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Dietary effect of sugarcane juice and glucose on the adult black soldier fly lifespan (Diptera: Stratiomyidae)

Thaddeus Zaabwe[1,2*], Elias Chirwa[1], Kumbukani Mzengereza[1], Jeremiah Kang'ombe[3], Abigail Ndhlovu[1], Chimango Mtekateka[1], Emmanuel Cishibanji Ayagirwelarha[1,4]

[1]*Department of Fisheries and Aquatic Sciences, Faculty of Environmental Sciences, Mzuzu University, P.O. Box 201, Luwinga, Mzuzu 2, Malawi.*
E-mail: zthaddeus@yahoo.com; chirwa.e@mzuni.ac.mw; mzengereza.k@mzuni.ac.mw; abigailannah01@gmail.com; mtekatekachima12@gmail.com

[2]*National Agricultural Research Organisation, Bulindi Zonal Agricultural Research and Development Institute, P.O. Box 101, Hoima, Uganda.*

[3]*Department of Aquaculture and Fisheries Science, Lilongwe University of Agriculture and Natural Resources, P.O. Box 219, Lilongwe, Malawi.*
E-mail: jkangombe@luanar.ac.mw

[4]*Department of Environmental Sciences, Faculty of Sciences, Université Catholique de Bukavu (UCB), 02 Avenue de la Mission, Bugabo, Bukavu, Democratic Republic of the Congo.*
E-mail: ayagirwelarha.cishibanji@ucbukavu.ac.cd

**Corresponding author: zthaddeus@yahoo.com*

ABSTRACT

The study aimed at enhancing adult *Hermetia illucens* (Linnaeus, 1758) lifespan and oviposition by using a range of liquid diet solutions. Adult black soldier flies (BSF) have a short lifespan ranging 7–9 days when given only water during rearing. In order to increase their lifespan, alternative energy-rich liquid diet solutions need to be introduced to the flies. In this study, water (control), milk, sugarcane juice and glucose were used. A total of 100 newly emerged 4-day-old flies were introduced into rearing cages and maintained under a 12:12 light/dark cycle. Each dietary treatment was replicated three times, with 110 ml of each liquid diet provided to flies through soaked cotton wool. The flies were reared under optimum temperature (28.62 ± 4.38 °C) and relative humidity (66.27 ± 10.48 %) conditions. Significant differences in the BSF lifespan and oviposition rates were observed, $p<0.001$. Sugarcane juice diet was associated with the highest survival rate (94.00 ± 0.02 %) and oviposition (43 552 ± 2504 eggs), and the lowest values (73.00 ± 0.04% and 6584 ± 2896) were observed under the water diet. Glucose and sugarcane juice doubled the fly lifespan to 14 days. Significant differences in the hazard ratio with reference to the water diet were noted, $p<0.001$. Glucose and sugarcane juice showed the lowest risks, with hazard ratios of 0.013 and 0.034, while the milk diet was associated with the highest hazard rate of 0.322. Sugarcane juice and glucose slow down the fly mortality, thus offering the best survival and high oviposition advantage. Our results show that utilisation of energy-dense diets such as glucose and sugarcane juice increase the lifespan of adult BSF populations.

KEYWORDS: BSF, diet, Diptera, *Hermetia illucens*, hazard rate, longevity, mass rearing, survival, waste management.

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INTRODUCTION

Black soldier fly (BSF), *Hermetia illucens* (Linnaeus, 1758), has been used globally as a cost-effective agent for turning a broad array of organic waste into valuable resources, including biodiesel and products of high nutritional value (see reviews in Surendra *et al*. 2016; Wang & Shelomi 2017; Scala *et al*. 2020; Siddiqui *et al*. 2022; Laursen *et al.* 2024), coupled with a low impact on the environment (Bosch *et al*. 2019; Rehman *et al*. 2023).

