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Opinion

Insect frass and exuviae to promote plant growth and health

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Beneficial soil microorganisms can contribute to biocontrol of plant pests and diseases, induce systemic resistance (ISR) against attackers, and enhance crop yield. Using organic soil amendments has been suggested to stimulate the abundance and/or activity of beneficial indigenous microbes in the soil. Residual streams from insect farming (frass and exuviae) contain chitin and other compounds that may stimulate beneficial soil microbes that have ISR and biocontrol activity. Additionally, changes in plant phenotype that are induced by beneficial microorganisms may directly influence plant–pollinator interactions, thus affecting plant reproduction. We explore the potential of insect residual streams derived from the production of insects as food and feed to promote plant growth and health, as well as their potential benefits for sustainable agriculture.

Highlights

Microbes with biocontrol or plant growth-promoting properties may reduce the use of synthetic pesticides and fertilisers and thereby support sustainable crop production.

While the persistence and effectiveness of introduced single microbial agents in the field are often insufficient, organic soil amendments can serve as substrates to promote naturally occurring beneficial microbes.

Insect-derived products such as exuviae and frass contain chitin and are rich in nitrogen as well as other nutrients that can improve soil quality and plant growth.

Insect exuviae are colonised by potentially plant-protective or plant growth-promoting bacteria when added to soil.

As the industrial production of insects for food and feed is increasing rapidly, large quantities of insect-derived products become available for soil amendment.

Insect-derived products affect species interactions

Terrestrial plant roots are embedded in soil, a biodiverse substrate rich in microorganisms [1]. These microbes affect plant phenotype and, consequently, plant-mediated interactions with herbivores and other members of the plant-associated community [1–4]. Soils also contain a high diversity of organic and inorganic substances that affect these plant-mediated interactions [5]. These substances may influence the community composition of soil microbiota and understanding the underlying mechanisms may allow to steer this process toward specifically promoting beneficial microbiota [6]. The use of organic soil amendments enhances soil microbial activity and modifies the microbial community composition. It increases soil fertility, consequently improving plant biomass and crop yield [7]. Exploiting these positive effects of organic materials on plant growth and resistance to herbivory can address rising environmental concerns about the use of artificial fertilisers and synthetic pesticides [8,9].

A novel organic soil amendment is emerging from the production of a new source of animal proteins, that is, the production of insects such as yellow mealworm (*Tenebrio molitor*), lesser mealworm (*Alphitobius diaperinus*), house cricket (*Acheta domesticus*), black soldier fly (*Hermetia illucens*), or housefly (*Musca domestica*) for food and feed [9]. This new industry can use organic residual streams as a resource [9,10] and is rapidly developing to an estimated market volume of 730 000 metric tons in 2030, having a compounded annual growth rate of 27.8% [11]. The production of insects for food and feed results in **insect residual streams** (see Glossary) such as insect **exuviae** and **frass**. These residual streams are considered a potential alternative to conventional fertilisers and pesticides [9]. An important component of insect exuviae is chitin, a high-molecular-weight amino-sugar polysaccharide that is also present in fungal cell walls and the exoskeleton of many crustaceans [12]. Chitin-containing soil amendments have been demonstrated to promote plant growth [13]. Likewise, the addition of insect frass to the soil has been shown to supply nitrogen

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and other nutrients to plants that increase their biomass and nutritional content [8]. Both chitin and insect frass amendments impact the soil microbiome composition and this may be an important factor in promoting plant growth and health [2]. However, information on the potential of insect-derived products to improve plant growth and their effects on the plant-associated community is limited. In this paper, we discuss the potential effects of adding insect-derived products to plants in five sections: effects on (i) beneficial soil microbes, (ii) plant growth, (iii) plant resistance, (iv) microbial antagonism against plant pathogens and insects, and (v) plant reproduction (Figure 1).

Beneficial soil microbes

Various plant beneficial soil microbes are commonly applied in agriculture and are often considered to be promising alternatives to agrochemicals [14]. Most notably, different soil bacteria have been found to possess a range of beneficial properties [15,16]. For example, so-called **plant growth-promoting rhizobacteria (PGPR)** can enhance resistance to pests and diseases [3,17]. Several strains have been reported to trigger **ISR** against pathogens and herbivores in their host plants or stimulate the attraction of the natural enemies of herbivores [3,18]. Furthermore, many beneficial bacteria control herbivores and plant pathogens via direct interactions. They can be pathogenic to insects or prevent the growth of other microbes and thus contribute to plant protection independently of the plant itself [15,16].

While the potential of beneficial microbes for sustainable agriculture is great, the outcomes of microbial applications in field-based crop production are often inconsistent [14]. Beneficial microbes are commonly inoculated into soil, the success of which depends on their establishment both in the soil and on plant roots. However, the colonisation by microbial inoculants can be constrained by competitive interactions with indigenous microbes [19]. The lack of capacity of the microbial inoculants to establish in the target environment may also result in a rapid decline in inoculant density [14].

