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Growth, survival, bioconversion performance, nutritional evaluation and palatability of black soldier fly (*Hermetia illucens* L.) larvae meal valorized from vegetable-waste and restaurant-waste for Japanese weatherloach, (*Misgurnus anguillicaudatus*)

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Abstract An evaluation of black soldier fly larvae parameters and its utilization as meal in the palatability test for juvenile loach were determined the species' acceptability to the new feed ingredient valorized from two types of waste substrates [vegetable waste (VW) and restaurant waste (RW)]. A higher survival rate was observed in RW (59.20 ± 4.92 %) than that in VW (17.05 ± 4.54 %). In terms of larval development, the RW had significantly a higher larval live weight (89.70 ± 28.00 mg), longer body length (19.79 ± 2.02 mm), and higher body thickness (4.88±0.65 mm) than those in VW. Conversely, the RW showed significantly higher protein conversion ratio (7.68 ± 0.06 %) compared to the VW (1.97 ± 0.93 %). In terms of moisture content, VW (87.59 ± 0.87 %) was significantly higher than RW (72.76 ± 2.02 %). In contrast, RW showed a significantly higher crude fat content (10.01 ± 0.30 %) compared to that of VW (4.83 ± 0.26 %). Moreover, palatability test revealed no significant difference between the test diets. Thus, the study suggested a prolonged feeding trial to determine the long-term adaptability of the said species to the new feed ingredient.

Keywords: Juvenile loach, Proximate composition, Test diet, Waste reduction

Introduction

The fish and animal feed industry needs a consistent feed input supply to maintain the increasing food production. For aquaculture, the compound feed consumption of 45.41 million tons of production of major fed species was estimated at 52.74 million tons and expected to grow to 58.96 million tons by 2025, which will need an average rate of 7.7% for feed inputs per year to target the estimated feed use of 69.57 million tons (Tacon *et al.*, 2022). Most of the ingredients utilized for aquaculture feeds are sourced mainly from capture

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fisheries for fish meal and fish oil and agricultural crops from plant-based ingredients (Boyd, 2015).

Access to feeds is a significant constraint on the aquaculture and livestock industries, as it is one of the most expensive components in food production. Feed costs typically account for 40-60% of production costs in semi-intensive and intensive aquaculture systems (Alqaisi *et al.,* 2017). The need to cut costs and reduce the carbon footprint of aquaculture feed ingredient production can possibly adapt insect farming for feed production. In 2000s, the incorporation of bugs into aquaculture feeding began wherein, the eight important species used in producing meals to replace fishmeal in aquafeeds are silkworms (*Bombyx mori*), black soldier fly (*Hermetia illucens*), housefly (*Musca domestica*), yellow mealworm (*Tenebrio molitor*), lesser mealworm (*Alphitobius diaperinus*), house cricket (*Acheta domesticus*), banded cricket (*Gryllodes sigillatus*), and Jamaican field cricket (*Gryllus assimilis*) (Alfiko *et al.*, 2022). European Union regulations only allow insect-based feeding to fish and carnivorous fur-producing animals except for livestock and are fed with authorized plant-based materials, vegetable and fruit residues, wheat bran, grass, brewery by-products and hay as feeding substrates (Gasco *et al.*, 2020).

The black soldier fly (BSF) is among the eight important species used in producing aquaculture feeds (Alfiko *et al.*, 2022). This insect belongs to the family Stratiomyidae and is native to America's tropical, subtropical and temperate regions. In Asia, China has been using BSF for waste management wherein an initial investment of CNY 15 million can process thirty tons of organic waste. At the same time, the global market for BSF is projected to reach $3.4 billion by 2030 (Foo, 2021). In the Philippines, FiveDOL Upcycling Corporation has already started an operation of an eco-agricultural start-up processing around 200 tons of waste per day (Sarmiento, 2021).

To date, the value of BSF larvae as a fish meal ingredient has been tested in a variety of fish species, including Japanese seabass (*Lateolabrax japonicus*) (Wang *et al*., 2019), Nile tilapia (*Oreochromis niloticus*) (Agbohessou *et al*., 2021; Were *et al*., 2022; Pérez-Pacheco *et al*., 2022), largemouth bass (*Micropterus salmoides*) (Peng *et al*., 2021; Xu *et al*., 2021; Fischer *et al*., 2022), European seabass (*Dicentrarchus labrax*) (Abdel-Tawwab *et al*., 2020), rainbow trout (*Oncorhynchus mykiss*) (Bolton *et al*., 2021), zebrafish (*Danio rerio*) (Zarantoniello *et al*., 2020; Lanes *et al*., 2021), barramundi (*Lates calcarifer*) (Hender *et al*., 2021), yellowtail (*Seriola quinqueradiata*) (Ido *et al*., 2021), lemon fin barb (Kamarudin *et al*., 2021), catfish (*Clarias gariepinus*) (Were *et al*., 2022), channel catfish (*Ictalurus punctatus*) (Yildirim-Aksoy et al., 2020), and salmonids (Weththasinghe *et al*., 2022).

According to a press release from the Bureau of Fisheries and Aquatic Resources, during World War II, Japanese introduced the fish, also known as dojo, yu-yu, panispis, or pikaw, to the Philippines. The Japanese weather loach (*Misgurnus anguillicaudatus*) has five barbs found around its mouth, a short dorsal fin, and a long body and three distinct characteristics, “missing a thin stripe from the pelvis to the abdomen”, ”odd spots located on the body, dark in color”, and “large spot, dark in color, located on the caudal peduncle base (upper section) and a caudal peduncle featuring some crests (low adipose)”, that feed on insect larvae, snails, worms, ostracods, cladocerans, fish eggs, larvae, snails and worms (Milton *et al*., 2018). The said fish species was once abundant in rice paddies in the Cordillera. However, as a result of commercialization, the species' production has been depleted and has a high market value. Because of its high protein content, the said species is also said to have played an important role in the nutrition of the populace.

