Global J. Environ. Sci. Manage. 10(2): 503-516, Spring 2024, Serial #38

Global Journal of Environmental Science and Management
(GJESM)

Homepage: https://www.gjesm.net/

Podcasts

ORIGINAL RESEARCH PAPER

Fermented palm kernel waste with different sugars as substrate for black soldier fly larvae

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ARTICLE INFO

Article History:
Received 03 September 2023
Revised 06 November 2023
Accepted 21 December 2024

Keywords:
Black soldier fly larvae
Nutritional value
Palm organic waste
Sugar

DOI: 10.22035/gjesm.2024.02.06

ABSTRACT

BACKGROUND AND OBJECTIVES: The palm industry generates several waste products. Some of this waste, such as palm kernel meal, has not been fully optimized for processing. Therefore, this study sought to determine whether fermented palm kernel meal with various types of sugar (fructose, glucose, maltose, and sucrose) added could be utilized as a substrate for black soldier fly larvae.

METHODS: This study investigated the use of fermented palm kernel meal with various types of sugar added at a proportion of five per cent as a substrate for black soldier fly larvae. Fermented palm kernel meal without added sugar was used as a control substrate. Seven-day-old larvae were fed fermented palm kernel meal as an experimental substrate for 22 days and harvested on the final day, when their weight and length were measured and they were processed into meal and oil to evaluate their nutritional composition.

FINDINGS: The addition of sugars to fermented palm kernel meal made no significant difference to the final weight or crude fat value of the larvae, but improved crude protein. The addition of glucose significantly increased the length of the larvae and increased their lauric acid value. However, glucose-added fermented palm kernel meal significantly reduced the relative percentage of total unsaturated fatty acids and the quantities of linolelaidic, α-linolenic, and nervonic acid compared to the larvae fed on substrates with other added sugars. Meanwhile, fructose-added substrate resulted in significantly higher crude protein and moisture values, but significantly lower ash and carbohydrate values than those of other groups. Sucrose-added substrate resulted in a considerable improvement in ash content; magnesium; the relative percentage of total unsaturated fatty acids; arachidic, erucic, and docosadienoic acid; phosphorus; sodium; and iron values in the larvae. The larvae grown in the substrate with added maltose had a significantly higher accumulation of phosphorus, sodium, and iron, but showed significantly lower palmitoleic acid than other larvae groups. Calcium and potassium were accumulated better in the larvae grown on fermented palm kernel meal with added either glucose, maltose, or sucrose than other substrates.

CONCLUSION: of the various waste products generated by the palm industry, some, including palm kernel meal, have not yet been entirely processed. This study's findings offer insights into managing the fermented palm kernel meal, which can be converted into valuable biomass with black soldier fly larvae, making the waste more sustainable and rich in nutrients. The addition of various sugars to fermented palm kernel meal improved the growth and nutritional value of the black soldier fly larvae. These results may help in building a process for the effective treatment of palm kernel meal for black soldier fly larvae production, which could further develop the feed industry and manage palm industry waste effectively by generating high protein meal as a step in creating a circular bioeconomy.

NUMBER OF REFERENCES
78

NUMBER OF FIGURES
3

NUMBER OF TABLES
5

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Note: Discussion period for this manuscript open until July 1, 2024 on GJESM website at the "Show Article".

