



## Assessment of the Short-Term Fertilizer Potential of Mealworm Frass Using a Pot Experiment

### David Houben<sup>1\*</sup>, Guillaume Daoulas<sup>2</sup> and Anne-Maïmiti Dulaurent<sup>1</sup>

<sup>1</sup> AGHYLE, UniLaSalle, Beauvais, France, <sup>2</sup> Ÿnsect, Evry, France

The forecasted growth of insect production in the next few years will generate high quantities of frass (insect excreta). Although frass is increasingly considered a potential fertilizer, the dynamics of nutrient supply by frass is still poorly understood. Here, we aimed at gaining insight into the short-term fertilizer value of frass from mealworm (Tenebrio molitor L.) in order to optimize its sustainable use in agroecosystems. Using a short-term pot experiment, we showed that, even though frass has a great potential to be used as a substitute of mineral NPK fertilizer, its N fertilizer potential is mediated by its rate of application. At 10 t ha<sup>-1</sup>, due to its fast mineralization coupled with improvement in microbial activity (assessed using Biolog EcoPlate), frass was as effective as mineral fertilizer to supply N to plant. By contrast, at 5t ha<sup>-1</sup>, the lower frass mineralization induced a reduced N uptake compared to its mineral control. Unlike N, frass was as effective as mineral fertilizer to supply P and K to plants irrespective of its application rate. This was attributed to the presence of P and K in a readily available form in frass. Taken together, our results indicate that mealworm frass supplies very rapidly N, P and K to plants but its effects on N dynamics should be better investigated to warrant its sustainable use as an alternative fertilizer for managing NPK nutrition in cropping systems.

Keywords: fertilizer, frass, insect excreta, mealworm, nitrogen mineralization, organic amendment, NPK, Tenebrio molitor

## INTRODUCTION

Fertilization of soils is essential to achieve the high yields required to feed an ever-increasing human population. However, the extensive use of chemical fertilizers leads to an increased consumption of energy and non-renewable resources (Chojnacka et al., 2020) while causing air and water pollution (Savci, 2012). In this regard, recent efforts have been channelized toward more sustainable resources for managing plant nutrition in cropping systems (Faucon et al., 2015). This has sparked a growing interest in the use of renewable feedstock from waste material to replace conventional fertilizers (Chew et al., 2019).

Recently, frass (insect excreta) has been considered a promising resource for managing plant nutrition in cropping system (Houben et al., 2020; Chavez and Uchanski, inpress). The growth of massive insect production in the near future to meet the need of finding alternative source of proteins (Derrien and Boccuni, 2018) is expected to produce frass in large quantities (Poveda, 2021). Due to its high content in nitrogen (N), phosphorus (P) and potassium (K) as well as the potential presence of beneficial microorganisms (Poveda et al., 2019), the use of frass as a fertilizer could help in reducing the use of agrochemicals. For instance, frass from black soldier

### OPEN ACCESS

### Edited by:

José Martinez, Institut National de recherche pour l'agriculture, l'alimentation et l'environnement (INRAE), France

### Reviewed by:

Margarita Ros, Spanish National Research Council, Spain M. Ángeles Gómez-Sánchez, University of Salamanca, Spain

\*Correspondence:

David Houben david.houben@unilasalle.fr

### Specialty section:

This article was submitted to Waste Management in Agroecosystems, a section of the journal Frontiers in Sustainable Food Systems

> **Received:** 25 May 2021 **Accepted:** 29 June 2021 **Published:** 28 July 2021

### Citation:

Houben D, Daoulas G and Dulaurent A-M (2021) Assessment of the Short-Term Fertilizer Potential of Mealworm Frass Using a Pot Experiment. Front. Sustain. Food Syst. 5:714596. doi: 10.3389/fsufs.2021.714596

1

fly (*Hermetia illucens* L.) was successfully used as an organic fertilizer to promote the growth of maize (Beesigamukama et al., 2020a; Gärttling et al., 2020) and ryegrass (Menino et al., 2021). In addition, frass from mealworm (*Tenebrio molitor* L.) showed great potential to be used as a partial or a complete substitute for mineral NPK fertilizer for the growth of barley (Houben et al., 2020) while stimulating soil microbial (Poveda et al., 2019) and earthworm activity (Dulaurent et al., 2020). However, as highlighted by Beesigamukama et al. (2020b), the fertilizer potential of frass may be affected by its rate of application in soil. Although the review by Chavez and Uchanski (inpress) shows promising results with the use of frass as a fertilizer, it also highlights that the optimal rate of frass application should be clarified because it may strongly differ between the couple of existing studies so far.