Various agencies have prioritized increased loach production in the Cordillera region. The loach was first artificially produced in Mayoyao in 2012. Hatchery's construction was achieved through the assistance provided in 2014 by the Japan International Cooperation Agency and the Japan Oversees Cooperation Volunteer. Mr. Juri Watanabe, a Japanese volunteer, managed the hatchery for five years. Mr. Watanabe is an aquaculture graduate from Kagoshima University and trained selected participants from the Municipal Agriculture Office and benefiting as many as 50 rice terraces farmers in Mayoyao (BFAR-CAR, 2021). Now, the successful rehabilitation of the loach hatchery by the Bureau under the “Bayanihan 2” or “Bayanihan to Recover as One” aids in increasing loach production.

Hence, this study was conducted to evaluate the nutritional value, bioconversion performance and palatability of the BSF larvae meal valorized from two types of wastes, the vegetable-waste and restaurant-waste for Japanese weather loach.

Materials and methods

Substrates for larval development

Two types of waste substrates for BSF larval rearing were used. The first treatment (VW) was composed of taro and cabbages. Meanwhile, RW was composed of rice, pork left-overs and fresh fish gills. Both wastes were sourced out from Baguio City, Philippines. To reduce water content, VW were stored for two-days at room temperature (Addeo *et al*., 2021). Both waste diets were cut into smaller pieces, placed in 6-plastic containers (h=28 cm, C_{mouth}=104 cm,

C_{base}=79 cm), and were offered in *ad libitum*. Moreover, the temperature in each replicate were monitored daily using a digital thermometer (TP01).

Black soldier fly hatchery rearing

Seventeen grams eggs were sourced out from Nueva Ecija, Philippines. These eggs were hatched on separate hatching container (h=28 cm, C_{mouth}=104 cm, C_{base}=79 cm) filled with 2 kg chicken feed mixed with 1.2 L water as the hatching substrate (60% water content) (El-Hack *et al*., 2020). Stale bread was used as a barrier in between the eggs and the hatching substrate to keep it dry until hatching.

Black soldier fly larval rearing & sampling

After hatching, a total of 30,000 5-day old larvae were used. The live weight, body length, body thickness was recorded from the 100 larvae samples from each replicate every 48 h and were returned to their respective containers (Addeo *et al*., 2021, Perez-Pacheco *et al*., 2022). Each container was covered with dark cloth tied with rubber on the lid. The rearing period ended after 14-days. The procedure was conducted at Benguet, Philippines from August 9-22, 2022.

Growth, survival and bioconversion performance

The survival rate (SR), waste reduction (WR), waste reduction efficiency (WRE) and waste conversion efficiency (WCE) were determined following the formulae used by Gold *et al*. (2020), Addeo *et al*. (2021) and Singh *et al*. (2021).

Proximate analyses of black soldier fly larvae

The samples of black soldier fly larvae were analyzed at FCA Laboratory in Baguio City following the standard methods provided by AOAC for crude protein (AOAC 2001.11), moisture content (AOAC 930.15) and ash content (AOAC 942.05). The crude fat content (AOAC 989.05) was analyzed by FAST Laboratories in Quezon City. Waste substrates, residue and frass samples were also sent to FAST Laboratories for moisture content and dry matter determination (vacuum oven drying at 100°C, 25 mmHg for 5-h).

Palatability test diet preparation

Following the procedure by the Eawag Aquatic Research (n.d), 15 g of larvae samples from each replicate were dried using a microwave oven which was powered to 1000 watts (575°C) for five minutes. Periodical stirring was done in intervals until the acceptable yield ranges from 25%-35% (Small-scale drying methods for Black Soldier Fly Larvae, Hu *et al*. 2017). The dried larvae were then grounded into fine powder using a laboratory mortar and pestle reaching a measurement range of 0.54-1.21 mm which can fit the mouth of the 90 pcs loach juveniles.

Moreover, a total of 3-test feed diets: control diet (100% commercial fry mash), VWF diet (60% commercial fry mash+40% BSF larval meal valorized from vegetable-waste), and RWF diet (60% commercial fry mash+40% BSF larval meal valorized from restaurant-waste), were prepared for the palatability test (Barrows, 2017).

Experimental fish and the feeding trial

The initial feeding trial was conducted from August 25-26, 2022 at Mayoyao Loach Hatchery in Ifugao, Philippines, in which 9-cylindrical aquaria (h=30.50 cm, C=53.34 cm, 1-liter capacity) were used. The experiment followed a complete randomized design with 3-treatments and 3-replicates each.

The loach started to feed exogenously 3-days after hatch (DAH) (Wang *et al*., 2009). Ninety pieces of 30-60 DAH loach juveniles ($mean_{weight}$= 20.00±10.00 mg, $mean_{TotalLength}$=2.42±2.40 mm, $mean_{StandardLength}$=1.87±2.22 mm, $mean_{HeadLength}$=0.66±0.70 mm) were randomly distributed in 9-aquaria and were acclimatized for an hour. The method described by Al-souti *et al.* (2019) was used to assess the palatability of the test diet measured by feed consumption (mg test feed/g fish biomass) in 5 minutes. Subsequently, 1 g test feed was introduced into each aquarium for the fish to consume.

After 5 minutes, the loach juveniles were removed from the aquaria while the uneaten feeds were sent to the FCA Laboratory for the moisture content analysis following AOAC 930.15. The results of the moisture analysis in dry basis (DM) were converted into moisture content (wet basis) following a formula [$MC_{wet}=(100MC_{dry})\div(100+M_{dry})$] (Nyquist and Stroshine, 2014). The data obtained were used in determining the amount of the unconsumed diets from each replicate. Furthermore, the two-day feeding trial was done twice a day (8:00-9:00, 16:00-17:00).

