

Review

# Insect Frass as a Sustainable Medium for Microalgae Growth: Bridging Waste Valorization and Eco-Physiological Innovation

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**Abstract:** The intensive use of chemical fertilizers has boosted agricultural yields but caused severe environmental concerns, including soil degradation, water pollution, and greenhouse gas emissions. Sustainable alternatives are therefore urgently needed. Insect frass, a nutrient-rich by-product of insect farming, and microalgae, with their ability to produce phytohormones and improve soil quality, have both been proposed as promising biofertilizers. While their individual applications are well documented, little attention has been given to their combined use. This review provides an updated synthesis of current knowledge on insect frass composition, the agronomic value of microalgae, and the first experimental evidence on the use of frass as a nutrient source for microalgal cultivation. Benefits and challenges are discussed, including nutrient variability, microbial safety, heavy metal accumulation, and production costs. The integrated perspective offered here highlights the potential of frass–microalgae systems to support circular bioeconomy models and reduce reliance on chemical fertilizers.

**Keywords:** circular bioeconomy; sustainable agriculture; waste valorization; biofertilizer; microalgae; frass

## 1. Introduction

Environmental sustainability is one of the main global challenges, as it involves striking a balance between economic growth and environmental protection [1]. This correlation is currently the subject of debate in the scientific community. A study [2] highlighted that economic growth is a fundamental requirement for development and environmental sustainability but often has negative effects on the environment. Modern agricultural practices are a clear example of this condition, particularly the widespread use of synthetic chemical fertilizers. These fertilizers are designed to optimize agricultural yields and ensure a stable food supply. However, at the same time, their excessive and prolonged use has led to various environmental and health concerns. Over-fertilization contributes to soil degradation, greenhouse gas emissions, and the bioaccumulation of toxic elements in food, which can be harmful to human health, with potential risks to food security [3].

To limit this disproportionate use and minimizing the environmental consequences of biodegradable waste, an economically sustainable approach to their management is needed. To this end, several studies investigated the application of insect frass as an organic fertilizer applied to soil. The bioconversion of organic waste represents an

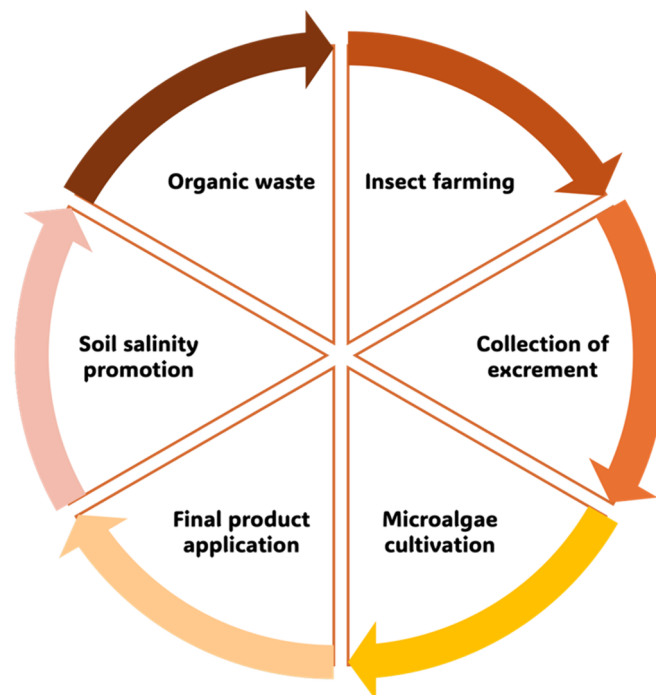


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environmentally sustainable and circular strategy that transforms biodegradable waste into natural fertilizers, contributing to the reduction of emissions and the achievement of a zero-waste economy [4]. The use of frass as an organic fertilizer is advantageous because it contains similar amounts of macro- and micronutrients compared to chemical fertilizers [5,6]. Moreover, the chemical and microbial composition of insect frass appears to play a significant role in the optimal functioning of the soil system, also offering solutions against biotic and abiotic stress [7–9]. Due to their high content of essential nutrients, their use as natural fertilizers for microalgae growth has been observed [10]. Their use provides microalgae with an optimal supply of carbon (C), nitrogen (N), and phosphorus (P), while also increasing their protein content.

From a broader perspective, these results highlight a sustainable cooperation between insect frass and microalgae. In this integrated system, insect frass primarily provides organic and inorganic nutrients (C, N, P), while microalgae biologically transform these inputs through assimilation and metabolic conversion, generating value-added biomass enriched in phytohormones and bioactive compounds suitable for biofertilizer production [11]. Several studies have identified this cooperation as a promising tool for reducing the excessive use of chemical fertilizers, improving soil health, and promoting environmental sustainability [6,12]. The overall process linking organic waste valorization, insect frass production, microalgae cultivation, and soil application is summarized in Figure 1.