Black soldier flies are known for their rapid growth and ability to convert organic waste into 'black gold', a nutrient-rich frass, which is essential for sustainable farming and circular economy (Beesigamukama *et al.* 2020; Gärttling & Schulz 2022; Lopes *et al*. 2022; Amorim *et al*. 2024) by improving crop production while lowering greenhouse gas emissions (Perednia *et al*. 2017) and reducing overall farm costs (Abiya *et al*. 2025). The use of BSF is on the rise in animal feed production, especially in aquaculture (Biasato *et al*. 2019; Çetingül & Shah 2022; Maulu *et al*. 2022; Melenchón *et al*. 2022).

The type, quality and quantity of food BSF larvae consume affect the rate of their development and the lifespan of the adult fly (Tomberlin *et al*. 2002; Gobbi *et al*. 2013; Thinn & Kainoh 2022). Supplementation of the BSF larval diet with locally available organic waste or protein-rich sources results in variation of the adult BSF lifespan (Brits 2016; Lalander *et al*. 2019). To ensure sustainable mass production, it is important to increase the adult BSF lifespan (and consequently, the egg-laying period), which normally lasts 7–14 days post-eclosion for wild populations, subject to water availability (Tomberlin *et al*. 2002). The adult BSF lifespan and fecundity are also affected by environmental conditions such as temperature, humidity and light regime (Tomberlin *et al*. 2009; Holmes *et al*. 2012; Chia *et al*. 2018; Singh *et al*. 2022). The adult BSF do not feed solids but may consume liquids, so such diet supplementation is pivotal for increasing the adult BSF lifespan and reproductive fitness. However, research on adult BSF lifespan is scanty, which contrasts with its importance during the rearing cycle (Macavei *et al*. 2020; Klüber *et al*. 2023).

One of the prerequisites for the BSF mass production is the use of diet supplementation technologies to enhance the adult BSF lifespan. This study addresses critical components of the diet besides the traditional water supplementation that can be fed to adult BSF to increase their lifespan.

MATERIALS AND METHODS

Black soldier fly egg collection, hatching and larval rearing

Fertilized BSF egg clutches were sourced from a base colony maintained at the BSF unit within the Department of Fisheries and Aquatic Science, Mzuzu University, Malawi. Egg incubation and hatching was carried out using moist maize bran (~65% moisture) placed in troughs 20×10×5 cm. Troughs were covered with a cloth and kept in a dark room until hatching. Four days after hatching, the neonate larvae (<1mm) were transferred and fed on a homogenous caloric and moist commercial poultry feed *ad libitum* for nursing. This was done to attain uniform larval size. The nursing extended for a period lasting 2 days. On day 6, the larvae were introduced to kitchen waste sourced from the university and surrounding restaurants. The waste was manually sorted, with fruit and vegetable pieces discarded to ensure its high nutritional value (Tomberlin *et al.* 2009), rid of excess of water by straining through a meshed net (0.8 mm) and kept in a semi-solid form to minimize larval drowning and increase nutrient extraction. The waste was kept airtight in 25 kg sisal sacs to prevent infestation by houseflies. To generate a base breeding colony of adult BSF, 20,000 manually counted uniform-sized larvae were reared on kitchen feed in plastic toughs (Model: FP 750/128, Kenpoly) measuring 60×30×8 cm under optimal temperature (28.62 ± 4.38 °C) and relative humidity (66.27 ± 10.48 %) (Barrett *et al.* 2023). Larvae were apportioned into 10 troughs, each holding a maximum of 2000 individuals, and reared for 3 weeks until initial pupation. Towards pupation, feed was reduced as larvae had amassed enough energy reserves. Manual sorted pre-pupae and pupae were placed in troughs with dry coffee husks as a substrate (≤3 cm deep). The troughs were placed in cage nets 50×80×50 cm with a mesh size of 1 mm for fly eclosion.

Experimental design

The experiment followed a Completely Randomized Design, while the rearing of flies followed procedures used by Nakamura *et al.* (2016). Rearing love cages measuring 80×50×50 cm and 1 mm mesh size were erected on wooden cage frames measuring 85×55×55 cm. A total of 300 newly hatched flies (<12 hours and 100 flies per replicate) were released in the rearing cage nets and monitored. Lethargic flies were discarded. Temperature (28.62 ± 4.38 °C) and relative humidity (66.27 ± 10.48 %) were monitored using a digital Thermo-Hygrometer (Model: TS-FT0421, China). Flies were reared under a 12:12 light/dark photoperiod.