To stimulate soil bacteria that possess biological control or ISR potential, the use of soil amendments that promote the activity and growth of beneficial endemic species has been suggested [19,20]. Amendment-mediated stimulation of indigenous microorganisms has a clear advantage over the employment of microbial inoculants because the enriched soil-borne microbes are well adapted to local soil conditions [19]. During decomposition in soils, mealworm exuviae were shown to stimulate a high diversity of chitinolytic bacteria, with a notable increase in the abundance of Bacilli (Y. Bai, Doctoral dissertation, Leiden University Repository, 2015, http://hdl.handle.net/1887/35971). Different members of this class of bacteria, such as *Bacillus thuringiensis*, *Bacillus cereus*, or *Lysinibacillus sphaericus*, are commercially provided for biological pest control [21]. Besides their ability to form spores, which facilitates production and storage, the success of these Bacilli as crop protection agents is also due to the fact that they can possess virtually all the beneficial properties mentioned previously. As prime examples, root-colonising *B. cereus* and *Bacillus subtilis* both promote plant growth, mediate ISR, and have antagonistic activity against a broad range of plant pathogens and pests [22,23]. In view of the increased abundance of Bacilli associated with applying insect exuviae as soil amendment, the utilisation of insect-derived products to promote plant growth and health seems to bear good prospects.

Plant growth

PGPR can enhance plant growth and productivity. Forming a symbiotic relationship with their host, they benefit from energy-rich root exudates. In turn, they may synthesise plant growth hormones such as cytokinins, auxins, and gibberellins or provide increased access to nutrients such as phosphorus or iron. For instance, they may solubilise minerals from sedimentary rocks or fix atmospheric nitrogen in soils where these nutrients would otherwise not be available to

Glossary

Exuviae: moulted exoskeletons of insects.
Frass: insect faeces.
Herbivore-induced plant volatiles (HIPVs): volatile compounds produced by plants in response to arthropod herbivory.
Induced systemic resistance (ISR): enhanced plant resistance against below- and aboveground pathogens and herbivores that is induced by root-colonising microbes.
Insect residual streams: by-products of farming insects for food and feed.
Microbe-associated molecular pattern (MAMP): molecules conserved in microbes that are recognised by plants.
Plant growth-promoting rhizobacteria (PGPR): plant symbiotic soil bacteria that colonise roots and enhance plant growth.