**Figure 1.** Schematic overview of the insect frass–microalgae–biofertilizer ecological network. Organic waste is converted through insect rearing into frass, which is subsequently used as a nutrient source for microalgae cultivation. The resulting microalgal biomass is applied to soil as a biofertilizer or biostimulant, contributing to nutrient recycling and soil health improvement within a circular bioeconomy framework.

In recent years, there has been growing interest in their correlation, as microalgae and insect excreta mutually contribute to resource recovery and the circular bioeconomy. For this reason, this review aims to collect and analyze the most recent evidence on their use as alternative fertilizers, highlighting their benefits, mechanisms of action, and future prospects, providing a comprehensive overview and guidance on more effective strategies for sustainable agriculture.

The literature considered in this review was collected using major scientific databases, including Google Scholar, Web of Science, and PubMed. The search was performed using combinations of keywords such as “insect frass”, “frass”, “microalgae biofertilizer”, “circular bioeconomy”, “waste valorization”, and “sustainable agriculture”.

The time window primarily covered studies published between 2014 and 2025 and also other relevant studies already assessed from different periods of time. Peer-reviewed articles written in English were considered, including original research articles, reviews, and relevant regulatory or policy-oriented studies. Publications were selected based on their relevance to the topics of insect frass production and characterization, microalgae cultivation and fertilization potential, and their integration within circular bioeconomy frameworks. Studies not directly related to agricultural or environmental applications were excluded.

## 2. Insects and Their Frass as a Source of Natural Fertilisers

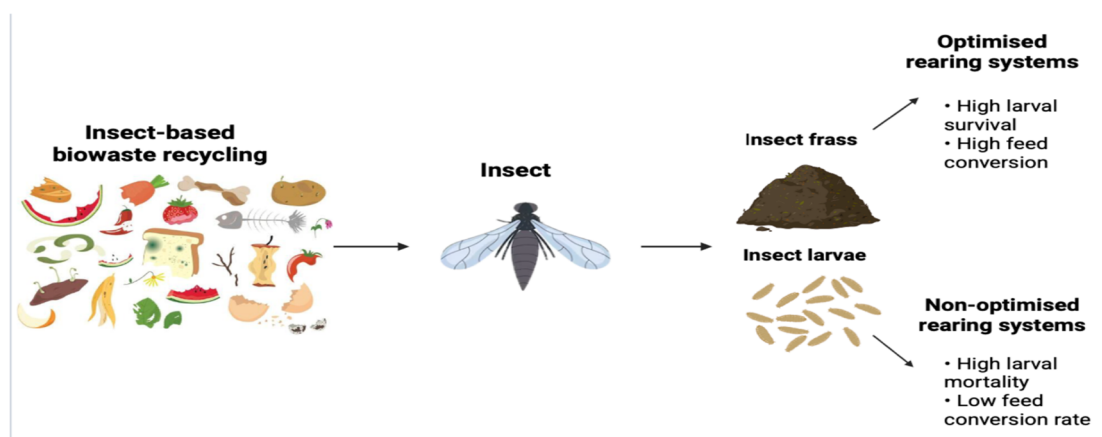
### 2.1. Towards Zero-Waste with Entomo-Composting

Insects represent an optimal solution within the bioeconomy for resource utilization and waste minimization [13,14]. This is due to their complex digestive system, which enable them to process large amounts of waste material, producing nutrient-rich excreta and beneficial microbes. These could be used as organic fertilizers. This approach has been explored because inappropriately treated biodegradable waste is considered a global threat from an environmental, social, and economic perspective [4,7]. Therefore, an economically sustainable approach to organic waste management is needed. With this in mind, researchers conducted studies highlighting insect-based bioconversion as an environmentally friendly approach to organic waste management [15]. Therefore, their use as organic fertilizer supports the transition from entomocomposting, which involves the decomposition of organic waste by insect larvae to produce larval biomass and excreta, to a net-zero waste emission process [16,17].

### 2.2. Main Insects Bred for Frass Production

To date, the insect species primarily studied and farmed are those authorized in the EU for food and feed production. Among these are *Hermetia illucens* (black soldier fly larvae), *Tenebrio molitor* (yellow mealworm), *Acheta domesticus* (house cricket), and *Alphitobius diaperinus* (lesser mealworm) [17]. Furthermore, the microbial composition of insect frass was also studied on other species, including *Oryctes rhinoceros*, *Pachnoda sinuata*, *Schistocerca gregaria*, *Bombix mori*, and *Gryllus bimaculatus* [7,8].

