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Growth of the Black Soldier Fly *Hermetia illucens* (Diptera: Stratiomyidae) on Organic-Waste Residues and Its Application as Supplementary Diet for Nile Tilapia *Oreochromis niloticus* (Perciformes: Cichlidae)

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Simple Summary: The need to identify sustainable and profitable resources for the elaboration of useful feeds in animal nutrition has been the focus of many studies. Insect-based feeds have been tested by introduction into the diets of pigs, poultry, and fish, with interesting results. Worldwide, solid waste management has increased because of economic and population growth, and most investigations have concentrated on identifying efficient solutions to reduce this waste. Aquaculture has experienced exceptional growth and is perceived as having the greatest potential to meet the growing demand for food in the world, by using aquatic species that serve as a protein source in the diet and therefore contribute to the food security of the population. There are studies with the use of black soldier flies (BSF) as a potential feed agent for animals with high protein content. The relevance of evaluating different food resources in the BSF-immature stages is important because the quantity and quality of the food provided to the fly larvae generate different responses on the growth and development of this insect used as animal feed.

Abstract: The black soldier fly, *Hermetia illucens* (BSF, Diptera: Stratiomyidae) is an insect with high protein value and a potential feed agent for animals aimed for human consumption. The growth parameters of BSF larvae reared on four substrates—restaurant-waste, fruit-waste, fish-waste, and commercial tilapia food—for 41 days before processing for inclusion into *Oreochromis niloticus* (Perciformes: Cichlidae, Nile tilapia) commercial fry diets at 30% (70:30) were determined. On fly larvae, the food substrate based on restaurant waste yielded the greatest larval weight and length. BSF larvae fed a fish-waste diet showed the shortest developmental time. The fruit-waste diet

induced the lowest weight and length in the fly larvae/pre-pupae (immature stage). The pre-pupal protein values were similar to commercial food. On fry-fish, the diets with pre-pupae grown on fish waste showed the greatest yields regarding weight (biomass), length, and nutritional content. These results suggest the BSF has the potential to be used in fish feed and provides an alternative for commercial cultivation.

Keywords: organic urban waste; fish-meal; economic alternative; *Hermetia*

1. Introduction

Worldwide, solid waste management has increased because of economic and population growth [1,2]. It is common to dispose of this waste in streets or open dumps, blocking drains, attracting disease vectors, and causing public health problems [3]. Recently, increasing research has been focused on developing efficient solutions to degrade and re-use these resources. One of them is using waste as a food substrate for insect populations, which can serve as a protein source for the production of animal feed [4]. Many studies have focused on the need to identify sustainable and profitable resources as functional foods in animal nutrition. There were interesting results when insect flour was incorporated as a protein source into diets for pigs, poultry, and fish [1,5–12]. Among the insects used in aquaculture feeding, arthropods belonging to the taxonomic orders Orthoptera, Coleoptera, Isoptera, Lepidoptera, and Diptera have been cited [7]. The Diptera order is one of the largest groups of insects, with approximately 100 families and 85,000 described species, most of which have economic importance because they participate in organic-matter degradation [13]. The Stratiomyidae family includes the Brachycera sub-order with 2600 species registered worldwide [14–16].

The black soldier fly *Hermetia illucens* L. (BSF, Diptera: Stratiomyidae) is distributed in the tropics and is found in humid and nutrient-rich ecosystems with an abundance of decomposing animal and plant remains; larval nutritional composition can reach up to 40% protein and 30% fat, and varies depending on the substrate used for growth [17,18]. There are also records of growth improvement in poultry, pigs, and commercial fish species fed with insect flour [8]. Protein and fat values observed in these flours meet the requirements established for fish feeding by the Organization of the United Nations for Food and Agriculture (FAO) [19].

BSF larvae have a significant protein content and possess a well-balanced amino acids and fats. Nutritional composition and fatty acid profiles are dependent on the developmental stage [20], making them a good nutritional source of animal feed. BSF is an excellent organic-waste bioconversion organism, but the type of substrate where BSF is reared is quite important for improved bioconversion activity, because of the nutrients needed to support its growth [21–23]. Several studies concentrate their aim on BSF larval nutrient assimilation in response to specific rearing conditions and substrates to improve their yield and quality [24,25]. In addition, BSF-derived products such as fats can be used as biodiesel [20], and oil can be used for medical proposes and as a skincare product [26]. Another valuable product gained from BSF is chitin—chitosan is widely used in medicine, cosmetics, and biotechnological research [27]. Hence the importance of black soldier fly research.

Several substrates such as manure, butchery, and vegetable wastes [28], fruits [29], kitchen waste [30], and coffee by-products (coffee silverskin) enriched with 10% of *Schizochytrium* sp. have been tested for BSF larval growth [31]. Each substrate confers different conditions for larvae growth and different nutrition values, development time, size, and biomass [32]. For instance, Scala et al. [33] found that larvae fed on diets containing spent grain contained 15% or more protein than those larvae reared only on fruit

Aquaculture has experienced exceptional growth and is perceived as having the greatest potential to meet the growing demand for food worldwide by using aquatic

species that serve as a protein source in the diet and therefore contributing to the food security of the population [34,35]. In Mexico, exploitation of Nile tilapia (*Oreochromis niloticus* L., Perciformes: Cichlidae) is an option that has been promoted to generate income for inhabitants of tropical areas. This has been accepted because of its ease of cultivation. However, rural producers in the southeast of the country face a constant problem with the cost, scarcity, and quality of commercial balanced foods [36]. The importance of evaluating different food sources for the BSF resides in the fact that factors such as moisture, nutritional composition, quantity, and quality of the food provided to the larvae may generate different responses in the development and growth of this insect and can also affect survival in larval stages and nutrient assimilation [28,37].

The present study investigated the effects on *O. niloticus* growth performance of partial dietary replacement of fishmeal by BSF prepupae cultured on four different organic substrates. A BSF stock colony was also set up to determine BSF survival and the development time for several stages, thus identifying parameters that would benefit the establishment of an optimized production breeding program (weight/biomass, length, and nutritional content). We hypothesized that larval development and biomass gain of fish depends on the nutrient content of the type of substrate on which the larvae grow.

2. Materials and Methods

2.1. Insect Rearing

The flies were reared in the facilities of the Applied Entomology Laboratory (LEA as in its Spanish abbreviation) of the Benito Juárez Autonomous University of Oaxaca (UABJO as in its Spanish abbreviation, 17°03'17.6" N, 96°43'37.6" W) in a 4 × 3 × 3 m concrete room with an artificial lighting system (Master-TL5 High Output Eco 109 LM/W Philips Lighting Holding B.V.), 12:12 light and dark cycle under a constant temperature of 27.00 ± 1.03 °C, and relative humidity of 51.77 ± 6.23% [38]. Eggs were obtained from a colony of flies established in the LEA-UABJO and reared in a 100 × 80 × 80 cm entomological cage where the larvae fed ad libitum. A mixture of 500 g of commercial tilapia food (Atilapia®, Aquaplus-Campi, Mexico) with 800 mL of distilled water in a plastic container (5 L) was prepared, then corrugated cardboard strips (to encourage oviposition into the holes of the cartons) on the food mixture were placed inside of the brooding cage to collect the eggs [39].

2.2. Substrates for Fly-Larvae Development

The larvae were developed on four different substrates: (1) restaurant-waste composed (approximately) of cooked chicken meat (5%), pork (5%), beef (5%), cooked tomato (20%), corn tortilla (50%), and rice (15%); (2) fruit-waste composed of cucumber (10%), pineapple (20%), watermelon (20%), mango (25%), papaya (20%), and carrot (5%) residues; (3) fish-waste composed of muscle (40%), skin (25%), and gills (35%) of *Solea solea* L. (Pleuronectiformes: Soleidae); and (4) commercial tilapia food (Atilapia®) as a control (40.13% crude protein, 5.96% crude fat, 12.00% moisture, and 10.03% ash) [37]. The chemical-nutritional characteristic of each substrate was performance.

2.3. Black Soldier Fly Growth

A cardboard strip with packets of BSF eggs ($n = 150$) was placed on 40 g of each food substrate in a 2 L plastic container. Three days after hatching, we recorded data every 24 h until 40% of the larvae had reached the pre-pupal stage and until the insects reached the adult phase [39]. Three replicates per food substrate were carried out. Ten (10) larvae were sampled in each replica and then returned to their respective container [37,38]. The length and gained weight for each larva were measured with a Vernier caliper (Model-CVQ1315) and an electronic scale, respectively (Scout® Pro, Ohaus Corporation, Germany).

