

Review

Microbial Community in the Composting Process and Its Positive Impact on the Soil Biota in Sustainable Agriculture

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Abstract: Conventional agriculture has important challenges to guarantee soil fertility and sustainable food production. Many agricultural soils in the world are degraded and multiple strategies are currently being developed to restore them. The study of beneficial soil microorganisms has attracted increasing interest due to their relevant role in sustainable agricultural development. The balance and maintenance of ecosystem services, such as biomass transformation, nutrient cycling, plant growth, and health, are directly dependent on soil microbial activity. Therefore, it is important to promote its establishment and propagation. An ancient technique that favors soil biodiversity is the production and application of compost. While numerous studies have focused on the benefits of plant cultivation, fewer studies have focused on the benefits for soil microbiota. The objective of this review was to elucidate the role of the microbiota in the composting process and its impact on soil microorganisms in agriculture. The review presents the advances in the knowledge and importance of microorganisms involved in the composting process and how compost promotes the maintenance and multiplication of beneficial microbial consortia and their ecosystem functions in agricultural soils, shifting towards a more sustainable and resilient agriculture.

Keywords: compost; composting application; soil microbial consortium; bacterial community; fungal community



Citation: Aguilar-Paredes, A.; Valdés, G.; Araneda, N.; Valdebenito, E.; Hansen, F.; Nuti, M. Microbial Community in the Composting Process and Its Positive Impact on the Soil Biota in Sustainable Agriculture. *Agronomy* **2023**, *13*, 542. <https://doi.org/10.3390/agronomy13020542>

Academic Editors: Jadwiga Stanek-Tarkowska and Yiqiang Li

Received: 9 December 2022

Revised: 7 February 2023

Accepted: 8 February 2023

Published: 14 February 2023



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1. Introduction

One of the main problems of agriculture is the loss of soil fertility, this generates a reduction in crop productivity, mainly due to the loss of ecosystem functions of soil microorganisms [1,2]. Soil comprises a highly dynamic reservoir of biodiversity, in which microorganisms, animals, and plants interact, sustaining the biodiversity of ecosystems. Biodiversity stabilizes ecosystems functioning under fluctuating environmental conditions, therefore, biodiversity increases ecosystem resilience and productivity under extreme climates and/or climatic events.

The soil microorganisms in agriculture regulate the ecosystem services [3], such as the control of organic matter decomposition, availability, nutrient cycling, soil electrical conductivity, pathogen control, pollutant degradation, and greenhouse gases (GHG) reduction. The knowledge of soil microbial communities contributes to the development of sustainable and environmentally friendly agriculture, and it is important to promote their care and proliferation, also in connection with the type of fertilization [4–6].

Compost is one of the most used and ancient fertilisers that promote soil biodiversity. Compost application is a beneficial practice for soil restoration [7]. It provides an important source of nutrients, improves soil structure [8–10], maintains and improves fertility, increases the productivity of agricultural soils [11], stimulates the rate of vegetation establishment, enhances root development [12], and allows the establishment of beneficial microorganisms and the suppression of soil pathogens [13,14]. The potential application of compost is largely derived from the quality of the production process and the final product obtained [15,16]. The technique to obtain compost is composting, a biological process mediated by different microorganisms, which transforms complex compounds from the biomass to simpler molecules through a process of oxidative or enzymatic hydrolysis, characterized by different thermophilic stages called mesophilic, thermophilic, cooling, and maturation phase, where the entire composting process occurs. The final product is a humus-like, stable, dark, and highly nutritious substrate, whose specific composition depends on the origin of the composted waste [10,17–20]. The establishment and activity of microbial communities in the composting process depends on many factors, such as starting material and environmental operating conditions (temperature, aerobiosis, moisture content, organic matter, and C/N ratio), both directly affecting the proliferation of different species [21,22].

Recently, the development of high-throughput sequencing technologies has provided a means to elucidate the soil microorganism communities present in the composting process [6,23]. It has been described, on the one hand, that bacteria play an important role, as they have a large specific surface area that allows them to rapidly absorb soluble substrates. Thus, the abundance of bacteria is usually much higher than that of fungi. However, on the other hand, the role of fungi within the composting process is equally relevant, as they produce a large quantity and variety of extracellular enzymes that allow the degradation of recalcitrant plant tissues, such as cellulose and lignin [23,24].

The present review focuses on composting process, identifies the main microorganisms involved in the process, and their impact on those microorganisms in agricultural soils that are favored by compost application. Studies with high impact in the area, spanning from 2000 to 2023, have been considered in this review. The latter is organised into three sections that include: The latter is organised into three sections that include:

Section 2: The Microbiota in the Composting Process;

Section 3: Conditions that affect the succession of microbial communities in the composting process;

Section 4: Promotion of soil microorganisms by compost application.

2. The Microbiota in the Composting Process

The dominant bacterial phyla in the composting process are: *Firmicutes*, *Actinobacteria*, *Proteobacteria*, *Bacteroidetes*, and *Chloroflexi* [25]. *Firmicutes* play an important role in lignocellulose degradation [26]; *Proteobacteria* are closely related to the mineralization of nitrogenous organic substrate; *Bacteroidetes* are involved in the degradation of a wide range of complex carbohydrates [27]; *Actinobacteria* are effective microbes for producing hydrolytic enzymes involved in the breakdown of lignocellulose and recalcitrant cellulose [28], in addition to the suppression of pathogenic microorganisms by the secretion of antibiotics [29]; *Chloroflexi* is related to the biogeochemical chlorine cycle, the degradation of soluble microbial products, and it has a crucial role in the degradation of hemicellulose under thermophilic conditions of the composting process [25,30]. The main microbial taxa present in the composting process are described in Figure 1.

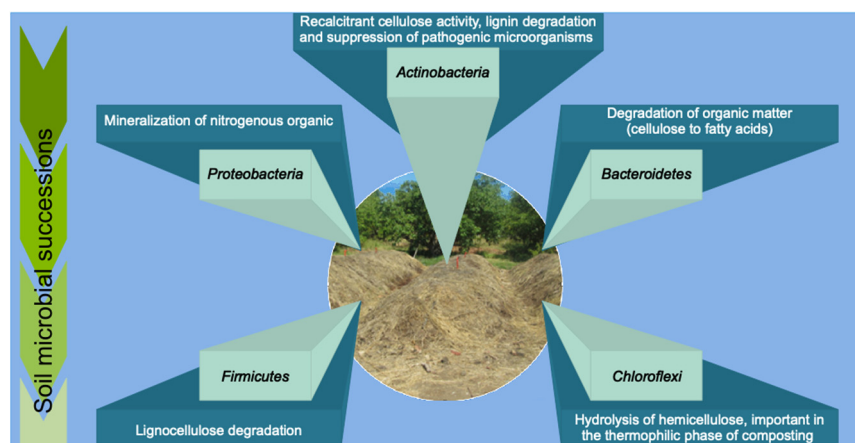


Figure 1. Dominant bacterial phyla in the composting process and their main functions.