Moreover, the dynamics of nutrient supply by frass is still poorly understood. Knowledge on short-term availability of N and, to a lesser extent, P and K, after application of organic fertilizers is pivotal to optimize fertilizer use with benefits for the farmer and the environment (Gutser et al., 2005). Application of organic fertilizer does not always involve a short-term increase in availability of plant nutrients and crop productivity due to, among others, microbial immobilization which subsequently reduces short-term nutrient availability to the plants (Geisseler et al., 2010). As a result, several studies have pointed out that the use of organic fertilizers may compromise crop yield as compared to inorganic fertilizers because of the reduced input of readily available plant nutrients and the absence of rapid and short-term beneficial effects on microbial properties (Pimentel et al., 2005).

Because the upscaling of the insect industry which currently takes place calls for more research on the fertilizer potential of frass (Berggren et al., 2019), the present study aimed therefore at gaining insight into the short-term fertilizer value of frass from mealworm in order to optimize the sustainable use of frass as an alternative to mineral fertilizers.

## MATERIALS AND METHODS

### Frass

Frass (ŸnFrass) from mealworm (*T. molitor* L.) was provided in the form of powder by Ÿnsect (Paris, France), an industrial company farming this insect at the large-scale. The mealworms were fed exclusively on raw materials authorized by French and European regulations for farm animal feeds. The frass was provided and used as such, that is with no chemical input, making it a fertilizer compatible with organic farming and not subject to any specific restrictions. **Table 1** shows the chemical characteristics of the mealworm frass used in this study.

### Soil

The studied soil was sampled in Beauvais (Northern France) and was classified as a Haplic Luvisol. Soil characterization was carried out by Houben et al. (2020) and revealed that the soil was a silt loam (USDA classification) with 16% sand, 67% silt, and 17% clay. Organic C was 1.54% and total N was 0.18%. Available concentrations as assessed using the acetate ammonium-EDTA extraction (Houben et al., 2011) were Ca 3869 mg kg<sup>-1</sup>, Mg 101 mg kg<sup>-1</sup>, K 292 mg kg<sup>-1</sup>, P 72 mg kg<sup>-1</sup>. The cation exchange capacity (CEC) was 12.5 cmol<sub>c</sub> kg<sup>-1</sup> and pH was 7.8.

### Treatments

The frass was applied to the soil at a rate of 5 and 10 t ha<sup>-1</sup> (hereafter called "Frass-5" and "Frass-10," respectively). Two mineral treatments adding the same quantity of N, P and K as in the Frass-5 and Frass-10 treatments were achieved by mixing the soil with appropriate amount of inorganic nutrients (NH<sub>4</sub>NO<sub>3</sub>, KH<sub>2</sub>PO<sub>4</sub> and KCl) in solution (hereafter called "NPK-5" and "NPK-10" treatments, respectively). Untreated (hereafter called "Control") soil was also part of the experimental design.

### Pot Experiment

A pot experiment was conducted to assess the effect of frass on the nutrient availability for plants. Plastic plant pots (11.5cm diameter, 10.5-cm height) were filled with 450 g of each mixture in four replicates. Before sowing, the pots were placed in a controlled dark room and the mixtures were equilibrated during one week at 80% WHC. After the equilibration period, the pots were transferred to a greenhouse glass and were arranged according to a randomized design. In each pot, 1.5 g seeds of Italian ryegrass (*Lolium multiflorum* Lam.) were sown. The trials were conducted under controlled greenhouse conditions (temperature 18–25°C, 16-h photoperiod) with daily sprinkler watering. After 4 weeks, shoots were harvested by cutting 1 cm above the soil with ceramic scissors, dried (60°C; 72 h), weighed and crushed. The nutrient concentration of the aerial parts was then analyzed by ICP-AES after *aqua regia* digestion.

