Effect of different waste substrates on the growth, development and proximate composition of black soldier fly (Hermetia illucens) larvae

Article *in* Livestock Research for Rural Development · July 2022

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Effect of different waste substrates on the growth, development and proximate composition of black soldier fly (*Hermetia illucens*) larvae

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Abstract

Black soldier fly larvae (BSFL) are a potentially sustainable source of protein for use as agriculture and aquaculture feeds, and a constructive means of disposing of organic wastes. Optimizing larval culture of these larvae depends on the refinement of husbandry methods. Towards this objective we investigated the use of different substrates on larval performance and proximate composition. Feeding and growth of BSFL on hotel wastes (HW), market wastes (MW) and mixed market and hotel wastes (MHW) were evaluated, with analysis of the efficiency of substrate consumption and waste load reduction. Mixed market and hotel wastes led to less larval production than either of the individual feed substrates ($p< 0.05$). Larvae aged < 3 days consumed waste loads substantially faster than older larvae and larvae feeding on hotel wastes grew faster and pupated earlier than those in the other two treatment groups ($p < 0.05$). Crude protein content was significantly higher in larvae reared on the mixed substrate. Mixed waste substrates is recommended as a the preferable choice for BSFL production for farming purposes.

Keywords: *black soldier fly, organic waste, protein source, sustainability, waste load*

Introduction

Black soldier fly (BSF; *Hermetia illucens*) is an insect endemic to tropical and high temperature regions (McCallan 1974) and this species is native to the southeast United States. It produces three generations every 12 months colonizing various decomposing and organic wastes (James 1935). BSF has five different stages of growth, namely egg, larva, prepupae, pupa and adult (Tomberline and Sheppard, 2002; Li et al 2011). Prepupae of BSF have a proximate composition of 44% dry matter, 42% protein and 35% fats (Hale 1973). Studies indicate that they are a good basis of nutrition for fish and can partially replace fishmeal in the diets of many fish species (Bondari and Sheppard 1987; Belghit, et al 2019; Madibana, et al 2020). For quite some time, researchers have developed different ways to manage organic waste decomposition including use of macro-fauna like black soldier fly larvae, earthworms, housefly among others (Diener et al 2009). Manure is the key food subject for many insect larvae in nature, used as a substrate for laying eggs and feeding of larvae during the developmental stages and converting them into biomass (Newton et al 2005).

Black soldier fly larvae (BSFL) can consume a wide variety of organic waste including agricultural waste (Manurung et al 2016), animal and human waste (Pujol-Liz et al 2008), food waste (Oonincx et al 2015a) and livestock waste (Oonincx et al., 2015b) for its growth and development. According to Dortmans et al 2017, municipal waste mostly consists of market waste (fruits and vegetables with at least 95% water content), agro-industrial wastes, food and restaurant waste. The agro-industrial wastes are generated from food processing wastes, spent grains and slaughterhouses. The BSF converts different organic wastes i.e. food wastes, animal manure and agro-industrial wastes from sources such as food processing industries and breweries into larval biomass and a mixture of frass (Lalander et al 2019; Gold et al 2020).

Various waste substrates contribute differently to the growth of BSF larvae. This includes municipal wastes (sewage sludge, usually a mixture of excreted matter and water on a sand or domestic scrap), poultry and pig manure, kitchen scraps and agricultural wastes (Dortmans et al 2017; Wang and Shelomi 2017). Previously, research has been done to investigate manure utilization and management in order to produce feeds of adequate quality for the larvae (Zhang et al 2021). However, human faecal wastes and animal manure have the potential to harbor pathogenic bacteria, viruses and parasites posing a substantial health concern. This has led the European Union (EU) to ban its use in BSF production (Green 2015). Recently, attention has shifted to higher quality feedstuffs to potentially enable BSF to serve as a feed ingredient for other farmed animals (Oonincx et al 2015a; Miranda et al 2019; Mazza et al 2020).

