Ciência Animal Brasileira
Brazilian Animal Science

DOI: 10.1590/1809-6891v27e-81644E

e-ISSN 1809-6891
Animal science | Research article

Use of black soldier fly (*Hermetia illucens*) larvae to feed *Colossoma macropomum* in floating cages in the Peruvian Amazon

[Uso de larvas da mosca-soldado-negro (*Hermetia illucens*) para a alimentação de *Colossoma macropomum* em gaiolas flutuantes na Amazônia Peruana]

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Received: February 04, 2025. Accepted: November 17, 2025. Published: January 16, 2026.
Editor: Rondineli P. Barbero

Abstract: This study was carried out at the "Centro de Investigaciones Fernando Alcántara Bocanegra" (CIFAB) of the "Instituto de Investigaciones de la Amazonía Peruana" (IIAP) to determine the growth variation of *Colossoma macropomum* "gamitana" fed with larvae of *Hermetia illucens* black soldier fly (BSF), termites, and conventional balanced feed. A total of 180 *C. macropomum*, with initial standard length of 3.5 ± 0.5 cm and weight of 2.5 ± 0.5 g, was used. Sixty fish per treatment were distributed in 9 floating cages, 20 fish in each cage. The floating cages were installed inside a fishpond from IIAP. Treatments (T) consisted of T1: fish fed with Black soldier fly larvae (BSFL), T2: fish fed with termites, and T3: fish fed with balanced feed. Black soldier fly larvae were obtained using orange waste for their production. Termites were collected from natural environments and balanced feed was purchased from a commercial company in Iquitos, Peru. The results showed that fish fed with balanced feed grew more (T1 = 3.61 ± 0.74; T2 = 1.65 ± 0.50; and T3 = 4.46 ± 0.08) and gained more weight (T1 = 8.87 ± 2.64; T2 = 2.75 ± 0.5; and T3 = 11.76 ± 0.26) than fish fed with Black soldier fly larvae (BSFL) and termites ($p<0.0001$). Regarding apparent feed conversion ratio, treatment T1 was more efficient than T2 (T1 = 1.52 ± 0.20; T2 = 6.00 ± 1.51; $p< 0.05$), but equally efficient as T3 (T1 = 1.52 ± 0.20; T3 = 1.58 ± 0.08a; $p>0.05$). The survival rate of fish fed with T1 was the same as that of T3 (95 %), whereas fish fed with T2 showed a survival rate of 60 %. Comparing T1 and T2, fish fed with (BSFL) showed adequate and superior zootechnical indices than fish fed with termites, being a promising alternative for the farming of *C. macropomum* in floating cages.

Keywords: balanced fish feed; Blackfin pacu; floating cage; termites; Stratiomyidae larvae.

Resumo: Este estudo foi realizado no Centro de Pesquisas Fernando Alcántara Bocanegra (CIFAB) do Instituto de Pesquisas da Amazônia Peruana (IIAP) para determinar a variação no crescimento de Colossoma *macropomum* (gamitana) alimentado com larvas de *Hermetia illucens* "mosca-soldado-negro" (MSN), térmitas e ração balanceada convencional. Foi utilizado um total de 180 indivíduos de *C. macropomum* com comprimento inicial padrão de 3.5 ± 0.5 cm e 2.5 ± 0.5 g de peso. Utilizaram-se 60 peixes por tratamento, distribuídos em 9 gaiolas flutuantes, com 20 peixes em cada gaiola. As gaiolas flutuantes foram instaladas dentro de um viveiro de peixes do IIAP. Os tratamentos (T) foram: T1: peixes alimentados com larvas de MSN, T2: peixes alimentados com térmitas, T3: peixes alimentados com ração balanceada. As larvas de MSN foram obtidas utilizando resíduos de laranja para sua produção. As térmitas foram coletadas em ambientes naturais, e a ração balanceada foi adquirida de uma empresa comercial em Iquitos, Peru. Os resultados mostraram que os peixes alimentados com ração balanceada cresceram mais (T1 = 3,61 ± 0,74; T2 = 1,65 ± 0,50; e T3 = 4,46 ± 0,08) e ganharam mais peso (T1 = 8,87 ± 2,64; T2 = 2,75 ± 0,5; e T3 = 11,76 ± 0,26) do que os peixes alimentados com MSNL e cupins (p<0,0001). Em relação à taxa de conversão alimentar aparente, o tratamento T1 foi mais eficiente do que o T2 (T1 = 1,52 ± 0,20; T2 = 6,00 ± 1,51; p< 0,05), mas igualmente eficiente quanto o T3 (T1 = 1,52 ± 0,20; T3 = 1,58 ± 0,08a; p>0,05). A taxa de sobrevivência dos peixes alimentados com T1 foi a mesma que a do T3 (95 %), ao contrário dos peixes alimentados com T2, que tiveram uma taxa de sobrevivência de 60 %. Comparando os tratamentos T1 e T2, os peixes alimentados com MSNL apresentaram índices zootécnicos adequados e melhores do que os peixes alimentados com cupins, sendo uma alternativa promissora para a criação de *C. macropomum* em gaiolas flutuantes.