Statistical analyses

The mean values for the survival, growth and bioconversion performance, and proximate composition of BSF larvae were subjected to Independent Samples T-test for the significant differences (P<0.01) between the two treatments. On the otherhand, palatability test diets were subjected to ANOVA to test for the significant differences (P<0.01) among the treatment means. Further, the HSD test was used to assess the difference between the treatment means. Analyses were performed using STAR Software for parametric tests and Jamovi (2.3.24.0) for non-parametric tests.

Results

Waste-substrate composition during and after the rearing trial

The amount of utilized wastes for VW was 2.82 kg and 3.91 kg for RW group. The waste composition, moisture content and the temperature range of the substrates used in the rearing trial is shown in Table 1. The mean moisture content of the waste used for VW (94.80%) was significantly higher compared to the RW (83.77%) (P < 0.01). The mean substrate temperatures at different times of the day for RWwere comparable to those in VW.

Table 1. Waste composition, moisture content and temperature of the waste substrates utilized (in *ad libitum*) during the 14-day larval rearing of BSF larvae

Parameter	Treatment	
	VW	RW
Waste composition, %	CC – 35.00 ± 0.00 TS – 27.00 ± 0.00 TL – 12.00 ± 0.00 C – 26.00 ± 0.00	RLO – 67.00 ± 0.00 CPLO – 5.00 ± 0.00 FFG – 28.00 ± 0.00
Moisture content, %	94.80 ± 0.98 ^{a}	83.77 ± 0.81 ^{b}
Temperature, °C		
08:00		
Temp. Range	20.90 ± 0.65 – 24.80 ± 0.02	20.70 ± 0.45 – 24.40 ± 0.20
Ave. temp.	22.30 ± 0.09 ^{a}	22.10 ± 0.75 ^{a}
12:00		
Temp. Range	20.90 ± 0.26 – 29.20 ± 0.35	21.20 ± 0.10 – 28.50 ± 0.25
Ave. temp.	24.90 ± 2.41 ^{a}	24.70 ± 2.32 ^{a}
18:00		
Temp. Range	20.50 ± 0.15 – 25.80 ± 0.25	20.80 ± 0.06 – 27.40 ± 0.46
Ave. temp.	23.00 ± 1.24 ^{a}	23.50 ± 1.54 ^{a}

*Means in a row superscripted with different letters are highly significant at P<0.01

CC: Chinese cabbage; TS: taro stalks; TL: taro leaves; C: cabbage; RLO: rice left-overs; CPLO: cooked pork left overs; FFG: fresh fish gills

Development of black soldier fly larvae

Based on the visual observation, 20% of the BSF larvae were grown into pre-pupae in RW whereas 10% was observed for VW as estimated after the 14-day larval rearing period. The average live weight, body length, and body thickness measured from 100 larvae per replicate every two days throughout the rearing trial are reported in Tables 2, 3 and 4, respectively. At the end of the rearing trial, T-test revealed that the mean larval live weight, mean body length and mean body thickness of RW group were significantly different from those in VW group ($P < 0.01$). The RW shows a higher mean larval live weight (Table 2), longer mean body length (Table 3) and higher mean body thickness (Table 4) than those in VW group.

Table 2. Mean (±SD) live weight (mg) of black soldier fly from 5 to 19 days of age

Age	Treatment	
	VW	RW
5 d	6.30 ± 3.40	5.60 ± 2.40
7 d	7.40 ± 3.10 ^{a}	6.00 ± 2.90 ^{b}
9 d	8.13 ± 3.50 ^{a}	7.60 ± 5.00 ^{b}
11 d	1.14 ± 9.60 ^{a}	1.95 ± 1.04 ^{b}
13 d	1.44 ± 6.50 ^{a}	2.85 ± 1.28 ^{b}
15 d	2.55 ± 1.57 ^{a}	3.42 ± 2.05 ^{b}
17 d	2.68 ± 1.64 ^{a}	6.60 ± 3.39 ^{b}
19 d	4.18 ± 2.06 ^{a}	8.97 ± 2.80 ^{b}

*Means in a row superscripted with different letters are highly significant at P<0.01

Table 3. Mean (±SD) body length (mm) of black soldier fly from 5 to 19 days of age

Age	Treatment	
	VW	RW
5 d	6.21 ± 1.12	6.14 ± 1.08
7 d	6.97 ± 1.27 ^{a}	6.42 ± 0.96 ^{b}
9 d	7.30 ± 1.19 ^{a}	7.03 ± 1.40 ^{b}
11 d	7.66 ± 1.65 ^{a}	9.91 ± 2.03 ^{b}
13 d	8.93 ± 1.50 ^{a}	11.68 ± 1.73 ^{b}
15 d	10.49 ± 2.36 ^{a}	12.05 ± 2.36 ^{b}
17 d	11.47 ± 1.48 ^{a}	15.67 ± 2.47 ^{b}
19 d	14.47 ± 2.09 ^{a}	19.79 ± 2.02 ^{b}

*Means in a row superscripted with different letters are highly significant at P<0.01

Table 4. Mean (±SD) body thickness (mm) of black soldier fly from 5 to 19 days of age

Age	Treatment	
	VW	RW
5 d	1.90 ± 0.45	1.84 ± 0.48
7 d	2.32 ± 0.52 ^{a}	1.87 ± 0.36 ^{b}
9 d	2.42 ± 0.58 ^{a}	2.15 ± 0.44 ^{b}
11 d	2.62 ± 0.81 ^{a}	2.78 ± 1.06 ^{b}
13 d	2.81 ± 0.79 ^{a}	3.07 ± 0.59 ^{b}
15 d	3.25 ± 1.14 ^{a}	3.64 ± 1.19 ^{b}
17 d	3.35 ± 0.63 ^{a}	4.31 ± 0.79 ^{b}
19 d	3.79 ± 0.80 ^{a}	4.88 ± 0.65 ^{b}

*Means in a row superscripted with different letters are highly significant at *P*<0.01

Survival and bioconversion performance

The survival rate and bioconversion performance of the BSF larvae during the rearing trial are summarized in Table 5. The RW group significantly shows a higher mean survival rate, mean growth rate, mean waste conversion efficiency, and mean protein conversion ratio than those in RW group (P < 0.01). However, the mean waste reduction rate and the mean waste reduction efficiency are comparable in both treatments.