# Incubation Experiment: C and N Mineralization

The kinetics of frass mineralization were followed during laboratory incubations of control and frass treatments, based on the French normalization (AFNOR, 2016). An amount equivalent to 100 g of dry soil mixture was incubated in 1.2 L hermetic jars kept in a dark room at 22°C. The experiment was conducted in four replicates and lasted 32 days. The water content of the mixtures was adjusted at field capacity with demineralized water and controlled during the incubation period. In each glass jar, C mineralized as  $CO_2$  was trapped

TABLE 1 | Chemical characteristics of frass (data from Houben et al., 2020).

Organic C g kg <sup>-1</sup>	Total N g kg <sup>-1</sup>	Total K g kg <sup>−1</sup>	Total P g kg <sup>-1</sup>	Soluble fraction %Corg	Hemicellulose- like fraction %Corg	Cellulose-like fraction %Corg	Lignin-like fraction %Corg
393	50	17	20	49.3	31	15.2	4.4

in 30 mL of 1 mol L<sup>-1</sup> NaOH. The C-CO<sub>2</sub> trapped in NaOH was determined using the alkali absorption/conductivity method (Rodella and Saboya, 1999). As recommended by Doublet et al. (2011), the dynamics of C mineralization in soil was calculated by subtracting C-CO<sub>2</sub> mineralized in the control treatment to C-CO<sub>2</sub> mineralized from the frass treatment and the results were expressed as a percentage of the total organic C (TOC) in frass. Similar to the recommendations by Flavel and Murphy (2006) for other organic amendments, the release of mineral N from frass was predicted on the basis of the strong relationship between C-CO<sub>2</sub> mineralized from frass and N mineralized from frass found in our previous study (Houben et al., 2020) (see details of the regression equation in **Supplementary Figure S1**).

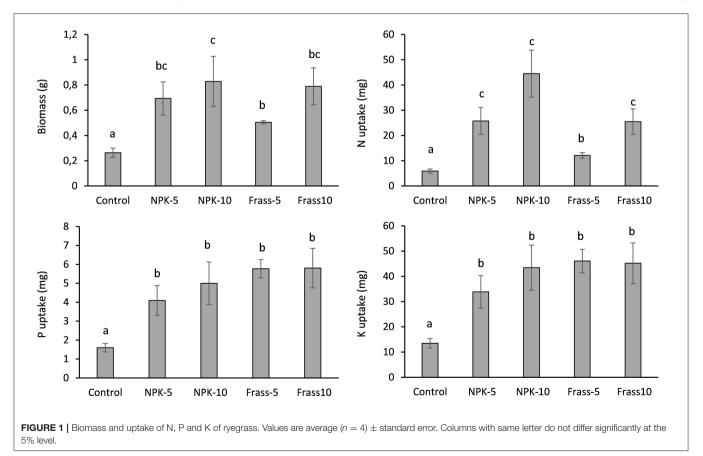
### **Community-Level Physiological Profiles**

The effect of frass on metabolic functions, including those involved in nutrient cycling, in soil was assessed by comparing the patterns of potential C source utilization by soil microbial communities under the Control, the NPK-10 and the Frass-10 treatments using Biolog Ecoplate. Briefly, each 96-well plate consisted of three replicates, each one comprising 31 sole C sources and a water blank. five grams of soil were shaken with 45 ml of sterile 0.85% NaCl for 30 min at 200 rpm and then diluted to 1:1000. Each plate well was inoculated with 150  $\mu$ L of the dilution and the plates were incubated at 25°C. Color development for each well was obtained in terms of optical density (OD) at 590 nm using an automated plate reader.