Diet preparation

Supplementary liquid diet treatments included water as a control, milk as treatment 1, sugarcane juice as treatment 2, and glucose as treatment 3. Liquid solutions were selected due to their low cost and availability. Moreover, sugarcane juice contained various vitamins, minerals and antioxidants, which were lacking in glucose. Each liquid diet (milk, sugarcane juice and glucose) was diluted with water to 50 % concentration. White cotton wool was soaked with 110 ml of each diet solution (Mohini's Zigzag cotton – absorbent wool I.P.) and placed in troughs 16×10.5×5 cm prior to feeding. Diet replenishment was done after every 12 hours. The diet costs are given in Malawian Kwacha (MK) (Table 1).

Adult fly oviposition

For fly oviposition, 25 smoothened round wooden sticks (18×8 cm) were stacked by rubber bands in pairs of 5 and set in cages. The sticks were set on top of fermented fish meal that was used as a fly attractant. To minimize desiccation, eggs were harvested at 07:00 or 17:00 after 2 days following oviposition.

Data collection

Survival

Treatments were checked three times a day (morning, afternoon and evening) for dead flies. The dead flies were removed and recorded. Survival estimates were calculated using the following formulae:

- *Survival function*

$$S(t) = 1 - Pr\ (T > t)$$

Where: T is the time to event (death), and $S(t)$ is the probability (Pr) that a fly would survive beyond time (t).

- *Cumulative hazard function*

$$H(t) \int_0^t h(t)du$$

Where: $h(t)$ is the instantaneous hazard rate at time (t), and $H(t)$ is the cumulative hazard rate.

- *Percentage change in hazard*

$$(1\text{-HR}) \times 100\%$$

Where: HR is the hazard ratio.

Egg counting

Egg collection was done by gently scrapping eggs off the oviposition sticks using the blunt side of a surgical blade. The harvested eggs were placed on a porcelain dish and weighed on a NUTRIFIT electronic pocket digital scale (Model WEHA0151H, China) to the nearest 0.001 g. In order to determine the average number of eggs in a clutch, 1 g of eggs was placed on wet glass slides with 70% ethanol and counted under a portable liquid crystal display digital microscope (Model G1000, China) from magnified microscope pictures. The average number of eggs determined in 1 g from was 38428. The total number of eggs in each liquid diet trial was calcualted as following:

Number of eggs ≈ Total weight of egg clutch (g) × 38428 eggs/g.

Statistical analysis

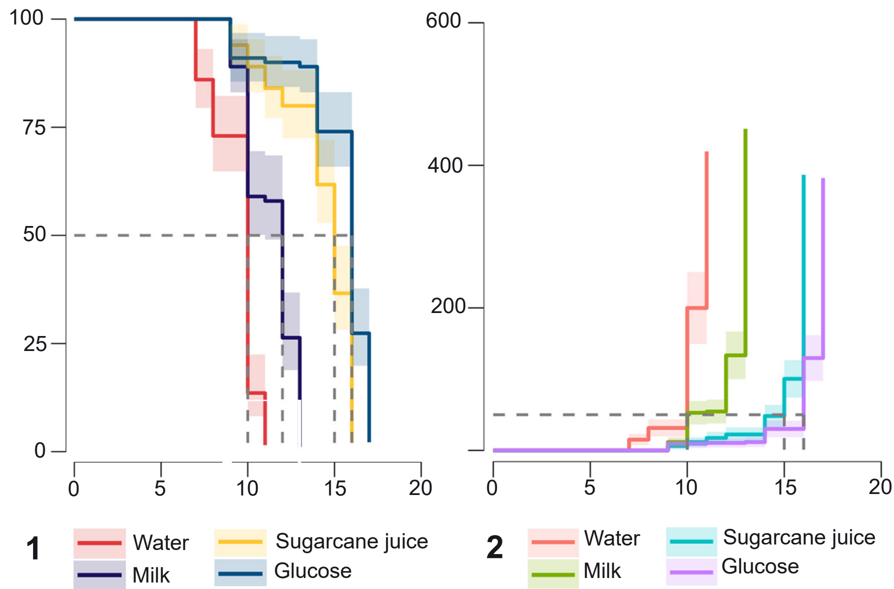
The survival probability of flies between treatments over time was estimated using the survival analysis and the non-parametric Kaplan-Meier technique (Kaplan & Meier 1958). Statistical significance between treatments was tested at $\alpha<0.05$ using a Log-rank Mantel-Haenszel test. The proportional hazard was calculated for estimates of hazard (death) across liquid diet treatments and compared at $\alpha<0.05$. This was done to ensure that the Cox proportional hazard regression model assumptions were met (Abd Elhafeez *et al*. 2021). Additionally, data on egg weight and egg number were subjected to normality and homogeneity of variances using Shapiro Wilk's and Levine's tests, respectively. This was followed by a one-way ANOVA to test for significant differences across treatment means. Tukey's Honest Significant

