

Utilization of black soldier flies to reduce grease waste and support zero waste

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Abstract

Purpose: This study analyzed the ability of Black Soldier Flies Larvae (BSFL) in reducing grease waste and the effect of variation in feed composition on the growth rate effect of larvae age at 8 days, 12 days, and 16 days.

Method: Before reaching the specified age (8, 12, or 16 days), the larvae are fed a mixture of expired bread and expired milk. BSFL at ages 8 days, 12 days, and 16 days are fed by pure grease waste or the mixing of grease waste and expired milk until turned into 20-day-old larvae. Then waste reduction is analyzed based on parameters such as feed consumption, waste reduction index, the efficiency of conversion of digested feed, and larvae biomass. Univariate analysis using SPSS was conducted to determine the significance of differences in waste reduction, feed consumption, conversion efficiency of digested feed, and larvae biomass among the experimental groups.

Results: The 16-day-old larvae fed with pure grease waste achieved a waste reduction index of 20.08 grams per day. Moreover, variations in feed composition influenced the growth rate of BSFL, with larvae fed a mixture of grease waste and expired milk exhibiting the largest weight (43 mg) and length (1.4 cm) among the 16-day-old larvae.

Conclusion: Black Soldier Fly Larvae at 16 days old displayed remarkable effectiveness in reducing grease waste from restaurants. This study underscores the potential of BSFL as a sustainable waste management and recycling solution for organic waste, particularly in the context of food waste.

Keywords: Black soldier flies; Sustainable waste management; Recycling; Organic waste management; Food waste

Introduction

Indonesia's population growth rate still dominates the world with a population of 270 million and is ranked

4th as the country with the largest population in the world. Along with the increasing population growth in Indonesia, there is also an increasing level of waste production. However, waste management and processing in Indonesia are still not optimal. This is evidenced by many cases of final waste disposal sites such as landfills that are already overloaded or almost overloaded due to suboptimal management and processing (Zahro et al. 2021).

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Among the various types of waste generated, waste from food establishments is a significant contributor. In a single day, eateries in malls or along roadsides engage in multiple cooking activities. Considering the dietary habits of urban Indonesians, oily foods that produce grease waste through frying are commonly consumed, resulting in a substantial amount of waste that requires proper treatment and processing before being discharged into the environment. There are many methods to reduce grease waste, one of which is the use of biodegradable technology that is now widely applied in Indonesia. Grease waste can be converted into a resource by composting it, but it takes more time, about 55 days (Chitthaluri and Rao 2022). One of the biodegradable technologies that can be applied to reduce grease waste is by utilizing insects as a bioconverter, which is Black Soldier Fly (BSF). The Black Soldier Fly (BSF), *Hermetia illucens* (Diptera: *Stratiomyidae*), is a fly species that lives in the tropics, where they can decompose organic materials (Dortmans et al. 2021). BSF can degrade organic waste by utilizing its larvae which will extract energy and nutrients from vegetable waste, food waste, animal carcasses, and manure as their food (Popa and Green 2012). The main purposes of cultivating BSF is to degrade organic waste (Sari, Taniwiryono, et al. 2022a; Sari, Taniwiryono, et al. 2022b; Sari, Azizi, et al. 2022), as well as producing various by-products such as liquid and solid fertilizer (Sari, Taniwiryono, et al. 2022c; Sari and Ridhani 2022). BSF has been proven to be a solution for organic waste and helps to achieve Zero Waste (Sari, Azizi, et al. 2022; Rohajawati and Sari 2022; Sari, Mumtaz, et al. 2022). Besides that, the cultivation of BSF can create and improve the local economy because it can be a source of protein and lipids (Sari et al. 2021; Sari and Ridhani 2022). BSF can be valorized to produce valuable by-products that are rich in protein and lipid, for animal feed or biodiesel (Franco et al. 2021; Makkar et al. 2014).