2.4. Nile Tilapia Growth Test

For fish-diet preparation, pre-pupae reared on the mentioned food substrates were used. Pre-pupae were dehydrated in an oven at 90 °C (Felisa EU) for 96 h and then crushed

using a mechanical mill and the obtained powder mixed with grounded commercial tilapia food (Atilapia®) at a ratio of 30% [40–42]. The resulting mixture was moistened with distilled water, obtaining a slurry with a semi-liquid consistency which then was mixed in a blender for 10 min. Flat surface metal trays were prepared, and a thin layer (0.2 mm) of the slurry was spread and allowed to dry at room temperature for 48 h. The flakes were removed with the help of a spatula and packed into flexible polyethylene zip-lock bags.

For this bioassay, hormonal 7-day-old *O. niloticus* fries were used. The fingerlings were obtained from Laboratorio Central Acuicola S.A., in the State of Campeche, Mexico (19°45′33.41″ N, 90°28′00.59″ W). The fish fry were acclimatized for two days in a 1000 L tank at the facilities of the Aquatic Ecology and Environmental Monitoring Laboratory of the Center for Studies in Sustainable Development and Use of Wildlife (CEDESU), at the Autonomous University of Campeche (UAC) (19°48′07.63″ N, 90°30′16.31″ W). For the experiment, 12 aquaria, with a capacity of 38 L each, were used. The aquaria were oxygenated using air pumps (Elite model A-802) and the oxygen concentration kept between 5.0 and 9.0 mg/L. The seeding density was 3.75 L/fish.

The fish were fed with 30% BSF-pre-pupae flour inclusion in commercial food with 100% commercial tilapia food (Atilapia®) as a control group. The fish-fry feed was carried out by supplying 5% of the total weight of the fish biomass at four-hour intervals, starting at 08:00 each day [43]. The amount of food was provided according to the increase in average weight and biomass of each sampling. The fish fry were randomly distributed in each aquarium, and each diet experiment was carried out in triplicate ($n = 3$). Sampling was carried out every six days, with total length and biomass gain (wet weight) of fish recorded with an ichthyometer (model KH-PISCIS-60-22) and an electronic scale (Scout® Pro, Ohaus Corporation, ± 0.001 g sensitivity). The salinity, dissolved oxygen, and water column temperature were recorded daily using a portable probe (HACH model HQ40d).

2.5. Nutritional Analysis of the BSF-Diet and Muscle of Nile Tilapia

For this, 100 g of the wet matter was used and placed on an aluminum tray. The samples were dried in a convection oven (PerfectBroil, CTO4400B, Black & Decker Corporation, Towson, MA, USA) at 100 °C for 15 min and cooled before recording dry weight. The samples were then placed in a mill (Fritsch Pulverisette-14), pulverized, and stored in zip-lock polyethylene bags. For moisture determination, 2 g of the ground matter was weighed and placed in the oven at 105 ± 0.05 °C for 24 h until a constant weight was achieved [44,45]. Moisture was calculated using the following formula:

$$(a) \text{ Moisture (\%)} = ((\text{wet weight} - \text{dry weight}) / (\text{wet weight})) \times 100$$

$$(b) \text{ Dry matter (\%)} = 100 - \text{Humidity (\%)}$$

2.6. Ash Determination

For this, 2 g of ground matter were weighed from each treatment and placed into a 50 mL crucible. These samples were then placed into a Felisa® MM 60 muffle at 500 °C for one hour to obtain carbon-free ash. Ash percentage was determined by the following formulae [46]:

$$(a) \text{ Wet base ash} = ((\text{ash weight} + \text{crucible weight}) - (\text{crucible weight} \times 100)) / (\text{weight of the sample})$$

$$(b) \text{ Dry base ash} = ((\text{wet base ash (\%)} / (100 - \text{moisture (\%)})) \times 100$$

2.7. Determination of Crude Fat Content

The crude fat content was determined according to the Mexican standard and AOAC [44] procedures by using the ethereal extract determination (Soxhlet) method for meat products. For this, 2 g of the sample was weighed, placed on filter paper, covered with a cotton portion, and placed into the extractor. A 1000 mL flat-bottomed balloon flask was placed in the lower area, and 80 mL of ether was added to the upper end of the condenser. The extraction

was carried out for six hours. The ether in the flask was evaporated and dried to constant weight. Fat content was quantified by weight difference using the following equation:

$$\text{Fat (\%)} = ((\text{PG} - \text{P})/\text{m}) \times 100$$

where PG is the mass in grams of the flask with fat, P is the mass in grams of the empty flask, and m is the mass in grams of the sample.

2.8. Determination of Proteins in the Diet Based on Pre-Pupae

A fresh sample of diets based on pre-pupae was used with bovine serum albumin as a standard at 1 mg/mL. Each sample was homogenized with a mortar. For this, 1 g of protein was measured with a digital weighing scale and mixed with 9 mL of sucrose solution. This mixture was centrifuged at 4000 rpm for 8 min. Then the supernatant was collected and 1 mL of protein sample was added to 4 mL of alkaline copper solution using a vortex shaker. These solutions were incubated at room temperature for 12 min. After this, 0.5 mL of Folin–Ciocalteu phenol was added to these solutions and mixed by using a vortex shaker. Finally, these solutions were incubated for 30 min at room temperature and in the dark. All absorbance readings were carried out in triplicate at 750 nm using a UV–VIS spectrophotometer (Thermo Scientific, Waltham, MA, USA) [47].

2.9. Determination of Proteins in the Muscle of Nile Tilapia

For this, 0.25 g of fish-muscle tissue samples from Nile tilapia was weighed and placed into an 800 mL balloon flask. The sample (6 mL) was digested in an oven at 400 °C with concentrated sulfuric acid in the presence of potassium sulfate and a catalyst (half a Kjeldahl tablet: K₂SO₄, 48.40%; Na₂SO₄, 48.30%; CuSO₄, 0.30%) to speed up the digestion and obtain nonvolatile ammonium sulfate. After cooling, 30 mL of distilled water was added, and the ammonium sulfate was converted to volatile ammonia gas by heating with 20 mL of sodium hydroxide. Then 10 mL of boric acid solution was added to the steam-distilled ammonia, which was trapped by forming ammonium borate. The amount of borate formed was determined by titration using hydrochloric acid [44]. The protein content was calculated using the following formulae:

$$\text{(a) Nitrogen (\%)} = ((\text{g nitrogen in sample})/(\text{g of sample})) \times 100$$

$$\text{(b) Protein (\%)} = \% \text{ N (total nitrogen content)} \times \text{Kjeldahl factor for meats (6.25)}$$

2.10. Ethical Standards

Our experiments were carried out according to the ethical procedures for the production, care, and use of laboratory animals of the Mexican National Standard [48].

2.11. Experimental Design and Statistical Analysis

All experiments were carried out using a completely randomized design with three replicates and data presented as the mean \pm standard deviation (SD) to provide an idea of the magnitude of the differences between means. The data were tested to verify the normality of errors (Shapiro–Wilk test) and homogeneity of variances (Bartlett test). One-way analysis of variance (ANOVA) was performed and mean comparison using the Tukey test ($p < 0.05$) as a post-hoc test carried out with the aid of Minitab version 18.1 to evaluate egg-hatching time, larvae–pupae developmental days, percentage of larval survival, larval–prepupal weight (g), and length (mm) for each substrate tested (restaurant waste, fruit waste, fish waste, and commercial food), along with the total caudal length (cm) and weight gain (biomass) of fish development for each diet containing 30% of larvae grown in each substrate and 100% commercial food as a control. In the study, these data were included as factors in the analysis. The nutritional composition percentages (moisture, ashes, protein, and fat) of BSF pre-pupae meal and muscle of *O. niloticus* fed with the diets were also evaluated.