The yield of the frass production process appears to be influenced by several factors, such as substrate pretreatment (pre-heating, microbial inoculation, nutrient balancing, pH and C/N ratio, moisture adjustment), substrate characteristics (nutritional quality, porosity, particle size, electrical conductivity, chemical contaminants), and rearing conditions (temperature, relative humidity, feeding rate, larval density) (Figure 2). Substrate pretreatment (e.g., shredding, fermentation, moisture adjustment) influences nutrient bioavailability and microbial load, directly affecting frass quality. Indeed, frass composition is highly dependent on the nature and processing of the feed substrate, with significant variation in nutrient content and microbiological characteristics observed across different feed types of insect rearing systems [18,19]. Moreover, the microbiological safety of frass is strongly impacted by post-harvest treatments and storage conditions, with untreated frass often exhibiting higher microbial counts that may not comply with regulatory criteria [20]. Substrate characteristics, such as protein and fiber content, determine nitrogen and mineral composition of the resulting frass, while rearing conditions (temperature, density, and duration) modulate both frass yield and hygienic safety [21]. Optimized systems generally produce nutrient-rich frass with lower pathogen risk, whereas non-optimized conditions result in heterogeneous and nutrient-poor outputs. Depending on the conditions, two different situations can be distinguished, i.e., a high yield is given by optimized systems, which provide nutrient-rich frass, and a lower yield is given by non-optimized systems, which provide nutrient-poor frass [16].



**Figure 2.** Schematic representation of insect-based biowaste recycling and frass production. Organic waste is converted through insect rearing into larval biomass and frass. Rearing conditions strongly influence system performance, with optimized systems characterized by high larval survival and feed conversion efficiency, and non-optimized systems associated with increased larval mortality and reduced feed conversion rates. These differences ultimately affect frass yield and quality. Created with BioRender, Toronto, ON, Canada, [www.biorender.com](http://www.biorender.com) (accessed on 24 October 2025).

### 2.3. Microbiological and Chemical Analysis of Insect Frass

Depending on the growth substrate that influences insect frass production, there appears to be a significant bacterial and fungal community, which varies from species to species. Therefore, it is important to focus on the microbiological composition of excrement to evaluate whether insect-derived fertilizers contain any pathogens [7].

According to a recent study [7], different concentrations of microorganisms were observed in eight distinct species. In particular, total and fecal coliforms are present in greater concentrations, especially in *Schistocerca gregaria* and *Gryllus bimaculatus* samples. In contrast, *Hermetia illucens* and *Bombyx mori* show very low values. The presence of *E. coli* is identified in significantly high concentrations within *S. gregaria* and *G. bimaculatus*; however, less significant concentrations have been detected in the other species. Enterococci show higher concentrations in *S. intermedia* and *B. mori* feces and negligible amounts in the remaining species. In the case of *B. cereus*, it appears to be present in high concentrations in almost all species, especially in *S. gregaria* and *S. intermedia*. *C. perfringens* is found in various species in varying concentrations, especially in *S. gregaria* and *H. illucens*. Concerning *Salmonella* spp., it is not detected in any sample. Based on what has emerged, it is interesting to note that *S. gregaria* and *G. bimaculatus* produce excrement with a high bacterial content, while other species, such as *H. illucens* and *B. mori*, seem to produce a microbiologically “cleaner” product. This suggests that the choice of species, in relation to the type of nutrients and farming conditions, can significantly influence the hygienic quality of frass and, consequently, its potential application as an agricultural fertilizer [7].

In addition to the microbiological aspect, the chemical aspect of frass has also been studied [19]. Insect excrement is particularly rich in macronutrients such as nitrogen, phosphorus, and potassium; secondary nutrients (calcium, magnesium, and sulfur); and micronutrients including manganese, copper, iron, zinc, boron, and sodium [22]. In a study recently conducted [23], *T. molitor* larvae fed only wheat bran had a nitrogen content of 5%, carbon of 39%, phosphorus of 2%, and potassium of 1.7%. The chemical composition varies depending on the diet. According to another study [24], a different diet was implemented, based on wheat bran and chayote (*Sechium edule*), such that the chemical composition varied, presenting a nitrogen content of 2.5% and a phosphorus content of 1.4%.

### 2.4. Production of Fertilizers Derived from Frass

The production of insect frass-based fertilizers occurs according to specific steps:

- (1) Breeding and bioconversion: selection of the substrate on which to rear the insects, which will determine the composition of the frass [25];
- (2) Frass harvesting: recovery of the by-product of bioconversion through sieving, consisting of larval excrement, larval exuviae, and residues of unconsumed raw material [18];
- (3) Hygienic treatment: performed to minimize any potential risks. It has been observed that a heat treatment at 70 °C for 60 min is appropriate for meeting the microbiological criteria for the application of insect excreta as a biofertilizer or soil improver [26];
- (4) Secondary maturation/composting: stabilization of the insect frass. The frass is composted for 5 weeks using the heap method to obtain a mature and stable product [27];
- (5) Frass characterization: chemical and microbiological analyses of frass [7,19];
- (6) Fertilizer application and evaluation: plant trials, effects on soil health and microbiota [28].

