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EXPLOITATION OF LOCALLY AVAILABLE ORGANIC WASTES FOR REARING BLACK SOLDIER FLY (*Hermetia illucens*) LARVAE AS A POTENTIAL PROTEIN INGREDIENT FOR POULTRY AND FISH FEEDS IN UGANDA

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AUTHORS' CONTRIBUTIONS
This work was carried out in collaboration among all authors. Author OH designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author CM managed the analyses of the study. Author OR managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

The animal industry especially aquaculture and poultry sectors contribute greatly to the livelihoods of households in sub-Saharan African countries. The ongoing depletion of fish stocks used as protein source requires substitution with alternative animal sources, e.g. black soldier fly (BSF) larvae, *Hermetia illucens*. Hence, this study assessed the growth performance of BSF larvae reared on locally and readily available organic wastes. Five organic waste types/substrates (cattle and swine dropping, chicken house rearing waste, decomposing avocado and millet brew wastes) were tested. Randomly selected third larval instar of BSF from each rearing substrate were measured: girth (in cm), length (in cm) and weight (in grams) using a vernier calliper and a weighing scale respectively. The findings indicate that in the 12 weeks period, millet brew wastes produced larvae with the widest girth, longest length and were heaviest, followed by decomposing avocado, waste from chicken rearing house, cattle and swine droppings. In conclusion, the growth performance of BSF larvae was influenced by substrates investigated. We recommend further studies to determine the nutritional composition of BSF larvae raised on each of the substrates investigated in this study.

Keywords: Black soldier fly; feeds; fish; *Hermetia illucens*; organic waste; poultry.

1. INTRODUCTION

Globally, animal industry especially poultry and aquaculture sector employ an estimated 1.3 billion people [1]. Additionally, livelihoods of approximately 600 million poultry and aquaculture farmers are directly supported by the sector [1,2]. Of these, 200 million are from sub-Saharan Africa [1]. Currently in Uganda, aquaculture is mainly practised using earthen ponds, cages and tanks. Throughout the country, there are approximately 25,000 ponds covering 10,000 hectares of land producing 15,000 tonnes of fish annually by about 12, 000 fish farmers [3].

Poultry and aquaculture sectors play important roles in human nutrition. For example, both poultry and fish provide a rich source of protein, whereas fish also supplies long-chain omega 3 fatty acids,

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eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) which are essential for optimal brain and neural system development in children [4,5]. Despite the positive contribution of poultry and aquaculture sectors in sub-Saharan Africa, feed constitutes 60-70% of the total production cost with protein being the most expensive component [6,7].

In Uganda, dried silver cyprinid (*Rastrineobola argentea*: common names: mukene and wanyjiri for Bantu and Luo speakers respectively), an indigenous pelagic fish species mainly available in Lake Victoria, is used as a protein source in fish and poultry feeds [6]. However, there is an increasing demand for *R. argentea* for human consumption and as a result, their catches are expected to reduce due to overfishing which may negatively affect fish and poultry sectors and indeed the entire animal industry [8].

With the current 3.2% natural increase of human population in Uganda, by 2050, the country is projected to have 95.6 million people from the 44.1 million in 2018 [9] hence, increased demand for animal protein will be inevitable. However, the unpredictable nature of climate change in sub-Saharan Africa may negatively impact on the pace of production of fish and poultry feeds in most African countries. Other sources of protein such as soybean and fish meal are unsustainable for many local poultry and fish farmers due to their high prices. There is need to adopt a low cost locally available alternative such as black soldier fly (BSF: *Hermetia illucens* L.; Diptera: Stratiomyidae) as an alternative protein source for poultry and fish feeds.

Several studies [10,11,12,13] have reported that BSF larvae convert organic waste into 42% crude protein and 29% fats, depending on the substrate type on which it is reared. Further, results from elsewhere [14,15] show that fish and chicken grow faster and are healthier with high quality meat when fed on BSF larval diet. Uganda is still at infant stages of exploring BSF larvae as an alternative protein source for fish and poultry feeds. Therefore, it important to identify and characterise a variety of locally and readily available organic substrates that can be used for rearing BSF and establish the best performing substrate for continuous mass production of BSF. In Uganda, the best locally and readily available organic substrate for rearing BSF larvae is unknown. This study assessed the growth performance of BSF larvae reared on five locally and readily available organic substrates.