Table 5. Bioconversion performance of BSF larvae during the 14-day rearing trial

Parameter	Treatment	
	VW	RW
Survival rate, %	17.05 ± 4.54 ^{a}	59.20 ± 4.92 ^{b}
Growth rate, % (Wet weight)	2.44 ± 0.74 ^{a}	39.70 ± 3.41 ^{b}
Waste reduction, % DM	19.50 ± 13.51 ^{a}	10.98 ± 1.74 ^{a}
Waste reduction efficiency, % DM	73.15 ± 23.42 ^{a}	87.65 ± 2.19 ^{a}
Waste conversion efficiency, % DM	1.30 ± 1.57 ^{a}	27.17 ± 3.42 ^{b}
Protein conversion Ratio, % DM	1.97 ± 0.93 ^{a}	7.68 ± 0.06 ^{b}

*Means in a row superscripted with different letters are highly significant at *P*<0.01

Proximate composition

The mean moisture content, crude ash, crude protein and fat content of the 5-day old (5 DOL) BSF used before the larval rearing trial were 78.17%, 2.24%, 12.69% and 2.14%, respectively. At the end of the larval growing period of 14 days, RW group showed a significantly higher mean crude fat content compared to the RW group (P < 0.01) (Table 6). On the other hand, the VW group has comparable mean moisture content, mean crude ash, and mean crude protein content to those in RW.

Table 6. Black soldier fly proximate composition after the 14-larval rearing trial

Parameter	Treatment	
	VW	RW
Moisture content, %	87.59 ± 0.87 [a]	72.76 ± 2.02 [a]
Crude ash, %	2.54 ± 0.19 [a]	2.21 ± 0.11 [a]
Crude protein, %	9.09 ± 2.14 [a]	11.89 ± 0.11 [a]
Crude fat, %	4.83 ± 0.26 [a]	10.01 ± 1.30 [b]

*Means in a row superscripted with different letters are highly significant at P<0.01

Palatability test

The test diet consumption (mg/g per fish biomass) ranged from 3.00% (0.70 ± 0.13 mg) - 15.00% (2.41 ± 1.76 mg) between replicates. ANOVA revealed the there is no significant difference between the amount of the test diet (1.77 ± 0.74 mg / 7.21%, 1.46 ± 0.75 mg / 5.96%, 1.96 ± 0.93 mg / 8%) provided to the juvenile loach (W= 0.02 ± 0.01 g, TL= 2.42 ± 2.40 mm, SL= 1.87 ± 2.22 mm, and HL= 0.66 ± 0.7 mm) after the two-day initial feeding trial (Table 7).

Table 7. Test diet consumption (mg/g fish biomass, %) of juvenile loach (30-60 DAH) during the 5-minute palatability test in a two-day initial feeding trial

	Control	Treatments	
		T1	T2
mg	1.77 ± 0.75	1.46 ± 0.46 [a]	1.96 ± 0.93 [a]
%	7.21	5.96	8.00

*Means in a row superscripted with different letters are highly significant at P<0.01

Discussion

The moisture composition of both waste substrate groups exceeded the ideal range (70-80%) described by Dortmans *et al*. (2017). The vegetable inputs used in the study are known to have high moisture contents (Adelanwa and Medugu, 2015, Chun *et al*., 2016, Zhang *et al*., 2021, Netam *et al.*, 2022) wherein VW group had a significantly higher moisture content (94.80 ± 0.98%). Additionally, the two-day storage period was insufficient to reduce the water content of the vegetable surplus before its utilization as waste substrate for BSF larval rearing. On the other hand, the average temperature readings between the two treatments were all comparable based from the specific time of the day. It was observed that the temperature ranges of both treatments during the 08:00 h (VW = 20.90 ± 0.66 – 24.80 ± 0.02 °C, RW = 20.70 ± 0.45 – 24.40 ± 0.20 °C), 12:00 h (VW = 20.90 ± 0.26 – 29.20 ± 0.35 °C, RW = 21.20 ± 0.10 – 28.50 ± 0.25 °C) and 18:00 h (VW = 20.50 ± 0.15 – 25.80 ± 0.25 °C, RW = 20.80 ± 0.058 – 27.40 ± 0.46 °C) were ≤ 24 – 30°C which are the ideal temperature range in rearing BSF larvae (Dortmans *et al.*, 2017). Larval stage can be prolonged for approximately four months in low temperature conditions (Salam *et al.*, 2022). Larvae reared at 27.6 °C and 32.2 °C are ≈30% heavier than larvae reared at 24.9 °C (Harnden and Tomberlin, 2016).