2.1. *Firmicutes*

Bacteria of the *Firmicutes* clade are mostly Gram-positive, non-filamentous bacteria, abundant in rhizospheres of plants [31], some species can form spores, resting stages that are inactive, strongly dehydrated, and highly resistant to environmental stresses [32].

Firmicutes species are widely studied for their beneficial effects on the development of sustainable agricultural systems. Among the benefits are the promotion of plant growth, helping them to acquire resources such as nitrogen, iron, phosphorus, or other minerals. They also allow the modulation of plant hormone production or the production of direct plant hormone analogues, and contribute to the biological control of plant pathogens (biocontrol agents) [31]. Other soil-related challenges addressed by the application of these microorganisms are soil desertification, decreased salt content in the soil, as well as contamination by organic substances (e.g., pesticides), and inorganic pollutants (e.g., metals) by breaking down recalcitrant molecules using their specific metabolic pathways [31].

Firmicutes have been considered the best-represented group of bacteria in the composting processes, due to, their participation in the decomposition of plant biomass [21,33]. *Firmicutes* are the dominant phylum during the initial stages of the composting process (mesophilic stage), and gradually decrease as the process progresses [34]. However, some *Firmicutes* can survive well at high-temperature conditions as thermotolerants, i.e., living at 30–50 °C, or as thermophiles i.e., living at above 50 °C [35].

The genera living at the widest temperature range (20–60 °C) are usually *Brevibacillus*, *Geobacillus*, *Bacillus*, and *Aeribacillus* [36–38]. Some members of *Firmicutes* play an important role in the degradation of lignocellulose, synthesizing various proteases and pectinases, and are able to degrade non-digestible carbohydrates such as cellulose [39,40], i.e., *Thermoanaerobacterium* (including the species formerly known as *Clostridium thermosaccharolyticum*), *Pseudoclostridium*, *Caldibacillus*, and *Thermohydrogenium*; they are strong producers of endoglucanases and exoglucanases belonging to GH1, 5 and 10 families [41].

2.2. *Actinobacteria*

Actinobacteria are a large group of Gram-positive bacteria with a cosmopolitan distribution. The actinobacteria species are distinguished from other bacteria by their morphology, resembling fungi because of their elongated cell branches into filaments as fungal hyphae by DNA rich in guanine and cytosine, and by generating the typical wet soil odor [42]. Thus, they show pleomorphic morphology and even coccoid elements, forming long filaments that extend through the soil and many of them produce spores that are easily detached.

Actinobacteria species are valuable microorganisms that play a crucial role in plant growth and yield and possess a multifunctional role beneficial for sustainable agricultural production. Among actinobacterial species, free-living symbiotic or diazotrophic bacteria able to solubilize potassium and/or phosphate, plant growth promoting and biocontrol

bacteria (antivirus, antifungal, etc.), and abiotic stress mitigators and plant probiotics have been described. Especially, species belonging to the genus *Streptomyces* are involved in crop growth and health [43]. *Streptomyces* species are considered inhibitors of phytopathogenic microorganisms since they can produce antibiotics [44]. It has also been considered as plant growth-promoting bacteria (PGPB) since they synthesize phytohormones, solubilize phosphate, or induce nitrogen fixation [45,46].

Actinobacteria have a special interest in compost production due to their ability to decompose plant biomass [47]. The Actinobacteria possess the ability to decompose the lignocellulose present in plant tissues, as well as chitin or insect exoskeletons, thanks to the extracellular enzymes they produce, such as alpha-amylase, glucoamylase, glucose isomerase, proteases, lignin peroxidase, among others [48]. They can grow in compost in both mesophilic and thermophilic conditions, up to 50–60 °C, and at neutral and alkaline pH.

Studies on Actinobacteria development in compost have shown a predominance of *Streptomyces* spp., with grey aerial mycelium, *Micromonospora* spp. [48], *Thermoactinomyces* [49], *Thermomonospora* and *Actinobifida* spp. Some species of actinobacteria are thermotolerant, living in the warmth of a hot active compost, where the members of the genus *Microbacterium* predominate [37,38]. The species of this genus live at the widest range of temperatures up to the long maturation stage of compost, and they spread their hyphae-like threads throughout the compost [37].

Thermoactinomyces spp. and *Microtetraspora* spp. usually colonise composts prepared from animal manure and straw, growing abundantly during the thermophilic phase and release many spores. Composts made from household green waste are often colonised by *Streptomyces* spp. and *Thermoactinomyces* spp., such as *T. vulgaris*, *T. thalophilus*, *S. rectivirgula*, *T. fusca*, *T. alba*, and *T. curvata*. Members of the genus *Thermoactinomyces* are particularly advantageous because, during the composting phase at high temperatures, they can degrade bioplastics such as polyethylene succinate (PES), poly(ϵ -caprolactone) (PCL), and poly(3-hydroxybutyrate) (PHB) [50].

2.3. Proteobacteria

Proteobacteria comprise the largest and most diverse phylum of bacteria and are of widespread phylogenetic, ecological, and pathogenic importance [51]. They are all Gram-negative, with an outer membrane containing lipopolysaccharide [52]. Bacteria within this phylum show considerable differences in morphology, motility, and metabolism. Although most Proteobacteria are motile with polar or peritrichous flagella, the unusual gliding movement has been documented, as in *Myxobacteria*, while some members of this phylum are non-motile [53].

The phylum Proteobacteria contains a diverse spectrum of bacteria of environmental importance, many of which are found in the soil rhizosphere of agricultural crops and weeds, and are characterised by a significant ability to survive the selective pressures of agriculture. They can retain moisture and are presumed to possess an important role in nutrient cycling. Proteobacteria are the exclusive producers of the quorum-sensing molecule *n*-acyl-homoserine lactone (ahl), which triggers beneficial responses to maintain soil health, but may also be harmful, as it may contribute to plant disease dynamics [54].

Their function within the composting process is the anaerobic degradation of organic matter [17], with an important role in the sulfur and nitrogen cycle, and participates in the carbon cycle [51,55,56]. Proteobacteria are the most abundant phylum detected at the beginning of the composting process, i.e., in the mesophilic stage, and their relative abundance decreases in the thermophilic stage as well as during the rest of the process [57]. The main examples of proteobacteria genus found in the mesophyll stage of composting are *Stenotrophomonas*, *Halotalea*, *Pseudomonas*, and *Acinetobacter* [57].

2.4. Bacteroidetes

The phylum *Bacteroidetes* is a very diverse bacterial phylum. These bacteria are all Gram-negative, covering a mixture of physiological types, from strictly anaerobic Bacteroides, to strictly aerobic Flavobacteria. They are non-motile, flagellated, or gliding motile, widely distributed in different habitats, especially in soil [58]. They have an important ecological role and are well-known degraders of polymeric organic matter [58].