3. Results

3.1. Survival and Development of BSFs

Significant differences in BSF development were found (Table 1). The restaurant-waste-based substrate extended the biological cycle of the BSFs to 40.50 ± 0.50 days (d) when compared to the rest of the food substrates, whereas the control group registered a cycle of 33.00 ± 0.81 d. BSF fed on fruit waste registered a development rate of 34.87 ± 0.85 d from egg to adult emergence. The fish-waste-based substrate induced a lower egg–adult development period than the rest of the treatments (31.33 ± 0.47 d). We suggest that the larvae could complete their cycle in a shorter period because of the amount of protein and fat provided by this waste. The control substrate showed the highest larval survival percentage at $98.88 \pm 1.92\%$, contrary to that recorded by restaurant-waste treatment, which yielded the lowest rate ($76.66 \pm 3.33\%$) ($F_{1,3} = 8,23$; $p < 0.008$; $r^2 = 75.53$). The fish-waste treatment registered a larval survival of $88.00 \pm 1.15\%$. Table 2 shows the chemical composition of the substrates used in the trial.

Table 1. Survival (%) and development (days) of BSFs reared on diverse food-waste-based substrates.

Waste-Based Substrate	Hatching Time (days)	Development Days			Larval Survival (%)
		Larvae	Pupae	Total	
Restaurant waste	2.98 ± 0.21 a	20.95 ± 0.75 a	15.87 ± 0.85 a	40.50 ± 0.50 a	76.66 ± 3.33 a
Fruit waste	2.88 ± 0.22 a	22.87 ± 0.74 b	8.87 ± 0.75 c	34.87 ± 0.85 b	76.86 ± 12.13 a
Fish waste	2.99 ± 0.10 a	15.66 ± 0.57 c	12.93 ± 0.72 ab	31.33 ± 0.47 bc	88.00 ± 1.15 b
Commercial Food	2.98 ± 0.21 a	14.62 ± 0.47 cd	16.25 ± 0.50 a	33.00 ± 0.81 bc	98.88 ± 1.92 c

Means followed by the same lower-case letter within each column are not significantly different based on Tukey test at $p < 0.05$. Commercial food: tilapia food (Atilapia). Larval development is the average amount of time required to reach the prepupal stage. Pupal development is the amount of time that it remained in that state until the adult emerged. Total development is the number of days elapsed between when the eggs hatched and the adult emerged.

Table 2. Substrate chemical composition.

Sample	Moisture (%)	Ash (%)	Protein (%)	Fat (%)
Restaurant waste	71.87 ± 0.73 b	4.06 ± 0.15 c	14.5 ± 0.30 d	12.36 ± 0.37 a
Fruit waste	69.67 ± 0.51 c	2.46 ± 0.15 d	15.73 ± 0.32 c	2.73 ± 0.15 c
Fish waste	80.56 ± 0.50 a	5.60 ± 0.30 b	48.70 ± 0.43 a	5.90 ± 0.20 b
Commercial food (Atilapia [®])	12.00 ± 0.10 d	10.03 ± 0.15 a	40.13 ± 0.32 b	5.96 ± 0.06 b

Means followed by the same lower-case letter within each column are not significantly different based on Tukey test at $p < 0.05$.

3.2. Weight (Biomass) and Length of Fly Larvae/Pupae

Substrate significantly affected the total weight of pre-pupae. Pre-pupae reared on restaurant waste were higher in biomass (29.9 g on average considering the total number of larvae and larval survival) registering a weight-gain of 0.25 ± 0.04 g, which represents an increase of 16 to 32% when compared to fruit and fish waste (19.59 and 27.72 g on average) ($F_{3,11} = 12.90$; $p < 0.001$; $r^2 = 25.01$). The fruit-waste substrate produced a significantly lower weight (0.17 ± 0.04 g) when compared to the rest of the treatments ($F_{3,11} = 29.00$; $p < 0.001$; $r^2 = 42.86$). There were significant differences between pairs of means from restaurant-waste and control groups vs. fruit and fish-waste groups. The first two treatments produced the highest weight (23.78 ± 1.86 g and 23.19 ± 2.61 g, respectively). BSF in all substrates showed a constant increment before harvesting. The larval length registered for restaurant-waste treatment was significantly lower (23.59 ± 1.84 mm) compared to the rest of the treatments (<12.2–19.45%) ($F_{3,11} = 18.06$; $p < 0.001$; $r^2 = 31.84$) (Table 3).

Table 3. Larval and pre-pupal length and weights of BSFs reared under different waste-based substrates.

Waste-Based Substrate	Larval wt (g)	Larval lt (mm)	Prepupal wt (g)	Prepupal lt (mm)
Restaurant waste	0.25 ± 0.04 a	23.59 ± 1.84 a	0.26 ± 0.05 a	23.78 ± 1.86 a
Fruit waste	0.17 ± 0.05 c	19.00 ± 3.33 b	0.17 ± 0.04 c	19.73 ± 1.75 b
Fish waste	0.21 ± 0.05 b	19.89 ± 2.58 b	0.21 ± 0.02 b	20.56 ± 1.62 b
Commercial Food	0.19 ± 0.05 bc	20.71 ± 2.25 b	0.26 ± 0.07 a	23.19 ± 2.61 a

wt, weight; lt, length. Means followed by the same lower-case letter within each column are not significantly different based on Tukey test at $p < 0.05$. Commercial food: tilapia food (*Atilapia*).

3.3. Bromatological Composition of the Food Based on Fly Pre-Pupae

During BSF base-diet preparation, data showed two main types of consistencies. Pre-pupae developed in fruit waste had a final consistency of powder with a fat percentage of $29.37 \pm 0.52\%$ ($F_{3,11} = 5031.57$; $p < 0.001$; $r^2 = 99.95$), larvae developed using restaurant and fish waste had a pasty consistency with a fat percentage of 46.54 ± 0.58 ($F_{3,11} = 756.57$; $p < 0.001$; $r^2 = 99.65$) and $46.50 \pm 0.45\%$ ($F_{3,11} = 30.00$; $p < 0.001$; $r^2 = 91.84$), respectively, while the control group registered $6.00 \pm 0.25\%$ ($F_{3,11} = 2276.77$; $p < 0.001$; $r^2 = 99.88$). Regarding protein content, the pre-pupae developed using the restaurant and fish-waste substrates presented the same percentage as the control group ($33.00 \pm 0.50\%$). Significant differences in the data for moisture and ash content between all the treatments were observed (Table 4).

Table 4. Nutritional composition (%) of BSF pre-pupae meal fed with different organic waste.

Sample	Moisture (%)	Ash (%)	Fat (%)	Protein (%)
Restaurant waste	3.99 ± 0.01 ^d	7.74 ± 0.01 ^c	46.54 ± 0.58 ^a	33.00 ± 1.00 ^a
Fruit waste	5.97 ± 0.03 ^b	13.88 ± 0.10 ^a	29.37 ± 0.52 ^b	28.00 ± 0.50 ^b
Fish waste	5.05 ± 0.03 ^c	6.32 ± 0.03 ^d	46.50 ± 0.45 ^a	33.00 ± 1.00 ^a
Commercial food	12.00 ± 0.25 ^a	9.00 ± 0.40 ^b	6.00 ± 0.25 ^c	33.00 ± 0.50 ^a

Means followed by the same lower-case letter (a, b, c, d) within each column are not significantly different based on Tukey test at $p < 0.05$. Commercial food: tilapia food (*Atilapia*).

3.4. Development of Nile Tilapia

A significant increase in fish biomass was found for fish fed with the fish-waste-based diet (gain biomass = 5.88 ± 1.88 g) when compared to the control group (1.22 ± 1.10 g). This diet presented the highest weight gain when compared to the restaurant-waste-based diet (3.77 ± 2.15 g) and fruit-waste-based diet treatments (4.14 ± 1.67 g) ($F_{3,11} = 10.54$; $p < 0.001$; $r^2 = 49.70$). Fish fed with the fish-waste-based diet reached a total length of 2.38 ± 0.71 cm, which was significantly higher than the rest of the treatments (0.62 ± 0.64 and 1.94 ± 0.66 cm) ($F_{3,11} = 9.58$; $p < 0.001$; $r^2 = 47.33$). In addition, a greater gain in caudal length was observed for the fish-waste group (2.10 ± 0.56 cm), while the smallest values were observed in the control group (0.49 ± 0.50 cm) ($F_{3,11} = 9.40$; $p < 0.001$; $r^2 = 46.84$) (Table 5).

