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## THE INFLUENCE OF BIOCHAR IN IMPROVING THE GROWTH RATE OF BLACK SOLDIER FLY LARVAE AND THE QUALITY OF FRASS

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### ABSTRACT

*The use of black soldier fly (BSF) technology is a good organic waste management technique that results in the production of valuable agricultural products. Waste management uses different technologies depending on the type of waste being handled. This study manages organic waste using BSF. The BSF larvae have a high ability to degrade organic waste, and after degrading the larvae are harvested for animal feed as a protein source. The quality of harvested BSF larvae depends on what they eat. In this study, biochar was blended with market wastes, and fed to BSF larvae to improve both the larvae's growth rate and the quality of the frass. Biochar absorbs moisture in the feed making the BSF larvae grow well in optimal moisture content. The results indicated that including 25% biochar in rearing BSF larvae improves their growth rate. The weight of the larvae obtained in each substrate on the harvested day were  $0.25 \pm 0.03$  and  $0.13 \pm 0.01$  g in BI and MW, respectively. The unconsumed feed (frass) from each substrate used to grow vegetables and the growth parameters recorded show that the plants grow better in BI than in frass from MW and BI. Plants grown in frass without biochar grow slowly. Therefore, for more productivity of BSF larvae biochar should be blended in the BSF feed. This also reduces the smell of the fermented BSF feed and improves the quality of the frass.*

**Keywords:** Black soldier fly larvae, Environmental management, Industrial wastes, Sisal decorticated waste, Waste treatment

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### INTRODUCTION

The availability of organic wastes which is a high feed value for black soldier fly (BSF) larvae consumption is still challenging (Bel and Warner, 2008; Ng *et al.*, 2016; Raksasat *et al.*, 2021). This is due to high competition from other livestock farmers which used waste as a feed source for pigs and other domestic animals (Tufvesson *et al.*, 2013). Value addition in the market wastes with less competition is the way to ensure the availability of BSF feed that is used to rear them. One of the value-addition methods is using the appropriate amount of biochar. Biochar is a lightweight black residue, made of carbon and ashes, remaining after the pyrolysis of biomass, and is a form of charcoal (Zhang *et al.*, 2020; Khan *et al.*, 2024).

The act of using biochar has a lot of potential including; improving the growth rate of BSF larvae, improving the quality of frass and reducing ammonia emissions and smells of the waste used to rear BSF larvae (Fangueiro *et al.*, 2021; Zhang *et al.*, 2023).

Previous studies have shown that the inclusion of biochar in the BSF larvae feed substrate has yielded good results including the large size and weight of the BSF larvae (gram/larvae) also the BSF larvae uniformity (Beesigamukama *et al.*, 2020; Beyers *et al.*, 2023). Moreover, in the basins containing substrate with biochar, there was no pungent smell of the fermented feed. In addition, the use of biochar in BSF substrate yields frass with high quality (Diatta *et al.*, 2020).

## MATERIALS AND METHODS

**Preparation of Biochar:** Biochar, the residual material that remains after the pyrolysis of either piece of wood or grasses (Zhang *et al.*, 2020), was prepared using a pit-digging method. Initially, a pit was dug, and dried wood pieces were gathered and filled into the pit to initiate a controlled fire. Additional wood was swiftly added as the flames consumed the older pieces, ensuring a continuous burning process. Once the desired pyrolysis level was achieved, water was carefully poured onto the fire to extinguish it. The resulting biochar was left undisturbed overnight to cool and mature, and it was subsequently collected the following morning, and fully prepared for incorporation into the feed for BSF larvae.

**Inclusion of Biochar in the Substrate:** The impact of biochar in the growth rate of BSF larvae can be obtained in both ways i.e. as a bulking agent and as inclusion in the substrate. In this experiment, three basins were prepared and in each, the following were included; A: 25% biochar blended with market waste B: 25% biochar on top of substrate (not mixed), This was done to observe if biochar will work best if it will be blended with BSF feed or if it will just put on top. The control of this experiment was 100% market waste as a substrate of BSF larvae.

**Black Soldier Fly Larvae Distribution and Sampling:** Approximately 10 grams of eggs were placed into three separate trays and allowed to hatch. Once the Black Soldier Fly (BSF) larvae reached five days of age, they were removed from the incubator and evenly distributed into each of two basins containing different feed types.