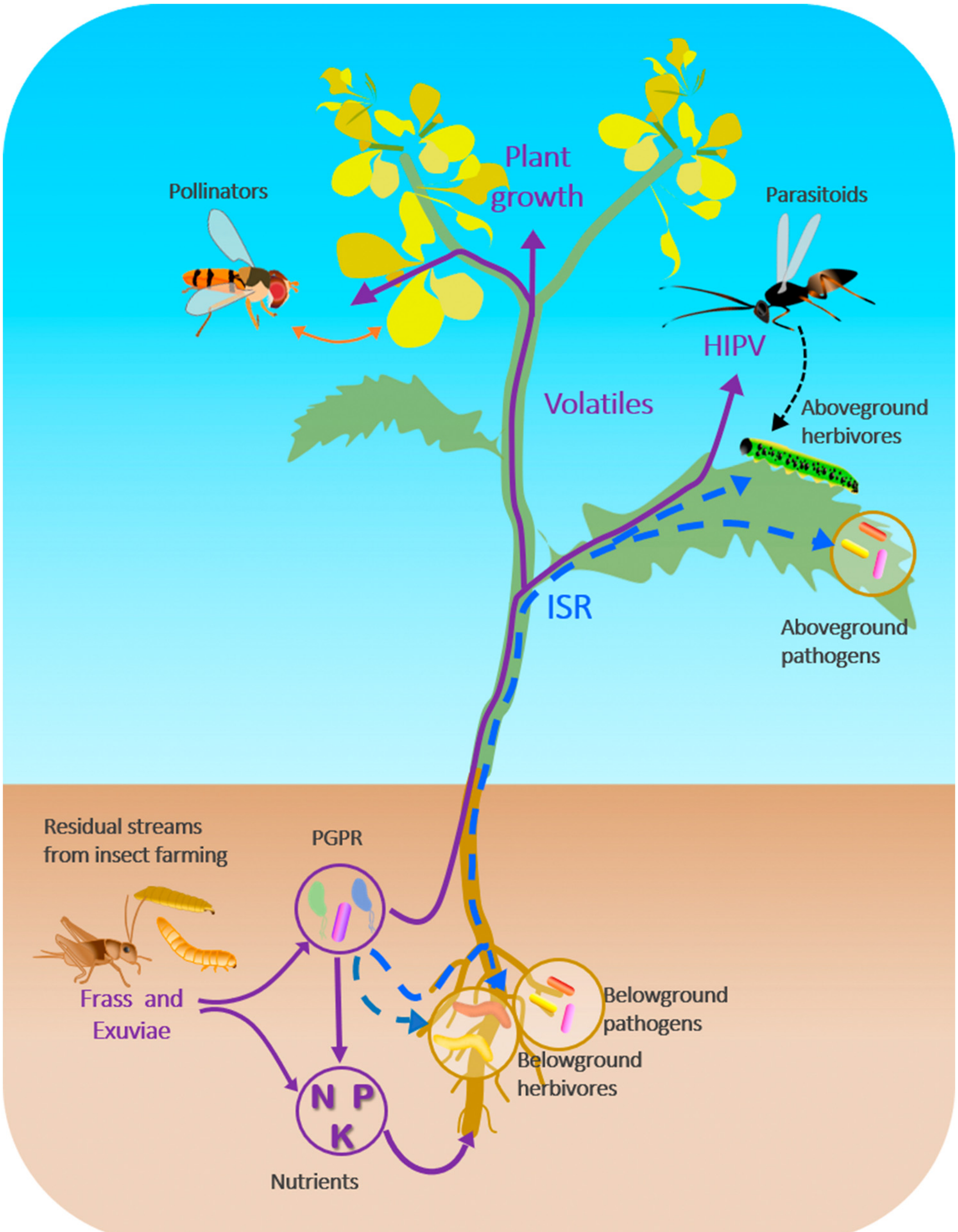

Figure 1. Schematic representation of potential pathways along which insect-derived products may affect plant growth and health. Arrows indicate the effects of organisms from the plant-associated community on the plant and each other. Unbroken arrows indicate positive effects; broken arrows indicate negative effects. Abbreviations: HIPVs, herbivore-induced plant volatiles; ISR, induced systemic resistance; PGPR, plant growth-promoting rhizobacteria.

plants [3]. Although PGPR are especially represented among *Bacillus* and *Pseudomonas* species, plant growth promotion is induced by different members of very diverse bacterial taxa [24]. Examples of other genera known to include growth-promoting strains are *Azospirillum*, *Burkholderia*, *Enterobacter*, *Flavomonas*, *Kluyvera*, *Paenibacillus*, *Rhizobium*, *Serratia*, and *Streptomyces*. Furthermore, associations with PGPR have been described for various plant species. For example, *B. subtilis* is known to promote the growth of cabbage, cotton, maize, pea, peanut, soybean, sweet pepper, and tomato among other crops [4,15].

While it has been noted that enhanced plant growth can also improve food supply for pests, it provides a means to compensate for possible yield losses at the same time [3]. By promoting plant growth, PGPR are thought to facilitate the allocation of resources to plant defence and, thus, additional protection to the host [25]. The high abundance of Bacilli promoted by insect exuviae suggests a stimulation of beneficial *Bacillus* or *Paenibacillus* spp. by these materials (Y. Bai, Doctoral dissertation, Leiden University Repository, 2015, http://hdl.handle.net/1887/35971). Similarly, others have attributed enhanced plant growth after soil amendment with insect frass not only to nutrient supply but to stimulated soil microbial activity as well [26,27].

Most of the emerging research on the fertiliser effects of insect residual streams focuses on frass rather than exuviae [8,26]. However, insect exuviae also contain a considerable amount of nitrogen, mainly in the form of chitin and proteins. Because plants lack the ability to utilise chitin directly, they rely on a cascade of microbial enzymatic activities to breakdown chitin, which releases compounds beneficial for plant growth such as plant-available nitrogen or short-chain chitin oligomers [28,29]. While the prospect of crustacean-derived chitin to enhance plant nutrient availability has been well documented [13,29], insect exuviae have not yet been investigated for this purpose. In addition to chitin, other compounds in insect exuviae such as proteins and lipids may be mediating increased plant performance [12]. The bacterial class Bacilli appeared to be strongly involved in the decomposition of mealworm exuviae in soil but not in the decomposition of purified shrimp chitin, whereas both contain high levels of chitin (Y. Bai, Doctoral dissertation, Leiden University Repository, 2015, http://hdl.handle.net/1887/35971).

Frass is defined as insect excrement, but in the context of the insect farming industry, it refers to a mix of predominantly insect faeces, remnants of shed exoskeletons, and undigested feed [10]. Frass is rich in readily extractable nutrients [30,31]. Frass deposition can result in a short-term pulse of plant-accessible nutrients due to stimulation of local activity of microbial decomposers [30], which can also accelerate the decomposition of recalcitrant organic matter [32]. Fragments of chitin-containing exuviae, which are present in frass as a minor component, may also provide additional benefits of frass application on plant growth and health [31]. In addition to improved plant productivity [26], frass application may also result in induced plant resistance to abiotic stresses [27]. These beneficial effects of frass are mainly ascribed to plant-accessible nutrients, although frass-associated microbes are also likely to play a role [8]. The microbes commonly present in frass are bacterial groups belonging to Gammaproteobacteria and Bacilli as well as fungal groups belonging to Ascomycota [27,33]. Similar microbial communities were also found in insect digestive tracts [34,35]. Frass-associated microbial isolates were shown to exhibit various PGPR traits, such as the capability to solubilise phosphate and produce siderophores [27]. These microbes may play a significant role in changing the natural soil community and improving plant growth, because the removal of microbes by sterilisation of frass resulted in lower plant yield compared with the use of nonsterilised frass [27].

Plant resistance

The addition of insect derivatives to soil may improve plant health not only by increasing plant growth and tolerance against herbivores but also by stimulating plant defence. The sparse research on how insect derivatives affect plant resistance shows that effects differ between plant species, insect species that produced the frass, and plant organ to which the frass was supplied [36]. For example, caterpillar frass suppresses caterpillar-induced defences in maize plants, while it increases the defence against pathogens and aphids [37]. However, the direct opposite is observed in rice plants exposed to caterpillar frass, where caterpillar-induced defences were increased and pathogen defences decreased [36].