Utilization of BSFL contributes to recycling of nutrients naturally, with some cultured animals including fish and chicken using it as food. Depending on the usage, larvae can be sold alive to birds/chicken/fish farmers or can be processed with other ingredients to make a nutritionally complete and balanced diet. Research shows that BSFL meal has a high level of protein, amino acid and other nutrients that is comparable to that found in fishmeal (Bondari and Sheppard 1981) (Cummins et al 2017). Being able to be reared in a variety of organic wastes, BSFL provides a potential approach to reduce the waste volume (Sheppard, Newton, Thompson and Savage 1994) and requires a small space for farming due to their small size nature, a study conducted by (Oonincx, et al 2015a). Feed protein source should meet optimum conditions for production like regular availability of the protein in quantity, economic value, human resource incompetence, environmental sustainability etc. and BSFL seem to fit these requirements. However its nutritional composition can be modified through a change in the composition of the waste substrate (Meneguz et al 2018). The growth, development and economic viability of an insect feed depend primarily on the substrate consumed by the insect throughout its development. So far, few studies have assessed the nutritional quality of BSFL as affected by diet in the developing world (Shumo et al 2019). A comprehensive analysis of the feed ingredient is vital to assess its nutritional value. BSFL studies on body composition have proved to be influenced by their production methods, processing and substrate consumption (Fasakin, Balogun, and Ajayi 2003), so investigating the nutritional composition of the kinds of available substrates key. Hotel and market waste are readily available and have been found to have high omega-3 fatty acid content (Spranghers et al 2017). The current study investigated the nutritive value of the hotel, market and a mixture of hotel and market waste substrates, their effects on growth and development of BSFL. Proximate compositions and relative concentrations of crude protein, crude fibre, crude ash, moisture, calcium and energy contents of BSFL fed on the different waste substrates were determined to. Identify a suitable substitute for fish meal protein in BSFL production for use as a fish feed ingredient.

Materials and methods

Experimental design

The experiment was conducted at Biobuu Limited, Kilifi County, Kenya. It was done in two phases which involved hatching of the eggs and rearing of the larvae. The first phase was conducted to quantify the larvae produced when BSF eggs are hatched in the different waste substrates and the second was conducted using larvae. Larvae were fed on the three experimental waste substrates to investigate waste utilization efficiency, production rate, development, growth performance and proximate composition. Three substrates were used in triplicates: Market waste (MW), hotel waste (HW) and a mixture of market and hotel waste (MHW). Eggs from BSF were drawn, hatched and the resulting larvae fed on the three experimental waste substrate treatments. In the first set experiment, a total of 90 g of BSF eggs were sourced from Biobuu Ltd. with each waste substrate holding 30 g of 5 g subdivisions. The performance of these larvae was monitored for one week.

After hatching, larvae were counted and distributed to the grow-out section in 46 feeding trays with MW for 15 trays, HW in 16 trays and MHW in 15 trays. The number of trays for each substrate distribution was based on the hatching performance of BSFL eggs. Feeding and re-feeding was done on day 1, day 5 and day 8 respectively. Larvae were then dehydrated in an oven at a temperature of 100°C for 18 hours in preparation for proximate analysis.

Waste sourcing, processing and diet preparation

The different waste categories were sourced from Kongowea market and hotels along the Kenyan coast using the company truck and transported to the site where sorting to remove plastic and hazardous materials was done to ensure that waste processed consists of organic and biodegradable materials. Market waste, hotel waste and mixture of market and hotel waste at a ratio of 50:50 was then separated, shredded by a hammer mill to reduce the waste particle size to fine particles. Market waste comprised of the decayed vegetables, meat, fish and fruits, the biodegradable hotel waste comprised of cooked food, vegetable and non-vegetable waste and the market and hotel waste was a mixture of the market and hotel waste substrate. Dewatering was done to reduce moisture by blending the different waste substrates with 300g of maize flour waste for every 3kg of waste substrate. At the beginning of the feeding trial, 5 days-old larvae (DOL) (baby larvae) were removed from the rearing unit, and were placed in the different waste substrates to grow to pupae. This was to ensure the BSF gets all the dietary nutrients required for its growth in all developmental stages. The different waste treatments MW, HW and MHW were used exclusively throughout the experimental period. The method of 10,000, 5-day-old larvae holding in a larvero (40x60x-17cm) feeding on 15 kg of wet waste (75% water) for 12 days was used as per Dortmans et al 2017. Waste was added on day 5 and 8 until the larvae developed sufficiently for harvesting on the last day (Day 13).