Their function within the composting process is the anaerobic degradation of ligno-cellulosic biomass [17], generating short-chain fatty acids [59]. Their abundance is higher during the cooling and maturation stage of the composting process, where cellulose and xylan are mainly used as a carbon source [60]. The main species of the Bacteroidetes associated with soil are *Flavobacterium*, *Pedobacter*, *Cytophaga*, and *Spirosoma* [58].

2.5. Chloroflexi

Members of *Chloroflexi* are filamentous Gram-negative chemo-lithotrophic or heterotrophic bacteria and have been reported as non-motile and motile (by gliding and flagella) [61]. Most *Chloroflexi* bacteria are moderate thermophiles, usually isolated from hot springs or identified in aquatic environments, including marine and freshwater sediments, and they have also been reported in soil [61]. They are difficult to isolate in pure culture, and so far, most cultured strains have been isolated from thermal habitats. Members of *Chloroflexi* are metabolically diverse but are notable for their ability to fix inorganic CO₂ and to aerobically oxidize carbon monoxide and nitrite. They also aerobically reduce nitrate and ferric iron [61].

Within the composting process, *Chloroflexi* is one of the main phyla in the maturation phase [25]. The phylum *Chloroflexi* is related to the biological nutrient removal process, the biogeochemical chlorine cycle, and the degradation of soluble microbial products [25], but its most important role is the degradation of hemicellulose through the hydrolysis of the internal glycosidic linkages of the heteroxylan backbone (endoxylanase) under the thermophilic phase of composting [30]; this explains its strong increase through the entire process, being the most abundant phyla in the mature compost [62,63].

The *Anaerolinaceae* family is a representative of the phyla *Chloroflexi* commonly found in compost. Members of the *Anaerolinaceae* family are known to participate in hydrocarbon degradation [64].

2.6. Fungi

Fungi are important decomposers of organic matter in nature and in the composting process. Fungi of the phylum *Ascomycota* are the most dominant throughout the composting process, followed by *Basidiomycota* and the subphylum *Mucoromycotina* have been reported in composting of cow manure, food, garden waste, sewage sludge, and corn straw [22,65–67]. Yeast species of the orders *Saccharomycetales* and *Tremellales* are found in greater numbers in the early stages of composting with the genera *Candida*, *Pichia*, and *Trichosporon* [68], fungal species reported in later stages include fungi of the genera *Arthrobotrys*, *Nectria*, *Thermomyces*, *Coprinus*, *Cryptococcus*, *Conocybe*, *Mortierella*, *Candida*, *Leucoagaricus*, *Malassezia*, *Phialopora*, and *Cercophora* [66,68,69].

It has been observed that in the composting process, in the thermophilic phase when the temperature reaches more than 65 °C, a significant development of fungi is not found. Temperature is one of the most important factors affecting fungal growth. Most fungi are mesophilic with an optimal temperature of 25–30 °C. Yeasts disappear during the thermophilic phase of composting, but when the temperature cools down to 54 °C, several genera of fungi capable of degrading cellulose, carboxymethylcellulose, hemicellulose, xylan, and arabinoxylan can be found again [33,66] as indicated in Table 1.

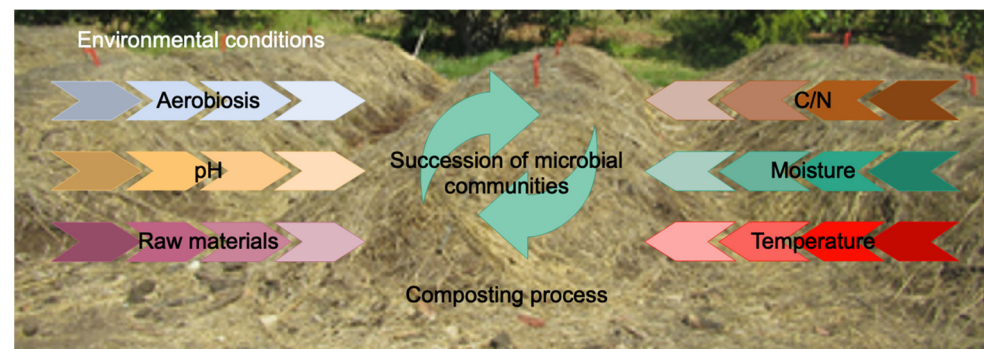
Table 1. Dominant fungal phyla in the different stages of the composting process.

Composting Stages	Dominant Phyla	Maximum Temperatures (°C)	References
Mesophilic	<i>Saccharomycetales Tremellales</i>	>25 °C	[68,69]
Thermophilic	Eurotiales Sordariales Hypocreales Microascales	>50 °C	[65]
Cooling	Eurotiales Sordariales	50–40 °C	[65]
Maturation		40 °C down to environment temperature	

Fungi can degrade cellulose and lignin more easily than actinomycetes and bacteria. Fungi degrade lignin better at low N levels, and although they can grow in wide pH ranges, they prefer a more acidic environment. Their extensive hyphal network permit to attack the organic residues that are not available to bacteria. [70–72].

3. Conditions That Affect the Succession of Microbial Communities in the Composting Process

The different microorganisms involved in compost production are grouped in communities that follow one another in a non-random fashion during the process, i.e., when the ability of a species to carry on its physiological functions is hampered by environmental variations (physical or chemical), other organisms become the main component of the process [73,74]. In general, communities are affected by environmental variations related to temperature, aerobiosis, feedstock, and humidity [75]. Controlling these conditions will directly impact the permanence of each community and, in turn, the duration of the process and the final quality of the compost [76] (Figure 2).

**Figure 2.** Environmental conditions influencing the composting process and soil microbial successions.

The other microorganisms that are present in the composting process, such as pathogens or inoculants, also modify the composting process in different ways.

3.1. Temperature

Temperature is one of the most important factors affecting the composting process. The latter is typically characterised into three stages: (a) mesophilic (temperature >25 °C), in which degradation of simple molecules such as sugars and amino acids, etc., occurs through the metabolism of mesophilic bacteria and fungi; (b) thermophilic (called also sanitization phase, with temperature >50 °C), where thermophilic microorganisms degrade organic matter (fats, cellulose, hemicellulose, and lignin); (c) maturation/stabilization (temperature <50 °C up to environment level) or cooling phase, characterised by lower microbial activity and a decrease of temperature. In this last stage, the compost mass is recolonized by mesophilic microorganisms that degrade residual sugars, cellulose, and

hemicellulose, materializing humic-type substances [77,78]. High temperatures result from strong microbial activity, which is possible in the presence of sufficient water, nutrients, and oxygen, releasing energy through respiration faster than it can be dissipated. Although the temperature range and the duration of each of the three phases vary according to the raw material, the addition of bulking agents, and the technology used, the optimal temperature range for each of the phases is (a) environment to 50 °C, (b) 50–65 °C for a minimum of 5–3 days to allow sanitization to occur, and (c) 65 °C to environmental for maturation/stabilization of the compost. There are variants to the process, from short (one week), to average (few weeks), to long-lasting (two-three months) industrial processes according to the amount of recalcitrant compounds such as lignocelluloses and the technology used. Table 2 summarizes the dominant species in the different stages of the composting process according to the temperature changes.

