems for farmed species including Nile tilapia. The quality and growth rate of fry directly affect total Nile tilapia production cost. Moreover, the culture of Nile tilapia fry requires high protein, threatening the sustainable production of quality feeds due to the use of expensive FM. Therefore, studies on alternative sources of protein to replace the expensive FM in Nile tilapia fry feed formulation, which meet the nutritional requirement for improved growth at affordable cost are required. Despite the importance of fry, studies on FM replacement with BSF-L are currently limited in fish literature. Xiao et al. (2018) reported that, replacing FM with BSF-L meal increased growth performance and decreased feed conversion ratio (FCR), but did not affect survival rate and body indexes in yellow catfish (*Pelteobagrus fulvidraco*) fry. Replacing FM with 50%, 75% and 100% housefly (*Musca domestica*) maggot meal in Nile tilapia sub adults affected water quality by decreasing concentrations of nitrite nitrogen and total phosphorus (Wang et al., 2017). For economic analysis, Wachira et al. (2021) reported higher gross profit margin for Nile tilapia fingerlings fed on 33% rather than 100% BSF-L meal, but did not record any significant differences on return on investment and the cost-benefit ratio. However, studies on the effects of BSF-L meal on growth performance of Nile tilapia fry, water quality and the corresponding economic analysis are currently limited.

The present study investigated the effects of partial or total replacement of expensive FM with cheap BSF-L meal on rearing Nile tilapia fry. The effects of BSF-L meal on Nile tilapia fry were assessed by determining growth performance, feed efficiency,

TABLE 1 The proximate composition and cost of each ingredient (dry matter) used to formulate the experimental diets in the present study

	Ingredient				
Composition (g kg^{-1})	FM	SBM	MG	WB	BSF-L
Dry matter	915.00	938.00	889.00	871.00	916.00
Protein	584.00	368.00	109.00	210.00	508.00
Lipid	122.00	164.00	41.00	42.00	194.00
Ash	152.00	77.00	53.00	36.00	69.00
Crude fiber	–	71.00	20.00	42.00	91.00
Cost (USD kg^{-1})	1.51	0.65	0.43	0.04	0.86

Abbreviations: BSF-L, black soldier fly larvae; FM, Fishmeal; MG, Maize germ; SBM, Soybean meal; WB, wheat bran.

organ indices, condition factor, water quality parameters and economic returns. Further, the study determined the optimum percentage inclusion of BSF-L meal required for maximum growth performance of Nile tilapia fry. It was hypothesized that inclusion of BSF-L meal in Nile tilapia fry could improve growth performance, feed efficiency, organ indices, condition factor, water quality parameters and economic returns. The information generated is critical to global fish aquaculture practitioners to evaluate the efficacy of replacing FM with BSF-L during early nursing stage of Nile tilapia fry for increased aquaculture production and the industry sustainability.

2 | MATERIALS AND METHODS

2.1 | Ethical statement

The authors confirm that the ethical policies of the journal, as noted on the journal's author guidelines page, have been adhered to. The study was carried out in accordance with the Tanzanian laws and guidelines for the care of experimental animals. Fish were anesthetized by immersing them into ice at a temperature of 4°C to avoid causing stress and pain before death.

2.2 | Source of black soldier fly larvae meal and diet formulation

The defatted BSF-L used in the present study was obtained from Recycler LTD Tanzania (Table 1). Other ingredients used in this study were silver cyprinid (*Rastrineobola argentea*), soybean, wheat bran (WB), maize germ (MG) and palm oil (PO). All the ingredients were obtained in dry form (except palm oil) from animal feed suppliers in Dar es Salaam, Tanzania. The silver cyprinid, soybean, WB and MG were mechanically grounded individually into fine powder by using a hammer mill with 0.8 mm mesh size to obtain fishmeal (FM), soybean meal (SBM), WB and MG powders. The ingredients were used to formulate five isonitrogenous diets (400 g kg^{-1}) by using a Pearson square method, in which FM was replaced by BSF-L at 0, 25, 50, 75 and 100% hereafter referred to as BSF-L0 (control), BSF-L25, BSF-L50, BSF-L75 and BSF-L100 (Table 2). The dry ingredients were mixed based on the five formulations to obtain experimental diets in powder form. The resulting diets were stored in plastic bags at 4°C until needed for feeding the Nile tilapia fry. Fifty grams of all the ingredients used and each of the formulated BSF-L diet were analyzed for proximate composition (moisture, protein, lipid, fibre, ash) by using standard methods (AOAC, 1995). Moisture was determined by oven drying at 105°C for 24 h. Crude protein (N × 6.25) was determined by the Kjeldahl method after digestion with concentrated H_2SO_4. Ash content was determined by incineration in a muffle furnace at 600°C for 16 h. Crude lipid was determined by the Soxhlet method using petroleum ether and crude fibre was determined by digestion with 1.25% NaOH and 1.25% H_2SO_4.

2.3 | Source of Nile tilapia fry and experimental set up

Newly hatched monosex Nile tilapia fry produced by using the YY technology were purchased from Big Fish Farm LTD, located at Kigamboni in Dar es Salaam, Tanzania. The Nile tilapia fry were acclimatized for five days before the start of the feeding trial. During acclimatization period, the Nile tilapia fry were not fed to ensure the yolk sac is exhausted. This study was conducted in 20 plastic tanks measuring 1 × 1 × 1 m with a capacity to hold water at volume of 1000 L (1 m^3). Each plastic tank was fixed with a hapa net with 1 mm mesh size measuring 1 × 1 × 1 m. Each tank was filled with dechlorinated freshwater to 800 m representing 80% of the total size of the tank by volume.