Feeding and refeeding of BSF

Eggs were set at the hatching shower where they hatched using a starter feed made of coconut waste and the different processed waste substrates. Hatching of eggs was done by introducing 5 g of eggs into 1,200 g starter feed for one week before moving to the main dish after 7 days to feed until they became mature adult larvae. Feeding of the larvae was done following established protocols (Dortmans et al 2017) for a period of 12 days with re-feeding on day 5 and day 8 with 3 kg of waste until harvesting. Caution was taken such that the feed could not exceed a depth of 5 cm, in order to allow larvae to process the food completely and with ease, and to avoid the attraction of unwanted flies to remaining uneaten feed.

BSF larvae harvesting and sampling

Harvesting was done after 12 days of feeding and refeeding, at which time separation of larvae from the unutilized organic residues took place. At this stage, the BSFL were at their maximum nutritional value. A vibration gauge machine was used for the residue and larvae separation because of its high vibration frequency compared to the manual sieves. Sampling of BSFL was done at every stage to record growth and mortality. Weight measurements of every BSFL on different substrates was recorded using ACS Digital scale with readability of 0.1g.

BSF proximate composition analysis

The proximate composition (crude protein, calcium, crude fibre, crude ash, moisture and energy) of the larvae reared on the three different substrates was analyzed. The BSFL were harvested, sacrificed by dipping into hot water, at temperatures of 50-70°C. They were then dried in an oven at 100°C for 18 hours, grinded and stored at - 4°C and samples were taken for analyses following standard methods by the Association of Official Analytical Chemist (A O A C 1995). To obtain the moisture content, the weight of the larvae was measured before and after being dried in an oven at 103°C for 30 min. The crude protein content was calculated by multiplying nitrogen content with conventional factor of 6.25 using the Kjeldahl method (Janssen et al 2017). Calcium was analyzed by the use of Atomic Absorption Spectrophotometer (Model AA6300, Shimadu Japan). Organic matrix was destroyed by ashing in the muffle furnace after dissolving in 6N HCL. To obtain the crude fibre content of the larvae, the weight of silica with larvae before ashing was multiplied by the weight of silica crucible with larvae after ashing divided by the larvae weight. Water and other volatile materials were condensed and organic materials burned in the presence of O_2 in air to CO_2, H_2O and N_2. Samples were boiled with sulphuric acid of 70% standard concentration, dry residues incinerated in the muffle furnace at 550 ± 25^{o}C to constant mass. Crude ash was determined by igniting larvae in a muffle furnace at 550^{o}C for 3 hours with a combustion oven (P300; Nabertherm Lillient hal, Germany) which was used to heat the samples. To obtain the energy content, samples were burned and heat released was quantified to a known mass of water in a calorimeter.

Substrates utilization

Four days old larvae in a 42 ml tube of a mean weight of 19.10 g (3,500-4,000 larvae) were placed in a 45 by 20 cm by 10 cm black, plastic feeding tray containing 3 kg of different waste substrates MW, HW and MHW. Organic manure was spread at the four corners of the tray to reduce moisture for larval feeding. Refeeding was done after five and eight days with an additional 3 kg of waste feed to allow for pupation of the larvae. The ability of the larvae to digest the different waste substrates (Conversion of Digested Feed) (ECD) was determined using the (Scriber and Slansky 1981) method as:- B = (I-F)-M and ECD = B/I-F; where B = Assimilated food used for growth (measured as pre-pupal biomass); I = Total food offered during experiment; F = Residue in the experimental tray (Undigested food + excretory products) and M = Assimilated food metabolized (calculated by mass balance). All calculations were done in grams (g). The higher the ECD the better the Feed Conversion Efficiency (FCE). Mass balance was used to create a biomass production structure to aid in predicting the substrates utilization. The larvae feed intake was divided into three outputs; weight of the diet, weight of undigested feed and weight of harvested larvae.