Palavras-chave: ração balanceada para peixes; tambaqui; gaiola flutuante; térmites; larvas de Stratiomyidae.

1. Introduction

Currently, Amazonian fish farming focuses on the breeding of native species such as *Colossoma macropomum*, *Piaractus brachypomus*, *Brycon amazonicus, Prochilodus nigricans*, and *Arapaima gigas*, species for which captive culture technologies are known and put into practice in the Peruvian Amazon. The recorded production of the sum of these species varies around 700 tons in Peru, and it is primarily destined for local consumption (1).

One of the species with the greatest technological advances in Loreto is *Colossoma macropomum*, popularly known in Peru as 'gamitana', considered the largest characid in the Amazon. Studies on the diet of juvenile individuals show a preference for consuming zooplankton, fruits, and seeds (1). This fish is farmed in various ways, using different environments and a range of farming technologies. According to Campos-Baca (1), its annual production in Loreto, Peru, is around 60 tons. Its culture in the Peruvian Amazon is mainly small-scale, and despite being the species with the greatest technological advances, many aspects still need improvement to ameliorate production processes (1).

The high cost of producing or acquiring fish feed is a particular problem that Amazonian fish farmers complain about, which results in high expenses for fish farmers and limits them from developing (1). The main input used to produce balanced feed is fishmeal, a product of marine origin that has a high value for low-income producers. Additionally, aquaculture centers are located far from the city, making it difficult to obtain this input (2).

Experiences from foreign countries in incorporating local inputs for fish feed production have been successful, using, for example, fruits, vegetables, and insects (3). Rural fish farmers in the Peruvian Amazon, mostly indigenous, spend their daily efforts on activities such as subsistence farming, fishing, hunting, and fruit gathering. Daily, they generate organic waste such as bananas, other vegetable peels, shells, seeds, and residues from wild fruits, which are discarded in the forest or thrown into rivers (4).

Among the insects globally used as animal protein, *Hermetia illucens*, popularly known as the ‘black soldier fly’, degrades organic matter and has a short production cycle, making its larvae great candidates for intensive and controlled production for use as animal protein [5]. According to Vargas-Arana [6], black soldier fly larvae meal fed with orange waste contains 28.5 % lipids, 15.8 % carbohydrates, and 38.9 % crude protein, making it an excellent feed option for Amazonian fish. Additionally, Sogbesan and Ugwumba [7] pointed out that the use of other insects, such as termites, can promote fish growth, as they contain 11.3 % lipids, 20 % carbohydrates, and 38.9 % crude protein. Knowing that both black soldier fly larvae and termites can be used in fish feed, the present study compared the growth and weight gain of *C. macropomum* fry fed termites and *H. illucens* larvae with those fed with conventional commercial feed, highlighting the significance of sustainable alternatives for feeding Amazonian fish.

