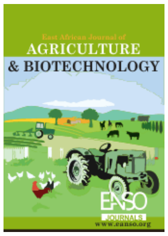

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Original Article

An Assessment of the Safety of BSF Larvae Reared on Different Substrates for Use in Animal Feeds – Chemical Contaminants

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ABSTRACT

The use of black soldier fly (BSF) larvae as a protein source for animal feed is gaining popularity. However, the safety of the larvae and the substrates used for rearing them has not been fully investigated, which is important for economic feasibility. This study aimed to evaluate the safety of BSF larvae and rearing substrates in Kenya by analysing chemical contaminants. To achieve this, the BSF larvae were reared on common production substrates, namely kitchen waste, fruit waste, and brewer's waste, in a Completely Randomised Design (CRD). Chemical contaminants such as cadmium, chromium, thallium, and lead were analysed using atomic absorption spectrometry (AAS). The data obtained were statistically analysed using the R statistical package version 4.0.5 and compared with recommended levels by KEBS, Codex Alimentarius, and the European Union (EU) standards. The study found that the larvae had lower levels of chemical contaminants (0.005±0.001 mg.kg-1 to 0.028±0.006 mg.kg-1) compared to the substrates used (0.008±0.0006 mg.kg-1 to 0.102±0.002 mg.kg-1). The average concentration of lead in BSFL was 0.0192±0.014 mg.kg-1 and 0.0391±0.037 for chromium. Cadmium was the only metal that showed an increase in concentration in the larvae reared on brewery waste (0.028±0.006) compared to larvae on other substrates. Based on the results, the study recommended the use of safe substrates such as kitchen and fruit waste for BSFL rearing.

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INTRODUCTION

The world population is projected to reach 10 billion persons by the year 2050 (United Nations, 2019). Population growth is estimated at 2.7%, 1.2% and 0.9% per year for Sub-Saharan Africa, Asia, and Latin America, respectively. On the other hand, global food demand is estimated to increase by a whopping 100% though agricultural production will only increase by a mere 60% during the same period. Therefore, the lives of approximately 805 million undernourished persons in underdeveloped countries are in jeopardy due to looming shortages in the production of major agricultural commodities such as maize, rice, wheat and soybean (Liu et al., 2017).

Sustainable meat production is in danger due to shortages in the primary ingredients used to make animal feeds, namely soybean and fishmeal. Potential plant alternatives are of inferior quality in terms of protein digestibility and amino acid composition (Tschirner & Simon, 2015). The plant proteins also contain anti-nutrients such as phytic acid, gossypol, lectins, protease inhibitors, amylase inhibitors, and goitrogens (Samtiya et al., 2020), require large land masses to produce and rely on sporadic weather conditions (Henchion et al., 2017). Most plant proteins are also utilised by humans as food (Henchion et al., 2017). Edible insects have been recommended to fill the gap.

Over the last thirty years, the black soldier fly (BSF) has been proposed as a suitable alternative protein source in animal feeds (Stamer, 2015; IDRC, 2019). The larvae have been authorised for use in poultry and aquaculture feeds in both African and European countries, courtesy of the high protein content (Cullere et al., 2018). The concern, however, is on establishment of sustainable production systems. Industrial-scale production is currently pegged on the use of organic waste as rearing substrates (Imathiu, 2020; Schmitt et al., 2019).

The ability of the larvae to consume a wide variety of organic materials and efficiently convert these into edible biomass promotes the circular economy concept (Lievens et al., 2021). However, BSFL are at risk of carrying biological and/or chemical contaminants from a variety of feeding substrates (Bouzari et al., 2015; EFSA, 2015). Whereas biological hazards can be eliminated via targeted thermal processing, which will, however, increase production costs (Schmitt et al., 2019), chemical hazards, like heavy metals and toxins, cannot be eliminated thermally (Bouzari et al., 2015). This, therefore, raises concerns about the safety of larval biomass reared on such substrates (Lievens et al., 2021). BSF larval rearing should therefore involve careful choice of feeding substrates to avoid the accumulation and consequent passing on of these harmful contents to humans (Nguyen et al., 2013). The current study, therefore, sought to evaluate the safety of both currently used BSFL rearing substrates and their respective larval products in terms of chemical contaminants.

MATERIALS AND METHODS

Location of the Study

The research was carried out at Jaramogi Oginga Odinga University of Science and Technology

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(JOOUST), located in Bondo town, Siaya County, western Kenya, approximately 62 kilometres (39 mi), by road, west of the city of Kisumu. The geographical coordinates of the University's main campus are 0°05'38.0” S, 34°15'31.0” E (Latitude: -0.093889; Longitude: 34.258611) (Ngonga et al., 2021).

The area experiences a tropical climate that significantly influences black soldier fly (BSF) farming conditions. The region follows a pattern of distinct wet and dry seasons, shaping the ideal environment for BSF cultivation. From March to October, the wet season prevails, characterised by increased rainfall and humidity (The County Government of Siaya, 2023). In contrast, the dry season, spanning from November to February, witnesses reduced rainfall. The main economic activities are fishing and conventional agricultural farming (The County Government of Siaya, 2023).

Source of Substrates

All of the substrates (including kitchen waste, fruit waste, and brewery waste) were sourced locally (either purchased at an agreed fee or collected for free) from local markets and restaurants and handled with precautions to ensure personal safety. These substrates had been chosen based on the ease of availability for the purpose of using them in large-scale industrial BSFL production (Shumo et al., 2019).

Source of Larvae for the Study

Larvae for the study were obtained from the JOOUST-INSEFOODS black soldier fly insectarium in Bondo main campus. The colony had been in existence since 2015 and was started from adults attracted from the wild (Nyakeri et al., 2016).

Adults were provided with cardboard grooves for ovipositing. Thereafter, the eggs were collected and transferred to a nursery where they were kept under controlled conditions of 30ºC with a relative humidity of 60-70%. The cardboard boxes containing eggs were placed on top of an empty basin for neonates to drop into immediately upon hatching (Woods et al., 2019).

Every day in the morning, the basins were checked for hatched larvae, and if present, these were weighed into groups of 1gram and transferred into feeding bottles containing wetted chicken mash (18% protein, 3.5% fat, 13% moisture) made by mixing 1 part feed:2 parts water for 5 days. The bottle lids were perforated and then covered with a porous paper to facilitate aeration. On the sixth day, the larvae were transferred to a bigger feeding basin, measuring 15 x 31 x 57 cm, that contained the experimental feeding substrates, namely, kitchen waste, fruit waste, and brewery waste (Ewusie et al., 2019). However, these plastic feeding bins were thoroughly cleaned before use, dried in the sun for 4-6 hours before stocking them with the feeding substrates. One gram of BSF eggs hatches to produce approximately 35000-40000 larvae.

Research Design

The study used an experimental research design. The design is used to establish a cause-and-effect relationship between two variables or among a group of variables (Vaus, 2006). The experimental research design has an experimental group and a control group, and study subjects are randomly assigned to the two groups. The researcher administers the treatment (independent variable) to the experimental /test group (manipulation) and not to the control group, and both groups are measured on the same dependent variable(s) of interest to gauge the difference due to the treatment and tries to control for confounding variables (Vaus, 2006).

To reduce variation, treatments were arranged in the larvarium using a completely randomised design. However, the study had different experimental procedures corresponding to the study objectives.

Larvae Feeding in the Larvarium.