Kinetic curves suggested that after 72 h incubation time, the wells with the most active microbial communities reached the asymptote of color development. Therefore, this point was considered as the optimal incubation time for further statistical analyses, as suggested by Doan et al. (2013). The C sources were grouped into six categories representing different substrate guilds according to Sala et al. (2010): amino acids (Larginine, L-asparagine, L-phenylalanine, L-serine, glycyl-L-glutamic acid, L-theronine,), amines (phenylethylamine, putrescine), carbohydrates (D-mannitol, glucose-1-phosphate, D,Lalphaglycerol phosphate, beta-methyl-D-glucoside, D-galactonic acid-gamma-lactone, ierythritol, D-xylose, N-acetyl-Dglucosamine, D-cellobiose, alpha-D-lactose), carboxylic acids (D-glucosaminic acid, D-malic acid, itaconic acid, pyruvic acid methyl ester, Dgalactouronic acid, alpha-ketobutiryc acid, gamma-hydroxybutyric acid), phenolic compound (2-hydroxy benzoic acid, 4-hydroxy benzoic acid) and polymers (Tween 40, Tween 80, alphacyclodextrine, glycogen). Substrate average well color development (SAWCD) values for each substrate categories were calculated with the same equation: AWCD=  $\sum ODi/N$ , where ODi is the corrected OD value of the substrates within the substrate category and N is the number of substrates in the category.

### **Statistical Analyses**

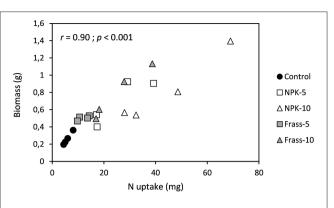
All recorded data were analyzed using descriptive statistics (mean  $\pm$  standard error) and normality was determined using



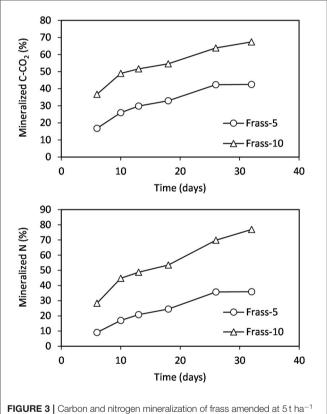
the Shapiro-Wilk test. One-way ANOVAs and Tukey's multiple comparison tests or Kruskal-Wallis and Mann-Whitney tests were used to compare biomass and nutrient concentrations in the shoot according to whether the distribution was normal or not, respectively. Pearson's correlation coefficient was used to analyze the relationship between biomass and N content. All statistical analyses were performed using R software version 3.5.0 and the package Rcmdr (Fox, 2005).

### **RESULTS AND DISCUSSION**

Recent studies have reported that frass application to soil might sustain plant biomass production (Beesigamukama et al., 2020a; Dulaurent et al., 2020; Houben et al., 2020; Schmitt and de Vries, 2020; Poveda, 2021). Here, we showed that, irrespective of the application rate, the biomass of ryegrass was significantly higher in the presence of frass than in the control (Figure 1). Using also ryegrass as the study plant, Kebli and Sina (2017) and Menino et al. (2021) found similar results with frass from black soldier fly. The authors concluded that higher ryegrass biomass after frass application was due to an improvement of plant N nutrition. In the present study, the strong relationship between biomass and plant N content corroborates these findings (Figure 2). Nitrogen is usually considered the key nutrient for plant growth and plant yield is closely related to the N supply (Marschner, 1995). Therefore, to be used as an alternative to inorganic fertilizers, organic resources are expected to provide a rate of available N similar to that provided by inorganic fertilization (Hernández et al., 2016). Compared to the Control, N uptake was significantly increased by the application of frass. This high short-term ability to supply N contrasts with other organic amendments which usually supply N more slowly (Delin et al., 2012; Cassity-Duffey et al., 2020). The amount of N released to plants by organic fertilizers depends on their chemical composition, including N content, C:N ratio, and contents of labile C, hemicellulose, cellulose, and lignin (Gutser et al., 2005; Mohanty et al., 2011). As shown in Figure 3, frass is rapidly mineralized after its incorporation into the soil which is related to its high labile C and its low recalcitrant C contents (Table 1), as previously suggested (Houben et al., 2020). The subsequent high N mineralization (Figure 3) can thus explain the rapid N supply by frass to plants. Higher N mineralization in the presence of frass was also reflected by BIOLOG Ecoplate results (Figure 4). In agreement with Chakraborty et al. (2011), the application of mineral N had compromised the ability of the soil microbial communities to catabolize amines as the AWCD values for these compounds was significantly lower in NPK-10. By contrast, the application of frass at 10 t ha<sup>-1</sup> restored the ability of microbes to catabolize amines compared with the control. It is noteworthy to mention that frass also stimulated significantly the substrate utilization of carboxylic acids compared with the NPK control. As observed for other organic amendments (Trabue et al., 2016), this likely reflects the fast decomposition of easily degradable organic compounds added by frass but also of more complex compounds, possibly originated from the soil organic matter (Kolton et al., 2017). According to Lazcano et al. (2013) who