Data analysis

All experimental data including initial and final weight of larvae of every substrate were recorded in an Excel spreadsheet after which analysis was carried out. All the data were analyzed using one-way analysis of variance (ANOVA). Comparison of means between groups was done by Tukey's HSD test. Statistical differences were considered at a significance level of ($P < 0.05$). Data were analyzed by Statistical Package for the Social Sciences (SPSS) version 23 for Windows.

Results

Substrate utilization rate

Hotel waste (HW) was more readily consumed and digested compared to MW and MHW substrate. The effectiveness of conversion decreased with increase in larval size. The 3-day-old larvae digested feeds at a faster rate compared to 6 day larvae being experimentally reared in the three different waste substrates. The young larvae took three days to digest 3 kg of feed while the 6-day-old larvae took five days to complete the same measure, hence confirming that efficiency of conversion decreased with increase in larval size.

Efficiency of conversion of food digestion (ECD) of the different substrates

In this study, the %ECD level investigated in HW substrate was seen to be high (17.96 ± 0.23%) compared to MW and MHW which had 14.10 ± 0.12% and 13.15 ± 0.14% respectively (Fig 1: a,b, and c). The ECD levels of all the treatments were significantly different to each other ($P < 0.05$).

* (B=Pre-pupal biomass I-F=Metabolism (Food and residue) ECD = (Conversion efficiency).

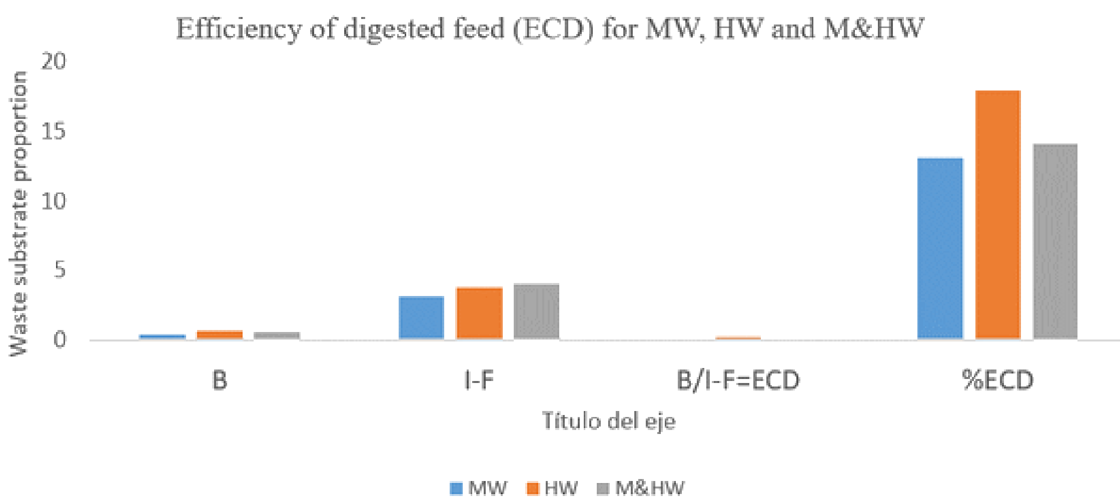


Figure 1. Substrate proportion converted into pre-pupal biomass used for energy conversion, and residue by BSFL on the different waste substrates: market waste (MW), hotel waste (HW) and mixture of hotel and market waste (MandHW)

Biomass of larvae produced during hatching

Hotel waste (HW) was more suitable for BSF hatching compared to MW and MHW. This was evident after 5 g of BSF eggs hatched in 3 kg of hotel waste (HW) produced an average of 5.89 ± 0.08 kg of young larvae in six days which was not significantly different to MW which produced 5.34 ± 0.03 kg. MHW produced a total of 4.60 ± 0.20 kg which was significantly lower than production in HW and MW (Table 1).