The basins were prepared as follows: Basin A: Mixture of 25 kilograms of biochar plus 75 kilograms of market waste. Basin B: 100 kilograms of market waste only. To ensure validity and reliability, each feed category was replicated thrice. All basins were raked from day 7 after the introduction of the 5-day-old larvae. At the end of the 16-day culture period in the basins, the BSF larvae were harvested.

Sampling of the larvae was conducted, including counting, weighing, and recording of data for subsequent analysis.

**Plant Growth Parameters:** Preparation of the plots was done before sowing the plant seed. The frass obtained after harvesting the BSF larvae was mixed with either biochar in a ratio of 1: 10 kg. The frass-biochar (P1) mixtures were placed in plastic containers and vegetables were planted in them. Other combinations were P2: Frass without biochar, and P3: empty soil (that serves as the control). The vegetable was sown and the plant growth was monitored. Plant growth parameters such as plant height, leaf length, seed and leaf width were measured in centimetres and recorded after every three days. For the number of leaves, counting was done and recorded. For the leaf width, a ruler was placed amid the leaf and the measurement was read. For the plant height, the ruler was placed on the surface of the soil to the top of the plant then the measurement was read and recorded.

**Frass Chemical Composition Assay:** The chemicals composition of frass (pH, EC (salts), dry matter, carbon, total nitrogen, phosphorus, potassium, calcium, magnesium, sulphur, manganese, iron, zinc, copper, boron, sodium, and C/N ratio) were analyzed using different methods such as; Potentiometric, Gravimetric, Walkley black, Dumas and Spectroscopy. In analyzing pH and EC potentiometric methods were used. This method is based on the measurement of the activity of hydrogen ions in an acid-base substance (Harvey, 2000; Risch *et al.*, 2015). In this study, the pH and EC of the frass were analyzed using the potentiometric method.

**Data Analysis:** The obtained data were statistically analyzed using one-way analysis of variance (ANOVA) and student's t-test to compare the means of the different treatments. Significant treatment means arising from ANOVA were separated using post hoc LSD at  $p < 0.05$ . Whereby if the t-statistic value was greater than the t-critical value there was a significant difference two treatment means at  $p < 0.05$ .

**RESULTS AND DISCUSSION**

**Black Soldier Fly Larvae Growth Rate:** The study found that BSF larvae growth rate was good in the basins containing blended market waste and 25% inclusion of biochar. On day 16 the weight of the larvae was  $0.13 \pm 0.02$  and  $0.25 \pm 0.04$  g in BSF larvae raised in market waste, and in 25% biochar plus market waste, respectively (Table 1).

**Table 1: The influence of biochar on the growth rate of black soldier fly larvae reared in different substrate**

Days in basin	Market waste (control)	Market waste plus Biochar	Correlation coefficient
7	$0.05 \pm 0.02$	$0.10 \pm 0.01^*$	0.94
8	$0.05 \pm 0.01$	$0.11 \pm 0.03^*$	0.93
9	$0.07 \pm 0.01$	$0.13 \pm 0.02^*$	0.91
10	$0.07 \pm 0.03$	$0.15 \pm 0.01^*$	0.94
11	$0.07 \pm 0.02$	$0.16 \pm 0.02^*$	0.96
12	$0.07 \pm 0.03$	$0.18 \pm 0.01^*$	0.98
13	$0.08 \pm 0.01$	$0.20 \pm 0.02^*$	0.97
14	$0.10 \pm 0.02$	$0.21 \pm 0.01^*$	0.98
15	$0.12 \pm 0.03$	$0.23 \pm 0.02^*$	1.00
16	$0.13 \pm 0.02$	$0.25 \pm 0.02^*$	1.01

*\*value on the same row for each treatment with an asterisk are significantly different ( $p < 0.05$ ) using t-test pairwise comparison*

This implied that biochar contributed to the growth rate of BSF larvae. Beesigamukama *et al.* (2020) in their study demonstrated that biochar amendment of agro-industrial waste enhanced black soldier fly larval biomass and quality frass fertilizer. This study to interpret the significant difference in the growth rate of BSF larvae reared in two different substrates, compared the t-statistic to the t-critical value. The result indicated that the t-statistic (10.72) obtained was greater than the t-critical (1.83). This means that there were significant differences between BSF larvae reared in market waste and market waste plus biochar. Biochar, a carbon-rich material produced from abandoned biomass has excellent stability, high porosity and extensive microstructure (Li *et al.*, 2020). It has been used in soil nitrogen recovery, nitrogen leaching reduction, and the improvement of anaerobic digestion efficiency (Dutta *et al.*, 2021). More importantly, biochar has been widely utilized as an amendment in compost processing (Qin *et al.*, 2022), which promotes composting processes,

alleviates the emissions of  $NH_3$  and  $N_2O$ , and improves the quality of final composting products

