



Frass derived from black soldier fly larvae treatment of biodegradable wastes. A critical review and future perspectives

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ABSTRACT

Inadequately treated biodegradable waste is considered an environmental, social and economic threat worldwide, which call for great attention. Waste treatment with larvae of the black soldier fly (BSF, *Hermetia illucens*) complies with the concepts of circular economy, as it enables the transformation of these wastes into marketable products, closing loops and promoting circularity. The processing residues of the treatment (frass) is constantly generated in waste management facilities in large volumes, and this product can be used as an organic fertilizer in agriculture, stimulating a transition to a circular economy. However, many aspects related to frass are still unknown, such as its varying composition of nutrients, microorganisms and bioactive compounds, its post-processing requirements for improved biological stabilization, its behavior in the soil and action in the plants' metabolism, among other aspects. In this review article, we highlight the potential of frass from BSF larvae treatment of biodegradable waste in the world market regarding its possible use as a fertilizer, summarize recent results with this novel product and point towards future research perspectives.

1. Introduction

The global generation of waste is expected to reach 3.4 billion tons by 2050, and up to 44% of the total accumulated waste currently comprise biodegradable materials globally, with a higher proportion in low to middle-income countries; of this accumulated waste today, most is either disposed in landfills (37%) or open dumps (33%) (Kaza et al., 2018). These ways of disposing biodegradable waste are considered major threats to the environment, due to the release of greenhouse gases (GHG) to the atmosphere and soil/water contamination with toxic compounds and nutrients from leachates, among other factors (Koda et al., 2017). Governments therefore have the responsibility to recognize the socio-economic and environmental risks and impacts associated with improper waste dumping, and must pursue more effective waste handling methods to ensure livability while moving towards a circular bioeconomy (Silva et al. 2017; Ahmad et al., 2020).

In high-income countries, most of the waste is collected and treated by government-funded initiatives, while in low- and middle-income countries, resource limitation hinders the development of adequate waste management to deal with the amount of waste generated (Yang et al., 2018). As a result, an informal sector arises, comprised of so-called

“waste-pickers” or “scavengers”, that take over the responsibility of waste recycling worldwide (Binion and Gutberlet, 2012). These actors play an important role in waste sourcing, handling and recycling, especially for the non-biodegradable waste, such as paper, plastic, metal and glass, while the organic fraction remains unmanaged (Linzner and Lange, 2013; Tong et al., 2021). Even after decades, this issue still remains, especially in low- and middle-income countries, where waste recycling is a common livelihood for the urban poor (Kaza et al., 2018). One major concern with the biodegradable waste management is the cost of collection and treatment, which often exceed the revenue derived from the generated products, rendering the treatment economically unviable (Lohri et al., 2017). Consequently, the poor financial outcome associated with waste recycling results in low motivation and incentives towards accomplishing the UN Sustainable Development Goals (SDG) and its targets, including reduced environmental impacts of cities (target 11.6) and eliminating waste dumping (target 6.3), which can be directly connected to waste management technologies (SGDs).

At the same time, the increasing world population result in not only increased waste generation, but also in a need to increase global food production substantially in the near future. According to McKenzie and Williams (2015), this means that the world population and policy-

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makers will face major challenges regarding the changes in the current paradigm of productivity, in terms of environmental safety, waste generation and increased food production. As natural sources of plant nutrients are depleted and some are non-renewable (e.g. phosphorus), and we surpass the planetary boundaries of reactive nitrogen (N) in the atmosphere (Steffen et al., 2015), harvesting nutrients from biodegradable materials, as a measure to recover them (Harder et al., 2021), become increasingly important (Gerten et al., 2020). In addition, the consumption of livestock products has increased steadily since the 1960s (González et al., 2020). This has encouraged unsustainable practices for feed production that have been demonstrated to have detrimental impact on the environment; around 30% of the global wild fish stocks are currently overfished (FAO, 2020), while monocultures of soybeans have severe impacts on land use and biodiversity (Green et al., 2019).

A promising method for treating biodegradable waste that could contribute to the three mentioned challenges has arisen in the past decade; black soldier fly (*Hermetia illucens*, (Diptera: Stratiomyidae)) (BSF) larvae treatment (Čičková et al., 2015; Gold et al., 2018). In this insect-based treatment, biodegradable waste is converted into two products: a larval biomass rich in proteins and lipids (Lalander et al., 2019; Ewald et al., 2020) that can be used in animal feed (Wang and Shelomi, 2017); and a processing residue, also called frass, which can be considered a fertilizer to be used for many purposes (Beesigamukama et al., 2020a). As two valuable products are generated, this technology complies with the principles of a circular bioeconomy, in which the waste from one process becomes the resource in another (Slorach et al., 2019).

Several studies have investigated the environmental impacts of the products obtained by the BSF larvae treatment of biodegradable materials. Considering the use of insect protein for feed and food, Smetana et al. (2016) reported that the environmental impact of traditional food sources (e.g. chicken meat and whey proteins) was 2–5 times higher than that of insect-based products. However, the benefits of this product were dependent on the feed substrate used to feed the larvae. Similarly, Salomone et al. (2017) highlighted that the frass produced by BSF larvae was one of the main outputs of the process, and it could replace traditional N fertilizers, thereby lowering the global warming potential associated with the use of any conventional N fertilizers. Smetana et al. (2019) also demonstrated that the BSF larvae frass gave lower environmental impacts in comparison to the production of other organic fertilizers, in relation to water use, energy expenditures, global warming potential, and other impact categories. It is noteworthy that the environmental benefits regarding the production of insect frass is closely related to the source of substrate used to feed the larvae, with lower impacts being reported when using non-utilized waste streams, instead of the traditional products such as soybean meal (Smetana et al., 2019).

Due to the constant production of frass in waste management plants, its abundance in plant nutrients and the potential of contributing to income-generation by the agribusiness sector, frass has started to gain attention in recent years (Gärtling et al., 2020). However, frass has been poorly explored in comparison to the larval biomass obtained in the same process. Specifically, there are numerous knowledge gaps in relation to the usage of frass and the benefits in agriculture and other cultivation related activities. Considering the convincing indices that this biodegradable waste treatment method can be environmentally sustainable (Smetana et al., 2021) and economically viable if undertaken in the correct manner, the full potential of frass should be thoroughly investigated, in order to fill existing knowledge gaps and stimulate the adoption of this technology. In this sense, societies could take a step in the transition from a linear to a circular bioeconomy (Ojha et al., 2020).

In November 2021, the International Platform of Insects for Food and Feed (IPIFF) elaborated a document to pave the way for a EU-wide insect frass standard (press release). This action resulted in a modification of the EU Regulation 142/2011, which included insect frass in a new category named “insect excrements”, with the requirement that frass

that is to be used as fertilizer has to be subjected to heat treatment at 70 °C for 60 min. According to Van Looveren et al., (2022), this is sufficient to reduce Enterobacteriaceae counts below the detection limit of 10 colony forming units (cfu) per gram of frass, result in the absence of *Salmonella* spp. in 25 g of product and reduce vegetative forms of *Clostridium perfringens*. This strong campaign by IPIFF is only one of the actions taken by different entities regarding frass, which is gaining prominent attention and is likely to be introduced on markets worldwide shortly. With this in mind and using the data reported on BSF larvae treatment of biodegradable materials, this review summarizes the current knowledge on the characteristics and use of frass obtained by treating biodegradable waste streams with the larvae of the black soldier fly. In addition, this review points towards salient knowledge gaps that still exist in relation to BSF larvae frass, and highlights the benefits of using frass as a supplement to improve plant nutrition and soil fertility.