2. MATERIALS AND METHODS

2.1 Establishment of the Starter Colony

The field experiments for this study were conducted in Koro Sub-county, Omoro district while chemical and nutritional properties of substrates were determined at Uganda National Bureau of Standards laboratory between August-December 2017. To establish the starter colony, five kilograms of kitchen waste were used in each of ten plastic buckets (five-litre capacity). In each bucket, ten card boxes with hollow crevices were cut (1 cm^2) and randomly placed vertically on the substrates with their hollow crevices facing upward. Two buckets with their contents were placed at each randomly selected domestic garbage collection points in the morning (0900 hrs) for the female gravid BSF to lay eggs in the hollow cardboard crevices. The set-ups were left to stand for six hours, then checked for presence or absence of BSF eggs. Card boxes with eggs in their hollow crevices were removed and taken for inoculation into the experimental substrates.

Fig. 1. a) Eggs of BSF and b) BSF larvae

2.2 Characterization of Experimental Substrates

Five locally and readily available organic substrates: bovine (cattle) dropping, decomposing avocado (*Persea americana*), swine droppings, waste from chicken rearing house (a mixture of feathers, sawdust and chicken faecal matter) and millet (*Eleusine coracana*) brew waste were each obtained from their single respective sources. Three replicates of ten kilograms of each organic substrate were inoculated in each 20-litre plastic grub tubs. In each tub, 400 eggs were inoculated in the substrate. The set up was left to stand for eggs to hatch to larvae and their growth monitored under natural conditions. The tubs were sheltered by translucent iron sheets to protect them from rainfall. Ten third instar larvae were randomly selected from each tub and their girth and length (cm), and weight (grams) measured using a vernier calliper and weighing scale (Mettler Toledo® XPR, Switzerland Instrument Corps) respectively.

2.3 Determination of Nutritional and Physico-Chemical Properties of the Substrates

For crude protein analysis, Feldsine, (2002) method was used [1]. This was achieved by weighing 0.5 g of each substrate and placing it in each test tube. Then, 5g of a catalyst (90% K_2SO_4 and 10% $CuSO_4$) was added into each test tube. For control, a blank test tube containing only the catalyst was used. Fifteen millilitres of concentrated sulphuric acid was then added and the mixture transferred into an acid hydrolyser for 3 hours. After digestion, each sample was titrated and the crude protein percentage content determined by multiplying the nitrogen content by 6.25.

For physical properties of substrate, APHA, [16] protocol was followed. To determine temperature, each thermometer was inserted into each replicate of substrate. On the other hand, pH was measured using a hand-held pH meter (pH-5012, Omega, UK). Measurements of temperature and pH were conducted in the morning (0800 hrs), midday (1200 hrs) and evening (1800 hrs) and the means of the recordings calculated. Ammonia (NH_4^+) was determined using the Aqualytic Ammonia VARIO kit (535650, Aqualytic, Germany) following the salicylate method.

2.4 Data Analysis

Data obtained were analysed using Predictive Analysis Software (PASW)/ SPSS version 21.0 for windows (SPSS Inc., Chicago, IL). Shapiro-Wilk tests for normality were performed followed by a non-parametric Kruskal Wallis test to compare growth performance of BSF larvae across substrate types. The same test (Kruskal Wallis) was used to compare water physico-chemical parameters and nutritional composition across substrates.

3. RESULTS

3.1 Performance of BSF larvae

BSF larvae grew in all the five organic substrates (bovine (cattle) and swine droppings, chicken house rearing waste, decomposing avocado and millet brew waste). However, their growth performance varied significantly across substrate types (χ^2 (df = 4) = 421.2, P < .001). A significant difference was observed between millet brew waste and the rest of the substrates (P = 0.03), Fig. 2. Millet brew waste recorded the best performance followed by decomposing avocado, waste from chicken house, bovine (cattle) droppings and the least was swine droppings. Millet brew waste substrate had the highest temperature (35.2°C), followed by decomposing avocado (30°C), waste from chicken house (28.3°C), bovine (cattle) droppings (27.9°C) and swine droppings (27.2°C). Similarly, growth rate of BSF to reach third instar larval stage was highest in millet brew waste substrate (18 days), followed by decomposing avocado (24 days), waste from chicken house (26 days), cattle and swine droppings (28 days).