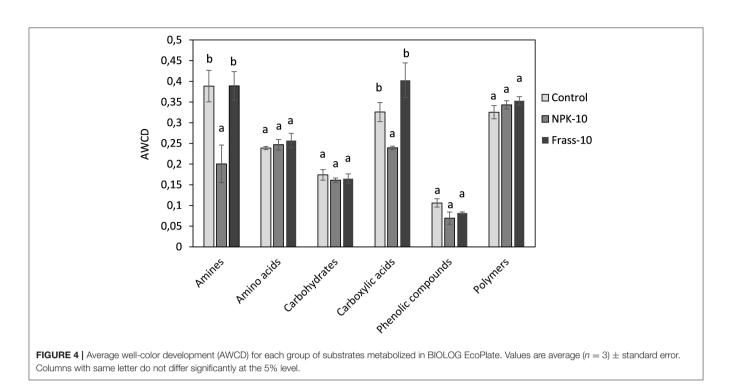
**FIGURE 2** | Relationship between biomass and nitrogen (N) uptake in ryegrass. Marked *r* correlation coefficient is the Pearson's correlation coefficient.



(Frass-5) and 10 t  $ha^{-1}$  (Frass-10).

reported similar findings for rabbit manure and vermicompost, frass might thus increase N availability not only by supplying N but also by promoting N turnover from organic matter through increased microbial activity.

Although frass increased N uptake by plants, its N fertilizer potential was interestingly mediated by its application rate. As shown in **Figure 1**, while N uptake was significantly lower in Frass-5 than in NPK-5, the application of frass at  $10 \text{ th} \text{ a}^{-1}$  (Frass-10) induced a similar N uptake compared to its mineral control



(NPK-10). The lower efficacy of frass to supply N when applied at 5 t  $ha^{-1}$  might be explained by its lower mineralization rate. As shown in Figure 3, the proportion of frass-derived C and N that was mineralized during the incubation experiment was lower when frass was applied at 5 t  $ha^{-1}$  than at 10 t  $ha^{-1}$ . This likely results from the higher input of easily-degradable C in the Frass-10 treatment, which in turn stimulates further microbial activity (Mohanty et al., 2011). Overall, the high mineralization of frass, especially at 10 t ha<sup>-1</sup>, suggests that frass application might be efficient to increase short-term soil N supply to plants. On the other hand, application of frass long before sowing or in excess with respect to the crop needs might cause a substantial loss of N mineralized, as reported for other organic fertilizers (Abbasi et al., 2007). Although in field studies in a longer run should be performed, these preliminary results indicate that frass might be well-suited for a synchronized application of N according to the plant demand. More specifically, application of frass with sowing could be recommended so that the quickly mineralized N may be readily utilized by crops.

Compared with the control, all treatments increased P and K uptake (**Figure 1**). More interestingly, the uptakes of P and K in the presence of frass or mineral fertilizer were similar, indicating that frass was as efficient as mineral fertilizer to supply P and K to plants. Investigating P and K amounts in barley shoot after the application of mealworm frass applied at 10 t ha<sup>-1</sup>, previous studies also concluded that frass was as efficient as mineral fertilizer to plants (Dulaurent et al., 2020; Houben et al., 2020), which was due to the rapid mineralization of frass and the presence of P and K in a readily available form. Our results confirm thus the potential of frass to be used as a fertilizer.

By contrast to N, high application rate had no significant effect on P and K uptakes compared to low application rate. These findings agree with Brod et al. (2012) who investigated the fertilizer potential of various organic by-products using a similar pot experiment with ryegrass. According to the authors, P and K uptake by ryegrass did not respond to high application because nutrient supply was higher than the plant requirements.