2. Contribution of BSF larvae to the circular bioeconomy

Biodegradable waste is generated in multiple human activities, from households to agricultural and industrial endeavors (Caldeira et al., 2019). These waste streams have high organic contents, macro- and micro-nutrients, and should be considered as valuable resources that could be recovered and reintroduced in other production chains. If the biodegradable waste, however, is disposed of in an inadequate way, it can become a threat to the environment. During its decomposition, these materials can release large amounts of nutrients that exceed the normal levels of nutrients in soil and water bodies, such as N, phosphorus (P) and potassium (K), as well as GHGs to the atmosphere (e.g. CO₂, CH₄ and N₂O), in addition to releasing pathogenic microorganisms and spreading heavy metals to the environment (Alvarenga et al., 2015; Ferronato et al., 2017). Therefore, adopting effective waste management technologies such as BSF larvae treatment could mitigate these negative impacts by avoiding inadequate disposal.

The contributions of BSF larvae treatment of biodegradable waste for a circular bioeconomy are very diverse. For instance, Bortolini et al. (2020) demonstrated that the larvae reduced chicken manure by 75% on a dry matter (DM) basis, and transformed this waste into added-value products that could be used in any form of cultivation. Similarly, Lalander et al. (2018) demonstrated that this technology could be more profitable in comparison to thermophilic composting of food waste and faeces. Complementarily, Mertenat et al. (2019) demonstrated that BSF larvae composting generated less than 2.5% of the GHG emissions of that generated in windrow composting for the same waste stream. While being more environmentally and economically sustainable than the conventional waste management methods, the products obtained (larval biomass and frass) also contribute directly to the circular bioeconomy. For instance, Rawski et al. (2021) fed BSF larvae with a mixture of fruits and vegetables, and the resulting protein ingredient (larvae meal) was used to feed an important fish in aquaculture, the Siberian sturgeon. The authors found that the replacements of up to 30% of fishmeal to BSF larvae meal resulted not only in economic gains, but also improving the health status of fish. Moreover, the frass was tested as feed ingredient for hybrid tilapia by Yildirim-Aksoy et al. (2020). The authors reported that inclusions of up to 30% in replacement of fishmeal resulted in higher growth and resistance to diseases, suggesting that the frass may increase the sustainability of aquaculture production. However, as the frass may contain pathogens derived originally from the contaminated waste streams (Lopes et al., 2020a), the hygiene aspects of frass as a fishmeal must be considered carefully. Also in terms of waste management, BSF larvae composting has demonstrated its usefulness and versatility; Song et al. (2021) reported a lower global warming potential of BSF larvae frass production in comparison to incinerating waste. The authors further recommended that aerated composting as a post-treatment of frass was useful in obtaining a waste-based fertilizer.

2.1. Waste reduction and nutrient recovery in BSF larvae composting

Several studies have demonstrated the outstanding capacity of BSF larvae in reducing waste volumes rapidly. For instance, Diener et al. (2011) reported a 79% reduction in municipal solid waste, while food/kitchen waste was reduced by 55–68%, on a DM basis (Nguyen et al., 2015; Lalander et al., 2019). Similarly, dairy manure has been found to be reduced by 58% (DM basis) (Myers et al., 2008), and pig manure by 56% (Newton et al., 2005). It is noteworthy that the waste reduction is highly dependent on the physico-chemical characteristics of the waste, with blends comprising more than one waste stream demonstrating higher reductions than that observed for a single waste stream (Nyakeri et al., 2017). More research is needed to understand the waste reduction potential and mechanism of BSF larvae treatment using different biodegradable wastes; for reviews, see Gold et al. (2018) and Surendra et al. (2020).

In a study conducted with a mixture of pig manure, dog food and human feces as feed substrate for BSF larvae, approximately 44% of the total N in the inflow material was lost in the form of NH_3 , and 29% of the total P content was reduced on a DM basis, while the remaining was recovered in the larval biomass and frass (Lalander et al., 2015). Lopes et al. (2020b) found a lower total N volatilization of up to 34%, when feeding BSF larvae with a blend of bread and aquaculture waste. It was likely that a lower pH (ranging from 5.7 to 6.8) in the BSF larvae composting process, shifted the $\text{NH}_4^+/\text{NH}_3$ equilibrium ($\text{NH}_4^+ [\text{aq}] \rightleftharpoons \text{NH}_3 [\text{gas}] + \text{H}^+$) towards forming NH_4^+ , reducing NH_3 volatilization (Fidjeland et al., 2016). Similarly, Pang et al. (2020) demonstrated losses of total N ranging from 2.1 to 30%, and between 10 and 24% reduction in total C, when feeding BSF larvae with a mixture of food waste and rice straw at different pH. According to Pang et al. (2020), most of the C is lost as CO_2 during the BSF larvae treatment, while a small fraction is volatilized as CH_4 . It has also been reported that the generation of CH_4 was dependent on the type of feed substrate during the BSF larvae composting process and as well as its moisture content, in which wetter substrates result in higher CH_4 emissions (Chen et al., 2019). As for N, most of this element is lost to the atmosphere in the form of NH_3 , while a smaller proportion is lost as N_2O (Pang et al., 2020). As discussed earlier, P and K are predominantly found in insoluble forms within the biodegradable matter and thus these elements are not lost via volatilization.

When feeding BSF larvae with a commercial diet (made up of 47% yeast concentrate from wheat, 47% starch from wheat and potato and 6% of a binding agent), inside an open-circuit climate respiration chamber, Parodi et al. (2020) evaluated the total emissions of C, N, P and K from a BSF larvae rearing system. The authors found that about 57% of the total C ended up in the frass, 20% in the larval biomass and 24% was lost by volatilization, mainly as CO_2 . Furthermore, they found that 62% of the N was recovered in the frass and 38% was found in the larvae, with N losses being $> 1\%$ of the total N in the inflow substrate. The authors suggested that these losses could potentially be avoided if the process would be halted prior to reaching the CO_2 peak. Furthermore, it has been reported that the bioconversion efficiency increases when the feed substrate is provided more frequently in smaller batches during the treatment than providing everything in bulk at the start (Lopes et al., 2020b). However, the delivery of substrate in batches may complicate the calculation of the emission patterns of the treatment. Providing a daily biomass conversion efficiency could simplify efficiency comparison of different treatment strategies. In addition, the process technical parameters such as larval density, larval feed load and feeding strategy greatly affect the overall process efficiency (Parra Paz et al., 2015; Lopes et al., 2020b), and also the concomitant emissions. The selection of these process parameters will determine the fly population size and also the overall emission of greenhouse gases (Guo et al., 2021). It is noteworthy that several studies have identified that the post-processing of frass (thermophilic composting) was the main contributor to the overall emissions arising from the BSF larvae treatment (Mertenat

et al., 2019; Guo et al., 2021). In summary, the environmental impact of this treatment is influenced by: waste type; process of waste treatment (process-parameters and feeding strategies); post-processing strategy and duration (frass and larvae).

