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Organic Agriculture

Edited by Shaon Kumar Das



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Published in London, United Kingdom



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<http://dx.doi.org/10.5772/intechopen.89775>

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First published in London, United Kingdom, 2020 by IntechOpen

IntechOpen is the global imprint of INTECHOPEN LIMITED, registered in England and Wales, registration number: 11086078, 5 Princes Gate Court, London, SW7 2QJ, United Kingdom
Printed in Croatia

British Library Cataloguing-in-Publication Data

A catalogue record for this book is available from the British Library

Additional hard and PDF copies can be obtained from orders@intechopen.com

Organic Agriculture

Edited by Shaon Kumar Das

p. cm.

Print ISBN 978-1-78984-668-3

Online ISBN 978-1-78984-669-0

eBook (PDF) ISBN 978-1-78984-720-8

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Meet the editor



Shaon Kumar Das is working as a Scientist SS (Agricultural Chemistry) at ICAR RC for NEH Region, Sikkim Centre, Tadong, Gangtok under the Indian Council of Agricultural Research, New Delhi. He received his M.Sc. in 2010 from IARI, New Delhi along with a gold medal for his outstanding contribution. He joined the Agricultural Research Service (ARS) in 2011 as a scientist. He completed his Ph.D in 2020 from Visva Bharati University in Soil Science and Agricultural Chemistry and worked on integrated organic nutrient management. He has published 27 national and 40 international research articles, 10 review articles in national and international journals, 30 popular articles, 28 extension folders, 2 books, and 25 book chapters in edited books. He received a DST-INSPIRE fellowship in 2010 from DST, Govt. of India. He has been awarded the best scientist award by the ICAR RC for the NEH Region, Umiam, Meghalaya in 2018. He received six young scientist awards and 3 best oral presentation awards from different reputable societies. He was the Principal Investigator (PI) of different externally funded projects like DST, DBT, NMHS, ICAR etc. He has been involved in the research on carbon sequestration, soil fertility management, soil acidity reclamation, and organic nutrient standardization of major hill crops.

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Preface

Organic crop production is the science and art of growing field crops, fruits, vegetables, and flowers by adopting the essential principles of organic agriculture in soil building and conservation, pest management, and heirloom variety conservation. This book provides detailed insights into organic farming in agriculture, biological efficacy in the management of plant diseases, organic nutrient management, socio-economic dimensions of adoption of conservation practices, nonchemical weed control, plant growth promoting fungi for phytostimulation, nanotechnological approaches, and finally vermicomposting. The book primarily focuses on research and development based organic agriculture and horticulture production technologies, and has attempted to abridge information on organic crop production of the major food grain crops. The book also contains comprehensive information on the various related dimensions of organic crop production.

It is a matter of great privilege and honor for me to express my esteemed gratitude to all my senior colleagues who conceived, detailed, and shaped the problem. Their sagacious guidance, persistent involvement, scholarly suggestions, relentless encouragement, prudent admonitions, constant and ungrudging help, and affectionate behavior has been a fountain of great inspiration to me. I implore my feeling of profound gratitude to the researchers for their keen interest, cordial attitude, generous help, and valuable suggestions during the entire course of work. I owe a great deal to all the scientific community for their wise counsel and steadfast help during this investigation. I avail myself of this opportunity to convey my heartfelt thanks to contributors of the book chapters for providing necessary facilities and valuable help during the entire span of work. I express my heartfelt thankfulness to my brother, mother, father, wife, and son for their encouragement, cordial attitude, and help during the entire span of my study. Candid thanks are due to all my seniors, juniors, and friends for their helping nature, company, inspiration, and moral support, which made my passage smooth over the rough terrain. Finally, I express my special thanks to all the staff of the IntechOpen from whom I received ever-willing help and moral support.

Shaon Kumar Das
Indian Council of Agricultural Research (ICAR),
India

Section 1

Organic Farming

Role of Organic Farming in Agriculture

*Muthuraman Yuvaraj, Peyandi Paraman Mahendran
and Eman Tawfik Hussien*

Abstract

Organic farming could be an all-encompassing generation administration framework that empowers and improves agroecosystem wellbeing, counting biodiversity, natural cycles, and soil biological activity. It stresses the role of management activities in preference to the use of off-farm data, considering that regional conditions require locally adapted systems. This can be achieved using agronomic, biological, and mechanical methods, in equal share to synthetic materials, to carry out any specific role inside the organization. Organic farming is still only a small industry, which represents only 2% of global food sales. However, it is growing in importance in the world. It is hard to get information due to lack of official statistics and the level of confidentiality of systems of organic produce. Soil practices such as crop rotations, organic fertilizers, symbiotic associations, cover crops, inter-cropping, and minimum tillage are central to organic practices. The static arrangements of soil are achieved by soil fauna and vegetation. Besides, cycling of nutrients and energy is enhanced by increasing the retentive abilities of the soil for nutrients and water.

Keywords: organic farming, vermicompost, soil health

1. Introduction

Organic farming is defined as a production system that avoids or largely eliminates the usage of synthetic compounded fertilizers, growth regulators, pesticides, and farm animal feed additives. To maximize feasible extent, the organic farming system depends on crop rotation, green manures, legumes, animal manures, crop residues, off-farming natural squanders, and aspect of biological pest control to preserve soil fertility and productivity to sustain plant and thereby curbing pests, diseases, and unwanted weeds. Organic farming methods are internationally regulated and legally executed by many countries, based in great part on the standards set by the International Federation of Organic Agriculture Movements (IFOAM), an international umbrella organization for organics established in 1972 [1].

After the launch of green revolution in India, substantial growth in the output of food grains was achieved. This was achieved through the utilization of improved crop varieties and higher levels of inputs of plant foods and plant protection chemicals. Merely there has been a rise in production, which was accomplished by the monetary value of soil health. The organic farming and ecological agriculture are one of the alternative agriculture systems to overcome the problems of soil degradation and declining soil fertility [2].

Soil organic matter is an essential soil component. These are residuals of dead plants, animals, and microbial tissues. Examples of organic manures are farmyard manure, compost, and green manure. They are added to the soil to the stock of organic matter. These added organics undergo a series of microbial decompositions, and finally, humus is formed. Humus is a light bulky amorphous material of dark brown to black color composed of organic compounds. Tropical soils characterized by low organic and loam less than of clay soils. The low organic matter is primarily due to climate, especially for high temperature and cultural practices, while organic matter contents increase with rainfall. In tropical and subtropical areas, there is much organic matter created, and it decays very quickly. Any organic matter added to the grounds will be decomposed (over 90% in a year), and thus, it is a Herculean job to produce the organic matter; content ranges from less than 1 to 15% [3].

2. Guidelines on production of organic produce

2.1 Duration of conversion period

Firstly, the formation of an organic controlling system and construction of soil fertility needs a temporal time known as change period. This change period may not continuously be the identical period to advance soil fertility and re-establish the equilibrium of the environment in all cases. It is defined as the time in which all the farm, including the livestock, is converted into organics. This was confirmed by the norms laid in the National Standards for organic products. The entire farm unit should be converted to organic in a phased manner, and the grower should present an alternative plan to the certification body when applying for accreditation.

Second step isolation belts are maintained all around the organic unit. This would drastically bring down the net area under organic cultivation. In view of this, a community approach is suggested to a group of continuous farms.

Thirdly, for existing conventional plantations, a changeover point for organic culture required a minimum of 3 years.

Finally, for a newly planted area, the first yield itself can be considered as organic, as the organic produce has a pre-bearing period of 1–2 years.

2.2 Farm designing

Firstly, an organic farm has to be a self-sustaining unit. Also, farm designing plays a very important role in optimizing the utilization of resources within the farm topography of the land and varieties of crops to be cultivated. Aside from these, border trees, compost yard, bounds, storage home, cattle shed, and farm-house can be suitably incorporated.

Secondly, the topography of the location of the cattle shed, compost yard, etc., should be decided. All the structures are better if at comparatively higher elevation than the cropped areas as it prevents water logging inside the cattle shed, store house, and office.

Finally, border trees: on the boundary of the farm, multipurpose border like Neem, Karanj or any other local trees of importance are planted 10 m apart. These border trees provide useful as wind break, abundant biomass for green manuring and composting, and preparation of pest management aids like Neem seed kernel extract to control insect pests.

2.3 Cattle shed

The cattle shed should be near the compost yard. The site for the cattle shed is better if it is at higher elevation. The shed should provide a comfortable and hygienic habitat to keep the cattle healthy and free of diseases. The floor may slope toward the dung channel to provide satisfactory drainage and facilitate collection of urine. Managers can be 0.75 m wide with all corners rounded off in cement. The cattle shed should be preferably oriented in east to west direction to have proper public discussion and with neem or peepal trees around it [4].

2.4 Store house

A storehouse should be maintained to stock the farm implements and the produce after harvest. The storehouse can be at least half a meter above the farm level and should throw a projection of 0.5 m at that height to protect the produce from rats and other rodents. The storehouse should be damp proof. The windows and doors could be lined with fine wire mesh [5].

2.5 Border plants

The planting of leguminous trees is suggested to be grown as border crops for biomass. The trees mentioned earlier would also enrich the soil fertility and help in the improvement of the soil structure [6, 7].

3. Components of organic farming system

The significant research in an organic farming system has been difficult to design due to the nature of organic philosophy. There is no shortage of essential component of research, which could be done to increase both nitrogen fixation (in legumes on component crops) and on weed and insect control measures. This research would touch only marginally on the basic questions inherent in organic philosophy [8, 9].

Principles of organic agriculture

- Use renewable resource in locally organized production system
- Increase balance between crop production and animal husbandry
- Give all livestock condition of life with due consideration for the basic aspects of their intake behavior
- Decrease all forms of pollution
- Processing organic products with renewable resources
- Production of fully biodegradable organic products
- Progress toward an entire production processing and distribution chain, which is both socially and ecologically responsible

Benefits of organic farming

- Organic agriculture helps to prevent environmental degradation and avoid a chain reaction in the environment from chemical sprays and dusts
- Organically grown crops are believed to be healthier and nutritional food for man and animals
- Organic fertilizer is considered as complete plant food. Organic matter restores the pH of the soil, which may become acidic due to the continuous application of chemical fertilizer.
- Organic manure is the principal component of organic farming to produce optimal conditions in the soil for high yields and good quality.
- Most of the organic manures are wastes or byproducts, which on accumulation may contribute to contamination. In this method of organic farming pollution is reduced.
- Organic farming is labor intensive in the nation which will also help in generating more employment in rural areas that will help in reducing economic inputs.
- As a whole, adoption of organic farming provides a better and balanced environment and better products and living conditions to the human beings.

4. Different available organic inputs

4.1 Organic droppings (manure)

The farm-yard manure (FYM) and vermicomposting, etc., are typically little in nutrient contented. So, great use rates are difficult to meet crop nutrient supplies. However, in numerous emerging nations (like India), the obtainability of organic manures is not sufficient for crop demands partly due to its wide use of cattle dung in energy production. Green manuring with *Sesbania* plant, cowpea, and green gram is effective to improve the soil content of organic matter. However, employment of inexperienced manuring has declined in the previous couple of decades because of intensive cropping and socioeconomic reasons. Taking these constraints, International Federation of Organic Agriculture Movement (IFOAM) and Codex Alimentarius have approved the use of some inorganic sources of plant nutrients like rock phosphate, basic slag, rock potash, etc., in organic farming systems [10].

4.2 Bacterial and fungal biofertilizers

Nitrogen fixation on the surface of the earth is mainly by microorganisms, representing 67.3% among all the bases of N fixation. Subsequent microorganism and plant life biofertilizers will be utilized as an ingredient of organic farming in numerous crops.

4.2.1 *Rhizobium*

The efficiency of nitrogen-fixing microorganisms viz. *Rhizobia* for legume crops e.g. *Rhizobium*, *Bradyrhizobium*, *Azorhizobium*, *Mesorhizobium*, and *Sinorhizobium*.

Legumes are infected by these bacteria all over the world. These rhizobia have a N₂-fixing capability up to 450 metric weight units N ha⁻¹ counting on host-plant species and microorganism strains. Carrier based inoculants will primarily be coated with seeds for the introduction of microorganism strains into soil [11].

4.2.2 *Azotobacter*

Nitrogen will be fixed by independent microorganisms in cereal crops with no interdependency. Such free-living bacteria are: *Azotobacter sp.* for dissimilar cereal crops; *Herbaspirillum spp* and *Acetobacter diazotrophicus*.

4.2.3 *Azospirillum*

The gram-positive bacteria *Azospirillum* colonizes in a remarkably kind of yearly and perennial floras. Studies indicate that *Azospirillum* will proliferate the development of crops like flowers, cotton, oak, tomato, sugar beet, pepper, carrot, wheat, and rice. The crop yield can upsurge from 5 to 30%. Inoculum of *Azotobacter* and *Azospirillum* will be created and applied as in humate origination finished seed coating.

5. Plant growth-promoting rhizobacteria

Numerous microorganisms promote plant growth area. The unit is jointly known as plant growth-promoting rhizobacteria (PGPR). PGPR improves plant growth by colonizing the root system. Huge inhabitants of microorganisms recognized in implanting material and roots develop an incomplete sink for nutrients in the rhizosphere [12].

6. Phosphorus-solubilizing microorganism (PSB)

Phosphorus is also an important nutrient similar to nitrogen for plants. This part is important for the nodulation by bacteria genus and even to nitrogen fixers, *Azolla* and BGA. Phosphorus-solubilizing microorganism (PSB) fungi create on the market are insoluble phosphorus to the plants. It will increase crop weight up to 200–500 metric unit/ha and so 30–50 kg Super Phosphate will be preserved. Most predominant phosphorus-solubilizing microorganism (PSB) belongs to the genera *Bacilli* genus.

6.1 Mycorrhizal fungi

Mycorrhizal fungi which cause root-colonizing increase tolerance to many severe metal contamination and drought. Mycorrhizal fungi improve soil quality additionally by having an on-the-spot influence on soil aggregation and also aeration and water dynamics. An interesting potential of these fungi is their ability to permit plant access to nutrient sources [13–15].

6.2 Blue Chlorophyta (BGA)

The BGA represents the most important, most numerous and cosmopolitan cluster of microscopic organisms that perform an oxygenic chemical process. These are as well-known as Cyanophyceae and cyanobacteria [15–18].

6.3 Azolla

A floating fern 'Azolla' hosts element fixing BGA *Anabaena azollae*. *Azolla* contains 3.4% nitrogen (on dry weight basis) and adds organic matter in soil. This biofertilizer is used for rice cultivation. There are six species of *Azolla* viz. *A. pinnata*, *A. microphylla*, *A. mexicana*, *A. filiculoides*, *A. nilotica*, and *A. caroliniana* [19–20].

7. Soil management in organic farming

It is essential to take care to build up comparison of average yields, standard deviations, and coefficients of variation over five successive years. This includes crops and soil fertility, based on three inextricably interrelated components of soil management. These components are physical (water-holding capacity, structure, etc.), chemical (nutrient dynamics, pH), and biological (soil biota). The soil fertility of organic farming, can be defined as: well-managed soil organic matter in the soil, good soil structure, diverse soil biota and a high nutrient and water-holding capacity by using compost and stable manure. The organic soil is one that can build up over time buffering capacity and resistance to an imbalance in growing conditions as part of the strategy to enhance the self-regulatory capacity of the farm ecosystem [21, 22].

8. Weed management in organic farming

Weeds are often cited as the most significant problem in organic farming systems, and they are certainly the problem that most concerns the farmers, who are looking at changing over their farm from a standard one into an associate-in-nursing organic one.

9. Botanical pesticide use in organic farming

9.1 Nicotine

Nicotine is obtained from tobacco or related *Nicotiana* species and is one of the oldest botanical insecticides in use today. It is also one of the most toxic to warm-blooded animals, and it is readily absorbed through the skin.

9.2 Sabadilla

Sabadilla is another botanical insecticide. It is extracted from the seeds of the sabadilla lilly. The veratrine alkaloid is the active ingredient. Sabadilla is a botanical insecticide with low toxicity. However, its dust can be extremely irritating to the eyes and can produce sneezing if inhaled.

9.3 Neem

Neem is a botanical pesticide that comes from the neem tree, which is native of India. This tree supplies at least two compounds, salannin and azadirachtin, that have insecticidal activity and other unknown compounds with fungicidal activity. Neem pesticide controls gypsy moths, western flower thrips, sweet potato white flies, leaf miners, loopers, caterpillars, and mealybugs.

10. Conclusions

The contemporary form of natural farming is being popular in the world in a timely fashion, in particular in developed countries. Organic farming device is a choice, and an appropriate management system would help to enhance the soil health environment and hence expand the productive ranges and the enhance quality of crops. The natural farming system utilizes extremely multifaceted and combined residing classifications to accomplish their cease of maintainable harvest and inventory output. Organic agriculture is a potential choice due to the fact that it enlivens the soil, strengthens the natural resource base, and sustains organic production at degrees to commensurate the carrying potential of the managed agro eco-system. In addition to this, export market can also be tapped by group initiatives in organic farming. In a country like India, food production has to grow steadily.

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Nanotechnological Approaches in Sustainable Agriculture and Plant Disease Management

Siddhartha Das and Sudeepta Pattanayak

Abstract

Every year approximately 30–50% of crops suffer with different kinds of biotic stresses. Rapidly growing agrochemical industries and their diverse products make the environment more toxic and simultaneously hazardous for plant health and soil health. Such types of agrochemicals are toxic, hazardous, carcinogenic, non-eco-friendly. Therefore, this is the ideal time to think about some more effective alternatives against those problems. Nanotechnological approaches bring the alternatives in the form of decreasing toxicity, improving shelf-life, increasing solubility for poorly water-soluble agrochemicals, minimum use with maximum effect, slow leaching efficiency with long-term effect with coupling of eco-friendly naturalistic way. The way of nanoparticle application in agriculture, specifically disease management, is unique, where it can be used singly or by coupling with fungicidal, herbicidal, insecticidal, RNA-interference molecules. Though it has such a positive impact, very few products will be commercially available in our market due to high price of particular products and well-established long field trial efficacy detection among insect, pest-pathogen, and environment. Application of nanomolecules in other progressive fields has been emerging, whereas advancement in agricultural applications needs to be boosted up through skilled knowledge transfer and basic understanding of its fundamental aspect.

Keywords: nanotechnology, IDM, sustainable, eco-friendly

1. Introduction

The population of the world is increasing in an alarming rate. The global demand for food production will need to double by 2050. But, the climate change such as prolonged drought, sudden increase of temperature, unpredictable rains, and floods are the main barrier to achieve this global food demand [1]. Moreover, significant crop losses, that is, 20–40% per year due to some biotic causes is another major problem in worldwide basis [2]. The biotic causes mostly include insects, nematodes, pathogens, weeds, human, and animal interaction. Earlier studies revealed that approximately 20 and 26% yield loss occurred due to pathogen and insect infestation [3]. Most developed and developing countries depend upon chemical pesticides to control the diseases and pest incidence as these are easily available and show the result quickly. Industrial business and marketing policy also sometimes cross their barrier to gain some profit. Therefore human made behavioristic alteration of our environment also considered as a crucial responsible factor.