2. Material and methods

The study was conducted at the Centro de Investigación Fernando Alcántara Bocanegra (CIFAB), of the Instituto de Investigaciones de la Amazonía Peruana (IIAP) in San Juan Bautista, Loreto, Peru (03° 48’, 48.9’’ S, 73° 19’, 2’’ W). One hundred eighty (180) *C. macropomum* individuals, with a standard length of 3.5 ± 0.5 cm and a weight of 2.5 ± 0.5 g, were selected from a 1,000-square-meter earthen pond. The inclusion criteria were fish measuring between 3 and 4 cm in length. Nine 1 m^3 floating cages made of polyethylene bags and plastic tubes were used. The distance between the top of each cage and the water surface was 20 cm. The cages were placed in a row inside a fishpond at CIFAB-IIAP (Fig. S1A). Three treatments (T) were used: T1 = fish fed with black soldier fly larvae (BSF), with a size between 4 – 5 mm; T2 = fish fed with termites, with a size between 4 – 5 mm, and T3 = fish fed with balanced feed. Both black soldier fly larvae and termites were offered whole. Each treatment consisted of three replicates, with 20 fish per replicate and a total of 60 fish per treatment. The experiment lasted 60 days. BSF larvae were produced in the production modules of CIFAB-IIAP (Fig. S1B), following the protocol established by Morey et al. [8].

The life cycle of BSF lasted 29 days. Eggs were observed five days after copulation between male and female specimens. The larvae phase lasted 15 days, the pre-pupa phase 7 days, and the pupa phase another 7 days, before emerging as adults that copulated to repeat the life cycle (Fig. 1). Termites were acquired from collections made in forested areas of CIFAB-IIAP. The commercial feed was purchased from external suppliers. According to the information on the feed label, the nutritional composition of the balanced feed was 32 % crude protein, 5 % fat, 8 % fiber, 10 % ash, and 10 % moisture, with 1.0 mm.