2.2. Production of BSF larvae frass from the treatment biodegradable waste

The amount of frass produced during the biodegradable waste treatment process with BSF larvae widely varies according to the physico-chemical characteristics of the waste. This data is not commonly presented in studies, as the production of larval biomass (protein-based product) has been considered the more interesting product. Most studies show the reduction of the organic materials used as feed substrate, rather than the “frass production efficiency” (Čičková et al., 2015; Gold et al., 2020a; Surendra et al., 2020). For instance, based on the data presented by Lalander et al. (2019), the total production (conversion efficiency) of frass, on a DM basis, when feeding BSF larvae with food waste was about 45% of the input DM, 40% for poultry manure, 52% for human feces and 15% for poultry feed. However, it is noteworthy that the “frass production” reported in studies should be linked with other process efficiency descriptions, in order to clarify if the process was well conducted, as large production of frass may well be a result of ineffective process yielding large amounts of non-consumed or non-digested organic materials. Thus, an effective production of frass should be evaluated along with other process parameters, such as the bioconversion efficiency, material reduction and final larval weight, among others. Based on the information of waste reduction presented in different studies, we calculated the frass produced per ton of processed waste (DM basis), presented in Table 1.

3. Frass characteristics

3.1. Nutritional composition of frass

The characteristics of frass from BSF larvae reported in literature indicated that the composition of plant nutrients varies somewhat in relation to the feed substrate provided to the larvae (Table 2). The total C and total N concentrations seem not to vary greatly and are around 37% and 3%, respectively. On the other hand, the concentrations of total P (1–5%) and total K (0.5–4.1%) varied significantly, and seemed to depend on the substrate used to feed the larvae. In addition, the micronutrients also vary depending on the feed substrate, while pH (around 7.5) and C/N ratio (around 15) appeared to vary less with substrate. Setti et al. (2019) supplied a standard laboratory diet for BSF larvae (“Gainesville House Fly” diet) and subsequently obtained frass containing 44 g kg^{-1} N, 52 g kg^{-1} P and 41 g kg^{-1} K. Similarly, Klammssteiner et al. (2020) fed BSF larvae separately with chicken feed, grass waste and fruit/vegetable waste, obtaining N concentrations ranging between 18.3 (fruits and vegetables) and 25.9 g kg^{-1} (chicken feed). In Lopes et al. (2020b), BSF larvae were fed exclusively on a carbohydrate source (bread waste). These larvae generated frass containing 15.2 g kg^{-1} of N, while small additions (5–15%) of a protein-rich waste stream (fish waste) resulted in increases in the nutrient content, ranging from 18.4 to 23.8 g kg^{-1} , demonstrating that the BSF larvae diet can be adjusted to modify the composition of the final product.

When feeding BSF larvae with almond byproducts of different compositions, Palma et al. (2020) obtained frass with 12.3–22.3 g kg^{-1} N, 17.9–44.6 g kg^{-1} K and 0.22–0.82 g kg^{-1} P. These differences were caused by the different carbohydrate, protein and fiber contents of the feed substrates. The higher levels of sugar/starch and proteins, and lower levels of fibers favored the accumulation of nutrients in frass. Sarpong et al. (2019) fed the BSF larvae with municipal solid waste of varying composition and found that the N, P and K contents increased substantially after the larvae consumed the substrate, reaching the respective concentrations of 4.8, 0.9 and 0.6 g kg^{-1} in the frass. Such an

Table 1
Frass produced in black soldier fly (*Hermetia illucens*) larvae composting of biodegradable materials.

Feed substrate	Material reduction (% DM)	Frass production per ton of waste (kg DM)	Larval density (larvae m ⁻²)	Composting time (d)	Reference
Household food waste	79.9	201	20,000	14	Lalander et al. (2020)
Fruits and vegetables	60.0	400	–	–	Giannetto et al. (2020)
Fruits and vegetables	65.2	348	6,000	20	Meneguz et al. (2018)
Fruits	70.8	292	6,000	22	Meneguz et al. (2018)
Bread waste	59.2	408	45,000	12	Lopes et al. (2020a, 2020b)
Poultry slaughterhouse waste	30.7	693	18,000	13–14	Gold et al. (2020a, 2020b)
Fish waste + bread waste	57.6–70.1	229–424	45,000	11–18	Lopes et al. (2020a, 2020b)
Chicken manure	75.6	244	7,000	–	Bortolini et al. (2020)
Cow manure	34.4–48.8	512–656	–	19–20	Rehman et al. (2019)
Fish waste + banana/orange peels	61.9–75.8	242–381	20,000	14–21	Isibika et al. (2021)
Bananas	63.4	366	45,000	14	Scala et al. (2020)
Apples	64.4	356	45,000	16	Scala et al. (2020)
Brewers spent grain	68.5	315	45,000	11	Scala et al. (2020)
Spent mushroom	42.3	577	–	20	Cai et al. (2019)
Canteen waste	37.9	621	2,000	9	Gold et al. (2020a, 2020b)
Vegetable canteen waste	58.4	416	2,000	9	Gold et al. (2020a, 2020b)
Mill byproducts	56.4	436	2,000	9	Gold et al. (2020a, 2020b)
Winery byproducts	53.0	470	6,000	22	Meneguz et al. (2018)
Brewery byproducts	38.7	613	–	15	Liu et al. (2018)

Table 2
Chemical attributes of different frass derived from black soldier fly (*Hermetia illucens*) larvae composting of biodegradable waste streams.