3.5. Nutritional Analysis of Muscle Tissue from Nile Tilapia

Bromatological analysis showed that the fish-waste-based diet had the highest protein percentages ($3.48 \pm 0.03\%$) ($F_{3,11} = 494.17$; $p < 0.001$; $r^2 = 99.46$), followed by the 100% commercial control group ($3.06 \pm 0.06\%$) ($F_{3,11} = 8.63$; $p = 0.007$; $r^2 = 76.40$) and the fruit-waste-based diet ($3.02 \pm 0.20\%$) ($F_{3,11} = 58.91$; $p < 0.001$; $r^2 = 95.67$). The restaurant-waste-based diet had the lowest protein content ($2.23 \pm 0.10\%$) ($F_{3,11} = 4.75$; $p = 0.035$; $r^2 = 64.05$). The diet with the highest amount of crude fat was the fish-waste-based diet with $3.45 \pm 0.03\%$, whereas we found the lowest fat content in the restaurant-waste-based diet ($1.51 \pm 0.10\%$) (Table 6).

Table 5. Chronology of *O. niloticus* development fed on diets based on 30% of BSF meals.

Variable	Diets	Fish Sample						Total Gain (Biomass)
		1st Day	6th Day	12th Day	18th Day	24th Day	30th Day	
TL (cm)	dRW	5.54 ± 0.32 b	6.25 ± 0.30 a	6.41 ± 0.52 a	6.71 ± 0.47 b	6.90 ± 0.59 b	7.33 ± 0.57 b	1.79 ± 0.86 a
	dFW	5.73 ± 0.52 ab	6.21 ± 0.28 a	6.53 ± 0.38 ab	6.86 ± 0.48 ab	7.01 ± 0.42 b	7.67 ± 0.50 b	1.94 ± 0.66 a
	dFiW	5.81 ± 0.42 ab	6.43 ± 0.44 a	6.88 ± 0.60 ab	7.23 ± 0.39 a	7.67 ± 0.46 a	8.18 ± 0.53 a	2.38 ± 0.71 ab
	dHW	6.04 ± 0.26 a	6.33 ± 0.43 a	6.50 ± 0.32 b	6.64 ± 0.30 b	6.67 ± 0.40 b	6.67 ± 0.44 c	0.62 ± 0.64 c
CL (cm)	dRW	4.43 ± 0.48 a	5.07 ± 0.28 a	5.13 ± 0.47 a	5.38 ± 0.39 a	5.52 ± 0.48 b	5.89 ± 0.56 bc	1.44 ± 0.90 a
	dFW	4.54 ± 0.46 a	5.08 ± 0.28 a	5.31 ± 0.30 a	5.53 ± 0.48 a	5.64 ± 0.39 b	6.20 ± 0.45 ab	1.65 ± 0.61 a
	dFiW	4.58 ± 0.34 a	5.24 ± 0.40 a	5.62 ± 0.50 a	5.86 ± 0.38 a	6.24 ± 0.36 a	6.68 ± 0.44 a	2.10 ± 0.56 ab
	dHW	4.88 ± 0.25 a	5.18 ± 0.41 a	5.24 ± 0.25 a	5.47 ± 0.33 a	5.34 ± 0.32 b	5.38 ± 0.37 c	0.49 ± 0.50 c
W (g)	dRW	2.73 ± 0.51 b	4.17 ± 0.75 a	4.47 ± 1.23 a	4.61 ± 0.97 b	5.25 ± 1.28 b	6.51 ± 1.68 bc	3.77 ± 2.15 a
	dFW	3.40 ± 0.86 ab	4.31 ± 0.54 a	4.61 ± 0.82 a	5.51 ± 1.31 ab	5.87 ± 1.24 b	7.56 ± 1.33 ab	4.14 ± 1.67 a
	dFiW	3.23 ± 0.76 ab	4.97 ± 1.05 a	5.81 ± 1.59 a	6.18 ± 1.15 a	7.66 ± 1.51 a	9.11 ± 1.67 a	5.88 ± 1.88 ab
	dHW	3.76 ± 0.44 a	4.57 ± 1.14 a	4.65 ± 0.74 a	5.04 ± 0.85 ab	5.03 ± 0.92 b	4.98 ± 1.12 c	1.22 ± 1.10 c

Means (±SE) of three replicate aquaria (10 fish/aquarium). Means followed by the same lower-case letter within each column by variable (TL, CL, W) are not significantly different based on Tukey test at $p < 0.05$. TL, total length (cm); CL, caudal length (cm); W, weight (g); dRW (30% BSF from RW); dFW (30% BSF from FW); dFiW (30% BSF from FiW); dHW (100% commercial food for tilapia); RW, restaurant-waste-based substrate; FW, fruit-waste-based substrate; FiW, fish-waste-based substrate.

Table 6. Nutritional composition in the muscle of *O. niloticus* fed with diets based on 30% flour from the black soldier fly.

Sample	Moisture (%)	Ash (%)	Protein (%)	Fat (%)
dRW (30% BSF from RW)	76.85 ± 0.08 a	2.50 ± 0.05 ab	2.23 ± 0.10 c	1.51 ± 0.10 d
dFW (30% BSF from FW)	75.33 ± 0.50 b	2.42 ± 0.10 ab	3.02 ± 0.20 b	3.22 ± 0.10 b
dFiW (30% BSF from FiW)	75.12 ± 0.10 b	2.29 ± 0.10 b	3.48 ± 0.03 a	3.45 ± 0.03 a
dHW (100% commercial food for tilapia)	75.93 ± 0.75 ab	2.55 ± 0.10 a	3.06 ± 0.06 b	1.80 ± 0.05 c

Means followed by the same lower-case letter within each column are not significantly different based on Tukey test at $p < 0.05$. RW, restaurant-waste-based substrate; FW, fruit-waste-based substrate; FiW, fish-waste-based substrate.

4. Discussion

A diet based on insect-derived products is a protein alternative of interest for quality feeding [49]. Furthermore, insects are efficient biological conversion agents, their reproduction can be carried out at large scales, and they present a sustainable option [50].

4.1. Survival and Development of BSF Larvae

The present study determined the variation and growth and development responses of BSF larvae fed with food substrates based on organic waste. There are previous studies that have described the use of balanced food for developing BSF larvae. For example, Tomberlin et al. [39] evaluated larval development of BSFs with commercial food for laying birds and two commercial feeds, without finding significant differences in prepupal development or survival. Samayoa et al. [51] reported that the average larval development time for the BSF at 28 °C was 23.03 days, while the average development from egg to adult was 47.44 days. Conversely, results obtained in the present study showed a larvae development on the fruit-waste-based substrate of 22.87 days. Tomberlin et al. [39] reported that the BSF reared at 27 °C had an average development time egg–adult of 41.50 days (41–43 days) with a larval stage of 23.30 days (22.50–24.10 days). These data are quite similar to our findings, in which the restaurant-waste-based substrate recorded a time of 40.50 ± 0.50 d for adult-egg development, and the fruit-waste-based diet recorded a time of 22.87 days in the larval stage, at an average temperature of 27.00 ± 1.03 °C and relative humidity of 51.77 ± 6.23%. Variations found in our study could be attributed to the fact that the development time depends on several factors, such as the composition and proportion of waste from the food substrates, the environmental conditions of development, and the size of test populations [52].

Oonincx et al. [53] reported survival of 72.00 ± 12.90 to 86.00 ± 18.00% in diets planned with high and low amounts of protein. These findings are in agreement with our results,

except for the case of commercial food. Barragan-Fonseca et al. [54] mentioned that larval survival of BSFs in meat waste including fish waste was $48.20 \pm 8.70\%$. In contrast, in our experiment, larva survival in fish waste was twice as high ($88.00 \pm 1.15\%$). It is, however, important to mention that variations in the development time of the immature stages of insects may vary according to the space and number of individuals used in the rearing since there is competition for these factors.

4.2. Biomass of BSF Pre-Pupae and Length of Larvae

In our study, diet significantly affected larval growth, and this may be related to specific physicochemical characteristics of each substrate, which makes the insect–substrate interaction complex [55]. Insects can biodegrade organic wastes into insect biomass [56], which shows that substrate conversion efficiency implies a balance in food consumption to survive [57], indicating that insects can adapt to any quality of substrate [58]. Barroso et al. [59] and Surendra et al. [60] mentioned that the pre-pupae collected contained between 41 and 44% of proteins that can feed animals. However, the use of larvae or pre-pupae depends on their nutritional composition, as the levels of protein and fat absorption may vary according to the growth medium [61,62].