**Plant Growth Parameters:** The measured parameters which include; the number of leaves, leaf width and plant heights showed that biochar improved the quality of the frass (Table 2). Plants grown in frass plus biochar ( $19.1 \pm 0.00$  cm) grow higher than plants grown in frass from market waste ( $14.1 \pm 0.00$  cm) and empty soil ( $9.5 \pm 0.00$  cm). From Table 2, there was a strong positive relationship between the parameters being compared; frass plus biochar (a), frass without biochar (b) and empty soil (c), since the correlation coefficients obtained data for all the measured parameters were approximately equal to 1. This result correlated with other studies which emphasized that biochar improved soil quality and hence plant growth (Yeboah *et al.*, 2009; Ding *et al.*, 2016).

**Frass Chemical Composition:** The plant growth in frass plus biochar had more chemical content as shown in Table 3. The results show that nitrogen was  $1.91 \pm 0.04$ ,  $0.80 \pm 0.05$  and  $1.50 \pm 0.04$  respectively in different treatments. Moreover, other results showed that frass plus biochar had a pH of  $9.22 \pm 0.03$ , this tends to control the toxicity of the soil especially those caused by aluminium. The coefficient correlation obtained in Table 3, showed that there were strong positive correlations between the parameters being compared; frass plus biochar (a), frass without biochar (b) and empty soil (c). Since the values were positive, it means that as one parameter increases, the other parameter tends to increase as well. Conversely, if one decreases, the other tends to decrease. The obtained results correlate with the study of Gärttling and Schulz (2022) which showed that frass had a slightly alkaline pH and NPK ratio of 1:0.9:1.1. Moreover, the high pH promotes the development of plant roots. The frass plus biochar can be more suitable in acidic soil. The most required nutrients for plant growth including carbon, calcium, zinc and copper observes to be at optimum level. This indicates that frass plus biochar contains the micro and macro elements at the recommended level.