Furthermore, low survival rates were observed in both groups (VW = 17.05±4.54%; RW = 59.20±4.92%) but the mean survival rate in RW group was significantly higher ($P < 0.01$) than that in the VW group. In the same manner, Isibika *et al*. (2021) reported a survival rate of more than 50% for BSF larvae fed with different substrates containing 25% fish waste but with varying banana peel and orange peel waste compositions. The moisture contents of the waste substrates (VW = 94.80%, RW = 83.77%) which had exceeded the ideal range (70 - 80%) described by Dortmans *et al.* (2017), affected the survivability and the growth rate of the two groups. Lalander *et al*. (2020) found that increasing the water content of the substrate reduced the survival rate of BSF larvae, from 97.20% survival in 76.00% water to 19.30% survival in 97.50% water. In the present study, mortalities were observed throughout the larval rearing period, revealing low larval harvest which resulted to low growth rate of both groups. However, the RW was significantly higher in growth rate ($P < 0.01$) than VW. Also, in terms of larval development, the RW showed a significantly higher larval live weight (89.70 ± 28.00 mg), longer body length (19.79 ± 2.02 mm) and higher body thickness (4.88 ± 0.65) ($P < 0.01$) than the VW group. Conversely, studies with growing periods of 15 – 40 days had reached larval weight of 150 – 320 mg (wet basis) (Liu *et al.*, 2017; Barragán-Fonseca *et al.*, 2018; Lalander *et al*. 2019).

The nutritional profile of vegetable-waste used must have impacted the BSF larval development in VW group. The taro foliage (leaves+stalk) (*Colocasia esculenta*) has high water, carbohydrate and fiber contents (Rashmi *et al*., 2018). Concentrations of neutral detergent fiber in cabbage and Chinese cabbage were reported to be 22.31% and 28.83% respectively (Song *et al.*, 2020). High cellulose content cannot be directly digested by BSF larvae (Deng *et al.*, 2022). The higher larval development for RW group can be attributed to the lower moisture content (83.77%) of the restaurant-waste substrate which is 3.00% higher than the ideal range. Cammack and Tomberlin (2018) reported that for BSF larvae reared on the diet, containing 21.00% protein and 21.00% carbohydrate, and hydrated to 70.00% moisture, developed the fastest and had the greatest survival (range 32.25 – 38.38 days and 57.00 – 62.00%, respectively).

The waste reduction efficiency of BSF larvae (n = 5000) was in the range, 46.26% - 90.07% between the number of replicates/observations (6) from the two groups. The maximum reduction efficiency was observed for RWT2 (87.65

± 2.19%) compared to VW (73.15 ± 23.42%). The result is consistent to the waste reduction rate (VW = 19.50 ± 13.51%, RW = 10.98 ± 1.74%) provided that the RW group consumed more waste than the VW group. Applying T-test, it revealed that the waste reduction rate and the waste reduction efficiency in both treatments groups were comparable. The present study shows a higher reduction rate compared to 48.41% (DM) reduction of maize straw (Gao *et al.*, 2019) and 48.70% - 56.60% (DM) reduction of dairy manure (Rehman *et al.*, 2017, 2019).

The waste conversion efficiency rate obtained from RW (27.17±3.41%) was significantly higher ($P < 0.05$) compared to that of VW (1.30 ± 1.57%). Taking into account, the low survival rates and low growth rates (due to high moisture content) observed from the two groups (VW = 17.05%, 2.44%; RW = 59.20%, 39.70%) have influenced the total larval weight gained affecting the low conversion of waste into larval biomass. In similar pattern, the RW group significantly ($P < 0.01$) showed a higher protein conversion ratio (7.68±0.06%) compared to that of VW (1.97 ± 0.93%). This result can be attributed to a higher total restaurant-waste consumed (2.70 kg) by RW group than by VW (1.88 kg) group.

In terms of larval moisture content, the VW group accumulated a significantly ($P<0.01$) higher moisture content (87.59 ± 0.87%) compared to the RW group (72.76 ± 2.02%). Before the 14-day larval rearing period, the moisture content of 5 DOL was 78.17%. The increase in the moisture content can be attributed from the high moisture content of the waste substrate diets (VW = 94.80%; RW= 83.77%) provided. Results on the crude ash (VW = 2.54 ± 0.19%; RW = 2.21 ± 0.10%) and crude protein contents (VW = 9.09 ± 2.14%; RW = 11.89 ± 0.11%) for both groups are comparable. The waste substrate diet for VW was composed of 39% taro, 35% Chinese cabbage and 12% cabbage. On the otherhand, the waste substrate diet for RW was composed of 67.00% cooked rice left-overs, 5.00% cooked pork meat left-overs, and 28.00% fresh fish gills. Unfortunately, the protein contents of the two waste-substrates were not determined. In a study by Danieli *et al.* (2019), diet formulation with increased crude protein content did not yield any practical advantage in terms of the larval growth, yield, composition, or fatty acid profile of BSF larvae.

Furthermore, the RW showed a significantly ($P<0.01$) higher crude fat content (10.01 ± 1.30%) compared to the VW (4.83 ± 0.26%). The higher crude fat from the RW group is possibly due to the consumption of fish gills and pork leftovers compared to the lone vegetable rations provided for RW group.

From the study of Wang *et al.* (2009), co-feeding dry feed and live feed increased the acceptance of formulated feed after 30 DAH. Thus, in this study, formulated feed (commercial feed/fry mash, particle size = 0.40 mm) was used in comparison to the BSF meal for the juvenile loach. After the two-day initial feeding trial, the result for the test diet consumption (mg/g per fish biomass) showed no significant difference from the commercial diet. Thus, it can be considered a pass (Barrows, 2017). Taking into account that the feeding rate ranged from 3.00% - 15.00% between replicates, the VW group (1.96 ± 0.93 mg, 8.00%) is higher and is close to the daily feeding rates (12.80%, 5.80%) obtained by Wang *et al.* (2008), using juvenile loach (30 DAH, 40 DAH) when fed with *Daphnia*.