Nguyen et al. [37] reported that black soldier larval length reared on substrates based on kitchen waste was 20.80 mm, for fruit and vegetable waste it was 18.70 mm, and for the substrate based on fish waste it was 19.00 mm. This is similar to our findings, in which restaurant-waste-based substrate yielded lengths of 23.59 mm, for fruit-waste-based substrate it was 19.00 mm, for fish-waste-based substrate, 19.89 mm, and for commercial food, 20.71 mm. Studies have shown that the type and amount of diet influence the growth (weight and height) of BSFs [63]. This is a key issue in the generation/development of adult or immature stages that allow the production of greater usable biomass [64,65], which can ensure a steady insect population [66].

Several studies have focused on the moisture content/development relationship (substrates with immature stages), and they have reported that a high moisture content increases larval growth, but decreases larval weight [67,68]. We observed this situation in the fruit-waste substrate, which presented the fastest development ($31.33 + 0.47$ d), but the lowest weight gain ($0.17 + 0.04$ g).

4.3. Bromatological Composition of Food Based on the BSF

St-Hilaire et al. [69] mentioned that the BSF larvae contain the fat omega-3 (3.0%), which makes it attractive to include in animal feed. In our study, the amount of protein was similar to commercial food in all treatments based on BSF pre-pupae (28–33%). However, Spranghers et al. [70] mentioned that their findings reached fat percentages between 39.9 and 43.1%, whereas Barragan-Fonseca et al. [54] reported a range between 37.0 and 63.0%. The percentage of ash found with the fruit-waste-based food was highest (13.88%) among the foods made with BSF pupae (6.32 and 9.00%), which was similar to the results found by Devic et al. [35]. Yet, the high ash content in the BSF-diet could be by deposition of calcium carbonate by the epidermis of the larval skin from the molting process [71]. In our study, moisture analysis was 12.0%, which was twice as high as in other studies (3.99 and 5.97%).

4.4. Development and Nutritional Analysis of the Nile Tilapia Muscle

We found significant differences in the growth and weight of *O. niloticus* when fed with integrated diets containing 70% of commercial food for tilapia and 30% of food based on fly pre-pupae. El-Saidy and Gaber [72] and Monentcham et al. [40] did not find significant variations in tilapia fed with 25 and 30% inclusion of BSF flours in fish feed until they increased the amount of feed to 1, 2, and 3% of weight daily, similar to the data recorded in our work. On the other hand, Sealey et al. [41] reported that diets including 25 and 50% of fly flour did not yield significant differences in the size and weight of fish, contrary to our finding.

Chatzifotis et al. [73] mentioned that the fat composition directly affected the proximal content of muscle in fish development. In our study, the fish-waste-based diet registered a 46.55% fat content and induced a greater gain in height and weight. Therefore, we recommend this level of fat for stable development of the fish, thus supporting the information cited by other authors mentioning that it is not convenient to completely replace the diet or make inclusions that exceed proportions of 40% of fly flour [42] unless partial defatting is carried out [9,54,74] to increase digestibility [75].

Cammack and Tomberlin [62] and Dzepe et al. [29] tested fruit waste and chicken-manure substrates with increasing moisture content (40 to 80%), observing that increasing the substrate moisture content increased larval feed, wet weight, development time, body size, and body thickness. Likewise, Dortmans et al. [76] commented that the ideal moisture content of food for BSF rearing was in the range of 70 to 80%, whereas, the lower threshold range was between 40 and 55%. Nguyen et al. [37] mentioned that larvae reared on substrates with high-fat content show accelerated development time since they more easily accumulate the necessary amount of fat to survive in the adult stage when they do not feed [77]. However, in our study, the fish-waste substrate had the second-lowest fat content, but the highest moisture and protein content, and had a faster development time compared to the other treatments.

Li et al. [78] mention that BSF larvae contain a high lipid content (26–35%), similar to that found in our study following biodegradation of substrates used for fly-larval rearing (29–46%), and state that this does not compromise fish development, which agrees with Ushakova et al. [79] who mentioned that BSF contain up to 45% of non-water-soluble compounds and recognize it as a biologically active natural resource. Lipid metabolism is an important development process of insects since it performs various functions in the organism such as energy storage, necessary for survival and reproduction in the adult stage [80], which are necessary factors when talking about the mass production of insects. Therefore, adding organic products with high-fat content in BSF-development substrates is an important factor for their production. However, the substrates to be used in BSF rearing, as well as the time of larval collection, must be carefully selected, since the fatty-acid composition of the substrate and the weight of the larvae affect fatty-acid composition in the larvae [81].

5. Conclusions

Results showed that an organic-waste mixture can produce high-quality BSF larvae that have the potential to substitute other sources of protein and lipids in commercial fish feed. The restaurant waste-based substrate extended the larval growth and the biological cycle of the BSFs, while the fish-waste-based substrate induced a lower egg–adult development period than the rest of the treatments. Pre-pupae reared on restaurant waste were higher in biomass, while the fruit-waste substrate produced a significantly lower weight. Regarding protein content, the pre-pupae developed using the restaurant and fish-waste substrates presented the same percentage as the commercial food. An increase in fish biomass was found for fish fed with the fish-waste-based diet. This diet presented the highest weight gain when compared to the restaurant and fruit-waste-based diet. Fish fed with a fish-waste-based diet reached a total length higher than the rest of the treatments. Bromatological analysis showed that the fish-waste-based diet had the highest crude fat and protein content. The nutritional composition of the food obtained from the fly pupae was similar in protein content to commercial food. Our findings indicate that rearing BSF pre-pupae on fish waste has the potential to deliver a high-quality insect resource with the potential for being incorporated into fish feed.

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Data Availability Statement: All the associated data are available in the manuscript.

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References

1. Van Huis, A. Potential of insects as food and feed in assuring food security. *Annu. Rev. Entomol.* **2013**, *58*, 563–583. [[CrossRef](#)] [[PubMed](#)]
2. Hoang, N.H.; Fogarassy, C. Sustainability Evaluation of Municipal Solid Waste Management System for Hanoi (Vietnam)—Why to Choose the ‘Waste-to-Energy’ Concept. *Sustainability* **2020**, *12*, 1085. [[CrossRef](#)]
3. Diener, S.; Solano, N.M.S.; Gutiérrez, F.R.; Zurbrügg, C.; Tockner, K. Biological treatment of municipal organic waste using black soldier fly larvae. *Waste Biomass Valori.* **2011**, *2*, 357–363. [[CrossRef](#)]
4. Smetana, S.; Palanisamy, M.; Mathys, A.; Heinz, V. Sustainability of insect use for feed and food: Life cycle assessment perspective. *J. Clean. Prod.* **2016**, *137*, 741–751. [[CrossRef](#)]
5. Veldkamp, T.; Van Duinkerken, G.; Van Huis, A.; Lakemond, C.M.M.; Ottevanger, E.; Bosch, G.; Van Boekel, T. *Insects as a Sustainable Feed Ingredient in Pig and Poultry Diets: A Feasibility Study (No. 638)*; Wageningen UR Livestock Research: Lelystad, The Netherlands, 2012.
6. Makkar, H.P.; Tran, G.; Heuzé, V.; Ankers, P. State-of-the-art on use of insects as animal feed. *Anim. Feed Sci. Technol.* **2014**, *197*, 1–33. [[CrossRef](#)]
7. Henry, M.; Gasco, L.; Piccolo, G.; Fountoulaki, E. Review on the use of insects in the diet of farmed fish: Past and future. *Anim. Feed Sci. Technol.* **2015**, *203*, 1–22. [[CrossRef](#)]
8. De Marco, M.; Martínez, S.; Hernandez, F.; Madrid, J.; Gai, F.; Rotolo, L.; Kovitvadhi, A. Nutritional value of two insect larval meals (*Tenebrio molitor* and *Hermetia illucens*) for broiler chickens: Apparent nutrient digestibility, apparent ileal amino acid digestibility and apparent metabolizable energy. *Anim. Feed Sci. Technol.* **2015**, *209*, 211–218. [[CrossRef](#)]
9. Dumas, A.; Raggi, T.; Barkhouse, J.; Lewis, E.; Weltzien, E. The oil fraction and partially defatted meal of black soldier fly larvae (*Hermetia illucens*) affect differently growth performance, feed efficiency, nutrient deposition, blood glucose and lipid digestibility of rainbow trout (*Oncorhynchus mykiss*). *Aquaculture* **2018**, *492*, 24–34. [[CrossRef](#)]
10. Bovera, F.; Loponte, R.; Pero, M.E.; Cutrignelli, M.I.; Calabro, S.; Musco, N.; Vassalotti, G.; Panettieri, V.; Lombardi, P.; Piccolo, G.; et al. Laying performance, blood profiles, nutrient digestibility and inner organs traits of hens fed an insect meal from *Hermetia illucens* larvae. *Res. Vet. Sci.* **2018**, *120*, 86–93. [[CrossRef](#)]
11. Moniello, G.; Ariano, A.; Panettieri, V.; Tulli, F.; Olivotto, I.; Messina, M.; Randazzo, B.; Severino, L.; Piccolo, G.; Musco, N.; et al. Intestinal Morphometry, Enzymatic and Microbial Activity in Laying Hens Fed Different Levels of a *Hermetia illucens* Larvae Meal and Toxic Elements Content of the Insect Meal and Diets. *Animals* **2019**, *9*, 86. [[CrossRef](#)]
12. Secci, G.; Addeo, N.F.; Rodriguez, L.F.P.; Bovera, F.; Moniello, G.; Parisi, G. *In vivo* performances, ileal digestibility, and physicochemical characterization of raw and boiled eggs as affected by *Tenebrio molitor* larvae meal at low inclusion rate in laying quail (*Coturnix japonica*) diet. *Poult Sci.* **2021**, *100*, 101487. [[CrossRef](#)] [[PubMed](#)]
13. Norouzi, R.; Manochehri, A.; Zarrin, S. External Ophthalmomyiasis Caused by *Lucilia sericata* (Diptera: Calliphoridae) Larva. *Iran. J. Parasitol.* **2017**, *12*, 466. [[PubMed](#)]
14. Gobbi, P.; Martínez-Sánchez, A.; Rojo, S. The effects of larval diet on adult life-history traits of the black soldier fly, *Hermetia illucens* (Diptera: Stratiomyidae). *Eur. J. Entomol.* **2013**, *110*, 461. [[CrossRef](#)]
15. Oliveira, F.; Doelle, K.; List, R.; O’Reilly, J.R. Assessment of Diptera: Stratiomyidae, genus *Hermetia illucens* (L., 1758) using electron microscopy. *J. Entomol. Zool. Stud.* **2015**, *3*, 147–152.
16. Benítez, A.J.A.; Barba, M.E.; Juárez, F.J. Composition and distribution of aquatic insects in streams of Tabasco, Mexico. *Hidrobiol.* **2016**, *26*, 509–518.