	Chemical properties													pH	C/N ratio	Reference
	C	N	P	K	Ca	Mg	Na	Fe	Cu	Mn	Zn					
	(%)				(g kg ⁻¹)			(mg kg ⁻¹)								
Gainesville diet	35.2	3.8	5.2	4.1	45	8.0	3.0	600	46.1	–	140	8.8	8.0	Setti et al. (2019)		
Distiller's grains	–	3.4	0.8	1.1	13	3.0	5.0	125	15	45	90	–	–	Yildirim-Aksoy et al. (2019)		
Brewery spent grain	38.6	3.6	0.5	0.3	9.7	1.0	–	310	25	109	182	7.3	10.7	Anyega et al. (2021)*		
Okara and wheat bran	37.1	4.8	1.0	0.9	1.3	0.1	–	26	2.2	4.2	0.1	7.5	7.7	Song et al. (2021)		
Okara and wheat bran	30.6	3.2	0.8	0.5	0.8	0.2	–	26	0.7	2.3	0.1	7.7	9.6	Song et al. (2021)*		
Household waste	35.8	2.2	0.5	0.7	10	0.9	0.8	240	10	10	10	7.4	16.6	Kawasaki et al. (2020)		
–	41.8	3.3	3.4	2.4	4.0	10	2.6	–	–	–	–	9.0	12.6	Gärttling et al. (2020)		
Wheat bran	35.7	2.8	1.4	2.3	–	0.3	–	15	8.9	19.4	15	6.8	16	Watson et al. (2021)		
Brewery spent grain	35.2	2.1	1.2	0.2	0.2	0.2	–	–	–	–	–	7.7	16.8	Beesigamukama et al. (2020b)		
Fresh okara	37.1	5.1	0.3	1.9	16.8	10.5	–	3.7	0.9	0.2	1.7	7.3	7.3	Chiam et al. (2021)		
Chicken manure	23.6	2.3	1.1	1.8	–	–	–	–	–	–	–	8.0	16.4	Liu et al. (2019)		
Pig manure	26.8	2.4	2.1	1.0	–	–	–	–	–	–	–	8.7	17.6	Liu et al. (2019)		
Chicken feed	47.9	2.6	–	–	–	–	–	–	–	–	–	6.2	18.5	Klammsteiner et al. (2020)		
Grass cuttings	44.3	2.4	–	–	–	–	–	–	–	–	–	5.4	18.2	Klammsteiner et al. (2020)		
Fruits and vegetables	48.8	1.8	–	–	–	–	–	–	–	–	–	5.6	26.6	Klammsteiner et al. (2020)		
Cow manure	27.7	1.9	1.0	0.2	–	–	–	–	–	–	–	8.4	15.1	Liu et al. (2019)		
Vegetables	38.7	2.8	1.5	3.3	15	7.0	0.3	896	19	149	137	8.6	13.8	Menino et al. (2021)		
Average	36.6	2.9	1.6	2.4	11.6	3.7	2.3	249.1	14.2	42.4	64.0	7.5	14.5			
Median	36.5	2.8	1.1	1.5	9.9	1.0	2.6	125.0	10.0	14.7	15.0	7.6	15.6			

*An asterisk indicates the adoption of thermophilic composting as a post-treatment for the frass. The references without an asterisk indicate that the referred study was conducted with fresh frass.

increase was hypothesized to occur because of the substrate's dry matter reduction over time and by the action of microorganisms excreted by the larvae in the growing media. Song et al. (2021) fed the BSF larvae with a mixture of okara and wheat bran, generating frass with 47.8 g kg⁻¹ N and 0.98 g kg⁻¹ K. Beesigamukama et al. (2020b) evaluated a frass produced by BSF larvae fed with brewer's spent grains that had 21 g kg⁻¹ N, 11.6 g kg⁻¹ P and 1.7 g kg⁻¹ K.

In general, the frass derived from BSF larvae has lower

concentrations of K in comparison to the other major nutrients. In some commercial BSF larvae frass (no specification on its origin), Choi et al. (2009) reported a K concentration of 0.1 g kg⁻¹, while Yildirim-Aksoy et al. (2019) reported 11 g kg⁻¹ of this same element in BSF larvae frass. There was no information about the feed substrate used during the larval growing period in these studies. The macronutrient K is not easily recovered from organic materials and it is sourced extensively from geological materials through mining (Manning, 2010). This and other

elements are usually found at low concentrations in BSF larvae frass (e.g. Mg, Mn and Cu), regardless of the feed substrate provided to the larvae (Table 2).

3.2. Provision of frass-derived nutrients for growing plants

Following the discovery of adequate plant nutrients in BSF larvae frass, many other investigations were duly conducted with the aim of understanding whether this product could be used as a fertilizer. To the authors' knowledge, the first study evaluating food waste-derived BSF larvae frass was carried out by Choi et al. (2009), who compared this product with a commercial fertilizer (unspecified origin) of similar nutrient composition, in terms of N, P, K and organic matter. The authors compared the number of leaves, leaf length and width and nutrient accumulation in Chinese cabbage plants and found that they were all of similar values, with exception for the P absorption by plants, which was lower when fertilized with frass. Alattar et al. (2016) tested BSF larvae frass as a fertilizer for maize plants, using a 1:2 (w/w) frass to soil mixture, without mentioning the nutrient composition of the frass. The authors reported that frass impacted plant growth (dwarf plants and fewer leaves) more negatively than did a microaerobic fermentation product made from the same feed substrate (food waste) used to feed the larvae. According to the authors, the reduced growth could be caused by the high concentrations of ammonia in the frass.

In a study conducted by Gärtling et al. (2020), three by-products from BSF larvae treatment of biodegradable wastes were tested as soil amendments in a pot trial with maize plants: the frass, larval skins and dead adult flies; in three levels of nutrient addition, 180 and 215 kg N ha⁻¹ and 75 kg P₂O₅ ha⁻¹ for each byproduct. The frass delivered poor growth results (yield, dry matter production, leaf area and nutrient use efficiency) in comparison to the other by-products and the controls (organic and chemical commercial fertilizers). The author attributed the poor fertilization property to the frass being an P-dominated fertilizer, rather than a N-dominated. In addition, the poor growth of the test crop was indicative that the frass may not have an optimal nutrient composition for certain crops. Kawasaki et al. (2020) assessed the fertilizing potential of BSF larvae frass in *Brassica rapa* and recommended an application rate of 1/20–1/30 of frass in relation to the amount of soil, in order to benefit growth, as plant growth was impaired with yellow leaves when applied at a higher application rate (1/10). Quilliam et al. (2020) tested BSF larvae frass made from poultry waste, brewery waste and green market waste as fertilizers for growing maize, pepper and shallots in a field experiment in Ghana. The authors conducted multiple field trials using 2.5 to 10 t ha⁻¹ of the three frass types alone and in combination with chemical (NPK type) fertilizers. Generally, the authors found similar growth responses in plants when compared to fertilization with chicken manure, which was the preferred local fertilizer. In addition, it was found that plants responded better when a combination of frass and the chemical NPK fertilizer was used.

Chiam et al. (2021), tested okara-derived BSF larvae frass as a fertilizer for lettuce plants, mixing frass with soil at 10, 20 and 30% concentrations (v/v). The frass had high concentrations of N (50 g kg⁻¹), P (0.3 g kg⁻¹) and K (2 g kg⁻¹). Interestingly, the general application (20%–30%) of frass resulted in poor growth of lettuce, except for when the frass level was at 10%. The authors speculated that this undesired growth response at high frass levels may be attributed to the low C/N ratio of the fertilizer (7.2), which induced rapid mineralization of nutrients in the soil. Contrary to this, Menino et al. (2021) observed a positive response and steady growth of ryegrass when applying six doses of BSF larvae frass as fertilizer, corresponding to 25–150% of the total demand of N by this species (estimated to be ca. 140 kg N ha⁻¹). In addition, increased fertility of soils amended with frass, in relation to higher soil organic matter, P₂O₅ and K₂O concentrations was found, which is something that is generally considered positive when using organic fertilizers rather than relying on chemical inputs.