Difference *post hoc* was used to separate statistically significant means at $\alpha<0.05$. Data were analyzed using Python version 3.12.2.

RESULTS

Survival rates

Statistically significant differences ($p<0.001$) in the BSF survival across liquid diet treatments were recorded (Fig. 1). Flies provided with water (red) exhibited the shortest lifespan, with 50% (median survival) mortality by day 8, and 100% mortality by day 11. Similarly, although flies fed with milk (purple) survived slightly longer than those on water, their median survival (dashed line) was reached by day 11, with no surviving flies by day 13. It is notable that the survival rate was high in sugarcane juice (yellow) and glucose (blue) trials. The median survival for BSF fed with sugarcane juice was achieved around day 13 and 0% survival by day 16. The median survival forthose fed with glucose was achieved on day 14. Results from life tables indicate that the fly population experienced a 100% risk on day 7 across all diets. However, the water diet on the same day had 14 events leading to a survival rate of 86±0.03%. This was followed by a drastic reduction risk to 9% with 8 events on day 11 hence lowering survival to 1.50±0.02% (Table 2).



Figs 1, 2. (1) Survival plot of adult black soldier flies fed various liquid diets, with days on the *x*-axis and survival rate (%) on the *y*-axis; (2) cumulative hazard plot of adult black soldier flies fed various liquid diets, with days on the *x*-axis and cumulative hazard (%) on the *y*-axis.

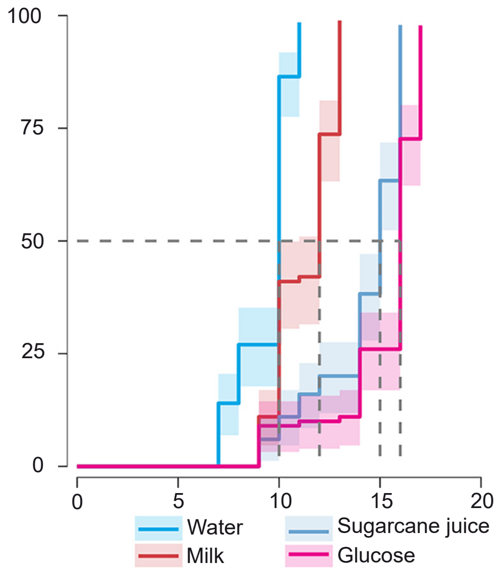


Fig. 3. Risk plot of adult black soldier fly deaths across various liquid diets, with days on the *x*-axis and risk (%) on the *y*-axis.

On the other hand, milk had a 100% risk for 2 days and subsequently reduced to 89.00±0.03% with 11 events occurring (Table 3). The high events (53) on day 13 reduced survival rate to 1.10±0.01%. Relatedly, as reflected in the milk treatment, sugarcane juice (Table 4) and glucose (Table 5) diets posed a 100% risk for the first 2 days, with 6 and 9 events occurring respectively.