## CONCLUSION

Insect production is forecasted to grow in the next few years in response to the increasing need of finding alternative sources of protein and this should generate high quantities of frass. Using a short-term pot experiment, this study indicates that frass has a great potential to be used as a substitute for mineral NPK fertilizer even though its N fertilizer potential is mediated by its rate of application. Indeed, at  $10 \text{ t ha}^{-1}$ , the fast frass mineralization coupled with improvement in microbial activity seemed to be enough to maintain the uptake of N by ryegrass as compared to mineral fertilization. By contrast, at 5 t  $ha^{-1}$ , the lower frass mineralization induces a reduced N uptake compared to its mineral control. Unlike N, frass was as effective as mineral fertilizer to supply P and K to plants, likely due to their presence in a readily available form in frass. Although this work must be considered a preliminary step which needs to be completed by studies on a longer term and with other crops, our findings also indicate that plant biomass responds mainly to the supply of N by frass. As a result, increasing N supply by frass by using higher application rate will result in better yield. On the other hand, due to its fast mineralization, higher application rate of frass might cause substantial loss of N to groundwater. The next challenge will be, therefore, to optimize the use of frass as a sustainable resource for managing NPK nutrition in cropping system.

### DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

## **AUTHOR CONTRIBUTIONS**

DH, GD, and A-MD: Conceptualization. DH and A-MD: methodology. DH and A-MD: investigation. DH: writing—original draft preparation. A-MD and GD: writing—review and writing—review and editing. All authors have read and agreed to the published version of the manuscript.

## REFERENCES

- Abbasi, M. K., Hina, M., Khalique, A., and Khan, S. R. (2007). Mineralization of three organic manures used as nitrogen source in a soil incubated under laboratory conditions. *Commun. Soil Sci. Plant Anal.* 38, 1691–1711. doi: 10.1080/00103620701435464
- AFNOR (2016). FD-U-44-163 Soil Improvers and Growing Media— Characterization of Organic Matter by Potential Mineralization of Carbon and Nitrogen. Paris: AFNOR.
- Beesigamukama, D., Mochoge, B., Korir, N., Musyoka, M. W., Fiaboe, K. K. M., Nakimbugwe, D., et al. (2020b). Nitrogen fertilizer equivalence of black soldier fly frass fertilizer and synchrony of nitrogen mineralization for maize production. *Agronomy* 10, 1395. doi: 10.3390/agronomy10091395
- Beesigamukama, D., Mochoge, B., Korir, N. K., Fiaboe, K. K. M., Nakimbugwe, D., Khamis, F. M., et al. (2020a). Exploring black soldier fly frass as novel fertilizer for improved growth, yield, and nitrogen use efficiency of maize under field conditions. *Front. Plant Sci.* 11:574592. doi: 10.3389/fpls.2020.574592
- Berggren, Å., Jansson, A., and Low, M. (2019). Approaching ecological sustainability in the emerging insects-as-food industry. *Trends Ecol. Evol.* 34, 132–138. doi: 10.1016/j.tree.2018.11.005
- Brod, E., Haraldsen, T. K., and Breland, T. A. (2012). Fertilization effects of organic waste resources and bottom wood ash: results from a pot experiment. Agric. Food Sci. 21, 332–347. doi: 10.23986/afsci.5159
- Cassity-Duffey, K., Cabrera, M., Gaskin, J., Franklin, D., Kissel, D., and Saha, U. (2020). Nitrogen mineralization from organic materials and fertilizers: predicting N release. *Soil Sci. Soc. Am. J.* 84, 522–533. doi: 10.1002/saj2.20037
- Chakraborty, A., Chakrabarti, K., Chakraborty, A., and Ghosh, S. (2011). Effect of long-term fertilizers and manure application on microbial biomass and microbial activity of a tropical agricultural soil. *Biol. Fertil. Soils* 47, 227–233. doi: 10.1007/s00374-010-0509-1
- Chavez, M., and Uchanski, M. (inpress). Insect left-over substrate as plant fertiliser. *J. Insects Food Feed* 1–12. doi: 10.3920/JIFF2020.0063
- Chew, K. W., Chia, S. R., Yen, H.-W., Nomanbhay, S., Ho, Y.-C., and Show, P. L. (2019). Transformation of biomass waste into sustainable organic fertilizers. *Sustainability* 11:2266. doi: 10.3390/su11082266
- Chojnacka, K., Moustakas, K., and Witek-Krowiak, A. (2020). Bio-based fertilizers: a practical approach towards circular economy. *Bioresour. Technol.* 295:122223. doi: 10.1016/j.biortech.2019.122223
- Delin, S., Stenberg, B., Nyberg, A., and Brohede, L. (2012). Potential methods for estimating nitrogen fertilizer value of organic residues. *Soil Use Manage*. 28, 283–291. doi: 10.1111/j.1475-2743.2012.00417.x
- Derrien, C., and Boccuni, A. (2018). "Current status of the insect producing industry in Europe," in Edible Insects in Sustainable Food Systems, eds. A.