### 2.5. Effects, Risks, and Limits

Insect frass fertilizers show positive effects on plant growth. Their production appears to be achieved by feeding insect larvae with different nutrients for different periods of time (Table 1) [7].

Overall, the nutrient concentrations in insect frass make it a viable and sustainable alternative to traditional organic fertilizers. In a recent research [29], macronutrient concentrations were evaluated in different species, as were micronutrient concentrations. These data confirm the high suitability of insect excreta-based fertilizers as a sustainable source of plant nutrients and as a quality fertilizer input for sustainable soil health management [29]. From these data, it can be highlighted that fertilizers composed of *T. molitor* and *B. mori* excreta have a poor nutritional profile, resulting in excreta with lower concentrations of micronutrients and macronutrients, leading to the production of fertilizers suitable for frequent use or on crops sensitive to salt or metals. Fertilizers produced from *O. rhinoceros* and *P. sinuata* excreta are particularly rich in micronutrients, such as iron, manganese, and aluminum, but low levels of macronutrients, such as nitrogen and potassium. As a result, they have an unbalanced mineral profile, producing fertilizers that are used as correctives for soils deficient in micronutrients, but only in minimal quantities. Fertilizers composed of *H. illucens* excreta, at the same time, have a complete and balanced nutritional profile, with high nitrogen, potassium, and sulfur contents, along with relatively high levels of iron and



sodium. Ultimately, *H. illucens* represents the best species capable of producing fertilizers with an optimal balance of macronutrients and micronutrients, providing plants with essential nutrients without the risk of significant toxicity [29].

**Table 1.** Insect species and substrates used in frass production studies.

Insect Species	Diet/Nutrient Substrate	Duration Conditions	Source
<i>Hermetia illucens</i>	Irish potato scraps and beer run-outs	2 weeks	[24]
<i>Oryctes rhinoceros</i>	Fresh bovine dung	4 weeks	[7]
<i>Pachnoda sinuate</i>	Fresh bovine dung	4 weeks	[7]
<i>Schistocerca gregaria forsskal</i>	Wheat and barley seedlings and wheat bran	/	[6]
<i>Bombix mori</i>	Mulberry leaves ( <i>Morus</i> spp.)	6 weeks	[8]
<i>Gryllus bimaculatus</i>	Wheat bran, soybeans, sweet potato vines, and weeds	/	[17]

Several studies analyze the use of insect excrement as fertilizer for plants, and the beneficial/toxic effects they produce are influenced by the composition, the amount used, and the type of plant. There are many possible beneficial effects, and this is related to the optimal dosage of excreta; conversely, if excessive dosages of excreta are applied, suppressive and deleterious effects are obtained [18]. These beneficial effects are diverse, i.e., increased plant growth, improved nutrient absorption, improved soil health, improved resistance to environmental stress, reduced use of chemical fertilizers, promotion of more sustainable practices, and a circular economy [30–35].

At the same time, the use of insect frass fertilizers presents limitations and critical issues. For instance, in the context of heavy metals, on muddy substrates, the excrement of *H. illucens* larvae can contain significant amounts of chromium and nickel, exceeding the limits set by the EU for organic fertilizers [36]. Furthermore, in a recent study [25] is indicated that frass could contain pathogenic bacteria if not subjected to optimal sanitation treatments. Another research [37] showed a high nitrogen content in *T. molitor* frass, which could impair plant germination.

In addition to all the limitations considered, the high cost of production hinders market expansion [38].

Ultimately, insect excrement-based fertilizers offer effective potential for improving plant growth, soil health, and agricultural sustainability. Despite this, their effectiveness depends on the quality of the source substrate and the application methods, as compositional variability, the risk of contaminants, and possible phytotoxic effects if used excessively still limit their fully standardized, large-scale application [28]. On the other hand, several studies investigated the use of insect excreta-based fertilizers as a potential source of nutrients for microalgae cultivation [6,12]. This process enables the production of microalgae with a high protein content, and this approach has been analyzed as the most sustainable and economically advantageous solution, fitting into the context of the circular bioeconomy [6]. While insect frass can be directly applied as an organic fertilizer, its nutrient content can be further valorized through biological conversion processes. In particular, microalgae cultivation represents a second upgrading step, enabling the transformation of frass-derived nutrients into high-value biomass with enhanced biostimulant and agronomic properties. This perspective provides the conceptual bridge between frass-based fertilization and microalgae-based biofertilizers discussed in the following section.