INTRODUCTION

Globally, the rate of organic waste production is increasing rapidly (Mohadesi *et al.,* 2023). Over 600 million tons of organic waste were generated annually in the early years of this century, and that number is projected to approach one billion tons by 2025 (Angulo-Mosquera *et al.*, 2021; Cudjoe *et al.*, 2021; Liu *et al.*, 2018; Widyarsana *et al.*, 2021). Organic waste, such as palm kernel meal (PKM), a byproduct of oil extraction from palm fruits, significantly contributes to Indonesia's agricultural waste stream (Hambali and Rivai, 2017). The oil palm, *Elaeis guineensis*, is extensively grown in Indonesia, where it was originally introduced in 1911. According to statistics obtained from the Directorate General of Estate Crops, Ministry of Agriculture of the Republic of Indonesia, oil palm plantations in 1970 were exclusively managed by state-owned and commercial businesses. However, in 1979, small-scale farm plantations were also in operation. The total area of oil palm plantations increased from 133,298 in 1970 to 11.3 million in 2015. Indonesia's oil palm plantations are widely scattered over 22 provinces, spanning the country from west to east, as the country's favourable climatic and soil conditions permit. In 2015, Indonesia generated 37.5 million metric tonnes (MMt) of palm oil, including 31.3 MMt of crude palm oil and 6.2 MMt of palm kernel oil, from an oil palm plantation area of 11.3 MMt (Dahniar and Rusniati, 2019; Effendi *et al*., 2022; Sharma *et al*., 2005). The oil palm plantations in Indonesia are concentrated in the provinces of Sumatra and Kalimantan. These provinces collectively own the biggest oil palm acreage in the country (Austin *et al*., 2017; Dharmawan *et al*., 2020). Production projections for 2030 indicated that 54 metric tonnes (Mt) of empty fruit bunches, 31 Mt of mesocarp fruit fibres, 15 Mt of palm kernel shells, 130 Mt of palm oil mill effluent, 115 Mt of oil palm fronds, and 59.7 Mt of oil palm trunks will be produced (Hambali and Rivai, 2017). As the activity of the palm oil industry rapidly increases, its organic waste, including palm oil mill effluent, palm oil decanter, and palm kernel meal, may result in environmental harm. Therefore, organic waste should be handled using biological methods that are environmentally friendly (Samimi, 2024). Past studies have revealed that one alternative approach to managing industrial organic waste through biological methods involves using insects to convert organic waste into valuable nutrient biomass. The preferred insect is the black soldier fly (*Hermetia illucens* Linnaeus (L.); Diptera: Stratiomyidae), whose larvae (BSFL) can convert industrial organic waste into biomass for animal feed. BSFL have been known for their utility in converting various forms of biological material waste into insect biomass (Kumar *et al*., 2021; Liu *et al*., 2022; Siddiqui *et al*., 2022). BSFL have a high capacity to bioconvert organic waste while releasing low amounts of greenhouse gases (Liu *et al*., 2021; Luperdi *et al*., 2023; Pang *et al*., 2020), marking their potential as a future alternative animal feed source (English *et al*., 2021; Higa *et al*., 2021; Nugroho and Nur, 2018; Wang and Shelomi, 2017). Additionally, rearing BSFL is considered a sustainable method to convert several types of waste biogenically (Nugroho *et al*., 2023; Santoso *et al*., 2023). BSFL have a high protein content of about 559.9 grams per kilogram (g/kg), crude lipids of about 18.6 g/kg, and a favourable amino acid balance (Al-Qazzaz *et al*., 2016). To raise BSFL, a high-nutrient, low-cost substrate is needed, such as PKM, although this lacks adequate nutrition. Inexpensive substrates that are rich in nutrients are used to boost BSFL development. Alternative methods, such as fermentation, may help to degrade cellulose wastes into usable ingredients for BSFL cultivation. The BSFL may be collected as they reach maturity and utilized as a source of protein and lipids. Insect meal made from larvae may be fed to livestock, and the larvae's lipid supply can also be transesterified into biodiesel to contribute to meeting the world's energy needs (Raksasat *et al*., 2020). Many different substrates and methods have been tried to raise BSFL to achieve the optimum nutrient, protein, and fat values (Fitriana *et al*., 2022; Lalander *et al*., 2020; Shumo *et al*., 2019). Previous research has shown that the nutritional content of palm industry waste might be enriched by fermentation, leading to better BSFL growth. At the best inoculum proportion of 0.5 millilitres (mL) per 10 grams (g) dry weight of palm industry waste, BSFL were 34 percent (%) heavier than the control and had high lipid content (24.7%) and protein value (24.7%) (Liew *et al*., 2022). The average individual weight of BSFL on fermented waste was 0.0619±0.004 g, which was substantially higher than the average individual weight of BSFL raised on a combination of duck dung and rice straw (0.0614±0.001 g), suggesting that BSFL might be employed as high-efficiency transformation agents in the conversion of organic duck manure to stable compost (Pamintuan *et al*., 2020). In addition, BSFL grown on expired milk substrate had a high probability of survival (96.5%) (Purba *et al*., 2021). BSFL that were reared on PKM waste with added fish pellets and fructose exhibited optimum fatty acid composition without reduced growth or survival (Nugroho *et al*., 2023). Although several studies have been conducted to ascertain the growth and nutrition value of BSFL on different substrates, the differences in growth and nutrient profiles of BSFL that were grown on fermented PKM (*f*PKM) combined with varying types of sugars have not been assessed. In this study, BSFL's efficiency in transforming locally produced *f*PKM with various added sugars into insect biomass was measured on a laboratory scale. These results may contribute to better knowledge of BSFL growth, nutrient composition, and lipid or fatty acid metabolism, illuminating the utility of insect cultivation with PKM waste and added sugar (fructose, glucose, maltose, and sucrose) for fat supply. This study predicted that the addition of various sugars would improve the growth and nutritional value of the BSFL and, in particular, that fructose and glucose would enhance the measured parameters better than other sugars. These findings were intended to contribute to BSFL rearing as related to the feed industry. This study was conducted in the Animal Physiology Development and Molecular Laboratory, Faculty of Mathematics and Natural Sciences, Mulawarman University, Samarinda, East Kalimantan, Indonesia, in 2023.

MATERIALS AND METHODS

Chemicals and BSFL source

The chemical substances (analysis grade) were bought from Merck KGaA (Darmstadt, Germany) and Sigma-Aldrich (Sigma Aldrich Incorporation, United State of America). Black soldier fly eggs were provided by a local farmer of BSFL, Ahasa, Samarinda, Limited (Ltd), East Kalimantan, Indonesia). After 4 days, the BSF eggs became young larvae. The young larvae were raised in a plastic growing chamber at about 28 degrees Celsius (°C) and a relative humidity of 60–70% until day 7. Broiler chicken pellets were used as the feeding medium during this stage.

Diet setup and trials

The PKM was obtained from Manunggal, Ltd., which is located in Kalimantan Timur, Indonesia. The PKM was fermented with effective microorganisms 4 (EM4) and the resulting feed was used as the control diet (*f*PKM). The fermentation process was performed by mixing 180 g of molasses, 279 mL of EM4, and 2.5 L of water with 2.5 kg of PKM. This mixture was placed in a plastic bag that was tied tightly closed. The mixture was allowed to stand for 4 days at room temperature to allow the fermentation process to proceed. This fermentation process may have decreased the fibre content of the PKM, as indicated by the reduction of the carbohydrate content from 60.11% to 38.69%, making the *fPKM* more digestible for the BSFL. The data about unfermented PKM were obtained from a previous study (Nugroho *et al*., 2023), while the fermented PKM data were derived from this experiment (Table 1). The treatment diets were formulated with *f*PKM to which various sugars (fructose (*f*PKMfru), glucose (*f*PKMglu), maltose (*f*PKMmal), and sucrose (*f*PKMsuc)) were added. Each sugar was added to the *f*PKM at a proportion of 5%. Based on previous studies, a 5% fructose proportion was the optimum level of added sugar for optimum BSFL growth and nutrient profile (Nugroho *et al*., 2023). Approximately 3,000 7-day-old BSFL were randomly split into five groups of three plastic chambers measuring 24x15x6 centimetres (cm) (length x width x height), each containing 200 larvae. Five different feeding experiments were run in triplicate in these containers. Water was added to each of the substrates to obtain the desired humidity level of 60–70%. In the BSFL rearing chamber room, the temperature was kept at about 28°C. During the experiment, an amount of substrate was given and adjusted to the BSFL in each chamber as described in a prior study with modifications (Hoc *et al*., 2020). On days 10–22, the BSFL were given 1 kilogram (kg) of adjusted substrate. This continued until 90% of the larvae became prepupa, which occurred at about day 22. The growth and survival of the BSFL were assessed on the final day of the experiment. The initial proximate analysis and mineral values of each substrate are shown in Tables 1 and 2.