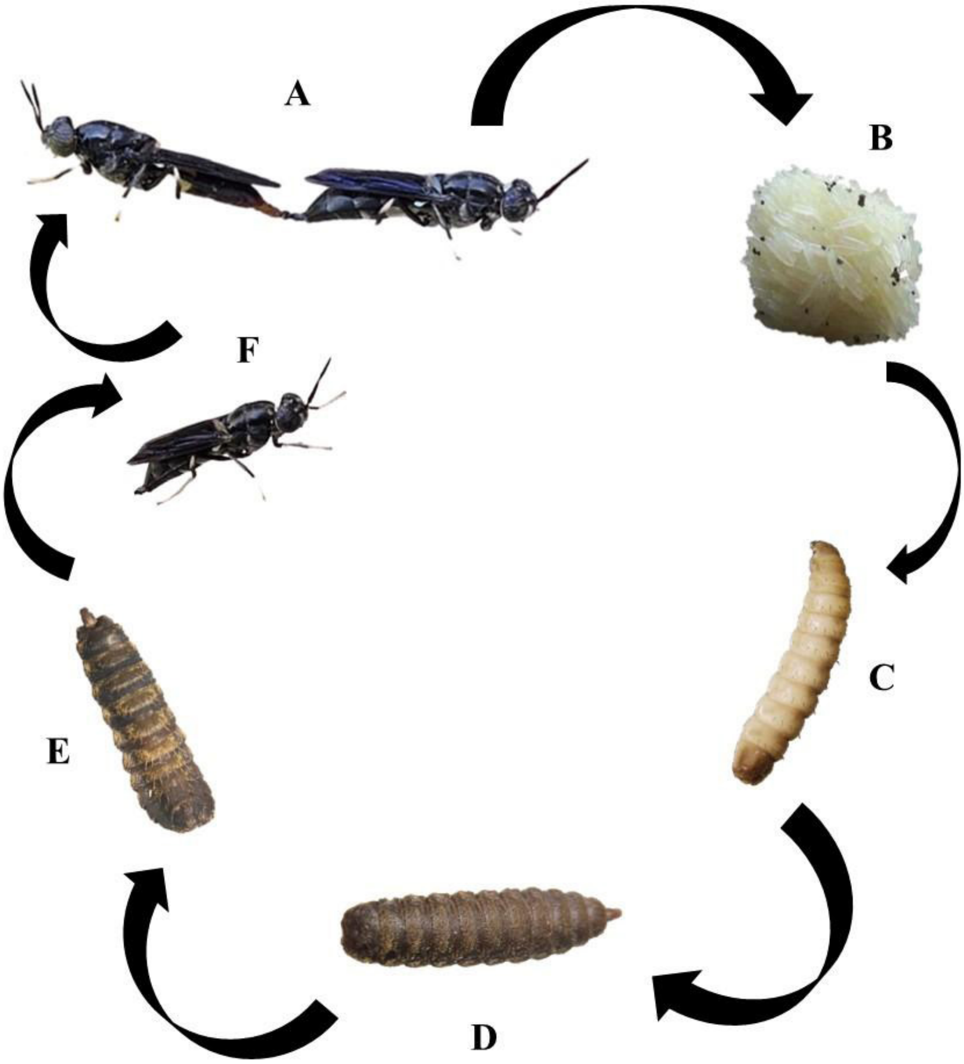

Figure 1. The life cycle of Black soldier fly *Hermetia illucens*. **A.** Male and female copulate. **B.** Females deposit their eggs into the organic matter. **C.** Larvae hatch and feed on the organic matter for approximately 15 days. **D.** Larvae develop into pre-pupae. **E.** Larvae develop into pupae. **F.** Pupae develop into adult specimens.

The daily amount of feed offered was determined using the following formula: Daily feed (g) = fish weight (g) x number of fish x (% feeding/100). The feeding frequency was twice a day (7:30 a.m. and 5:30 p.m.). Growth and weight gain were evaluated every 15 days through sampling, during which biometric data such as standard length and weight were taken (Fig. S2A, S2B). After sampling, fish were returned to their respective experimental units, and the daily feed amount was recalculated as mentioned above.

After 60 days of experimentation, the final data were taken in a last sample where the final values of growth and weight gain were recorded. Subsequently, the data were analyzed, and the zootechnical indices were calculated as described by Castell and Tiews [9]:

(a) Average initial weight of the fish - AIW (g)

AIW = [total weight of the sample (kg) x number of fish in the sample] × 1000

(b) Final average fish weight FAF (g)

FAF is equal to [total weight of the sample (kg) x number of fish in the sample] × 1000

(c) Average weight gain (g) - AWG
AWG = average final weight - average initial weight

(d) Daily growth in weight - DGW (DGW)
DGW = Weight gain/time in days

(e) Initial Biomass - iBIO (kg)
iBIO = Average initial weight (g) x Number of seeded fish

(f) Final Biomass - fBIO (kg)
fBIO = Average final weight x Number of harvested fish

(g) Biomass gain - BG (kg)
BG = Final biomass - Initial biomass.

(h) AFCR (Apparent feed conversion ratio)
AFCR = Amount of feed consumed / Biomass gained

(i) Survival rate
S% = [N° of fish present at the end of the experiment / N° of initial fish] x 100.

Water quality values were recorded daily, and temperature, oxygen, conductivity, and pH were measured using a Hanna HI 9146 multi-parameter instrument (Fig. S2C). The data were stored in Microsoft Excel spreadsheets. The Analysis of Variance test (ANOVA) was performed to determine significant differences between treatments using the statistical program BioEstat 5.0 for Windows. After rejecting the null hypothesis using ANOVA, Tukey's test was applied for pairwise comparisons of means. Tukey's test was chosen for its ability to control the family-wise error rate when comparing multiple means, making it suitable for our experimental design with three treatments. The significance level (α) used for all statistical tests was 0.05, meaning that results with a p-value < 0.05 were considered significant. Before performing the ANOVA, the assumptions of homogeneity of variances and normality of errors were verified. Homogeneity of variances was assessed using Levene's test, and normality of errors was verified using the Shapiro-Wilk test.

Concerning permission to work with biological samples, the IIAP has the respective permits for collection, handling, and experimentation with fish, as indicated by the documents: Directorial Resolution (R.D) N° 217-2016-GRL-DIREPRO and PTH-068-16-PEC-SANIPES.

3. Results

The highest growth (cm) and weight gain (g) were reported in fish fed with balanced feed (T3), followed by those fed with BSF larvae (T1), and finally, fish fed with termites (T2) (Fig. 2). There were significant differences ($p < 0.0001$) between all treatments. The values of the other zootechnical indices evaluated showed the same trend, with the highest values for fish from T3, followed by fish from T2 and T1, respectively (Table 1).