Chitin is recognised by plants as a **microbe-associated molecular pattern (MAMP)**, eliciting diverse defence responses in plants including, but not limited to, systemic expression of defence-related genes [38,39], programmed cell death [40], and release of reactive oxygen species [38,41]. Its efficiency in stimulating plant defences against pathogens after application as a soil amendment or as a foliar spray has been shown in numerous systems [29,42]. Therefore, the addition of chitin-rich insect residual streams to agricultural soil is expected to benefit plant resistance.

In addition to inducing plant defences directly via chitin, insect residual streams have the potential to stimulate PGPR. Besides increasing tolerance to herbivory by promoting plant growth, PGPR may also sensitise plants for enhanced defence against a broad range of below- and above-ground attackers. This systemic response to root colonisation by beneficial microorganisms is called ISR [3,17,24]. To activate ISR, plant roots must recognise beneficial microbes through MAMPs, such as cell surface molecules and compounds that are excreted by these microbes [43,44]. Subsequent ISR signalling throughout the root and shoot system is dependent on the plant hormones jasmonic acid (JA) and ethylene (ET) [17,43]. Typically, no direct change in defence-related gene expression is found, but rather a faster and stronger response upon pathogen or insect attack [3,17,24]. The phenomenon enabling more effective responses to biotic and abiotic stresses via physiological changes in the plant is known as priming [3]. A wide variety of root-associated symbionts, including *Pseudomonas*, *Bacillus*, *Trichoderma*, and mycorrhiza species have been shown to prime the plant immune system, without directly activating costly defences [17]. For example, soil inoculation with different PGPR, including *Bacillus* species, has been shown to mediate ISR of plants against various insects, such as root-feeding beetle larvae or shoot-feeding aphids and whiteflies [4,23]. Furthermore, PGPR may affect the recruitment of natural enemies of herbivorous insects, by modifying the blend of **herbivore-induced plant volatiles (HIPVs)** (Figure 1). Soil amendment with the rhizobacterium *Pseudomonas simiae* or with several *Bacillus* species, for example, resulted in an altered plant volatile blend and increased recruitment of parasitoids of two aphid species and a caterpillar by *Arabidopsis thaliana* and *Brassica oleracea*, respectively [18,22]. Selectively stimulating PGPR by adding insect residual streams to the soil may thus induce systemic resistance in the plant, reducing herbivore performance and increasing recruitment of natural enemies.

Microbial antagonism against plant pathogens and insects

In addition to plant-mediated mechanisms, insect residual streams can also exert positive effects on plant survival through the stimulation of native soil microbes with natural biological control activity. Several greenhouse and field studies have shown that the application of chitin-containing amendment coincided with a reduction in disease incidence caused by root-infecting fungi, such as *Verticillium dahliae* [45], *Fusarium oxysporum* [46], and *Rhizoctonia solani* [47]. The key mechanism for this suppression of pathogens is attributed to increased abundance and activity of chitinolytic bacteria and fungi, particularly members of Actinobacteria, Gammaproteobacteria, Bacilli, and Mortierellomycetes [48,49]. The chitinases produced by these microbes, in combination with other cell wall-degrading enzymes and antibiotics, can weaken and disrupt the developing cell wall of fungal pathogens [45,50]. In a similar way, chitinases can affect the development of root herbivores and have been shown to reduce larval feeding and biomass when ingested. The underlying mechanism is thought to be the degradation of chitin in the insect midgut peritrophic matrix [51]. Chitinolytic activity is only one of many mechanisms underlying microbial antagonism. Several native soil microbial species have the inherent capacity to produce a wide array of bioactive metabolites to neutralise detrimental organisms (Box 1).

Box 1. Microbial secondary metabolites with biocontrol activity

By means of toxic or inhibitory allelochemicals and proteins, beneficial microbes can control various soil pests and pathogens directly. For instance, the compounds produced by different *Bacillus* spp. are known to have insecticidal, antibiotic, or nematicidal properties [15,21]. The well-known Cry and Cyt proteins of *B. thuringiensis* are potent insect-specific toxins that are effective against various members of the Coleoptera, Diptera, and Lepidoptera. Similarly, certain cyclic lipopeptides produced by *B. subtilis* have insecticidal activity against fruit flies and mosquitoes. However, *B. subtilis* is mainly known for its antimicrobial activity, which is due to the production of various antibiotic peptides. While lantibiotics, for example, have strong antibacterial activity, different cyclic lipopeptides of *B. subtilis* are involved in the suppression of fungal and oomycete plant pathogens such as *R. solani* or *Pythium aphanidermatum* [15]. Besides *Bacillus* species, soil bacteria that are entomopathogenic or inhibit the growth of plant pathogens can be found in many other genera. Examples are plant growth-promoting *Kluyvera* and *Pseudomonas* species, which can exhibit oral toxicity to insects or suppress plant diseases [16,52].