BSFL growth

The body weight (g) and length (cm) of 30 BSFL that were randomly selected from each chamber were measured on the final day of the study using a digital microscale (Sartorius, Beijing, China) and digital

Table 1: Proximate analysis of initial substrates for BSFL rearing

Proximate analysis (%, as-is basis)	*f*PKM	*f*PKMfru	*f*PKMglu	*f*PKMmal	*f*PKMsuc
Crude protein	11.65	9.25	9.26	9	9.6
Crude fat	4.6	6.69	2.35	2.69	2.44
Ash	3.57	2.95	2.8	2.72	2.83
Moisture	41.49	47.37	48.02	45.84	46.41
Carbohydrate	38.69	33.74	37.57	39.75	38.72

*f*PKM = Fermented Palm Kernel Meal; fru = fructose; glu = glucose; mal = maltose; suc = sucrose. Various types of sugars were added at level 5% into *f*PKM.

Table 2: Mineral value of initial substrates for BSFL rearing

Mineral (%, as-is basis)	*f*PKM	*f*PKMfru	*f*PKMglu	*f*PKMmal	*f*PKMsuc
Phosphorous (P)	0.39	0.4	0.41	0.4	0.4
Calcium (Ca)	0.48	0.48	0.47	0.47	0.47
Potassium (K)	0.75	0.75	0.75	0.74	0.75
Magnesium (Mg)	0.04	0.04	0.04	0.04	0.04
Sodium (Na)	0.17	0.18	0.18	0.16	0.15
Iron (Fe)	57.5	57	57	57.5	57

*f*PKM = Fermented Palm Kernel Meal; fru = fructose; glu = glucose; mal = maltose; suc = sucrose. Various types of sugars were added at level 5% into *f*PKM.

callipers (Tools, Ltd., Shanghai, China).

Proximate analysis

On the final day of the study, all of the BSFL in each group were milled after they were freeze-dried for 72 hours. To evaluate the dry matter content of pre-dried samples, they were dried at 103°C for 16 hours. Subsequently, a three-hour incineration process was conducted at a temperature of 550°C to ascertain the ash composition of each group. The Kjeldahl technique was employed to determine the total nitrogen content (Marco *et al*., 2021; Scheiner, 1976). The crude fat value was measured using the hydrolysis method in HCl and subsequent extraction in light petroleum.

Mineral profiling

The concentrations of minerals, including P, Ca, K, Mg, Na, and Fe, were quantified from the BSFL substrate and the BSFL after they were reared for 22 days on *f*PKM with various types of sugar added. The samples were ashed at a temperature of 500°C for 5 hours to achieve a steady weight. A muffle furnace (BF-02/15, SM Indo, Banten, Indonesia) was used for this procedure. After ashing, the samples were allowed to cool to ambient temperature. Then, a volume of 5 mL of a 1 molar (M) solution of nitric acid (HNO_3) was introduced. The resulting solution was subjected to filtration and transferred into a volumetric flask with a capacity of 100 mL that had been filled to its maximum capacity with a solution of 1M HNO_3. The mineral value was quantified with an atomic absorption spectrophotometer (Model AA6300, Shimadzu, Japan).

Fatty acid profiling

The frozen samples of BSFL were subjected to freeze-drying until a consistent weight was achieved. This was followed by lipid extraction with the direct methylation procedure (Ramos-Bueno *et al*., 2016). The fatty acid value of the samples was evaluated by subjecting them to methylation with 14% of boron trifluoride (BF3) to produce fatty acid methyl esters (FAME). Gas-liquid chromatography was performed with an Agilent 7890A instrument equipped with a DB-23 column at 30 metres (m) in length, 0.25 millimetres (mm) in diameter, and 0.20m of film thickness. The chromatographic analysis was conducted under the specified conditions. The term 'injection' refers to the process of introducing a substance, typically a liquid. The experimental setup consisted of a system with a volume of 1 litre (L). The inlet heater was set at 260°C, and the split ratio was maintained at 35:1. The detector heater was operated at 280°C. The hydrogen and airflow rates were set at 40 millilitres per minute (mL/min) and 400 mL/min, respectively. A purge flow of 25 mL/min was used. The gas used in the experiment was of 99.999 per cent purity. Atmospheric pressure was measured at 0.4 megapascals (MPa), while the nitrogen (N_2) pressure ranged from 0.5 to 0.8 MPa, and the hydrogen (H_2) pressure ranged from 0.3 to 0.4 MPa. The fatty acid composition was determined by calculating the fraction of identified fatty acids.

Statistical analysis

All quantitative data obtained are represented as average ± standard error mean (SEM). A one-way analysis of variance (ANOVA, performed with SPSS software, USA) was applied to evaluate statistical differences and Duncan's multiple-range post-hoc test (DMRT) at a significance of $p<0.05$ (Samimi and Nouri, 2023) was used to assess differences in significance among the experimental groups. Microsoft Excel 2022 (Microsoft Inc., USA) was used to depict graphical data.