In addition to having higher concentrations of organic matter in

frass-amended soils, another beneficial aspect of using frass as a bio-fertilizer concerns the rates of plant available nutrients. In a recent study, Beesigamukama et al. (2021a) applied a five-week composted frass in an acric ferralsol and evaluated the mineralization of nutrients over 125 days. They reported net immobilization during 30–60 days in the amended soil, but over time, the release of N (mostly in the form of ammonium), P and Mg in the soil was significantly higher than the control treatment, without any fertilizer. These data were indicative that the nutrients in the frass were mineralized. The immobilization/mineralization of nutrients from organic fertilizers is highly dependent on its characteristics, such as C/N ratio, biological stability, among other factors (Chen et al., 2014). It is therefore plausible that due to the great variability of frass compositions and their associated structural matrices observed in the literature (Table 2), the behavior of these products in the soil will be highly variable as well.

In a study conducted by Rummel et al. (2021), different BSF larvae frass were applied in a Haplic Luvisol of silty loam texture at doses of 170 and 510 kg N ha⁻¹. The authors reported that the frass with the lowest C/N ratio resulted in N mineralization and accumulation. In addition, they found that the dynamics of mineralization over time, for both C and N, depended on the quality of the substrate used to feed the larvae. The rates of N mineralization/immobilization from frass in the soil have been found to directly related to the amount of ammonium N in the frass, likely because this is the preferred source by microorganisms (Beesigamukama et al., 2021a). Accordingly, Rummel et al. (2021), reported that a high provision of C and N in the soil immediately after frass application led to elevated N₂O emissions, and depending on the form of C in frass, it could contribute less to the microbial biomass in the soil, and consequently to the soil organic carbon sequestration. Rummel et al. (2021) concluded that due to the widely varying composition and its behavior in the soil, frass fertilizing capacity is still not completely understood. As a consequence of this, frass could potentially have a higher carbon footprint than expected.

3.3. Stability of frass and the adoption of post-treatments

Both positive and negative observations have been reported for growth studies involving frass. It is plausible that the poor growth could be attributed to the lack of stability of the frass-compost in some of the studies. In some cases, the frass was post-composted (Chirere et al., 2021; Song et al., 2021; Anyega et al., 2021), while in other studies, it was used as a freshly produced frass (Beesigamukama et al., 2020a; Menino et al., 2021). An important consideration when applying organic fertilizers during cultivation is the compost's maturity and stability. Even though a compost, such as frass, contains adequate amounts of nutrients for crop production, a general lack of stabilization and phytotoxicity can result in poor growth if the compost is not stabilized (Setti et al., 2019). Generally, it is advocated that a compost should be stabilized before being added to the soil, in order to promote the degradation of its organic matter and the mineralization of nutrients. Otherwise, these nutrients may be unavailable in the soil for root assimilation, causing negative nutritional anomaly in plants (Bernal et al., 2009; Chen et al., 2014).

The stabilization of organic fertilizers is related to the decomposition and transformation of organic matter, resulting in lower C/N ratio and phytotoxicity (Raj and Antil, 2011; Gavilanes-Terán et al., 2016). The composting process with BSF larvae is typically rapid (12–15 days), and as a result the frass-compost is often immature and biologically unstable (Setti et al., 2019). Therefore, it is preferred that this product should be given some sort of post-treatment (e.g. thermophilic composting), in order to stabilize it and making it suitable as a biofertilizer for cultivation.

One of the early research on the use and stability of BSF larvae frass as a soil amendment was reported by Alattar et al. (2016). They evaluated the growth of corn (*Zea mays*) in soil amended with BSF larvae processing residues from kitchen scrap. The authors blended the frass

with soil in a 1:2 ratio (weight weight basis), without detailing the chemical composition of the frass, and cultivated the plants for ten weeks. Stunted growth of plants was found, which according to the authors occurred due to the presence of phytotoxic compounds in the frass, even though they did not evaluate these compounds.

Subsequent studies have evaluated the phytotoxicity of BSF larvae frass. [Xiao et al. \(2018\)](#) evaluated the frass production from BSF larvae treatment of chicken manure inoculated with *Bacillus subtilis*. They found that the phytotoxicity levels of the fertilizer, tested in Chinese cabbage and rape seeds, were reduced over time, reaching a germination index of above 66% after 13 days of BSF larvae treatment. Similarly, [Setti et al. \(2019\)](#) assessed the phytotoxicity of BSF larvae frass from a treatment using wheat bran, alfalfa meal and corn meal as feed substrate for the larvae, using lettuce (*Lactuca sativa*) seeds. They found that the germination indexes were above 70%, indicating no sign of phytotoxicity. Conversely, older plants showed poor growth when receiving high levels of BSF larvae frass.

When treating chicken, pig and cow manure with BSF larvae, [Liu et al. \(2019\)](#) demonstrated high levels of phytotoxicity, by means of a seed germination test, in the treatment with chicken manure, while the other manures generated a stabilized frass with reduced electrical conductivity and higher germination index. The authors hypothesized that the lack of maturity in the chicken manure-derived frass occurred because of the high electrical conductivity and concentration of N-NH_4^+ in the feed substrate. As highlighted by [Lopes et al. \(2019\)](#), such an initial and high concentration of N-NH_4^+ could be reduced by further composting the frass.

The stability and behavior of organic fertilizers in soil can be assessed by a number of ways in addition to performing bioassays (e.g. germination tests); the C/N ratio can also give some useful insights. The C/N ratio of fertilizers is recommended to be between 20 and 40, in order to avoid the possible immobilization of certain mineral nutrients in the soil, causing poor growth ([Chen et al., 2014](#)). The C/N ratio of BSF larvae frass varies somewhat depending on the feed substrate used for the larvae; the frass always has a lower C/N ratio than the feed substrate provided ([Sarpong et al., 2019](#)). For instance, the C/N ratio of household biodegradable waste was reduced from 48 to 17 during the BSF larvae composting process ([Kawasaki et al., 2020](#)), while the C/N ratio of almond byproducts was reduced from 73 to 20 ([Palma et al., 2020](#)). According to [Beesigamukama et al. \(2021b\)](#), the feed substrates used for BSF larvae growth should have a C/N ratio ranging between 15 and 30, in order to generate a stable, and non-phytotoxic frass.