Table 1. Amount of larvae (kg) extracted from 5 g of BSF eggs reared on market waste (MW), hotel waste (HW) and market and hotel waste (MHW) substrates

Waste substrates	Mean larvae weight (kg)
Market waste (MW)	5.34 ± 0.03^{b}
Hotel waste (HW)	5.89± 0.08^{a}
Market and hotel waste (MHW)	4.60 ± 0.20^{c}

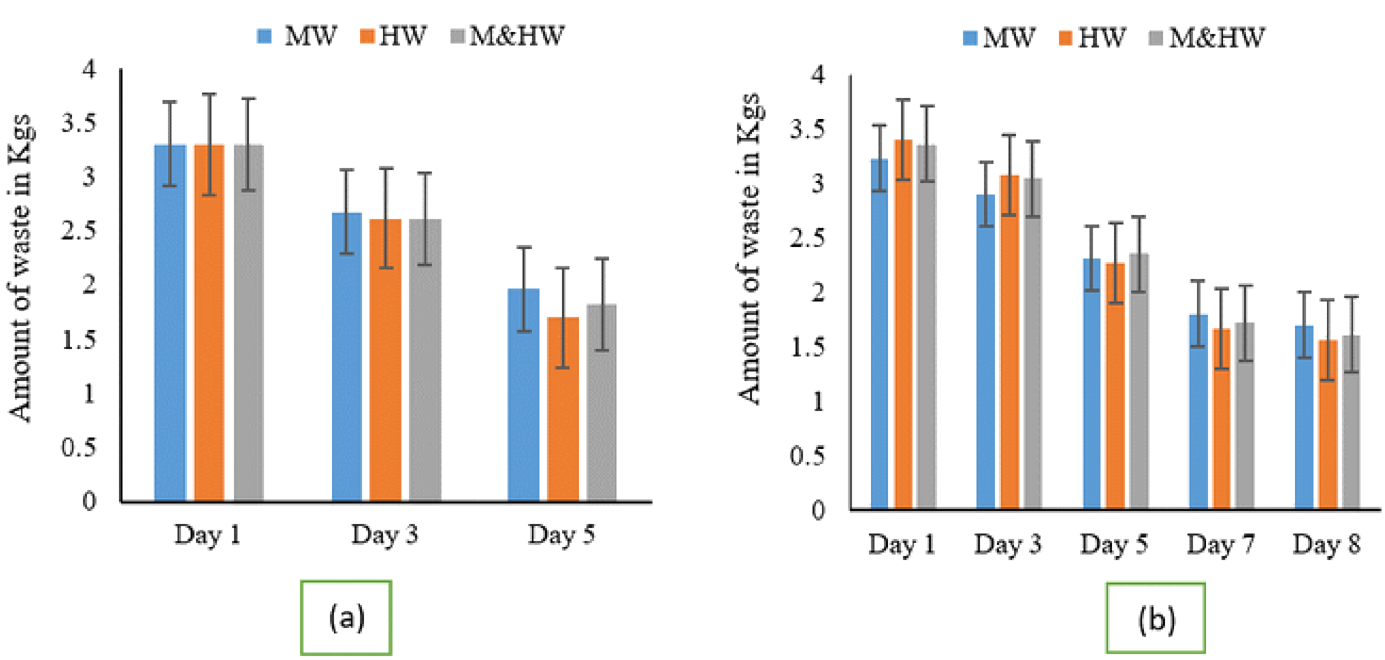
** Values expressed as Mean ±SE*

BSFL development period

To determine the period taken for larvae development, larvae were given the same proportions of feed. It was observed that larvae fed on MW substrate required 10 days to develop recording a significantly longer time ($p < 0.05$) compared to larvae fed on HW and MHW. Larvae fed on MW and MHW took 8 and 7 days to develop into pupae and were not significantly different among the treatments ($p > 0.05$) (Table 2) while larvae fed on HW took 7 days to develop into pupae. Utilization of feed waste was dependent on the size of the larvae and as such, the smaller the larvae size, the shorter the time it took to digest the different feed substrates. This was observed after young larvae of 3 days took only 4 days to digest 3 kg of wastes compared to average sized-larvae of 7 days which took 9 days to digest 3 kg of feed ($p < 0.05$).