Plastic storage racks measuring 15 x 31 x 57 cm were used to contain 6kg of the different treatment substrates, namely, kitchen waste, fruit waste, and

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brewery waste (Ewusie et al., 2019). The BSF larvae in each bottle were transferred onto the top of the respective feeding substrates as shown in Figure 4. All substrates were replenished weekly with fresh 6kg batches. Each experimental treatment was replicated three times, and the basins were kept in the larvarium at a temperature range of 25-30°C and a moisture content of 65-70% in the rearing substrate. Moisture was regulated by adding water to increase it or adding frass to dry the substrate if above the range (Ewusie et al., 2019).

Throughout the rearing process, the basins were monitored daily for signs of prepupae (Ewusie et al., 2019). At the first sign of prepupae, all the larvae in the basin were harvested by sieving off the frass and small-sized larvae using a screen mesh measuring 3.5mm. This ensured uniformity in the size of the collected larvae, which were then sampled for chemical analysis. All the larvae for a treatment were then pooled together and thoroughly shaken to mix them up.

During each feeding replenishment, 1 kg of substrate per treatment was set aside, dried and stored in airtight zip-lock bags. At the end of the feeding period, these samples were pooled together, treatment-wise, for further chemical analysis.

Larvae and Substrate Processing for Analysis.

To eliminate any unwanted physical contamination from the samples, stones and other debris in the larvae and substrates were sorted and picked out. The larvae were then sun-dried in the house to a moisture content of 10% using a sun drier. Each treatment had approximately 105,000-120,000 larvae (6kg-7.5kg) as each replicate basin had approximately 35,000-40,000 larvae whose final weight upon drying reduced to 2.5-3.5kg. On the other hand, the amount of dry substrate per treatment reduced from 14kg to a range between 6kg-8kg. Larvae and substrate samples were then ground into powder treatment-wise using a blender (Khind, BL1016/300W - 1L) and stored in airtight zip-lock bags for further analysis. Before introducing samples from the next treatment, the blender was disassembled, and the blades and glassware were thoroughly cleaned by wiping using dry towels to eliminate cross-contamination.

Figure 1: Dried Larvae (Day 1 - 4 of Drying)

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The Analysis of Chemical Contaminants.

Sample Preparation and Analysis of Heavy Metals.

For heavy metal analyses, two grams of each sample were placed in a digestion flask. This was followed by the addition of 10% nitric acid and perchloric acid to the sample. The mixed sample was then heated until the brown-red colour turned colourless, and it was subsequently cooled down and filtered using filter paper. The filtrate was then transferred to a volumetric flask and diluted with distilled water, yielding the final processed and diluted sample for testing.

The Atomic Absorption Spectrometry (AAS) method was employed to determine and quantify the available heavy metals in the samples, including Cadmium (Cd), Chromium (Cr), Thallium (Tl), Arsenic (Ar), and Lead (Pb). AAS is an analytical technique that measures the concentrations of elements qualitatively and quantitatively (Kulkarni, 2018).

The AAS instrument was calibrated with standard solutions of known concentrations to determine the concentration of each heavy metal. Sample introduction was performed with precision, following the laboratory's standard operating procedures (SOPs) (Siemer, 2018). The samples were atomised, converted into gaseous atoms, and their absorption was measured. Unique absorption lines for each heavy metal were used for identification and quantification. The absorbance of the samples was then compared to the calibration curve, which correlated absorbance values with known concentrations of heavy metal standards. The software computed the concentration of each heavy metal within the samples, presented in parts per million (ppm). Quality control measures were implemented to ensure data accuracy and prevent potential errors. The findings were meticulously recorded and reported, providing a detailed overview of the heavy metal concentrations within the filtrate.

Bioaccumulation Factor

When assessing feed conversion, the buildup of chemicals in the larvae is of serious importance. Again, as far as the applications of the larvae go, the accumulation of desirable nutrients could be advantageous, whereas the accumulation of undesirable nutrients, such as heavy metals, is undesirable. On a dry matter (DM) basis, a bio-accumulation factor (BAF) greater than 1 indicates that the element from the feeding substrate has been bio-accumulated by the insect. In this study, BAF was calculated as follows:

BAF = Concentration of heavy metal in the organism (DM) / Concentration of heavy metal in the given feed (DM).

Statistical Analysis

Statistical analysis was performed using the R statistical package version 4.0.5. The tables and Histograms were used to show a graphical representation of data. The homogeneity of variances and normality tests were performed to test ANOVA assumptions. For all analyses, means were separated using a Bonferroni adjustment at a 95% confidence level, and the obtained levels were compared against the recommended levels in the Codex Alimentarius 2020. The study utilised the Welch Two-Sample t-test to compare the means of two independent groups and the Two-Factorial ANOVA to investigate the individual and combined effects of two categorical independent variables on a continuous dependent variable. Data is expressed as mean ± standard deviation.

Two-Factorial ANOVA Model

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + \varepsilon_{ijk}$$

Equation *1*)

Equation 1: In this model,

- Yijk represents the individual data points (Dependent variable).
- μ is the overall population mean.

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- αi represents the effect of the i-th level of each factor group.
- Bj represents the effect of the j-th level of the factor group.
- (αβ)ij represents the interaction effect between the i-th and j-th levels.
- Ɛijk represents the random variability or error within each group.

RESULTS.

Chemical Contaminants (Heavy Metal Accumulation)

Table 1 presents the results related to the concentration of heavy metals in the substrates used for rearing Black Soldier Fly (BSF) larvae. There was a significant difference in the heavy metal content between the rearing substrates, ranging from 0.007 mg.kg-1 to 0.104 mg.kg-1. There was no detection of thallium and arsenic compounds in all the treatments after being subjected to the AAS machine for analysis. In all the treatments, the results showed significantly higher levels of contaminants compared to the harvested BSFL levels. In Fisher's LSD test with an alpha value of 0.05, a pairwise comparison of all the mean treatments revealed significant differences in their respective means.

Table 1: Chemical Composition of the Three Types of Substrates Used for BSFL Rearing.

Component (Mg/100g)	FW	BW	KW
Lead	0.033^{b}±0.003	0.048^{a}±0.0006	0.013^{c}±0.0025
Cadmium	0.018^{a}±0.002	0.011^{b}±0.001	0.008^{c}±0.0006
Chromium	0.03^{b}±0.001	0.102^{a}±0.002	0.015^{c}±0.0006
Thallium	ND	ND	ND
Arsenic	ND	ND	ND

Values represent heavy metal levels in ppm. FW: Fruit Waste, BW: Breweries Waste, KW: Kitchen Waste, ND: Not Detected.

Table 2 shows the results findings for the chemical composition for BSF larvae reared on the three substrates. The body mass of the harvested BSF larvae exhibited a significant difference in heavy metal content across all treatments. The concentrations ranged from 0.004 mg.kg-1 to 0.028 mg.kg-1. Thallium and arsenic compounds were not detected in the larvae after AAS analysis.

Unlike in the substrates, the results showed consistently lower levels of contaminants in the larvae with an average concentration of 0.0192±0.014 mg.kg-1 for lead and 0.0391±0.037 mg.kg-1 for chromium. Only cadmium increased in concentration in the larvae reared on brewery waste in the corresponding substrate used, which caused the treatment to increase on average by 0.0031±0.005 mg.kg-1. Pairwise comparisons using Fisher's LSD test (with an alpha value of 0.05) revealed significant differences among all mean treatments, except for the chromium metal content in larvae (LSD 0.0025), where larvae raised on brewery waste and fruit waste treatments were statistically similar.

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Table 2: Chemical Composition of the BSF Larvae Reared on Three Types of Substrates.