## FUNDING

This study received funding from Ÿnsect. The funder was not involved in the study design, collection, analysis, interpretation of data, the writing of this article or the decision to submit it for publication.

## ACKNOWLEDGMENTS

We thank Céline Roisin, Aurore Coutelier and Vincent Hervé for technical assistance.

### SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fsufs. 2021.714596/full#supplementary-material

Halloran, R. Flore, P. Vantomme, and N. Roos (Cham: Springer International Publishing), 471-479. doi: 10.1007/978-3-319-74011-9\_30

- Doan, T. T., Jusselme, D. M., Lata, J.-C., Van Nguyen, B., and Jouquet, P. (2013). The earthworm species *Metaphire posthuma* modulates the effect of organic amendments (compost vs. vermicompost from buffalo manure) on soil microbial properties. A laboratory experiment. *Eur. J. Soil Biol.* 59, 15–21. doi: 10.1016/j.ejsobi.2013.08.005
- Doublet, J., Francou, C., Poitrenaud, M., and Houot, S. (2011). Influence of bulking agents on organic matter evolution during sewage sludge composting; consequences on compost organic matter stability and N availability. *Bioresour. Technol.* 102, 1298–1307. doi: 10.1016/j.biortech.2010.08.065
- Dulaurent, A.-M., Daoulas, G., Faucon, M.-P., and Houben, D. (2020). Earthworms (*Lumbricus terrestris* L.) mediate the fertilizing effect of frass. *Agronomy* 10:783. doi: 10.3390/agronomy10060783
- Faucon, M.-P., Houben, D., Reynoird, J.-P., Mercadal-Dulaurent, A.-M., Armand, R., and Lambers, H. (2015). Advances and perspectives to improve the phosphorus availability in cropping systems for agroecological phosphorus management. *Adv. Agron.* 134, 51–79. doi: 10.1016/bs.agron.2015.06.003
- Flavel, T. C., and Murphy, D. V. (2006). Carbon and nitrogen mineralization rates after application of organic amendments to soil. J. Environ. Qual. 35, 183–193. doi: 10.2134/jeq2005.0022
- Fox, J. (2005). The R commander: a basic-statistics graphical user interface to R. J. Stat. Softw. 14, 1–42. doi: 10.18637/jss.v014.i09
- Gärttling, D., Kirchner, S. M., and Schulz, H. (2020). Assessment of the N- and Pfertilization effect of black soldier fly (Diptera: Stratiomyidae) by-products on maize. *J. Insect Sci.* 20:8. doi: 10.1093/jisesa/ieaa089
- Geisseler, D., Horwath, W. R., Joergensen, R. G., and Ludwig, B. (2010). Pathways of nitrogen utilization by soil microorganisms—a review. *Soil Biol. Biochem.* 42, 2058–2067. doi: 10.1016/j.soilbio.2010.08.021
- Gutser, R., Ebertseder, T., Weber, A., Schraml, M., and Schmidhalter, U. (2005). Short-term and residual availability of nitrogen after long-term application of organic fertilizers on arable land. *J. Plant Nutr. Soil Sci.* 168, 439–446. doi: 10.1002/jpln.200520510
- Hernández, T., Chocano, C., Moreno, J.-L., and García, C. (2016). Use of compost as an alternative to conventional inorganic fertilizers in intensive lettuce (*Lactuca sativa L.*) crops—effects on soil and plant. *Soil Tillage Res.* 160, 14–22. doi: 10.1016/j.still.2016.02.005
- Houben, D., Daoulas, G., Faucon, M.-P., and Dulaurent, A.-M. (2020). Potential use of mealworm frass as a fertilizer: impact on crop growth and soil properties. *Sci. Rep.* 10:4659. doi: 10.1038/s41598-020-61765-x
- Houben, D., Meunier, C., Pereira, B., and Sonnet, P. (2011). Predicting the degree of phosphorus saturation using the ammonium acetate–EDTA soil test. *Soil Use Manage*. 27, 283–293. doi: 10.1111/j.1475-2743.2011.00353.x