Based on the description above, Black Soldier Flies Larvae (BSFL) is a species that can help reduce grease waste. The aim of the research is to find the age of BSFL that maximizes the waste reduction index (WRI) and to find out what effects will occur if samples of grease waste are given to BSFL at a specified age (8,12, 16 days old).

Materials and methods

Selecting BSFL at specific ages is crucial for achieving desired outcomes in terms of growth and developmental characteristics (Intayung et al. 2021). These particular stages of larval development offer optimal conditions for rearing and utilizing BSFL in various applications. This research analyzed the ability of BSFL aged 8, 12, and 16 days to reduce grease waste and the effect of applying grease waste on their growth. The research was conducted at TPS 3R in Midang Village, Gunungsari District, West Lombok Regency, West Nusa Tenggara, Indonesia.

10 grams of BSF eggs were used in each biopond, obtained from the same site as the research, which is TPS 3R Midang Village. To analyze the ability of BSF to reduce organic waste, the substrates were divided into two variations, namely 1) 83% grease waste (5 kg) + 17% expired milk (1 kg), and 2) 100% grease waste (6 kg), given at multiple feedings of 0.5 kg twice a day in the morning and evening. This feeding protocol was replicated three times for each test variation to enhance the statistical robustness of the findings.

Before reaching the desired age for each test variation, the larvae were given feed consisting of a mixture of 10 kg of expired milk and 50 kg of expired bread, which was sufficient to feed the newly hatched larvae until the desired age (8,12, or 16 days).

Feed collection and preparation

The testing process in this study involved taking larval food samples first, which consisted of expired bread,

expired milk, and grease waste. The food samples were obtained from different locations. The expired bread, which was already available at the research location in TPS 3R in Midang Village, was easy to obtain. All of the expired bread was moldy, and some bread was still soft or slightly hard. Since the expired bread was still wrapped in plastic, it needed to be sorted to take only the bread. 50 kg of expired bread was used as a mixed composition in the pre-feed for newly hatched larvae in this study. Expired bread was used to compose the feed mixture that would be given before the study with test samples. Expired bread is one of the expired foods that can increase the growth and weight of larvae (Ewald et al. 2020).

Another sample used was expired liquid milk with an expiration date of more than one month, obtained from a distributor located in the city of Mataram. This expired milk did not need to be sorted in the packaging as it was already stored in jerry cans. However, there were lumps in the milk, which had to be stirred before use. These clots indicated protein damage caused by bacterial contamination of *Staphylococcus aureus* (Dyah and Arini 2017). The expired milk was used as a component in the feed mixture, and the amount used in this study was 15 kg.

The next sample used was grease waste, which was obtained from a shopping center in Mataram City. The obtained samples were separated from the waste grease and water and put into a large black plastic bag. The grease waste used had a total weight of 55 kg. This waste grease has a sticky texture, is quite moist and oily, and has a very pungent odor.

Sample variation

Variations of the test samples were put into the 1×1×0.2 m biopond. The treatment was carried out by using variations in the age of the larvae, Shown in Table 1. The experiment ended after 20 days of observation since the newly hatched larva.

Table 1. Treatment variation

Larvae Age	Treatment
8-days-old	Sample 1A: 5 kg of pure grease waste + 1 kg of expired milk
	Sample 1B: 6 kg of pure grease waste
12-days-old	Sample 2A: 5 kg of pure grease waste + 1 kg of expired milk
	Sample 2B: 6 kg of pure grease waste
16 days-old	Sample 3A: 5 kg of pure grease waste + 1 kg of expired milk
	Sample 3B: 6 kg of pure grease waste

Waste reduction efficiency

The waste reduction efficiency aims to determine BSFL's ability to reduce organic waste based on its type. Therefore, it is necessary to analyze feeding consumption, waste reduction index, and digestible feed conversion efficiency. The three analyzes are described in more detail as follows:

Feed consumption

Feed consumption (Substrate Reduction) is the amount of feed consumed by BSFL during treatment. Feed consumption is obtained by subtracting the feed mass at the beginning of the treatment and the feed mass at the end of the treatment and then dividing it by the feed mass at the start of the treatment (Hakim et al. 2017). Feed consumption is expressed in percent with the following formula (Pliantiangtam et al. 2021):

Substrate Reduction (SR)

$$= \frac{\text{Distributed substrate (gr)} - \text{Residual substrate (gr)}}{\text{Distributed substrate (gr)}} \times$$

100%

Waste Reduction Index (WRI)

Waste Reduction Index (WRI) is the ability of the larvae to reduce waste with consideration of the feeding time of the larvae, where the higher the number or WRI value, the higher the ability of the

larvae to reduce waste (Diener et al. 2009). The WRI value can be calculated using the equation formula, namely (Pliantiangtam et al. 2021):

WRI=

$$\frac{(\frac{\text{Distributed substrate (gr)} - \text{Residual substrate (gr)}}{\text{Distributed substrate (gr)}}) \times 100\%}{\text{Days of trial (day)}}$$

The Efficiency of Conversion of Digested Feed (ECD)

ECD is the efficiency of converting waste ingested by larvae during the rearing period. Calculations based on the Scriber method, namely (Diener et al. 2009):

$$ECD = \frac{B}{(I-F)}$$

Information:

B: weight gain of one larva during the eating period; obtained from weight reduction at the larvae's end with the larvae's initial weight (mg).

I: the total feed offered during the experiment (mg).

F: the weight of the remaining waste and the resulting material excretion (mg).

BSFL growth test

The growth of BSFL is known by calculating the biomass of the larvae. The formula for calculating larval biomass is as follows:

$$\text{Larvae Biomass} = \frac{\text{total weight of larvae (mg)}}{\text{larva count}}$$

Statistical analysis

To analyze the data, a statistical analysis was performed using the Statistical Package for the Social Sciences (SPSS) software. Analysis of variance (ANOVA) was performed to identify the significant differences between treatments ($P < 0.05$). When means differ significantly, the Tukey post-hoc test was performed. The recorded values from the three runs were averaged to obtain representative and reli-

able results. This approach allowed for a comprehensive assessment of the impact of age and substrate variation on the ability of BSFL to reduce grease waste and their subsequent larval growth. By conducting the research three times and reporting average values, the study aimed to provide more accurate and robust findings regarding the efficacy of BSFL at different ages in reducing grease waste. The utilization of statistical analysis using SPSS further enhanced the validity and reliability of the results obtained.

Result and discussion

Parameter observation

Newly hatched larvae phase

The food samples offered at this phase are a mix of expired bread and expired milk. Thorough observation is not carried out at this phase, but rather to keep the larvae growing regularly until they are 8 days, 12 days, and 16 days old, at which point further observations will be carried out. Based on the observations, it is known that the larvae can live and develop if nourishment in the form of expired bread and milk is provided. There was no significant difference in larval growth over the first three days of feeding, but on the fourth day, BSFL began to grow larger. The larvae fed a mixture of expired bread and milk are white and continue to grow in size until the diet composition changes at a specific age.

Eight-day-old Larvae phase

Food was replaced on the eighth day in Samples 1A and 1B, with the food being a mixture of grease waste and expired milk in Sample 1A and pure grease waste in Sample 1B. Feeding is done twice a day, in the morning and evening, until the larvae can be harvested after 20 days. Observations showed that the larvae had a rather large appetite during the first to

third day's feeding phase. However, it was observed that the larvae's appetite started to decline on the fourth day of feeding. The larvae's delayed feed consumption and the food substrate that was still present the next day serve as indicators of this. On the fifth day of observation, however, dead larvae were discovered. Because the media conditions were too sticky, larvae in reactor testing with food in the form of waste grease suffered from lower rates of survival (Klammsteiner et al. 2021).