Component (Mg/100g)	FW –Larvae	BW-Larvae	KW-Larvae
Lead	$0.027^{a}\pm0.001$	$0.024^{b}\pm0.001$	$0.018^{c}\pm0.002$
Cadmium	$0.012^{b}\pm0.002$	$0.028^{a}\pm0.006$	$0.0063^{c}\pm0.0012$
Chromium	$0.011^{a}\pm0.0012$	$0.013^{a}\pm0.002$	$0.005^{b}\pm0.001$
Thallium	ND	ND	ND
Arsenic	ND	ND	ND

Values represent heavy metal levels in ppm. FW: Fruit Waste, BW: Breweries Waste, KW: Kitchen Waste, ND: Not Detected.

Table 3: Chemical Composition of the Three Types of Substrates as Compared to BSF Larvae.

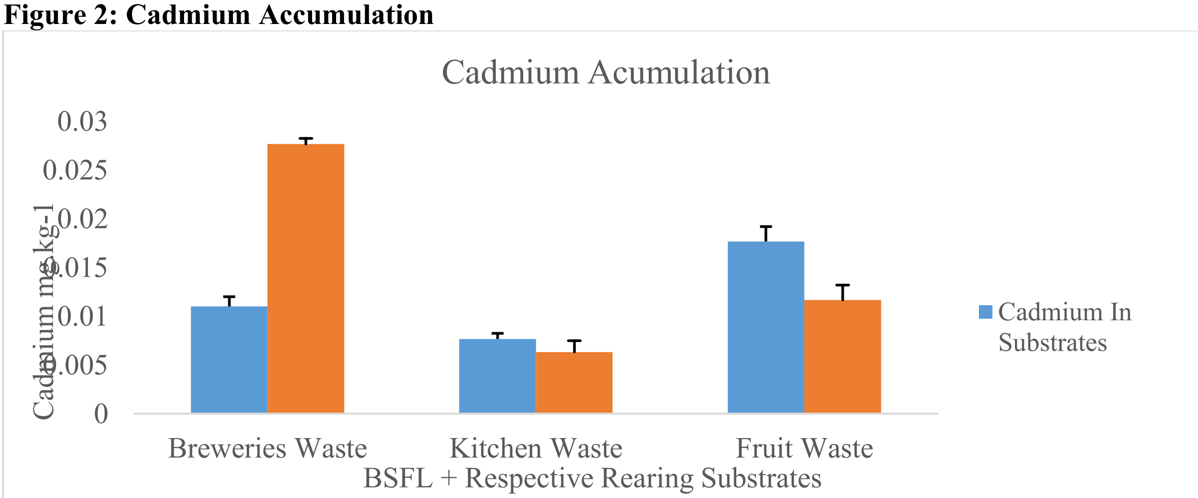
Component (Mg/100g)	FW	BW	KW	FW –Larvae	BW-Larvae	KW-Larvae
Lead	$0.033^{b}\pm0.003$	$0.048^{a}\pm0.0006$	$0.013^{c}\pm0.0025$	$0.027^{a}\pm0.001$	$0.024^{b}\pm0.001$	$0.018^{c}\pm0.002$
Cadmium	$0.018^{a}\pm0.002$	$0.011^{b}\pm0.001$	$0.008^{c}\pm0.0006$	$0.012^{b}\pm0.002$	$0.028^{a}\pm0.006$	$0.0063^{c}\pm0.0012$
Chromium	$0.03^{b}\pm0.001$	$0.102^{a}\pm0.002$	$0.015^{c}\pm0.0006$	$0.011^{a}\pm0.0012$	$0.013^{a}\pm0.002$	$0.005^{b}\pm0.001$
Thallium	ND	ND	ND	ND	ND	ND
Arsenic	ND	ND	ND	ND	ND	ND

Values represent heavy metal levels in ppm. FW: Fruit Waste, BW: Breweries Waste, KW: Kitchen Waste, FW –Larvae: Larvae reared on FW, BW-Larvae: Larvae reared on BW, KW-Larvae: Larvae reared on KW, ND: Not Detected.

Cadmium

Figure 2 shows results on Cd content in BSF larvae and their respective rearing substrates. The Cd content in feeding substrates ranged from 0.007 mg.kg^{-1} to 0.019 mg.kg^{-1} DW. In particular, the lowest Cd concentration was observed in the feeding substrate made from kitchen waste, whereas the highest concentration was found in the feeding substrate made from fruit waste. The levels of Cd in the larvae were not in the same order as the feeding substrates, which ranged from 0.005 mg.kg^{-1} to 0.028 mg.kg^{-1} in concentration. Larvae raised on brewery waste had a Cd content that was significantly higher (0.0277 mg.kg^{-1}) than larvae raised on other substrates ($P = 1.23e\text{-}06$), with kitchen waste having the lowest levels (0.0063 mg.kg^{-1}).

Figure 2: Cadmium Accumulation



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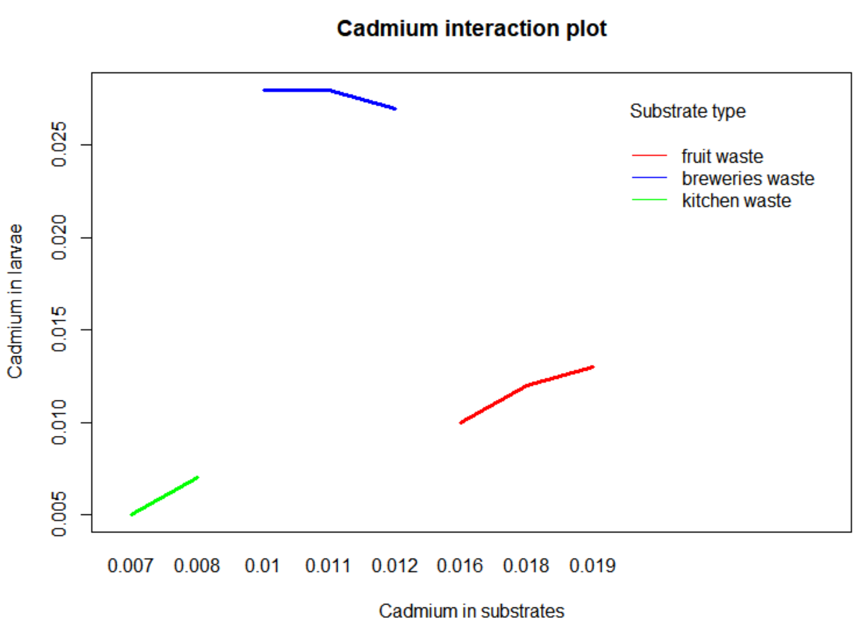
Furthermore, the ANOVA analysis (Table 4) revealed significant effects and interactions between "substrate type" used for rearing the BSF larvae and "cadmium levels in substrates" in relation to 'cadmium levels in larvae". It showed that the choice of the substrate type in this study significantly influenced cadmium levels in larvae ($F = 6656.00$, $p < 0.001$). Similarly, cadmium levels in substrates significantly affected cadmium accumulation in larvae ($F = 61.36$, $p = 0.00433$), underscoring the importance of monitoring and controlling cadmium levels in substrates in bioconversion processes. The significant interaction between these factors ($F = 39.82$, $p = 0.00692$) indicated that the impact of cadmium levels varied across different substrate types (Figure *3*).

Table 4: ANOVA Table for Chemical Composition of the Three Types of Substrates as Compared to BSF Larvae and Their Interaction Effect on Cadmium Content.