RESULTS AND DISCUSSION

In recent decades, numerous industries have produced large amounts of waste due to human activity and rapid population expansion. According to past studies, 3.4 billion metric tonnes of solid trash are expected to be produced globally by 2050. To address trash disposal, authorities must immediately create low-cost, effective technologies that are environmentally friendly (Samimi and Shahriari-Moghadam, 2023). However, thus far, only 20% of waste is recycled, while the rest is landfilled. Waste presents a serious hazard to people, animals, and the environment when it is simply dumped in open-air landfills in poor nations (Ashokkumar *et al*., 2022). One of the waste products that can potentially affect the environment is PKM from the palm oil industry. Previous studies have mentioned that palm kernel meal, both fermented and unfermented, has been studied for its effects on the growth performance of broiler chickens (Alshelmani *et al*., 2016). BSFL that are reared on bioorganic waste, such as PKM, exhibit high nutritional protein and oil content. Compared to other substrates, such as food waste (Fu *et al*., 2022) or manure (Awasthi *et al*., 2020), PKM is a much more promising substrate. PKM as a BSFL substrate has lower potential pathogenicity and bioplastic contamination. The BSFL reared on PKM also offer higher nutritional value than those raised on other substrates. This study evaluated the effects of *f*PKM substrate with various added sugars on BSFL growth and nutritional composition. The weight and length of the BSFL fed *f*PKM with added sugars for 22 days can be seen in Figs. 1 and 2. The addition of various sugars in *f*PKM had no significant effect on the final weight of the BSFL, while the addition of glucose to the *f*PKM resulted in a final BSFL length (1.64 cm) that was significantly ($p<0.05$) higher than the lengths of the BSFL fed on other media.

The study of various sugar additions to insects' diets has been limited. Nevertheless, dipteran larvae, such as BSFL, appear capable of sustaining growth even when provided with diets lacking or low in carbohydrates or sugar. This is attributed to the insects' ability to derive the necessary energy for maintenance and weight increase from proteins (Brookes and Fraenkel, 1958). In this study, the various types of added sugar did not affect the final weight of the BSFL. This finding is supported by past studies that reported that the inclusion of monosaccharides, disaccharides, and starches did not have any significant effect on the growth indices

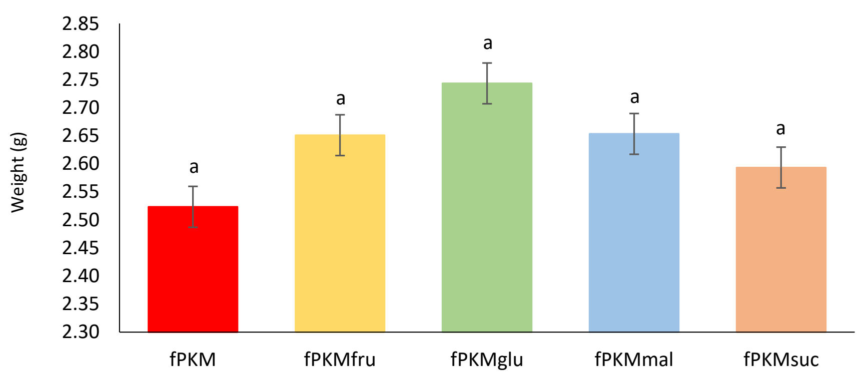

Fig. 1: Mean±SE weight of BSFL fed fermented palm kernel meal (*f*PKM) with various types of sugars at level 5% (fru = fructose; glu = glucose; mal = maltose; suc = sucrose) for 22 days

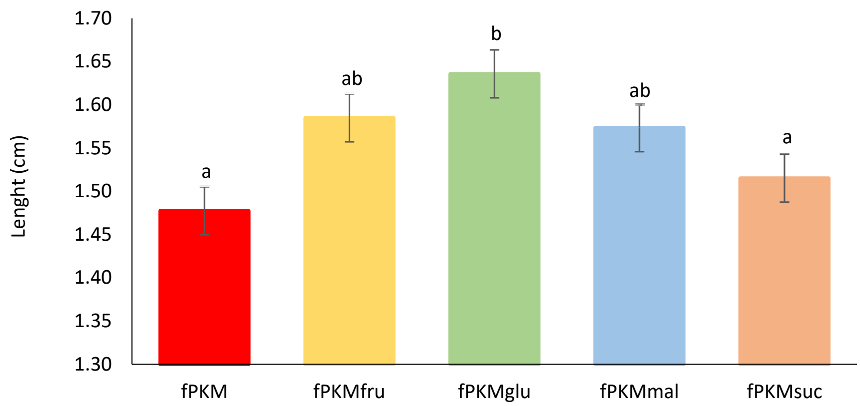

Fig. 2: Mean±SE length (cm) of BSFL after 22 days grown in fermented palm kernel meal (*f*PKM) with various types of sugars (fru = fructose; glu = glucose; mal = maltose; suc = sucrose). Various types of sugars were added at level 5% into *f*PKM. Significant differences (*P<0.05*) among the groups are exhibited by different letters (a, b) in each bar

Table 3: Proximate value of BSFL meal grown in fermented palm kernel meal (*f*PKM) added various sugar for 22 days