After the five days of acclimatization, 2400 visually healthy Nile tilapia fry were bulk weighed (0.001 g) and randomly stocked into the 20 plastic tanks (120 fry per tank, four replicates per diet). The Nile tilapia fry stocking density of 0.12 fry L^{-1} used represents a semi-intensive production for this species (Huang & Chiu, 1997). During the culture period, 75% of water was replaced by reducing the water level from 800 to 200 m and replenished with fresh dechlorinated water every two weeks due to small size of the fry and little amount of diets fed. No aeration was provided during the entire study period. Overall, the initial water quality parameters measured were optimum for Nile tilapia fry and recorded as follows: temperature 29.98 ± 0.03 to 30.03 ± 0.10°C; dissolved oxygen 7.99 ± 0.19 to 8.14 ± 0.15 mgL^{-1}; and pH 8.58 ± 0.08 to 8.35± 0.12.

2.4 | Feeding trial and data collection

Before stocking, the Nile tilapia fry in each tank were bulk weighed by using a sensitive weighing balance (EWS-H+, Endel, Dubai, India) to determine their mean initial weight (g). The BSF-L diets were randomly assigned to the experimental tanks in four replicates. The fish in each tank were hand-fed to apparent visual satiation four times a day for 12 weeks. During the entire study period, fish sampling was conducted

TABLE 2 Feed formulation, proximate composition and cost of the experimental diets used during the study

	Experimental diets				
Ingredient (g kg^{-1})	0 BSF-L	25 BSF-L	50 BSF-L	75 BSF-L	100 BSF-L
Fish meal	423.50	317.60	211.80	105.90	0
Soybean meal	282.40	282.40	282.40	282.40	282.40
Maize germ	109.80	109.80	109.80	109.80	109.80
Wheat bran	164.70	164.70	164.70	164.70	164.70
Palm oil	9.80	9.80	9.80	9.80	9.80
Black soldier fly larvae	0	105.90	211.80	317.60	423.50
Vitamin premix[a]	4.90	4.90	4.90	4.90	4.90
Mineral premix[b]	4.90	4.90	4.90	4.90	4.90
Total	1000	1000	1000	1000	1000
Composition (g kg^{-1})					
Dry matter	907.00	891.00	888.00	885.00	882.00
Protein	398.00	396.00	393.00	391.00	389.00
Lipid	128.00	129.00	135.00	136.00	138.00
Ash	129.00	114.00	106.00	105.00	102.00
Crude fiber	97.00	73.00	64.00	57.00	48.00
Cost (USD)	0.995	0.927	0.859	0.790	0.722

[a]Vitamin Premix (per 100 g premix): thiamin hydrochloride, 0.15 g; retinol acetate, 0.043 g; Ca pantothenate, 0.3 g; riboflavin, 0.0625 g; niacin, 0.3 g; ascorbic acid, 0.5 g; biotin, 0.005 g; pyridoxine hydrochloride, 0.225 g; para-aminobenzoic acid, 0.1 g; folic acid, 0.025 g; α-tocopherol acetate, 0.5 g; cholecalciferol, 0.0075 g; menadione, 0.05 g; inositol, 1 g.
[b]Mineral Premix x (per 100 g premix): NaH_2PO_4, 10.0 g; $CaCO_3$, 10.5 g; KH_2 PO_4, 21.5 g; $Ca(H_2PO_4)_2$, 26.5 g; KCl, 2.8 g; $AlCl_3{\cdot}6H_2O$, 0.024 g; $MgSO_4{\cdot}7\,H_2O$, 10.0 g; MnSO4 6 H_2O, 0.143 g; KI, 0.023 g; $ZnSO_4$ $7H_2O$,0.476 g; $CuCl_2$ 2 H_2O, 0.015 g; $CoCl_2{\cdot}6\,H_2O$, 0.14 g; Calcium lactate, 16.50 g; Fe-citrate, 1 g.

every two weeks by collecting randomly 30 fry from each tank by using a seine net (12.00 mm mesh size), and their weights were measured as described above. The amount of feed was calculated and adjusted on a biweekly basis, according to the measured average weight of sampled fish in each tank.

2.5 | Determination of growth performance, feed efficiency, condition factor and survival rate

At the end of experiment, all survived Nile tilapia fry in each tank were starved for 24 h. The Nile tilapia fry in each tank were counted for determination of survival rate by using the formula:

$$\text{Survival rate (\%)} = \frac{\text{Final number of fry}}{\text{Initial number of fry}} \times 100$$

The Nile tilapia fry in each tank were bulk weighed to determine final mean weight (g). The weight data obtained were used to calculate growth performance by using the following formulae:

$$\text{Specific growth rate (SGR \% day}^{-1}) = \frac{(\ell\text{n Final weight} - \ell\text{n Initial weight})\text{ g}}{\text{Time (days)}} \times 100$$

$$\text{Total weight gain (g)} = \text{Final weight (g)} - \text{Initial weight (g)}$$

The amount of diet fed and weight of fish obtained were used to calculate FCR by using the formula:

$$\text{FCR} = \frac{\text{Total weight of dry diet fed (g)}}{\text{Total wet weight gain of the fry (g)}}$$

A sample of 30 Nile tilapia fry from each tank were collected and individually weighed by using a sensitive weighing balance (EWS-H+, Endel, Dubai, India) and measured for total length by using a measuring board. The weight (g) and length (cm) of each fish were used to determine condition factor by using the formula:

$$\text{Condition factor (g cm}^{-3}) = \frac{\text{Weight of individual fry (g)}}{\text{Total length (cm)}^3\text{ of individual fry}} \times 100$$