Unfortunately, natural levels of antagonistic microbial activity are often insufficient to be effective and consistent [53]. However, the selective enrichment of beneficial microbes, for example, by the addition of chitin-rich soil amendments can serve to enhance pest and disease suppression [45]. Chitin-containing organic amendments can be applied as an inoculant carrier of beneficial microbes to improve their efficacy. In some cases, combined application of chitin-containing material with beneficial microbes resulted in synergistic positive effects in terms of plant growth and disease suppression. When used as a seed treatment, formulations of *B. subtilis* in combination with chitin-containing materials showed a steady increase in *B. subtilis* over time and a better control of *Aspergillus niger* (causing crown rot) and *Fusarium udum* (causing wilt) in groundnut and pigeon pea plants, respectively [54]. Similarly, *B. thuringiensis* is known to use chitin as a carbon source and the application of chitin to stimulate its growth has been suggested. Furthermore, co-application of *B. thuringiensis* and chitinase has been shown to increase its insecticidal activity, for example, against *Choristoneura fumiferana* caterpillars [13]. These studies suggest that chitinous amendments can enhance the establishment and antagonistic activity of introduced biocontrol strains and render the soil environment more suitable for the successful establishment of introduced biocontrol agents.

Plant reproduction

Changes in the soil and rhizosphere microbiome induced by insect-derived products may impact plant phenotype such as floral phenology [2]. Marigold plants grown in soil inoculated with *B. subtilis* produced more and heavier flowers, with a significantly increased colour intensity [55]. Similarly, the addition of chitin and its derivative, chitosan, can affect flowering phenology [13,56], speeding up flower production by as much as 15 days [57]. These effects were related to increases in chitinolytic microorganisms [13]. The increase in nutrient availability as a result of microbial activity stimulated by the insect-derived amendments may allow the plant to increase resource investment in flower production [58]. For example, addition of nutrients has been found to increase the number of flowers, flowering duration, and nectar quantities in scarlet trumpet plants [58]. Also, petunia flowers showed increases in corolla size, display size, and consequently, in the number of flower visitors in response to an increase in soil nitrogen [59]. These patterns suggest that plants are likely to alter their phenotype in response to the availability of nutrients influenced by insect-derived products, affecting plant–pollinator interactions and directly influencing plant fitness and yield [60]. While such effects have not been examined for insect-derived materials, their nutrient content and potentially stimulating effect on Bacilli seem promising. Impacts of soil microbes on flowering phenology are also expected to influence plant reproduction (Figure 1). Although the effect of flowering duration on plant reproduction varies between different plant species, pollinator visitation and subsequent seed set increased with flowering duration in plants with unspecialised flowers [61].

To the best of our knowledge, the effects of insect residual streams on flower traits, interactions with pollinators and, consequently, plant reproduction have not been reported in the literature. First evidence has recently been collected that indeed amending soil with insect residual streams can influence plant–pollinator interactions and increase plant reproduction (K.Y. Barragán-Fonseca *et al.*, unpublished).

Concluding remarks and future perspectives

As the insect farming industry is growing rapidly, new companies as well as companies already established in the biocontrol sector have entered the market for insects as animal feed. With the development of regulatory frameworks and the recent authorisation of insects as components of pig and poultry feed in the European Union [62], the use of insects in feed is expected to increase rapidly [63]. At the same time, large amounts of insect residual streams will become available. The application of these residual streams as soil amendments can further contribute to a sustainable and circular agriculture. In the light of legislation that becomes more and more restrictive for the use of synthetic pesticides, these products can provide alternatives to support the development of sustainable pest management [64].

The use of insect-derived products represents a tremendous opportunity to enhance crop productivity within circular agriculture (Figure 2). The stimulation of important functional groups like PGPR and antagonists of pathogens influences the functioning of more complex ecological networks. Beneficial soil bacteria can not only boost plant growth but also cause changes in plant physiology, attracting mutualist insects, such as pollinators and natural enemies, and