But they do not think those chemicals are harmful to the ecosystem, poisonous to beneficial insects, create pest resurgence, 90% loss of chemical during or post application in field leading to more economical loss for farmers. By keeping all the aforesaid reality based problem in mind, a new potential concept and technique known as nanotechnology is developed by the scientists, which is cost-effective, reliable, eco-friendly, and very effective. Although, this concept has not gained more focus in agriculture compared to pharmacology and medicine, but still it takes part a major role in plant breeding, nano sensors, plant hormone delivery, limited application of chemicals, plant health management etc. [4–6].

Nanotechnology refers to the technology related to application and manipulation of nanoparticles, that is, very small particles or materials having one or more dimensions, that is, 1–100 nm and fashioned with exclusive physical, chemical, and biological properties [7]. Several scientists have worked on the desired characteristics of nanoparticles such as pore shape, size etc. for accurate and specific application through adsorption or encapsulation of effective pesticide [8]. Application of nanoparticles can be functionalized in two ways, that is, first, nanoparticles directly involves as a plant protectant and second, nanoparticles used as carrier. On the first way researchers have tested the viability of the nanomolecules and mode of action against a particular pathogen. On the second way nanoparticles used with the existing fungicides or pesticide, herbicides, RNAi-mediated coupled component, to boost its activity. Nanoparticles used as a carrier have some positive sites like it can increase solubility of the coupled component, increase shelf-life, boosting site- and target-specific activity, reduce toxicity level, and maintain environmental safety. The implementation and utilization of fruits of nanotechnological applications, in terms of plant disease detection and management, gene editing and transformation is at infancy stage till now due to insufficiency of knowledge and skills. In the present context, the use of nanotechnology in plant disease management and allied sectors of agriculture is a real challenging task, which is objectified and framed with the present chapter.

Context of sustainable agriculture and nanotechnology

- 1. Application of nanotechnology to increase crop productivity and quality:** A variety of nanoparticles were tested previously to study their efficacy for boosting the yield as well as quality of the crop. It was observed that the nanoparticles modify enzymatic action, electron transport system, and influence the nutrition uptake. Carbon nanoparticles are reported to increase the yield of bitter melon by enhancing the biomass, water content, fruit weight, length, and number. Lentil seeds show high germination rate and growth when treated with silica-based nanoparticles. Titanium dioxide nanoparticles can boost the water uptake, breakdown of organic compounds, photosynthesis capacity, etc. [9].
- 2. Increase of photosynthetic efficiency through nanotechnological application:** Seeds incorporated with nanoparticles can promote the photosynthetic activity in plants. The nanoparticles respond to a specific wavelength of light and increase the optical potential of leaves which in result effect the hills reaction. Titanium dioxide-based nanoparticles can enhance the water and nutrient uptake, light absorbance and activity of Rubisco activase enzyme resulting more photosynthetic activity in plants. These nanoparticles induce the plants to be photosynthetically more active, grow faster, resulting in quality food production in less period of time [9, 10].
- 3. Improvement in water retention and management:** Nanotechniques can be imposed in agriculture to reduce the loss through evaporation, irrigation, and

to stabilize soil horizons in addition to lessening the ecotoxicity. Nanoparticles or nanotubes are found to be effective in retaining the water inside the hollow core for longer period by modifying the xylem vessel mechanism and metabolism of plant cells. Due to the partial solubility and dispersible characteristics, the water is retained inside plant cells for more time. In 2009, Corredor et al. demonstrated carbon nanotube treatment in pumpkin plant to analyze the water retention capacity and observed the accumulated carbon nanotubes inside the plant cell, which act as a water transporter. Due to the cohesive force, the water moves continuously through the nanotubes in plant cell forming a nanosized water stream [11].

4. Significance in the field of plant disease management: Recent days, nanotechnology is taking part a major role in plant disease management due to its eco-friendly nature and potentiality. Among all nanobased particles, silver nanoparticles stand out in the frontline in plant defense. These nanoparticles disturb the cell DNA, metabolic activity, electron transport chain, nutrient uptake of micro-organisms leading to death. The pathogenic fungi which can be controlled by using silver nanoparticles are *Colletotrichum gloeosporioides*, *Alternaria solani*, *Fusarium oxysporum*, *F. solani*, *Macrophomina phaseolina*, *Rhizoctonia solani*, *Aspergillus niger* etc. Silica-silver based nanoparticles are reported to inhibit the growth of bacteria *Pseudomonas syringae*, *Xanthomonas campestris* pv. *vesicatoria* up to 100%. Copper-based nanoparticles were found to be effective against bacteria *Xanthomonas oryzae*pv. *Oryzae*, *Xanthomonas campestris* pv. *phaseoli* and fungi *Fusarium solani*, *Alternaria solani*, *Aspergillus flavus* etc. [12, 13].

A. Limitations of different applied disease management strategies:

Drawbacks or limitation of all applied disease management practices are depicted under **Table 1**.

B. Types of nanoparticles for plant disease management: Nanoparticles can be used to manage the plant disease through two different mechanisms: as protectants where the nanoparticles only protect the plants and as carrier where the nanoparticles contain potential pesticides or some other active compounds [2]. Schematic representations of mode of action of these two modes are shown under **Figure 1**.

I. Nanoparticles used as protectants: Potential nanoparticles used alone to protect the plants from pathogenic micro-organisms and applied directly to plant and plant parts. Several metal nanoparticles like copper, silver, zinc oxide, titanium dioxide is experimented for their antagonistic effects against all pathogenic bacteria, virus, fungus, and concluded as successful potential protectants [14–16]. These nanoparticles have more shelf life, easily soluble in water, and show site specific uptake as compared to conventional chemicals.

a. Silver nanoparticle: It is in the frontline due to its “green synthesis” production mechanism in bacteria, yeast, fungi, and plants [17]. From previous studies, it is reported that silver nanoparticles have antifungal effect against *Alternaria alternata*, *Sclerotinia sclerotiorum*, *Macrophomina phaseolina*, *Botrytis cinerea*, *Rhizoctonia solani*, etc. by well diffusion assay [18] while antiviral effect on sun hemp rosette virus when sprayed on leaves and bean yellow mosaic virus when applied after infection [19].

Techniques	Physical Method	Chemical method	Biological method
Strategies applied	Soil solarization	Fungicide, pesticide or related agrochemicals	Resistant varieties, Use of biocontrol agents
Limitations	<ul style="list-style-type: none"> • Longer duration • Selective towards thermophilic and thermotolerant pathogens • Climate dependent • Availability of crop-free fields for long period • Survival of pathogen in deeper soil zone • Pollution from plastic residue after treatment 	<ul style="list-style-type: none"> • Resistance to fungicide or pesticide • Harmful effect on non-target organisms • Non ecofriendly • Carcinogenic • Non-economic 	<ul style="list-style-type: none"> • Wide host range of pathogen • Survival of pathogen for long period • Costly • Lack of proper awareness and training

Table 1.
Limitations of different disease management practices.

- b. **Nano carbon:** The carbon nanoparticles not only show antimicrobial activity in plant health management but also positively affect the plant growth. These nanoparticles from graphene oxide sheets have shown antimicrobial activity against *Aspergillus niger*, *A. oryzae*, and *Fusarium oxysporum* *in vitro* [20]. Many researchers have anticipated about the mechanism behind the inhibition of microbial growth by carbon nanomaterials and explained that the nano edges of graphene oxide come in direct contact with the chemical compounds present in cell wall of fungi and bacteria making them inactive [21–23].
- c. **Titanium dioxide nanoparticle:** When this nanoparticle is used as fertilizer was known to show antibacterial, antiviral and insecticidal characteristics.
- d. **Poly dispersed gold nanoparticles:** It was reported to inactivate the Barley yellow mosaic virus and develop plant resistance [24, 25].
- e. **Chitosan nanoparticles** control alfalfa mosaic virus, *Fusarium* spp., *Botrytis* spp., *Pyriularia grisea* while show limited effect against bacteria [26]. The mechanisms behind the antimicrobial properties of chitosan are inhibition of the growth of pathogen, protein synthesis, and ATPase activity, agglutination, interruption of cell membrane, disruption of nutrient flow etc. Chitosan nanoparticles have also been reported to control aphids, root knot nematode, and cotton leaf worm.

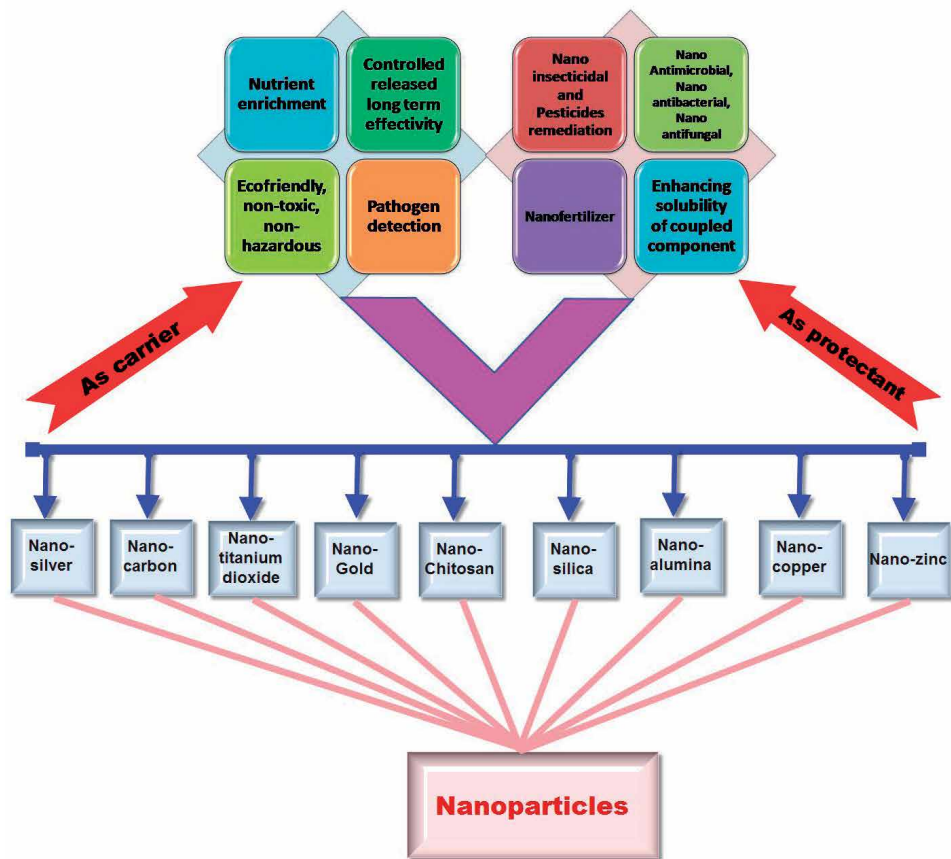


Figure 1.
 Schematic representation of different nanoparticles and their mode of action in term of carrier and protectant.

II. Nanoparticles that act as carriers: Nanoparticles are engineered in such a way that these can carry, encapsulate, or absorb active potential compounds to develop effective agricultural pesticides. These carrier nanoparticle-based pesticides have less toxicity, slow leaching capacity, and highly target specific. The below-mentioned nanoparticles are mainly used as carrier:

a. **Silica nanoparticles:** These nanoparticles have specific size, shape, and structure making them able to deliver the active compounds in the target site [27]. They are mainly round or circular like structure having a pore like holes, for example, mesoporous silica nanoparticles (MSNs) or porous hollow silica nanoparticles (PHSNs). The active compounds are mainly loaded in the inner hole to keep them protect and proper release in target sites. The outer shell guards the active compounds against degradation by UV light.

b. **Nano alumino-silicate:** The major advantages for the use of nano alumino-silicate are that it is biologically active, eco-friendly, and more effective than other nanoparticles. The alumino-silicate nanotube containing active compounds when sprayed on plants, the insects present on plants intake the active compounds contained nanotubes [28].

- c. **Chitosan nanoparticles:** Due to the low solubility in aqueous media, chitosan nanoparticles are applied by mixing with some organic or inorganic or copolymer compounds to increase its solubility [29]. These compounds stick to the stem and leaf epidermis facilitating them for prolonged effect in target sites [27]. Presence of amines and hydroxyl groups, enable the nanoparticles to enhance its properties [30].
- d. **Nanocopper:** Copper is one of the most effective and potential chemicals in combating wide range of plant diseases. For the first time in 2013, Giannousi et al. studied the potential antimicrobial characteristics of nanocopper particles and concluded that the nanocopper particles are more promising than other copper-based chemicals [31]. From past studies, it was reported to control or minimizes the growth of the pathogen of Fusarium wilt of tomato, *Verticillium* wilt of eggplant, Leaf spot, leaf blight diseases etc. [32, 33]. Biosynthesized nanocopper derived from *Streptomyces griseus* was observed to limit the growth of *Poria hypolateritia* causing root rot disease of tea [34].
- e. **Nanozinc:** The characteristic antimicrobial properties of nanozinc particles are mostly studied in vitro condition. It was reported to control bacteria, fungi, for example, *Alternaria alternata*, *Botrytis cinerea*, *Sclerotinia sclerotiorum*, *Rhizopus stolonifera*, *Rhizoctonia solani*, *Mucor* spp., *Fusarium oxysporum*, and *Penicillium* spp. [35, 36]. Disease suppression in protected cultivation and green house condition through nanozinc is also possible.
- f. **Solid lipid nanoparticles (SLNs):** These nanoparticles contain lipids, which remain in solid state at room temperature. Therefore, the lipophilic compounds show its prolonged effect by slow releasing of active compounds without adding any extra organic solvents [37]. The limitations are that the active compound may escape during storage [38].
- g. **Layered double hydroxides (LDHs):** These nanoparticles break down when come in contact with water or carbon dioxide [39]. The positively charged LDH nanoparticles make the active compounds easier to move through the plant cell wall.
- h. **Nanoemulsions:** Nanoemulsions are combination of more than one liquid that does not mix easily. These nanoparticles contain active ingredients in the droplets of diameter 500 nm or less and lessen the degradation of chemicals or active ingredients inside [35].
- i. **Dendrimers:** These nanoparticles are branched, similar to tree like structure, having a central core, which is occupied by functional groups [1]. Very few reports and research have done on dendrimers related to plant pathology. It can be referred as a great delivery vehicle as it can transport chemicals or pesticides in basipetal manner after a foliar spray. It can enhance the permeability in diseased plant cells.

III. Application of nanoparticles to boost/develop commercial agro-protectants (herbicide/fungicide/insecticide)

a. **Nanoparticles as carriers for herbicides:** Nanocarrier-based herbicide studied mainly confined to reduce the environmental toxicity, which occurs due to conventional herbicides. The herbicides, carrier-based nanoparticles with target pest and toxicity is listed in the below **Table 2**.

b. **Nanoparticles as carriers for fungicides:** The mostly used nanoparticle carriers for fungicidal use are silica, chitosan, polymer mixes etc. Nanoparticles for carrier-based fungicides are much popular as they can reduce the loss through volatilization, increase solubility and release chemicals at target sites slowly. The list of nanocarrier-based fungicide, its target pest with crop details, is given in below **Table 3**.

c. **Nanoparticles as carriers for insecticides:** Nanoparticles can be used to lower the toxicity level while increasing its solubility in water. Additionally, these particles lessen the volatilization loss resulting effective protection on targeted sites. The nanoparticles as carrier of insecticide, its target and crop details given in the below-mentioned **Table 4**.

IV. Application of nanoparticles in gene delivery system and to moderate gene expression:

The nanoparticles can be referred as molecular cargo as they can easily transfer the genetic materials such as nucleic

Nanoparticle	Herbicide	Target pest	Toxicity or soil leaching	Reference
Chitosan	Paraquat (L)	Maize, Mustard	Chinese hamster ovary cells <i>A. cepa</i> , Soil sorption	[40]
SLN	Simazine (C)	<i>R. raphanistrum</i>	Mouse fibroblast cells Maize	[41]
Chitosan/tripolyp hosphate	Imazapic (B) and Imazapyr (B)	Black-jack (<i>B. pilosa</i>)	Chinese hamster ovary cells <i>A. cepa</i> Soil biota	[42]
Chitosan	Diuron (C)	<i>E. crusgalli</i>	Maize	[43]
Nanosized rice husk	2,4-D (I)	Maize	<i>Brassica Sp.</i> Soil sorption	[44]
Polymer	Metolachlor	<i>D. sanguinalis</i>	Preosteoblast cell line (mammal)	[45]
Polymeric	Simazine (C)	-	<i>C. elegans</i>	[46]

Table 2.
 Nanocarrier-based herbicidal characterization and their target pests [40–46].

Nanoparticle	Fungicide	Target pest	Crop	Reference
Silver	Tebuconazole	<i>Bipolaris maydis</i>	-	[47]
	Propineb			
	Fludioxonil			
PVP and PVP copolymer	Chlorothalonil, Tebuconazole	<i>Gloeophyllum trabeum</i>	Southern pine sapwood	[48]
Gold	Ferbam	-	Tea	[49]
PVC	Chlorothalonil, Tebuconazole	Turkey tail (<i>T. versicolor</i>) <i>G. trabeum</i>	Southern yellow pine and Birch pine	[50]
Bacterial ghosts	Tebuconazole	<i>L. nodorum</i> , <i>E. graminis</i> , <i>S. fuliginea</i> and <i>P. teres</i>	Wheat, Barley and cucumber	[51]
Chitosan	Carbendazim	<i>F. oxysporum</i> and <i>A. parasiticus</i>	Cucumber, Maize, Tomato	[52]

Table 3.
List of nanocarrier-based fungicides and their application in plant disease management [47–52].

Nanoparticle	Insecticide	Target pest	Crop	Reference
Silica	Chlorfenapyr	<i>H. armigera</i> , <i>P. xylostella</i>	<i>Brassica chinensis</i>	[53]
Sodium alginate	Imidacloprid	Leafhopper (Jassids)	-	[54]
Dendrimers	Thiamethoxam	Cotton bollworm cells and larvae (<i>H. armigera</i>)	-	[55]
Chitosan	Azadirachtin	Tobacco cutworm (<i>S. litura</i>) culture ovarian cell lines	-	[56]
PHSN	Avermectin	<i>P. xylostella</i> larvae	<i>Brassica oleracea</i>	[57]
Zinc oxide and chitosan	Azadirachtin	-	Groundnut bruchid (<i>C. serratus</i>)	[58]
Polydopamine	Avermectin	-	Cotton leaves and corn leaves	[59]

Table 4.
Nano-protectant with insecticidal activity [53–59].

acids to the target site and induce to express the new genetic material in the recipient. Surface-functionalized vertically arranged carbon nanofibers act as molecular cargo and carry the plasmid DNA to the

desired site, expressing the characters in the recipient similar to other conventional methods. Researchers had developed a fluorescent labeled starch nanoparticle-based transgene vector that is able to enter the plant cell and nuclear membrane to deliver the genetic materials. In this system, ultrasound and fluorescent label help in gene delivery by producing transient pores and visual tracking of transgene respectively. Some nanoparticles carry the genetic material by holding it tightly so that it will not get detached from the nanoparticle thereby expressing for short time without integrating to the genetic material of the recipient. Apart from this, sometimes nanoparticle-based gene transfer show negative effect by modifying some genes.