- Kebli, H., and Sina, S. (2017). Potentiel agronomique d'un engrais naturel à base de digestats de larves de mouches. *Recherche agronomique suisse* 8, 88–95.
- Kolton, M., Graber, E. R., Tsehansky, L., Elad, Y., and Cytryn, E. (2017). Biochar-stimulated plant performance is strongly linked to microbial diversity and metabolic potential in the rhizosphere. *New Phytol.* 213, 1393–1404. doi: 10.1111/nph.14253
- Lazcano, C., Gómez-Brandón, M., Revilla, P., and Domínguez, J. (2013). Short-term effects of organic and inorganic fertilizers on soil microbial community structure and function. *Biol. Fertil. Soils* 49, 723–733. doi: 10.1007/s00374-012-0761-7
- Marschner, H. (1995). Mineral Nutrition of Higher Plants, 2nd Edn. London: Academic Press. doi: 10.1016/B978-012473542-2/50001-8
- Menino, R., Felizes, F., Castelo-Branco, M. A., Fareleira, P., Moreira, O., Nunes, R., et al. (2021). Agricultural value of black soldier fly larvae frass as organic fertilizer on ryegrass. *Heliyon* 7:e05855. doi: 10.1016/j.heliyon.2020.e05855
- Mohanty, M., Reddy, K. S., Probert, M. E., Dalal, R. C., Rao, A. S., and Menzies, N. W. (2011). Modelling N mineralization from green manure and farmyard manure from a laboratory incubation study. *Ecol. Model.* 222, 719–726. doi: 10.1016/j.ecolmodel.2010.10.027
- Pimentel, D., Hepperly, P., Hanson, J., Douds, D., and Seidel, R. (2005). Environmental, energetic, and economic comparisons of organic and conventional farming systems. *BioScience* 55, 573–582. doi: 10.1641/0006-3568(2005)055<0573:EEAECO>2.0.CO;2
- Poveda, J. (2021). Insect frass in the development of sustainable agriculture. A review. *Agron. Sustain. Dev.* 41:5. doi: 10.1007/s13593-020-00656-x
- Poveda, J., Jiménez-Gómez, A., Saati-Santamaría, Z., Usategui-Martín, R., Rivas, R., and García-Fraile, P. (2019). Mealworm frass as a potential biofertilizer and abiotic stress tolerance-inductor in plants. *Appl. Soil Ecol.* 142, 110–122. doi: 10.1016/j.apsoil.2019.04.016
- Rodella, A. A., and Saboya, L. V. (1999). Calibration for conductimetric determination of carbon dioxide. *Soil Biol. Biochem.* 31, 2059–2060. doi: 10.1016/S0038-0717(99)00046-2

- Sala, M. M., Arrieta, J. M., Boras, J. A., Duarte, C. M., and Vaqué, D. (2010). The impact of ice melting on bacterioplankton in the Arctic Ocean. *Polar Biol.* 33, 1683–1694. doi: 10.1007/s00300-010-0808-x
- Savci, S. (2012). An agricultural pollutant: chemical fertilizer. *IJESD* 3, 73–80. doi: 10.7763/IJESD.2012.V3.191
- Schmitt, E., and de Vries, W. (2020). Potential benefits of using *Hermetia illucens* frass as a soil amendment on food production and for environmental impact reduction. *Curr. Opin. Green Sustain. Chem.* 25:100335. doi: 10.1016/j.cogsc.2020.03.005
- Trabue, S. L., Kerr, B. J., Bearson, B. L., Hur, M., Parkin, T., Wurtele, E. S., et al. (2016). Microbial community and chemical characteristics of swine manure during maturation. *J. Environ. Qual.* 45, 1144–1152. doi: 10.2134/jeq2015.09.0446

**Conflict of Interest:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

**Publisher's Note:** All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Copyright © 2021 Houben, Daoulas and Dulaurent. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.