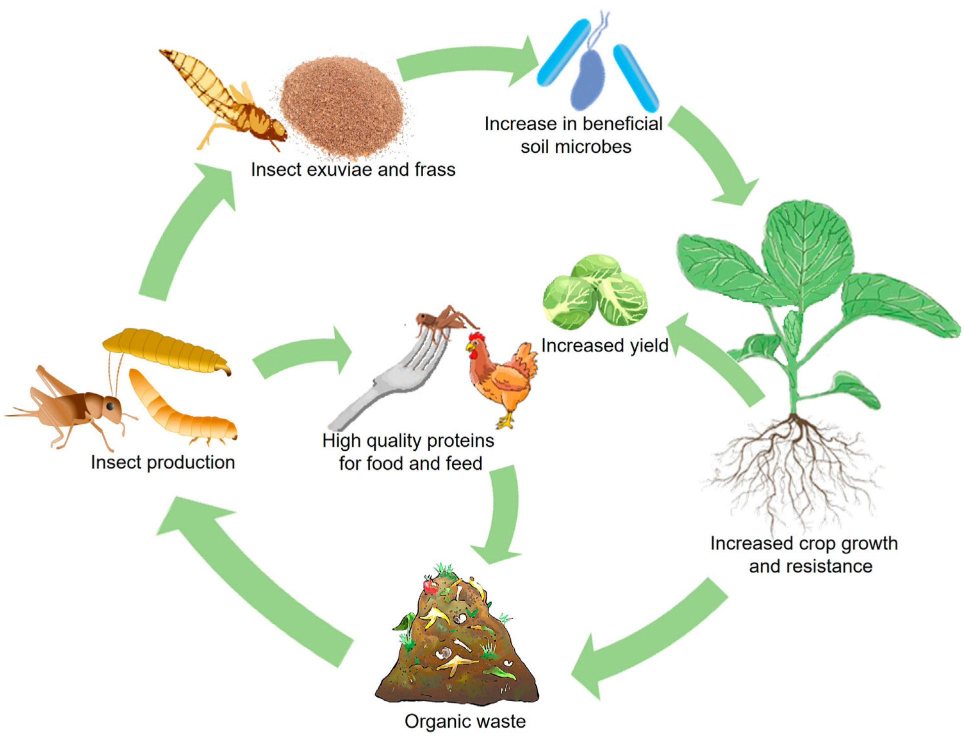

Figure 2. Schematic representation of insect production in a circular food production system. Insects can transform organic waste into high-quality animal protein for food and feed. Here, we discuss the possible use of insect residual streams as soil amendments to stimulate beneficial microbiota and improve soil and crop health.

Outstanding questions

Does amending soil with insect residual streams lead to an increase in PGPR and crop growth? If so, what are the underlying mechanisms?

Does addition of insect residual streams to soil result in ISR in crop plants? If so, is this strong enough to reduce herbivore performance on these plants?

Do plants growing on soils treated with insect residual streams recruit more parasitoids upon herbivory than plants growing in untreated soil?

What is the effect of soil amendment with insect residual streams on crop growth and community development of rhizosphere microbes and aboveground herbivores under field conditions?

Can the addition of insect residual streams alter flowering plant traits exploited by pollinators? If so, how does this affect pollinator behaviour and ultimately plant fitness?

How does soil amendment with insect residual streams affect microbial communities in different soil compartments?

Does soil amendment with insect residual streams reduce the performance of belowground insect herbivores?

How does exposure to insect residual streams in the soil affect the microbes associated with soil-borne pathogens and belowground insect herbivores?

Is soil amendment with insect residual streams compatible with existing agronomic practices?

suppressing insect pests. To better understand these complex dynamics, studies on the effects of insect-derived products on insect–plant–microbe interactions should be expanded. In this way, strategies for applying insect residual streams to control pests while maximising positive effects and avoid negative side-effects on plant traits that are relevant for beneficial insects may be developed. These evaluations should be conducted not only under highly controlled conditions but also in agroecosystems where environmental conditions are variable. Furthermore, the benefits of insect-derived products for agricultural systems compared with conventional management practices need further attention (see Outstanding questions). An improved understanding of key steering factors that are relevant for the successful application of insect residual streams may aid their adoption as a novel approach to develop resilient crop production systems.

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Declaration of interests
No interests are declared.