A short post-composting period of four days of BSF larvae frass compost was done by [Chirere et al. \(2021\)](#), resulting in higher growth of Swiss chard in comparison to unfertilized soil and having similar growth in relation to inorganic NPK fertilizer. However, the authors did not measure any stability-related characteristic of the frass, besides mentioning its C/N ratio (ranging from 8.2 to 9.0); nor was the fresh frass evaluated for any potential phytotoxicity. This was however done by [Song et al. \(2021\)](#), who compared the impact of fresh BSF larvae frass (3 weeks old) with naturally composted and aerated composted frass (both 8 weeks old) on the growth of *B. rapa*, and found that application rates above 10% of the fresh frass caused stunted growth and reduced biomass production in the plants. However, after composting the frass for five weeks, applications of up to 40% resulted in better growth, further suggesting the need for stabilizing this product before applying it to the soil. Similarly, [Anyega et al. \(2021\)](#) composted frass for 112 days and compared its fertilizing potential (in tomatoes, kale and French beans) with other soil amendments, such as commercial organic fertilizers and chemical fertilizers, under greenhouse and field conditions. Generally, it was found that a combination of composted frass and NPK fertilizers delivered the best results in terms of growth, crop yield, N uptake and nutrient use efficiency, in comparison to unfertilized soil and soil fertilized exclusively with frass. However, the sole application of composted BSF larvae frass also resulted in positive results for the three species tested. The same combinational approach of mixing various

composts and NPK fertilizers generally produced positive growth results. For example, [Song et al. \(2015\)](#) demonstrated that the inclusion of vermicompost enhanced the beneficial effects of beneficial microorganisms on soil properties and crop yield.

One major question regarding the stabilization of organic fertilizers is the formation of humic substances over time. Humic substances (e.g. humic and fulvic acids) are mixtures of multiple organic compounds derived from the decomposition of organic materials in the soil, constituting the largest share of the soil organic matter and playing a paramount role in the fertility of soils and plant nutrition ([Canellas et al., 2015](#)). The formation of humic substances during composting of organic materials is one of the main indicators of compost stability (resistance to decomposition) and maturity (use for a determined purpose) ([Zhou et al., 2014](#)). These substances contribute to several soil fertility parameters by regulating soil acidity, improving the cation exchange capacity, increasing the water holding capacity, improving the uptake of nutrients and stimulating plant growth ([Sutton and Sposito, 2005](#); [Canellas and Olivares, 2014](#); [Conselvan et al., 2018](#); [Abbott et al., 2018](#); [Olaetxea et al., 2018](#)). The relation between humic substances and BSF larvae treatment of biodegradable waste is unclear and deserves further investigation. [Liu et al. \(2020\)](#) investigated the impact of BSF larvae composting of a mixture of chicken, pig and cow manure, with emphasis on the humification and speciation of trace elements (Cu and Zn), and reported that fulvic acids were reduced over time, in contrast to humic acids, which were not reduced. Using the same type of feed substrate, [Wang et al. \(2021\)](#) demonstrated that BSF larvae were efficient in increasing the humification degree of manures. [Song et al. \(2021\)](#) composted BSF larvae frass for eight weeks and identified several benefits of the post-treatment in this product (e.g. higher nutrient content, higher stability and lower environmental impact), even though the concentration of humic substances was not assessed. The authors found high concentrations of ethers in the aerated-composted frass and hypothesized that this finding indicate the formation of humic acids and other metabolites in the compost.

Another possibility for the further stabilization of frass, even though it has not yet been investigated, is using this material for vermicomposting with earthworms. Earthworms play an active role in the degradation of biodegradable materials, and these organisms are well known to accelerate the stabilization of organic matter ([Frederickson et al., 1997](#); [Kaviraj and Sharma, 2003](#)). The biologically unstable frass from BSF larvae composting could be submitted to vermicomposting. Other possibilities for the use of frass could be taken into consideration, such as providing BSF larvae frass to mealworms, an insect species with a longer life cycle in comparison to BSF that also generates a nutrient-rich and stable fertilizer ([Houben et al., 2021](#)), or using frass in anaerobic digestion for biogas production, which could render an economically feasible process ([Lalander et al., 2019](#)).

3.4. Microbial composition of the BSF larvae frass

One of the major unanswered questions regarding the use of organic fertilizers in agriculture concerns the microbiological composition of these products, and their capacity in altering the soil microbiome and benefiting plants in multiple ways ([Lugtenberg and Kamilova, 2009](#); [Singh et al., 2011](#); [Pérez-Montaño et al., 2014](#)). The presence of beneficial microorganisms in the soil has been demonstrated to develop higher nutrient use efficiency, resistance to abiotic stress conditions and improve plants growth ([Poveda et al., 2019](#)). The microbiota of BSF larvae frass was assessed by [Wynants et al. \(2019\)](#) and [Gold et al. \(2020b\)](#), and both studies revealed that the microbial composition of frass changes according to the feed substrate supplied to the larvae. Similarly, [Kawasaki et al. \(2020\)](#) fed BSF larvae with household organic waste and found that the resulting frass had a varied composition of microorganisms, both in comparison to the initial household waste used to feed the larvae and to the other fertilizers (cow, horse and poultry manure), with higher abundances of *Sporosarcina* spp., *Corynebacterium*

spp. and *Bacillus* spp. Nevertheless, the authors highlighted the importance of performing in-depth evaluations of the microbiota of BSF larvae frass, as a few microorganisms found in the frass could cause diseases in plants, such as members of the family Xanthomonadaceae, belonging to the phylum Proteobacteria. These studies did not mention the possible benefits of frass' microbial composition on plant growth by enhancing the soil-microbe-plant interactions. Specifically, the studied frass contained a great abundance of *Lactobacillus* spp., *Bacillus* spp., *Actinobacteria* spp., and *Pseudomonas* spp.; these microbial genera are well known to be beneficial for plant growth (Lugtenberg and Kamilova, 2009; Babalola, 2010; Ahemad and Kibret, 2014).

Current agricultural practices are highly dependent on chemical fertilizers, which are known to deteriorate cropland soil fertility when used in the long-term (Bünemann et al. 2006; Choudhary et al., 2018). The microbiological compositions of organic fertilizers could benefit more sustainable production systems. As highlighted by several major reviews (Ahmad et al., 2020; Pérez-Montaño et al., 2014; Pathania et al., 2020), a diverse set of microorganisms, especially bacteria, play important roles as plant growth-promoting rhizobacteria (PGPR), by enhancing crop productivity, stimulating plant growth and even suppressing pathogens. These microorganisms act in the rhizosphere, which is the fine region of soil that is influenced by the secretions of plant roots (root exudates) and can be stimulated by the input of organic fertilizers, benefiting the soil and the plant as a whole (Lugtenberg and Kamilova, 2009; Berendsen et al., 2012). Based on the extensive knowledge on the enrichment of soil microorganisms by the use of organic fertilizers (Treonis et al., 2010; Abbott et al., 2018), it is likely that BSF larvae frass contains different groups of PGPRs that could enhance crop quality, and increase the sustainability of traditional cultivation, replacing the use of conventional chemical fertilizers.

While multiple groups of beneficial microorganisms can improve soil quality, plant performance and crop yields (Balestrini et al., 2017; Maçik et al., 2020), other assemblages of microorganisms could compose a plausible risk when using organic fertilizers in agriculture, such as pathogenic bacteria and fungi. Several worldwide legislations (EU Regulation 2019/1009; Brasil, 2006; EPA, 2020) prohibit the application of fertilizers containing *Salmonella* spp., *Escherichia coli*, thermotolerant coliforms and other microorganisms, at certain concentrations on cropland. Similarly, pathogenic fungi and/or their spores should not be present in organic fertilizers, in order to avoid cross contaminations to the field (Tournas, 2005).