The low number of events signified high survival rates compared to water and milk for the same days. The highest number of events occurred on day 16 and 17 for the sugarcane jiuce and glucose trials, with survival rates of 2.10±0.02 and 2.20±0.02, respectively. There was a significant delay in events occurring between day 9 and 13 during the sugarcane juice and glucose trials, which indicated better survival rates. From the cumulative hazard plot (Fig. 2), all diet groups started with a low risk (flat curves) until day 7 when the hazard (death) kicked in during the control trial. A steep rise in the hazard occured on day 10 in the water and milk treatments (earliest and highest spikes) indicating faster mortality. Water (orange) indicated the highest cumulative hazard by day 10. Milk (green) showed a similar pattern with a high cumulative hazard after day 12. Sugarcane juice (blue) and glucose (purple) reflected a gradual increase with the slowest cumulative hazard. The instantaneous probability (risk) of death across flies is shown in Fig. 3. Water-fed flies (light blue) showed the earliest rise in risk starting just before day 10 followed with a sharp rise in milk (red). Sugarcane juice (blue) and glucose (pink) diets showed delayed and gradual risks.

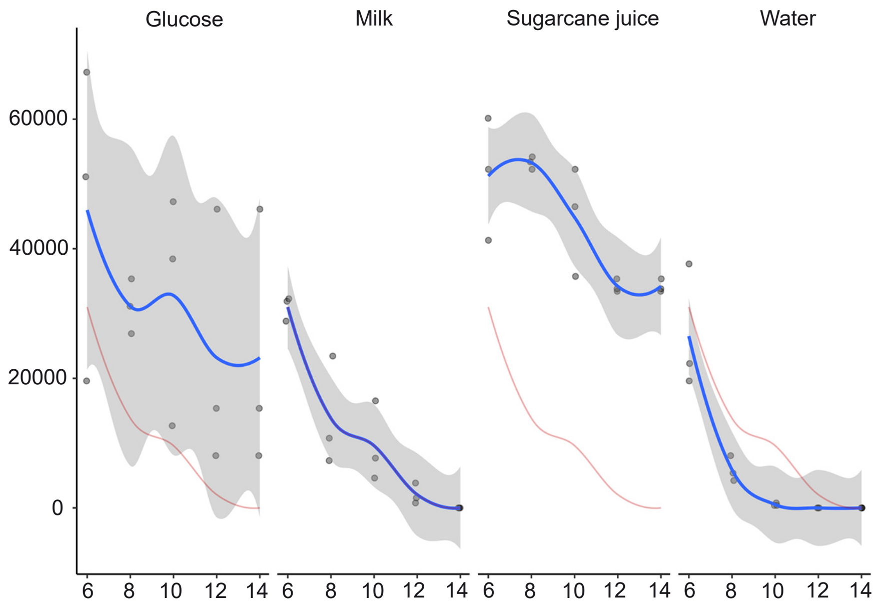


Fig. 4. Adult black soldier fly oviposition over rearing time, with oviposition duration (days) on the *x*-axis and total number of eggs on the *y*-axis.

During the control trial (light blue), the 50% median risk mark (dashed line) was achieved fastest on approximately day 10, yet milk (orange) reached the same level around day 13. The sugarcane juice (grey) and glucose (pink) trials showed the least risk, with the median risk attained on day 15 and 16, respectively. Under the water and milk treatment, steep vertical jumps indicated sudden mass deaths, while sugarcane juice and glucose diets favoured prolonged survival. Results (Table 6) show significant ($p<0.01$) differences in the survival outcomes of BSF subjected to the diets of milk, sugarcane juice and glucose, with water as a reference diet. Glucose has the most noticeable effect of reducing the hazard by 98.7%, followed by sugarcane juice at 96.6% and milk at 67.8%. Overall, the dietary liquid treatments have improved survival rates.