References
1. Berendsen, R.L. *et al.* (2012) The rhizosphere microbiome and plant health. *Trends Plant Sci.* 17, 478–486
2. Heinen, R. *et al.* (2018) Effects of soil organisms on aboveground plant-insect interactions in the field: patterns, mechanisms and the role of methodology. *Front. Ecol. Evol.* 6, 106
3. Pineda, A. *et al.* (2010) Helping plants to deal with insects: the role of beneficial soil-borne microbes. *Trends Plant Sci.* 15, 507–514
4. Friman, J. *et al.* (2021) Bidirectional plant-mediated interactions between rhizobacteria and shoot-feeding herbivorous insects: a community ecology perspective. *Ecol. Entomol.* 46, 1–10
5. Rowen, E. *et al.* (2019) Managing fertility with animal waste to promote arthropod pest suppression. *Biol. Control* 134, 130–140
6. Bonanomi, G. *et al.* (2018) Organic amendments, beneficial microbes, and soil microbiota: toward a unified framework for disease suppression. *Annu. Rev. Phytopathol.* 56, 1–20
7. Bonanomi, G. *et al.* (2020) Repeated applications of organic amendments promote beneficial microbiota, improve soil fertility and increase crop yield. *Appl. Soil Ecol.* 156, 103714
8. Poveda, J. (2021) Insect frass in the development of sustainable agriculture. A review. *Agron. Sustain. Dev.* 41, 5
9. van Huis, A. (2021) Prospects of insects as food and feed. *Org. Agric.* 11, 301–308
10. Fowles, T.M. and Nansen, C. (2020) Insect-based bioconversion: value from food waste. In *Food Waste Management: Solving the Wicked Problem* (1st edn) (Närvänen, E. *et al.*, eds), pp. 321–346, Springer International Publishing, Cham
11. Anon (2019) Edible insects market by product type (whole insect, insect powder, insect meal, insect type crickets, black soldier fly, mealworms), application (animal feed, protein bar and shakes, bakery, confectionery, beverages) - Global Forecast to 2030. https://www.researchandmarkets.com/reports/4757400/edible-insects-market-by-product-type-whole
12. Roer, R. *et al.* (2015) Exoskeletons across the Pancrustacea: comparative morphology, physiology, biochemistry and genetics. *Integr. Comp. Biol.* 55, 771–791
13. Sharp, R.G. (2013) A review of the applications of chitin and its derivatives in agriculture to modify plant-microbial interactions and improve crop yields. *Agronomy* 3, 757–793
14. Kaminsky, L.M. *et al.* (2019) The inherent conflicts in developing soil microbial inoculants. *Trends Biotechnol.* 37, 140–151
15. Cawoy, H. *et al.* (2011) *Bacillus*-based biological control of plant diseases. In *Pesticides in the Modern World - Pesticides Use and Management* (Stoytcheva, M., ed.), pp. 273–302, IntechOpen
16. Kupferschmied, P. *et al.* (2013) Promise for plant pest control: root-associated pseudomonads with insecticidal activities. *Front. Plant Sci.* 4, 287
17. Pieterse, C.M. *et al.* (2014) Induced systemic resistance by beneficial microbes. *Annu. Rev. Phytopathol.* 52, 347–375
18. Pangesti, N. *et al.* (2015) Rhizobacterial colonization of roots modulates plant volatile emission and enhances the attraction of a parasitoid wasp to host-infested plants. *Oecologia* 178, 1169–1180
19. Mazzola, M. and Freilich, S. (2017) Prospects for biological soil-borne disease control: application of indigenous versus synthetic microbiomes. *Phytopathology* 107, 256–263
20. de Boer, W. (2017) Upscaling of fungal-bacterial interactions: from the lab to the field. *Curr. Opin. Microbiol.* 37, 35–41
21. Francesca, N. *et al.* (2015) Biological activity of *Bacillus* spp. evaluated on eggs and larvae of red palm weevil *Rhynchophorus ferrugineus*. *Ann. Microbiol.* 65, 477–485
22. Gadhave, K.R. *et al.* (2016) Plant growth-promoting *Bacillus* suppress *Brevicoryne brassicae* field infestation and trigger density-dependent and density-independent natural enemy responses. *J. Pest. Sci.* 89, 985–992
23. Gadhave, K.R. and Gange, A.C. (2016) Plant-associated Bacillus spp. alter life-history traits of the specialist insect *Brevicoryne brassicae* L. *Agric. For. Entomol.* 18, 35–42
24. Hol, W.H.G. *et al.* (2013) Getting the ecology into interactions between plants and the plant growth-promoting bacterium *Pseudomonas fluorescens*. *Front. Plant Sci.* 4, 81
25. Pangesti, N. *et al.* (2013) Two-way plant mediated interactions between root-associated microbes and insects: from ecology to mechanisms. *Front. Plant Sci.* 4, 414
26. Houben, D. *et al.* (2020) Potential use of mealworm frass as a fertilizer: impact on crop growth and soil properties. *Sci. Rep.* 10, 4659
27. Poveda, J. *et al.* (2019) Mealworm frass as a potential biofertilizer and abiotic stress tolerance-inductor in plants. *Appl. Soil Ecol.* 142, 110–122
28. de Tender, C. *et al.* (2019) Peat substrate amended with chitin modulates the N-cycle, siderophore and chitinase responses in the lettuce rhizobiome. *Sci. Rep.* 9, 9890
29. Shamshina, J.L. *et al.* (2020) Agricultural uses of chitin polymers. *Environ. Chem. Lett.* 18, 53–60
30. Lovett, G.M. and Ruesink, A.E. (1995) Carbon and nitrogen mineralization from decomposing gypsy moth frass. *Oecologia* 104, 133–138
31. Watson, C. *et al.* (2021) Excellent excrement? Frass impacts on a soil's microbial community, processes and metal bioavailability. *Appl. Soil Ecol.* 168, 104110
32. Zimmer, M. and Topp, W. (2002) The role of coprophagy in nutrient release from feces of phytophagous insects. *Soil Biol. Biochem.* 34, 1093–1099