The treatment of contaminated biodegradable waste with BSF larvae has been proven to be effective in significantly inactivating different strains of *Salmonella* spp. and *E. coli*, even though the remaining frass still holds small concentrations of these bacteria (Lopes et al., 2020a). Erickson et al. (2004) observed the inactivation of *Salmonella enterica* (up to 4-log₁₀ reduction) and *E. coli* (up to 5-log₁₀ reduction) in chicken manure, while Lalander et al. (2015) reported significant reductions of *Salmonella* spp. (>7 log₁₀) in a blend of dog food, human faeces and pig manure, while no inactivation of thermotolerant coliforms were found. Moreover, other biological contaminants, such as the eggs of *Ascaris* spp., have been found to not be affected by BSF larvae treatment (Lalander et al., 2013). Similarly, fungi-derived contaminants (e.g. aflatoxins) are partially reduced in the BSF larvae treatment, but these toxins remain in the frass, as well as in the larval biomass after the treatment of contaminated biodegradable waste (Bosch et al., 2017), which could compromise its use in agriculture due to safety reasons. Therefore, future research must focus on the need for post-treatments of BSF larvae frass, aiming not only to stabilize it biologically, but also at inactivating chemical and biological contaminants in this product.

4. Frass as a source of bioactive compounds

In addition to the macro- and micronutrients, organic fertilizers are usually a good source of growth promoting compounds that in recent years have gained great attention, namely bioactive compounds. These

biologically active compounds are responsible for another lesser-known aspects of plant nutrition (e.g. signaling), other than the provision of mineral nutrients as building blocks for structural growth (Yakhin et al., 2017). Bioactive compounds include a set of substances, such as peptides, amino acids, humic substances and phytohormones, as well as a diverse group of microorganisms (Du Jardin, 2015; Yakhin et al., 2017; Xu and Geelen, 2018). When applied in the soil, these substances stimulate the natural processes of the soil-rhizosphere medium and enhance plants' metabolism, thereby improving growth as a whole. Based on the knowledge existing on the processing residues of other invertebrates, BSF larvae frass could also be an abundant source of bioactive compounds that can be harvested and used in sustainable agriculture practices (Wong et al., 2020; Poveda, 2021) (Fig. 1).

4.1. Plant biostimulants

Plants are often subjected to biotic and abiotic stresses that affect their metabolism, preventing crops to achieve their full potential in terms of productivity. In order to avoid these negative impacts, plants should be provided with adequate amounts of water, nutrients and plant growth regulators (including bioactive substances), compounds that are not nutritive but affect plants physiology in various ways (Yakhin et al., 2017; Cortleven et al., 2019; Gupta et al., 2020; Schmitt and de Vries, 2020). These compounds are represented by several substances, such as phytohormones (e.g. auxins, gibberelins, cytokinins), humic substances (e.g. humic and fulvic acids), and many other sets of molecules (Dodd et al., 2010; Calvo et al., 2014; Wong et al., 2020). In addition, some groups of microorganisms are also classified as biostimulants, usually named plant growth-promoting microorganisms (PGPM), due to their ability to modulate multiple metabolic functions and physiological mechanisms in the plant metabolism (Pérez-Montaño et al., 2014; Sofo et al., 2014). According to the European biostimulants industry council (EBIC), the definition of biostimulants is "substances and/or microorganisms whose function when applied to plants or the rhizosphere is to stimulate natural processes to enhance/benefit nutrient uptake, nutrient efficiency, tolerance to abiotic stress and crop quality".

Multiple biostimulants have been reported in the worm cast and frass of invertebrates, namely earthworms and mealworms, respectively (Zhang et al., 2014; Aremu et al., 2015; Poveda et al., 2019). Specifically, as humic acids are found in frass, the biochemical changes associated with humic acid metabolism and its interaction with soil microorganisms are known to release several biostimulants like auxins and cytokinins (Pizzeghello et al., 2013; Conselvan et al., 2018; Nardi et al., 2018). It is therefore plausible that the occurrence of phytohormonal type of biostimulants in frass is through its humic acid metabolism (Olaetxea et al., 2018). As previously mentioned in this review, BSF larvae frass is rich in microorganisms (Gold et al., 2020b), some of which may have biostimulatory capacity. For instance, multiple species of *Bacillus* spp. produce phytohormones with beneficial traits for plants, such as auxins (promotes growth and increases drought tolerance), cytokinins (stimulate root exudation), jasmonic acid (induce salinity stress tolerance), gibberelins (increases seed germination, enhance nutritional metabolites, regulates endogenous phytohormones, induce thermotolerance), among others (Poveda and González-Andrés, 2021).

Vermicompost, the resulting product of organic materials assimilated by earthworms, is one of the most used and studied organic fertilizer in the world (Van Groenigen et al., 2014; Wong et al., 2020). Despite its relatively low mineral nutrient content in comparison to chemical fertilizers, it has been demonstrated to bring multiple benefits to plants, due to the presence of multiple biostimulants (Wong et al., 2020). Similarly, the frass from mealworms (*Tenebrio molitor*) present several groups of PGPM with biostimulant action, such as *Bacillus* spp. (promotes root development and nutrient assimilation) (Ab Aziz et al., 2015; Poveda and González-Andrés, 2021), *Pseudomonas* spp. (produces phytohormones, biocontrol of plant diseases) (Sivasakthi et al., 2014),

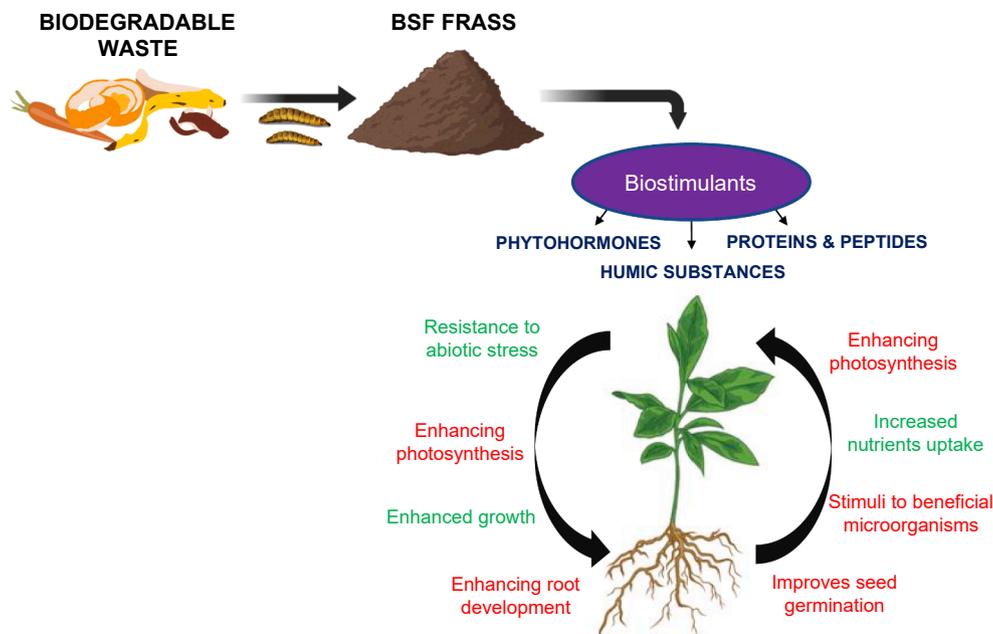


Fig. 1. Schematic representation of the action of biostimulants derived from black soldier fly larvae (*Hermetia illucens*) frass. The beneficial traits of frass are shown in green (already demonstrated in frass from other insects) and red (not yet reported for insect frass).