Table 2. Feed consumption by young larvae (3 days) and medium larvae (6-days) old larvae subjected into 3 kilograms of waste feed

Substrate	3-days old larvae			6-days old larvae				
	Day 1	Day 3	Day 5	Day 1	Day 3	Day 5	Day 7	Day 8
Market waste (MW)	3.3	2.61	1.96 ±0.00^{a}	3.22	2.90	2.31	1.80	1.70 ± 0.01 ^{a}
Hotel Waste (HW)	3.3	2.62	1.70 ± 0.01^{c}	3.40	3.07	2.27	1.66	1.56 ± 0.01^{c}
Market and Hotel waste (MHW)	3.3	2.61	1.82 ± 0.00^{b}	3.36	3.04	2.35	1.72	1.61 ± 0.01^{b}



**Values expressed as Mean ±SE*

Figure 2. Graphs showing waste utilization trend of (a) small (3-days old larvae) and (b) medium sized (6-days old larvae)

BSFL growth performance

When 3-day old larvae of 19.10 g were subjected to different waste substrates to pupation, the highest mean weight gain was recorded in the larvae reared on HW substrate (0.68 ± 0.12 kg), which were significantly larger ($p < 0.05$) than the larvae reared on MHW (0.567 ± 0.03 kg and MW (0.41± 0.01 kg). BSFL fed on HW were larger in size compared to those fed on MW and MHW (Table 3).

Table 3. Growth performance of BSF larvae subjected in Market waste (MW), Hotel waste (HW) and mixture of Market and hotel waste (MHW) substrates

Waste substrates	Initial average weight (kg)	Final average weight (kg)	Average weight gained (kg)
Market waste (MW)	0.02	0.41 ± 0.01^{c}	0.39 ± 0.01^{c}
Hotel waste (HW)	0.02	0.68 ± 0.01^{a}	0.66 ± 0.01^{a}
Market and hotel waste (MHW)	0.02	0.57± 0.03^{b}	0.55 ± 0.03^{b}

**Values expressed as Mean ±SE*

Proximate composition of BSFL produced in three different waste substrates

Proximate composition of the BSF cultured in different substrates is presented in (Table 4). Crude protein and calcium were higher in larvae fed on MHW substrate at 51.52% and 3.50% respectively. Crude protein and calcium were significantly different among the treatments ($p< 0.05$). Crude fibre, ash and energy were significantly higher in larvae fed on MW substrate ($p<0.05$) and moisture content was highest in larvae fed on HW fed larvae with 54.78%. Moisture was significantly different in all the larvae from the different treatment groups ($p< 0.05$).

Table 4. Table showing proximate composition and mineral content of BSF larvae fed on market waste (MW), hotel waste (HW) and market and hotel waste (MHW) substrates

Parameter (% dry matter)	Market waste (MW)	Hotel waste (HW)	Market and Hotel waste (MHW)
Crude protein	46.52±0.55^{b}	45.29±0.52^{c}	51.52±0.01^{a}
Crude fibre	4.92±0.01^{a}	4.12±0.01^{b}	4.02±0.01^{c}
Crude Ash	10.08±0.01^{a}	9.03±0.02^{c}	9.55±0.02^{b}
Moisture	50.05 ±0.01^{c}	54.78±0.02^{a}	51.06±0.02^{b}
Calcium as Ca	3.30±0.06^{b}	3.15±0.03^{c}	3.50±0.01^{a}
Energy Kcal/kg	3,924.18 ±0.01^{a}	3,801.89±0.00^{c}	3,847.06±0.01^{b}

**Values expressed as Mean ±SE*

Discussion

Substrates utilization rate, food conversion efficiency

In this study, HW was more rapidly utilized by the BSF larvae compared to MW and MHW. The three different substrate ECD levels ranged from 13.2% to 18.0% with the HW having the highest ECD (18.0%). This indicated that BSF larvae that consumed low quality food (unbalanced food) had a lower Efficiency of Conversion of food digestion (ECD) than the one that consumed a balanced diet meal. These results are in agreement with previous studies by (Supriyatna et al 2016), who observed ECD level of 12 to 21% with cassava peel substrate, and Abduh et al 2017 who reported ECD levels of 12.5% to 25.9% while using rubber seed. Contrary to the present study, (Manurung et al 2016) found lower ECD levels of 5.69% to 10.85% while using rice straw and (Abduh et al 2017) obtained an ECD increment of 6.3% to 26.3% ECD using Screw pine (*Pandanus tectorius)* fruits. However, (Kinasih et al 2018) investigated the growth performance of BSFL *(Hermetia illucens)* fed on plant based organic waste and the ECD level was high (48.1 % and 49.54%) with a low nutrient diet substrate.