It has been observed that high temperatures in the composting process have a positive effect on promoting compost maturity and removing harmful organisms (pathogenic bacteria, weeds, and intestinal worms, among others). In this context, an innovative hyperthermophilic composting (HTC) technology has been developed. The temperature reaches approximately 90–100 °C [79], which is 20–30 °C higher than that of conventional composting. The technique is beneficial in maximally decreasing pathogen load, increasing the rate of organic matter degradation and nutritional nitrogen retention, and thus, a favorable compost maturity in a shorter period [80]. In addition, HTC accelerates humic acid formation [81], microplastic degradation [82], a decrease of CH₄ emission [83], decreases N₂O emission [84] elimination of antibiotic resistance genes and mobile genetic elements (plasmids, transposons) that can occur within HTC, thus minimizing the horizontal gene transfer which might represent a risk for both human and environmental health [85].

HTC can be divided into three stages: hyperthermophilic (≥ 80 °C), thermophilic (50–80 °C), and maturation (<50 °C). The most abundant genus in the hyperthermophilic stage is thermus, which produces many enzymes (hydrolytic and catalase), stable at high temperatures, allowing it to considerably exceed the upper limit of growth compared to most organisms. During the thermophilic stage, there is a shift in the microbial community being more abundant in *Planifilum* (*Firmicutes*) [85].

Table 2. Dominant bacterial phyla during the different stages of the composting process.

Composting Stages	Dominant Phyla	Maximum Temperatures (°C)	References
Mesophilic	<i>Actinobacterias</i> <i>Firmicutes</i> <i>Proteobacterias</i>	>25 °C	[78]
Thermophilic	<i>Actinobacterias</i> <i>Firmicutes</i>	50–65 °C	[86]
Cooling	<i>Bacteroidetes</i>	50 to 40 °C	[86]
Maturationn	<i>Chloroflexi</i>	40 °C to room temperature	[86]

3.2. Aerobiosis

Aeration rate has an important effect on microbial activity and degradation rate. Aeration is directly related to O₂ concentration and is one of the most influential factors during composting. An adequate pO₂ not only promotes the decomposition of organic waste but also reduces greenhouse gas (GHG) emissions as CH₄ [87–89]. Intermittent aeration was found to increase the efficiency of the O₂ supply compared to the continuous aeration system [90]. When excessive, aeration leads to cooling and reduced thermophilic conditions [88]. In addition, it causes water losses, hinders moisture control [87], and increases NH₃ and N₂O losses, though it produces higher mitigation of CH₄ emissions [91] affecting the establishment of some microorganisms. Controlling the relative abundance of the phylum *Firmicutes* has been shown to be closely related to reducing greenhouse gas emissions [6].

3.3. pH

In the early stages of the composting process, a decrease in pH is observed, and it is suggested that this may be due to NH_3 volatilization and microbial nitrification which produces higher amounts of CO_2 and acids [92,93]. A very low pH could represent a challenge when composting is carried out at the industrial level, as the transition from the mesophilic to the thermophilic phase could be negatively affected [94]. However, in the later stages of the composting process, there is an increase in pH that could be attributed to the degradation of nitrogen-containing compounds as ammonia is released and accumulates in the compost piles [95,96]. A pH range of 7.5–8.5 during composting supports the decomposition of organic matter by microorganisms. For other authors, [97] the optimum pH range is established between 5.5–8.

3.4. Humidity

Humidity content greatly affects the physical and chemical properties of waste materials during organic matter degradation [98]. The initial humidity content of animal manure composting is between 60–65%, and that of the final products is between 30–50% [99]. In the case of other plant-derived biomasses such as olive husks, the humidity can be as high as 60–70% at the start [100] (in piles or windrow composting) and 25% with air-dried final compost in temperate regions. In the composting process, the population of microorganisms, mainly bacteria, increases with moisture content [20] as water is a medium for dissolved nutrients necessary for microbial metabolism and physiological activity. On the other hand, excessive humidity triggers anaerobic conditions because the pore spaces of solid matrices are filled with water instead of air [101]. It should be borne in mind that drying the stabilized compost is crucial for its proper conservation and also to avoid microbial re-colonization. Therefore, it would be desirable to store the final product dried at 18% whenever feasible at the industrial level.

3.5. Raw Materials/Composting Methods

The feedstock as well as physicochemical parameters alter the succession of the microbial community in the composting process [55]. It has been clarified that composts with higher amounts of cellulose and lignin are the first to enter the cooling phase. These residues have a high organic carbon content, low heavy metal content, and low nitrogen, phosphorus, and total potassium content; in general, the C/N ratio is greater than 25. Therefore, it is recommended to mix them with nutrient-rich materials such as food waste or manure, among others [78,102]. It is recommended to reduce the particle size of the lignocellulosic biomass to make the mixture homogeneous and to increase its porosity, thus, the oxygen content and circulation by the addition of bulking agents. This will favor the establishment and proliferation of microorganisms that degrade the organic matter. It is important also to adopt composting management to separate waste collection at the source. Indeed, composting is far better and less dangerous for human health and the environment when the organic fraction of waste is separated at the source to avoid mixing it with other undesirable fractions of municipal solid waste.

There are multiple methodologies to produce compost that are determined by the experience of the producer, the availability of suitable raw materials, and bulking agents for composting or co-composting, the size of the area where the process is carried out (decentralized at a small scale or centralized at semi-industrial or industrial scale), in piles or windrows, with or without forced ventilation, and the times of the composting process. Bacterial and fungal communities respond to both compost recipes and composting methods [103]. The methodology for combining the starting materials, the size of the piles, the frequency of turning, etc., will particularly affect the microbial communities [86]. More studies are required to systematize the different methods and recipes of composting and their impact on microbial communities.