2.6 | Determination of Nile tilapia fry organ indices

Thirty Nile tilapia fry were sampled randomly from each tank and sacrificed for tissue collection. The sampled fry were anesthetized by immersing them into ice at a temperature of 4°C to avoid causing stress and pain before death. The Nile tilapia fry collected were individually weighed and measured for total length as described above. The Nile tilapia fry were dissected and the liver and intestine were carefully removed. The weight of the liver and that of the intestine were measured by using an analytical balance (ME104/02, Mettler Toledo, Shanghai, China). Moreover, each intestine was carefully stretched and its length was measured by using a measuring ruler. The individual fry, liver and intestine weights together with the intestine length were used to determine organ indices by using the following formulae:

$$\text{Hepatosomatic index (HSI, \%)} = \frac{\text{Weight of liver (g)}}{\text{Individual fish weight (g)}} \times 100$$

$$\text{Intestine index (\%)} = \frac{\text{Intestine weight (g)}}{\text{Individual fish weight (g)}} \times 100$$

$$\text{Zihler's index} = \frac{\text{Intestine length (mm)}}{10\left[\text{Fish weight (g)}^{1/3}\right]}$$

2.7 | Determination of physico-chemical water quality parameters

Water quality parameters were measured in each culture tank throughout the trial. Water temperature and dissolved oxygen (DO) were recorded twice per day at around 1000 hrs and 1600 hrs every one week by using a hand-held dissolved oxygen and temperature meter (OxyGuard Handy Polaris GB, Farum Denmark). The water pH was also measured twice daily every one week at around 10:00 hrs and 04:00 hrs by using a pH meter (HANNA model No. HA-01092845, Romania). All the measurements were done before and after feeding.

A 1 L plastic bottle was used for collecting the water samples for quantifying nitrate, ammonia and total suspended solids (TSS). The water samples for ammonia, TSS and nitrate were collected from each tank twice daily at around 1000 hrs and 1600 hrs every one week. The collected water samples were immediately taken to the Tanzania Fisheries Research Institute (TAFIRI) laboratory at the study site for further processing and analysis. In the laboratory, 500 mL of water samples each for ammonia, TSS and nitrate analyses were immediately filtered by using Whatman GF/C glass microfiber filters with 0.45 μm prior to analysis. The filters for TSS determination for each water sample per tank were heated at 105°C by using a drying oven (XMTD-8222, Jinghong, Shanghai, China). Nitrate, ammonia and TSS were measured by using a Spectrophotometer (L8, Double Beam UV-VIS Spectrophotometer, INAESA, Shanghai, China).

2.8 | Economic analysis

A partial enterprise budget was used to assess the cost-effectiveness of BSF-L diets used in the present study. The cost of diet was calculated by using market prices of the ingredients used (Table 1) taking into consideration the processing cost (Limbu, 2020). The analysis was based on local market retail prices converted into US$ (USD 1 = TZS 2323.46 in the course of the study period). During calculation, tank and labour costs were assumed to be constant. The income generated after selling fingerlings and the cost of feed were used to calculate incidence cost and profit index according to Obirikorang et al. (2020).

2.9 | Statistical analyses

All data were tested for normality and homogeneity of variances by using Shapiro-Wilk and Levene's tests, respectively. Data for total weight gain, SGR, FCR, Zihler's index, dissolved oxygen, nitrate, ammonia and TSS were normally distributed while those for pH were normally distributed after $\log_{10}$ transformation (Table S1). Data for condition factor, intestine index, water temperature, incidence cost and profit index were skewed even after $\log_{10}$ transformation. Therefore, one-way analysis of variance (ANOVA) was used to test for statistical differences on the obtained data for total weight gain, SGR, Zihler's index, dissolved oxygen, nitrate, ammonia, TSS and pH among the BSF-L diets. When statistical differences were detected, Tukey's post hoc test was used to compare significant differences among treatments. Data for condition factor, intestine index, water temperature, incidence cost and profit index were tested by using Kruskal-Wallis (H) test while specific comparisons between treatments were conducted by using Mann-Whitney (*U*) test as reported previously (Limbu & Kyewalyanga, 2015). Linear, quadratic and cubic orthogonal polynomial contrasts were performed for data obtained on total weight gain and SGR, in order to understand the appropriate equation for estimating the optimum percentage of BSF-L meal inclusion for replacing FM in Nile tilapia fry (Table S2). Cubic equation was used for polynomial regression analysis to assess the optimum percentage level of BSF-L meal required for maximum growth because it had higher R^2 values and significantly lower p values than linear and quadratic equations (Table S2). In all analyses, significant levels were judged at a $p \leq 0.05$. Percentage data were arcsine-transformed prior to analysis and reversed afterwards for reporting purposes as performed previously (Limbu et al., 2017). The obtained results are presented as means ± standard error of the mean (SEM). All statistical analyses were performed by using Statistical Package for the Social Sciences (SPSS) for windows version 20 (IBM, Armonk, NY, USA).