3.2 Nutritional Composition of Substrates

The nutritional composition of the substrates varied significantly ($\chi 2$ (df = 4) = 365.4, P < .001). Decomposing avocado substrate had significantly low crude protein compared to swine droppings, bovine (cattle) droppings, chicken house waste and millet brew waste. Overall, bovine (cattle) substrate produced larvae with the highest crude protein compared to the rest of the substrates. On the hand, bovine (cattle) waste had significantly low dry mater compared to the rest of the substrates, while millet brew waste had significantly low ash content (Table 1).

3.3 Physico-Chemical Properties of Substrates

All the measured physico-chemical parameters (temperature, pH and ammonia) varied significantly across substrate types investigated ($\chi 2$ (df = 4) = 441.4, P = .002). Millet brew waste had significantly high temperature compared to the rest of the substrates, while swine droppings had significantly low temperature compared to chicken house waste, millet brew waste and decomposing avocado. On the other hand, millet brew waste had significantly high pH compared to the rest of the substrates, while bovine (cattle) waste had significantly high ammonia (Table 2).

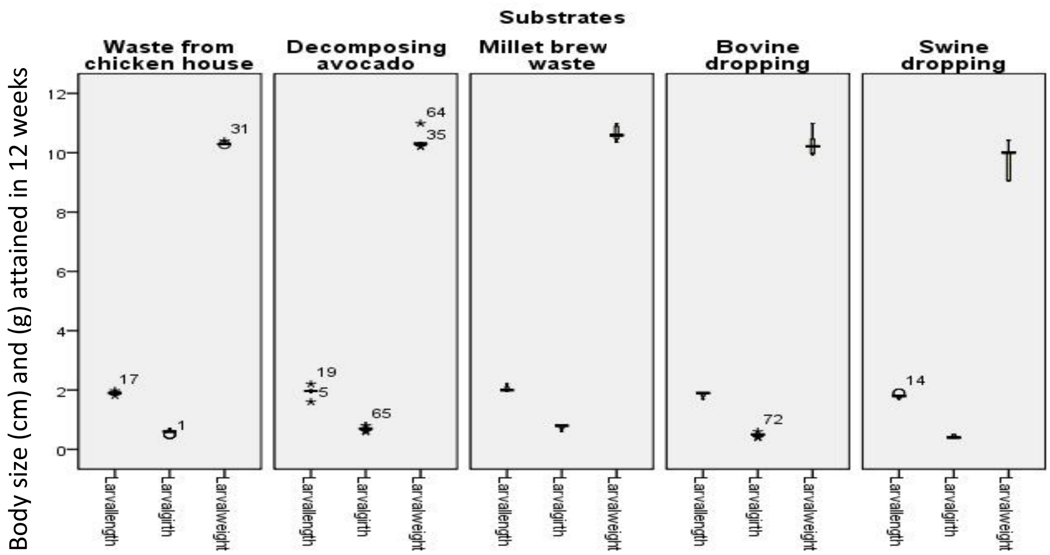

Fig. 2. Growth performance of BSF larvae reared on different substrates over a period of 12 weeks

Table 1. Nutrient composition (ash, protein and dry matter) of different substrates (Mean± Standard deviation)

Substrate type	Parameters		
	Ash (%)	Crude protein (%)	Dry matter (%)
Swine droppings	18.91±0.5[a]	39.4±0.3[a]	40.67±0.3[a]
Bovine (cattle) droppings	18.75±0.6[a]	55.71±0.2[b]	30.52±0.8[b]
Chicken house waste	14.25±1.1[b]	39.55±0.2[a]	40.99±1.4[a]
Millet brew waste	8.98±0.4[c]	44.31±20.3[c]	36.29±1.5[c]
Decomposing avocado	12.14±0.3[d]	38.62±0.1[d]	37.16±0.6[dc]

Mean values with the same letter within a column are not significantly different (p < .05).