Proximate analysis (%, as-is basis)	*f*PKM	*f*PKMfru	*f*PKMglu	*f*PKMmal	*f*PKMsuc
Crude protein (%)	50.66±0.39[b]	52.26±0.29[c]	49.90±0.33[ab]	49.35±0.48[a]	50.65±0.02[ab]
Crude fat (%)	19.85±0.05[a]	20.75±0.06[b]	20.63±0.04[b]	20.64±0.02[b]	20.63±0.04[b]
Carbohydrate (%)	16.50±0.36[b]	12.41±0.33[a]	15.98±0.28[b]	16.60±0.42[b]	16.12±0.20[b]
Ash (%)	11.47±0.01[a]	11.65±0.06[b]	11.54±0.01[a]	11.73±0.03[b]	11.84±0.03[c]
Moisture (%)	1.51±0.01[a]	2.92±0.04[d]	1.95±0.03[c]	1.69±0.04[b]	1.91±0.01[c]

Data shown as average ± SEM (standard error mean). Significant differences between groups (*P < 0.05*) is indicated by different superscripts following the average± SEM in the same row. *f*PKM = Fermented Palm Kernel Meal; fru = fructose; glu = glucose; mal = maltose; suc = sucrose. Various types of carbohydrate were added (5%) into *f*PKM.

of BSFL (Cohn *et al*., 2022). The addition of sugars, such as galactose or xylan, resulted in a decrease in crude lipid levels. This suggests that the presence of various types of sugar in the diet of BSFL does not induce growth but may affect their proximate and fatty acid values.

Proximate analysis

The BSFL reared on *f*PKM with added fructose showed significantly higher ($p<0.05$) crude protein and moisture values but had the significantly lowest ($p<0.05$) ash and carbohydrate values compared to BSFL in other groups. The addition of any type of sugar to the *f*PKM did not significantly affect the BSFL crude fat value compared to other sugar-added substrates but increased it compared to the BSFL reared on unaltered *f*PKM (Table 3).

The use of the *f*PKM with various sugar additions resulted in the biosynthesis of a substantial amount of crude protein, between 49.35±0.48% and 52.26±0.29%, in the BSFL meal. This study's results suggest that *f*PKM with added fructose as a BSFL substrate that is fed for 22 days produces larvae with a significantly higher protein value than the other tested substrates. The crude protein level of the BSFL in this study exhibited similarities to the defatted BSFL meal from BSFL cultivated on food waste, which may contain various sugars (Ebeneezar *et al*., 2021). Similar findings were reported in a previous study, which found that the addition of fructose to PKM resulted in high protein values in the BSFL (Nugroho *et al*., 2023). This high protein content was notably greater than the protein content seen in BSFL produced using industrial agriculture waste products (39–48%) (Zulkifli *et al*., 2022), as well as those reared on rice straw (34.62%) (Pamintuan *et al*., 2020). BSFL are often reared on a substrate of organic waste to achieve a substantial protein yield (Kishawy *et al*., 2022). The considerable protein content found in BSFL makes them a viable alternative to fish meal.

This substitution is particularly advantageous given fish meal's higher cost, limited availability, and unsustainable nature. Recently, there has been a growing trend in the aquafeed business towards partially or completely substituting fish meal with other high-protein sources, such as BSFL meal (Mikołajczak *et al*., 2022; Opiyo *et al*., 2023; Zhao *et al*., 2023). The findings of this study affirm that growing BSFL on *f*PKM with or without added sugar may be a viable approach to implementing BSFL farming in the feed business. Notably, the addition of any type of sugar to the *f*PKM substrate for BSFL resulted in significantly higher amounts of crude fat content, from 20.63±0.04% to 20.75±0.06%, compared to BSFL reared on unaltered *f*PKM. One possible method by which sugars such as fructose, glucose, maltose, and sucrose may increase BSFL fat content is by influencing an insect gene for lipid metabolism that is related to lipid synthesis through a de novo pathway (Bergstrom, 2023; Biolchini *et al*., 2017; Thompson and Redak, 2000; Van Handel, 1966). However, the inclusion of fructose in the *f*PKM reduced the carbohydrate content in the BSFL. When sucrose was added to the *f*PKM, a considerable improvement in ash content was seen. The highest moisture content (2.92±0.04%) was observed in BSFL that were reared on *f*PKM with added fructose. The BSFL grown on *f*PKM with added maltose or sucrose exhibited a significantly higher accumulation of phosphorus, sodium, and iron than those of other groups. Calcium and potassium were accumulated better in the BSFL reared on *f*PKM with added either glucose, maltose, or sucrose. However, magnesium was only significantly elevated in the BSFL reared on *f*PKM with added sucrose (Table 4).

BSFL can accumulate abundant mineral deposits from a wide range of substrates (Daş *et al*., 2023; Raksasat *et al*., 2020; Shumo *et al*., 2019). The addition of sugar to the BSFL substrate was shown to increase mineral accumulation from PKM, which contains minerals in quantities of 835–6,130 parts per million (ppm) (Alimon, 2004; Bárcena-Gama *et al*., 2022). Furthermore, the BSFL raised on substrates with different added sugars exhibited diverse mineral deposit patterns, indicating a high mineral turnover (Paul *et al*., 2023; Seyedalmoosavi *et al*., 2023). Some previous studies have stated that a high-fructose diet was associated with altered phosphorus metabolism in BSFL. There is also a correlation between fructose consumption and decreased blood phosphorus values (Mayes, 1993; Milne and Nielsen, 2000; Wong, 2022). This study found nine fatty acids in the BSFL raised on *f*PKM with various types of sugar added (Table 5). This finding aligns with past studies, which reported that approximately nine fatty acids were detected in BSFL grown on PKM with added fructose (Nugroho *et al*., 2023). The lauric acid level in BSFL increased (22.60±0.34%) when the BSFL were raised on *f*PKM with added glucose. However, adding glucose to the *f*PKM results in significantly reduced values of linolelaidic (21.47±0.36%), *α*-linolenic (13.68±0.32%), and nervonic (1.21±0.00%) acids compared to other groups of BSFL raised on *f*PKM with other added sugars. The inclusion of sucrose in the *f*PKM significantly improved the values of arachidic (5.025±0.23%), erucic (5.45±0.22%), and docosadienoic (1.89±0.10%) acids. The significantly highest myristoleic acid value (5.50±0.05%) was found in the BSFL reared on *f*PKM with added maltose. However, the BSFL grown on *f*PKM with added maltose and sucrose showed significantly lower palmitoleic acid (25.44±0.39 and 25.41±0.30%, respectively) values than other groups of BSFL fed either unaltered *f*PKM or *f*PKM with added fructose or glucose.