3.6. C/N

During composting, microbes break down organic compounds to obtain energy for metabolism and acquire nutrients to sustain their growth. However, C and N are the most crucial: C is used as an energy source, while N is used to build cell structure. A nutritional balance in the form of an optimal C/N ratio is essential to formulate an efficient compost mix. According to some authors [104], the ideal C/N ratio of the initial mix to be composted should be 25–30, although higher C/N ratios such as 36, as in the case of virgin olive pomace [105] allowed to obtain the same final compost, at farm level, though in a slower composting time. In the latter case, due to the high lignocellulose content along with the presence of high content of polyphenols and fats (both inhibitory microbial activities), starter cultures were used at the beginning to counteract inhibition. As composting progresses over time, variations in C/N projected organic degradation rate governed by the extent of carbon transformed to CO₂ [106]. On the one hand, at a lower C/N ratio, N will be available in excess and will be lost as ammonia gas, resulting in an undesirable odor. In addition, a large amount of soluble basic salt is released, making the soil unfavorable for plant growth [107]. On the other hand, a higher C/N ratio slows down the composting rate and generates nutrient deficiency due to excessive substrate accumulation [106]. Other authors [108] found that a higher C/N (30:1) appears to promote the relative abundance of nitrogen-fixing bacteria while compressing denitrifying bacteria.

Therefore, to optimise C/N for composting purposes, a variety of approaches can be used, including the addition of bulking agents to the waste (e.g., sawdust, rice husks, peanut shells, and wood chips) and the homogenization of the waste prior to composting to improve the porosity of the initial mix [78]; the addition of microbial starter cultures to high-lignin containing materials can be beneficial to reduce the process to industrially acceptable times [109,110].

3.7. Pathogens in the Composting Process

One of the challenges of the composting process is the inactivation of pathogenic bacteria coming mainly from animal manure, which is essentially one of the main objectives [50,111]. The temperature inside a compost pile reaches values above 70 °C, reducing the maximum contaminants limit allowed by most international regulations, or eliminating pathogenic bacteria and viruses [111–113]. In the composting process, the application of raw or poorly composted organic manure or poor management of the process can induce the spread and dispersal of pathogenic bacteria such as *Escherichia coli*, *Enterococcus* sp., and *Salmonella* sp. [114]. This is the case of the sheet and the trench composting: in the first one, the organic material is spread on the soil or ground, allowing it to decompose naturally, while the second one consists in filling of organic materials into trenches 15–20 cm deep [113]. Inactivation, or destruction of indicator pathogenic organisms, is expected to occur if all compost particles maintain temperatures above 55 °C for at least three days (which occurs within the thermophilic phase of the composting process) [115]. This is the case of in-vessel composting, which is the largest scale of this type of process and is made in large size closed metal tanks, drums, or concrete containers, with a rotation of the compost material at 3–5 rpm, destroying the pathogens in 3–4 d [113].

Moreover, it has been reported that the elimination of pathogenic bacteria depends on the initial substrate, how long the thermophilic stage lasts, and at what temperature it is maintained. Pathogen survival has been reported (Table 3), attributed to uneven heating temperatures (the lower temperature at the surface of the composting piles), lack of homogenization due to inadequate mixing, contact with leachates, and addition of raw materials or young compost to mature compost [116].

Table 3. Effects of time and temperature on pathogenic bacteria in different raw materials during the thermophilic stage of composting.

Substrate	Process Time (days)	Temperature (°C)	Bacteria Pathogens	Reference
Chicken manure + peanut hulls	57	55	<i>E. coli</i> <i>Salmonella</i> spp. <i>Listeria innocua</i>	[114]
Bovine manure	28	70	<i>E. coli</i> <i>Salmonella</i> spp.	[112]
Meat waste	98	67	<i>E. coli</i> <i>Salmonella</i> spp.	[117]
Food waste	70	60	<i>E. coli</i> <i>Salmonella</i> spp.	[118]

3.8. Enrichment of the Composting Process with External Inoculums

Traditional composting is always time-consuming, so several strategies have been tested to improve the efficiency of the process. Among them, indirect and direct biological enhancers or improvers have been studied. Indirect improvers are responsible for regulating the physicochemical properties of the compost [55,119] related to the application of animal manure and additives such as sawdust, corn, rice straw, and rice husk, among others. The latter depends on the number of indigenous microorganisms in the compost. When the number of microorganisms is insufficient, many dysfunctions are generated, including low decomposition capacity, prolonged fermentation period, and reduced nutrient content in the compost products [120].

In this context, microbial inoculation has become an interesting research topic due to its ability to accelerate the degradation of agricultural residues, as well as its advantages, e.g., low application cost, absence of secondary contamination, and ease of operation [121]. Beneficial microorganisms can be isolated from soil microbial communities, then cultured and selected according to specific degradative functions, and used as inoculums. They can also be applied directly to substrate mixtures such as soil, cow manure, and straw, among others [106]. Compost stimulators can be composed of a single strain [122], a mixture of strains [123], or mature compost [124]. Table 4 summarizes some studies conducted on the use of direct regulators in the composting process.

Table 4. Studies on direct biological regulators and their effects on composting.

Substrate	Inoculum	Effect on the Compost Process	Reference
Peat-based substrate	Three different commercial	Improve nutrient status and flower development	[125]
Green compost	Arbuscular mycorrhizal fungi inocula	Improve plant growth on yield and quality of fruits in tomato	[126]
Sugarcane bagasse substrate (fresh, dry, and compost)	Arbuscular mycorrhizal fungal-rhizobacterial mixed inoculum <i>Funneliformis mosseae</i> (sensu <i>Glomus mosseae</i>)	Increase in plant growth	[127]
Food scraps and dry leaves	Mature and commercial compost	Lower C/N ratio, higher germination rate, higher compost fraction less than 12.5 mm, and higher volatile mass reduction.	[124]
Municipal green waste	Arbuscular mycorrhizal fungus	Increase plant growth and/or nutrition	[128]
Municipal solid waste	<i>Clostridia</i> sp.	Improved anaerobic digestion of cellulosic biomass	[129]
Food waste	Lactic acid bacteria, yeasts, and phototrophic bacteria.	Accelerate the decomposition process	[130]

Table 4. Cont.