### 3. The Role of Microalgae-Based Fertilizers

Through growing scientific research aimed at discovering the multiple benefits of microalgae-based fertilisers, it has been observed that their use is efficient for agricultural productivity, and thus for businesses, as well as for human health. For instance, in the field of agriculture, it has been found that the implementation of biofertilizers drastically reduces the use of chemical fertilisers, which are useful for increasing crop yields but at the same time cause soil depletion. Biofertilizers, on the other hand, help prevent soil degradation and contribute to reducing CO<sub>2</sub> emissions into the atmosphere. Also, they have the advantage of being produced from living organisms and can even be modified, thus optimising their characteristics in relation to different crops [11].

### 3.1. Application of Microalgae as Biofertilizers

Microalgae are an important resource in both freshwater and marine ecosystems as photosynthetically active organisms. Thanks to the high biodiversity that characterises these microorganisms and their rapid growth rate, recent scientific studies have shown that they play a key role in numerous applications [39]. In fact, microalgae are widely used for their biological characteristics and their ability to produce bioactive metabolites [40] in biotechnology, nutraceuticals, and environmental well-being, as they can also be used for wastewater treatment. In particular, they are useful in crucial phases such as those aimed at removing hard-to-degrade pollutants and excess nutrients such as nitrogen and phosphorus, heavy metals and pharmaceutical residues. Therefore, they represent an eco-friendly alternative to traditional chemical methods [41].

#### 3.1.1. Application and Economic Value of the Main Microalgae Species

*Chlorella* sp. and *Arthrospira* sp. are among the major species of microalgae, accounting for approximately 90% of global biomass production. They are also widely used as a sustainable resource in various sectors, ranking among the most emerging and relevant bioeconomic systems [39]. Starting from organic carbon molecules, these microorganisms have the ability to absorb and oxidise these molecules to produce energy [42]. Moreover, they are among the most valued microalgae species for their nutritional and therapeutic qualities [43]. From a nutritional point of view, among the 14 most relevant species of *Chlorella* sp. are *Chlorella vulgaris* and *Chlorella sorokiniana* [44]. Among the most exploited species of *Arthrospira* sp. for commercial cultivation are: *Arthrospira maxima*, *Arthrospira fusiformis*, and *Arthrospira platensis*. It is important, however, to clarify that *Spirulina* sp. and *Arthrospira* sp. are considered to be very similar but taxonomically distinct species of cyanobacteria [45], but the commercial term “spirulina” refers only to *Arthrospira* sp. [46].

Therefore, from a commercial point of view, a significant expansion of the microalgae market is expected, particularly for the nutraceutical sector [39]. The *Chlorella* sp. market is expected to grow at a compound annual growth rate (CAGR) of 6.3% over the period 2021–2028, reaching an estimated value of 412.3 million by 2028 [39]. The market for the *Arthrospira* sp. species is significantly larger and is expected to grow to 1.1 billion by 2030, with a CAGR of 9.4% over the period 2023–2030 [39]. Furthermore, in 2021, Europe retained the largest share of the global *Chlorella* sp. market [39].

#### 3.1.2. Value of Microalgae in Plant Growth: Production of Phytohormones and Bioactive Metabolites

It has been observed that the biomass produced by microalgae can be converted into nutrients useful for sustaining their own development and that of other plant species [47,48]. In this context, phytohormones intervene as chemical messengers to regulate key physiological and biochemical processes in higher plants. For example, studies have found that, even at extremely low concentrations, phytohormones are able to modulate metabolic functions such as nutrient absorption, photosynthesis, and nucleic acid synthesis [49,50]. In this regard, phytohormone-rich microalgae extracts have proven to be particularly effective as a basis for the development of biostimulants for use in agriculture. Phytohormones are commonly divided into five main classes: auxins, abscisic acid, cytokinins, gibberellins and ethylene, together with their respective precursors and analogues [51]. Auxins are particularly effective in maturing the metabolism of microalgae, promoting their growth, increasing biomass, and synthesising bioactive molecules, even at very low concentrations [52]. Depending on the plant species, growth stage, and culture conditions, their chemical composition and concentration can vary considerably. Within microalgae, the main chemical compounds associated with auxins are indole-3-acetic acid (IAA) and indole-3-butanoic acid (IBA) [53]. Depending on the level of IAA and its derivatives, the stimulating or inhibitory effects on plant metabolism may vary [54]. IAA, IBA, indole-3-propionic acid (IPK), and indole-3-acetamide (IAM) have been identified in 46 species of microalgae belonging to the Cyanophyta and Chlorophyta families [53].

Another relevant class of phytohormones is that of cytokinins (CK). It has been demonstrated that these substances are essential for inducing cell division, stimulating cell growth processes, and increasing photosynthetic activity in plant organisms. Furthermore, it has been verified that under stressful conditions, CKs have a protective effect on physiological activities and, in particular, on photosynthesis [55]. As regards the biosynthesis of CKs in microalgae, this is dependent on nitrogen, as the latter influences the production of RNA, specific proteins, and polypeptides [56]. In addition, the content and structure of CKs in microalgae are strongly influenced by lighting regimes and the availability of carbon sources [55,57].