Twelve-day-old Larvae phase

The reactors known as Sample 2A and Sample 2B received the same treatment on the 12th day, feeding the larvae with a combination of grease waste and expired milk for Sample 2A and pure grease waste for Sample 2B. According to studies of larvae at 12 days old, the larvae seem to consume the substrate more quickly than larvae at 8 days old. However, some dead larvae were also discovered the second day after being fed a diet of pure grease waste and expired milk.

Sixteen-day-old Larvae phase

16-day-old larvae with the reactor names Sample 3A and Sample 3B were the subject of the most recent observation. The larvae were treated the same way whether they were 8 days, 12 days, or 16 days old. The 16-day-old larvae consumed the provided substrate more quickly throughout the feeding phase than the 8-day and 12-day-old larvae. However, a similar incident occurred with the 12-day-old larvae, in which a number of 16-day-old larvae were discovered dead.

Waste reduction analysis

To calculate feed consumption, the remaining feed given to the larvae after a total of 20 days was weighed and compared with the feed at the beginning of the treatment. The following are the results of waste reduction analysis in experimental variations using grease waste after 20 days of observations (Table 2):

Table 2. Waste reduction analysis

Larvae Age	Food Variation	Substrate Reduction (%)		WRI (gr/day)		ECD (%)	
		Mean	Std. Deviation	Mean	Std. Deviation	Mean	Std. Deviation
8 days	5 kg Grease + 1 kg Expired Milk	91.3900	.09000	7.6233	.00577	23.5000	.15000
	6 kg Grease	91.3900	.09000	7.6167	.00577	21.6167	.14295
12 days	5 kg Grease + 1 kg Expired Milk	88.4433	.07506	11.4267	.00577	46.1067	.22502
	6 kg Grease	85.8900	.02000	11.1800	.19925	30.9333	.20502
16 days	5 kg Grease + 1 kg Expired Milk	78.8167	.01528	22.6033	.42771	159.1467	.28290
	6 kg Grease	80.3500	.02000	21.8967	.37005	132.3700	.24000

Feed consumption

The feed consumption values for pure grease waste and a mixture of grease + expired milk ranged from 78.83% to 91.39%. The highest feed consumption values were observed in samples 1A (5 kg grease + 1 kg expired milk) and 1B (6 kg grease) with a value of 91.39%. On the other hand, the lowest feed consumption value was recorded in sample 3A (5 kg grease + 1 kg expired milk) at 78.83%. The data from Table 2 indicate that higher feed consumption values correspond to a greater amount of bait consumed by the BSFL. The results also demonstrate that 8-day-old larvae consume more feed compared to 16-day-old larvae. Furthermore, the average larva consumes more feed when provided with a variation of 5 kg waste grease + 1 kg expired milk. This can be attributed to the higher protein and carbohydrate content of expired milk. Additionally, this variation contains more water content compared to pure waste grease. It is worth

noting that feed consumption with wet organic waste can reach up to 99.36% (Sari, Taniwiryo, Pratiwi, et al. 2022). Previous studies have also indicated that larvae are generally tolerant of food with a water content of 60-90%, which has a specific particle size that makes it easier to digest (Dortmans et al. 2017). In contrast, the feed consumption value for a variation of 6 kg of pure waste grease is smaller. This can be attributed to the presence of organic matter remnants from grease waste, which contain oily and weak substances mixed with chemicals and lack the necessary nutrients required by BSFL. However, BSFL can consume almost all types of waste, but it is important to determine the origin and composition of the waste (Mohd-Noor et al. 2017).

The analysis of the substrate reduction data in Table 3 shows that the model is highly significant ($p < 0.001$). The factors being tested, which are the age of the larvae and the food variation, have a significant effect on substrate reduction.