**Table 2: The influence of biochar on plant growth cultivated in different substrate**

Days	Parameters	Frass plus biochar (a)	Frass without biochar (b)	Empty soil (c)	Correlation coefficient		
					a-c	b-c	a-b
<b>7</b>	Number of leaves (cm)	5.00 ± 0.00 <sup>b</sup>	4.00 ± 0.00 <sup>ab</sup>	3.00 ± 0.00 <sup>a</sup>	0.89	0.89	0.87
	Leaf width (cm)	2.20 ± 0.00 <sup>b</sup>	1.30 ± 0.00 <sup>a</sup>	2.00 ± 0.00 <sup>ab</sup>	0.89	0.89	0.87
	Plant height (cm)	4.00 ± 0.01 <sup>a</sup>	4.50 ± 0.00 <sup>ab</sup>	5.20 ± 0.00 <sup>b</sup>	0.87	0.87	0.86
	Leaf length (cm)	3.01 ± 0.03 <sup>b</sup>	2.10 ± 0.01 <sup>a</sup>	2.90 ± 0.02 <sup>b</sup>	0.90	0.88	0.86
<b>10</b>	Number of leaves	5.00 ± 0.00 <sup>b</sup>	4.00 ± 0.00 <sup>a</sup>	4.00 ± 0.00 <sup>a</sup>	0.89	0.87	0.82
	Leaf width (cm)	3.00 ± 0.00 <sup>b</sup>	1.80 ± 0.01 <sup>a</sup>	2.00 ± 0.00 <sup>a</sup>	0.89	0.87	0.82
	Plant height (cm)	8.00 ± 0.01 <sup>b</sup>	7.00 ± 0.00 <sup>ab</sup>	6.50 ± 0.00 <sup>a</sup>	0.87	0.85	0.87
	Leaf length (cm)	4.60 ± 0.01 <sup>b</sup>	3.20 ± 0.01 <sup>ab</sup>	2.70 ± 0.01 <sup>a</sup>	0.87	0.85	0.87
<b>13</b>	Number of leaves	6.00 ± 0.00 <sup>b</sup>	5.00 ± 0.00 <sup>a</sup>	5.00 ± 0.00 <sup>a</sup>	0.86	0.83	0.86
	Leaf width (cm)	3.20 ± 0.00 <sup>b</sup>	1.80 ± 0.00 <sup>a</sup>	2.10 ± 0.00 <sup>ab</sup>	0.86	0.83	0.86
	Plant height (cm)	8.50 ± 0.00 <sup>b</sup>	7.00 ± 0.00 <sup>ab</sup>	6.50 ± 0.00 <sup>a</sup>	0.82	0.78	0.82
	Leaf length (cm)	5.20 ± 0.02 <sup>c</sup>	4.20 ± 0.01 <sup>b</sup>	3.50 ± 0.02 <sup>a</sup>	0.82	0.78	0.82
<b>16</b>	Number of leaves	6.00 ± 0.00 <sup>b</sup>	6.00 ± 0.00 <sup>b</sup>	5.00 ± 0.00 <sup>a</sup>	0.80	0.75	0.87
	Leaf width (cm)	4.00 ± 0.00 <sup>b</sup>	3.00 ± 0.01 <sup>a</sup>	4.00 ± 0.00 <sup>b</sup>	0.79	0.74	0.87
	Plant height (cm)	9.00 ± 0.00 <sup>c</sup>	7.50 ± 0.00 <sup>a</sup>	8.00 ± 0.00 <sup>b</sup>	0.76	0.73	0.86
	Leaf length (cm)	7.20 ± 0.01 <sup>c</sup>	6.50 ± 0.01 <sup>b</sup>	4.00 ± 0.02 <sup>a</sup>	0.78	0.80	0.86
<b>19</b>	Number of leaves	7.00 ± 0.01 <sup>b</sup>	7.00 ± 0.00 <sup>b</sup>	6.00 ± 0.00 <sup>a</sup>	0.78	0.78	0.82
	Leaf width (cm)	4.50 ± 0.00 <sup>a</sup>	5.00 ± 0.00 <sup>b</sup>	4.70 ± 0.00 <sup>ab</sup>	0.79	0.79	0.82
	Plant height (cm)	10.00 ± 0.00 <sup>b</sup>	10.00 ± 0.00 <sup>b</sup>	9.00 ± 0.00 <sup>a</sup>	0.81	0.80	0.87
	Leaf length (cm)	7.40 ± 0.01 <sup>b</sup>	7.00 ± 0.02 <sup>b</sup>	4.30 ± 0.01 <sup>a</sup>	0.85	0.76	0.87
<b>22</b>	Number of leaves	8.00 ± 0.00 <sup>b</sup>	8.00 ± 0.01 <sup>b</sup>	7.00 ± 0.00 <sup>a</sup>	0.84	0.71	0.86
	Leaf width (cm)	9.00 ± 0.00 <sup>c</sup>	7.30 ± 0.00 <sup>b</sup>	5.20 ± 0.00 <sup>a</sup>	0.93	0.76	0.86
	Plant height (cm)	13.00 ± 0.00 <sup>c</sup>	10.00 ± 0.00 <sup>b</sup>	9.00 ± 0.00 <sup>a</sup>	0.97	0.73	0.82
	Leaf length (cm)	7.50 ± 0.01 <sup>b</sup>	7.20 ± 0.01 <sup>b</sup>	5.00 ± 0.02 <sup>a</sup>	0.93	0.76	0.82
<b>25</b>	Number of leaves	9.00 ± 0.00 <sup>b</sup>	9.00 ± 0.00 <sup>b</sup>	7.00 ± 0.00 <sup>a</sup>	0.88	0.64	0.97
	Leaf width (cm)	9.20 ± 0.01 <sup>b</sup>	10.50 ± 0.00 <sup>c</sup>	6.30 ± 0.00 <sup>a</sup>	1.00	1.00	0.93
	Plant height (cm)	19.10 ± 0.00 <sup>c</sup>	14.10 ± 0.00 <sup>b</sup>	9.50 ± 0.00 <sup>a</sup>	0.90	0.68	0.88
	Leaf length (cm)	7.60 ± 0.01 <sup>b</sup>	7.40 ± 0.01 <sup>b</sup>	5.10 ± 0.02 <sup>a</sup>	0.75	0.75	1.00

<sup>abc</sup> = Means within rows with different superscript letters differ significantly ( $p < 0.05$ )