Brassinosteroids, jasmonic acid, polyamines, salicylates, and signalling peptides are other types of hormones and growth regulators of fundamental importance [58]. In fact, brassinosteroids play a role in cell division, elongation, and differentiation of the vascular system; jasmonic acid improves resistance when microalgae are

subjected to unfavourable environmental conditions [59]; while salicylates may be involved in the activation of defensive responses [60].

### 3.2. Mechanism of Action of Microalgae on Soil and Plants

Nowadays, the efficiency of the global agricultural sector depends heavily on the use of synthetic fertilisers and pesticides, with negative consequences for the environment and health [61–66]. According to several studies, the use of microalgae can help change this scenario in several ways [67–75]. One reason for using microalgae is that they represent a rich source of organic carbon when used in the soil [67]. This advantage is particularly important in agriculture, as the consumption of organic carbon accounts for a significant part of the deterioration in soil quality and fertility. In fact, microalgae can absorb organic carbon into their biomass through photosynthesis, and some strains release exopolysaccharides (EPS), molecules that act both as a source and as sequesters of carbon itself [67,76]. In fact, EPS can form a protective layer on the surface of the soil, which promotes the retention of organic carbon, nitrogen, and phosphorus and also acts as a water reserve in case of drought. In addition, microalgae and cyanobacteria have proven to be extremely effective in recovering soils deteriorated due to high salinity [77] and add organic matter and nitrogen to the soil, promoting particle aggregation, greater permeability, and aeration [78,79]. In addition, microalgae promote the growth of other plants through three main classes of substances produced from them: biofertilizers, biostimulants, and biopesticides [69,80].

The use of these compounds in agriculture offers multiple benefits, contributing not only to environmental protection but also to improving soil health and, consequently, crop quality [11]. The concept of biofertilizers has progressed over time, and today we can define them as compounds that include living microorganisms, which stimulate plant growth and thus increase the nutrient resources in the plant rhizosphere [81]. Unlike chemical fertilizers, the use of biofertilizers in the soil helps to develop a more balanced and differentiated soil ecosystem, promoting the nutrient cycle and optimizing both soil structure and fertility.

An excellent example is the study conducted on *Chlorella vulgaris* as a biofertilizer in tomato cultivation (*Solanum lycopersicum* L.). In this investigation, a marked improvement in the growth and yield of tomato plants [82] was noted thanks to the use of *C. vulgaris*, as well as an improvement in the nutritional composition and shelf life of the fruit. Similarly, further improvements were observed in the use of *C. vulgaris* on the growth of common beans (*Phaseolus vulgaris*), where the biofertiliser led to a 26.9% increase in the total length and height of the beans, a 37.28% increase in dry weight, and an increase in protein and carbohydrate content during the vegetative phase. Furthermore, during the fruiting phase, an increase in the number of pods per plant (from 3.4 to 5.2), the number of seeds/pod (from 2.6 to 3.5), and the dry weight of the pod (from 0.67 to 0.95 g·plant<sup>−1</sup>) was measured [83]. In line with these results, *C. vulgaris* extract had positive effects on the growth of lettuce seedlings, increasing fresh and dry weight, chlorophyll, carotenoids, protein content, and ash in shoots [84]. Furthermore, biofertiliser extracts from *C. vulgaris* and *S. platensis* have been shown to improve the growth of mung beans (*Vigna radiata*), including shoot and root length and weight during the flowering stage [85].

These treatments also improved the physical characteristics of the plant, such as water and oil absorption, and positively affected soil pH and minerality [39]. Recently, it has been confirmed that *Arthrospira* sp. contributed to increased plant growth and stress tolerance [86], probably due to the synergistic action of various bioactive molecules, including phytohormones, polysaccharides, vitamins, and amino acids. Microalgae are particularly suitable for removing contaminants due to their high bioabsorption potential. The structural composition of the carbohydrates located in the cell wall of microalgae promotes the bioabsorption of toxic pollutants [87,88]. A new study [89] showed that bioabsorption plays a key role in the elimination of pesticides such as atrazine, carbofuran, dimethoate, and simazine by living *C. vulgaris*. Furthermore, the physicochemical properties of molecular size and structure have a significant impact on the bioabsorption process. In fact, the increase in the size of algae particles leads to greater surface availability, thus promoting the bioabsorption of pesticides [90]. Therefore, microalgae can effectively eliminate pesticides through bioabsorption [91].