3 | RESULTS

3.1 | Growth performance, feed efficiency, condition factor, survival rate and organ indices

The results showed that, feeding the Nile tilapia fry with BSF-L75 diet improved weight gain (Figure 1). Feeding the Nile tilapia fry on varying proportions of BSF-L diets affected significantly the SGR ($p = 0.042$) and total weight gain ($p = 0.041$). Indeed, the BSF-L75 diet increased significantly SGR ($p < 0.034$) and total weight gain ($p < 0.033$) of Nile tilapia fry than those fed on BSF-L25 diet (Table 3). However, the fish fed on BSF-L75 diet had no significant difference in SGR to those fed on BSF-L0 ($p = 0.435$), BSF-L50 ($p = 0.128$) and BSF-L100 ($p = 0.798$) diets. Equally, the fish fed on BSF-L25 diet had no significant difference

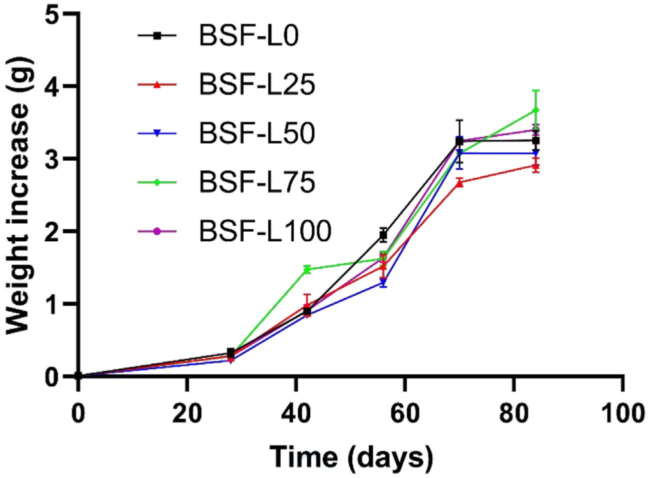

FIGURE 1 Weight increase of Nile tilapia fry fed on varying proportions of black soldier fly larvae (BSF-L) diets. Values are mean ± SEM, $n = 30$

in SGR to those fed on BSF-L0 (p = 0.554), BSF-L50 (p = 0.947) and BSF-L100 (p = 0.240) diets. The Nile tilapia fry fed on BSF-L0 diet had no significant difference in SGR to those fed on BSF-L50 (p = 0.923) and BSF-L100 (p = 0.966) diets. Likewise, the Nile tilapia fry fed on BSF-L50 and BSF-L100 diets had no significant difference in SGR (p = 0.607). The fish fed on BSF-L75 diet had no significant difference in total weight gain to those fed on BSF-L0 (p = 0.399), BSF-L50 (p = 0.119) and BSF-L100 (p = 0.759) diets. Moreover, fish fed on BSF-L25 diet had no significant difference in total weight gain to those fed on BSF-L0 (p = 0.585), BSF-L50 (p = 0.953) and BSF-L100 (p = 0.261) diets. Feeding the Nile tilapia on BSF-L0 diet had no significant difference in total weight gain to those fed on BSF-L50 (p = 0.931) and BSF-L100 (p = 0.967) diets. Furthermore, the Nile tilapia fry fed on BSF-L50 and BSF-L100 had no significant difference in total weight gain (p = 0.625).

Feeding the Nile tilapia fry with the BSF-L diets affected significantly the feed efficiency ($p < 0.001$). The Nile tilapia fry fed on BSF-L75 diet reduced FCR than those fed on BSF-L0 (p = 0.006), BSF-L25, BSF-L50 and BSF-L100 diets (Table 3; $p < 0.001$). Similarly, the Nile tilapia fry fed on BSF-L0 diet reduced FCR than those fed on BSF-L25 and BSF-L50 diets ($p < 0.001$). Moreover, the Nile tilapia fry fed on 100% BSF-L diet reduced FCR than those fed on BSF-L25 diet (p = 0.003). The fish fed on BSF-L0 and BSF-L100 diets had no significant difference in FCR (p = 0.120). Moreover, feeding the fish on BSF-L25 and BSF-L50 diets did not affect significantly the FCR (p = 0.494). Additionally, the fish fed on BSF-L50 and BSF-L100 diets had no significant difference in FCR values (p = 0.062). Interestingly, inclusion of BSF-L diet at all levels used in this study did not affect significantly condition factor (p = 0.280) and survival rate (p = 0.686) of the Nile tilapia fry (Table 3).

Feeding the Nile tilapia with the experimental diets affected significantly the HSI (p = 0.027), Zihler's index (p = 0.020) and intestine index (p = 0.050) (Table 3). Feeding the Nile tilapia fry with the BSF-L50 diet increased HSI than those fed on BSF-L100 diet (p = 0.021). However, the fish fed on BSF-L50 diet had no significant difference in HSI to those fed on BSF-L0 (p = 0.400), BSF-L25 (p = 0.993) and BSF-L75 (p = 0.819) diets. Equally, the fish fed on BSF-L100 diet had no significant difference in HSI to those fed on BSF-L0 (p = 0.747), BSF-L25 (p = 0.141) and BSF-L75 (p = 0.280) diets.

The Nile tilapia fry fed on BSF-L75 diet increased significantly Zihler's index than those fed on BSF-L0 diet (p = 0.034). However, the fish fed on BSF-L0 diet had no significant difference in Zihler's index to those fed on BSF-L25 (p = 0.971), BSF-L50 (p = 0.977) and BSF-L100 (p = 0.151) diets. Equally, the fish fed on BSF-L75 diet had no significant difference in Zihler's index compared to those fed on BSF-L25 (p = 0.233), BSF-L50 (p = 0.990) and BSF-L100 (p = 0.990) diets. The Nile tilapia fry fed on BSF-L50 diet had significantly higher intestine index than those fed on BSF-L0 diet (p = 0.035). Feeding the Nile tilapia on BSF-L0 diet did not affect significantly the intestine index compared to those fed on BSF-L25 (p = 0.166), BSF-L75 (p = 0.597) and BSF-L100 (p = 0.694) diets. Similarly, the Nile tilapia fry fed on BSF-L50 diet did not affect significantly the intestine index than those fed on BSF-L25 (p = 0.693), BSF-L75 (p = 0.391) and BSF-L100 (p = 0.157) diets.