Table 2. Physico-chemical properties of different substrates (Mean± Standard deviation)

Substrate type	Parameters		
	Temperature (°C)	pH	Ammonia-N (mgL^{-1})
Swine droppings	27.2±0.3[a]	6.4±0.4[a]	371.2±0.4[a]
Bovine droppings	27.9±0.4[a]	6.5±0.2[a]	468.2±0.5[b]
Chicken house waste	28.3±1.6[b]	6.2±0.6[a]	392.5±1.6[c]
Millet brew waste	35.2±0.6[c]	8.6±20.4[c]	390.1±1.1[c]
Decomposing avocado	30±0.6[d]	7.3±0.2[d]	236.8 ±0.8[e]

Mean values with the same letter within a column are not significantly different (p < .05)

4. DISCUSSION

In sub-Saharan Africa, high demand for silver cyprinid (*Rastrineobola agentina*) as a source of protein by both animal industry (fish, and poultry sectors) and humans has led to increased cost of fish meal as a source of feed for poultry and fish. An alternative substitution is required, since protein source derived from plants (soya beans, *Glycine max L*; and maize, *Zea mays L*) are of lower protein quality [17] and are, as well being consumed by humans. Protein source from the black soldier fly (BSF) larvae, *Hermetia illucens*, is considered as a promising alternative [2]. This study presents the first account on exploring and establishing the best locally and readily available organic substrates that can be used for rearing BSF in Uganda.

Results from this study show that substrate types affect growth performance of BSF larvae, concurring with other findings [18,19]. Overall, millet brew wastes produced larvae with the widest girth, highest weight and were longest, reaching 3rd instar within 18 days. Besides other food values, millet contains carbohydrate as its main nutritional composition, requiring amylase enzymes for their breakdown [20]. Although BSF larvae uses a cocktail of enzymes to digest different substrates, previous studies by Kim et al. [20] indicated that amylase is the most dominant. It is possible that the high titre of these enzymes in BSF larvae [20] could have fore-played the breakdown of polysaccharides in the millet brew waste into soluble monosaccharides, hence their quick uptake alongside other nutritional requirements, allowing for fastest growth. Other studies [12,11] have reported that avocado contains high level of vitamins, mono-saturated fats and less carbohydrates; wastes from chicken house contain high level of fiber; while bovine droppings have higher levels of carbohydrates.

Although other nutritional composition investigated in this study were not reported by Spranghers et al. (2017) and Osuga et al. [8,11], Lalander et al. [18] on the other hand reported that swine droppings also has high protein content. The highest composition of protein in bovine waste compared to other wastes may be attributed to the combination of diet that they feed on. It would be expected that the best performing organic substrates would be those with high protein composition. However, it is possible that other factors such as temperature and texture of the substrates contributed to the differences in growth performance observed in this study.

The development of most insect larvae depends on optimum temperature [21] and pH, for insects whose larvae require water [22]. The substrates used in this study were dumped wastes, for example, fermented millet brew waste. It is possible that residual fermentation processes continued during larval rearing, hence the high temperature of the substrate observed, which favoured growth of BSF larvae in millet brew waste. During development, BSF larvae feed from the top of the substrates downward, which require wiggling through a substrate with loose texture as was observed with millet brew waste.

5. CONCLUSION

From this study, it can be concluded that the type of substrate has great influence on BSF production, such as total larval yield, larval body weight, girth, and length. The best production results were achieved with millet brew wastes compared to decomposing avocado, swine droppings, and (bovine) cattle droppings. We recommend that at the initial stages where large numbers of adult flies and eggs are required, farmers should emphasize on raising BSF larvae on millet brew waste. Further, studies to determine the nutritional composition of BSF larvae raised on each of the substrates investigated in this study should be conducted for BSF larvae to supplement fish meal in diets for fish and poultry.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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