Sphingobacterium spp. (solubilizes P and increases tolerance to abiotic stress) (Ahmed et al., 2014), in addition to showing benefits such as N fixation, phosphate and K solubilization, auxins production, among others (Poveda et al., 2019).

Although BSF larvae frass has been evaluated in various fertilization-linked studies, the topic on biostimulants has not been addressed, with a single exception; Antonov et al. (2020), evaluated frass as a biostimulant (using a 1% aqueous extract) in rubber trees, and reported a 20% higher yield in the acquisition of pine resin. However, the authors did not present any characteristics of the frass, for either nutrients or bioactive compounds. Nevertheless, several studies have demonstrated positive effects on plants growth and metabolism when using BSF larvae frass as an organic fertilizer, as mentioned before, in particular after a post-treatment of the frass has been undertaken. It is noteworthy that the microbial characterization of BSF larvae frass revealed multiple groups of microorganisms with possible biostimulant action (Wynants et al., 2019; Gold et al., 2020b; Klammersteiner et al., 2020; Tan et al., 2021), even though the authors did not further explore this subject in their studies. Nonetheless, multiple microorganisms are plant growth promoting, but not all strains of a certain genus or even species shall have an equivalent competence in relation to its biostimulatory action (Huang et al., 2013; Poveda and González-Andrés, 2021). For detailed information on biostimulants in agriculture, consult Xu and Geelen (2018) and Yakhin et al. (2017).

4.2. Biocontrol

A final aspect that should be considered when using BSF larvae frass is the potential for suppressing diseases in plants. The plant disease suppression ability of frass could be attributed to the presence of certain biostimulants (bioactive compounds and certain microorganisms) and chitin-rich compounds. Schmitt and De Vries (2020) highlighted that finding biostimulants/biocontrol properties in BSF larvae frass could reduce the world's reliance on unsustainable pesticides. The biocontrol of plant diseases through biostimulants has been extensively reviewed (for reviews, see Naseem et al., 2014; Ahmad et al., 2020; Cortleven et al. 2019; Hamid et al. 2021; Gupta and van Staden 2021) and thus, the discussion here focus on the BSF chitinous exuvia.

The BSF larvae undergoes multiple developmental stages, passing

through six or seven larval instars (of which the prepupa is the final instar), followed by the pupal stage, from which an adult emerges (Tomberlin et al., 2002; Gligorescu et al., 2018). At each instar, the larvae molt and their chitinous exuvia remained in the frass. Chitin is known to display disease suppressive functions in the soil; for examples, plant-parasitic fungi (Postma and Schilder, 2015) and nematodes (Oka, 2010). The mechanism behind this suppression seems to be related to the presence of chitin in the eggshells of nematodes (Warton and Jenkins, 1978), as well as in the cell wall of fungal plant pathogens (Nobel et al., 2000). Thus, when chitin-rich materials are added to the soil, it could lead to an increased number of chitinolytic microorganisms in the soil enhancing the inactivation of fungi and nematodes (Oka, 2010).

The content and structural forms of chitin in BSF was thoroughly studied during all developmental stages of this fly (larvae, prepupae, pupae and adults) (Wang et al., 2020). However, only a few studies investigated the benefits of BSF-derived chitin in practical applications. For instance, Vilela et al. (2020) demonstrated a possible beneficial effect in the modulation of the immune system of broilers, when feeding the animals with BSF larvae meal. Similarly, Kroeckel et al. (2012) found positive effects in growth performance of juvenile turbot when feeding the fish with a BSF prepupae meal, which according to the authors was made possible because of a higher feed intake, due to the presence of chitin. Regardless of these beneficial effects for animals, to the best of the authors' knowledge, BSF chitin has not been evaluated in any cultivation practices. Considering the beneficial effects of chitin against fungal pathogens and other pests (Oliveira Jr. et al., 2008; Sharp, 2013), and the presence of chitin in the frass from BSF composting (Klammersteiner et al., 2020), it is likely that this product display beneficial effects in the soil–plant system, in relation to pathogen control.

5. Conclusions and future perspectives

One of the main goals of this review was to assess the potential of BSF larvae frass as an organic fertilizer to be used in sustainable cultivation, in light of the new development across all green (e.g. agriculture, horticulture, ecological restoration) and brown (e.g. soil health, natural soil fertility, erosion management) sectors globally.

Frass from BSF larvae composting of biodegradable wastes is a very promising product that still requires multi-disciplinary investigative

strategy, as highlighted in this review. The composition of frass is highly variable and, in particular, the concentrations of P, K and micronutrients were highly dependent on the feed substrate. There is evidence that the frass does not have an optimal nutrient composition for some crops (P-dominated fertilizer). The addition of another, N-dominated input to the frass could potentially be a practical solution to produce a well-balanced fertilizer. Nutrient supplementation to frass-based fertilizer is another way to be investigated in future studies, as it may be needed in order to meet the precise nutrient requirements of the crops.

There are several studies indicating the potential of BSF larvae frass to increase yield, while others reported negative growth associated with plausible phytotoxicity of the frass. BSF larvae frass is a biologically unstable product, due to the rapid composting process and the presence of substances with potential phytotoxic properties. In order to enhance the compatibility of this product as a fertilizing amendment, frass likely has to undergo some sort of post-treatment for stabilization. Alternatively, one could consider mixing the frass with another fertilizer product to achieve better matrix stabilization while enhancing the combined fertilizer efficacy (Sani and Yong, 2022). Future research should investigate the impact of post-treatment processes, such as thermophilic composting, including the duration of thermophilic, mesophilic and maturation stages, temperatures reached, in terms of reduction of phytotoxicity and the formation of humic substances. However, the emissions from the different post-treatments must be taken into consideration. In addition, frass could be submitted to vermicomposting with earthworms or provided to mealworms as feed to achieve a higher stabilization state.

Considering the enormous attention that bioactive substances and beneficial microorganisms in organic materials have received in recent years, BSF larvae frass should also be investigated in terms of the formation of substances such as humic and fulvic acids, phytohormones, short-chain proteins and amino acids, among others. The presence of biostimulants and plant growth-promoting rhizobacteria and fungi in BSF larvae frass is yet to be unraveled. This is one of the most relevant research topics currently, which could enhance the deployment of frass as a pivotal circular bioeconomy product for the development of sustainable cultivation across all green and brown sectors globally. A holistic approach, connecting the different steps of the process with the different aspects of interest, has to be undertaken for a more in-depth understanding of the full potential of this waste derived fertilizer product.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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