Id	Df	Sum Sq.	Mean Sq.	F-Value	P-Value	Significant
Substrate Type	2	0.0007396	0.0003698	6656.00	3.38e-06	***
Cadmium in Substrates	1	0.0000034	0.0000034	61.36	0.00433	**
Substrate Type: Cadmium in Substrates	2	0.0000044	0.0000022	39.82	0.00692	**
Residuals	3	0.0000002	0.0000001			
Signif. Codes		0 '***'	0.001 '**'	0.01 '*'	0.05 '.'	0.1 ' ' 1

*Significant effect on Cadmium content; F value= F statistic; p value: significance level

Figure 3: Cadmium Interaction Plot.



A Welch Two-Sample t-test between BSF larvae and feeding substrates (Table 5), for each treatment, showed a statistically significant mean difference for Cd content at 95% confidence level for brewery waste (p-value = 8.87e-05 with a t value = -25) with the highest mean (0.028 $mg.kg^{-1}$) obtained from larvae and for fruit waste (p-value = 0.0086 and t value = 4.8107) with the highest mean (0.018 $mg.kg^{-1}$) obtained from the feeding substrate. Kitchen waste treatment showed no significant difference in mean Cd content (p-value = 0.1734

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and t-value = 1.7889) between the larvae and the feeding substrate.

Table 5: A Welch Two-Sample t-test for Each Cadmium Treatment (between BSFL and Feeding Substrates).

Data	Cadmium ($mg.kg^{-1}$)	t- Value	DF	P-Value	Significance @ 95%	Interpretation
BW **BW – Larvae**	0.011 0.028	- 25	3.2	8.87e-05	<0.05	Significant
KW **KW – Larvae**	0.0077 0.0063	1.7889	2.9412	0.1734	0.05	Not Significant
FW **FW - Larvae**	0.018 0.0117	4.8107	4	0.008581	<0.05	Significant

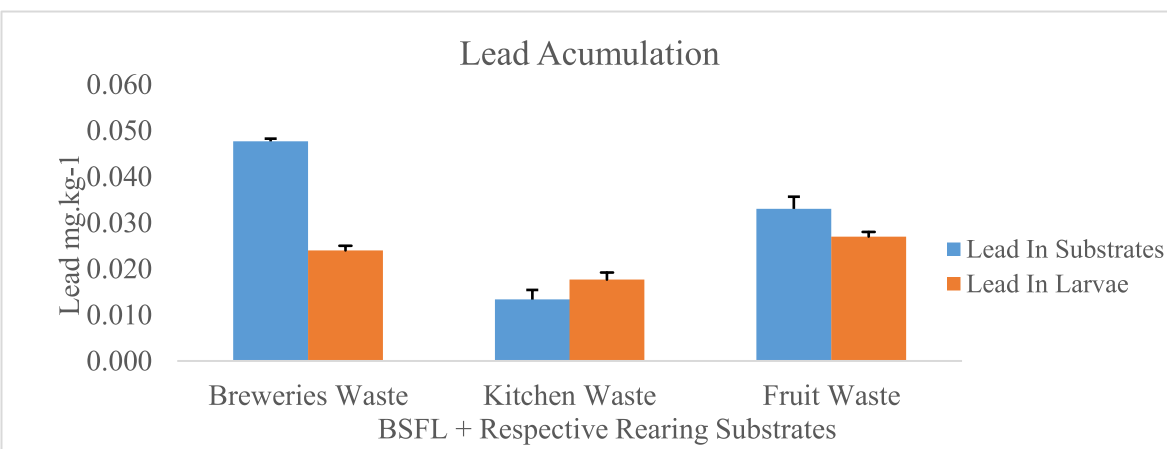
t-value = Statistical Measure; DF=Degrees of freedom; P-value = Statistical significance; < =significant; > = Not significant

Lead

Figure 4 shows results on Pb content in BSF larvae and their respective rearing substrates. Pb levels in feeding substrates varied from 0.011 $mg.kg^{-1}$ to 0.048 $mg.kg^{-1}$ DW, with a mean value averaging 0.031 $mg.kg^{-1}$. In particular, the lowest Pb concentration, 0.0133 ± 0.0021 $mg.kg^{-1}$, was found in the feeding substrate made from kitchen waste, while the highest concentration, 0.0477±0.0006 $mg.kg^{-1}$, was found in the substrate made from brewery waste.

Pb content in larvae (from 0.016 $mg.kg^{-1}$ to 0.028 $mg.kg^{-1}$ DW, with an average of 0.023 $mg.kg^{-1}$) was very low with respect to Pb content in the feeding substrates used, with the exception of kitchen waste, which was high in the larvae. The concentrations of Pb in larvae varied statistically significantly (P = 0.000214), with fruit waste showing the highest concentration (0.027 $mg.kg^{-1}$ dw) and kitchen waste showing the lowest concentration (0.018 $mg.kg^{-1}$).

Figure 4: Lead Accumulation



The ANOVA analysis results (Table 6) showed that the substrate type used for rearing BSF larvae had a significant impact on the lead content in larvae (F = 29.008, p = 0.0109), which indicates that different substrates contributed to the lead content in the larvae. However, the lead levels in substrates did not

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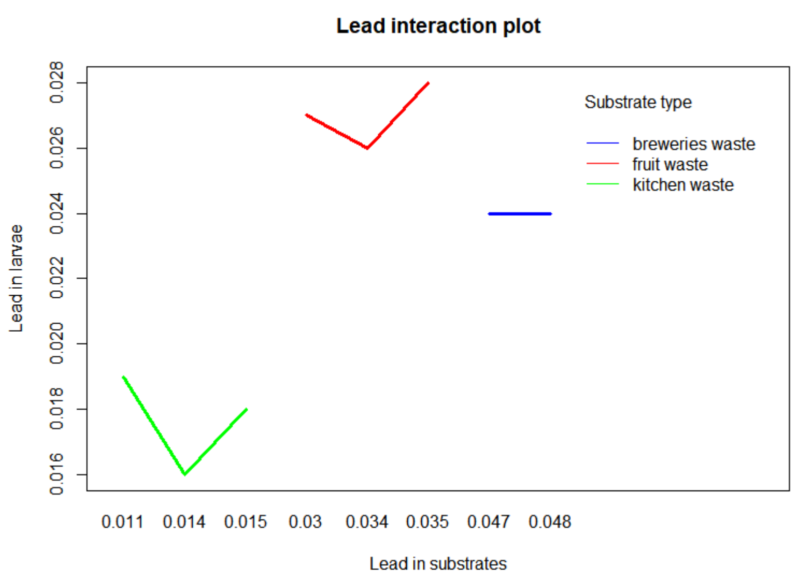
have a significant effect on the lead content in larvae (F = 0.130, p = 0.7425). Moreover, the interaction (Figure *5*) between substrate type and lead levels in substrates did not significantly affect the lead content in BSF larvae (F = 0.281, p = 0.7732).

Table 6: ANOVA Table for Chemical Composition of the Three Types of Substrates as Compared to BSF Larvae and Their Interaction Effect on Lead Content.

Id	Df	Sum Sq.	Mean Sq.	F-Value	P-Value	Significant
Substrate Type	2	1.362e-04	6.811e-05	29.008	0.0109	*
Lead in Substrates	1	3.000e-07	3.000e-07	0.130	0.7425	
Substrate. Type: Lead in Substrates	2	1.320e-06	6.600e-07	0.281	0.7732	
Residuals	3	7.040e-06	2.350e-06			
Signif. Codes	0 ‘***’	0.001 ‘**’		0.01 ‘*’	0.05 ‘.’	0.1 ‘ ’ 1
*Significant effect on Lead content; F value= F statistic; p value: significance level						

Figure 5: Lead Interaction Plot.