17. Newton, G.L.; Sheppard, D.C.; Watson, D.W.; Burtle, G.J.; Dove, C.R.; Tomberlin, J.K.; Thelen, E.E. The black soldier fly, *Hermetia illucens*, as a manure management/resource recovery tool. In Proceedings of the Symposium on the State of the Science of Animal Manure and Waste Management, San Antonio, TX, USA, 5–7 January 2005.
18. St-Hilaire, S.; Sheppard, C.; Tomberlin, J.K.; Irving, S.; Newton, L.; McGuire, M.A.; Sealey, W. Fly prepupae as a feedstuff for rainbow trout, *Oncorhynchus mykiss*. *J. World Aquac. Soc.* **2007**, *38*, 59–67. [[CrossRef](#)]
19. Food and Agriculture Organization. *The State of World Fisheries and Aquaculture, Opportunities and Challenges*; FAO: Rome, Italy, 2014; 243p. Available online: <http://www.fao.org/3/a-i3720e.pdf> (accessed on 11 September 2020).
20. Giannetto, A.; Oliva, S.; Lanes, C.F.C.; Pedron, F.A.; Savastano, D.; Baviera, C.; Parrino, V.; Lo Paro, G.; Spanò, N.C.; Cappello, T.; et al. *Hermetia illucens* (Diptera: Stratiomyidae) larvae and prepupae: Biomass production, fatty acid profile and expression of key genes involved in lipid metabolism. *J. Biotechnol.* **2020**, *307*, 44–54. [[CrossRef](#)]
21. Kinasih, I.; Suryani, Y.; Paujiah, E.; Ulfa, R.A.; Afiati, S.; Adawiyah, Y.R.; Putra, R.E. Performance of black soldier fly, *Hermetia illucens*, larvae during valorization of organic wastes with changing quality. *IOP Conf. Ser. Earth Environ. Sci.* **2020**, *593*, 012040. [[CrossRef](#)]
22. Wang, S.Y.; Wu, L.; Li, B.; Zhang, D. Reproductive potential and nutritional composition of *Hermetia illucens* (Diptera: Stratiomyidae) prepupae reared on different organic wastes. *J. Econ. Entomol.* **2020**, *113*, 527–537. [[CrossRef](#)]
23. Tanga, C.M.; Waweru, J.W.; Tola, Y.H.; Onyoni, A.A.; Khamis, F.M.; Ekesi, S.; Paredes, J.C. Organic Waste Substrates Induce Important Shifts in Gut Microbiota of Black Soldier Fly (*Hermetia illucens* L.): Coexistence of Conserved, Variable, and Potential Pathogenic Microbes. *Front. Microbiol.* **2021**, *12*, 635881. [[CrossRef](#)]
24. Shumo, M.; Osuga, I.M.; Khamis, F.M.; Tanga, C.M.; Fiaboe, K.K.M.; Subramanian, S.; Ekesi, S.; van Huis, A.; Borgemeister, C. The nutritive value of black soldier fly larvae reared on common organic waste streams in Kenya. *Sci. Rep.* **2019**, *9*, 1–13. [[CrossRef](#)] [[PubMed](#)]
25. Chia, S.Y.; Tanga, C.M.; Khamis, F.M.; Mohamed, S.A.; Salifu, D.; Sevgan, S.; Fiaboe, K.K.M.; Niassy, S.; van Loon, J.J.A.; Dicke, M.; et al. Threshold temperatures and thermal requirements of black soldier fly *Hermetia illucens*: Implications for mass production. *PLoS ONE* **2018**, *13*, e0206097. [[CrossRef](#)] [[PubMed](#)]
26. Verheyen, G.R.; Ooms, T.; Vogels, L.; Vreysen, S.; Bovy, A.; van Miert, S.; Meersman, F. Insects as an alternative source for the production of fats for cosmetics. *J. Cosmet. Sci.* **2018**, *69*, 187–202. [[PubMed](#)]
27. Nagdalian, A.A.; Pushkin, S.V.; Dmitrievich, L.A.; Dmitrievna, T.L.; Rzhepakovsky, I.V.; Andreevich, T.P. Bioconversion of nutrients and biological active substances in model systems chlorella-insect-livestock. *Entomol. Appl. Sci. Lett.* **2018**, *5*, 103–110.
28. Addeo, N.F.; Vozzo, S.; Secci, G.; Mastellone, V.; Piccolo, G.; Lombardi, P.; Parisi, G.; Asiry, K.A.; Attia, Y.A.; Bovera, F. Different Combinations of Butchery and Vegetable Wastes on Growth Performance, Chemical-Nutritional Characteristics and Oxidative Status of Black Soldier Fly Growing Larvae. *Animals* **2021**, *11*, 3515. [[CrossRef](#)]
29. Dzepe, D.; Nana, P.; Kuitche, H.M.; Kimpara, J.M.; Magatsing, O.; Tchuinkam, T.; Djouaka, R. Feeding strategies for small-scale rearing black soldier fly larvae (*Hermetia illucens*) as organic waste recycler. *SN Appl. Sci.* **2021**, *3*, 252. [[CrossRef](#)]
30. Nguyen, T.; Tomberlin, J.K.; Vanlaerhoven, S. Ability of black soldier fly (Diptera: Stratiomyidae) larvae to recycle food waste. *Environ. Entomol.* **2015**, *44*, 406–410. [[CrossRef](#)]
31. Chemello, G.; Zarantonello, M.; Randazzo, B.; Gioacchini, G.; Truzzi, C.; Cardinaletti, G.; Riolo, P.; Olivotto, I. Effects of black soldier fly (*Hermetia illucens*) enriched with *Schizochytrium* sp. on zebrafish (*Danio rerio*) reproductive performances. *Aquaculture* **2022**, *550*, 737853. [[CrossRef](#)]
32. Chia, S.Y.; Tanga, C.M.; Osuga, I.M.; Mohamed, S.A.; Khamis, F.M.; Salifu, D.; Sevgan, S.; Fiaboe, K.; Niassy, S.; van Loon, J.; et al. Effects of waste stream combinations from brewing industry on performance of Black Soldier Fly, *Hermetia illucens* (Diptera: Stratiomyidae). *PeerJ* **2018**, *6*, e5885. [[CrossRef](#)]
33. Scala, A.; Cammack, J.A.; Salvia, R.; Scieuzo, C.; Franco, A.; Bufo, S.A.; Tomberlin, J.K.; Falabella, P. Rearing substrate impacts growth and macronutrient composition of *Hermetia illucens* (L.) (Diptera: Stratiomyidae) larvae produced at an industrial scale. *Sci. Rep.* **2020**, *10*, 19448. [[CrossRef](#)]
34. Martins, C.; Eding, E.H.; Verreth, J. The effect of recirculating aquaculture systems on the concentrations of heavy metals in culture water and tissues of Nile tilapia *Oreochromis niloticus*. *Food Chem.* **2011**, *126*, 1001–1005. [[CrossRef](#)]
35. Devic, E.; Leschen, W.; Murray, F.; Little, D.C. Growth performance, feed utilization and body composition of advanced nursing Nile tilapia (*Oreochromis niloticus*) fed diets containing Black Soldier Fly (*Hermetia illucens*) larvae meal. *Aquac. Nutr.* **2018**, *24*, 416–423. [[CrossRef](#)]
36. Poot-López, G.R.; Gasca-Leyva, E.; Olvera-Novoa, M.A. Nile tilapia production (*Oreochromis niloticus* L.) using tree spinach leaves (*Cnidioscolus chayamansa* McVaugh) as a partial substitute for balanced feed. *Lat. Am. J. Aquat. Res.* **2012**, *40*, 835–846.
37. Nguyen, T.T.; Tomberlin, J.K.; Vanlaerhoven, S. Influence of resources on *Hermetia illucens* (Diptera: Stratiomyidae) larval development. *J. Med. Entomol.* **2013**, *50*, 898–906. [[CrossRef](#)] [[PubMed](#)]
38. Harnden, L.M.; Tomberlin, J.K. Effects of temperature and diet on black soldier fly, *Hermetia illucens* (L.) (Diptera: Stratiomyidae) development. *Forensic Sci. Int.* **2016**, *266*, 109–116. [[CrossRef](#)]
39. Tomberlin, J.K.; Sheppard, D.C.; Joyce, J.A. Selected life-history traits of black soldier flies (Diptera: Stratiomyidae) reared on three artificial diets. *Ann. Entomol. Soc. Am.* **2002**, *95*, 379–386. [[CrossRef](#)]
40. Monentcham, S.E.; Pouomogne, V.; Kestemont, P. Influence of dietary protein levels on growth performance and body composition of African bonytongue fingerlings, *Heterotis niloticus* (Cuvier, 1829). *Aquac. Nutr.* **2010**, *16*, 144–152. [[CrossRef](#)]