V. Nanoparticles used for abiotic stress tolerance: Nanoparticles are reported to reduce or tolerate abiotic stress like salt stress, heavy metal toxicity, biotic stress etc. In addition to this, nanoparticles can provide mechanical strength to plants, promote germination and seedling growth, help in nutrient and water uptake, etc. Zinc, copper, and iron nanoparticles are observed up to 40 times less lethal than their salts. Hence, these nanoparticles widen the scope for more productivity than other salt-based applications [60]. In 2014, Sabaghnia and Janmohammadi reported that SiO₂ nanoparticles enhanced germination percentage, shoot and root length, seedling fresh and dry weight in lentil plant [61]. In 2017, Taran et al. concluded that the drought tolerance in winter wheat can be alleviated by inducing higher antioxidant enzyme activity when treated with the colloidal solution of Cu and Zn nanoparticles [62].

C. Preparation of silver nanoemulsion

- a. **Wet chemistry method:** This method is one of the best methods in preparation of silver nanoemulsion as it combines in molecular level and versatile in nature. This method is first time carried out by Guzman et al. in 2009. In this method, silver nitrate, hydrazine hydrate, and sodium dodecyl sulfate act as metal precursor, reducing agent, and stabilizing agent respectively. The creation of silver nanoparticles was observed with the help of UV-Vis absorption spectroscopy [63].
- b. **Ion implantation method:** This method was carried out by Popok and his coworkers in 2005. In this method, silver ions were implanted to synthesize the metal nanoparticles in SiO₂ by using 30 KeV energy and ion current density of 4–15 $\mu\text{A}/\text{cm}^2$. The analysis of this new compound was done by using optical spectroscopy and atomic force microscopy [53].
- c. **Physical vapor deposition:** This method was explained by Lin and his coworkers in 2003. In this method, the overall size of silver trifluoroacetate reduced up to 7–11 nm under isoamyl ether and oleic acid solution by properly maintaining the temperature variation. This process is widely accepted and very smooth to manage [54].

5. Nanotechnological applications to reduce postharvest loss: Nanotechnology can be used to limit the post-harvest loss of solid, liquid or processed food

items and beverages. A sensory coating is applied on the product through wrapping or encapsulation method, which slightly modifies the color of the product by improving the taste and shelf-life of the food item. Thin layer of nanoparticles protects the food products from spoilage by holding moisture. Some of the nanoparticles proven promising in lessening the post-harvest loss are carbon nanoparticles, zinc oxide etc., which are mainly antibacterial [9].

6. **Nanotechnological applications in agriculture and animal husbandry:** In livestock management and animal husbandry, nanochips are installed in poultry and livestock to check their health and behavior through sensory technique. Quality of the poultry and livestock products can also be tested by using this method. The quality of meat or any contaminated particles present in it can be checked to avoid microbial spoilage [64]. In case of milk and milk products, **pasteurization** and improved nutrition to increase its quality are the possible ways through nanotechnological system. If nano carrier-based particles are employed in egg production, it will lessen pathogenic infection and transport the nutrients properly resulting quality egg production [65].
7. **Nanotechnological applications in fishery and aquaculture:** Nanotechnology is found to be one of the effective methods in aquaculture in order to increase the nutritional quality like protein and oil and also to promote the health of the fish and other sea foods [9]. Sensory-based drug delivery system was proved to be promising in detection of pathogen present in fish thereby promoting good health and quality nutrition. Nanoparticles mixed in water can also detect the presence of algae or any other micro-organisms and control eutrophication by reducing the phosphorus compounds [66]. Additionally, the encapsulated nanoparticles can effectively be used for fish feed, promoting the fish growth, increasing water quality etc. [67].
8. **Waste management approaches through nanotechnological applications for sustainable agriculture:** Researchers have done enough study on sustainable management of both solid and liquid waste in such a way that it can reduce the soil and water contamination. Scientists observed that implementation of nanotechnology, for example, bionanomaterials and nanobiomimics in waste management is a promising solution. Nanofibers can effectively be used to prepare absorbable substances that can degrade the waste materials in less time without leaving any toxic substances in addition to improve the production and quality of biomass [9].
9. Concerns about nanotechnology in terms of environmental protection as well as human development:
 - a. **Toxicological and environmental safety concerns:** The more use of nanoparticles can lead to negativity on agriculture as many nanosubstances have proven toxic to some beneficial micro-organisms. The uncontrolled application of nanobased agrochemicals can usher to environmental pollution and disturbances in food chain. Before mass release of any nanoparticles, it should be checked properly for safety protocols of toxic composition and its residual effect [9]. A previous study has reported that the pure aluminum-based nanoparticles inhibit root growth of many plants while aluminum oxide-based nanoparticles act as a pollutant and hamper crop growth [68]. Similarly, nanozinc oxide-based products

improve the food quality of soybean while nano-CeO₂ has reported to reduce the soybean yield and nitrogen fixing potential [69]. Therefore, the nanobased products should be in controlled use with prior precaution measures and existing regulatory frameworks to lessen the negative effects on agriculture.

b. Legal and regulatory action: Nonhomologous legal protocols refer to nano entities. In some developing countries like India, some compounds or particles with known application characteristics, if modified to nanoparticles, it will not be registered under patent. Section 3d of Indian patent act limited or restricted the common or already known materials from patent registration thereby opening the ways to do more research on it [70]. These types of laws have widened the paths to find out the scope of nanotechnological application, which will lead to sustainable agriculture.

c. Socioeconomic perspective: The nanotechnological application not only used to get more benefits in case of farmers as well as entrepreneur but also to take our agricultural system toward sustainability. All the nanoproducts should be properly labeled, which will make the farmers to choose the required products. Nanoparticles and nanofibers can improve the efficacy of agrochemicals in less period of time. The use of natural occurring nano formulations is very old. The potentiality of these formulations is very large, which can be known from the patents registered from last year's [9].

2. Summary and future scope

Nanotechnology is one of the emerging methods in agriculture that can bring revolution to bring sustainability. Nanotechnology is not only the potential solution for agriculture but also in other allied sectors like animal husbandry, dairy, poultry, fishery etc. The use of nano-components is one of the potentials, eco-friendly and economical method by using the entire proper regulatory framework. This method should be followed properly by keeping in view of all ethical, regulatory, toxic, and policy concerns. The nanoparticles should come in to market after repeated experiment both in vitro and lab condition.

Nanotechnology helps to developed multiple new methods for green house to field disease management as well as new area of molecular editing and manipulation for plants and pathogenic stress. Therefore diverse nanoparticles like nano-Ag, nano-Cu, nano-Zn used as potential arsenal against various destructive diseases which causes severe yield loss. One of the most important or striking feature is that considerable amount of reduction of metals compared to inorganic agrochemicals. Using of carbon nanoparticles to control disease is also under trial. Every time one thing must be remembered whenever any metal or metallic ions we are applying for nanomaterial preparation, that must be consumable through food chain additionally nontoxic and nonhazardous. It is also found that mechanism of action for each disease, different diseases on the same host and different diseases on various hosts are not the same way of treatment. Because the mode of action is different for each cases. Application of nano-globule in organic farming system is slowly upgrading and matter under consideration of developing countries agricultural policy. Some countries used copper oxychloride, copper hydroxide and copper oxide in their organic farming system in very lesser amount. Application of nanocopper in

organic farming system found to be very effective for its sustainable, environment friendly and long term approach. Researchers are trying to develop micro and macro nutrient enriched plant growth nano promoter. This type of nano-complex not only gives protection against various destructive diseases but also enhancing the plant growth with its slow leaching compatibility mode. Application of different metallic ions and carbon-nanomer under genetic delivery system is the modern topic of research. Introduction of modern tools like quantum dots and biosensor (with combination of nanotechnology) in plant pathology can greatly replace conventional ELISA technique for plant viral detection.

The use of nanobased pesticide, herbicide and fungicide, is an attractive advancement in plant health management keeping in focus of the healthy environment. It has the ability to deliver the pesticide to the specified target with low evaporation loss, improve the solubility, overcome resistance of pesticide in insects and pests, and have better shelf-life. Due to lacking of long-term based in vivo experimental trial, observation or monitoring, more research should be carried out on the particular field. To brand this method as a successful one, material scientists and biologists should cooperate with each other to gain more deep knowledge on complex nanosystems. The reliable, efficient nanoparticles should be chosen that can be biocompatible and no harmful to the plant system. Before releasing, any nanoparticles-based products in to the market, their toxicity, impact on environment, and other risk assessment factors should be analyzed properly. This method is yet to reach to the farmers mostly in developing countries. Therefore, government should work on this to create awareness among the farmers. The journey of nanotechnology in plant disease management and different other sectors of agriculture is under progressive research and wide adaptation through farmers level. The multidisciplinary nanotechnological approaches needs support from government and government-induced schemes or policies and also support from funding agencies, which will lead to a sustainable agriculture system by lessening the global food production as a challenge for future.

Conflict of interest

The authors solemnly and confidently declared that they have no conflict of interest. They are not attached with any other academic or research institutes, except the mentioned affiliation in terms of contribution. Additionally, they also declared that they are not attached with any kind of financial interests or non-financial (or personal interests) interests with any other organization or person. The subject matter of this chapter is totally original and unique or if taken from any other literature properly cited under the references of this manuscript.

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Section 2

Nutrient Management

Packages of Organic Nutrient Management as Soil Policy for Upgrading Cropping System to Restore Soil Productivity

Shaon Kumar Das and Ravikant Avasthe

Abstract

The indigenous farming systems are, by and large, organically practiced. Organic farming systems facilitate the buildup of soil organic matter, reducing risk of erosion and runoff and enhancing nutrient storehouse in soils for plants. Rapid developments in organic farming promotion necessitated continuous flow of technology to meet day-to-day challenges. Farmyard manure (FYM), compost, and green manure are the most important and widely used bulky organic manures. Manuring with different short-duration legumes is suitable for maintenance of soil quality in terms of adding nitrogen to soil. Sustainable quantity of potassium can be maintained by vegetative mulching with crop residues. The use of balanced dosages of mixed compost at 5–10 t/ha along with 2 t/ha dolomite increases yield of maize, rice, mustard, and soybean. This article briefly describes about the integrated organic nutrient management as soil policy for upgrading cropping system to restore soil productivity.

Keywords: organic farming, balanced dose, soil, policy, manure, cropping system

1. Introduction

Sikkim enjoys a wide range of climate, physiographic, geology, and vegetation that influence formation of different kinds of soils. Hills of Sikkim mainly consist of gneissose and half-schistose rocks, producing generally poor and shallow brown soils [1, 2]. The soil is coarse, with large concentrations of iron oxide, and ranges from neutral to acidic, making it lacking in mineral nutrients. This type of soil tends to support evergreen and deciduous forests [3]. Rock consists of phyllites and schists, which is much younger in age and is highly susceptible to weathering and erosion [4–8]. This combined with the state's heavy rainfall causes extensive soil erosion and the loss of soil nutrients through leaching. Soils of Sikkim belong to 3 orders, 7 suborders, 12 great groups, and 26 subgroups. It is observed that inceptisols are dominant (42.84%) followed by entisols and mollisols occupying 42.52 and 14.64%, respectively. Percentage area under Zn deficiency ($<0.6 \text{ mg kg}^{-1}$) in Sikkim is 15.69% (202.35 sq. km) of the geographic area having highest Zn deficiency in South Sikkim district (82.07 sq. km, 19.1% of TGAD) followed by East (56.84 sq. km, 13.3% of TGAD), West (48.91 sq. km, 15.7 of TGAD), and North (14.53 sq. km,

11.8% of TGAD). Percentage area under Mn deficiency ($<3.5 \text{ mg kg}^{-1}$) in Sikkim is 10.16% (131.02 sq. km) of the geographic area having highest Mn deficiency in South Sikkim (48.72 sq. km, 11.3 of TGAD) followed by East (34.52 sq. km, 8.1% of TGAD), North (28.82 sq. km, 23.13% of TGAD), and West (18.96 sq. km, 6.1% of TGAD). Total degraded area in Sikkim is 60,000 ha (9% of TGA), of which West Sikkim is highly degraded, followed by South Sikkim and North Sikkim [9–13]. Erosional hazard has affected about 2000 ha (0.28% of TGA of the state). South Sikkim is the worst affected district, followed by West Sikkim and North Sikkim [14, 15]. Sikkim being hilly state practicing terraced agriculture on an extensive scale could successfully control soil erosion [16].

2. Integrated organic nutrient management practices

The major challenge in organic agriculture is the availability of huge quantities of organic inputs for satisfying the farm demand. The use of animal excreta-based manure alone is not sufficient for meeting the nutrient needs of the crops. It is, therefore, necessary to utilize all the sources available on and off farm effectively [17]. The resource components available for nutrient management in organic horticulture are the following: farmyard manure (FYM), crop residue, weed biomass, green manures, biofertilizers, composts/phospho-compost, vermicomposting, oil cakes, mulching/cover crop, liquid manures, biodynamic preparation, botanicals, legumes in cropping sequence, and certified commercial products. Maintenance of soil fertility may be achieved through organic matter recycling, enrichment of compost, vermicomposting, animal manures, urine, farmyard manure, litter composting, use of botanicals, green manuring, etc. Biofertilizers like *Azolla*, *Azospirillum*, *Azotobacter*, *Rhizobium* culture, PSB, etc. can be used. Sawdust from untreated wood, calcified seaweed, limestone, gypsum, chalk, magnesium rock, and rock phosphate can be used [18]. Various sprays like vermiwash, liquid manure, etc. can be used in crops for nourishing the soil and plant. Farmyard manure, compost, and green manure are the most important and widely used bulky organic manures. Partially decomposed FYM has to be applied 3–4 weeks before sowing, while well-decomposed FYM should be applied immediately before sowing [19]. Manuring with different short-duration legumes is suitable for maintenance of soil quality in terms of adding nitrogen to soil. Nitrogen addition by sun hemp (150–200 kg/ha N) and dhaincha (125–175 kg/ha N) is highly beneficial for the succeeding crops and even for the subsequent crops too. Crop residue can also produce 2.47 kg N, 0.53 kg P, and 8.87 kg K per ha. Edible oil cakes of mustard and nonedible oil cakes from neem, karanj, and castor can serve the dual purpose of manure and bio-pest control [20–25]. Vermicompost can be used for a wide variety of horticultural, ornamental, and vegetable crops at any stage. Generally vermicompost is applied at 3–5 t/ha in row zones for field crops, whereas, for fruit crops, it is preferred to use the same mixing with equal amount of FYM in periodic interval. The general recommendation dose of vermicompost is 6–8 t/ha for field crop and 3–5 t/ha for subtropical fruits [26, 27]. In case of soil application, desired strain of biofertilizer is normally mixed with 20 times well-decomposed FYM to maintain uniformity of mixture and applied in furrows. However, for seedling treatments, biofertilizer slurry is made (1:10 ratio) in water, and roots are emerged in suspension for about 30 minutes. For cereals like, maize, baby corn, buckwheat, upland rice, and finger millet, it was suggested to apply 10–20 t/ha FYM along with 5.0 t/ha vermicompost, whereas, for low P and low K, the dosages are 6–12 t/ha FYM and 3–4 t/ha vermicompost. It is suggested that goat/pig/poultry at 3.0 t/ha along with FYM at 5.0 t/ha is a good source of organic zinc supplement in zinc-deficient soils. For spices like ginger,

turmeric, and large cardamom, it is suggested to apply well-decomposed FYM along with neem cake at 3.0 t/ha and biofertilizer slurry in rows at planting time in variable dosages under low NPK situations [28]. The temperate climate with high organic matter is highly suitable for fruits like mandarin, chayote, strawberry, pear, etc. in the state. Application of well-decomposed FYM along with neem cake and vermicompost at variable dosages during land preparation and biofertilizer treatment before transplanting can be beneficial for improving fruit quality even under the stress of NPK in soils [29].

3. Year-round cropping systems of major crops for lower and mid hills (300: 2000 m amsl)

For rainfed areas, the predominant cropping systems are maize + beans-vegetable pea; maize + beans-barley; maize + beans-rajma; maize + beans-buckwheat; maize + beans-toria; soybean-buckwheat; and soybean-toria. For irrigated areas, the predominant cropping systems are maize (green cobs)-pahenlo dal-buckwheat; maize-vegetable pea; rice-vegetable pea-maize (green cobs); rice-fenugreek (leafy vegetable)-baby corn; rice-sunflower-*dhaincha* (green manuring); and rice-vegetable pea. Important vegetable cropping systems under low-cost plastic tunnels are broccoli-spinach-coriander-broccoli-coriander system; broccoli-coriander-cabbage-radish-coriander system; coriander-radish-fenugreek-spinach-coriander system; cabbage-local rayo sag-broccoli-coriander system; cabbage-spinach-broccoli-coriander system; and coriander-radish-fenugreek-cauliflower-pak choi system [30, 31]. Important vegetable cropping sequences for low-cost plastic rain shelter are tomato-pea-tomato system; bitter gourd-pea-tomato system; bottle gourd-capsicum-pea system; and sponge gourd-pea-tomato system. Important vegetable cropping sequences for low-cost polyhouse are cucumber-cabbage-tomato system; capsicum-broccoli-tomato system; and cucumber-cauliflower-tomato system. Important vegetable cropping sequences for open condition are okra-pea-cole crops system; okra-cole crops-local rayo sag/leafy vegetables system; dalley chili + local rayo sag/leafy vegetables as intercrop; okra-garlic-local rayo sag/leafy vegetables system; ginger-pea system; and okra-potato-local rayo sag/leafy vegetables system. **Table 1** represents the organic nutrient available in Sikkim from all possible sources.

4. Nutrient management in major crops of Sikkim

1. Maize (*Zea mays* L.): Application of dolomite at 2 t/ha + mixed compost at 2.5 t/ha + neem cake at 0.5 t/ha + vermicompost at 2.5 t/ha (ICAR Sikkim, 2011). Apply FYM at 15 t/ha 20 days before planting along with 150 kg rock phosphate. Neem cake at 150 kg/ha for nutrient supply and control of soil-borne insect pests. Green manuring: sun hemp and dhaincha another alternative. Seed inoculant: *Azospirillum*, *Azotobacter*, and PSB at 20 g/kg seed.
2. Rice (*Oryza sativa* L.): Apply FYM at 10 t/ha to supplement recommended dose of N + P + K for maintaining soil fertility. Practice of raising a pre-kharif crop like green gram, cowpea, sun hemp, or *Sesbania* for use as green manure. Biofertilizers (blue-green algae or *Azolla*) capable of providing 20–25 kg N/ha. Neem cake at 150 kg/ha provides protection against soilborne diseases and improves nutrition of rice crops. 5 t FYM + 2 t vermicompost + green manures/weed biomass before 20 days transplanting and 250–300 kg neem cakes during transplanting of rice crop are best nutrient management options. Mixed

Sl. no.	Animal	Livestock population of Sikkim (19th livestock census 2012)	Manure production rate per animal per day in kilograms	Amount of manure produced per day (in kilograms)	Manure/year (tons)	Manure/year On dry weight basis (in tons)
1	Cattle	140,467 CB—126,519 Ind.—13,948	25–30	3,511,675–4,214,010	1281761.375– 1538113.65	384528.4125–461434.095
2	Buffalo	703	25–30	17,575–21,090	6414.875–7697.85	1924.4625–2309.355
3	Sheep	2634	2–3	5268–7903	1922.82–2884.595	576.846–865.3785
4	Goat	113,364	2–3	226,728–340,092	82755.72– 124133.58	24826.716–37240.074
5	Pig	29,907	5	149,535	54580.275	16374.0825
6	Yak	4036	25	100,900	36828.5	11048.55
7	Poultry	451,966	0.6	271179.6	98980.554	29694.1662
Total				4282860.6–5104709.6	1563243.9– 1863219	468973.17–558965.7

Table 1.
Organic nutrient available in Sikkim from all possible sources.

compost at 15 t/ha + neem cake at 1 t/ha. Green manure crops like dhaincha, sun hemp, and cowpea capable of accumulation of 4–5 t/ha of dry biomass and 100 kg N₂/ha in 50–60 days.