TABLE 3 Growth performance, feed efficiency, condition factor, survival rate and organ indices of Nile tilapia fry fed on varying proportions of black soldier fly larvae (BSF-L) meal

	Experimental diets				
Parameter	BSF-L0	BSF-L25	BSF-L50	BSF-L75	BSF-L100
SGR (% day^{-1})	2.10 ± 0.02^{a,b}	2.06 ± 0.02^{a}	2.08 ± 0.02^{a,b}	2.15 ± 0.03^{b}	2.12 ± 0.01^{a,b}
Total weight gain (g)	3.25 ± 0.13^{a,b}	2.90 ± 0.10^{a}	3.06 ± 0.16^{a,b}	3.66 ± 0.27^{b}	3.39 ± 0.07^{a,b}
FCR	0.96 ± 0.01^{a}	1.09 ± 0.01^{b}	1.06 ± 0.02^{b,c}	0.89 ± 0.02^{d}	1.01 ± 0.01^{a,c,e}
Condition factor (g cm^{-3})	1.44 ± 0.03^{a}	1.47 ± 0.04^{a}	1.54 ± 0.04^{a}	1.51 ± 0.05^{a}	1.55 ± 0.04^{a}
Survival rate (%)	96.46 ± 1.75^{a}	93.54 ± 3.97^{a}	92.71 ± 2.79^{a}	96.46 ± 1.61^{a}	96.88 ± 1.81^{a}
HSI (%)	0.25 ± 0.03^{a,b}	0.32 ± 0.06^{a,b}	0.34 ± 0.04^{b}	0.29 ± 0.04^{a,b}	0.19 ± 0.02^{a}
Zihler's index (mm g^{-1})	1.25 ± 0.13^{a}	1.37 ± 0.15^{a,b}	1.68 ± 0.13^{a,b}	1.77 ± 0.13^{b}	1.69 ± 0.13^{a,b}
Intestine index (%)	14.63 ± 0.58^{a}	16.21 ± 0.78^{a,b}	16.54 ± 0.50^{b}	14.89 ± 0.57^{a,b}	14.37 ± 0.47^{a,b}

Note: Values in each row with different superscript alphabets (a, b, c, d or e) are statistically different ($p \leq 0.05$). SGR = specific growth rate, FCR = feed conversion ratio and HSI = hepatosomatic index.

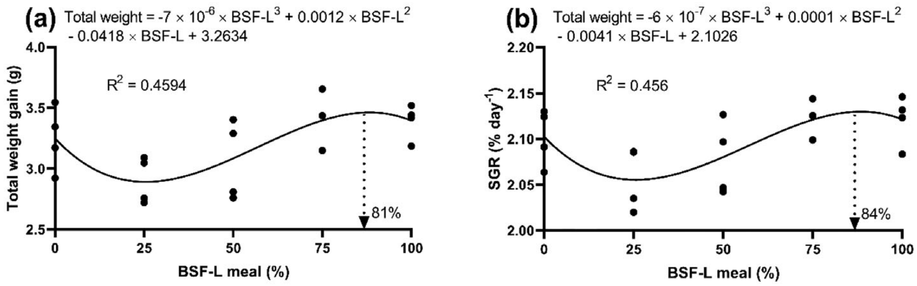

FIGURE 2 The optimum percentage of black soldier fly larvae (BSF-L) meal inclusion required for growth performance of Nile tilapia fry. (a) Optimum percentage of BSF-L meal inclusion estimated based on total weight gain and (b) Optimum percentage of BSF-L meal inclusion estimated based on specific growth rate (SGR)

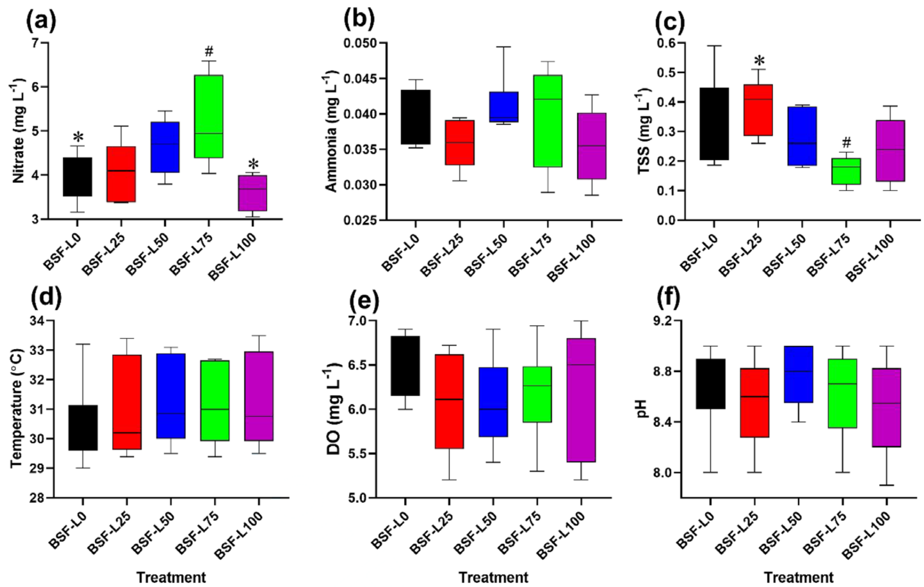

FIGURE 3 Chemical and physical quality parameters measured in water used for rearing Nile tilapia fry fed on varying proportions of black soldier fly larvae (BSF-L) diet. (a) Nitrate, (b) ammonia, (c) total suspended solids (TSS), (d) water temperature, (e) dissolved oxygen and (f) pH. Values are mean ± SEM, $n = 8$ for (a) to (c) and $n = 24$ for (d) to (f). Box-plots with different symbols (* and #) are statistically different ($p \leq 0.05$) and those without any symbol are not significant different ($p > 0.05$)

3.2 | The optimum percentage of BSF-L meal required for maximum growth performance

We determined the optimum percentage of BSF-L inclusion required for maximum growth of Nile tilapia fry. We found that Nile tilapia fry requires between 81% and 84% BSF-L meal for maximum growth performance based on total weight gain (Figure 2a) and SGR (Figure 2b), respectively.