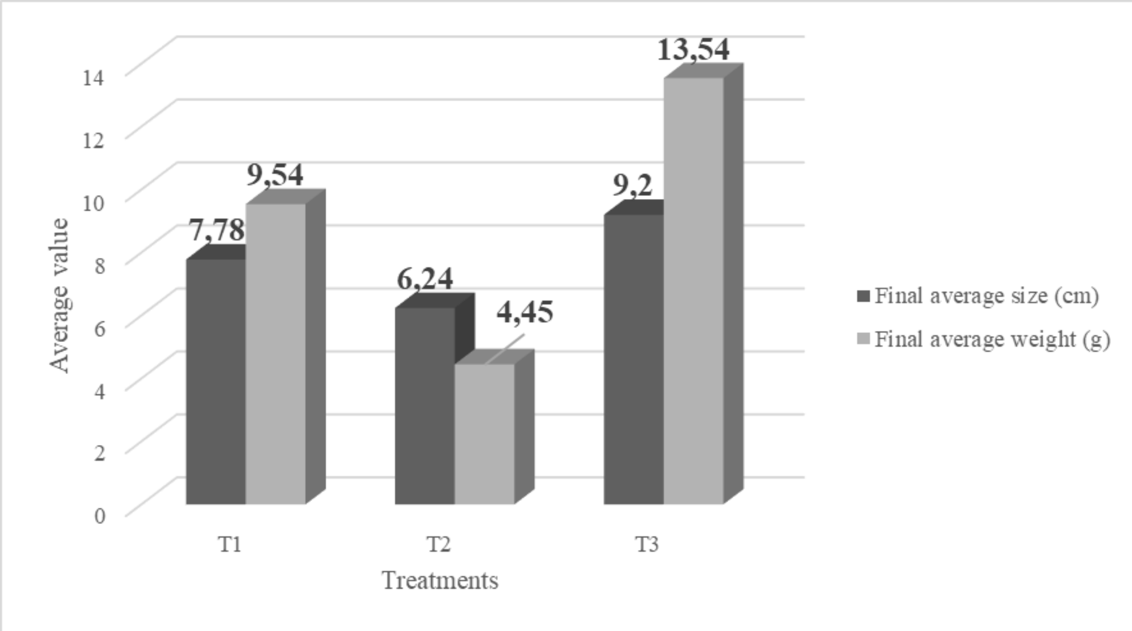

Figure 2. Values of Final average size (cm) and Final average weight (g) of *Colossoma macropomum* feed with three types of feed. T1 = BSF larvae, T2 = termites, and T3 = balanced feed.

Table 1. Values of the zootechnical indices calculated on *Colossoma macropomum* feeding using three types of feed: Black soldier flye larvae – BSF (T1); termites- Te (T2) and balanced food – BF (T3).

	Treatments			
Zootechnical indices	**T1 = BSF**	**T2 = Te**	**T3 = BF**	**p value**
Initial length (cm)	4.35 ± 0.56	4.63 ± 0.73	4.74 ± 0.60	> 0.05
Final length (cm)	7.78 ± 1.48[a]	6.24 ± 0.94[b]	9.20 ± 0.92[c]	< 0.0001
Initial weight (g)	1.29 ± 0.57	1.68 ± 0.94	1.78 ± 0.90	> 0.05
Final weight (g)	9.54 ± 5.34[a]	4.45 ± 2.18[b]	13.54 ± 4.13[c]	< 0.0001
Initial daily feed (g)	38.7	50.4	53.4	-
Length gain (cm)	3.61 ± 0.74[a]	1.65 ± 0.50[b]	4.46 ± 0.08[c]	< 0.0001
Weight gain (g)	8.87 ± 2.64[a]	2.75 ± 0.54[b]	11.76 ± 0.26[c]	< 0.0001
Daily weight gain (g)	0.15 ± 0.04[a]	0.05 ± 0.01[b]	0.20 ± 0.01[c]	< 0.05
Initial Biomass (Kg)	0.02 ± 0.01	0.03 ± 0.01	0.04 ± 0.01	> 0.05
Final Biomass (Kg)	0.12 ± 0.01[a]	0.05 ± 0.02[b]	0.27 ± 0.01[c]	< 0.05
Biomass gain (Kg)	0.10 ± 0.02[a]	0.02 ± 0.01[b]	0.24 ± 0.01[c]	< 0.05
Feed intake (Kg)	0.15 ± 0.01[a]	0.12 ± 0.04[a]	0.37 ± 0.03[b]	< 0.05
Apparent feed conversion ratio	1.52 ± 0.20[a]	6.00 ± 1.51[b]	1.58 ± 0.08[a]	< 0.05
Survival rate	95 %[a]	60 %[b]	95 %[a]	< 0.05