The incorporation of sucrose in the *f*PKM for

Table 4: Mineral value of defatted BSFL meal reared in fermented palm kernel meal (*f*PKM) and various types of carbohydrate addition for 22 days (n = 3).

Minerals (%, as-is basis)	*f*PKM	*f*PKMfru	*f*PKMglu	*f*PKMmal	*f*PKMsuc
Phosphorus (P)	3.73±0.03[a]	4.30±0.06[b]	4.70±0.00[c]	4.93±0.07[d]	5.00±0.06[d]
Calcium (Ca)	1.27±0.03[a]	2.23±0.03[b]	3.47±0.03[c]	3.53±0.03[c]	3.53±0.03[c]
Potassium (K)	3.73±0.07[a]	4.53±0.03[b]	5.30±0.06[c]	5.40±0.06[c]	5.27±0.09[c]
Magnesium (Mg)	3.27±0.07[a]	4.47±0.03[bc]	4.50±0.06[bc]	4.67±0.03[cd]	4.63±0.03[d]
Sodium (Na)	1.73±0.03[a]	2.13±0.03[b]	2.63±0.01[c]	2.50±0.00[d]	2.63±0.03[d]
Iron (Fe)	1.00±0.06[a]	1.53±0.03[b]	2.47±0.03[c]	2.63±0.03[d]	2.63±0.03[d]

Data shown as average ± SEM (standard error mean). Significant differences between groups ($P < 0.05$) is indicated by different superscripts following the average± SEM in the same row. *f*PKM = Fermented Palm Kernel Meal; fru = fructose; glu = glucose; mal = maltose; suc = sucrose. Various types of carbohydrate were added (5%) into *f*PKM.

Table 5: Fatty acid value (%) of BSFL grown in fermented palm kernel meal (*f*PKM) with various types of sugars addition

Fatty acids	*f*PKM	*f*PKMfru	*f*PKMglu	*f*PKMmal	*f*PKMsuc
Lauric acid (C12:0)	17.02±0.25[a]	19.77±0.28[b]	22.60±0.34[c]	20.68±0.76[b]	19.18±0.541[b]
Myristoleic acid (C14:1)	4.65±0.05[b]	3.83±0.19[a]	4.22±0.19[a]	5.50±0.05[c]	4.07±0.09[a]
Palmitoleic acid (C16:1)	28.51±0.14[b]	28.18±0.33[b]	27.69±0.21[b]	25.44±0.39[a]	25.41±0.30[a]
Linolelaidic acid (C18:2n6t)	23.34±0.05[b]	22.49±0.29[ab]	21.47±0.36[a]	23.52±0.29[b]	21.84±0.52[a]
α-Linolenic acid (C18:3n3)	14.84±0.33[b]	14.02±0.13[ab]	13.68±0.32[a]	13.98±0.57[ab]	14.85±0.07[b]
Arachidic acid (C20:0)	3.08±0.05[a]	3.67±0.12[ab]	3.26±0.19[a]	4.14±0.33[b]	5.025±0.23[c]
Erucic acid (C22:ln9)	4.11±0.06[a]	4.20±0.02[a]	4.21±0.12[a]	3.74±0.22[a]	5.45±0.22[b]
Docosadienoic acid (C22:2)	1.18±0.03[a]	1.25±0.04[a]	1.62±0.267[a]	1.30±0.17[a]	1.89±0.10[b]
Nervonic acid (C24:ln9)	3.24±0.08[d]	2.55±0.18[cd]	1.21±0.00[a]	1.67±0.42[ab]	2.27±0.22[bc]

Data shown as average ± SEM (standard error mean). Significant differences between groups ($P < 0.05$) is indicated by different superscripts following the average± SEM in the same row. *f*PKM = Fermented Palm Kernel Meal; fru = fructose; glu = glucose; mal = maltose; suc = sucrose. Various types of carbohydrate were added (5%) into *f*PKM.

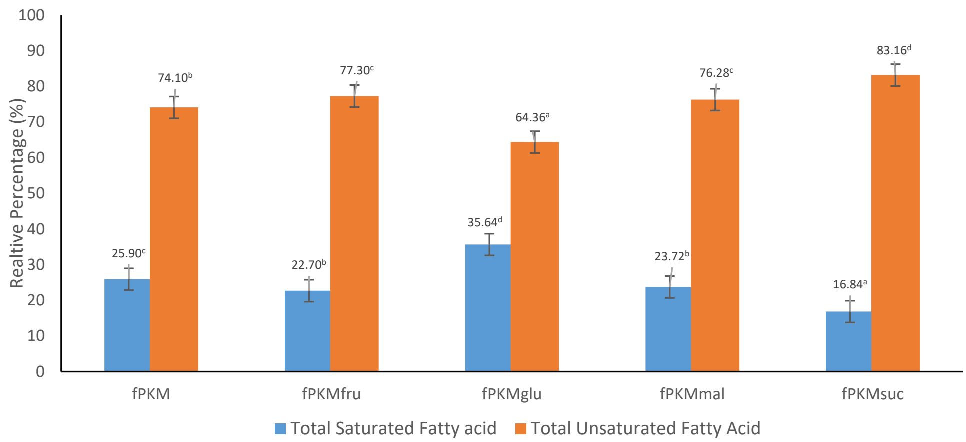

Fig. 3: Relative percentage of fatty acids in the BSFL grown in the fermented palm kernel meal (*f*PKM) added various sugars for 22 days. *f*PKM = Fermented Palm Kernel Meal; fru = fructose; glu = glucose; mal = maltose; suc = sucrose. Various types of sugars e were added at level 5% into fPKM