33. Boiocchi, F. *et al.* (2017) Insect frass in stored cereal products as a potential source of *Lactobacillus sanfranciscensis* for sourdough ecosystem. *J. Appl. Microbiol.* 123, 944–955
34. Mogouong, J. *et al.* (2020) Gut microbiome of the emerald ash borer, *Agrilus planipennis* Fairmaire, and its relationship with insect population density. *FEMS Microbiol. Ecol.* 96, fiaa141
35. Muratore, M. *et al.* (2020) The gut bacterial communities across six grasshopper species from a coastal tallgrass prairie. *PLoS One* 15, e0228406
36. Ray, S. *et al.* (2016) Lessons from the far end: caterpillar frass-induced defenses in maize, rice, cabbage, and tomato. *J. Chem. Ecol.* 42, 1130–1141
37. Ray, S. *et al.* (2020) Asymmetry in herbivore effector responses: caterpillar frass effectors reduce performance of a subsequent herbivore. *J. Chem. Ecol.* 46, 76–83
38. Parada, R.Y. *et al.* (2018) Optimization of nanofibrillation degree of chitin for induction of plant disease resistance: elicitor activity and systemic resistance induced by chitin nanofiber in cabbage and strawberry. *Int. J. Biol. Macromol.* 118, 2185–2192
39. Ramonell, K. *et al.* (2005) Loss-of-function mutations in chitin responsive genes show increased susceptibility to the powdery mildew pathogen *Erysiphe cichoracearum*. *Plant Physiol.* 138, 1027–1036
40. Newman, M.A. *et al.* (2013) MAMP (microbe-associated molecular pattern) triggered immunity in plants. *Front. Plant Sci.* 4, 139
41. Kishimoto, K. *et al.* (2010) Perception of the chitin oligosaccharides contributes to disease resistance to blast fungus *Magnaporthe oryzae* in rice. *Plant J.* 64, 343–354
42. Ramírez, M. *et al.* (2010) Chitin and its derivatives as biopolymers with potential agricultural applications. *Biotecnol. Apl.* 27, 270–276
43. van Wees, S.C. *et al.* (2008) Plant immune responses triggered by beneficial microbes. *Curr. Opin. Plant Biol.* 11, 443–448
44. Segarra, G. *et al.* (2009) MYB72, a node of convergence in induced systemic resistance triggered by a fungal and a bacterial beneficial microbe. *Plant Biol.* 11, 90–96
45. Cretoiu, M.S. *et al.* (2013) Chitin amendment increases soil suppressiveness toward plant pathogens and modulates the actinobacterial and oxalobacteraceal communities in an experimental agricultural field. *Appl. Environ. Microbiol.* 79, 5291–5301
46. Randall, T.E. *et al.* (2020) Changes of *Fusarium oxysporum* f.sp. *lactucae* levels and soil microbial community during soil biosolarization using chitin as soil amendment. *PLoS One* 15, e0232662
47. Andreo-Jimenez, B. *et al.* (2021) Chitin- and keratin-rich soil amendments suppress *Rhizoctonia solani* disease via changes to the soil microbial community. *Appl. Environ. Microbiol.* 87, 11
48. Debode, J. *et al.* (2016) Chitin mixed in potting soil alters lettuce growth, the survival of zoonotic bacteria on the leaves and associated rhizosphere microbiology. *Front. Microbiol.* 7, 565
49. Wieczorek, A.S. *et al.* (2019) Ecological functions of agricultural soil bacteria and microeukaryotes in chitin degradation: a case study. *Front. Microbiol.* 10, 1293
50. de Boer, W. *et al.* (2001) Growth of chitinolytic dune soil beta-subclass *Proteobacteria* in response to invading fungal hyphae. *Appl. Environ. Microbiol.* 67, 3358–3362
51. Veliz, E.A. *et al.* (2017) Chitinase-producing bacteria and their role in biocontrol. *AIMS Microbiol.* 3, 689–705
52. Laurentis, V.L. *et al.* (2014) *Kluyvera ascorbata*: a plant growth-promoting bacteria (PGPB) to manage *Plutella xylostella* (L., 1758) (Lepidoptera: Plutellidae). *Int. J. Res. Agric. Food Sci.* 1, 340–343
53. Garbeva, P. *et al.* (2004) Microbial diversity in soil: selection of microbial populations by plant and soil type and implications for disease suppressiveness. *Annu. Rev. Phytopathol.* 42, 243–270
54. Manjula, K. and Podile, A.R. (2001) Chitin-supplemented formulations improve biocontrol and plant growth promoting efficiency of *Bacillus subtilis* AF 1. *Can. J. Microbiol.* 47, 618–625
55. Flores, A.C. *et al.* (2007) Yield and quality enhancement of marigold flowers by inoculation with *Bacillus subtilis* and *Glomus fasciculatum*. *J. Agric. Sustain.* 31, 21–31
56. Salachna, P. *et al.* (2015) The effect of chitosan coating of bulbs on the growth and flowering of *Ornithogalum saundersiae*. *Acta Hortic.* 1104, 115–118
57. Ohta, K. *et al.* (1999) Chitosan treatment affects plant growth and flower quality in *Eustoma grandiflorum*. *Hort. Sci.* 34, 233–234
58. Burkle, L.A. and Irwin, R.E. (2009) The effects of nutrient addition on floral characters and pollination in two subalpine plants, *Ipomopsis aggregata* and *Linum lewisii*. *Plant Ecol.* 203, 83–98
59. Rebolleda-Gómez, M. *et al.* (2019) Gazing into the anthosphere: considering how microbes influence floral evolution. *New Phytol.* 224, 1012–1020
60. Cardoza, Y. *et al.* (2012) Effects of soil quality enhancement on pollinator-plant interactions. *Psyche* 2012, 581458
61. Lázaro, A. *et al.* (2013) How do pollinator visitation rate and seed set relate to species' floral traits and community context? *Oecologia* 173, 881–893
62. European Union (2021) Commission Regulation (EU) 2021/1372 of 17 August 2021. *Off. J. Eur. Union* 64, L295/1-17. https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=OJ:L:2021:295:FULL&from=EN
63. International Platform of Insects for Food and Feed (2021) An overview of the European market of insects as feed. https://ipiff.org/wp-content/uploads/2021/04/Apr-27-2021-IPIFF_The-European-market-of-insects-as-feed.pdf
64. Torgerson, K.L. *et al.* (2021) Towards circular agriculture – exploring insect waste streams as a crop and soil health promoter. *J. Insects Food Feed* 7, 357–368