Additionally, a Welch Two Sample t-test for each treatment (between BSFL and feeding substrates) (Table 7) revealed a statistically significant mean difference for Pb content at a 95 percent confidence level for brewery waste (p-value = 2.897e-05 with a t value of 35.5) and fruit waste (p-value = 0.04532 and t value of 3.7) with the highest mean (0.048 mg.kg-1) and (0.033 mg.kg-1) obtained from the feeding substrate, respectively. The highest mean (0.018 mg.kg-1) was obtained from the larvae in the kitchen waste treatment, which had a significantly different Pb content (p-value = 0.04865 and t-value =-2.9).

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Table 7: A Welch Two-Sample t-test for Each Lead Treatment (between BSFL and Feeding Substrates).

Data	Lead (mg.kg^{-1})	t- Value	DF	P-Value	Significance @ 95%	Interpretation
BW **BW – Larvae**	0.048 0.024	35.5	3.2	2.897e-05	<0.05	Significant
KW **KW – Larvae**	0.013 0.018	-2.9	3.7	0.04865	<0.05	Significant
FW **FW – Larvae**	0.033 0.027	3.7	2.56	0.04532	<0.05	Significant

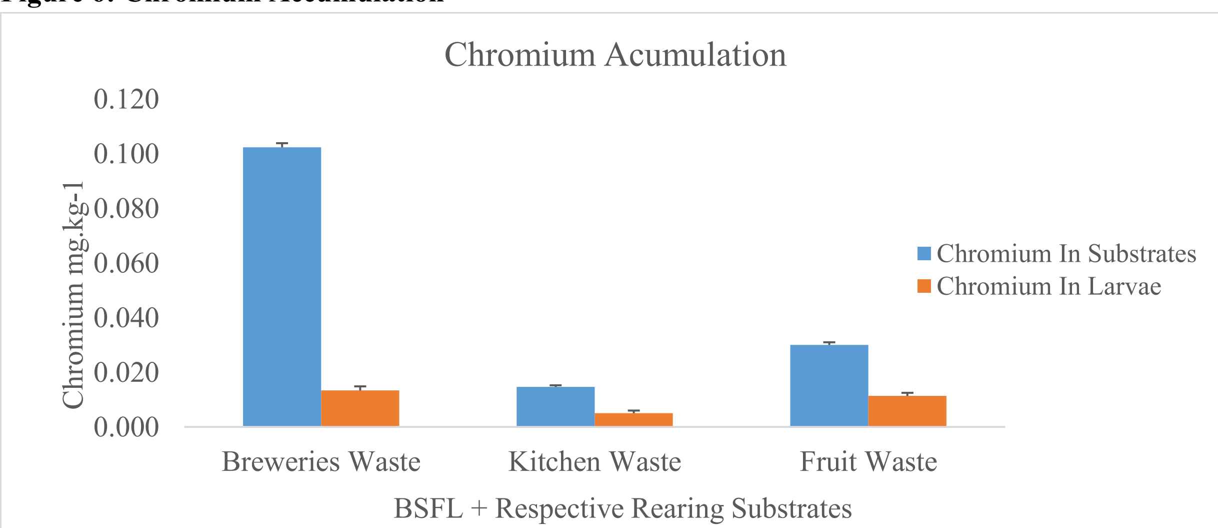
t-value = Statistical Measure; DF=Degrees of freedom; P-value = Statistical significance; < =significant; > = Not significant

Chromium

Figure 6 shows results on Cr content in BSF larvae and their respective rearing substrates. The Cr content in feeding substrates ranged from 0.014 mg.kg^{-1} to 0.104 mg.kg^{-1} DW, with an average of 0.049 mg.kg^{-1}. In particular, brewery feeding substrate showed the highest statistically significant difference (P = 1.73e-10) Cr content of 0.102±0.0015 mg.kg^{-1}, whereas kitchen waste substrate showed the lowest Cr concentration of 0.015±0.0006 mg.kg^{-1}.

In larvae, Cr content (from 0.004 mg.kg^{-1} to 0.015 mg.kg^{-1} DW, with an average of 0.01 mg.kg^{-1}) was very low with respect to Cr content in feeding substrates. Larvae reared on various substrates showed statistically significant differences (P = 0.000438) in Cr content, with brewery waste having the highest Cr content and kitchen waste having the lowest concentration of (0.0133 mg.kg^{-1} dw) and (0.001 mg.kg^{-1}), respectively. The results of a Fisher-LSD test (0.0025) at 0.05 alpha revealed that the levels of chromium in fruit waste and brewery waste treatment were statistically equivalent.

Figure 6: Chromium Accumulation



The ANOVA analysis (Table 8) of chromium content in BSF larvae showed significant effects and interactions. Specifically, the substrate type used for rearing the larvae had a highly significant influence on chromium content in the larvae (F = 146, p = 0.00103), which underscored the undeniable impact of the chosen type of substrate for larvae rearing on chromium accumulation in the

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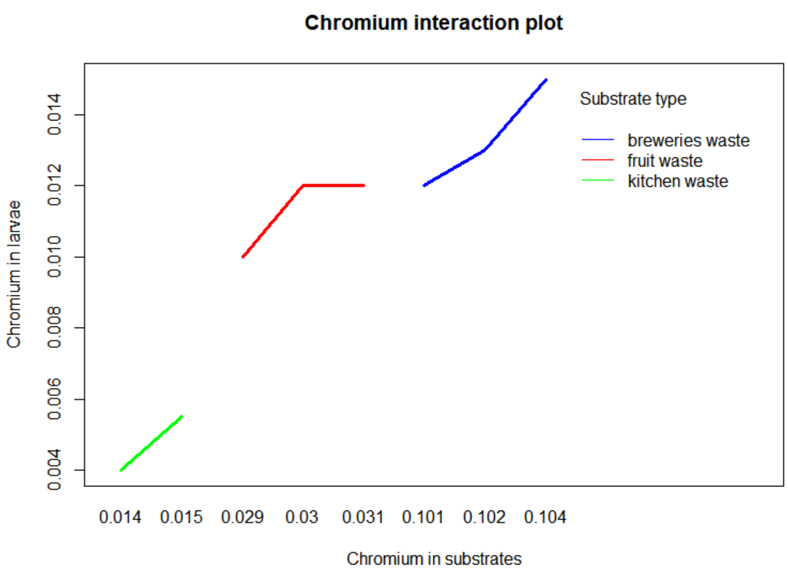
larvae. On the other hand, the larvae's chromium content was not significantly affected by the "chromium levels in substrates" (F = 20.610, p = 0.02001). The interaction (Figure 7) between substrate type and chromium levels in substrates was also not significant (F = 0.195, p = 0.83264).

Table 8: ANOVA Table for Chemical Composition of the Three Types of Substrates as Compared to BSF Larvae and Their Interaction Effect on Chromium Content.

Id	Df	Sum Sq.	Mean Sq.	F-Value	P-Value	Significant
Substrate Type	2	1.136e-04	5.678e-05	146.000	0.00103	**
Lead in Substrates	1	8.020e-06	8.020e-06	20.610	0.02001	*
Substrate Type: Lead in Substrates	2	1.500e-07	8.000e-08	0.195	0.83264	
Residuals	3	1.170e-06	3.900e-07			
Signif. Codes	0 '***'	0.001 '**'		0.01 '*'	0.05 '.'	0.1 ' ' 1
*Significant effect on Chromium content; F value= F statistic; p value: significance level						

Figure 7: Chromium Interaction Plot.