Overall BSF eggs hatching rate

The overall hatching rate of BSF eggs in this study varied with the different waste substrates. HW substrate produced a significantly higher volume of larvae compared to MW and MHW. This could be related to a more preferable nutrient profile (e.g. protein) in a HW substrate as compared with MW, which may have nutritional deficiencies or other imbalances (Chu 2018). The MW substrate could have low contents of fats and proteins rendering the substrate nutritionally inadequate and thereby resulting in reduced production of BSFL. It could also lead to slow consumption of feed by the BSFL to recompensate deficiencies in the nutrients, mainly proteins and carbohydrates (Lee et al 2004; Banks et al 2014).

BSF larval development period

When 5-day old larvae were fed with HW, they took a shorter time to develop into desirable size larvae (7 days) than MW (10 days) and MHW (8 days). It was also observed that young larvae took a significantly shorter time to reduce waste load than larger larvae. Young larvae of 4 days were more vibrant and active compared to adult larvae which could have led to loss of energy hence ability to consume and reduce the waste load at a faster rate. Barragan-Fonseca et al (2018) observed an increment in BSF development period under nutrient limitation in studying the influence of larval density and dietary nutrient concentration on performance, body protein, and fat contents of BSFL (*Hermetia illucens*) and Spranghers et al (2017) reported an achievement of shortest development time to BSF larvae fed with chicken feed. Similarly, deficiency of a certain class of nutrient (fat, proteins and carbohydrates) in a diet could have led to a prolonged development period of larvae subjected in MW, a pattern similarly observed by Green et al (2003).

BSF Larvae growth performance

Larvae fed on HW had a significantly better growth performance compared to MW and MHW. The large sized larvae in the HW substrate could have resulted from the protein rich nature of HW substrate as compared to MW and MHW substrate however, nutritional content of the substrates may vary depending on the waste substrate materials. Larvae weight was observed to be proportional to the food nutritional composition. In a study conducted by (Tschriner and Simon 2015) to investigate the influence of different growing substrates and processing on the nutrient composition of BSFL destined for animal feed, foods with higher protein sources content was seen to produce heavy larvae compared to those with more complex carbohydrates. Research reported by Nguyen et al (2013), Oonincx et al (2015b) and Simon et al (2011) recorded high growth rates on on BSF fed on a diet high in protein, and higher growth rates in response to higher fat content than diets of low fats. The hotel waste may contain more protein leading to better growth performance of the larvae.

Proximate composition of larvae subjected in three different waste substrates

The crude protein concentration in the feeds partially influnces the quality of livestock and fish feeds. Larvae reared in MHW substrate had 51.52% crude protein content. Generally, crude protein (CP) level observed in this study ranged between 45-52% which is higher than the 39% obtained by (Spranghers at al 2017) on BSFL fed on fruits and vegetables. (Sheppard et al 1994) obtained a value of 42% CP in their study on a value added manure management system by the use of BSF. The CP range obtained from this study was higher than levels reported by Sheppard et al (1994); Spranghers et al (2017) and Nguyen et al (2015) but closely in agreement with the results of Jannsen et al (2017) and Caligiani et al (2018). The *Hermatia illucens* nutrition and growth is strongly influenced by the feed nutrition it consumes (Tschimer and Simon 2015). The high content of crude protein could be a result of the rich protein and carbohydrates ingredients in the mixture of hotel and market waste substrates.