### 3.3. Limitations

Microalgae have demonstrated remarkable capabilities in agriculture, thanks to their ability to deliver useful substances and support environmentally friendly farming practices. Nevertheless, there are still several restrictions that prevent their use in agriculture. One of the main restrictions is the high cost of producing microalgae biomass and removing significant metabolites [69]. Another high cost is the supply of nutrients, including nitrogen, phosphorus, and carbon dioxide, which are essential for algae growth [92]. Recently, it has been observed that microalgae production through wastewater treatment has several disadvantages, as microalgae have the property of storing heavy metals and other contaminants present in wastewater, and their improper use can lead to the

introduction of these pollutants into agricultural soil, potentially causing damage to crop development and human health [3]. A comparative summary of nutrient profiles, microbial risks, production costs, and sustainability aspects of chemical, frass-based, and microalgae-based fertilizers is provided in Table 2.

**Table 2.** Comparison between chemical, frass-based, and microalgae-based fertilizers.

Parameter	Chemical Fertilizers	Frass-Based Fertilizers	Microalgae-Based Fertilizers
N Content	High, Mineral	Moderate–High, Organic	Moderate
P Content	High	Moderate	Low–Moderate
K Content	High	Moderate	Low
Micronutrients	Limited	Rich And Variable	Rich (Fe, Zn, Mg)
Microbial Risk	Absent	Possible If Untreated	Low–Moderate
Production Cost	High (Energy-Intensive)	Medium	High
Environmental Impact	High	Low	Low
Circular Bioeconomy	No	Yes	Yes

#### 4. Frass and Microalgae as Living Fertilizer

A key challenge in environmental sustainability is the replacement of significant amounts of chemical fertilizers with nutrients from waste or residual flows, and one example is the use of insect frass. In this context, insect frass represents a promising organic nutrient source derived from the bioconversion of organic waste, while microalgae offer an additional biological upgrading step capable of transforming these nutrients into value-added biomass. The integration of insect frass and microalgae could be applied to make microalgae production more sustainable, cost-effective, and part of a circular bioeconomy [6].

From a process perspective, the frass microalgae system can be described as a sequential chain in which organic waste streams are first converted into insect biomass and frass through insect rearing. The collected frass, after appropriate pre-treatment steps such as extraction, dilution, or sterilization, is subsequently used as a nutrient source for microalgae cultivation. The resulting microalgal biomass can then be applied directly as a biofertilizer or further processed into biostimulant formulations, allowing the biological upgrading of waste-derived nutrients while reducing dependence on external mineral inputs.

##### 4.1. Insect Frass Used for Microalgae Cultivation

Microalgae cultivation using waste streams as an alternative nutrient source is currently a major area of interest for environmental sustainability. Numerous studies have focused on the application of insect excreta for low-cost microalgae cultivation, aimed at developing a circular bioeconomy [12]. In a new study [12], the frass considered was obtained from the rearing of subadult house crickets (*Acheta domesticus*). In this case, their composition showed a crude protein content of 36%, which corresponded to a total nitrogen content of 6% in the excreta. This was done to evaluate the nutrient supply that the frass could provide, without adding any additional requirements. The resulting frass was used to cultivate three species of microalgae, i.e., one freshwater, *Chlorella vulgaris*, and two marine, *Phaeodactylum tricornutum* and *Nannochloropsis salina*.

Another study highlighted the use of another insect species, *T. molitor*, for microalgae cultivation. Mealworms were raised on organic waste for feed and food containing a high protein content, and were only sieved before use. Given that the amount of nutrients was essential for microalgae growth, to make the frass nutrients more available, they were eluted with water to remove the insoluble fraction, and the soluble fraction was sterilized to avoid any bacterial contamination. In this case, based on the type of sterilization performed, autoclaving and sterile filtration, it was observed that autoclaving was an optimal method for recovering nutrients from *Tenebrio molitor* frass, with a higher dissolved organic carbon content of approximately 26% and a higher total dissolved nitrogen concentration of approximately 31%, compared to the application of sterile filtration. The phosphate-phosphorus concentration was also evaluated, which was found to be the same in both treatments (0.8%). This was evaluated with the aim of being able to apply the frass obtained from *Tenebrio molitor* for the cultivation and growth of *C. vulgaris* [6].

##### 4.2. Microalgae Species Produced with Insect Frass

The species cultivated with insect frass are diverse. In a study [12], three species of microalgae were considered, including one freshwater species, *Chlorella vulgaris*, and two marine species, *Nannochloropsis salina* and *Phaeodactylum tricornutum*, derived from the frass cultivation of an insect species, *Acheta domesticus* [93,94]. Given the high protein content of the frass considered, nitrogen, being an essential macronutrient for their growth,

was found in frass-water media mainly in its reduced form of ammonia, with concentrations below the toxic effect [95,96]. Their cultivation allowed the growth of species-specific microalgae.