3. Rapeseed and mustard (*Brassica* sp.): Apply FYM at 10 t/ha or vermicompost at 5 t/ha during last field preparation. Vermicompost along with Azotobacter and PSB considerably enhances mustard yield. Apply different oil cakes at 0.5 to 1.0 t/ha to meet demand of micronutrient and S demand of the crop. Mixed compost at 5 t/ha + vermicompost at 1.0 t/ha + neem cake at 1.0 t/ha + dolomite at 1.0 t/ha was recommended (ICAR Sikkim, 2011).
4. Soybean (*Glycine max*) (L.) Merr.: Being a leguminous crop, require less N than other crops. Apply FYM at 5–10 t/ha and incorporate into soil during final land preparation. Apply neem cake at 1 t/ha + mixed compost at 2.5 t/ha + dolomite at 1 t/ha (ICAR Sikkim, 2011). Seed inoculation with *Bradyrhizobium japonicum* culture (500 g/75 kg seed) + PSB/PSM (6.5 g/ kg seed).

Buckwheat (*Fagopyrum esculentum* Moench.): Application of vermicompost at 1.5 t/ha recorded the higher grain yield of buckwheat. Efficient crop in extracting phosphorus of low availability from the soil. Azophos seed treatment (APST) + mixed compost at 5 t/ha + neem cake at 0.5 t/ha (ICAR Sikkim, 2011).

5. Baby corn (*Zea mays* L.): Well-decomposed FYM at 10 t/ha should be applied 20 days before sowing of crop. Baby corn should be inoculated with N-fixing nonsymbiotic microorganism like *Azospirillum*, *Azotobacter*, etc. and PSB at 20 g/kg seed.
6. Finger millet (*Eleusine coracana*): Apply 5 t FYM/ha 15 days prior to sowing of the crop. Biofertilizers like *Azospirillum brasilense* (N-fixing) and *Aspergillus awamori* (P-solubilizing) apply at 25 g/kg seed.
7. Black gram (*Vigna mungo* L.): FYM or mixed compost at 5 tons/ha enhances the yield. Seed inoculation with *Rhizobium* strains increases seed yield and uptake of nutrients. Additional nutrient may be supplied through water-soluble organic granules at 5 kg/acre mixed with FYM, vermicompost, or mixed compost.
8. Large cardamom (*Amomum subulatum* Roxb.): If the land is not terraced, soil base may be made by cutting topsoil from upper half and placed on lower half followed by mulching. At plant base, mulching with easily degradable organic materials is good for conserving both moisture and soil. Mulching improves soil physical condition and fertility. Dried organic matter, leaves, weeds, etc. can be used as mulch. During planting, pits are filled with topsoil mixed with FYM at 1–2 kg/pit. FYM/compost at 5 kg/plant at least twice a year in April to May and August to September is beneficial.
9. Ginger (*Zingiber officinale* L.): Well-decomposed and dried cattle manure or compost at 25–30 t/ha + neem cake at 2 t/ha + biofertilizer (*Azospirillum* + PSB) at 5–6 kg/ha helps in reducing incidence of rhizome rot of ginger and increases yield. Two months after planting, vermicompost at 5 t/ha should also be applied for better growth and production. Since edible part is rhizome, prior to planting of seed rhizome in soil, a half foot layer (6") of leaf increases production of ginger by loosening soil texture around seed rhizome at later stages.

10. Turmeric (*Curcuma longa* L.): Needs heavy manuring. Apply FYM at 15–20 t/ha along with 250 kg neem cake or vermicompost at 10 t/ha. Integrated application of FYM at 10 t/ha and vermicompost at 5 t/ha along with 250 kg neem cake. O.M. along with biofertilizers like Azospirillum and Bacillus for better nutrition. Dolomite at 2 t/ha to ameliorate soil acidity.
11. Mandarin (*Citrus reticulata* Blanco.): Young plants manured once/year, bearing plants twice/year (June to July and after harvesting in December to January) at 10–20 kg FYM/tree or 2–2.5 kg vermicompost/tree. Micronutrients through foliar sprays of water-soluble organic sources or nano-fertilizers at 0.2%. Dolomite at 100–200 g/plant for every second year. Neem cake at 2 t/ha during active growth stage in July to August.
12. Kiwifruit (*Actinidia chinensis*): Plants are heavy nitrogen feeders. Apply well-decomposed FYM at 25–30 t/ha and neem cake at 2 t/ha after vines have several inches of new growth during early spring. During active fruit growth stage, vermicompost at 2 kg/plant should also be given for better growth, production, and fruit quality.

Cole crops (*Brassica* spp.): Well-decomposed FYM or compost should be applied at 5.0 kg/m² along with neem cake at 200 g/m² at the time of final land preparation. Root dipping of seedlings in Azospirillum + PSB (20%) for 15 minutes at the time of planting. Additional application of vermicompost in cole crops at 1 kg/m² further improves production.
13. Potato (*Solanum tuberosum* L.): Proper soil fertility management alone accounts for 20.7% of all yield contributing factors. Well-decomposed and dried cattle manure or compost at 25–30 t/ha and neem cake at 2 t/ha should be applied.

5. Identified crops for marketing outside state from Sikkim

The most important crops which have been identified in Sikkim as commercial crop for marketing outside state are large cardamom, ginger, turmeric, buckwheat, cymbidium (flower), and tea. **Table 2** represents the marketing of organic produce in Sikkim.

Agency	Jurisdiction of marketing	Products
Sikkim Marketing Federation (SIMFED)	Within and outside Sikkim	Sikkim mandarin, kiwi, ginger, turmeric, buckwheat, rajma, and vegetables
Farmers Producers Organizations	Within Sikkim	Vegetables
Nature's gift (private entity)	Outside Sikkim	Ginger, turmeric, buckwheat

Table 2.
Marketing of organic produce in Sikkim.

6. Strategies for increasing organic farm productivity in Sikkim

Single-cropping should be avoided and preferably 2–3 crops should be grown together. If for any reason it is not possible to grow mixed or intercrops, then grow


different crops in adjacent plots to maintain diversity. At any given time, legumes must occupy at least 30% of total cropping area. The legumes are nitrogen-fixing and can also be good source of mulching from the crop residues. High-yielding varieties require high nutrient inputs; they should be replaced with improved varieties suitable for organic management [31–35]. The same crop or same cropping sequence should not be repeated in the same field in two consecutive seasons/years (except for some legume crops such as mung bean or cowpea), and the field must be rotated every 2–3 years. Adoption of conservation tillage practices for improving soil quality and conserving soil moisture. Cover cropping, in situ residue management and restoration of degraded lands for soil moisture conservation, and improved C-sequestration should be practiced [36, 37]. Integrated farming systems and watershed development with animal, fishery, and suitable cropping for soil and moisture conservation and nutrient recycling should be practiced. The use of water-saving and nutrient-saving technologies, viz., system of rice intensification (SRI) and aerobic rice, should be popularized [38]. Rainwater harvesting: in situ (land configuration, mulching with locally available biomass, etc.) and ex situ (ponds, micro-water harvesting structures like *jalkund*, etc.) for ensuring year-round high-value crop production [38]. Adoption of conservation irrigation practices like drip, sprinklers, etc. in situ biomass management in shifting cultivation instead of biomass burning for improving soil carbon economy and hydrology should be practiced. Adoption of low-cost plastic tunnels, low-cost plastic rain shelters, and greenhouse (low cost) for year-round production of high-value low-volume vegetable crops should be promoted. Sufficient application of organic matter is crucial for soil fertility management especially for achieving satisfactory yields with good-quality product. Integration of integrated farming system is a necessity for organic farming [39]. It is also important to strengthen the animal husbandry section with main emphasis on poultry and piggery because majority of the population consume meat [40]. Besides, both are more profitable ventures. Composting of locally available biomass and construction of vermibeds for vermicomposting is also essential. Need-based crop diversification which allows more crops per unit area per unit time and per drop of water with due consideration of market demand should be enhanced. Introduction of new oilseeds and pulse crops which have yield potentials to meet the pulses and oilseed requirement of the region should be promoted. Recycling of all kinds of biomass and crop residues for minimizing the dependence of nutrient requirement from outside should be practiced [24, 41–45]. Adoption of soil conservation measures and careful soil cultivation that does not lead to soil erosion and conserves the soil moisture should be practiced. Integrated organic nutrient management strategies should be adopted. Uses of biofertilizers, green manuring, and concentrated organic manures like neem cake should be used for proper nutrition. Preventive measures should be adopted to manage pests, diseases, and weeds [46]. Awareness should be created for off-season vegetable production on scientific lines. There should also be an adoption of cool transport chain, pre-cooling units, packing houses, short- and long-term cold stores, etc. for minimizing the postharvest losses. Extension network for dissemination should be strengthened and the adoption of appropriate knowledge/technologies monitored [47, 48]. Agri Export Zone should be identified by the government for export of organic products, and contract cultivation/cooperative farming should be encouraged.

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Vermicomposting: An Effective Option for Recycling Organic Wastes

Tamanreet Kaur

Abstract

Urbanization and industrialization resulted in rapid increase in volume of solid waste; its management has become one of the biggest problems today. Solid wastes can be disposed off by methods like land filling, incineration, conversion into biogas, recycling, and composting, but its overproduction has led to inappropriate disposal practices such as their indiscriminate and inappropriately timed application to agricultural fields that ultimately leads to water and soil pollution. However, if handled properly, these organic wastes can be used for vermicomposting; it is an effective recycling technology that improves the quality of the products which is disinfected, detoxified, and highly nutritive. It is a low cost, eco-biotechnological process of waste management in which earthworms are used to cooperate with microorganisms in order to convert biodegradable wastes into organic fertilizer. Earthworms excreta (vermicast) is a nutritive organic fertilizer rich in humus, NPK, micronutrients, beneficial soil microbes; nitrogen-fixing, phosphate solubilizing bacteria, actinomycets, and growth hormones auxins, gibberlins and cytokinins, is a suitable alternative to chemical fertilizers, being an excellent growth promoter and protector for crop plants. Thus, vermiculture not only results in management of solid waste but also produces excellent nutrient enriched vermicompost. Vermicompost is beneficial for sustainable organic agriculture and maintaining balanced ecosystem.

Keywords: organic waste, earthworms, vermicompost, nutrients, sustainable organic agriculture

1. Introduction

Increasing world population has resulted in higher consumption of goods and services that has driven a substantial increase of organic wastes originating from households, industry, and agriculture [1]. Much of the organic wastes are highly infectious as they contain a variety of pathogenic microorganisms. Dumping of organic wastes in open areas generates serious environmental issues such as the accumulation of heavy metals in soil, pollution of ground and surface waters due to leaching and run-off of nutrients. These organic wastes when applied directly to agricultural fields cause soil environment-related problems including phytotoxicity [2]. These wastes represent a valuable organic resource, which could be

recycled and transformed into nutrient rich fertilizer and/or soil conditioner [3–5]. Moreover growing awareness about adverse effects of agricultural chemicals on human health has increased interest in organic agriculture [6]. Organic agriculture also promotes ecological conservation due to judicious use of natural resources [7–9]. In demand for safe and sustainable strategies to treat organic wastes includes best known practices of composting and vermicomposting for biological stabilization of solid organic wastes by transforming them into a safer and more stabilized material that can be used as a source of nutrients and soil conditioner in agricultural applications [10–12]. Vermicomposting is one of the most efficient means to mitigate and manage environmental pollution problems [13]. Recently, many studies are being done to establish vermicompost as one of the preferred organic substitutes to chemical fertilizers [14, 15]. Vermicompost is more rich in NPK, micronutrients and beneficial soil microbes (nitrogen fixing and phosphate solubilizing bacteria and actinomycetes), an excellent growth promoter and protector for crop plants [16, 17] than compost [18, 19].

1.1 Vermicomposting- a preferred approach in organic farming

Vermicomposting (vermis from the Latin for worm) is a mesophilic process [20] which involves a joint action of earthworms (active at 10–32°C) and mesophilic microbes [21] for the conversion of organic wastes into a valuable end product known as vermicompost. Whereas, composting involves the degradation of organic waste by microorganisms under controlled conditions, in which the organic material undergoes a characteristic thermophilic stage that allows sanitization of the waste by elimination of pathogenic microorganisms [22]. Composting is also used to treat manures, green wastes or municipal solid wastes [23]. However, vermicomposting gives a higher-quality end product than composting due to joint action of enzymatic and microbial activities that occur during the process [24]. This process is faster than traditional composting as the material passes through the earthworm gut, whereby the resulting earthworm castings are rich in microbial activity and plant growth regulators, and fortified with pest repellence attributes as well [25, 26]. Compared to traditional composting method, vermicomposting also results in mass reduction, shorter processing time, and high levels of humus with reduced phytotoxicity [27]. Thus, vermicompost is considered an ideal manure for organic agriculture as it is nutrient rich and contains high quality humus, plant growth hormones, enzymes, and substances that are able to protect crops against pests and diseases [28, 29]. Moreover, vermicompost has high porosity, aeration, drainage, and water-holding capacity [20]. In addition to increased N availability, C, P, K, Ca and Mg plant nutrient availability in the earthworm casts are also found [30]. Plant growth hormones namely cytokinins and auxins are found in organic wastes processed by earthworms [31]. They also release certain metabolites, such as vitamin B, vitamin D and similar substances into the compost [32]. Thus, earthworms accelerate the mineralization rate and convert the manures into casts with higher nutritional value and degree of humification than traditional method of composting [33]. The composition of commonly available nutrients in vermicompost is as follows: Organic carbon 9.5–17.98%, Nitrogen 0.5–1.50%, Phosphorous 0.1–0.30%, Potassium 0.15–0.56%, Sodium 0.06–0.30%, Calcium and Magnesium 22.67–47.60 meq/100 g, Copper 2–9.50 mg/kg, Iron 2–9.30 mg/kg, Zinc 5.70–11.50 mg/kg, Sulfur 128–548 mg/kg [34]. Hence, vermicomposting enables biological transformation of wastes into a valuable organic fertilizer [35, 36]. Vermicompost is popularly called as black gold and has become one of the major components of organic farming system [26].

2. Role of earthworm in vermicomposting

2.1 Biology of earthworm

Earthworms are invertebrates belonging to the phylum Annelida, class Oligochaeta and family Lumbricidae. The earthworms are long, elongated, cylindrical, soft bodied animals with uniform ring like structures consisting of segments along the length of their body outwardly highlighted by circular grooves called annuli. On the ventral surface of sides of the body each segment bears four pairs of short, stubby bristles, or setae used for its movement. Earthworms have an opening at the anterior end is mouth and the one at the posterior is anus. Earthworms possess both male and female gonads, so are called as hermaphrodites. They deposit their eggs in a cocoon without any larval stage. At the time of egg laying, the sexually mature worms contain a distinctive epidermal ring just beneath the anterior segments called, clitellum, which has gland cells to form a viscid, girdle like structure known as cocoon. The number of fertilized ova in each cocoon has 1–20 lumbricid worms.

2.2 Classification of earthworm

There are about 3320 species of earthworms all over the world [37], but hardly 8–10 species are suitable for vermicompost preparation. Earthworms have been extensively utilized for the recycling of a variety of organic wastes like municipal solid wastes [38] wheat straw [39], sewage sludge [40], forestry waste [41], vegetable waste [42], farmyard manure [43], sorghum stalk, wheat straw, paddy straw [44], coir pith [45]. Renowned scientists, Charles Darwin called earthworms as the ‘unheralded soldiers of mankind’, and Aristotle described them as the ‘intestine of earth’, as they could digest a wide range of organic materials [46, 47]. On the basis of morpho-ecological characteristics, earthworms have been classified into three categories [48]; Anecic (Greek word “out of the earth”) – these are burrowing worms that only come to the surface at night to drag food down into their permanent burrows deep within the mineral layers of the soil. Endogeic (Greek word “within the earth”) – these are also burrowing worms but their burrows are typically more shallow and they feed on the organic matter inside the soil, so they come to the surface only rarely. Epigeic (Greek word “upon the earth”) – these worms live on the surface litter and feed on decaying organic matter. They do not have any permanent burrows. These “decomposers” are the type of worm used in vermicomposting. Two tropical species, African night crawler, *Eudrilus eugeniae* (Kinberg) and Oriental earthworm, *Perionyx excavates* (Perrier) and two temperate ones, red earthworm, *Eisenia andrei* (Bouche) and tiger earthworm, *Eisenia fetida* (Savigny) are extensively used in vermicomposting [49–51]. Most vermicomposting facilities and studies are using the worms *E. andrei* and *E. fetida* due to their high rate of consumption, digestion, and assimilation of organic matter, tolerance to a wide range of environmental factors, short life cycles, high reproductive rates and endurance and resistance during handling [52]. A few other species *Drawida nepalensis*, *Lampito mauritrr*, *Dichogaster* spp., *Polypheretima elongate*, *Amyntas* spp. *Dendrobaena octaedra*, *Eisenia hortensis* [53] have also been used for composting under specific conditions.

2.3 How does earthworm facilitate vermicomposting?

Earthworms promote the growth of “beneficial decomposer aerobic bacteria” in organic waste material and also act as a grinder, crusher, chemical degrader and a biological stimulator of waste material [54, 55]. Earthworm hosts millions of decomposer

(biodegrader) microbes [56], hydrolytic enzymes and hormones that helps in rapid decomposition of complex organic matter into vermicompost in a relatively smaller duration of 1–2 months [57] as compared to traditional composting method which takes nearly 5 months [58]. The mechanism of formation of vermicompost by earthworms occurs in following steps; organic material consumed by earthworm is softened by the saliva in the mouth of the earthworms. Food in esophagus is further softening and neutralization by calcium and physical breakdown in muscular gizzard results in particles of size $<2\ \mu$, thereby giving an enhanced surface area for microbial processing. This finally ground material is exposed to various enzymes such as protease, amylase, lipase, cellulase and chitinase secreted in lumen by stomach and small intestine [12]. Moreover, microbes associated with intestine facilitate breaking down of complex biomolecules into simple compounds. Only 5–10% of the ingested material is absorbed into the tissues of worms for its growth and the rest is excreted as vermicast. The vermicast is a good organic fertilizer and soil conditioner. High-quality vermicast can be produced by worms such as the red wigglers (*E. fetida*) as it contains humus with high levels of nutrients that has good potential for the production of organic fertilizer. Vermiwash is a liquid fertilizer and used as a foliar spray produced by passing water through columns of vermiculture beds [59].