a, b, c = identical letters mean that there is no statistical difference between the treatments. Different letters mean that there is a statistically significant difference between the treatments.

Regarding the physical and chemical water parameters, the average values recorded throughout the experiment were as follows:

For treatment 1: Temperature 29.06 ± 0.69 °C; dissolved oxygen 6.41 ± 0.53 mg/L; conductivity of 67.82 ± 2.89 µS/cm; 6.77 ± 0.31 UI of pH. For treatment 2: Temperature 29.15 ± 0.82 °C; dissolved oxygen 6.30 ± 0.60 mg/L; conductivity of 69.10 ± 2.99 µS/cm; 6.88 ± 0.40 IU of pH. For treatment 3: Temperature 29.20 ± 0.70 °C; dissolved oxygen 6.5 ± 0.55 mg/L; conductivity of 68.9 ± 2.91 µS/cm; 6.85 ± 0.35 IU of pH.

4. Discussion

The results showed that fish fed with balanced feed (T3) had significantly higher growth (4.46 ± 0.08 cm) and greater weight gain (11.76 ± 0.26 g) compared to fish fed with BSF larvae (T1) and termites (T2) ($p<0.0001$). Similar findings were reported in a study on juvenile *C. macropomum* using whole black soldier fly larvae (treatment 1), balanced feed (treatment 2), and a mixture of balanced feed + whole black soldier fly larvae (treatment 3). In that study, fish fed with balanced feed + black soldier fly had similar growth and weight gain values to fish fed only with balanced feed. However, lower growth values were recorded for fish fed only with black soldier fly larvae, although the difference amounted to only 80 g over 120 days of experimentation [10]. Balanced feed is specifically formulated to meet the nutritional needs of fish at each life stage. This ensures that fish receive all the essential nutrients for optimal growth. Additionally, balanced feed is usually processed to facilitate digestion and nutrient absorption. This can be an advantage compared to BSF larvae and termites, which may be less digestible for fish. Although the influence of factors such as feed palatability (fish preference for taste and texture) and feed presentation (particle size, buoyancy, etc.) was not tested in this study, they could also have influenced the results. Therefore, complementary studies testing the impact of these factors would be important in elucidating the above questions.

The BSF larvae treatment (T1) exhibited a better feed conversion ratio compared to the termite treatment (T2), indicating that fish fed with BSF larvae were more efficient at converting feed into growth and weight gain. Fish fed with BSF larvae showed a higher survival rate (95 %) than those fed with termites (60 %). A higher survival rate means that more fish were able to grow and gain weight during the study period. Overall, BSF larvae proved to be a promising alternative to termites for feeding *C. macropomum*, as they provided more adequate nutrition, a superior feed conversion rate, and a higher survival rate, contributing to better growth and weight gain. Black Soldier Fly larvae are high in protein and rich in minerals, essential amino acids, and unsaturated fatty acids [7], a nutritional composition crucial for fish growth and development. Although termites also contain protein, the quality and balance of nutrients may not be as optimal as in BSF larvae, influencing the results obtained in the present study.

In the present investigation, fish fed with T3 (balanced feed) grew and gained weight faster than fish fed with BFS larvae (T1) and termites (T2). However, the values of the zootechnical indices between fish fed with T3 and T1 are not so different compared to fish fed with T2. Similar results were reported in Bahia, Brazil, where the author used BFS larvae to feed *C. macropomum* and registered higher zootechnical indices in fish fed with balanced feed. However, fish fed with BFS larvae presented values close to those found in the present study [11].