41. Sealey, W.M.; Gaylord, T.G.; Barrows, F.T.; Tomberlin, J.K.; McGuire, M.A.; Ross, C.; St-Hilaire, S. Sensory analysis of rainbow trout, *Oncorhynchus mykiss*, fed enriched black soldier fly prepupae, *Hermetia illucens*. *J. World Aquac. Soc.* **2011**, *42*, 34–45. [[CrossRef](#)]
42. Renna, M.; Schiavone, A.; Gai, F.; Dabbou, S.; Lussiana, C.; Malfatto, V.; De Marco, M. Evaluation of the suitability of a partially defatted black soldier fly (*Hermetia illucens* L.) larvae meal as ingredient for rainbow trout (*Oncorhynchus mykiss* Walbaum) diets. *J. Anim. Sci. Biotechnol.* **2017**, *8*, 57. [[CrossRef](#)]
43. Günther, J.; Jiménez-Montealegre, R. Effect of the probiotic *Bacillus subtilis* on the growth and food utilization of tilapia (*Oreochromis niloticus*) and prawn (*Macrobrachium rosenbergii*) under laboratory conditions. *Rev. Biol. Trop.* **2004**, *1*, 937–943.
44. AOAC Association of Official Analytical Chemist. *Official Methods of Analysis of AOAC International*, 18th ed.; Association of Officiating Analytical Chemists International: Gaithersburg, MD, USA, 2005; p. 771.
45. *NMX-F-428-1982 Alimentos-Determinación de Humedad (Método Rápido de la Termobalanza)*; Diario Oficial de la Federación: Mexico; Available online: https://www.dof.gob.mx/nota_detalle_popup.php?codigo=719128 (accessed on 10 November 2021). (in Spanish)
46. Barroso, F.G.; Sánchez-Muros, M.J.; Segura, M.; Morote, E.; Torres, A.; Ramos, R.; Guil, J. Insects as food: Enrichment of larvae of *Hermetia illucens* with omega 3 fatty acids by means of dietary modifications. *J. Food Compos. Anal.* **2017**, *62*, 8–13. [[CrossRef](#)]
47. Lowry, O.H.; Rosebrough, N.J.; Farr, A.L.; Randall, R.J. Protein measurement with the Folin phenol reagent. *J. Biol. Chem.* **1951**, *193*, 265–275. [[CrossRef](#)]
48. NOM-062-ZOO-1999. *Especificaciones Técnicas para la Producción, Cuidado y uso de los Animales de Laboratorio*; Diario Oficial de la Federación: Mexico; pp. 107–167. Available online: <https://administracion.cinvestav.mx/Secretar%C3%ADadePlaneaci%C3%B3n/Coordinaci%C3%B3ndeServiciosdeApoyo/Coordinaci%C3%B3nGeneraldeServiciosExperimentales/UPEAL/NOM-062-ZOO-1999.aspx> (accessed on 10 November 2021). (in Spanish).
49. Schiavone, A.; Cullere, M.; De Marco, M.; Meneguz, M.; Biasato, I.; Bergagna, S.; Dezzutto, D.; Gai, F.; Dabbou, S.; Gasco, L.; et al. Partial or total replacement of soybean oil by black soldier fly larvae (*Hermetia illucens* L.) fat in broiler diets: Effect on growth performances, feed-choice, blood traits, carcass characteristics and meat quality. *Ital. J. Anim. Sci.* **2016**, *16*, 93–100. [[CrossRef](#)]
50. Nijdam, D.; Rood, T.; Westhoek, H. The price of protein: Review of land use and carbon footprints from life cycle assessments of animal food products and their substitutes. *Food Policy* **2012**, *37*, 760–770. [[CrossRef](#)]
51. Samayoa, C.A.; Chen, W.T.; Hwang, S.Y. Survival and development of *Hermetia illucens* (Diptera: Stratiomyidae): A biodegradation agent of organic waste. *J. Econ. Entomol.* **2016**, *109*, 2580–2585. [[CrossRef](#)]
52. Zhou, F.; Tomberlin, J.K.; Zheng, L.; Yu, Z.; Zhang, J. Developmental and waste reduction plasticity of three black soldierfly strains (Diptera: Stratiomyidae) raised on different livestock manures. *J. Med. Entomol.* **2013**, *50*, 1224–1230. [[CrossRef](#)]
53. Oonincx, D.G.; Van Broekhoven, S.; Van Huis, A.; van Loon, J.J. Feed conversion, survival and development, and composition of four insect species on diets composed of food by-products. *PLoS ONE* **2015**, *10*, e0144601. [[CrossRef](#)]
54. Barragan-Fonseca, K.B.; Dicke, M.; van Loon, J.J. Nutritional value of the black soldier fly (*Hermetia illucens* L.) and its suitability as animal feed—a review. *J. Insects Food Feed.* **2017**, *3*, 105–120. [[CrossRef](#)]
55. Caruso, D.; Devic, E.; Subamia, I.W.; Talamond, P.; Baras, E. *Technical Handbook of Domestication and Production of Diptera Black Soldier Fly (BSF) Hermetia Illucens, Stratiomyidae*; Penerbit IPB Press: Kampus IPB Taman Kencana Bogor, 2014; p. 158.
56. Wang, Y.S.; Shelomi, M. Review of black soldier fly (*Hermetia illucens*) as animal feed and human food. *Foods* **2017**, *6*, 91. [[CrossRef](#)]
57. Diener, S.; Zurbrugg, C.; Tockner, K. Conversion of organic material by black soldier fly larvae: Establishing optimal feeding rates. *Waste Manag. Res.* **2009**, *27*, 603–610. [[CrossRef](#)]
58. Xue, M.; Pang, Y.H.; Wang, H.T.; Li, Q.L.; Liu, T.X. Effects of four host plants on biology and food utilization of the cutworm, *Spodoptera litura*. *J. Insect Sci.* **2010**, *10*, 22. [[CrossRef](#)] [[PubMed](#)]
59. Barroso, F.G.; de Haro, C.; Sánchez-Muros, M.-J.; Venegas, E.; Martínez-Sánchez, A.; Pérez-Bañón, C. The potential of various insect species for use as food for fish. *Aquaculture* **2014**, *422*, 193–201. [[CrossRef](#)]
60. Surendra, K.C.; Olivier, R.; Tomberlin, J.K.; Rajesh Jha, R.; Khanalet, S.K. Bioconversion of organic wastes into biodiesel and animal feed via insect farming. *Renew. Energy* **2016**, *98*, 197–202. [[CrossRef](#)]
61. Li, W.; Li, M.; Zheng, L.; Liu, Y.; Zhang, Y.; Yu, Z.; Ma, Z.; Li, Q. Simultaneous utilization of glucose and xylose for lipid accumulation in black soldier fly. *Biotechnol. Biofuels* **2015**, *8*, 1–6. [[CrossRef](#)] [[PubMed](#)]
62. Cammack, J.A.; Tomberlin, J.K. The impact of diet protein and carbohydrate on select life-history traits of the black soldier fly *Hermetia illucens* (L.) (Diptera: Stratiomyidae). *Insects* **2017**, *8*, 56. [[CrossRef](#)]
63. Čičková, H.; Newton, G.L.; Lacy, R.C.; Kozánek, M. The use of fly larvae for organic waste treatment. *Waste Manag.* **2015**, *35*, 68–80. [[CrossRef](#)] [[PubMed](#)]
64. Myers, H.M.; Tomberlin, J.K.; Lambert, B.D.; Kattes, D. Development of black soldier fly (Diptera: Stratiomyidae) larvae fed dairy manure. *Environ. Entomol.* **2014**, *37*, 11–15. [[CrossRef](#)]
65. Purschke, B.; Scheibelberger, R.; Axmann, S.; Adler, A.; Jäger, H. Impact of substrate contamination with mycotoxins, heavy metals and pesticides on the growth performance and composition of black soldier fly larvae (*Hermetia illucens*) for use in the feed and food value chain. *Food Addit. Contam. Part A.* **2017**, *34*, 1410–1420. [[CrossRef](#)]
66. Holmes, L.A.; Vanlaerhoven, S.L.; Tomberlin, J.K. Substrate effects on pupation and adult emergence of *Hermetia illucens* (Diptera: Stratiomyidae). *Environ. Entomol.* **2013**, *42*, 370–374. [[CrossRef](#)]