Substrate	Inoculum	Effect on the Compost Process	Reference
Food waste	mesophilic yeast <i>Pichia kudriavzevii</i>	Promote the degradation of organic matter and accelerated the composting process.	[122]
Pig manure and corn straw	<i>Acinetobacter pittii</i> , <i>Bacillus subtilis</i> sub sp. <i>Stercoris</i> y <i>Bacillus altitudinis</i>	Prolonging thermophilic stage, increasing germination rate	[131]
Municipal solid waste, sawdust, brown matter, and grasses	<i>Bacillus subtilis</i> , <i>B. amyloliquefaciens</i> , <i>B. nakamurai</i> y <i>B. velezensis</i>	Shorter time to reach the thermophilic stage, lower C/N, higher cellulose hydrolysis	[123]
Citrus peels (CP)	Mixing of bacteria from high temperature CP compost samples and citrus orchard soil.	Increased pile temperature, accelerates cellulose and pectin degradation	[132]
Cow dung and wheat straw	Bacterial consortium	Inhibited gaseous emissions and enhanced carbon and nitrogen sequestration	[36]
Green and industrial waste	Arbuscular mycorrhizal fungus <i>Rhizoglyphus irregularis</i>	Improve plant growth and development	[133]
Mushroom and wood chip residues	<i>Aspergillus</i> , <i>Penicillium</i> , <i>Nocardia</i> , <i>Arthrobacter</i> , <i>Sphingobacterium</i> , <i>Streptomyces</i> , <i>Novosphingobium</i> , <i>Flavobacterium</i> , <i>Pseudomonas</i> , <i>Cladosporium</i> y <i>Bacillus</i> bacteria (<i>Bacillus subtilis</i> , <i>Serratia marcescens</i>), yeasts (<i>Rhodotorula mucillaginosa</i> , <i>Candida butyrii</i>), actinobacteria	Heat the compost pile and thus initiate the composting process at low temperatures. Prolong the thermophilic stage and promote the decomposition of cellulosic biomass.	[134]
Olive wet husks	(<i>Streptomyces drozdowiczii</i> , <i>S. griseofulvus</i> , <i>S. rochei</i>), and microfungi (<i>Phanerochaete chrysosporium</i> , <i>Chaetomium globosum</i> , <i>Trichoderma atroviride</i>)	Faster start and shorter maturation time of the process of composting. Deeper humification rate. Better detoxification of the starting material.	[109]

4. Promotion of Soil Microorganisms by Compost Application

4.1. Fungal Community

Soil fungi play a fundamental role in agricultural ecosystems, with multiple ecosystemic functions and several taxonomic groups. Although there are few recent studies on the impact of compost on the fungal community, it is important to highlight the important role they play in soil fertility, with both symbiont and saprophyte groups being involved in soil organic matter and nutrient cycling [135,136].

It has been observed that compost can increase the diversity of fungi present in soil finding up to 66 operational taxonomic units (OTUs) [137,138]. On the other hand, more recent studies have shown that the diversity and richness of the fungal community can be reduced when using high doses of compost [135]. For this reason, further studies are needed to elucidate the effect of composts on the soil fungal community using various doses of composts additionally, different agronomic management of amendment, and different raw materials.

It has been observed that the application of compost modifies the functional structure of soil fungal communities by increasing the groups of saprophytic fungi, which are abundantly found in the composting process. It has also been observed that pathogenic fungi decrease after compost application probably due to the pathogen-inhibitory properties of compost and the modification of soil pH [70,135].

4.2. Arbuscular Mycorrhizal Fungi (AMF)

Arbuscular mycorrhizae are the result of the symbiosis between *Glomeromycota* fungi and most plants, including major crops [139]. These fungi provide multiple benefits to

associated plants, which mainly include greater access to nutrients (nitrogen, phosphorus, zinc, and magnesium) and water in the soil, as well as defense against soil pathogens, due to the stimulation of the secondary metabolism of plants, for which has multiple ecosystem functions, in return plants can allocate a 4–20% of assimilable C to the fungi [140].

The formation and establishment of arbuscular mycorrhizae in crops depends on agricultural practices that impact the soil, such as the use of agrochemicals, tillage, and type of crop [3]. In general, intensive practices harm the establishment of symbiosis, and therefore its ecosystem functionalities for crops [141]. On the other hand, sustainable, conservative, regenerative, and organic agriculture encourages the formation of this symbiosis, because they promote the biodiversity of soil microorganisms [142].

Composting represents a widely used agricultural practice and its relationship with arbuscular mycorrhizae has been studied with different raw materials and with different plants [128]. The compost, according to the composition of its raw materials, contains the spores of various species of arbuscular mycorrhizal fungi, this is because spores are found in the guanos, as well as in the development sites of the composting piles. These spores can resist the temperatures of the composting process and later germinate when in contact with the roots of the crops [143].

It has been observed in different experiments that composting promotes plant mycorrhization, and increases the formation of spores and extra-radical mycelium, probably due to the delivery of available organic matter up to 50%. This favors the establishment of mycorrhization and stimulates the formation of symbiosis so that the plant can access the microsites where the nutrients delivered by compost are found [127,144–146].

In most of the studies where mycorrhizae and compost have been evaluated, it has been observed that these two practices are not incompatible, but rather act synergistically, benefiting them by delivering a greater amount of nutrients (80–89% of N and P) to the plants and therefore increasing the yields of the crops [128,147], but the doses of compost to be used should be considered, since it has also been observed that an excess at the amount of compost can inhibit mycorrhization; in fact, in high concentrations of nutrients such as nitrogen and phosphorus, the symbiosis does not develop properly [126,133,148].

The different species of arbuscular mycorrhizal fungi have different effects on the crops, due to the multifunctionality of the mycorrhizae, for this reason, the starting materials of the compost and crops must be carefully selected to propose certain species [128,146].

4.3. Bacterial Community in Agricultural Soil

Agricultural soil bacterial communities are essential for maintaining soil health and plant productivity. Soil bacteria perform multiple metabolic activities that allow, among others, the degradation of organic compounds, mineralization of nutrients, nitrification, and dissimilatory reduction of nitrate to ammonium and transform organic residues into plant nutrients such as amino acids, ammonium, phosphate, and potassium, among others [149]. Studies suggest that an optimal ecology of agricultural techniques as manure application, organic farming, biochar modification, tillage management and improvement of the soil carbon-nitrogen ratio promotes the bacteria and activity, increasing the efficiency of nutrient use by plants [150].

The diversity of bacteria in agricultural soils is diverse and depends largely on soil nutrition as shown for Proteobacteria, *Bacteroidetes*, *Actinobacteria*, and *Firmicutes* which are fast-growing copiotrophs when nutrients are abundant, while *Acidobacteria* is a slow-growing oligotrophic bacterium but adaptable to low nutrients [149].

Organic amendment, biostimulation, and biodegradation using nutrient-rich organic substrates are recommended techniques to stimulate the establishment and activity of bacteria that degrade organic matter and release inorganic nutrients to plants through mineralization, which in turn improves the quality of agricultural soils. In general, it is suggested that healthy bacterial communities in soils could be maintained by adding organic matter at near-neutral pH levels [149]. It has been shown that the soil after compost application is dominated by bacteria. Compost treatment has been reported to impact the

composition and activity of the soil microbial community, especially increasing the presence of Gram-positive bacteria and rhizobacterial populations [151]. Compost fertilizes the soil with nutrients that enhance the growth of organotrophic bacteria, oligotrophic bacteria, and actinobacteria, finding up to 383 operational taxonomic units (OTUs) [152]. Of the benefits of bacterial communities in agricultural soils has been proposed, the application of bacterial fertiliser to provide a variety of nitrogen-fixing bacteria and phosphorus-solubilizing bacteria that could improve the soil microbial community structure [153].