2.4 Suitable environmental conditions for earthworms

Optimum conditions of temperature 15–20°C (limits 4–30°C), Moisture content 80–90% (limits 60–90%), Oxygen – Aerobicity, Ammonia content of the waste Low: $<1\ \text{mg}\cdot\text{g}^{-1}$ (0.016 oz.1b–1), Salt content Low $< 0.5\%$ and pH of 5–9 are preferred for stable life cycle of earthworm.

2.5 Starter food for multiplication of earthworms

To attain the desired earthworm population their starter food includes 1:1 mixture of cow dung and decaying leaves in a cement tank/wooden box/plastic bucket with proper drainage facilities and on attaining sufficient number of earthworms, subsequently other sources of organic wastes can be provided. Compost worms being voracious feeders, consume in excess of their body weight each day but they prefer some foods to others. Manures are the most commonly used worm feedstock, with dairy and beef manures generally considered the best natural food for *E. fetida* [60]. The unit should be kept in shade. Sufficient moisture level should be maintained by occasional sprinkling of water. Within 1–2 months, the worms multiply 300 times, which can be used for large scale vermicomposting.

3. Methods of vermicomposting

Earthworms are used to convert organic waste material into dark brown nutrient rich humus that is a good source of manure for plants. Worms can also degrade specific pollutants and might allow community formation of useful microorganisms. Three commonly used methods for vermicomposting are discussed below:

1. Bin composting: The most common method for small scale composting is bin composting method. The bin can be constructed of several materials such as wooden/plastic/recycled containers like bathtubs and barrels. A vermicompost bin may be in different sizes and shapes, but its average dimensions are $45 \times 30 \times 45\ \text{cm}$. Around 10 holes with 1–1.5 cm in diameter holes in bottom, sides and cap of bin is useful for aeration and drainage.

2. Pit composting: For large scale composting, pits of sizes 2.5 m × 1 m × 0.3 m under thatched sheds with sides left open are advisable. The bottom and sides of the pit should be made hard with a wooden mallet.
3. Pile composting: Pile method is mostly used for vermicomposting in larger scale. The piles can be made in porch place like greenhouse or in a floor with some facilities for drainage in warm climate. The pile size may vary in length and width, however, its height is average height of bin used for bin composting.

4. Conventional steps involved in vermicomposting

Prior to the vermicomposting process, it is preferred to assign pre-composting of organic waste (thermophilic composting), which comprises a short period of high temperature for facilitating mass reduction, waste stabilization, and pathogen reduction [61, 62]. Thermophilic composting results in sanitization of organic wastes and elimination of toxic compounds [63]. Although pathogen removal occurs during transit in the worm gut [64] but thermophilic composting prior to vermicomposting is advisable to avoid the earthworm mortality. Then, after some days of high temperature, pre-mature compost is cooled by spreading it as thin layers on vermicomposting beds. Vermicomposting can be done either in containers, pits or piles.

1. **Materials required for vermicomposting:** Carbon and nitrogen-rich organic materials, spade, ground space, stakes, hollow blocks, plastic sheets or used sacks, water (according to the season) and water sprinklers, shading materials, nylon net or any substitute to cover the beds, and composting earthworms.
2. **Site Selection:** Vermicompost production can be done at any place which is having shades, cool and has high humidity. For instance, abandoned cattle shed, or poultry shed or unused buildings or artificial shading could also be provided.
3. **Shredding of organic waste material:** The collected organic waste material should be processed for shredding along with mechanical separation of the metal, glass and ceramics that should be kept aside.
4. **Pre-digestion of organic waste material:** Pre-digestion of organic waste should be done for at least 20–25 days prior by mixing the waste material along with raw material (e.g., cattle dung slurry). Regular watering is required for partially digesting it and making it fit for earthworm consumption. Raw material to be used includes for composting – cow dung, crop residues, farm wastes, vegetable market wastes and fruit wastes. Cow dung should be at least 20–25 days old to avoid excess heat generation during the composting process. Moreover addition of higher quantities of acid-rich substances such as citrus wastes should be avoided. It is important to mix carbonaceous with nitrogenous organic materials at the right proportions to obtain a C: N ratio of about 30:1, as it results in product of highest stability, the best fertilizer-value and with lowest potential for environmental pollution. For example, rice straw and fresh manure are mixed at about 25:75 ratio by weight. When the material with higher carbon content is used with C:N ratio exceeding 40:1, it is advisable to add nitrogen supplements to ensure its effective decomposition.

5. **Earthworm bed preparation:** An hospitable living environment for worms called bedding is prepared. Bedding is a material that provides the worms with a relatively stable habitat with following characteristics:

i. **High absorbency:** As earthworms breathe through their skins and therefore bedding must be able to absorb and retain water fairly well. Worms die if its skin dries out.

ii. **Good bulking potential:** Worms respire aerobically and different bedding materials affect the overall porosity of the bedding, including the range of particle size and shape, the texture, and the strength and rigidity of its structure. If bedding material is too dense or packs too tightly, then the flow of air is reduced or eliminated. This overall effect is referred to as the material's bulking potential.

iii. **Low protein and/or nitrogen content/high Carbon:** Earthworms consume their bedding as it breaks down and it is very important for this process to be slow. High protein/nitrogen levels can result in rapid degradation of bedding and its associated heating, creating inhospitable or fatal conditions. High carbon content is required as earthworms and microbes in the feed mixtures activate microbial respiration and degradation of organic wastes, thereby increasing the loss of organic carbon during the vermicomposting process [65, 66]. Various bedding materials according to absorbency, bulking potential and C:N are listed in **Table 1**.

6. **Vermiculture bed:** Vermiculture bed can be prepared by placing a first layer of saw dust, newspaper, straw, coir waste, sugarcane trash etc. at the bottom of tub/container. Newspaper is one of bedding materials that is high in absorbency whereas for the sawdust the level of absorbency is poor to medium. A second layer of moistened fine sand of 3 cm thick should be spread over the culture bed followed by a layer of garden soil (3 cm). The floor of the unit should be compacted to prevent earthworm's migration into the soil.

7. **Loading of organic waste mixture in bed:** Third layer of the pre-digested organic waste prepared is added. Thereafter a thin layer of cow dung mixture is placed on the surface of waste material as starter food for compost worms. Then compost worms are to be added without spreading them out. Earthworms consume various organic wastes and reduce the volume by 40–60%. Earthworm eats waste equivalent to its body weight, and produces cast about 50% of the wastes, it consumes in a day.

8. **Composting process:** After addition of compost worms wait for at least 15 days for the thermophilic process to end. During this process there is a rapid increase in temperature followed by a gradual decrease. During this period turning the material 2–3 times at 4–5 days interval is required. Its temperature should be maintained at 30°C, when temperature approaches ambient temperature (<35°C) covering is to be removed and for temperature maintenance, upturning and regular sprinkling of water is advisable. Prominent precautionary measures include; Composting pit should be covered with nylon net or any substitute material to serve as barrier against predators like ants, birds, lizards as it may disturb the activity of earthworm, Blockage of side air vents should be avoided as it can quickly lead to putrefaction and extreme

weather conditions such as frost, heavy rainfall, drought and overheating should be avoided. No smell comes out of composting site if the right products or bedding and feed are used. The vermicompost once formed completely will give the smell of moist soil. Maturity could be judged visually also by observing the formation of granular structure of the compost at the surface of the tank. Next step is to make a heap in sunlight on a plastic sheet and keep it for 1-2 hours. The worms will gather at the bottom of heap. After removing vermicompost on top, the worms settled down at the bottom can be carefully collected for use in the next batch of vermicomposting.

Bedding Material	Absorbency	Bulking Pot.	C:N Ratio
Horse manure	Medium-good	Good	22-56
Peat moss	Good	Medium	58
Corn silage	Medium-Good	Medium	38-43
Hay-general	Poor	Medium	15-32
Straw-general	Poor	Medium-Good	48-150
Straw-oat	Poor	Medium	48-98
Straw-wheat	Poor	Medium-Good	100-150
Paper from municipal waste stream	Medium-Good	Medium	127-178
Newspaper	Good	Medium	170
Bark-hardwoods	Poor	Good	116-436
Bark-softwoods	Poor	Good	131-1285
Corrugated cardboard	Good	Medium	563
Lumber mill waste-chipped	Poor	Good	170
Paper fibre sludge	Medium-Good	Medium	250
Paper mill sludge	Good	Medium	54
Sawdust	Poor-Medium	Poor-Medium	142-750
Shrub trimmings	Poor	Good	53
Hardwood chips, shavings	Poor	Good	451-819
Softwood chips, shavings	Poor	Good	212-1313
Leaves (dry, loose)	Poor-Medium	Poor-Medium	40-80
Corn stalks	Poor	Good	60-73
Corn cobs	Poor-Medium	Good	56-123
Paper mill sludge	Good	Medium	54
Sawdust	Poor-Medium	Poor-Medium	142-750
Shrub trimmings	Poor	Good	53
Hardwood chips, shavings	Poor	Good	451-819
Softwood chips, shavings	Poor	Good	212-1313
Leaves (dry, loose)	Poor-Medium	Poor-Medium	40-80
Corn stalks	Poor	Good	60-73
Corn cobs	Poor-Medium	Good	56-123

Table 1.
List of some of the commonly used earthworm bedding material.

5. Effect of abiotic factors on vermicomposting

The most important abiotic factors which affect vermicomposting process include moisture, pH, temperature, aeration, pH value, ammonia and salt content.

- 1. Moisture:** A strong relationship exists between the moisture content of organic wastes and the growth rate of earthworms. In a comparative study on vermicomposting process and earthworm's growth at different temperature and moisture ranges showed that 65–75% is most suitable range of moisture at all ranges of vermicomposting temperature [67]. The bedding used for vermicomposting must be able to hold sufficient moisture as earthworms respire through their skins and moisture content in the bedding of less than of 45% can be fatal to the worms. Although epigenic species, *E. fetida* and *E. andrei* can survive moisture ranges between 50% and 90%, but they grow more rapidly between 80% and 90% [20, 68]. The bacteria also plays vital role in vermicomposting. Its activity decreases in moisture content lower than 40% and it almost stops in lower than 10% [69].
- 2. Temperature:** Earthworm's activity, metabolism, growth, respiration and reproduction are greatly influenced by temperature [70]. The temperature for the stable development of earthworm population should not exceed 25°C [71]. Although *E. fetida* cocoons survive extended periods of deep freezing and remain viable [72] but they do not reproduce and do not consume sufficient food at single digit temperatures. It is generally considered necessary to keep the temperatures preferably 15°C for vermicomposting efficiency and 20°C for effective reproductive vermiculture operations. Temperatures above 35°C will cause the worms to leave the area or if they cannot leave, they will quickly die. Bacterial activity is also greatly depended on temperature as it multiplies by two per each 10°C increase in temperature and is quite active around 15–30°C.
- 3. Aeration:** Earthworms are oxygen breathers and cannot survive in anaerobic conditions. They operate best when compost material is porous and well aerated. Earthworms also help themselves by aerating their bedding by their movement through it. *E. fetida* have been reported to migrate in high numbers from oxygen depleted water saturated substrate, or in which carbon dioxide or hydrogen sulfide has accumulated.
- 4. pH value:** The pH value is also one of the important factors affecting the vermicomposting process [73]. Epigenic worms can survive in a pH range of 5–9 [74]. The pH of worm beds tends to drop over time. If the food source/ bedding is alkaline, than pH of bed drop to neutral or slightly alkaline and if the food source is acidic than the pH of the beds can drop well below 7. The pH can be adjusted upwards by adding calcium carbonate or peat moss for adjusting pH downward can be introduced into the mix. Although microorganisms which are active in vermicomposting which can maintain their activity even in lower pH of around 4 but recommended pH range for compost is around 6.5–7.5.
- 5. Ammonia and salt content:** Earthworms cannot survive in organic wastes containing high levels of ammonia. Worms are also very sensitive to salts and they prefer salt contents less than 0.5% [75]. However, many types of manures

have high salt contents and if they are to be used as bedding, they should be leached first to reduce the salt content, it is done by simply running water through the material for a period of time [60].

6. Common methods of vermicompost harvesting

The vermicompost is ready within 60–90 days and ultimately the material becomes black, granular, lightweight, moderately loose, crumbly and humus-rich. Watering must be avoided two to three days before emptying the beds to facilitate the separation of worms from the compost. Common procedures for harvesting the vermicompost are briefly described below. Any method may be adopted exclusively by preference. Moreover, two or more methods may be applied on the same pile. Except for the first method, the rest are intended for bulk harvesting.

6.1 Manual harvesting of Vermicompost

This method is practiced if one wants to collect small amounts of vermicast just a few days after the compost pile is stocked with composting worms. In this case only top layer is covered with a thin layer of vermicast and rest of pile has not fully decomposed. The vermicast on top of the pile are simply gathered by hand/trowel and transferred directly into a container. This method is recommended if there is need of organic soil amendment in preparing a fertile potting mix. With time, as vermicompost is collected at the bottom of the pile it is further collected by hand.

6.2 Vermicompost harvesting by pyramidal heap

The vermicompost is first gathered to form a pyramid like heap within the composting enclosure provided that the heap is exposed to light or it is transferred on to a flat surface elsewhere in open sun on a plastic sheet or a sack. This method of harvesting vermicompost takes the advantage of the earthworm's sensitivity towards light as they will tend to move deep into the pyramid. Vermicompost from the bottom, sides, and top surface of the heap is then collected by hand or with a trowel. After the first cycle of vermicompost collection, a few minutes are passed to provide sufficient time for the earthworms to move deeper and another cycle is commenced. For faster rate of harvesting vermicompost, the original heap is divided into several smaller heaps.

6.3 Screening or sieving of vermicompost

This method of vermicompost harvesting is done manually with tool consists of mesh wire nailed on wood called sieve. A small portion from vermicompost pile spread on flat floor is transferred into a sieve and it is shaken so that fine vermicompost falls on the ground. Any undecomposed substrates and earthworms are retained in the screener and the worms are separated manually.

6.4 Vermicompost harvesting by inducing the migration of earthworms

This method of harvesting vermicompost is based on earthworms' ability to detect sources of food. Earthworms have the habit of abandoning the pile exhausted of food and moving towards fresher palatable source. Despite many modifications in this technique, but the basic principle is the same to provide fresh or more palatable food to cause the migration of earthworms from the exhausted pile to the new food source.

7. Storing and packing of vermicompost

The harvested vermicompost should be stored in dark and cool place as sunlight will lead to loss of moisture and nutrient content. Moreover, harvested vermicompost material should be stored in open rather than packed in sacs. Packing should be done at the time of selling and laminated sac is always advisable. During compost storage in open place, periodical sprinkling of water should be done to maintain moisture level and beneficial microbial population. Vermicompost can be stored for longer periods of one year without loss of its quality, if its moisture is maintained at 40% level.

8. Role of vermicompost on soil fertility

The key role of vermicompost is change in physical, chemical and biological properties of soil by earthworm activities and they thus called as soil managers [59]. It substantially improves soil structure, texture, aeration and prevents soil erosion. It increases the macropore space ranging from 50 to 500 μm , resulting in improved air-water relationship in the soil thereby favorably affecting plant growth [76]. It also favorably affects soil pH, its microbial population and soil enzyme activities [77]. Moreover, vermicompost is rich source of nutrients such as nitrates, phosphates and exchangeable calcium and soluble potassium [30]. Apart from adding mineralogical nutrients, vermicompost is also rich in beneficial micro flora such as N-fixers, P-solubilizers, cellulose decomposing micro-flora, etc. It also reduces the proportion of water soluble chemical, which causes possible environmental contamination [78]. Mucus excreted by earthworm's digestive canal produces some antibiotics and hormone-like biochemicals thereby boosting plant growth [70] and enhancing the decomposition of organic matter in soil [79]. Vermicompost has been reported to have favorable influence on the growth and yield parameters of several crops like paddy, sugarcane, brinjal, tomato, and okra [59]. Thus, vermicompost acts a soil conditioner [80] and a slow-release fertilizer [81] that ultimately improves soil structure, soil fertility, plant growth and suppresses diseases caused by soil-borne plant pathogens, increases crop yield [82–84].

9. Conclusion

Chemical fertilizers are produced from “vanishing resources” of earth and crops grown on chemical fertilizers have low and contaminated nutrient value in comparison to grown naturally or organic way. To preserve the agro-ecosystem and protect human health from the harmful chemical fertilizers ‘Ecological Agriculture and Organic Farming’ has to be promoted as the new emerging concept of “Organic Farming” focuses mainly on production of chemical free foods. Organic farming with use of organic fertilizers like “vermicompost” could substitute the chemical fertilizers and can reduce the economic cost and may also lead to organic products which fetches higher price in the market.

Conflict of interest


No conflict of interest is indulged.

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Section 3

Biological Aspects

Application and Mechanisms of Plant Growth Promoting Fungi (PGPF) for Phytostimulation

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Abstract

Plant growth-promoting fungi (PGPF) constitute diverse genera of nonpathogenic fungi that provide a variety of benefits to their host plants. PGPF show an effective role in sustainable agriculture. Meeting increasing demand for crop production without damage to the environment is the biggest challenge nowadays. The use of PGPF has been recognized as an environmentally friendly way of increasing crop production. These fungi have proven to increase crop yields by improving germination, seedling vigor, plant growth, root morphogenesis, photosynthesis, and flowering through either a direct or indirect mechanism. The mechanisms of PGPF involve solubilizing and mineralizing nutrients for easy uptake by plants, regulating hormonal balance, producing volatile organic compounds and microbial enzyme, suppressing plant pathogens and ameliorating abiotic stresses. Successful colonization is an intrinsic factor for most PGPF to exert their beneficial effects on plants. A certain level of specificity exists in the interactions between plant species and PGPF for root colonization and growth promoting effects. There is a gap between the number of reported efficacious PGPF and the number of PGPF as biofertilizer. Efforts should be strengthened to improve the efficacy and commercialization of PGPF. Hence, this chapter summarizes valuable information regarding the application and mechanisms of PGPF in sustainable agriculture.