**Table 3: The influence of biochar in improving soil quality**

Parameters	Unit	Frass plus biochar (a)	Frass without biochar (b)	Empty soil (c)	Correlation coefficient		
					a-c	b-c	a-b
pH		9.22 ± 0.03 <sup>c</sup>	6.00 ± 0.04 <sup>a</sup>	8.50 ± 0.02 <sup>b</sup>	0.99	0.90	0.90
EC (Salts)	mS/cm	10.12 ± 0.02 <sup>c</sup>	0.75 ± 0.05 <sup>a</sup>	1.50 ± 0.03 <sup>b</sup>	0.99	0.90	0.89
Dry matter	%	58.71 ± 0.02 <sup>b</sup>	32.46 ± 0.03 <sup>a</sup>	60.0 ± 0.02 <sup>c</sup>	0.99	0.90	0.89
Carbon	%	33 ± 0.01 <sup>b</sup>	13.0 ± 0.04 <sup>a</sup>	60.0 ± 0.04 <sup>c</sup>	0.99	0.90	0.89
Total Nitrogen	%	1.91 ± 0.04 <sup>c</sup>	0.80 ± 0.05 <sup>a</sup>	1.50 ± 0.04 <sup>b</sup>	0.99	0.90	0.89
Phosphorus	%	0.91 ± 0.03 <sup>c</sup>	0.20 ± 0.06 <sup>a</sup>	0.75 ± 0.05 <sup>b</sup>	0.99	0.89	0.89
Potassium	%	3.21 ± 0.04 <sup>c</sup>	0.40 ± 0.03 <sup>a</sup>	2.00 ± 0.02 <sup>b</sup>	0.99	0.89	0.90
Calcium	%	2.17 ± 0.06 <sup>c</sup>	0.60 ± 0.04 <sup>a</sup>	1.50 ± 0.03 <sup>b</sup>	0.99	0.89	0.89
Magnesium	%	0.61 ± 0.04 <sup>b</sup>	0.20 ± 0.05 <sup>a</sup>	0.80 ± 0.05 <sup>c</sup>	0.99	0.89	0.90
Sulphur	%	0.73 ± 0.05 <sup>c</sup>	0.20 ± 0.03 <sup>a</sup>	0.50 ± 0.03 <sup>b</sup>	0.99	0.89	0.89
Manganese	ppm	229 ± 0.02 <sup>a</sup>	200 ± 0.04 <sup>a</sup>	800 ± 0.03 <sup>b</sup>	0.99	0.90	0.89
Iron	ppm	11500 ± 0.04 <sup>c</sup>	9572 ± 0.05 <sup>b</sup>	3467 ± 0.03 <sup>a</sup>	1.00	0.89	0.89
Zinc	ppm	201 ± 0.03 <sup>b</sup>	40.0 ± 0.03 <sup>a</sup>	10000 ± 0.02 <sup>c</sup>	1.00	0.97	0.89
Copper	ppm	40.9 ± 0.04 <sup>b</sup>	8.00 ± 0.06 <sup>a</sup>	400 ± 0.03 <sup>c</sup>	1.00	1.00	0.89
Boron	ppm	49.2 ± 0.03 <sup>b</sup>	20.0 ± 0.05 <sup>c</sup>	140 ± 0.02 <sup>c</sup>	1.00	1.00	0.90
Sodium	ppm	4192 ± 0.02 <sup>c</sup>	2100 ± 0.03 <sup>a</sup>	3000 ± 0.03 <sup>b</sup>	1.00	0.97	0.89
C/N ratio		17.9 ± 0.02 <sup>ab</sup>	10.0 ± 0.04 <sup>a</sup>	20.0 ± 0.01 <sup>b</sup>	1.00	0.65	0.90

*abc* = Means within rows with different superscript letters differ significantly ( $p < 0.05$ )

**Conclusion:** The results of this study reveal that biochar improves the growth rate of BSF larvae and the quality of the frass. The weights of the larvae were higher in BSF larvae reared in market waste plus biochar than in market waste only. Moreover, the vegetables planted using frass plus biochar as a fertilizer grew better than those in frass from market waste and empty soil. Food insecurity might be associated with different factors including poor soil fertility. The utilization of frass plus biochar will increase crop production and ensure food security. Therefore, the findings obtained in this study show that the inclusion of biochar in the substrate used to rear BSF larvae improves the growth rate of larvae and the quality of frass. This optimization will enable BSF stakeholders to get high-value feed at reasonable costs which yields more profit through obtaining large amounts of harvested BSF larvae. Moreover, the farmers will get organic fertilizer (frass) with sufficient nutrients required.

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