BSFL resulted in the significantly highest relative percentage of total unsaturated fatty acid (83.16%), but the lowest total saturated fatty acid (16.84%). Conversely, adding glucose to the *f*PKM for BSFL resulted in the significantly lowest relative percentage of total unsaturated fatty acid (64.36%) (Fig. 3).

The lipid content of BSFL may be elevated as a result of sugar in their diets, as the experimental diets boosted the biosynthesis of specific saturated fatty acids, including capric acid (C10), lauric acid (C12), and myristic acid (C14) (Hoc *et al.*, 2020). This study also revealed a notable proportion of certain fatty acids in the BSFL that were grown on *f*PKM. These fatty acids include lauric acid (C12:0), myristoleic acid (C14:1), palmitoleic acid (C16:1), linolelaidic acid (C18:2n6t), *α*-linolenic acid (C18:3n3), arachidic acid (C20:0), erucic acid (C22:ln9), docosadienoic acid (C22:2), and nervonic acid (C24:ln9). Previous studies reported that lauric acid (C12:0) was the predominant fatty acid in BSFL (Nugroho *et al.*, 2023; Shumo *et al.*, 2019), differentiating BSFL from other insect species, such as *Acheta domesticus* (Linnaeus) and *Alphitobius diaperinus* (Pfanzer) (Oonincx *et al.*, 2015). Furthermore, the bioconversion of a significant amount of carbohydrates into lauric acid is attributed to the activity of BSFL (Spranghers *et al.*, 2017). This study demonstrated that the incorporation of glucose into the *f*PKM substrate resulted in an increase in the concentration of lauric acid in the BSFL. Glucose may be involved in fatty acid synthesis via a de novo biosynthesis pathway (Prager *et al.*, 2019; Stanley-Samuelson *et al.*, 1988). The researchers highlighted the possible role of certain enzymes in the metabolic processes to produce BSFL fatty acids. Certain fatty acids, namely decanoic, lauric, and myristic acid, were exclusively detected in deuterated states. Conversely, palmitic, palmitoleic, and oleic acids were observed in both deuterated and non-deuterated forms. This suggests that BSFL can partially synthesize these fatty acids through biosynthetic pathways, rather than solely accumulating them from their diet. Fatty acids play a crucial role in insects by facilitating the synthesis of pheromones for communication and protective compounds (Blomquist *et al*., 2012; Moriconi *et al*., 2019; Pei *et al*., 2019). Furthermore, BSFL oil contains a significant concentration of lauric acid (C12:0), which resembles the composition of coconut oil (Li *et al*., 2016; Ushakova *et al*., 2016). This study also provides evidence that BSFL can efficiently convert *f*PKM with or without added sugars into significant quantities of palmitoleic acid. Dipterans may be distinguished from the members of other insect orders by their elevated palmitoleic acid levels, which often exceed 15% of their total fatty acid composition (Aguilar, 2021). The presence of palmitoleic acid is prevalent in the lipid composition of the larvae of five of the eight species belonging to the order Lepidoptera (Thomas and Kiin-Kabari, 2022). Linolelaidic acid (C18: 2n6t), which is classified as an omega-6 trans fatty acid, was present in significant quantities in the BSFL that were grown on *f*PKM with added glucose. Past research has reported the occurrence of omega-6 trans fatty acids in black soldier fly prepupae (Giannetto *et al*., 2020). The analysis of fatty acids revealed the occurrence of arachidic acid, erucic acid, and docosadienoic acid in the BSFL grown on *f*PKM with added sucrose. This finding is similar to those of previous studies revealing that rearing BSFL on sugar-beet pulp, bakery waste, and fruit and vegetable waste, which may contain sucrose, significantly affects the levels of arachidic acid, erucic acid, and docosadienoic acid present in the BSFL (Fischer and Romano, 2021; Magee *et al*., 2021; Shumo *et al*., 2019). Furthermore, the addition of maltose resulted in a notable increase in myristoleic acid, sometimes referred to as a mono-unsaturated fatty acid (MUFA). Therefore, the incorporation of maltose into BSFL substrate during industrial farming might provide advantageous outcomes in terms of MUFA production, particularly the production of myristoleic acid. The precise mechanisms by which various sugars affect BSFL's fatty acid metabolism remain largely unexplored. Nevertheless, the variation in the fatty acid compositions of BSFL during the prepupal phase may influence the regulation of genes associated with lipid metabolism throughout larval growth (Giannetto *et al*., 2020). Moreover, there are several interconnected pathways linking various types of sugar, especially glucose, and lipid metabolism (Parhofer, 2015).