To our knowledge, this is the first study that evaluated the palatability of BSF, *H. illucens* larvae meal valorized from kitchen waste and food waste for Japanese weatherloach, *M. anguillicaudatus* at juvenile stage. Based from the palatability test, BSF larvae meal valorized from vegetable-waste composed of 39% taro, *C. esculenta*, 35% Chinese cabbage, *B. rapa subsp. pekinensis*, and 12% cabbage *B. oleracea Linn. var. capitate*, is the most suitable for juvenile loach with the highest feeding rate of 8% recorded. The nutritional profile of BSF larvae fed with the mentioned vegetable rations have the same crude protein and ash content with the BSF larvae group fed with restaurant-waste composed of 67% cooked rice left-overs, 5% cooked pork meat left-overs, and 28% fresh fish gills, but with different amounts of crude fat and moisture content. Furthermore, the moisture contents of both vegetable-waste (94.80%) and restaurant-waste (83.77%) must be lowered to possibly upturn the low survival (T1= 17.05±4.54%; T2= 59.20±4.92%) and growth rates (T1= 2.44±0.74%; T2= 39.70±3.41%) of the BSF larvae affecting its bioconversion performance. It is then suggested that an application of pre-treatment techniques for each waste substrate classification prior to BSF larval rearing should be explored to improve larval performances.

Likewise, since mixed-aged of 30-60 DAH loach juveniles were used, the present study suggests a longer rearing trial to qualify the BSF meal as an effective loach co-feed component for juvenile loach diet to determine the long-term adaptability of the said species to the new ingredients.

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References

Abd El-Hack, M. E., Shafi, M. E., Alghamdi, W. Y., Abdelnour, S. A., Shehata, A. M., Noreldin, A. E., Ashour, E. A., Swelum, A. A., Al-Sagan, A. A., Alkhateeb, M., Taha, A. E., Abdel-Moneim, A.-M. E., Tufarelli, V. and Ragni, M. (2020). Black soldier fly (*Hermetia illucens*) meal as a promising feed ingredient for poultry: A comprehensive review. Agriculture, 10:339.

Abdel-Tawwab, M., Khalil, R. H., Metwally, A. A., Shakweer, M. S., Khallaf, M. A. and Abdel-Latif, H. M. R. (2020). Effects of black soldier fly (*Hermetia illucens* L.) larvae meal on growth performance, organs-somatic indices, body composition, and hemato-biochemical variables of European sea bass, *Dicentrarchus labrax*. Aquaculture, 522: 735136.

Addeo, N. F., Vozzo, S., Secci, G., Mastellone, V., Piccolo, G., Lombardi, P., Parisi, G., Asiry, K. A., Attia, Y. A. and Bovera, F. (2021). Different combinations of butchery and vegetable wastes on growth performance, chemical-nutritional characteristics and oxidative status of black soldier fly growing larvae. Animals, 11:3515.

Adelanwa, E. B. and Medugu J. M. (2015). Variation in the nutrient composition of red and green cabbage (*Brassica oleracea*) with respect to age at harvest. Journal of Applied Agricultural Research, 7:183-189.

Agbohessou, P. S., Mandiki, S. N. M., Gougbédji, A., Megido, R. C., Hossain, Md. S., De Jaeger, P., Larondelle, Y., Francis, F., Lalèyè, P. A. and Kestemont, P. (2021). Total replacement of fish meal by enriched-fatty acid *Hermetia illucens* meal did not substantially affect growth parameters or innate immune status and improved whole body biochemical quality of Nile tilapia juveniles. Aquaculture Nutrition, 27:880-896.

Alfiko, Y., Xie, D., Astuti, R. T., Wong, J. and Wang, L. (2022). Insects as a feed ingredient for fish culture: Status and trends. Aquaculture and Fisheries, 7:166-178.

Alqaisi, O., Ndambi, O. A. and Williams, R. B. (2017). Time series livestock diet optimization: Cost-effective broiler feed substitution using the commodity price spread approach. Agricultural and Food Economics, 5:25.

Al-Souti, A., Gallardo, W., Claereboudt, M. and Mahgoub, O. (2019). Attractability and palatability of formulated diets incorporated with chicken feather and algal meals for juvenile gilthead seabream, *Sparus aurata*. Aquaculture Reports, 14:100199.

Barragan-Fonseca, K. B., Dicke, M. and Van Loon, J. J. A. (2018). Influence of larval density and dietary nutrient concentration on performance, body protein, and fat contents of black soldier fly larvae (*Hermetia illucens*). Entomologia Experimentalis et Applicata, 166:761-770.

Barrows, F. T. (2017). Palatability. Retried from https://f3fin.org/protocols/Palatability.pdf

Bolton, C. M., Muller, N., Hyland, J., Johnson, M. P., Souza Valente, C., Davies, S. J. and Wan, A. H. L. (2021). Black soldier fly larval meal with exogenous protease in diets for rainbow trout (*Oncorhynchus mykiss*) production meeting consumer quality. Journal of Agriculture and Food Research, 6:100232.

Boyd, C. E. (2015). Overview of aquaculture feeds. In: Davis DA ed. Feed and feeding practices in aquaculture, Cambridge, Woodhead Publishing, pp.3-25.

Bureau of Fisheries and Aquatic Resources (2021). Project proposal: Construction of the regional/rice terraces dojo (yu-yu, weather loach) hatchery & training center, Mayoyao Ifugao.

Cammack, J. and Tomberlin, J. (2017). The impact of diet protein and carbohydrate on select life-history traits of the black soldier fly *Hermetia illucens* (L.) (Diptera: Stratiomyidae). Insects, 8:56.

Chun, J., Kim, N., Seo, M., Jin, M., Park, S. U., Valan Arasu, M., Kim, S. and Al-Dhabi, N. (2016). Molecular characterization of glucosinolates and carotenoid biosynthetic genes in Chinese cabbage (*Brassica rapa* L. ssp. *Pekinensis*). Saudi Journal of Biological Sciences, 25:71-82.