3.3 | Chemical and physical water quality parameters

The BSF-L diets used affected significantly the nitrate concentration in the water ($p = 0.007$; Figure 3). The diet with BSF-L75 enhanced significantly the availability of nitrate in culture water compared to the BSF-L0 ($p = 0.033$) and BSF-L100 ($p = 0.012$) diets (Figure 3a). However, the water in the tanks used to culture the Nile tilapia fry while feeding them with BSF-L75 diet did not affect significantly nitrate levels compared to those reared under BSF-L25 ($p = 0.067$) and BSF-L50 ($p = 0.657$) diets. Equally, the water used to culture the Nile tilapia fry while feeding them with BSF-L100 diet did not affect significantly nitrate levels compared to those fed on BSFL-0 ($p = 0.941$), BSF-L25 ($p = 0.840$) and BSF-L50 ($p = 0.201$) diets. The Nile tilapia fed on BSF-L0 diet did not affect significantly nitrate level in the water than those fed on BSF-L25 ($p = 0.998$) and BSF-L50 ($p = 0.482$) diets. Feeding the Nile tilapia fry with BSF-L25 and BSF-L50 diets did not affect nitrate level in the culture water ($p = 0.667$).

Feeding the Nile tilapia fry with all the BSF-L diets used in this study did not cause any significant differences in ammonia levels in

TABLE 4 Economic analysis of Nile tilapia fry fed on varying proportions of black soldier fly larvae (BSF-L)

	Experimental diets				
Parameter	BSF-L0	BSF-L25	BSF-L50	BSF-L75	BSF-L100
Amount of meal used (kg)	0.36	0.35	0.36	0.37	0.40
Price of meal (USD)	1.00	0.93	0.86	0.79	0.72
Processing cost of meal (USD)	0.25	0.23	0.21	0.20	0.18
Total cost of meal (USD)	1.25	1.16	1.07	0.99	0.90
Total fingerling weight (kg)	0.38	0.33	0.34	0.42	0.39
Fingerling price (USD)	0.13	0.13	0.13	0.13	0.13
Incidence cost (USD)	1.19 ± 0.01^a	1.26 ± 0.01^b	1.14 ± 0.02^c	0.88 ± 0.02^d	0.91 ± 0.01^d
Profit index	0.11 ± 0.00^a	0.10 ± 0.00^b	0.11 ± 0.00^c	0.15 ± 0.00^d	0.14 ± 0.00^d

Note: Values in each row with different superscript alphabets (a, b, c or d) are statistically different ($p \le 0.05$).

$$\text{Incidence cost (USD)} = \frac{\text{Amount of meal used (kg)} \times \text{Total cost of meal (USD)}}{\text{Total fingerling weight (kg)}}$$

$$\text{Profit index} = \frac{\text{Total fry weight (kg)} \times \text{Fingerling price (USD)}}{\text{Amount of meal used (kg)} \times \text{Total cost of meal (USD)}}$$

the culture water (Figure 3b; $p = 0.258$). Contrary, the Nile tilapia fry fed on the BSF-L diets affected significantly the TSS in the culture water (Figure 3c; $p = 0.018$). The Nile tilapia fry fed on BSF-L75 diet reduced significantly the TSS compared to the culture water in tanks reared under the BSF-L25 diet ($p = 0.019$). However, the water in the tanks used to culture the Nile tilapia while feeding them with BSF-L75 diet had no significant difference in TSS levels to those reared under BSF-L0 ($p = 0.060$), BSF-L50 ($p = 0.426$) and BSF-L100 ($p = 0.842$) diets. Equally, the water used to culture the Nile tilapia fry while feeding them with BSF-L0 diet had no significant difference in TSS levels to those fed on BSF-L25 ($p = 0.966$), BSF-L50 ($p = 0.876$) and BSF-L100 ($p = 0.485$) diets. Feeding the Nile tilapia fry with BSF-L25 did not affect the TSS in the culture water compared to those fed on BSF-L50 ($p = 0.559$) and BSF-L100 ($p = 0.218$) diets. Likewise, the fish fed on BSF-L50 diet and those fed on BSF-L100 diet did not affect significantly the TSS in the water ($p = 0.961$). The feeding of Nile tilapia fry with the BSF-L diets at all levels used in this study did not affect significantly water temperature (Figure 3d; $p = 0.334$), dissolved oxygen (Figure 3e; $p = 0.970$) and pH (Figure 3f; $p = 0.470$) of the culture water.