Oviposition

Sugarcane juice and glucose diets significantly ($p<0.001$) increased egg mass and egg number (Table 7). Higher egg masses and numbers were found in the sugarcane juice trial, compared to low egg masses and numbers under the control conditions. A significantly higher egg weight and the total egg number were noted for the sugarcane juice and glucose diets, $p<0.001$. The water and milk trials yielded lower egg masses and fewer eggs. The egg mass ranged as follows: 0.1–0.98 g (water),

Table 1. Cost of diets (per litre or kilogram) used in the experiment in MK. (USD 1 = MK 1751)

Diet	Composition (%)	Cost
Milk	100	2500
Sugarcane juice	100	1400
Water (control)	100	1200
Glucose (food grade)	50 g/l of water	1000

Table 2. Life table for adult BSF fed on water diet.

Time (days)	Risk (%)	Events	Survival (%)
7	100	14	86.00±0.03
8	86	13	73.00±0.04
9	70	0	73.00±0.04
10	70	57	13.60±0.03
11	9	8	1.50±0.02

Table 3. Life table for adult BSF fed on milk diet.

Time (days)	Risk (%)	Events	Survival (%)
7	100	0	100
8	100	0	100
9	100	11	89.00±0.03
10	89	30	59.00±0.05
11	56	1	57.90±0.05
13	24	53	1.10±0.01

Table 4. Life table for adult BSF fed on sugarcane juice diet.

Time (days)	Risk (%)	Events	Survival (%)
7	100	0	100
8	100	0	100
9	100	6	94.00±0.02
10	94	5	89.00±0.03
11	89	5	84.00±0.04
13	79	4	80.00±0.04
14	79	18	61.70±0.05
15	59	24	36.60±0.05
16	35	33	2.10±0.02

Table 5. Life table for adult BSF fed on glucose diet.

Time (days)	Risk (%)	Events	Survival (%)
7	100	0	100
8	100	0	100
9	100	9	91.00±0.02
10	91	0	91.00±0.02
11	91	1	90.00±0.03
13	90	1	89.00±0.03
14	89	15	74.00±0.04
15	73	0	74.00±0.04
16	73	46	27.40±0.05
17	25	23	2.20±0.02

Table 6. Hazard estimates across liquid diet treatments with reference to water, with *p* values recorded as ** very significant and *** highly significant.

Treatment	Hazard ratio	Change (%)	*p* value
Milk	0.322	67.8	<0.01**
Sugarcane juice	0.034	96.6	<0.001***
Glucose	0.013	98.7	<0.001***

Table 7. Oviposition of adult black soldier flies fed liquid diets. *Values (mean±SE) in rows with different superscripts are statitsically signficant at $p<0.05$ according to Tukey's *HSD*.

	Liquid diets				Polynomial contrasts		
Parameter	Water	Glucose	Sugarcane juice	Milk	Linear	Cubic	Quadratic
Total egg weight (g)	0.17±0.08[a]	0.81±0.12[b]	1.13±0.07[b]	0.29±0.08[a]	0.02	<.001	0.03
Total number of eggs	6584±2896[a]	31 255±4647[b]	43 552±2504[b]	11 298±3136[a]	0.01	<.001	0.02

0.02–0.84 g (milk), 0.87–1.57 g (sugarcane juice) and 0.21–1.75 g (glucose). The number of eggs per clutch ranged as follows: 12–476 (water), 24–576 (milk), 512–1093 (sugarcane juice) and 124–1060 (glucose). Additionally, the total number of eggs per harvest averaged 10973 (water), 14122 (milk), 43552 (sugarcane juice) and 33272 (glucose). High oviposition peaks were achieved on day 6 for water, milk and glucose diets (Fig. 4). However, oviposition under the sugarcane diet peaked on days 7 and 8. A drastic fall in the oviposition under the water and milk diets occured from day 7. For example, by day 9 in the water diet, no oviposition was observed, since 99% of the flies had died.