Danieli, P. P., Lussiana, C., Gasco, L., Amici, A. and Ronchi, B. (2019). The effects of diet formulation on the yield, proximate composition, and fatty acid profile of the black soldier fly (*Hermetia illucens* L.) prepupae intended for animal feed. Animals, 9:178.

Deng, B., Zhu, J., Wang, G., Xu, C., Zhang, X., Wang, P. and Yuan, Q. (2022). Effects of three major nutrient contents, compost thickness and treatment time on larval weight, process performance and residue component in black soldier fly larvae (*Hermetia illucens*) composting. Journal of Environmental Management, 307:114610.

Dortmans B. M. A., Diener S., Verstappen B. M. and Zurbrügg C. (2017). Black soldier fly biowaste processing - a step-by-step guide. Eawag: Swiss Federal Institute of Aquatic Science and Technology, Dübendorf, Switzerland, pp.7-8.

Fischer, H., Romano, N., Renukdas, N., Kumar, V. and Sinha, A. K. (2022). Comparing black soldier fly (*Hermetia illucens*) larvae versus prepupae in the diets of largemouth bass, *Micropterus salmoides*: Effects on their growth, biochemical composition, histopathology, and gene expression. Aquaculture, 546:737323.

Foo, Y. L. (2021). A primer on the black soldier fly market in Asia. Mana Impact.

Gao, Z., Wang, W., Lu, X., Zhu, F., Liu, W., Wang, X. and Lei, C. (2019). Bioconversion performance and life table of black soldier fly (*Hermetia illucens*) on fermented maize straw. Journal of Cleaner Production, 230:974-980.

Gasco, L., Biancarosa, I. and Liland, N. S. (2020). From waste to feed: A review of recent knowledge on insects as producers of protein and fat for animal feeds. Current Opinion in Green and Sustainable Chemistry, 23:67-79.

Gold, M., Cassar, C. M., Zurbrügg, C., Kreuzer, M., Boulos, S., Diener, S. and Mathys, A. (2020). Biowaste treatment with black soldier fly larvae: Increasing performance through the formulation of biowastes based on protein and carbohydrates. Waste Management, 102:319-329.

Harnden, L. M. and Tomberlin, J. K. (2016). Effects of temperature and diet on black soldier fly, *Hermetia illucens* (L.) (Diptera: Stratiomyidae), development. Forensic Science International, 266:109-116.

Hender, A., Siddik, M., Howieson, J. and Fotedar, R. (2021). Black soldier fly, *Hermetia illucens* as an alternative to fishmeal protein and fish oil: Impact on growth, immune response, mucosal barrier status, and flesh quality of juvenile barramundi, *Lates calcarifer* (Bloch, 1790). Biology, 10:505.

Ido, A., Ali, M.-F.-Z., Takahashi, T., Miura, C. and Miura, T. (2021). Growth of yellowtail (*Seriola quinqueradiata*) fed on a diet including partially or completely defatted black soldier fly (*Hermetia illucens*) larvae meal. Insects, 12:722.

Isibika, A., Vinnerås, B., Kibazohi, O., Zurbrügg, C. and Lalander, C. (2021). Co-composting of banana peel and orange peel waste with fish waste to improve conversion by black soldier fly (*Hermetia illucens* (L.), Diptera: Stratiomyidae) larvae. Journal of Cleaner Production, 318:128570.

Kamarudin, M. S., Rosle, S. and Md Yasin, I. S. (2021). Performance of defatted black soldier fly pre-pupae meal as fishmeal replacement in the diet of lemon fin barb hybrid fingerlings. Aquaculture Reports, 21:100775.

Lalander, C., Diener, S., Zurbrügg, C. and Vinnerås, B. (2019). Effects of feedstock on larval development and process efficiency in waste treatment with black soldier fly (*Hermetia illucens*). Journal of Cleaner Production, 208:211-219.

Lalander, C., Ermolaev, E., Wiklicky, V. and Vinnerås, B. (2020). Process efficiency and ventilation requirement in black soldier fly larvae composting of substrates with high water content. Science of the Total Environment, 729:138968.

Lanes, C. F. C., Pedron, F. A., Bergamin, G. T., Bitencourt, A. L., Dorneles, B. E. R., Villanova, J. C. V., Dias, K. C., Riolo, K., Oliva, S., Savastano, D. and Giannetto, A. (2021). Black soldier fly (*Hermetia illucens*) larvae and prepupae defatted meals in diets for zebrafish (*Danio rerio*). Animals, 11:720.

Liu, X., Chen, X., Wang, H., Yang, Q., Li, W., Cai, M., Li, Q., Mazza, L., Zhang, J., Yu, Z. and Zheng, L. (2017). Dynamic changes of nutrient composition throughout the entire life cycle of black soldier fly. PLOS ONE, 12:0182601.

Milton, J., Paray, B. A. and Rather, I. A. (2018). A review on the biology and physiology of loach *Misgurnus anguillicaudatus* in China. Indian Journal of Geo Marine Sciences, 47:759-765.

Netam, U., Thakur, P., Nair Singh, B. and Kar, S. (2022). Morphological characterization of taro [*Colocasia esculenta var. antiquorum* (L.) Schott.]. Genotypes, 11:2070-2076.

Nyquist, C. E. and Stroshine R. L. (2014). Relationship between wet basis and dry basis moisture content.

Peng, K., Mo, W., Xiao, H., Wang, G. and Huang, Y. (2021). Effects of black soldier fly pulp on growth performance, histomorphology and lipid metabolism gene expression of *Micropterus salmoides*. Aquaculture Reports, 20:100737.