4.4. Plant Growth Promoting Bacteria (PGPR)

Plant growth-promoting rhizobacteria (PGPR) are an important group of microbial soil communities that enhance plant establishment and growth [45]. They could fix nitrogen (N), dissolve phosphorus, chelate iron, inhibit root ethylene production pressure, and produce plant growth hormones, antibiotics, and antifungal compounds [154]. Furthermore, PGPRs enhance plant development and tolerance to different types of stresses by inducing the activity of several antioxidant enzymes, such as catalase (CAT), peroxidases (POD), and polyphenoloxidases (PPO), among others [155,156].

Compost treatment increases the presence of PGPR bacteria [151], stimulating the enzymatic activity of PGPR. It has also been reported that integrated composting with rhizobacteria inoculation benefits soil fertility, especially in the environment where plants grow [157].

Another study has shown that the co-application of PGPR and compost alleviate plant toxicity to heavy metals such as lead, possibly due to the enhancement of physiological machinery and antioxidant homeostasis that both components provide to plants. It is suggested that co-application is feasible for the management of contaminated soils [158]. However, further in situ studies are needed to confirm the long-term efficacy of the co-application of compost and PGPR [159].

4.5. Other Microorganisms

Microalgae are microscopic algae, found as part of phytoplankton and in nearly all aquatic, terrestrial, and sub-aerial surfaces, including all types of soil. They provide several benefits; they participate in nutrient cycling promoting plant growth by improving nutrient availability. They have also been shown to produce bioactive substances such as phytohormones, form root associations, and protect plants from phytopathogens and pests. They also fix carbon dioxide (CO₂) and some produce exopolysaccharides (EPS) that improve soil structure. Cyanobacteria are considered biofertilizers because of their known ability to fix atmospheric nitrogen (N₂) in the soil and solubilize immobilized phosphorus (P) [160].

Cyanobacteria are the most diverse and studied microalgae in the rhizosphere since they can form symbiotic associations with plants. The establishment and growth of cyanobacteria are favored by low light conditions and high availability of P, neutral to slightly alkaline pH values, and temperatures of 30–35 °C [161]. However, their density can be seen as affected by alternating dry and wet periods and invertebrate grazing. Agricultural practices can also affect the growth of indigenous strains in situ, such as the use of chemical fertilizers with N and the increase in ammonium sulfate rates due to chemical fertilizers [160,162]. There are many studies to identify the microalgae in rice fields because of the wet condition, the most important genera of cyanobacteria found are *Anabaena*, *Nostoc*, *Gloeotrichia*, *Microcystis*, *Chroococcus*, *Oscillatoria*, *Lyngbya*, and *Phormidium* [160].

The growth of microalgae biomass depends on the supplies of CO₂, light, and nutrients mainly N, P, and potassium (K), with low P and N requirements of the order of 0.2 µmol/L. They also require other trace metals such as zinc (Zn) and magnesium (Mg). These requirements can be met with livestock manure and sewage [163]. Therefore, it is believed that the integration of microalgae cultivation with compost is a potential technique for the development of microalgae and its potential use as a biofertilizer.

5. Discussion

Composting is an important technique in sustainable agriculture for the transformation of organic matter in fertiliser or organics amendments favoring soil fertility and pathogen suppression, while promoting microbial biodiversity in the soil. This will impact both the quantity and quality of crops and foods. It is currently widely used in organic and conventional agriculture throughout the world (Figure 3), which indicates how the composting process is regulated by the succession of microorganisms and their environmental conditions while promoting sustainable agriculture [164]. There are limitations to the study of composting as it is developed at different latitudes. On the one hand, we find different techniques to produce it and different starting materials. As the methodology is not standardized, we can find a wide variety of factors that affect the correct development of composting. Therefore, it is necessary to standardize the methodology and locally monitor the microorganisms found in the compost. It is also necessary to promote studies in poorly represented areas, to determine the microbial diversity contained in these composts and the impact on local communities of soil microorganisms. Furthermore, it is also important to assess whether local microorganisms may have a pathogenic effect on crops or people [65].

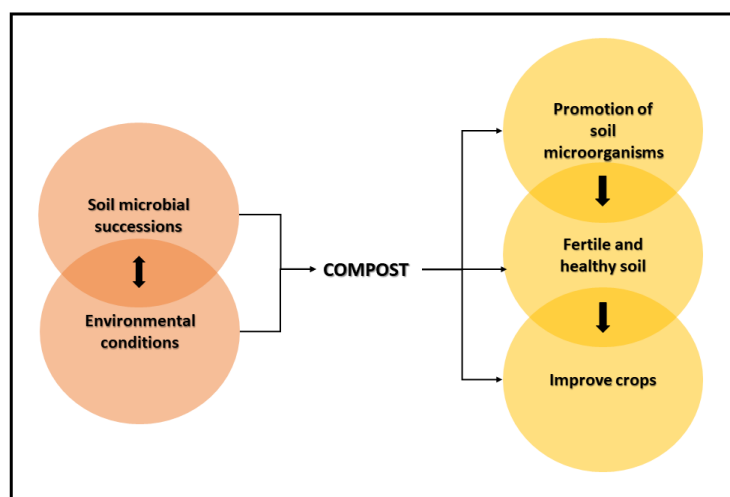


Figure 3. The effect of compost and its components promotes sustainable agriculture.

The composting process is determined by a set of successions of bacteria and fungi from diverse taxonomic groups. This wide microbial diversity gives it the properties that benefit soil fertility and health. Due to the environmental conditions, the different composting processes that exist, the different raw materials, and the initial microbial composition, it is difficult to generalize the microbial composition of the composting process [165].

Studies in general show that the most relevant microorganisms in the composting process are bacteria and archaea with respect to the number of propagule or abundance, but fungi also play a fundamental role in the degradation of more complex materials that are low humidity, acidic, or low in nitrogen for bacterial decomposition, although many of the fungal populations in the composting process are not yet well characterized taxonomically [68].

Bacteria, being microorganisms with faster life cycles, can develop and rapidly absorb energy from raw materials and new substrates generated in the initial stages of composting. Undoubtedly, there are other microorganisms participating in the composting process that have been scarcely considered in studies such as viruses and microalgae [69].

The study of microorganisms in composting has been developed for decades by traditional culture methods, but it should be considered that most microorganisms are difficult to cultivate. In that sense, molecular techniques and massive sequencing complement help to characterise the complete communities of compost microorganisms [103].

The environmental conditions in which compost is developed will be fundamental for the success of the process. It is necessary to standardize and investigate these conditions to achieve the greatest local microbial development in composting, mainly considering the new methodologies and technologies used to produce the compost, the time length of the process, and the different composting phases [103].

In the case of an aerobic composting process, the main determinant of the microbial community will be temperature, although pH and humidity will also determine the success of different populations of microorganisms, especially fungi. The raw material used to develop the compost will be fundamental to determine the biodiversity that we can find in the composting process, even if much of it is not expressed due to the increase in temperature [33].