DISCUSSION

The survival analysis demonstrated significant differences in the adult BSF longevity and risk among the four liquid diet treatments. Sugarcane juice and glucose diets showed the highest survival rates for the days of observation, with a 50% survival beyond the 14th day. These results indicate the efficacy of sugarcane juice and glucose in sustaining the adult BSF longevity, which can be ascribed to the immediate availability of energy from the simple sugars of the liquid solutions. Such sugars sustain necessary metabolic processes at the adult stage when the feeding level is low. Conversely, flies given water show the shortest lifespan with a sharp drop in survival starting at approximately day 5 and complete deaths by day 11 marking the flies' maximum lifespan. The low survival rates can be attributed to the absence of nutritional value in water, and it cannot be recommended as a sole diet for adult BSF. Longevity of adult BSF is dependent on carbohydrate-rich diets (Bertinetti *et al.* 2019), particularly those with simple sugars that appear to extend the lifespan (Klüber *et al.* 2023) and to be more palatable to adult BSF. Energy-rich carbonydrate diets may at least double the adult BSF lifespan to over 30 days, prolong the oviposition period and boost the BSF reproductive fitness (Thinn & Kainoh 2022; Barrett *et al.* 2025). In our study, the sugarcane juice and glucose treatments have yielded the highest survival rates in relation to the milk and water diets. The high survival rates under the carbohydrate diets can further be linked to their simple structures and bioavailability, leading to a greater energy retention and utilization. Although the nutrient content of milk is obviously higher compared to that of water, it might have not satisfied the energy requirements of adult flies sufficiently. In our study, although the milk diet performed better than water, its inability to get on par with the sugarcane juice and glucose could be linked to the milk's predisposition to solidify and adult fly's inability ingest solid foods (Bruno *et al.* 2019). More so, milk's rapid spoilage under experimental conditions might have contributed to microbial contamination and fermentation, potentially accelerating fly mortality.

The high survival rates to 11 days and over 20 days across treatments can be linked to the food type offered to the BSF larvae during rearing. The BSF larvae used in this experiment were fed on kitchen waste high in protein and fat content. Corroborating this is a study by Bruno *et al.* (2019) who highlighted that highly nutritious diet of the BSF larvae enhances the lifespan of adult flies, which do not have a well-developed digestive system and on energy reserves accumulated during the larval stage (Bruno *et al.* 2025). Overall, the flies survived more than their maximum lifespan reaching the 50% quartile at approximately >9 days. The BSF larvae gut microbiom facilitates digestion of food, thus improving the adult fly lifespan (Cammack & Tomberlin 2017), but this parameter has not been monitored in our study.

The oviposition significantly increased under the sugarcane juice diet in comparison to other diets. Various proteins, carbohydrates, vitamins, minerals, flavanoids and antioxidants in sugarcane juice are phisiologically beneficial. Differences in the carbohydrate content among the sugarcane jiuce, glucose and milk are responsible for the limited energy available for oviposition under the milk and water diets. Although the presence of protein in the diet is important in sustaining fly survival, the protein-rich milk diet has been associated with low oviposition. This is in contrast to Bertinetti *et al.* (2019), who observed high oviposition in milk-fed BSF. The differences in milk freshness and microbial dynamics could account for the disparity in oviposition. While progressive spoiling results in casein breakdown and the creation of off-odour that may discourage oviposition (Klüber *et al.* 2023), fresh or fermenting milk can release attractive volatiles that encourage oviposition. Therefore, differences in oviposition responses among our trials may be explained by differences in milk management and its spoiling stage. Casein itself may not be suitable for the adult BSF, although it is effectively consumed by the BSF larvae (Yu *et al.* 2023).

CONCLUSIONS

Overall, the findings demonstrate that the diet composition has a profound impact on the adult black soldier fly lifespan and oviposition. The use of glucose and sugarcane juice significantly enhance the adult BSF lifespan/survival and oviposition. This study emphasizes that the use of energy-dense diets such as glucose and sugarcane juice may have practical implications in sustaining the adult BSF populations and improving oviposition in extensive or mass-rearing systems. In order to complement this study, future research focusing on the addition of metabolic markers, utilization of sugarcane and glucose at different proportions, and mating activity of flies is recommended.

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