67. Cheng, J.Y.K.; Chiu, S.L.H.; Lo, I.M.C. Effects of moisture content of food waste on residue separation, larval growth and larval survival in black soldier fly bioconversion. *Waste Manag.* **2017**, *67*, 315–323. [[CrossRef](#)]
68. Palma, L.; Ceballos, S.J.; Johnson, P.C.; Niemeier, D.; Pitesky, M.; Vander Gheynst, J.S. Cultivation of black soldier fly larvae on almond byproducts: Impacts of aeration and moisture on larvae growth and composition. *J. Sci. Food Agric.* **2018**, *98*, 5893–5900. [[CrossRef](#)] [[PubMed](#)]
69. St-Hilaire, S.; Cranfill, K.; McGuire, M.A.; Mosley, E.E.; Tomberlin, J.K.; Newton, L.; Irving, S. Fish offal recycling by the black soldier fly produces a foodstuff high in omega-3 fatty acids. *J. World Aquac. Soc.* **2007**, *38*, 309–313. [[CrossRef](#)]
70. Spranghers, T.; Ottoboni, M.; Klootwijk, C.; Obyn, A.; Deboosere, S.; De Meulenaer, B.; De Smet, S. Nutritional composition of black soldier fly (*Hermetia illucens*) prepupae reared on different organic waste substrates. *J. Sci. Food Agric.* **2017**, *97*, 2594–2600. [[CrossRef](#)] [[PubMed](#)]
71. Newton, G.; Booram, C.; Barker, R.; Hale, O. Dried *Hermetia illucens* larvae meal as a supplement for swine. *J. Anim. Sci.* **1977**, *44*, 395–400. [[CrossRef](#)]
72. El-Saidy, D.M.; Gaber, M.M. Effect of dietary protein levels and feeding rates on growth performance, production traits and body composition of Nile tilapia, *Oreochromis niloticus* (L.) cultured in concrete tanks. *Aquac. Res.* **2005**, *36*, 163–171. [[CrossRef](#)]
73. Chatzifotis, S.; Panagiotidou, M.; Papaioannou, N.; Pavlidis, M.; Nengas, I.; Mylonas, C.C. Effect of dietary lipid levels on growth, feed utilization, body composition and serum metabolites of meagre (*Argyrosomus regius*) juveniles. *Aquaculture* **2010**, *307*, 65–70. [[CrossRef](#)]
74. Kroeckel, S.; Harjes, A.G.; Roth, I.; Katz, H.; Wuertz, S.; Susenbeth, A.; Schulz, C. When a turbot catches a fly: Evaluation of a pre-pupae meal of the Black Soldier Fly (*Hermetia illucens*) as fish meal substitute—Growth performance and chitin degradation in juvenile turbot (*Psetta maxima*). *Aquaculture* **2012**, *364*, 345–352. [[CrossRef](#)]
75. Schiavone, A.; De Marco, M.; Martínez, S.; Dabbou, S.; Renna, M.; Madrid, J.; Gasco, L. Nutritional value of a partially defatted and a highly defatted black soldier fly larvae (*Hermetia illucens* L.) meal for broiler chickens: Apparent nutrient digestibility, apparent metabolizable energy and apparent ileal amino acid digestibility. *J. Anim. Sci. Biotechnol.* **2017**, *8*, 51. [[CrossRef](#)]
76. Dortmans, B.M.A.; Egger, J.; Diener, S.; Zurbrügg, C. *Black Soldier Fly Biowaste Processing—A Step-by-Step Guide*, 2nd ed.; Swiss Federal Institute of Aquatic Science and Technology: Dübendorf, Switzerland, 2021.
77. Coppe, P.A.; Montali, A.; Bruno, D.; Tettamanti, G. Metabolic adjustment of the larval fat body in *Hermetia illucens* to dietary conditions. *J. Asia-Pac. Entomol.* **2017**, *20*, 1307–1313.
78. Li, S.; Ji, H.; Zhang, B.; Zhou, J.; Yu, H. Defatted black soldier fly (*Hermetia illucens*) larvae meal in diets for juvenile Jian carp. (*Cyprinus carpio* var. Jian): Growth performance, antioxidant enzyme activities, digestive enzyme activities, intestine and hepatopancreas histological structure. *Aquac.* **2017**, *477*, 62–70. [[CrossRef](#)]
79. Ushakova, N.A.; Brodskii, E.S.; Kovalenko, A.A.; Bastrakov, A.I.; Kozlova, A.A.; Pavlov, D.S. Characteristics of lipid fractions of larvae of the black soldier fly *Hermetia illucens*. *Dokl. Biochem. Biophys.* **2016**, *468*, 209–212. [[CrossRef](#)] [[PubMed](#)]
80. Kaczmarek, A.; Boguś, M. The metabolism and role of free fatty acids in key physiological processes in insects of medical, veterinary and forensic importance. *PeerJ* **2021**, *9*, e12563. [[CrossRef](#)] [[PubMed](#)]
81. Ewald, N.; Vidakovic, A.; Langeland, M.; Kiessling, A.; Sampels, S.; Lalander, C. Fatty acid composition of black soldier fly larvae (*Hermetia illucens*)—Possibilities and limitations for modification through diet. *Waste Manag.* **2020**, *102*, 40–47. [[CrossRef](#)] [[PubMed](#)]