Table 3. Tests of between-subjects effects dependent variable: Substrate reduction (%)

Source	Type I Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Model	133702.521a	4	33425.630	36469.629	.000	1.000	145878.514	1.000
Ages	133702.001	3	44567.334	48625.982	.000	1.000	145877.947	1.000
Food	.520	1	.520	.568	.464	.039	.568	.108
Error	12.831	14	.917					
Total	133715.353	18						

a. R Squared = 1.000 (Adjusted R Squared = 1.000); b. Computed using alpha = .05

The descriptive statistics provide information about the mean substrate reduction and standard deviation for each combination of larvae age and food variation. For larvae aged 8 days, both the 5 kg Grease + 1 kg Expired Milk and 6 kg Grease variations show a mean substrate reduction of 91.39%. For larvae aged 12 days, the 5 kg Grease + 1 kg Expired Milk variation

has a mean reduction of 88.44%, while the 6 kg Grease variation has a mean reduction of 85.89%. Lastly, for larvae aged 16 days, the 5 kg Grease + 1 kg Expired Milk variation has a mean reduction of 78.82%, while the 6 kg Grease variation has a mean reduction of 80.35%. The results suggest that the age of the larvae has a significant impact on substrate

reduction. There are significant differences in substrate reduction between larvae aged 8 days and 16 days, as well as between larvae aged 12 days and 16 days. However, there is no significant difference in substrate reduction between larvae aged 8 days and 12 days.

On the other hand, the food variation does not show a significant effect on substrate reduction. The substrate reduction percentages for the two food variations, 5 kg Grease + 1 kg Expired Milk and 6 kg Grease, do not significantly differ from each other.

The experiments demonstrate varying levels of substrate reduction based on larvae age, indicating that younger larvae tend to have higher substrate reduction percentages. However, the specific food variation used does not significantly influence the substrate reduction. While anaerobic digestion and composting are common biotreatment methods for grease waste, they require more time compared to the use of BSFL (Chitthaluri and Rao 2022; Lemus and Lau 2002; Long et al. 2012). Moreover, unlike many pests that consume waste, BSFL do not carry bacteria or diseases, and their larvae can inactivate harmful bacteria such as *Escherichia coli* and *Salmonella enterica* (Erickson et al. 2004). Therefore, the use of BSFL can be a viable and effective solution for managing grease waste while also promoting environmental sustainability.

Waste Reduction Index (WRI)

Based on the updated information from Table 2, the Waste Reduction Index (WRI) values were analyzed. The highest WRI value was 22.60 g/day, observed in 16-day-old larvae with sample 5 kg Grease + 1 kg Expired Milk. Conversely, the lowest WRI value was 7.62 g/day, found in 8-day-old larvae with sample 5 kg Grease + 1 kg Expired Milk and 6 kg Grease. This

discrepancy can be attributed to the age of the larvae and the feeding variations, where the WRI value decreased for 8-day-old larvae. Therefore, the timing of feeding and the composition of the feed mixture can influence the WRI values, with higher values associated with specific feeding conditions (Pasymi et al. 2022).

Upon examining Table 4, the comparison of larvae age displayed a significance value (P) of 0.000, indicating a significant effect of larvae age on the Waste Reduction Index (WRI) values. Post hoc tests using Tukey's method further confirmed significant differences among all age groups. Specifically, there were significant differences between larvae aged 8 days and 12 days (P = 0.001), larvae aged 8 days and 16 days (P = 0.000), as well as larvae aged 12 days and 16 days (P = 0.000) regarding their WRI values.

On the other hand, the comparison of feed variations, including 5 kg of waste grease + 1 kg of expired milk and 6 kg of pure grease waste, yielded a significance value (P) of 0.998. This indicates that there was no significant difference in WRI values between the different feed variations. Therefore, further post hoc tests were not necessary.

In summary, the statistical analysis demonstrated a significant influence of larvae age on the WRI values, while the feed variations did not significantly affect the WRI values. Several studies have shown the effectiveness of using BSFL for organic waste management. For example, BSFL are effective at reducing the volume and mass of various types of organic waste, including food waste, manure, and slaughterhouse waste (Lalander et al. 2019). Furthermore, a previous study investigated the potential use of BSFL to reduce the volume of organic waste with the highest waste reduction index achieved at a feeding rate of 7.5 g larvae per 100 g of organic waste per day (Diener et al. 2009).