Welch Two-Sample t-test (Table 9) for each treatment (between BSFL and feeding substrates) showed a statistical mean difference for Cr content at 95% confidence level for brewery waste (p-value = 2.311e-07 with a t value = 71.4) with the highest mean (0.102 mg.kg-1) obtained from the feeding substrate and for fruit waste (p-value = 3.452e-05, and t value = 21.2) with the highest mean (0.03 mg.kg-1) obtained from the feeding substrate. The Cr content in kitchen waste treatment showed a significant difference (p-value = 0.0005004 and t-value = 14.5), with the feeding substrate having the highest mean (0.015 mg.kg-1).

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Table 9: A Welch Two-Sample t-test for Each Chromium Treatment (between BSFL and Feeding Substrates).

Data	Chromium ($mg.kg^{-1}$)	t-Value	DF	P-Value	Significance @ 95%	Interpretation
BW **BW – Larvae**	0.102 0.013	71.4	4	2.311e-07	<0.05	Significant
KW **KW – Larvae**	0.005 0.015	14.5	3.2	0.0005	<0.05	Significant
FW **FW - Larvae**	0.03 0.0113	21.2	3.92	3.452e-05	<0.05	Significant

t-value = Statistical Measure; DF=Degrees of freedom; P-value = Statistical significance; < =significant; > = Not significant

Bioaccumulation Factor

When assessing feed conversion, the buildup of chemicals in the larvae is of serious importance. Again, as far as the applications of the larvae go, the accumulation of desirable nutrients could be advantageous, whereas the accumulation of undesirable nutrients, such as heavy metals, is undesirable. On a dry matter (DM) basis, a bio-accumulation factor (BAF) greater than 1 indicates that the element from the feeding substrate has been bio-accumulated by the insect. It was calculated as follows:

Equation 2:

BAF = Concentration of heavy metal in the organism (DM) / Concentration of heavy metal in the given feed (DM).

Equation 2)

Table 9 displays the bioaccumulation factor (BAF) of the three heavy metals in BSFL from the three different feeds. Arsenic and Thallium are left out of Table 1 and Table 2, because there was no detection of them in the Larvae and feeding substrates; therefore, no bioaccumulation. The Values highlighted in bold are BAFs that are greater than 1, which indicates that the larvae accumulated the elements from the feeding substrates. In this study, Pb bioaccumulated only in larvae reared on Kitchen waste (1.3±0.7) and did not bioaccumulate in the Larvae samples reared on Breweries and Fruit waste substrates, while Cd accumulated in larvae reared on Brewery waste (2.5±0.6) and not in the larvae reared on Fruit waste and Kitchen waste. There was no accumulation of Cr in any of the Larvae samples (Table 9).

Table 10: Bioaccumulation Factor (BAF) for Heavy Metals for BSF Larvae Reared on the Three Types of Feeding Substrates. Calculated on a Dry Weight Basis.

Component (Mg/100g)	BSFL – FW	BSFL – BW	BSFL - KW
Lead	0.8±0.4	0.5±1.7	**1.3±0.7**
Cadmium	0.7±1.0	**2.5±0.6**	0.8±2.0
Chromium	0.5±1.2	0.1±1.0	0.3±1.7

Values represent BAF levels for heavy metal levels in BSFL. BSFL-FW: Larvae reared on FW, BSFL-BW: Larvae reared on BW, BSFL-KW: Larvae reared on KW.

DISCUSSION

Chemical Contaminants

Cadmium

Cadmium (Cd) is, without doubt, one of the highly toxic, carcinogenic heavy metals that pose serious health risks to people when exposed to it. The Cd concentrations in BSF larvae in this

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assessment (0.005 to 0.028 mg.kg^{-1}) were incredibly low compared to those in earlier studies ***(0.11- 0.24)*** (Truzzi et al., 2020***;*** Bessa et al., 2021), which could be explained by the lower Cd concentrations of the feeding substrates used in this study. The Cd content of feeding substrates and larvae did not significantly correlate with one another (r = 0.0653, p = 0.8675). As a result, the Cd content of the feeding substrates had no significant effect on the Cd content of the larvae.

The BAF of Cd (0.7–2.5) of BSF larvae in this study was actually lower as compared to previous studies (2.5–12.27) (Schmitt et al., 2019***;*** Bessa et al., 2021; Meyer et al., 2021). Despite having a lower BAF, Cd displayed higher BAF values across all treatments than the other heavy metals, with brewery waste having the highest Cd BAF value (2.5) (Table *10*). Even though the Cd bioaccumulation in the larvae raised on fruit and kitchen waste was below one, indicating that they did not bioaccumulate in the larvae, it is clear that Cd is highly accumulative (Schmitt et al., 2019; Bessa et al., 2021). The large number of calcium channels in the guts of BSF larvae contributes to the high uptake of cadmium, which results in higher cadmium BAF values than for other heavy metals (Diener et al., 2015; Bessa et al., 2021). As was the case in this study, these discrepancies were not present in other studies where Cd bioaccumulated from all feeds used (Bessa et al., 2021***;*** Truzzi et al., 2020). However, more research is needed to better understand the factors that affect or influence bioaccumulation, as this will help explain why it accumulates from some feeds but not from others. Cd content in tested BSF Larvae was from 0.0063 to 0.03 mg.kg^{-1}, which was lower than the legal limit (Table *11*) as indicated by KEBS-DKS 2922-2:2020| Second |Edition of edible insects and EFSA, (2012) a scientific report on cadmium dietary exposure in the European population. Then, tested larvae from all the treatments were safe from the point of view of Cd content in animal feed (EFSA, 2012; European Commission, 2013; CXS 193-1995, 2019; KEBS, 2020).

Table 11: Heavy Metals Content in BSF Larvae and Comparison with Legal Limit for Food in KEBS, 2020, (CXS 193-1995, 2019) EFSA Journal, 2020 and European Commission, 2013

Component(mg.kg^{-1})	BSFL-FW	BSFL-BW	BSFL-KW	Legal Limit			
				KEBS	CXS	EFSA	EC
Lead	0.027^{a}±0.001	0.024^{b}±0.001	0.018^{c}±0.002	0.5	0.1, 0.2	--	5
Cadmium	0.012^{b}±0.002	0.028^{a}±0.006	0.0063^{c}±0.0012	0.1	0.1, 0.2	2.5	0.5
Chromium	0.011^{a}±0.0012	0.013^{a}±0.002	0.005^{b}± 0.001	--	--	0.4	0.5
Thallium	ND	ND	ND				
Arsenic	ND	ND	ND				

Values represent heavy metal levels in ppm. BSFL-FW: Larvae reared on Fruit Waste, BSFL-BW: Larvae reared on Brewery Waste, BSFL-KW: Larvae reared on Kitchen Waste, ND: Not Detected.

Lead

Lead is an environmental contaminant that arises both naturally and, to a greater extent, as a result of human activity. Lead exposure in humans primarily comes from food, but it can also be found in water, air, soil, and dust (EFSA, 2010).

Lead levels in the larvae in this study varied between 0.016 and 0.028 mg.kg-1, which was inconsistent with earlier findings. This might be because the feeding substrates used in the current study had lower Pb concentrations than those used in earlier research (Truzzi et al., 2020). There was a strong correlation between the Pb content of the feeding substrates and the larvae ($r = 0.693$, $p = 0.0385$), unlike Truzzi et al. (2020), who experienced no correlation between the Pb in the feeding substrates and the BSF Larvae ($r = 0.04352$, $p = 0.9115$). The Pb content of the feeding substrates in this study, consequently, had a sizable impact on the Pb content of the larvae, as also demonstrated by Diener et al. (2015).