Studies in general show that the most relevant microorganisms in the soil that impact the composting process are the bacterial community, although the fungal community produced in the composting process is also modified and persists in soils after compost application [166].

The active proliferation of fungal and bacterial cells inside and around the particles of the compost is a crucial moment for eliciting the beneficial effects of compost as a microbiome source when delivered to soil or around crop plants. From each well-colonized compost particle, the microbiota relevant as an ecosystem service provider will proliferate and colonize the neighboring micro- and macro-aggregates, becoming an established soil or rhizospheric population. While the effect of arbuscular mycorrhizal fungal on bacterial transport and organic phosphorus mineralization of organic phosphorus or the colonization of legume roots by symbiotic dinitrogen-fixing bacteria is known [167], the effects of compost-derived fungi on the spread of other microorganisms remains poorly studied. The same applies to the factors affecting the migration of microbial cells from compost particles to soil aggregates.

Knowing the microbial diversity of the composting process and its impact on soil communities it is relevant to standardize and verify the reproducibility, performance, and quality of the final product. Evaluating the microbiology in the finished compost will also be important to determine the existence of possible pathogens or microorganisms harmful to human health [65].

Extensive research supporting the application of external inoculants in agriculture has promoted the use of consortia of microorganisms to improve the composting process [83,86,130,132,134], as well as, to modify and improve soil microbial diversity and soil fertility [4,168]. A clear example is the same use of microbial consortia present in the composting process, previously studied in this review.

The benefits of compost microbial consortia have also been studied for their application in other similar processes. One example is from Kinet et al. [129], who isolated and studied a microbial consortium isolated from compost to enhance the initial hydrolysis step in anaerobic digestion. The exogenous consortium was able to adapt and maintain the cellulolytic degradation potential during the initial step [129]. Further examples are the discovery of the positive effect that compost generates on the biodiversity of arbuscular mycorrhizal fungi, which in turn are considered bioindicators of soil fertility [169], and the use of microbial starters which enhanced transformation and increased earlier bacterial diversity in composting of olive mill solid waste [105].

Despite the above and from an ecological point of view, it would be important to promote the development and application of native inocula with higher taxonomic diversity than the use of commercial inocula, which have low biodiversity and whose species are not locally adapted [170]. The contraindications for the application of little-studied commercial inocula could be, among others, the non-adaptation of the microorganisms to the environmental conditions of the soil, or the overexpression of a species that could prevent the establishment of other microorganisms, thus reducing biodiversity.

To study the effects of the application of external inocula on soil biodiversity, it is possible to use new molecular tools, such as pyrosequencing and massive sequencing.

These techniques have been used to provide rapid and efficient knowledge of the microbial successions present during composting [23,171]; they have also allowed us to understand the effect of compost on the microbial diversity of agricultural soils and its relationship with fertility [23,169,172].

While the benefits of composting and the microorganisms involved in the composting process are widely recognized, more research is needed in this area. It is important to know or predict the composition and nutritional concentration of the compost according to the starting material and extend the use of this practice in agriculture. Equally relevant is to quantify the effect that this amendment has on the microbial consortia that inhabit agricultural soils [128].

Likewise, it is important to investigate the processes of translocation of nutrients from organic amendments to plants and the environment where arbuscular mycorrhizae and bacterial biota play an important role in the transport and acquisition of minerals and organic matter [173,174].

The use of compost in agriculture provides a positive impact on the mitigation of climate change and the reduction of greenhouse gases. This is because it generates a reduction in the use of pesticides and (partial or total) replacement of chemical fertilisers, and the improved structure of soils reduces the requirement for irrigation water, improved tilt, and workability. All these benefits are associated with lower consumption of fossil fuels and a reduction in greenhouse gases [175].

The knowledge will allow the safe development and implementation of the use of mycorrhizae and PGPR together with composting and greatly promote the growth, yield, physiology, and nutritional quality of crops generating greater growth of the crops and better yields (Figure 4).

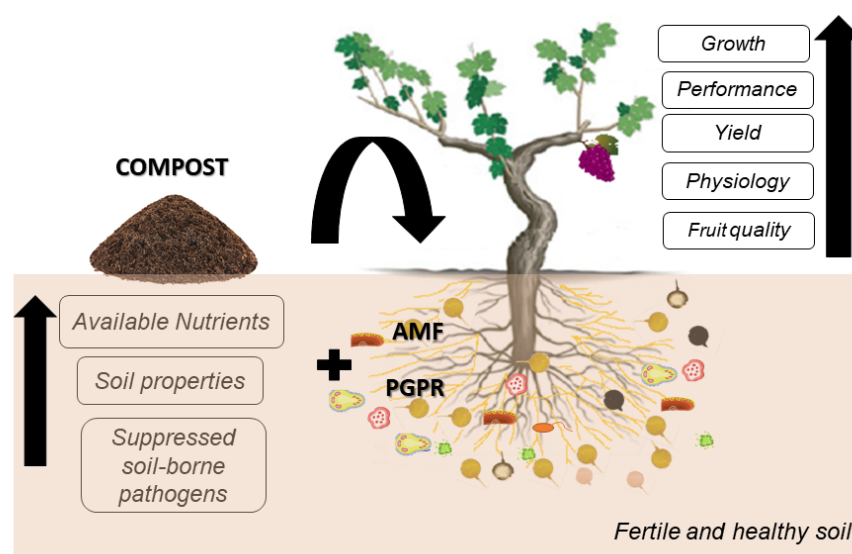


Figure 4. The effect of compost and soil microorganisms on crops for the development of fertile and healthy soils.

6. Conclusions

In conclusion, soil microorganisms play a very important role in composting and in obtaining abundant and healthy agricultural products. Their benefits allow for maintaining the balance and ecosystem services of the soil, and therefore its care and proliferation are important. The information in this review allows us to identify and learn about the main soil microorganisms that participate in and benefit from one of the most important practices of sustainable agriculture, composting. Knowledge of the biological processes that occur in composting will allow us to know the effectiveness of compost on crops and promote its use in sustainable agriculture. Delving into research on the application of the amendment

in different crops and the study of soil microorganisms and their associations with the plant environment is still an important research objective.

Author Contributions: Conceptualization, supervision, investigation, and formal analysis A.A.-P. and G.V.; resources, project administration and funding acquisition A.A.-P. All authors participated in writing the original draft, revising, and editing. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by CONICYT (ANID) I + D Ciencia y territorio 2019. NO. R19F10005 and VII Concurso de Fortalecimiento y Desarrollo de Centros Regionales de Desarrollo Científico y Tecnológico—2022, NO. R22A0002.

Data Availability Statement: Not applicable.

Acknowledgments: The authors acknowledge the agricultural systems Granja Crisol, Aroma SPA, and Casablanca Organics.

Conflicts of Interest: The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

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