3.4 | Incidence cost and profit index

The BSF-L diets used affected significantly the incidence cost and profit index (Table 4; $p = 0.001$). Feeding the Nile tilapia fry with BSF-L75 and BSF-L100 diets reduced significantly the incidence cost compared to those fed on BSF-L0, BSF-L25 and BSF-L50 diets ($p > 0.021$). Similarly, the Nile tilapia fry fed on BSF-L50 diet reduced significantly the incidence cost than those fed on BSF-L0 and BSF-L25 diets ($p > 0.021$). The Nile tilapia fed on BSF-L0 diet also had significantly lower incidence cost than those fed on BSF-L25 diet ($p > 0.021$). Feeding the Nile tilapia fry with BSF-L75 and BSF-L100 diets did not affect significantly the incidence cost ($p = 0.083$). On the contrary, the Nile tilapia fry fed on BSF-L75 and BSF-L100 diets increased significantly the profit index compared to those fed on BSF-L0, BSF-L25 and BSF-L50 diets (Table 4; $p = 0.001$). Moreover, the Nile tilapia fry fed on BSF-L50 diet increased significantly profit index than those fed on BSF-L0 and BSF-L25 diets ($p = 0.021$). The Nile tilapia fed on BSF-L0 diet had significantly higher profit index than those fed on BSF-L25 diet ($p = 0.021$). Inclusion of BSF-L75 and BSF-L100 diets in the Nile tilapia fry did not affect significantly the profit index ($p = 0.083$).

4 | DISCUSSION

Feeding represents one of the most important aspects of aquaculture industry because it contributes to growth, survival and wellness of the cultured fish. Thus, aquaculture industry sustainability depends on reducing feeding cost while improving or without affecting the survival rate, growth performance and cultured fish well-being. The present study investigated the effects of partial or total dietary replacement of expensive FM with cheap BSF-L diets on rearing Nile tilapia fry. The results showed that feeding the Nile tilapia fry with BSF-L75 diet improved growth performance. The optimum percentage of BSF-L meal inclusion for Nile tilapia fry maximum growth performance was estimated to range between 81 and 84% based on total weight gain and SGR, respectively. The increased growth performance of the fry fed on BSF-L75 diet was due to increased feed efficiency. In this study, the fry fed on BSF-L75 diet had lower FCR, indicating efficient utilization of nutrients contained in the diet. The use of *H. illucens* larvae meal is known to increase feed efficiency resulting in decreased FCR in other fish species such as Atlantic salmon postsmolt (Lock et al., 2016) and Siberian sturgeon fingerlings (Rawski et al., 2020). The increased feed utilization of BSF-L has been related to defatting of insect meals, which improved digestibility and utilization of nutrients (Basto et al., 2020). Improved growth performance after BSF-L meal feeding has also been reported in yellow catfish fry (Xiao et al., 2018), rice field eel (*Monopterus albus*) juvenile (Hu et al., 2020) and Siberian sturgeon fingerlings (Rawski et al., 2020). It is noteworthy that replacing FM by using BSF-L diets at all levels used did not affect survival rate of the fry in this study, similar to results obtained in European sea bass (Abdel-Tawwab et al., 2020) and Nile tilapia (Wachira et al., 2021). These results indicate that, replacing FM by using BSF-L meal at 81 to 84% improves growth performance of Nile tilapia fry by enhancing nutrients utilization without affecting the survival rate of the fish.

Organ indices are critical indicators on the effects of dietary treatments in cultured fish. The fish intestine is an important organ involved in digestion of ingested feeds (Ganguly & Prasad, 2012), while the liver plays key functions on nutrients metabolism (Brusle & Gonzàlez i Anadon, 2017). In the present study, the Nile tilapia fry fed on BSF-L50 diet had higher HSI than those fed BSF-L100 diet, while those fed on BSF-L75 diet had higher Zihler's index compared to those fed on BSF-L0 diet. Similarly, inclusion of BSF-L meal increased HSI in adult and juvenile Atlantic salmon in which FM was replaced by 100 (Lock et al., 2016) and 85% (Belghit et al., 2018), respectively. However, replacing FM with BSF-L diet at all levels did not affect intestine index and condition factor of the Nile tilapia fry. These results are similar to previous studies, in which inclusion of BSF-L meal up to 100%, did not affect Atlantic salmon condition factor (Lock et al., 2016).

The increased HSI for Nile tilapia fry fed on BSF-L50 diet is most likely due to higher hepatic and intraperitoneal fat contents as reported previously (Lock et al., 2016). Unfortunately, hepatic and intraperitoneal fat contents were not measured in this study due to the small size of the fry. Interestingly, feeding the Nile tilapia fry on BSF-L75 diet and those fed on BSF-L50 and BSF-L100 diets did not affect the HSI. Higher and lower HSI are undesirable in cultured species because they cause cirrhosis and hepatotoxicity (Limbu et al., 2021). Noticeable, the Nile tilapia fry fed on BSF-L75 diet recorded higher Zihler's index. The precise reason(s) for longer intestine in fish fed on BSF-L diet awaits further studies to scrutinize. A higher Zihler's index indicated longer digestive tract in the Nile tilapia fry fed on BSF-L75 diet. Longer digestive tract extend the retention period of ingested diets in the digestive tract, thereby increasing exposure to the digestive processes (Obirikorang et al., 2020). It is more likely that, increased exposure of the diets to digestive processes due to higher Zihler's index in the fry fed on BSF-L75 diet led to increased digestion efficiency and nutrient utilization reported before in this study, which led to increased growth performance.

In general, the results on growth and organ indices indicate that, feeding the Nile tilapia fry with BSF-L75 diet improves growth performance, feed efficiency and intestinal length without affecting the liver and condition factor. Thus, farmers with access to BSF-L meal can include a range from 81% to 84% for improved growth performance and liver and intestine functions of their Nile tilapia fry.