Keywords: seed germination, seedling vigor, root morphogenesis, yield, root colonization, formulation

1. Introduction

The world's population exceeded ~7 billion just after 2010, and still continues to grow fast. Roughly, 83 million people are added to the world's population every year and with this pace of growth, the global population is projected to reach around 9.7 billion by 2050, ~24% higher than today [1]. In order to feed this large population, crop production must increase by approximately 25–70% above current production levels [2]. Intensification of agriculture is considered a potential solution. By relying on intensive use of fertilizers, pesticides and other inputs, agricultural intensification increases the productivity of existing farmland and delivers more food to the added population. However, the chemical-based crop

intensification produces more food in a way that the future production potential of farmland is being undermined and the environment is being affected. An increasingly degraded soil, overwhelming health hazards from soil and water pollution, disturbed natural microbial populations are a few of the direct implications in chemical-intensive agriculture. To avoid these potentially harmful effects of agrochemicals in agriculture, alternative approaches must be persuaded. An ecocentric approach that provides both environmental and economic benefits is increasingly needed. Organic farming is one of many such approaches that promote agroecosystem health, ensuring sustainable intensification in agriculture.

The uniqueness of microorganisms and the dynamic part played by them in sustaining agricultural ecosystems have made them likely candidates for playing a central role in organic-based modern agriculture. Fortunately, plant roots harbor an abundant association of beneficial microorganisms. Root exudates are the largest source of carbon that attracts the microbial populations and allow them to forge an intimate association with host plants [3]. In response, the rhizosphere microbial populations play versatile roles in transforming, mobilizing and solubilizing soil nutrients, which are crucial for plant growth and development. Among the diverse rhizosphere microbial population, fungi known as plant growth promoting fungi (PGPF) are receiving a growing attention in recent days. Over the decades, varieties of PGPF have been studied including those belong to genera *Trichoderma*, *Penicillium*, *Phoma* and *Fusarium* [4]. Studies have shown that PGPF modulate plant growth and enhance resilience to plant pathogens without environmental contamination [5]. The positive effects of PGPF on plant and environment make them well fitted to organic agriculture.

The course of plant growth promotion by PGPF is a complex process and often cannot be attributed to a single mechanism. A variety of direct and indirect mechanisms, including solubilization of minerals, synthesis of phytohormones, production of volatile organic compounds, exploitation of microbial enzymes, increases in nutrient uptake, amelioration of abiotic stresses and suppression of deleterious phytopathogens are involved. These wide arrays of interconnected mechanisms help PGPF maintaining rhizosphere competence and stability in host performance. Compared to the large number of PGPF identified in the laboratory, only a small fraction of them is in agricultural practice worldwide. Inconsistent performance of the inoculated PGPF under field conditions limits the commercial application of them. Development of appropriate formulation could improve the performance in the field and pave the way for commercialization of the PGPF. An ideal formulation of PGPF should fit with existing application technologies, protect biological actives from stress, ensure viability, remains unaffected after storage under ambient conditions, ensure microbial actives in the field and be cost effective [6].

Considering the aspects discussed above, the need for superior PGPF to supplement inorganic chemical fertilizers as one of the crucial steps of moving toward organic farming practices has been highlighted. Inclusion of new techniques in these processes has been vital to the development of novel PGPF applications. This review will therefore attempt to shed light on the recent findings related to the impact of PGPF on plant growth and yield, duration of their effects, host specificity of the cooperation, root colonization mechanisms, their modes of action and commercial formulation for enhancement of plant growth and yield. The knowledge produced from this review could be very useful to those who are apprehensive about environmental protection and agricultural sustainability.

2. Plant growth promoting fungi (PGPF)

Plants have intricate relationships with an array of microorganisms, particularly rhizosphere fungi and bacteria, which can lead to an increase in plant vigor, growth and development as well as changes in plant metabolism [7]. The group of rhizosphere fungi that colonize plant roots and enhance plant growth is referred to as PGPF [4]. PGPF are heterogeneous group of nonpathogenic saprotroph fungi. They can be separated into endophytic, whereby they live inside roots and exchange metabolites with plants directly, and epiphytic, whereby they live freely on the root

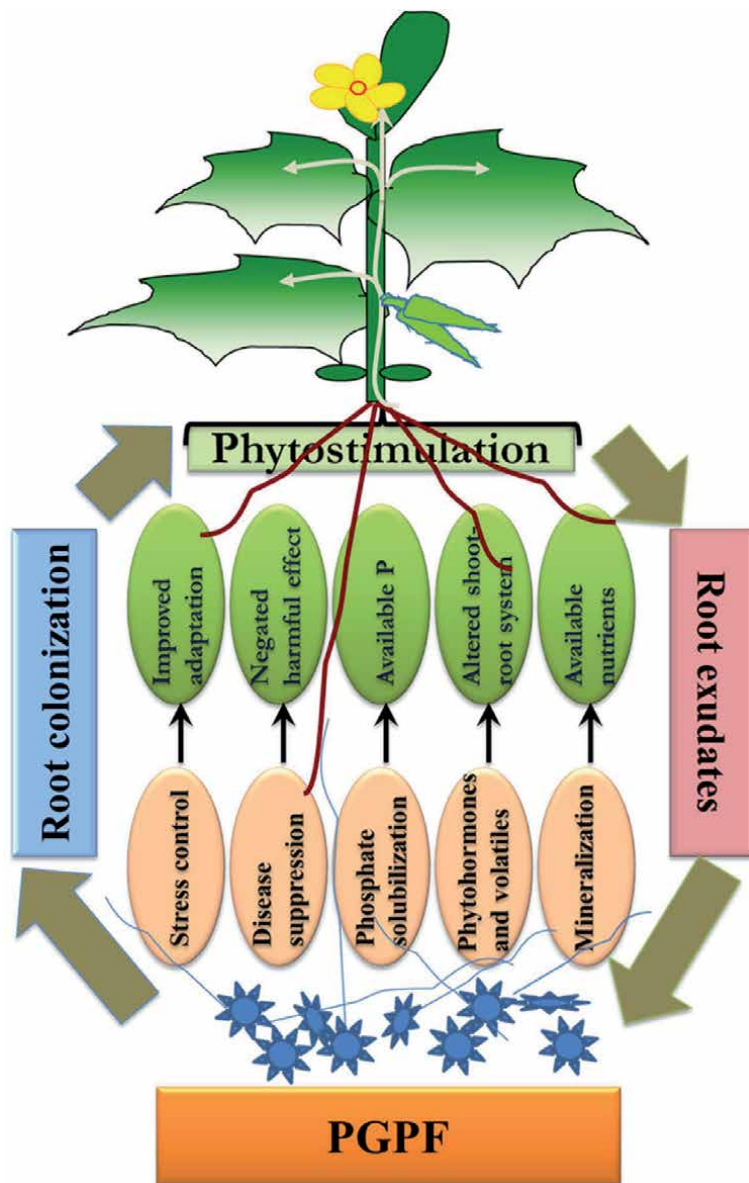


Figure 1. Beneficial interaction between plant and plant growth promoting fungi (PGPF). PGPF can modulate plant growth and development through the production of phytohormones and volatile compounds. PGPF also influence plant nutrition via solubilization of phosphorus and mineralization of organic substrates. PGPF modify plant functioning against biotic and abiotic stresses by negating their harmful effects.

surface and free-living PGPF, which live outside plant cells, i.e., in the rhizosphere [5]. PGPF establish a non-obligate mutualism with a broader range of host plants. That is why symbiotic mycorrhizal fungi are not considered as PGPF, although they are known to improve growth of the plants [8]. Moreover, PGPF encompass a diverse taxonomic group in comparison to mycorrhiza. They are often involved in a range of complex interactions with plants and develop distinct strategies to mediate improvements in seed germination, seedling vigor, plant growth, flowering and productivity of host plants (**Figure 1**). PGPF are not only associated with the root to mediate positive effects on plant growth and development but also have beneficial effects on suppressing phytopathogenic microorganisms [9]. Not every organism identified as PGPF will improve plant growth under all conditions or in association with all plant hosts [10]. Some PGPF biocontrol inoculants usually contain necrotrophic mycoparasites such as *Trichoderma* spp. [11], while a limited number such as *Sphaerodes mycoparasitica* is biotrophic mycoparasitic agent [12]. Therefore, PGPF are considered one of the potential active ingredients in both biofertilizer and mycofungicide formulation.

3. The nature and composition of PGPF

PGPF are common root-associated and soil-borne fungi from diverse genera. Fungi reported as PGPF include Ascomycetes, Basidiomycetes and Oomycetes [5]. Some strains of hypovirulent binucleate *Rhizoctonia* (HBNR) are known to be PGPF [13]. PGPF also include isolates of mycelial fungi that do not produce any spores, generally known as sterile black fungus (SBF), sterile dark fungus (SDF) and sterile red fungus (SRF) [14]. The non-sporulating PGPF are often difficult to identify and mostly lack formal taxonomic status. Among the PGPF *Aspergillus*, *Fusarium*, *Penicillium*, *Phoma* and *Trichoderma* have a wide distribution and are, by far, the most extensively reported (**Table 1**). Each of the genera has a variety of species. *Aspergillus*, *Fusarium*, *Penicillium* and *Phoma* were frequently found in the rhizosphere or in the roots of plants. Instead, *Trichoderma* were mostly isolated from soil. Among the rhizosphere population, PGPF have a high relative abundance. A total of 619 (44%) out of 1399 fungal isolates collected from rhizosphere of six different plants were PGPF, while frequency of occurrence of PGPF in zoysiagrass, wheat, corn and eggplant rhizosphere were 46, 47, 38 and 10%, respectively [4]. This indicates that abundance of PGPF varies largely according to the host rhizosphere. Similarly, the dominating fungal genus is not necessarily the dominating PGPF in the rhizosphere population. The order of the frequency of the main genera among 1399 fungal isolates was *Fusarium* > *Trichoderma* > sterile fungi > *Penicillium* > *Pythium* > *Rhizoctonia* > *Mucor*, while that of PGPF from each plant genus was: *Trichoderma* (~82%) > *Pythium* (~75%) > *Penicillium* (~69%) > *Alternaria* (~63%) > *Fusarium* (~44%) > sterile fungi (40%) > *Mucor* (~38%) [4]. The important characteristics of these fungi are their high rhizosphere competence and ability to promote plant growth.

Initial search for identification of PGPF was concentrated to rhizosphere fungi. Recent studies have demonstrated the potential of phyllosphere fungi as PGPF. The phyllosphere, which consists of the above ground surfaces of plants, is one of the most prevalent microbial habitats on earth. Phyllosphere fungi can act as mutualists promoting plant growth and tolerance of environmental stressors [53]. A few of other fungi isolated from tree bark, decorticated wood and water damaged building functioned as PGPF [43, 49]. More interestingly, the fungal entomopathogens also show potential to be PGPF and promote plant growth [54]. PGPF seem to have a cosmopolitan occurrence.

PGPF	Original source of isolation	References
<i>Alternaria</i> sp.	<i>Zoysia tenuifolia</i> , <i>Rosa rugosa</i> , <i>Camellia japonica</i> , <i>Delonix regia</i> , <i>Dianthus caryophyllus</i> , <i>Rosa hybrid</i>	[4, 15]
<i>Aspergillus</i> sp., <i>As. fumigatus</i> , <i>As. niger</i> , <i>As. terreus</i> , <i>As. ustus</i> , <i>As. clavatus</i>	<i>Capsicum annuum</i> , <i>Glycine max</i> , <i>Cicer arietinum</i> , <i>Elymus mollis</i> , <i>Solanum tuberosum</i> , <i>Nymphoides</i> <i>peltata</i>	[16–21]
<i>Aureobasidium pullulans</i>	Dark chestnut soil	[22]
<i>Chaetomium globosum</i>	<i>Capsicum annuum</i>	[23]
<i>Cladosporium</i> sp., <i>Cladosporium sphaerospermum</i>	<i>Cucumis sativus</i> , <i>Glycine max</i>	[24, 25]
<i>Colletotrichum</i> sp.	<i>Rosa rugosa</i> , <i>Camellia japonica</i> , <i>Delonix regia</i> , <i>Dianthus caryophyllus</i> , <i>Rosa hybrid</i>	[15]
<i>Exophiala</i> sp.	<i>Cucumis sativus</i>	[26]
<i>Fusarium</i> sp., <i>F. equiseti</i> , <i>F. oxysporum</i> , <i>F.</i> <i>verticillioides</i>	<i>Cynodon dactylon</i> , <i>Lygeum spartum</i> , <i>Zoysia tenuifolia</i> , <i>Musa</i> sp. and other environment	[27–32]
Non-sporulating sterile fungi	<i>Zoysia tenuifolia</i>	[14]
<i>Penicillium</i> sp., <i>Pe. chrysogenum</i> , <i>Pe. citrinum</i> , <i>Pe. klockeri</i> , <i>Pe. menonorum</i> , <i>Pe. resedanum</i> , <i>Pe.</i> <i>simplicissimum</i> , <i>Pe. janthinellum</i> , <i>Pe. viridicatum</i>	Halophyte, <i>Ixeris repenes</i> , <i>Cicer</i> <i>arietinum</i> , <i>Elymus mollis</i> , <i>Capsicum</i> <i>annuum</i> , <i>Zoysia tenuifolia</i>	[9, 16, 22, 33–40]
<i>Phoma</i> sp., <i>Phoma herbarum</i> , <i>Phoma multirostrata</i>	<i>G. max</i> , <i>Rosa rugosa</i> , <i>Camellia</i> <i>japonica</i> , <i>Delonix regia</i> , <i>Dianthus</i> <i>caryophyllus</i> , <i>Rosa hybrid</i> , <i>Zoysia</i> <i>tenuifolia</i>	[4, 14, 15, 34, 41, 42]
<i>Phomopsis</i> sp., <i>Phomopsis liquidambari</i>	<i>Rosa rugosa</i> , <i>Camellia japonica</i> , <i>Delonix regia</i> , <i>Dianthus caryophyllus</i> , <i>Rosa hybrid</i> , <i>Bischofia polycarpa</i> bark	[15, 43]
<i>Purpureocillium lilacinum</i>	Soil	[44]
<i>Rhizoctonia</i> spp.	Orchid, <i>Lycopersicon lycopersicum</i> , and soil	[13, 45, 46]
<i>Rhodotorula mucilaginosa</i>	Soil	[22]
<i>Talaromyces wortmannii</i>	Soil	[40]
<i>Trichoderma asperellum</i> , <i>T. atroviride</i> , <i>T. hamatum</i> , <i>T. harzianum</i> , <i>T. longibrachiatum</i> , <i>T. pseudokoningii</i> , <i>T. viride</i> , <i>T. virens</i>	Soil, wood and damaged building	[34, 47–52]

Table 1.
 Different fungi reported as plant growth promoting fungi (PGPF) with their original source of isolation.

4. Impact of PGPF on plant growth promotion

PGPF exhibit traits beneficial to plant and as such, their capacity to enhance plant growth and development is well founded. PGPF mediate both short- and long-term effects on germination and subsequent plant performance. Improvement in germination, seedling vigor, shoot growth, root growth, photosynthetic efficiency, flowering, and yield are the most common effects decreed by PGPF. A particular PGPF may condition plant growth by exerting all or one or more of these effects.

4.1 Impact of PGPF on seed germination and seedling vigor

Seed germination and germinant growth are critical developmental periods of the young plantlet until it begins producing its own food by photosynthesis. Treatment with PGPF, particularly of the genus *Aspergillus*, *Alternaria*, *Trichoderma*, *Penicillium*, *Fusarium*, *Sphaerodes* and *Phoma* has been reported to improve seed germination and seedling vigor in different agronomic and horticultural crops (Table 2). Scarified seeds inoculated with spores from *Aspergillus* and *Alternaria* had significant increases in germination of Utah milkvetch (*Astragalus utahensis*) *in vitro*, and in greenhouse and fall-seeded plots near Fountain Green and Nephi [55]. The *Aspergillus*-treated seeds performed out seeds inoculated with *Alternaria*. An increase of 30% in seedling emergence was observed in cucumber plant raised upon the treatment of *T. harzianum* [47]. Application of *T. harzianum* also significantly increased seed germination, emergence index, seedling vigor and successful transplantation percentage in muskmelon compared to the untreated controls [59]. Early seedling emergence and enhanced vigor were observed in bacterial wilt susceptible tomato cultivar treated with *T. harzianum*, *Phoma multivirata*, and *Penicillium chrysogenum* compared to untreated controls [34]. The culture filtrate of *Penicillium* was as effective as the living inocula in improving seed germination of tomato [70]. Significantly, higher germination and vigor index were observed in Indian spinach, when seeds were sown in sterilized field soil amended with wheat grain inoculum of *Fusarium* spp. PPF1 [27]. *Sphaerodes mycoparasitica*, a biotrophic mycoparasite of *Fusarium* species, improved wheat seed germination and seedling growth *in vitro* compared to *T. harzianum*, while under phytotron conditions, both *S. mycoparasitica* and *T. harzianum* had positive impact on wheat seedlings growth in the presence of *F. graminearum* [12]. These results show the positive impact of PGPF on seed germination and seedlings growth of a wide arrays of hosts.