Table 4. Tests of between-subjects effects of waste reduction index

Source	Type I Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Model	4085.814 ^a	4	1021.454	13012.937	.000	1.000	52051.747	1.000
Ages	4085.353	3	1361.784	17348.625	.000	1.000	52045.876	1.000
Food	.461	1	.461	5.870	.030	.295	5.870	.616
Error	1.099	14	.078					
Total	4086.913	18						

a. R Squared = .997 (Adjusted R Squared = .996); b. Computed using alpha = .05

The efficiency of Conversion Of Digested Feed (ECD)

The ECD (Digestible Feed Conversion Efficiency) values in this study ranged from 21.74% to 159.31%. As shown in Table 2, the ECD value for feeding with 5 kg of waste grease + 1 kg of expired milk was higher compared to feeding with 6 kg of waste grease alone. A lower feed consumption resulted in a decrease in the conversion of feed into BSF larval biomass, leading to a decrease in the ECD value, which can impact larval weight (Hakim et al. 2017).

The quality of the feed provided affects the ECD value of insect larvae growth (Ahmad 2001). Poor feed quality can lead to a lower ECD value, which

indicates that pure waste grease has less nutritional value than the mixture of grease waste and expired milk.

BSFL, as polyphagous insects, can utilize various food sources using enzymes involved in digestion. Enzymes such as amylase, lipase, and protease actively assist in the digestive processes in the BSFL gut (Kim et al. 2011). Most digestive enzymes work well at an early age and then decrease at the end of the larval stage (Intayung et al. 2021). However, the results presented in Table 5 show a difference. This indicates that BSFL can more abundantly and easily digest a mixture of expired bread and expired milk during the early larval stage compared to variations of grease waste as they become adult larvae.

Table 5. Tests of between-subjects effects of dependent variable: ECD (%)

Source	Type I Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Model	140389.644a	4	35097.411	1054.392	.000	.997	4217.568	1.000
Ages	139428.963	3	46476.321	1396.236	.000	.997	4188.707	1.000
Food	960.681	1	960.681	28.861	.000	.673	28.861	.999
Error	466.016	14	33.287					
Total	140855.660	18						

a. R Squared = .997 (Adjusted R Squared = .996); b. Computed using alpha = .05

The statistical analysis compared the age of the larvae and revealed a significance value (P) of 0.007, indicating a significant effect of larvae age on the ECD value. Post hoc tests using Tukey's method indicated significant differences between larvae aged 8 days and 16 days ($P = 0.007$), as well as between larvae aged 12 days and 16 days ($P = 0.011$), suggesting variations in ECD values among these age groups. However, there was no significant difference in the ECD value between larvae aged 8 days and 12 days ($P = 0.583$).

Table 6. Larvae Growth

Larvae Age	Food Variation	Larva Weight (mg)		Larvae Length (mm)	
		Mean	Std. Deviation	Mean	Std. Deviation
8 days	5 kg Grease +1 kg Expired Milk	27.0000	.00000	.4286	.00000
	6 kg Grease	25.0000	.00000	.2857	.00000
12 days	5 kg Grease +1 kg Expired Milk	35.0000	.00000	.3000	.00000
	6 kg Grease	32.0000	.00000	.2000	.00000
16 days	5 kg Grease +1 kg Expired Milk	43.0000	.00000	.2727	.00000
	6 kg Grease	40.0000	.00000	.1818	.00000