In addition, the high protein content, digestibility, and amino acid profile of BFS larvae have been targeted as an input in fish diets. This meal has successfully replaced other conventional sources of protein worldwide: catfish (*Ictalurus punctatus*) [12, 13] blue tilapia (*Oreochromis aureus*) [14], Nile tilapia (*Oreochromis niloticus*) [15], sabaki tilapia (*Oreochromis spilurus*) [16], rainbow trout (*Oncorhynchus mykiss*) [17], Atlantic salmon (*Salmo salar*) [18], turbot (*Psetta maxima*) [19], yellow catfish (*Tachysurus fulvidraco*) [12], and African catfish (*Clarius gariepinus*) [20].

The results of the present research indicate the use of BFS larvae as a nutritionally adequate feed for the growth of *C. macropomum*. Arana et al. [6] reported that larvae and larval meals of black soldier fly larvae in the Peruvian Amazon are good sources of protein, minerals, essential amino acids, and unsaturated fatty acids with high nutritional value. These authors also noted that BFS larvae fed with orange bagasse contain high levels of protein (40-44 %) and lipids (27-31 %), as well as significant contents of potassium, calcium, iron, and manganese. Considering that the commercial balanced feed used in the Peruvian Amazon for *C. macropomum* in its initial stages has a protein level between 32-38 %, the use of a natural and low-cost feed with a similar or superior nutritional profile represents a promising alternative for the sustainable development of Amazonian fish farming.

This indicates that feeding *C. macropomum* with BFS larvae is a viable option due to the positive effects on growth and weight gain observed in this study. Furthermore, it is a cheap alternative that aligns with circular economic technologies, being a sustainable production method for low-income rural fish farmers who could utilize the organic waste generated daily as a substrate to produce BFS larvae, which can then be used to feed farmed fish.

The recorded water quality parameters were within the normal and adequate range for the rearing of Amazonian fish, indicating that the use of the three types of feed did not negatively influence the physical and chemical characteristics of the water. Similar results for water physical and chemical parameters were reported by Almeida (11) and Ordoñez et al. (10).

5. Conclusion

This study evaluated the feasibility of using *H. illucens* (BSF) larvae as an alternative feed for *C. macropomum* in the Peruvian Amazon, comparing them with termites and commercial feed. The results showed that, although commercial feed produced the highest growth and weight gain, fish fed with BSF larvae performed significantly better than those fed with termites, as evidenced by improved feed conversion (1.52 ± 0.20 vs. 6.00 ± 1.51; $p<0.05$) and a significantly higher survival rate (95 % vs. 60 %). These findings suggest that BSF larvae represent a promising and sustainable source of animal protein for *C. macropomum* aquaculture. Their potential lies in the use of organic waste as a production substrate, contributing to a circular economy and reducing dependence on traditional ingredients such as fishmeal. Although balanced feed remains the most efficient option in terms of growth, black soldier fly larvae have emerged as a viable alternative, especially in contexts where sustainability and cost reduction are priorities. Further research is recommended to optimize BSF-based diets and evaluate their long-term impact on the health and quality of the final *C. macropomum* product.

Supplementary material

Figure S1. A. Experimental units for feeding *Colossoma macropomum*. B. Production modules for BSF larvae.
Figure S2. A, B. Biometric sampling conducted every fortnight. C. Measurement of physical and chemical water parameters.

Conflict of interest statement

The authors declare no conflicts of interest.

Data availability statement

The complete dataset supporting the results of this study is available upon request from the corresponding author.

Author contributions

Conceptualization: Morey, A. M. M., Bardales, J. V.; Formal analysis: Arellano, S.A., Chu, A.R., Reáteguil, J. N., Guimaraes, J. L. C., Guzmán, V. H. P. N.; Supervision: Morey, A. M. M., Isern, R. I., Bardales, J. V.; Editor: Morey, A. M. M., Isern, R. I., Viana, D. C.

Generative AI use statement

The authors did not use generative Artificial Intelligence tools or technologies in the creation or editing of any part of this manuscript.

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