Bioaccumulation of Pb did not occur in all but one sample (BSFL fed on kitchen waste, BAF 1.3), with BAF ranging from 0.5–0.8. Previous studies had similar BAF ranges of 0.03–0.99 (Diener et al., 2015; Meyer et al., 2021). While other studies had a higher BAF range of 1.11–3.2 (Purschke et al., 2017; Truzzi et al., 2020; Bessa et al., 2021; Meyer et al., 2021), in this study, the BAF for larvae fed on kitchen waste can be explained due to the nature of the feeding substrates, which are prone to lead contamination exposure (EFSA, 2010; EFSA, 2010; Purschke et al., 2017). According to KEBS-DKS (2020) Second Edition of Edible Insects, CODEX STAN 193-1995, (2019) and European Commission (2013), the tested Pb content in BSF larvae was between 0.016 and 0.028 mg.kg1, which was less than the permissible limit (Table *11*). The tested larvae from all the treatments were then found to be safe in terms of the amount of Pb in the animal feed (European Commission, 2013; CXS 193-1995, 2019; KEBS, 2020).

Chromium

Chromium is another necessary nutrient in the diet, which is also used as a feed supplement to boost animal immunity. However, when consumed in amounts above 200 mg at a time, chromium can be hazardous and have mutagenic and genotoxic effects (Gao et al., 2017).

In this study, the Cr levels in the larvae were too low to raise any safety concerns, with Cr ranges of 0.004 to 0.015 mg.kg1 compared to legal limits (Table *11*). These values were too low compared to those of the previous studies (due to the low Cr concentration in the feeding substrates), which ranged from 0.22 to 0.52 $mg.kg^{-1}$ but were close to the larvae reared on the uncontaminated feeding substrate of 0.064 $mg.kg^{-1}$ (Bessa et al., 2021; Kuppusamy et al., 2020). This study finds a significant correlation between the Cr contents in the feeding substrates and the larvae ($r = 0.771$, $p = 0.0151$), indicating that the Cr contents of the feeding substrates had a significant effect on the Cr content of the larvae.

All the bioaccumulation values in this study were less than one (range 0.1–0.5) in all the treatments (Table *10*), indicating that Cr did not bioaccumulate in the larvae. This supports earlier findings from studies in which Cr did not bioaccumulate (Gao et al., 2017), although Bessa et al. (2021) discovered that there was low bioaccumulation in some of the treatments (1.2). According to this study's findings regarding the content of Cr in animal feed (Table 17), all of the tested larvae from all treatments were safe (EFSA Journal, 2020; European Commission, 2013).

CONCLUSION AND RECOMMENDATIONS

Conclusions

The study was conducted to investigate whether BSF larvae reared on different substrates are safe from the accumulation of heavy metals for use in animal feeds. The study found that the feeding substrate influenced the heavy metal accumulation. The results show that cadmium and lead are the most concerning heavy metals in terms of potential bioaccumulation, and their levels in the larvae varied based on the feeding substrates used. While the Cd concentrations in the larvae were incredibly low, the BAF values were higher than those of other heavy metals, indicating that Cd is highly accumulative. In contrast, Pb bioaccumulation did not occur in all except for Kitchen waste, with a BAF value of 1.3±0.7. The tested larvae from all the treatments were found to be safe from the point of view of Cd, Cr, Ar, TI and Pb content in animal feed, as they were below the permissible limit. However, it is important to note that these findings

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do not mean that all BSF larvae raised on different substrates will be safe for animal feed consumption, as the levels of heavy metal accumulation can vary based on the specific feeding substrate used.

Recommendations

The findings of this study highlight the potential risks and benefits of using black soldier fly larvae (BSFL) as a source of food and feed. While BSFLs are a nutrient-rich and sustainable alternative to conventional protein sources, such as fishmeal and soybean meal, they can also accumulate heavy metals, which can pose a risk to human and animal health.

Therefore, it is recommended that food safety regulations and guidelines be strictly followed during the rearing and processing of BSFLs, particularly with regard to heavy metal content. Measures such as regular testing for heavy metals, adopting proper processing methods, proper handling, storage, and processing of raw materials should be implemented to minimise the risk of accumulation.

Additionally, there is a need for further research to be conducted to better understand the factors that influence the bioaccumulation of heavy metals in BSFLs, particularly for highly toxic and carcinogenic metals such as cadmium (Cd) and lead (Pb). Strategies to mitigate heavy metal accumulation in BSFLs, such as using low-heavy-metal substrates like kitchen and fruit waste instead of brewery waste, should also be explored. Furthermore, continued monitoring of heavy metal levels in BSFLs should be conducted to ensure compliance with regulatory limits and identify any emerging risks. This is particularly important for vulnerable populations, such as infants and children, who are more susceptible to the potential health risks associated with exposure to environmental contaminants like Pb.

By ensuring the safety and quality of BSFLs, their use as a source of food and feed can be a viable and sustainable strategy to improve the nutritional status of populations and reduce the environmental impact of conventional livestock production. However, this will require comprehensive and standardised methods for assessing their safety and quality, as well as addressing the regulatory and cultural barriers to their adoption. This thesis can contribute to the ongoing discourse on the potential of insects as food and feed, as well as to the development of evidence-based policies and practices that promote their safe and sustainable use.

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REFERENCES

Bessa, L. W., Pieterse, E., Marais, J., Dhanani, K., & Hoffman, L. C. (2021). Food safety of consuming black soldier fly (Hermetia illucens) larvae: Microbial, heavy metal and cross-reactive allergen risks. *Foods*, *10*(8). https://doi.org/10.3390/FOODS10081934

Bouzari, A., Holstege, D., & Barrett, D. M. (2015). Mineral, Fiber, and Total Phenolic Retention in Eight Fruits and Vegetables: A Comparison of Refrigerated and Frozen Storage. *Journal of Agricultural and Food Chemistry*, *63*(3), 951–956. https://doi.org/10.1021/JF504890K

Cullere, M., Tasoniero, G., Giaccone, V., Acuti, G., Marangon, A., & Zotte, A. D. (2018). Black soldier fly as dietary protein source for broiler quails : meat proximate composition, fatty acid and amino acid profile, oxidative status and sensory traits. *Animal, The International Journal of Animal Biosciences*, *12*(3), 640–647. https://doi.org/10.1017/S1751731117001860

CXS 193-1995. (2019). *Codex Alimentarius Commission. General Standard for Contaminants and Toxins in Food and Feed*

Article DOI : https://doi.org/10.37284/eajab.8.2.3541

CXS 193-1995. Adopted in 1995, revised in 1997, 2006, 2008, 2009, amended in 2010, 2012, 2013, 2014, 2015, 2016, 2017, 2018, 2019. https://www.fao.org/fao- who- codexalimentarius/sh- proxy/en/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252FStandards%252FCXS%2B193-1995%252FCXS_193e.pdf

Diener, S., Zurbrügg, C., & Tockner, K. (2015). Bioaccumulation of heavy metals in the black soldier fly, Hermetia illucens and effects on its life cycle. *Journal of Insects as Food and Feed*, *1*(4), 261– 270. https://doi.org/10.3920/JIFF2015.0030

EFSA. (2010). EFSA Panel on Contaminants in the Food Chain (CONTAM); Scientific Opinion on Lead in Food. EFSA Journal 2010; 8(4):1570. [151 pp.]. doi:10.2903/j.efsa.2010.1570. *EFSA Journal*, *8*(4), 1– 151. https://doi.org/10.2903/j.efsa.2010.1570

EFSA. (2012). *European Food Safety Authority, SCIENTIFIC REPORT OF EFSA Cadmium dietary exposure in the European population 1*. *10*(1), 1– 37. https://doi.org/10.2903/j.efsa.2012.2551.