4.2 Impact of PGPF on shoot growth

The most common form of growth promotion by PGPF is the augmented shoot in colonized plants. Shoot growth promotion has been shown by a great diversity of PGPF across a large number of plant species. Isolates of *Aspergillus*, *Trichoderma*, *Penicillium*, and *Fusarium* were capable of enhancing the shoot growth in model plant *Arabidopsis* [9, 20, 28, 33, 48]. Different species of *Aspergillus* are known to support shoot growth in chickpea [16], Chinese cabbage [56], cucumber [17], soybean [18, 65] and wheat [76]. Species of nonpathogenic *Fusarium* were reported to stimulate shoot growth in Indian spinach [27] and banana [29]. Application of barley grain inoculum of *Penicillium viridicatum* GP15-1 to the potting medium resulted in 26–42% increase in stem length, 37–46% increase in shoot fresh weight and 100–176% increase in shoot dry weight of cucumber plants [35]. Similarly, inoculation of cucumber plants with *Pe. menonorum* KNU3 increased cucumber shoot dry biomass by as much as 52% [36]. Stimulated shoot growth by *Penicillium* spp. was also reported in tomato [69], Waito-c rice [37, 38], chili [23, 39] and sesame [74]. Application of *T. longipile* and *T. tomentosum* increased shoot dry weight of cabbage seedlings by 91–102% in glasshouse trials [57]. Likewise, cottonseeds pretreated with *T. viride* showed four-fold increases in shoot length elongation and an almost 40-fold increase in plant dry weight compared to the control [66]. Augmented shoot growth by *Trichoderma* has also been reported in chickpea [16], wheat [79], maize [78], cucumber [60] and other plant species (Table 2). Isolates of *Phoma* were found to be an efficient stimulator of plant shoot [15, 41, 62]. A few hypovirulent *Rhizoctonia* isolates were able to induce significantly higher fresh leaves and stems weights in tomato plants grown in greenhouse [13]. Enhancement of shoot growth was also observed

Test crop	PGPF strain	Improvement	References
<i>Arabidopsis thaliana</i>	<i>Trichoderma virens</i> Gv. 29-8	Biomass, lateral root development	[48]
	<i>Penicillium janthinellum</i> GP16-2	Shoot biomass, leaf number	[33]
	<i>Pe. simplicissimum</i> GP17-2	Shoot biomass, leaf number	[9]
	<i>Fusarium oxysporum</i> NRRL 38499, NRRL 26379 and NRRL 38335,	Shoot-root growth	[28]
	<i>Aspergillus ustus</i>	Shoot growth, lateral root, root hair numbers	[20]
<i>Astragalus utahensis</i>	<i>Aspergillus</i> spp., <i>Alternaria</i> spp.	Seed germination	[55]
<i>Basella alba</i>	<i>Fusarium</i> spp. PPF1	Germination, seedling vigor, shoot-root growth, leaf area, leaf chlorophyll content	[27]
<i>Brassica campestris</i>	<i>Talaromyces wortmannii</i> FS2	Shoot fresh weight	[40]
<i>B. chinensis</i>	<i>A. niger</i> 1B and 6A	Plant dry weight, N and P content	[56]
<i>B. oleracea</i> var. <i>capitata</i>	<i>T. longipile</i> , <i>T. tomentosum</i>	Shoot dry weight, leaf area	[57]
<i>Capsicum annum</i>	<i>Pe. resedanum</i> LK6	Shoot length, biomass, chlorophyll content, photosynthesis	[39]
	<i>Chaetomium globosum</i> CAC-1G	Plant biomass, root-shoot growth	[23]
<i>Cicer arietinum</i>	<i>A. niger</i> BHUAS01, <i>Pe. citrinum</i> BHUPC01, <i>T. harzianum</i>	Plant growth	[16]
	<i>T. harzianum</i> T-75	Yield	[58]
<i>Cucumis melo</i>	<i>T. harzianum</i> Bi	Germination, seedling health, vigor	[59]
<i>Cucumis sativus</i>	<i>Pe. simplicissimum</i> GP17-2	Root-shoot growth	[4]
	<i>Pe. viridicatum</i> GP15-1	Root-shoot length, biomass	[35]
	<i>T. harzianum</i> GT3-2	Root-shoot growth	[60]
	<i>F. equiseti</i> GF19-1	Root-shoot growth	[61]
	<i>Aspergillus</i> spp. PPA1	Root-shoot length, biomass, leaf area, chlorophyll content	[17]
	<i>Exophiala</i> sp. LHL08	Plant growth under drought and salinity	[26]
	<i>Phoma</i> sp.	Root-shoot growth, yield in the field	[62]
GiSeLa6® (<i>Prunus cerasus</i> × <i>P. canescens</i>)	<i>Phoma</i> sp. GS8-2, GS8-3	Root-shoot growth	[63]
	<i>T. harzianum</i> T-22	Root growth, development	[64]
<i>Glycine max</i>	<i>A. fumigatus</i> HK-5-2	Shoot growth, biomass, leaf area, chlorophyll contents, photosynthetic rate	[65]
	<i>A. fumigatus</i> LH02	Shoot growth, biomass, leaf area, chlorophyll contents, photosynthetic rate	[18]
	<i>Phoma herbarum</i> TK-2-4	Plant length, biomass	[41]
<i>Gossypium arboreum</i> L	<i>T. viride</i>	Root-shoot length, plant dry weight	[66]

Test crop	PGPF strain	Improvement	References
<i>Helianthus annuus</i>	<i>Trichoderma</i> sp., <i>Aspergillus</i> sp., <i>Penicillium</i> sp., <i>Phoma</i> sp., <i>Fusarium</i> sp.	Seed germination, seedling vigor	[67]
<i>Lactuca sativus</i>	<i>F. oxysporum</i> MSA 35	Root-shoot growth, chlorophyll content	[68]
<i>Lycopersicon lycopersicum</i>	<i>T. harzianum</i> TriH_JSB27, <i>Phoma multirostrata</i> PhoM_JSB17, <i>T. harzianum</i> TriH_JSB36, <i>Pe. chrysogenum</i> PenC_JSB41	Seedling emergence, vigor	[34]
	<i>T. harzianum</i> T-22	Seed germination under stress	[69]
	<i>Penicillium</i> spp.	Seed germination, root-shoot growth	[70]
	<i>F. equiseti</i> GF19-1	Plant biomass, root-shoot growth	[71]
<i>Musa</i> sp.	<i>F. oxysporum</i> V5W2, Eny 711o, Emb 2.4o	Yield	[29]
<i>Nicotiana tabacum</i>	<i>Alternaria</i> sp., <i>Phomopsis</i> sp., <i>Cladosporium</i> sp., <i>Colletotrichum</i> sp., <i>Phoma</i> sp.	Root-shoot growth, chlorophylls, soluble sugars, plant biomass	[15]
<i>Pinus sylvestris</i> var. <i>mongolica</i>	<i>T. harzianum</i> E15, <i>T. virens</i> ZT05	Seedling biomass, root structure, soil nutrients, soil enzyme activity	[72]
<i>Saccharum officinarum</i>	<i>T. viride</i>	Yield	[73]
<i>Sesamum indicum</i>	<i>Penicillium</i> spp. NICS01, DFC01	Root-shoot growth, chlorophylls, proteins, amino acids, lignans	[74]
<i>Solanum tuberosum</i>	<i>A. ustus</i>	Root-shoot growth, lateral root, root hair numbers	[20]
<i>Spinacia oleracea</i>	<i>F. equiseti</i>	Plant biomass, root-shoot growth	[75]
<i>Suaeda japonica</i>	<i>Penicillium</i> sp. Sj-2-2	Plant length	[38]
	<i>Cladosporium</i> sp. MH-6	Shoot length	[24]
	<i>Pe. citrinum</i> IR-3-3	Root-shoot length	[37]
	<i>Phoma herbarum</i> TK-2-4	Plant length	[41]
<i>Triticum aestivum</i>	<i>T. harzianum</i> , <i>T. koningii</i>	Plant biomass, root-shoot growth.	[4]
	<i>Sphaerodes mycoparasitica</i>	Seed germination, seedling growth	[12]
	<i>A. niger</i> NCIM	Shoot and total plant length ratio	[76]
<i>Vinca minor</i>	<i>T. harzianum</i>	Flowering, plant height, weight	[77]
<i>Zea mays</i>	<i>T. harzianum</i> T22	Shoot growth, area and size of main and secondary roots	[78]

Table 2. Effect of different plant growth promoting fungi (PGPF) on seed germination, plant growth and yield in various plants.

by *Talaromyces wortmannii* in cabbage [40], *Chaetomium globosum* in chili [23], *Colletotrichum* sp. in tobacco and *Exophiala* sp. in cucumber [26]. The results from these studies are consistent with numerous field and growth chamber experiments that have shown that PGPF inoculants can mediate shoot growth improvement.

4.3 Impact of PGPF on photosynthesis

The plant growth promotion in some plant-PGPF interaction is occasionally associated with improvement in state and function of the photosynthetic apparatus of plants. Treatment with *T. longipile* and *T. tomentosum* increased leaf area of cabbage by 58–71% in glasshouse trials [57]. Tomato plants grown with HBNR isolates had significantly higher leaf fresh weight than control plants in greenhouse [13]. Arabidopsis grown in soil amended with *Pe. simplicissimum* GP17-2 and *Pe. janthinellum* GP16-2 were more greener and had approximately 1 more leaflet per plant than control plants 4 weeks after treatment [9]. *Penicillium* spp. also enhanced leaf chlorophyll content in cucumber and chili [36, 39]. Soil amendment with *Aspergillus* spp. PPA1 and *Fusarium* spp. PPF1 significantly increased leaf area and leaf chlorophyll content in cucumber and Indian spinach, respectively [27]. Improvement in leaf number, leaf area and leaf chlorophyll levels would contribute to increases in photosynthesis rate and net accumulation of carbohydrate in plants.

4.4 Impact of PGPF on root growth and architecture

Roots are vital plant organs that remain below the surface of the soil. The root system is important for plant fitness because it facilitates the absorption of water and nutrients, provides anchorage of the plant body to the ground and contributes to overall growth of plants. Root functions as the major interface between the plant and the microbes in the soil environment. The bulk of previous studies have evidenced the immense ability of PGPF in enhancement of root growth in different plants (Table 2). Plants forming association with PGPF show faster and larger root growth resulting in a rapid increase in the root biomass [27, 35, 50, 57]. Moreover, root length, root surface area, root diameter and branch number are under direct influence of intimate interaction with PGPF. Application of *T. vires* ZT05 increased root length, root surface area, average root diameter, root tip number and root branch number of pines by 25.11, 98.19, 5.66, 45.89 and 74.42%, respectively [72]. *A. ustus* is known to cause alterations in the root system architecture by promoting the formation of secondary roots in Arabidopsis and potato [20]. In maize (*Zea mays*), *Trichoderma* inoculation enhanced root biomass production and increased root hair development [78]. The abundance in root hair formation significantly increases root surface area, suggesting that PGPF inoculants could enhance the potential for plant roots to acquire nutrients under nutrient-limited conditions.

4.5 Impact of PGPF on flowering

The application of PGPF may influence the number, size and timing of flower in flowering plants. *Tagetes* (marigolds) grown with companion of *Pe. simplicissimum* flowered earlier and had greater flower size and weight [80]. Steamed or raw soil infested with *T. harzianum* hastened flowering of periwinkle and increased the number of blooms per plant on chrysanthemums [77]. Under greenhouse conditions, *T. harzianum* TriH_JSB27 and *Pe. Chrysogenum* PenC_JSB41 accelerated the flowering time in tomato [34]. Similarly, root colonization by the nematophagous fungus *Pochonia chlamydosporia* hurried flowering in *Arabidopsis thaliana* [81]. Root colonization by *Piriformospora indica* also results in early flowering in *Coleus forskohlii*, bottle gourd and *Nicotiana tabacum* [82]. Flowering time has commercial significance for crops and ornamental plants by shortening crop duration and improving productivity. A short duration crop would have several advantages over a long duration crop, even with equal total yields such as require less water, expose less to stresses and

increase the availability of the land for subsequent cropping. This indicates that PGPF improve the plasticity of complex plant traits.

4.6 Impact of PGPF on yield

PGPF show promising ability to promote growth through extensive improvements and betterment of fundamental processes operating in the plants, all of which directly and indirectly contributes to the crop yield increase. Inoculation of banana (cv. Giant Cavendish and Grand Nain) with *F. oxysporum* resulted in 20–36% yield increase in the field [29]. Soil treatment with *T. harzianum* alone or in combination with organic amendment and fungicide significantly improved seed yield in pea [83] and chickpea [58]. Similarly, soil treatment with *T. viride* produced significantly the highest number of fruits per plant, number of seeds per fruit, fruit weight and dry weight of 100 seeds as compared to untreated control [84]. The beneficial association of plants with nonpathogenic binucleate *Rhizoctonia* spp. resulted in increase in yield of carrot, lettuce, cucumber, cotton, radish, wheat, tomato, Chinese mustard and potato [13, 45, 46]. These results demonstrate that PGPF hold great promise in the improvement of agriculture yields.

5. Duration of sustained plant growth promotion effect by PGPF

The duration of biofunctional activities of PGPF in plants is a key factor for their effective application in the field. Naturally, a legitimate question may arise whether PGPF isolates that have shown promising effects on early growth stage of plants, could also affect the middle or late ontogenetic stages and ultimately contribute to yield increases at harvest. As for potato, an increase in leaf, shoot, and tuber weight was observed by a nonpathogenic isolate (No. 521, AG-4) of *Rh. solani* 63–70 days after planting, while it was not expressed in yield at harvest [85]. Conversely, increased growth responses of wheat plants treated with PGPF were observed during seedling (2 weeks after sowing), vegetative (4 weeks), pre-flowering (6 weeks), flowering (10 weeks) and seed maturation stages (14 weeks) [4]. The isolates of *Phoma* sp. (GS6-1, GS7-4) and non-sporulating fungus (GU23-3), increased plant height, ear-head length and weight, seed number and plant biomass at harvest [79]. Again, isolates of *Phoma* sp. and non-sporulating fungus significantly increased plant length, dry biomass, leaf number and fruit number of cucumber cv. Jibai until 10 weeks post planting in greenhouse trials [62]. These isolates were equally effective in promoting growth and increasing yield of cucumber at 6 and 10 weeks post planting in the field [62]. There are other PGPF, which as well have shown the ability to confer long-term growth benefits to different plants. Rice and pea plants inoculated with *Westerdykella aurantiaca* FNBR-3, *T. longibrachiatum* FNBR-6, *Lasiodiplodia* sp. FNBR-13 and *Rhizopus delemar* FNBR-19 showed a stimulatory increase of growth for 8 weeks in the greenhouse [86]. Similarly, a single inoculation with inoculum of *Penicillium* and *Pochonia* affected the whole life cycle of tomato and Arabidopsis, respectively, accelerating the growth rate, shortening their vegetative period and enhancing seed maturation [34, 81]. As such, majority of PGPF strains are able to induce sustained beneficial effects on plant growth. The basis of sustained effects of PGPF on plants is not fully understood. One possibility is that the fungus continues to colonize the root system and establishes a life-long colonization with crop roots. The ability of PGPF to confer sustained benefit to plant is of great agriculture importance in terms of improving crop yield.

6. Host specificity of the plant growth-promoting cooperation

Although plants harbor a diverse community of fungi, a preferential interaction exists between certain PGPF and a particular host. Once a particular host mutualizes this fungus, it undergoes host-specific adaptations. The outcome of such adaptations is a highly specialized and finely tuned mutualism, leading to improved responsiveness to each other needs. Evidences show that PGPF that induce growth in one plant species do not necessarily have the same effect in other species [5]. Some PGPF exert general growth promotion effects in several plant species, other fungi only do so in specific host plant. A field study showed that most of eight non-sporulating PGPF isolates enhanced the growth of one wheat variety, whereas a few isolates enhanced the growth of the other variety [87]. Moreover, at least four isolates increased yields of both varieties. Thus, the efficacy of the PGPF isolates depended upon the wheat variety in addition to their inherent growth promoting abilities. Similarly, many of the zoysiagrass PGPF isolates promoted growth of bentgrass [4], in contrast to a few isolates enhanced growth in soybean [88]. Similarly, nine isolates belonging to *Phoma* sp. and one non-sporulating fungus caused consistent plant length enhancement in cucumber cv. Shogoin fushiharii compared to nine isolates except the non-sporulating fungus in cv. Aodai Kyuri. Again, plant length enhancement in cv. Jibai was shown by eight *Phoma* sp. and one non-sporulating fungus compared to five *Phoma* sp. isolates in cv. Ociai fushinari [62]. Identically, *Pe. simplicissimum* GP17-2 and *F. equiseti* 19–1 demonstrated sufficient growth-promoting effects on different host plants [4, 9, 60], but did not have effect on *Lotus japonicas* [89]. The outcome of the plant-PGPF interaction, therefore, depends on the plant and PGPF species. It is likely that the specific interaction develops during long-term co-evolution, as it has been observed for compatible and incompatible interactions of pathogens with plants [90]. Moreover, certain components of root exudates may attract and interact microbe specifically and allow it colonize the roots.

7. Mechanisms of plant growth promotion

The course of plant growth promotion by PGPF is complex and often cannot be attributed to a single mechanism. Various mechanisms that are known to modulate plant growth and development can be either direct or indirect. Direct growth promotion occurs when substances produced by the fungi or nutrient available by them facilitate plant growth. On the other hand, the ability of fungi to suppress plant pathogens and to ameliorate stress are considered major indirect mechanisms of plant growth promotion by PGPF. A particular PGPF may affect growth and development of plants using one or more of these mechanisms (Table 3).

7.1 Phosphate solubilization

Phosphorus is the second most important and frequently limiting macronutrient for plant growth and productivity. It is an important component of the key macromolecules in living cells and thereby, required for wide array of functions necessary for the survival and growth of living organisms. Despite the abundance of phosphorus in agricultural soils, the majority occurs in an insoluble form. Phosphorus forms complex compounds by reacting with iron, aluminum or calcium depending on the soil types and becomes insoluble and unavailable to plants [102]. To circumvent this problem, phosphate-solubilizing PGPF can play an important role dissolving insoluble P into the soluble form and making it available for plants. PGPF produce

Mechanisms	Specific activities	PGPF strain	References
Phosphate solubilization	Solubilized P by acid phosphatase and alkaline phosphatase	<i>F. verticillioides</i> RK01, <i>Humicola</i> sp. KNU01	[30]
	Solubilized P from rock phosphate and Ca-P by organic acid	<i>A. niger</i> 1B and 6A	[56]
	Solubilize P from tricalcium phosphate (TCP)	<i>A. niger</i> BHUAS01, <i>Pe. citrinum</i> BHUPC01, <i>T. harzianum</i>	[16]
	Solubilized P by organic acid activities	<i>Pe. oxalicum</i> NJDL-03, <i>Aspergillus niger</i> NJDL-12	[91]
	Phytase-mediated improvement in phytate phosphorus	<i>A. niger</i> NCIM	[76]
	Increased HCO ₃ -extractable P (23% increase)	<i>Pe. bilaiae</i> RS7B-SD1	[92]
Mineralization of organic substrate	Increased production of NH ₄ -N and NO ₂ -N in soil	<i>T. harzianum</i> GT2-1, <i>T. harzianum</i> GT3-1	[4]
	Increased availability of ammonium nitrogen from barley grain	<i>Phoma</i> sp.GS6-1, GS6-2, GS7-3, GS7-4, GS8-6, GS10-1, GS10-2, sterile fungus GU23-3	[87]
	Solubilize minerals such as MnO ₂ and metallic zinc	<i>T. harzianum</i> Rifai 1295-22	[93]
	Increased availability of ammonium nitrogen from barley grain	<i>Phoma</i> sp. GS8-1, GS8-2, GS8-3, Sterile fungus GU21-1	[62]
	Increased concentration of Cu, P, Fe, Zn, Mn and Na in roots Increased concentration of Zn, P and Mn in shoot	<i>T. harzianum</i> strain T-203	[47]
	Increased soil organic carbon, N, P and K content	<i>T. viride</i>	[73]
	Increased availability of macro and micronutrients and organic carbon	<i>T. harzianum</i> strain Th 37	[94]
Phytohormone and enzyme production	Auxin-related compounds (indole-3-acetic acid, IAA)	<i>T. virens</i> Gu. 29-8	[48]
	Gibberellins (GA1 and GA4) production	<i>A. fumigatus</i> HK-5-2	[65]
	GAs production	<i>Pe. resedanum</i> LK6	[39]
	GAs production	<i>Penicillium</i> sp. Sj-2-2	[38]
	GAs production	<i>Cladosporium</i> sp.MH-6	[24]
	GAs production	<i>Pe. citrinum</i> IR-3-3	[37]
	GAs and IAA production	<i>Chaetomium globosum</i> CAC-1G	[23]
	GAs production	<i>Exophiala</i> sp. LHL08	[26]
	GAs production	<i>Phoma herbarum</i> TK-2-4	[41]
	GAs production	<i>A. fumigatus</i> HK-5-2	[65]
	GAs production	<i>A. fumigatus</i> LH02	[18]
	IAA production	<i>T. harzianum</i> T-22	[64]
	Zeatin (Ze), IAA, 1-aminocyclopropane-1-carboxylic acid (ACC)	<i>T. harzianum</i>	[95]