Minerals like calcium play a critical role in the BSF's physiological status including enzymatic activities, muscle weight reduction, metabolic reactions and bone formation (Hafeez et al 2015). Lack of minerals could lead to loss of muscles, stunted growth and unusual portrait. The range value of calcium obtained in this study (3.15% to 3.50%) was lower compared to (2.4% to 5.8%) as reported by Finke(2013); Dierenfeld and King (2008); Newton at al (2005) in BSFL cultured in poultry deposit. This difference could be brought about by the BSFL outer skin released as calcium carbonate leading to high levels of calcium and ash (Barragan-Fonseca et al 2017) and (Newton et al 1977). Most nutritive values of MHW were observed to be high compared to MW and HW. However, the lower moisture content was recorded in larvae fed on MHW compared to MW (51.06) and HW (54.78). The HW substrates have ample kitchen waste thereby high nutritional value, high moisture content than MW and MHW. The MW is said to have high water content (95%) as described by Dortmans et al (2017) and most of its contents (vegetables and fruits) might have been exposed to pesticides, which should be carefully checked be done before use.

The crude ash content of MW was significantly higher (9.55%) compared to HW and MHW. This is in line with the findings of Meneguz et al (2018) who investigated the effect of rearing substrate on growth performance, waste reduction efficiency and chemical composition of BSFL but contradicts the findings of Spranghers et al (2017) whose study on nutritional composition of BSF prepupae reared on different organic waste substrate found a crude ash composition of 100%, 197%, 96% and 27% on Chicken feed, digested waste, vegetable waste and restaurant waste respectively. The energy content of larvae in this study was higher on the MW substrate with 3.924 K/Cal with MHW and HW having 3,847 and 3,801 K/Cal respectively. This showed that the substrate nutritional component had an effect on the growth and production of BSFL. Diets high in energy content could be rich in carbohydrates and *vice versa* (Barragan-Fonseca et al 2018) which is a vital aspect of a healthy diet. It provides body with glucose that is used as an energy source in support of physiological functions and physical activity. According to Chamberlain et al., (1993), energy density of diets may affect animal performance since non-fibre carbohydrates have starch, simple sugars, fructan, and soluble fibre. Spranghers et. al.,2017 observed that BSFL cultured on a rich energy substrate with low ash and fibre consequently appear to have a low ash content. The crude fibre results were not significantly different in the three waste substrates. MW had the highest crude fibre content of 4.92% followed by HW whose content was 4.12% and MHW which had 4.02%. This is lower than results of (Mahmud et al 2020) who reported 9.07% as the lowest crude fibre and 10.50% as the highest crude fibre on BSFL reared for 20 days. The rearing period could have been a reason for the low content of the crude fibre in the BSFL. The longer the rearing period the lower the crude fibre content of *Hermetia illucens* (Manurung et al 2016). The BSFL of the HW substrate in this study took a short time to develop into prepupae (7 days) compared to MW and MHW substrate.

Conclusion

- The results of this study indicate that nutritional composition of waste substrates affects the growth performance and development of BSFL and that growth trend of larvae slows with the increase in the larval size in all the different waste substrates.
- The quality and quantity of substrate play a substantial role in determining the body composition of the larvae. Substrates with quality protein and carbohydrates lead to enhanced development of BSF larvae, with high protein and fat content.
- It is therefore recommended that Black Soldier Fly Larvae (BSFL) should be cultured in MHW to obtain a more optimal protein content for fish feed. At the same time, culture in the hotel waste (HW) substrate would be likely to result in more fatty larvae.

Ethical statement

All procedures carried out for the BSF production followed the approvals by the Institutional Animal care and use committee (IACUC) of Kenya Agricultural and Livestock Research Organization (KALRO)- Veterinary Science Research Institute (VSRI); approval Code No.: KALROVSRI/IACUC019/25092020.

Funding

This work was carried out with financial support from ICIPE through the Bioinnovate Africa program funded by Swedish International Development Cooperation Agency (SIDA) grant contribution number 51050076 under the project "Using Black Soldier Fly Larvae as an environmentally sustainable source of affordable protein for Chicken and Fish Feed".

Acknowledgement

We acknowledge Biobuu Kenya Ltd in Kilifi through Mr. Kigen Compton for provision of working space, test animals and the guidance offered during the experiment. The authors would like to acknowledge the BioInnovate Africa Fellowship for Women Scientists (BA-FWS) program for the sponsorship to undertake the fellowship at Biobuu Kenya Ltd in Kilifi.

Competing Interests

The authors state that there is no conflict of interest to declare

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Received 12 May 2022; Accepted 14 May 2022; Published 1 July 2022