The results obtained from a recent research [12] show that cultivating freshwater microalgae, including *C. vulgaris*, with insect frass is an optimal and environmentally sustainable method, as it does not compromise microalgal growth. On the other hand, cultivating marine microalgae with insect frass, including *N. salina* and *P. tricornutum*, proved limiting, as the conditions were suboptimal with respect to pH and nutrient imbalance. In this regard, cultivating *C. vulgaris* with insect frass from a different species (*T. molitor*) was studied.

This study revealed that the growth of *C. vulgaris* exhibited several key components for its development, determined by cultivation with *T. molitor* frass, including total protein composition and microbial composition. From this, it was possible to deduce that the protein quality obtained from *C. vulgaris* grown with frass was satisfactory for their growth, allowing them to adapt and stabilize in the culture medium. Depending on the culture medium, *C. vulgaris* was able to efficiently utilize nitrogen, primarily due to the mineralization of organic compounds [97–99]. Similarly, however, phosphorus uptake is influenced by its bioavailability and the ability of *C. vulgaris* to accumulate it in the form of intracellular polyphosphates. From a microbial perspective, it was observed that culture media containing *T. molitor* frass favor the development of high bacterial counts, but without inducing negative effects on the growth of *C. vulgaris*. Indeed, they could lead to the creation of beneficial symbiotic relationships between bacteria and microalgae [6].

#### 4.3. Nutrient Conversion Pathways and System Limitations

Unlike direct soil application of untreated frass, frass–microalgae systems rely on a sequence of biological transformations that can change both nutrient form and bioavailability. In aqueous frass-derived media, part of the organic nitrogen pool can be progressively mineralized by the associated microbial community, supporting algal uptake of inorganic N while also sustaining bacteria–algae cross-feeding interactions in the phycosphere. Such interactions are known to influence nutrient cycling and, depending on environmental conditions and partners, may enhance algal productivity and stability rather than simply increasing microbial load. Regulatory considerations within the European Union [100]. Phosphorus dynamics also reflect active biological regulation. Microalgae can respond to fluctuating phosphate availability by modulating uptake systems and by storing phosphorus intracellularly in polymeric forms (polyphosphate), a strategy widely documented across microalgal taxa and relevant for media derived from waste streams where P availability may vary over time [101]. Despite these mechanistic advantages, several barriers currently limit transferability beyond lab scale. The most recurrent issues include frass heterogeneity (linked to insect diet and rearing conditions), the need for pre-treatment steps to manage turbidity and microbial safety, and nutrient imbalances that can constrain growth depending on the target strain (particularly in non-freshwater settings). On the downstream side, harvesting and dewatering remain among the most energy- and cost-intensive steps of microalgal production, often representing a practical bottleneck for scale-up [102].

#### 4.4. Regulatory Considerations within the European Union

Regulatory alignment is another key factor determining whether integrated frass–microalgae products can realistically reach the market. In the European Union, the Fertilising Products Regulation (EU) 2019/1009 establishes the rules for placing CE-marked EU fertilising products on the market, including requirements related to safety, labelling, and limits for certain contaminants and pathogens [103]. In practice, frass- and microalgae-derived products may fall under different functional categories (e.g., organic fertilisers, soil improvers, plant biostimulants), but their eligibility depends on how the product is defined, processed, and documented with respect to the Regulation's component material categories and conformity assessment routes. As a result, even when the agronomic rationale is strong, regulatory uncertainty and inconsistent interpretation can slow down commercialization, particularly for novel “integrated” formulations that combine multiple bio-based components [103].

### 5. Conclusions

Insect excrement and microalgae are complementary and highly promising biological resources for the development of new-generation fertilisers, which combine nutrient supply with improved soil health and environmental sustainability. Their integration into circular bioeconomy models, in which insect excrement serves as both fertiliser and growth substrate for microalgae, offers a concrete strategy for nutrient recycling, waste valorisation and reduced dependence on synthetic fertilisers. In the future, research efforts should move beyond proof-of-concept studies towards standardised production and application frameworks. Priority directions include: chemical and microbiological standardisation of excrement and microalgal biomass to reduce compositional

variability; optimisation of integrated production systems to improve scalability, economic viability and life-cycle performance; and risk assessment and regulatory alignment, particularly with regard to microbial safety and compliance with fertiliser legislation. Furthermore, the combined use of insect excrement and microalgae opens up new application scenarios in precision agriculture, customised biofertiliser formulations and biorefinery-based nutrient recovery systems.

### Author Contributions

C.F.: conceptualization; G.C. and E.C., writing—original draft preparation; E.C., S.S. and G.C.: visualization, investigation; C.F. and M.G.R.: supervision; C.F. and M.G.R.: validation; F.I., M.G.R. and C.R.M.: writing—reviewing and editing. All authors have read and agreed to the published version of the manuscript.

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### Use of AI and AI-Assisted Technologies

During the preparation of this work, the authors used Perplexity to correct English grammar. After using this tool, the authors reviewed and edited the content as needed and takes full responsibility for the content of the published article.

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