EFSA Journal. (2020). EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), Bampidis V, Azimonti G, Bastos ML, Christensen H, Dusemund B, Kos Durjava M, Kouba M, L_opez-Alonso M, L_opez Puente S, Marcon F, Mayo B, Pechov_a A, Petkova M, Ra. *EFSA Journal*, *18*(2). https://doi.org/10.2903/J.EFSA.2020.6026

European Commission. (2013). Commission Regulation (EU) No 1275/2013 of 6 December 2013 amending Annex I to Directive 2002/32/EC of the European Parliament and of the Council as regards maximum levels for arsenic, cadmium, lead, nitrites, volatile mustard oil and harmful botanical im. *Official Journal of the European Union*, *328*(1275), 86–92. https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32013R1275&from=EN

Ewusie, E. A., Kwapong, P. K., Ofosu-Budu, G., Sandrock, C., Akumah, A. M., Nartey, E. K., Tetegaga, C., & Agyakwah, S. K. (2019). The black soldier fly, Hermetia illucens (Diptera: Stratiomyidae): Trapping and culturing of wild colonies in Ghana. *Scientific African*, *5*, 134. https://doi.org/10.1016/j.sciaf.2019.e00134

Gao, Q., Wang, X., Wang, W., Lei, C., & Zhu, F. (2017). *Influences of chromium and cadmium on the development of black soldier fly larvae*. *March*. https://doi.org/10.1007/s11356-017-8550-3

Henchion, M., Hayes, M., Mullen, A. M., Fenelon, M., & Tiwari, B. (2017). *Future Protein Supply and Demand: Strategies and Factors Influencing a Sustainable Equilibrium*. https://doi.org/10.3390/foods6070053

KEBS. (2020). *DRAFT KENYA STANDARD Edible Insects Part 2 : Products containing Specification edible insects —. 2*.

Kulkarni, N. M. (2018). Citation of this article Nitin Madhukar Kulkarni. Determination of Heavy Metals in Animal Feed by Inductive Coupled Plasma-Optical Emission Spectrometry (ICP-OES). *Int. Arch. App. Sci. Technol*, *9*, 58–61. https://doi.org/10.15515/iaast.0976-4828.9.4.5861

Kuppusamy, G., Kong, C. K., Segaran, G. C., Tarmalingam, E., Herriman, M., Ismail, M. F., Khan, T. M., Low, L. E., & Goh, B. H. (2020). Hummingbird-leaves-reared black soldier fly prepupae: Assessment of nutritional and heavy metal compositions. *Biology*, *9*(9), 1–11. https://doi.org/10.3390/BIOLOGY9090274

Lievens, S., Poma, G., De Smet, J., Van Campenhout, L., Covaci, A., & Van Der

Borght, M. (2021). Chemical safety of black soldier fly larvae (Hermetia illucens), knowledge gaps and recommendations for future research: a critical review. *Journal of Insects as Food and Feed*, *February*, 1–14. https://doi.org/10.3920/jiff2020.0081

Liu, X., Chen, X., Wang, H., Yang, Q., Ur Rehman, K., Li, W., Cai, M., Li, Q., Mazza, L., Zhang, J., Yu, Z., & Zheng, L. (2017). Dynamic changes of nutrient composition throughout the entire life cycle of black soldier fly. *PLoS ONE*, *12*(8). https://doi.org/10.1371/journal.pone.0182601

Meyer, A. M., Meijer, N., Hoek-van den Hil, E. F., & van der Fels-Klerx, H. J. (2021). Chemical food safety hazards of insects reared for food and feed. *Journal of Insects as Food and Feed*, *7*(5), 823– 831. https://doi.org/10.3920/jiff2020.0085

Ngonga, C. A., Gor, C. O., Okuto, E. A., & Ayieko, M. A. (2021). Growth Performance of Acheta Domesticus and Gryllus Bimaculatus Production Reared under Improvised Cage System for Increased Returns and Food Security. *Journal of Insects as Food and Feed*, *7*(3), 301–310. https://doi.org/10.3920/JIFF2020.0082

Nguyen, T. T. X., Tomberlin, J. K., & Vanlaerhoven, S. (2013). Influence of Resources on Hermetia illucens (Diptera: Stratiomyidae) Larval Development. *Journal of Medical Entomology*, *50*(4), 898–906. https://doi.org/10.1603/ME12260

Nyakeri, E. M., Ogola, H. J., Ayieko, M. A., & Amimo, F. A. (2016). An open system for farming black soldier fly larvae as a source of proteins for smallscale poultry and fish production. *Https://Doi.Org/10.3920/JIFF2016.0030*, *3*(1), 51– 56. https://doi.org/10.3920/JIFF2016.0030

Purschke, B., Scheibelberger, R., Axmann, S., Adler, A., & Jäger, H. (2017). *Impact of substrate contamination with mycotoxins, heavy metals and pesticides on the growth performance and composition of black soldier fly larvae (Hermetia illucens) for use in the feed and food value chain*. https://doi.org/10.1080/19440049.2017.1299946

Samtiya, M., Aluko, R. E., & Dhewa, T. (2020). Plant food anti-nutritional factors and their reduction strategies: an overview. *Food Production, Processing and Nutrition 2020 2:1*, *2*(1), 1–14. https://doi.org/10.1186/S43014-020-0020-5

Schmitt, E., Belghit, I., Johansen, J., Leushuis, R., Lock, E.-J., Melsen, D., Kathirampatti, R., Shanmugam, R., Loon, J. Van, & Paul, A. (2019). Growth and Safety Assessment of Feed Streams for Black Soldier Fly Larvae: A Case Study with Aquaculture Sludge. *Animals*, *9*, 189. https://doi.org/10.3390/ani9040189

Shumo, M., Osuga, I. M., Khamis, F. M., Tanga, C. M., Fiaboe, K. K. M., Subramanian, S., Ekesi, S., van Huis, A., & Borgemeister, C. (2019). The nutritive value of black soldier fly larvae reared on common organic waste streams in Kenya. *Scientific Reports 2019 9:1*, *9*(1), 1–13. https://doi.org/10.1038/s41598-019-46603-z

Siemer, D. D. (2018). Atomic Absorption Spectrometry. *Materials Characterization*, 43–59. https://doi.org/10.31399/asm.hb.v10.a0001730

The County Government of Siaya. (2023). *Department of Water, Sanitation, Environment, Natural Resources and Climate Change, Siaya County Climate Change Action Plan (2023-2028). The County Government of Siaya, Republic of Kenya.*

Truzzi, C., Annibaldi, A., Girolametti, F., Giovannini, L., Riolo, P., Ruschioni, S., Olivotto, I., & Illuminati, S. (2020). A

Article DOI : https://doi.org/10.37284/eajab.8.2.3541

chemically safe way to produce insect biomass for possible application in feed and food production. *International Journal of Environmental Research and Public Health*, *17*(6). https://doi.org/10.3390/ijerph17062121

Tschirner, M., & Simon, A. (2015). Influence of different growing substrates and processing on the nutrient composition of black soldier fly larvae destined for animal feed. *Journal of Insects as Food and Feed*, *1*(4), 249–259. https://doi.org/10.3920/JIFF2014.0008

United Nations. (2019). World population projected to reach 9.8 billion in 2050, and 11.2 billion in 2100. *UN Department of Economic and Social Affairs*, 8–12.

Vaus, D. de. (2006). *Research Design: Sage benchmarks in social research methods.* 358. https://catalogue.nla.gov.au/catalog/3308406

Woods, M. J., Hoffman, L. C., & Pieterse, E. (2019). *Artificial diets for neonatal black soldier fly (Hermetia illucens) larvae*. *5*(2), 99–105. https://doi.org/10.3920/JIFF2018.0028