Pérez-Pacheco, R., Hinojosa-Garro, D., Ruíz-Ortíz, F., Camacho-Chab, J., Ortega-Morales, B., Alonso-Hernández, N., Fonseca-Muñoz, A., Landero-Valenzuela, N., Loeza-Concha, H., Diego-Nava, F., Arroyo-Balán, F. and Granados-Echegoyen, C. (2022). 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). Insects, 13:326.

Rashmi, D. R., Anitha, B., Sahair, R. A., Raghu, N., Gopenath, T. S., Chandrashekrappa, G. K. and Basalingappa, K. M. (2018). An overview of taro (*Colocasia esculenta*): A review. Academia Journal of Agricultural Research, 6:346-353.

Rehman, K. U., Rehman, A., Cai, M., Zheng, L., Xiao, X., Somroo, A. A., Wang, H., Li, W., Yu, Z. and Zhang, J. (2017). Conversion of mixtures of dairy manure and soybean curd residue by black soldier fly larvae (*Hermetia illucens* L.). Journal of Cleaner Production, 154:366-373.

Rehman, K. U., Ur Rehman, R., Somroo, A. A., Cai, M., Zheng, L., Xiao, X., Ur Rehman, A., Rehman, A., Tomberlin, J. K., Yu, Z. and Zhang, J. (2019). Enhanced bioconversion of dairy and chicken manure by the interaction of exogenous bacteria and black soldier fly larvae. Journal of Environmental Management, 237:75-83.

Salam, M., Shahzadi, A., Alam, F., Nabi, G., Shi, D., Ullah, W., Ammara, S., Ali, N. and Bilal, M. (2022). Effect of different environmental conditions on the growth and development of black soldier fly larvae and its utilization in solid waste management and pollution mitigation. Environmental Technology and Innovation, 28:102649.

Sarmiento, B. S. (2021). A startup deploys black soldier flies in the Philippines' war on waste. Mongabay.

Singh, A., Srikanth, B. H. and Kumari, K. (2021). Determining the black soldier fly larvae performance for plant-based food waste reduction and the effect on biomass yield. Waste Management, 130:147-154.

Song, K. H., Woo, J. S., Kim, J. R., Ryu, G. L., Baek, Y. C., Oh, Y. K., Kwak, W. S. and Park, K. K. (2020). Nutritional value and in situ degradability of fruit-vegetable byproducts and their feeding effects on performance of growing Hanwoo steers. Asian-Australasian Journal of Animal Sciences, 33:973-980.

Tacon, A. G. J., Metian, M. and McNevin, A. A. (2022). Future feeds: Suggested guidelines for sustainable development. Reviews in Fisheries Science & Aquaculture, 30:135-142.

Wang, G., Peng, K., Hu, J., Yi, C., Chen, X., Wu, H. and Huang, Y. (2019). Evaluation of defatted black soldier fly (*Hermetia illucens* L.) larvae meal as an alternative protein ingredient for juvenile Japanese seabass (*Lateolabrax japonicus*) diets. Aquaculture, 507:144-154.

Wang, Y., Hu, M., Wang, W. and Cao, L. (2009). Effects on growth and survival of loach (*Misgurnus anguillicaudatus*) larvae when co-fed on live and microparticle diets. Aquaculture Research, 40:385-394.

Wang, Y., Hu, M., Wang, W., Cao, L., Yang, Y., Lü, B. and Yao, R. (2008). Transpositional feeding rhythm of loach *Misgurnus anguillicaudatus* from larvae to juveniles and its ontogenesis under artificial rearing conditions. Aquaculture International, 16:539-549.

Were, G. J., Irungu, F. G., Ngoda, P. N., Affognon, H., Ekesi, S., Nakimbugwe, D., Fiaboe, K. K. M. and Mutungi, C. M. (2022). Nutritional and microbial quality of extruded fish feeds containing black soldier fly (*Hermetia illucens* L.) larvae meal as a replacement for fish meal for Nile Tilapia (*Oreochromis niloticus*) and African sharptooth catfish (*Clarius gariepinus*). Journal of Applied Aquaculture, 34:1036-1052.

Weththasinghe, P., Hansen, J. Ø., Mydland, L. T. and Øverland, M. (2022). A systematic meta-analysis based review on black soldier fly (*Hermetia illucens*) as a novel protein source for salmonids. Reviews in Aquaculture, 14:938-956.

Xu, F. M., Hou, S. W., Wang, G. X., Gong, J. Y., Zhou, L., Huang, Y. H., Huang, X. D. and Liu, L. (2021). Effects of zymolytic black soldier fly (*Hermetia illucens*) pulp as dietary supplementation in largemouth bass (*Micropterus salmoides*). Aquaculture Reports, 21: 100823.

Yildirim-Aksoy, M., Eljack, R. and Beck, B. H. (2020). Nutritional value of frass from black soldier fly larvae, *Hermetia illucens*, in a channel catfish, *Ictalurus punctatus*, diet. Aquaculture Nutrition, 26:812-819.

Zarantoniello, M., Randazzo, B., Gioacchini, G., Truzzi, C., Giorgini, E., Riolo, P., Gioia, G., Bertolucci, C., Osimani, A., Cardinaletti, G., Lucon-Xiccato, T., Milanović, V., Annibaldi, A., Tulli, F., Notarstefano, V., Ruschioni, S., Clementi, F. and Olivotto, I. (2020). Zebrafish (*Danio rerio*) physiological and behavioural responses to insect-based diets: A multidisciplinary approach. Scientific Reports, 10:10648.

Zhang, J., Shi, Z., Gao, Z., Wen, Y., Wang, W., Liu, W., Wang, X., and Zhu, F. (2021). Identification of three metallothioneins in the black soldier fly and their functions in Cd accumulation and detoxification. Environmental Pollution, 286:117146.

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