The growth performance and survival rate of cultured fish are also affected by chemical and physical parameters of the water. Water quality parameters are affected by fish metabolism on absorbed nutrients from diets resulting in faecal matter and urine production, and decomposition of uneaten feeds. Therefore, ingredients which are environmentally friendly that can replace FM in fish feeds without affecting water quality parameters are important for aquaculture sustainability. Accordingly, we assessed some chemical and physical water quality parameters in the culture water. We found that, feeding the Nile tilapia fry with BSF-L75 diet enhanced the availability of nitrate and decreased TSS but did not affect ammonia in the culture water. Insect diets are known to reduce concentrations of some water quality parameters such as nitrite nitrogen and total phosphorus following FM replacement by housefly (*Musca domestica*) maggot meal (Wang et al., 2017). Currently, the ability of BSF-L meal to increase nitrate and reduce TSS in the culture water requires further study to scrutinize. Increased nitrate concentration in the water act as a source of nutrient for natural productivity. Although we did not measure the abundance of natural food organisms due to limited research facilities, we hypothesized that the availability of nitrate possibly increased natural productivity. Notably, ammonia, which is a harmful water quality parameter to fish was low among the BSF-L diets used in this study. The average ammonia levels obtained in this study ranged from 0.035 ± 0.002 to 0.041 ± 0.002 mgL^{-1} in the water for Nile tilapia fry fed on BSF-L50 and BSF-L100 diets, respectively, while the average mean acute toxicity of ammonia for 32 freshwater species is 2.79 mgL^{-1} (Randall & Tsui, 2002). These results indicate that the levels of BSF-L diets used in this study did not generate high levels of toxic ammonia. Taken together, the use of BSF-L75 diet enhanced nitrate availability for natural productivity and reduced TSS without generating high levels of toxic ammonia.

Economic returns are integral consideration for modern aquaculture farmers because feeds account for more than half of the operating cost (Limbu, 2020). Therefore, reducing feed cost is critical to the aquaculture production profitability and sustainability. In this study, feeding the Nile tilapia fry with BSF-L75 and BSF-L100 diets reduced incidence cost by 31.97% and 28.77% and increased profit index by 3.97% and 3.44%, respectively compared to 100% FM (BSF-L0 diet). Wachira et al. (2021) reported higher gross profit margin for Nile tilapia fingerlings fed on 100 BSF-L diet. Moreover, FM replacement with partial defatted black soldier larvae meal (PD-BSLM) at 25%, 50% and 100% in Nile tilapia juvenile decreased feed cost, increased profit/kilogram gain and resulted in higher economic efficiency (Kishawy et al., 2022). The lower incidence cost and higher profit index for Nile tilapia fry fed on BSF-L75 and BSF-L100 diets are due to the lower cost of BSF-L meal as a cheaper nutrient-rich alternative protein ingredient. During the study period, the cost of BSF ingredient was 0.86 USD kg^{-1} compared to 1.51 USD kg^{-1} for FM (Table 1), which resulted in 20.50% and 27.34% cheaper diets for the BSF-L75 and BSF-L100, respectively (Table 2). Indeed, the formulated diets cost decreased as the levels of BSF-L replacement increased (Table 2). Similarly, BSF-L decreased the diet cost at increasing levels in European sea bass diets from 0.71 USD kg^{-1} to 0.60 USD kg^{-1} diet (Abdel-Tawwab et al., 2020). Taken together, feeding the Nile tilapia fry with BSF-L75 diet reduces feed cost by 30% leading to higher economic returns of 4%. For this reason, Nile tilapia fry farmers with access to BSF-L are advised to include 75% in the diets to reduce the feeding cost.

Notewithstanding the important scientific knowledge generated in this study, we wish to inform that, our results should be conceived in the context of the following limitations. First, despite the fact that we did not analyze natural food organisms in the culture tanks and hepatic and intraperitoneal fat contents, our results are supported by other aspects in this study and previous results (Limbu et al., 2016; Lock et al., 2016). Second, our results are based on a laboratory set up, whereby conditions such as water quality are easily monitored and controlled compared to practical large scale commercial production with little or no control of variables. Therefore, to enhance further our understanding on application of BSF-L meal in cultured fish fry, future on-farm studies at production scale are required.

5 | CONCLUSION

This study revealed that feeding Nile tilapia fry with BSF-L75 diet improves growth, feed efficiency, liver and intestinal organs and enhances nitrate for increased natural productivity. The use of BSF-L75 diet reduces 30% of feed cost leading to 4% higher economic returns. Feeding the Nile tilapia fry with BSF-L75 does not affect survival rate, condition factor, water temperature, dissolved oxygen and pH based on the current experimental setup. For the first time, this study provides novel knowledge on the potential of using BSF-L meal to reduce feed cost in cultured Nile tilapia fry at hatchery level.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTION

The experiment was conceived by Samwel Mchele Limbu, Amon Paul Shoko, Eusebia Ernest Ulotu, Siwema Amran Luvanga and Mary Adhiambo Opiyo. Amon Paul Shoko, Eusebia Ernest Ulotu, Siwema Amran Luvanga and John Obedy John planned and executed the experiment. Samwel Mchele Limbu, Amon Paul Shoko, Eusebia Ernest Ulotu, Siwema Amran Luvanga, Fridah Mukiri Munyi and John Obedy John conducted the sample preparations, chemical analyses and data analysis. Amon Paul Shoko supervised the study. Samwel Mchele Limbu drafted the manuscript. All authors critically examined, revised and approved the final version of the manuscript for submission.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon a reasonable request.

PEER REVIEW

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ORCID

Samwel Mchele Limbu https://orcid.org/0000-0001-6063-2745
Amon Paul Shoko https://orcid.org/0000-0002-2174-8857
Eusebia Ernest Ulotu https://orcid.org/0000-0003-1154-5295
Fridah Mukiri Munyi https://orcid.org/0000-0003-2513-3558
Mary Adhiambo Opiyo https://orcid.org/0000-0001-9660-5888

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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