The data presented in Table 6 indicate that the weight of BSFL is influenced by the nutritional composition of the feed provided during their growth period. It is observed that larvae fed with a mixture of grease waste and expired milk have a higher weight compared to larvae that consume only pure grease waste. The highest biomass in this experiment was observed in sample 3A (5 kg of grease waste + 1 kg of expired milk) with 16-day-old larvae weighing 43 mg, while the lowest biomass was observed in sample 1B (6 kg of grease waste) with 8-day-old larvae weighing 25 mg. The statistical analysis revealed a significant effect of larval age on their biomass, with a significance value (P) of 0.009. The results are summarized in Table 7, which provides an overview of the biomass variations observed among different age groups of

black soldier fly larvae. Furthermore, the comparison of feed variations, including 5 kg of waste grease + 1 kg of expired milk and 6 kg of pure grease waste, yielded a significance value (P) of 0.776, indicating no significant difference in ECD values between these feed variations. Therefore, further post hoc tests were not required.

Larvae growth analysis

The following are the results of observations on larval growth in experimental variations using grease waste (Table 6).

The data presented in Table 6 indicate that the weight of BSFL is influenced by the nutritional composition of the feed provided during their growth period. It is observed that larvae fed with a mixture of grease waste and expired milk have a higher weight compared to larvae that consume only pure grease waste. The highest biomass in this experiment was observed in sample 3A (5 kg of grease waste + 1 kg of expired milk) with 16-day-old larvae weighing 43 mg, while the lowest biomass was observed in sample 1B (6 kg of grease waste) with 8-day-old larvae weighing 25 mg. The statistical analysis revealed a significant effect of larval age on their biomass, with a significance value (P) of 0.009. The results are summarized in Table 7, which provides an overview of the biomass variations observed among different age groups of black soldier fly larvae. The post hoc test using Tukey's method further confirmed significant differences among different larval ages. Specifically, there was a significant difference in biomass between larvae aged 8 days and 16 days ($P = 0.008$). Similarly, larvae aged 12 days and 16 days also showed a significant difference in biomass ($P = 0.050$). However, no significant difference was found in biomass between larvae aged 8 days and 12 days ($P = 0.059$). The comparison of feed variations revealed a non-significant effect between variations in larvae feed, specifically between 5 kg of waste grease + 1 kg of expired milk and 6 kg of pure grease waste, with a significance value (P) of 0.695. Therefore, further tests (post hoc tests) are not necessary to determine significant differences in larval biomass between these feed variations.

Table 7. Tests of between-subjects effects of biomass (mg)

Source	Type I Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Model	21155.000a	4	5288.750	74042.500	.000	1.000	296170.000	1.000
Ages	21123.000	3	7041.000	98574.000	.000	1.000	295722.000	1.000
Food	32.000	1	32.000	448.000	.000	.970	448.000	1.000
Error	1.000	14	.071					
Total	21156.000	18						

a. R Squared = 1.000 (Adjusted R Squared = 1.000); b. Computed using alpha = .05

The data in Table 6 demonstrates that larval growth, specifically the length of the larvae, is influenced by the type of feed they consume during their growth period. It is evident that larvae fed with a mixture of grease waste and expired milk exhibit longer lengths compared to larvae that consume pure grease waste. The larvae fed with 5 kg of waste grease + 1 kg of expired milk showed the longest body size, particularly at 16 days of age. On the other hand, the larvae fed with 6 kg of waste grease exhibited the shortest body size, specifically at 8 days of age.

Furthermore, the analysis reveals that larval growth is also influenced by the age of the larvae when they receive the feed. Older larvae tend to have greater length than younger larvae. This aligns with the statement that BSFL increases in weight and size with age, especially when provided with adequate and nutritious feed. The 16-day-old larvae consumed a larger quantity of the mixture of expired bread and expired milk, which contains carbohydrates, protein, calcium, and fiber. In contrast, the 8-day-old larvae primarily consumed various grease waste. This finding supports the notion that the growth and composition of BSF larvae depend on the type of waste used as a feed source (Saragi and Bagastyo 2015; Joly and

Nikiema 2019). This is in line with the statement that the growth and content of BSF larvae depend on the type of waste used as a feed source. Materials that are rich in protein and carbohydrates will promote good growth for the larvae (Mohd-Noor et al. 2017). Expired bread contains nutrients in the form of carbohydrates (Dyah and Arini 2017). Meanwhile, expired milk contains protein, fat, minerals, vitamins, and amino acids (Dyah and Arini 2017; Sulistia et al. 2021; Josefin Purba et al. 2021). According to previous study, larvae reared on a balanced diet of protein (21%) and carbohydrates (21%) developed the fastest with the least amount of food and had the highest survival rate (Cammack and Tomberlin 2017). Thus, the nutritional source is also essential to extend BSFL lifespan (Lupi et al. 2019).