CONCLUSION

The worldwide rate of organic waste generation is undergoing significant, rapid growth. In the early part of this century, the annual amount of organic waste generated exceeded 600 million tons. According to projections, this quantity is expected to reach about one billion tons by 2025. Palm kernel meal, a byproduct of palm fruit oil extraction, is a significant component of Indonesia's agricultural waste stream. Meanwhile, the oil palm plantation area continues to grow across 22 provinces in Indonesia, covering the nation from east to west. BSFL present an alternative approach to managing organic waste, such as by converting PKM into insect biomass that may then be included in animal feed. The addition of various sugars to *f*PKM is suggested for rearing BSFL to provide nutritional compounds, such as proteins, lipids and various fatty acids, as well as to support mineral accumulation in the BSFL biomass. Specifically, the use of *f*PKM with 5% fructose added to feed BSFL for 22 days improves the final protein content in the BSFL. The addition of sugars to *f*PKM did not significantly affect the final weight of the larvae or the crude fat value, but it did enhance the larvae's crude protein content. The inclusion of glucose resulted in a substantial increase in larval length, as well as an increase in the concentration of lauric acid in the BSFL. However, the addition of glucose to the *f*PKM led to a notable decrease in the relative proportion of total unsaturated fatty acids, such as linolelaidic, α-linolenic, and nervonic acid. Conversely, the addition of fructose resulted in enhanced crude protein and moisture content, while resulting in BSFL that exhibited the lowest levels of ash and carbohydrates of the measured groups. The addition of sucrose to the *f*PKM resulted in significant increases in the BSFL's ash content, magnesium levels, relative percentage of total unsaturated fatty acids, arachidic acid, erucic acid, and docosadienoic acid, as well as phosphorus, sodium, and iron values. The larvae grown on the *f*PKM supplemented with maltose showed improved accumulation of phosphorus, salt, and iron. However, these larvae had a considerably lower value of palmitoleic acid compared to the larvae in other groups. Conversely, the larvae that were grown on *f*PKM supplemented with glucose, maltose, or sucrose exhibited a greater accumulation of calcium and potassium. These findings may be beneficial for feed manufacturers seeking alternatives to fish meal and fish oil. Furthermore, exploring alternative sources of animal feed has become crucial in recent years, and palm kernel waste has emerged as a promising feed alternative option. Palm kernel waste is rich in essential nutrients and is an economical and sustainable feed choice for livestock. Its high fibre content aids digestion, while its protein content contributes to animals' growth and muscle development. Moreover, the use of palm kernel waste as animal feed reduces the environmental impacts associated with the oil processing industry's waste disposal activity. However, certain precautions must be taken, such as proper treatment to remove any harmful substances from PKM before incorporating it into feed. Thus, with proper management and utilization, palm kernel waste presents a valuable resource for animal nutrition, supporting the overall objective of sustainable agriculture.

AUTHOR CONTRIBUTIONS

R.A. Nugroho performed the literature analysis, experimental activities, writing of the manuscript, and analyzed the manuscript critically. R. Aryani contributed in the literature analysis, data, and information collection, and writing of the manuscript. E.H. Hardi performed the data and information collection, data handling, and validation. H. Manurung prepared the experimental activities, data handling, and validation. R. Rudianto performed the experimental activities and writing of the manuscript. W.N. Jati performed the experimental activities, writing of the manuscript and administration.

ACKNOWLEDGEMENT

The authors acknowledge the support of this work by [Contract Number 300/UN17.L1/HK/2023] from the Ministry of Education, Culture, Research, and Technology for the 2023 Fiscal Year. For its assistance, the authors additionally acknowledge Department of Biology, Faculty of Mathematics and Natural Sciences, Mulawarman University, Samarinda, East kalimantan, Indonesia.

CONFLICT OF INTEREST

The author declares that there is no conflict of interests regarding the publication of this manuscript. In addition, the ethical issues, including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, and redundancy have been completely observed by the authors.

PUBLISHER'S NOTE

GJESM Publisher remains neutral with regard to jurisdictional claims in published maps and institutional afflictions.

ABBREVIATIONS

°C	Degree Celsius
%	Percent
±	Plus minus
ANOVA	Analysis of Variance
BF3	Boron trifluoride
BSFL	Black soldier fly larvae
C10	Capric acid
C12	Lauric acid
C14	Myristic acid
C14:1	Myristoleic acid
C16:1	Palmitoleic acid
C18:2n6t	Linolelaidic acid
C18:3n3	α–Linolenic acid

C20:0	Arachidic acid
C22:2	Docosadienoic acid
C22:ln9	Eruchic acid
C24:ln9	Nervonic acid
Ca	Calcium
cm	Centimeter
CoA	Coenzyme A
DMRT	Duncan Multiple Range Test
EM4	Effective microorganisms 4
FAME	Fatty acid methyl esters
Fe	Iron
Fig	Figure
fPKM	Fermented palm kernel meal
fPKMfru	Palm kernel meal fructose
fPKMglu	Palm kernel meal glucose
fPKMmal	Palm kernel meal maltose
fPKMsuc	Palm kernel meal sucrose
g	Gram
g/kg	Gram per kilogram
H_2	Hydrogen
HNO_3	Nitric acid
K	Potassium
kg	Kilogram
L	Linnaeus
L	Liter
Ltd	Limited
M	Molar
m	Meter
Mg	Magnesium
mL	Mililiter
mL/g	Mililiter per gram
mL/min	Milliliter per minute
mm	Milimeter
MMT	Million metric ton
MPa	Megapascals
Mt	Metric ton
MUFA	Mono-unsaturated fatty acid
N_2	Nitrogen
Na	Sodium
P	Phosphorous
PKM	Palm kernel meal
ppm	Part per million
SDGs	Sustainable development goals
SEM	Standard error mean
TFA	Trans fatty acid

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HOW TO CITE THIS ARTICLE

Nugroho, R.A.; Aryani, R.; Hardi, E.H.; Manurung, H.; Rudianto, R.; Jati, W.N., (2024). Fermented palm kernel waste with different sugars as substrate for black soldier fly larvae. Global J. Environ. Sci. Manage., 10(2): 503-516.

DOI: 10.22035/gjesm.2024.02.06

URL: https://www.gjesm.net/article_709461.html