The statistical analysis shown in Table 8 revealed a significant effect of larval age on length, with a significance value (P) of 0.022. The post hoc test using Tukey indicates significant differences in length between larvae aged 8 days and 16 days, as well as between larvae aged 8 days and 12 days. However, there was no significant difference in larval length between larvae aged 12 days and 16 days.

Table 8. Tests of between-subjects effects of larval growth (mm)

Source	Type I Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Corrected Model	.116 ^a	5	.023	.	.	1.000	.	.
Intercept	1.392	1	1.392	.	.	1.000	.	.
Ages	.058	2	.029	.	.	1.000	.	.
Food	.056	1	.056	.	.	1.000	.	.
Ages * Food	.002	2	.001	.	.	1.000	.	.
Error	.000	12	.000					
Total	1.508	18						
Corrected Total	.116	17						

a. R Squared = 1.000 (Adjusted R Squared = 1.000); b. Computed using alpha = .05

In contrast, the analysis of variations in larval feed, comparing 5 kg of waste grease + 1 kg of expired milk and 6 kg of pure grease waste, shows no significant difference in larval length. The significance value (P) is 0.588, indicating that variations in larval feed do not have a significant effect on larval length.

It is worth noting that the larval lengths observed in this study are considerably larger than those obtained from a previous study conducted at TPS 3R in Midang Village, where larvae reared on expired food bait ranged from 200 mm to 250 mm in length and weighed around 200 mg (Sasongko 2022). The variation in feed mixture is crucial for larval biomass, and different types of waste can be utilized to achieve optimal results. Compared to the largest variation in this study, this size is five times larger. Hence, the selection of feed mixtures is crucial for larval biomass. Several types of waste that can be used are chicken feed with a biomass production of 200 mg/larva/day, vegetable and fruit waste of 130 mg/larva/day, dairy cow dung of 70 mg/larva/day, and palm kernel meal of 64 mg/larva/day (Joly and Nikiema 2019).

Overall, the findings indicate that larval growth and length are influenced by both the composition of the feed and the age of the larvae. Larvae fed with a combination of grease waste and expired milk exhibit greater length, while older larvae generally exhibit larger sizes. The selection of appropriate feed variations is essential for achieving desired larval biomass in BSFL rearing.

Conclusion

Based on the conducted tests and analysis, it can be concluded that Black Soldier Fly (BSF) larvae have the ability to effectively reduce grease waste. The age of the larvae was found to have a significant impact on the waste reduction index. The highest waste reduction rate of 20.08 grams per day was observed in 16-day-old larvae, specifically in sample 3B with 6 kilo-grams of grease waste, while the lowest value of 7.62 grams per day was recorded in 8-day-old larvae in samples 1A and 1B. The study did not find any significant effect of feed variations, such as 5 kilo-grams of waste grease + 1 kilogram of expired milk or

6 kilograms of pure grease waste, on the biomass and length of the BSF larvae. However, it was observed that the provision of variations in feed composition influenced the growth rate of the larvae. Specifically, the mixture of grease waste and expired milk resulted in larvae with the highest and longest weights, measuring 43 milligrams and 1.4 centimeters, respectively, in 16-day-old larvae. Future research could take steps to evaluate the nutritional value of BSFL reared on grease waste as an ingredient of animal feed.

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Compliance with ethical standards

Conflict of interest The authors declare that there are no conflicts of interest associated with this study.

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