Mechanisms	Specific activities	PGPF strain	References
Suppression of deleterious pathogens	Suppressed damping off caused by <i>Pythium irregular</i> , <i>Pythium</i> sp., <i>Pythium parvoecandrum</i> , <i>Pythium aphanidermatum</i> and <i>Rhizoctonia solani</i> AG4	Sterile fungus GSP102, <i>T. harzianum</i> GT3-2, <i>F. equiseti</i> GF19-1, <i>Pe. simplicissimum</i> GP17-2	[4]
	Induced systemic resistance against <i>Colletotrichum gramimicola</i>	<i>T. harzianum</i> T22	[78]
	Suppressed bacterial wilt disease caused by <i>Ralstonia solanacearum</i>	<i>T. harzianum</i> TriH_JSB27, <i>Phoma multivirata</i> PhoM_JSB17, <i>T. harzianum</i> TriH_JSB36, <i>Pe. chrysogenum</i> PenC_JSB41	[34]
	Suppressed Fusarium wilt caused by <i>Fusarium oxysporum</i> f. sp. <i>ciceris</i>	<i>T. harzianum</i> T-75	[58]
	Suppressed <i>Fusarium graminearum</i>	<i>Sphaerodes mycoparasitica</i>	[12]
	Suppressed damping off caused by <i>Rhizoctonia solani</i> AG4	<i>Pe. viridicatum</i> GP15-1	[35]
	Suppressed nematodes <i>Pratylenchus goodeyi</i> and <i>Helicotylenchus multicinctus</i>	<i>F. oxysporum</i> V5W2, Eny 7.11o and Emb 2.4o	[29]
	Suppressed seedling mortality by <i>Rhizoctonia solani</i>	<i>T. harzianum</i> isolate T-3	[83]
Amelioration of abiotic stress	Increased tolerance to salt stress	<i>T. harzianum</i> T-22	[69]
	Mitigation of oxidative stress due to NaOCl and cold stress	<i>T. harzianum</i> Rifai strain 1295-22	[96]
	Enhanced maize seedling copper stress tolerance	<i>Chaetomium globosum</i>	[97]
	Minimized Cu-induced electrolytic leakage and lipid peroxidation	<i>Pe. funiculosum</i> LHL06	[98]
	Increased tolerance to drought stress	<i>T. atroviride</i> ID20G	[99]
Volatile organic compounds (VOCs)	Produced abundant classes of VOCs (sesquiterpenes and diterpenes)	<i>F. oxysporum</i> NRRL 26379, NRRL 38335	[28]
	Produced mainly terpenoid-like volatiles including β -caryophyllene	<i>Talaromyces wortmannii</i> FS2	[40]
	Produced 2-methyl-propanol and 3-methyl-butanol	<i>Phoma</i> sp. GS8-3	[100]
	Produced abundant amount of isobutyl alcohol, isopentyl alcohol, and 3-methylbutanal	<i>T. viride</i>	[101]

Table 3. Different mechanisms of plant growth promotion used by various plant growth promoting fungi (PGPF).

phosphate-solubilizing enzymes such as phytases and phosphatases and organic acids, which liberate P from insoluble phosphates. The most efficient phytase and phosphatase producing PGPF belong to the genera *Aspergillus*, *Trichoderma*, and *Penicillium* [103]. The order in terms of phytate hydrolysis efficacy was *Aspergillus* > *Penicillium* > *Trichoderma* [104]. *Fusarium verticillioides* RK01 and *Humicola* sp. KNU01 solubilized phosphate by increasing activities of acid phosphatase and alkaline phosphatase, and promoted soybean growth significantly [30]. The phosphate solubilizing fungi possess greater phosphorus solubilization ability than bacteria,

especially under acidic soil conditions [105]. The main reason is most fungi are eosinophilic, and have relatively higher growth in acidic environments than bacteria [106]. The acidity has significant influence on organic acid-mediated phosphate solubilizing activities of *Pe. oxalicum* NJDL-03 and *A. niger* NJDL-12 [91]. However, acidification is not always the major mechanism of P solubilization by *T. harzianum* Rifai 1295-22 (T-22), where pH of cultures never fell below 5.0 and no organic acids were detected [93]. Some of the reported PGPF such as *Aspergillus niger* has twin abilities of P mineralization and solubilization [104]. The fungus releases P both from organic and inorganic sources. These suggests that specific PGPF may have specific activity in solubilizing phosphate and making it available for crop growth.

7.2 Substrate degradation (mineralization)

Microorganisms primarily mediate soil nutrient pathways. Microbial mineralization of nutrients from organic matter is crucial for plant growth. Some PGPF promote plant growth, but do not produce plant hormones or solubilize fixed phosphate. Among *Pe. radicum*, *Pe. bilaiae* (strain RS7B-SD1) and *Penicillium* sp. strain KC6-W2, the strongest growth promotion in wheat, medic, and lentil was shown by *Penicillium* sp. KC6-W2, while the only significant P increase (~23% increase) was found in *Pe. bilaiae* RS7B-SD1-treated plants [92]. Similarly, seven *Trichoderma* isolates significantly improved the growth of bean seedlings; despite some of them do not possess any of the assessed growth-promoting traits such as soluble P, indole acetic acid (IAA) and siderophores [107]. These PGPF are believed to encourage plant growth by accelerating mineralization in the soil. Fungi have better substrate assimilation efficiency than any other microbes and are able to break down complex polyaromatic compounds such lignin and humic or phenolic acids [108]. A close relationship was found between the cellulose and starch degradation activity of PGPF for decomposing barley grain and their subsequent growth promotion effect in plants [109]. Application of *T. harzianum* strain Th 37 increased the availability of macro and micronutrients and organic carbonate in the ratoon initiation stage in sugarcane [94]. Colonization of *T. harzianum* in cucumber roots enhanced the availability and uptake of nutrients by the plants [47]. Cucumber plants grew better and produced more marketable fruits due to an increase in soil nutrients caused by PGPF, and accumulated more inorganic minerals like Ca, Mg, and K in aerial shoots [62]. PGPF are also directly involved the degradation of the nitrogenous organic materials through ammonization and nitrification. Formation of $\text{NH}_4\text{-N}$ and $\text{NO}_2\text{-N}$ in soil was accelerated during soil amendment with PGPF-infested barley grains [109]. More interestingly, the fungal entomopathogen *Metarhizium robertsii*, when established as a root endophyte, was shown to translocate nitrogen from a dead insect to a common bean plant host, suggesting this PGPF's potential to acquire mineral nutrients from organic matter and promote plant growth [54]. Nutrient release by mineralization could explain why PGPF other than mycorrhizae improve plant growth when added to soil.

7.3 Phytohormone production

Phytohormones are involved in many forms of plant-microbe interactions and also in the beneficial interactions of plants with PGPF. The commonly recognized classes of phytohormones produced by PGPF are the auxins (IAA) and gibberellins (GAs) (Table 3). IAA, the most studied auxin, regulates many aspects of plant growth, in particular, root morphology by inhibiting root elongation, increasing lateral root production, and inducing adventitious roots [48]. The *T. harzianum* T-22-mediated root biomass production and root hair development in maize is

believed to operate through a classical IAA response pathway [78]. Similarly, a direct correlation exists between increased levels of fungal IAA and lateral root development in *Arabidopsis* seedlings inoculated with *T. virens* [48].

GAs are well known for their role in various developmental processes in plants, including stem elongation. Shoot elongation of waito-c rice seedlings by culture filtrates of *Pe. citrinum* IR-3-3 and *A. clavatus* Y2H0002 was attributed to the activity of physiologically active GAs existing in the culture filtrates [19, 37]. Biochemical analyses of *Penicillium* sp. LWL3 and *Pe. glomerata* LWL2 culture filtrates that enhanced the growth of Dongjiin beyo rice cultivar and in GA-deficient mutant *Waito-C* revealed the presence of IAA and various GAs [110]. Similarly, production of bioactive GAs correlated with enhanced growth of *Waito-C* under salinity by *Penicillium* sp. Sj-2-2 [38]. GA also played key roles during root colonization by *P. indica* in pea roots [111].

Another phytohormone through which PGPF mediate plant growth is cytokinin, especially the Zeatin. Zeatin production has been documented in *Piriformospora indica*, *T. harzianum* and *Phoma* sp., and the fungi that also produce other phytohormones [95, 112, 113]. *P. indica* produces low amounts of auxins, but high levels of cytokinins. *Trans-Zeatin* cytokinin biosynthesis was found crucial for *P. indica*-mediated growth stimulation in *Arabidopsis* [112]. This evidence suggests that PGPF often mediate the various growth and developmental processes in plants by influencing the balance of various plant hormones.

7.4 Microbial ACC deaminase

PGPF produces a crucial enzyme ACC (1-aminocyclopropane-1-carboxylic acid) deaminase. ACC deaminase cleaves the ethylene precursor, 1-aminocyclopropane-1-carboxylic acid (ACC), into NH₃ (ammonia) and α -ketobutyrate [114]. The ACC deaminase regulates the plant growth by cleaving ACC produced by plants and thereby minimizing the ethylene level in the plant, which when present in high concentrations can lead to a reduced plant growth [115]. ACC deaminase is an inducible enzyme encoded by *acdS* genes of fungi and bacteria [116]. ACC deaminase appears to be central to the functional interactions of some plant-PGPF. *T. asperellum* T203 produced high levels of ACC deaminase and showed an average 3.5-fold induction of the *acdS* gene [117]. When ACC deaminase expression is impaired in the fungus *T. asperellum* T203, the plant growth promotion abilities of this organism are also decreased [51]. The root colonizing bacteria *T. harzianum* T22 no longer promote canola root elongation after its *acdS* gene is knocked out [64]. Production of ACC deaminase was reported in some other fungi, which include *Issatchenkia occidentalis* [118], and *Penicillium citrinum* and a stramnopile, *Phytophthora sojae* [119, 120]. The ACC deaminase-producing microbes have competitive advantages in the rhizosphere over nonproducing microorganisms because the enzyme acts as a nitrogen source for them [116]. Moreover, bacteria and fungi that express ACC deaminase can lower the impact of a range of different stresses that affect plant growth and development [114]. These show that ACC deaminase is not only related to plant growth promotion abilities of the microbes, but also play additional roles in the rhizosphere.

7.5 Suppression of deleterious microorganisms by PGPF

The key indirect mechanism of PGPF-mediated plant growth promotion is through their activities as biocontrol agents. PGPF protect and empower plants to resist harmful pathogens and ensure their better growth. The mechanisms by which PGPF suppress growth or activity of invading pathogens in crop plants

include antibiosis, competition for nutrient and space, mycoparasitism and induced systemic resistance (ISR) [121]. PGPF of diverse genera promoted growth of field-soil grown cucumber by counteracting damping off pathogen *Pythium* sp. through microbial antagonism [4]. Banana plants inoculated with PGPF *F. oxysporum* significantly suppressed nematode pathogens *Pratylenchus goodeyi* and *Helicotylenchus multincinctus* resulting in up to ~20 to 36% increase in banana yields [29]. The mycoparasite *Sphaerodes retispora* has been reported to improve the plant dry weight and to decrease plant mortality in the presence of *F. oxysporum* [122]. Similarly, under phytotron conditions, seed germination, root biomass, total biomass, root length, and total length of *F. graminearum*-infected wheat were noticeably increased with the treatments of *S. mycoparasitica* and *T. harzianum*, as compared to inoculation with *F. graminearum* alone. Both mycoparasites prevented colonization and reduction in root growth by the pathogen [12]. PGPF compete with the pathogen for colonization niche on roots [79]. Other mechanisms of disease suppression by PGPF are, therefore, likely to include competition with pathogens for infection sites on the root surface. Moreover, there is a long and growing list of PGPF such as *Trichoderma*, *Penicillium*, *Fusarium*, *Phoma*, and non-sporulating fungi, which can protect crop plants against pathogens by eliciting ISR [14, 31, 123, 124]. Although many fungal strains to act as PGPF and elicit ISR, it is not clear how far both mechanisms are connected. These microbes may use some of the same mechanisms to promote plant growth and control plant pathogens.

7.6 Rhizoremediation and stress control

The microbial association of plants has a major influence on plant adaptation to abiotic stresses such as salinity, drought, heavy metal toxicity, extreme temperatures and oxidative stress. Recent studies indicate that fitness benefits conferred by certain PGPF contribute plant adaptation to stresses [125]. There are reports of enhanced plant growth because of the association of PGPF with plants, even when plants are under suboptimal conditions [126]. Root colonization by *T. atroviride* ID20G increased fresh and dry weight of maize roots under drought stress [99]. Supplementation of *T. harzianum* to NaCl treated mustard seedlings showed elevation by 13.8, 11.8, and 16.7% in shoot, root length and plant dry weight, respectively as compared to plants treated with NaCl (200 mM) alone [127]. The fungus *Pe. funiculosum* significantly increased the plant biomass, root physiology and nutrients uptake to soybean under copper stress [98]. These fungi have been known to produce plant growth regulators (like GAs and auxins) and extend plant tolerance to abiotic and biotic stresses [23, 125]. Recurrently, *T. harzianum* T22 has little effect upon seedling performance in tomato, however, under stress; treated seeds germinate consistently faster and more uniformly than untreated seeds [69]. A few other fungi like *Microsphaeropsis*, *Mucor*, *Phoma*, *Alternaria*, *Peyronellaea*, *Steganosporium*, and *Aspergillus* are known to grow well in polluted medium and protect plants from adverse effects of metal stress [128]. There are numerous similar examples of PGPF ameliorating abiotic stresses and promoting plant growth. Despite significant differences between different stresses, cellular responses to them share common features. Enhanced resistance of PGPF-treated plants to abiotic stresses is explained partly due to higher capacity to scavenge ROS and recycle oxidized ascorbate and glutathione [99, 127]. The increase in proline content is found to be very useful in providing tolerance to these plants under stress [129]. Both enzymatic (peroxidase, catalase, superoxide dismutase, ascorbate peroxidase, monodehydroascorbate reductase, dehydroascorbate reductase, glutathione reductase, glutathione S-transferase and guaiacol peroxidase), and non-enzymatic (ascorbic acid, reduced glutathione, oxidized glutathione) antioxidants are induced by PGPF further

enhance the synthesis of these phytoconstituents and defend the plants from further damage [127].

7.7 Production of volatile organic compounds (VOCs)

Microorganisms produce various mixtures of gas-phase, carbon-based compounds called volatile organic compounds (VOCs) as part of their normal metabolism. The comparative analysis of experimental data has shown that volatile metabolites make a much greater contribution to the microbial interactions than non-volatile ones [130]. Recent studies reveal that VOC emission is indeed a common property of a wide variety of soil fungi, including PGPF. Some of these VOCs produced by PGPF exert stimulatory effects on plants. A PGPF, *Talaromyces wortmannii* emits a terpenoid-like volatile, β -caryophyllene, which significantly promoted plant growth and induced resistance in turnip [40]. The identified VOCs emitted by *Phoma* sp. GS8-3 belonged to C4-C8 hydrocarbons, where 2-methyl-propanol and 3-methyl-butanol formed the main components and promoted the growth of tobacco seedlings [100]. These two components were also extracted from PGPR [131]. On the other hand, 3-methyl-butanol has been reported from *T. viride* [101]. The other most abundant VOCs from *T. viride* were isobutyl alcohol, isopentyl alcohol, farnesene and geranylacetone. *Arabidopsis* cultured in petri plates in a shared atmosphere with *T. viride*, without direct physical contact was taller with more lateral roots, bigger with augmented total biomass (~45%) and earlier flowered with higher chlorophyll concentration (~58%) [101]. Moreover, volatile blends showed better growth promotion than individual compounds [132]. Volatile compounds produced by PGPF are also heavily involved in induce systemic resistance toward pathogens [100].

8. Pattern and process of root colonization by PGPF

Root colonization is considered as an important strategy of PGPF for plant growth promotion. Root colonization is the ability of a fungus to survive and proliferate along growing roots in the presence of the indigenous microflora over a considerable period [35]. The fungus that colonizes plant root effectively is more rhizosphere competent than others [107]. Rhizosphere competence is a necessary condition for a fungus to be an efficient PGPF. Re-isolation frequency of the fungus from the colonized roots is an indirect measure of its root colonizing ability and thereby, its rhizosphere competence. In such studies, *Pe. simplicissimum* GP17-2 and *Pe. viridicatum* GP15-1 were re-isolated from *Arabidopsis* Col-0 roots 3 weeks after planting at high frequencies which were found to be >90% (**Figure 2**). Similarly, the re-isolation frequency of *Pe. janthinellum* GP16-2 from the roots of Col-0 plants was recorded to be, on average, 85% [33]. *Aspergillus* spp. PPA1 was re-isolated from the roots of cucumber plants at a frequency of 95–100% 3 weeks after planting [17], indicating a rapid and efficient root colonization by the PGPF. However, a slow root colonization by PGPF was also reported, as it was the case with *Phoma* sp. GS8-2, which achieved maximum colonization on cucumber roots at 10 weeks [62]. The relative growth rate of the fungi and roots seems to determine the length of time required for maximum root colonization.

Some PGPF selectively colonize host roots and promote growth. Isolates of *Phoma* and sterile fungi showed poor ability to colonize the soybean roots and were unable to enhance the growth of soybean [79]. Similarly, *T. koningi* colonized roots and enhanced growth of *Lotus japonicas*, but *Pe. simplicissimum* and *F. equiseti* did not [89]. It was observed that *T. koningi* induced a transient and decreased level of defense gene expression in *L. japonicas* during its entry into the roots, while a



Figure 2.

Re-isolation of *Penicillium simplicissimum* GP17-2 and *Penicillium viridicatum* GP15-1 at higher frequencies from colonized roots of *Arabidopsis thaliana* ecotype Col-0 3 weeks after sowing.

stimulated expression of these genes was induced by *Pe. simplicissimum* and *F. equiseti* [89]. *T. koningi* resembles symbiotic fungi, while *Pe. simplicissimum* and *F. equiseti* act similar to fungal pathogens in activating host defense. This shows that legumes selectively avoid some PGPF and thus allow only specific PGPF to interfere.

There are also PGPF, in particular, the non-sporulating sterile fungi that lack root colonization ability, but they are able to promote growth and yield of plants [62, 133]. This indicates that root colonization is not an indispensable condition for growth promotion by all PGPF. Some chemical factor(s) produced by them might be responsible for growth promotion.

The colonization of the root system of by PGPF is not always homogenous; the density of PGPF varies in different parts of the root system. The colonization of roots by the majority of PGPF appears to be higher in the upper than in the middle and lower root parts of roots, [35, 133]. The lower part was always less colonized by PGPF, especially during first 2 weeks of colonization. This is probably due to the faster growth of the roots than of the hyphae. Moreover, the main zone of root exudation is located behind the apex [134]. However, some PGPF can keep up with root growth and colonize the entire root system [35]. Only fungi with large nutrient reserves can move to the root and along the root over larger distances [135].

Anatomical data show that PGPF may colonize root tissues internally and establish a mutualistic relationship with host. *F. equiseti* GF19-1 produced abundant hyphal growth on the root surface, formed appressoria-like structures and grew in the intercellular space, not inside the cell [31]. *T. harzianum* CECT 2413 exhibited profuse adhesion of hyphae to the tomato roots and colonized the epidermis and cortex. Intercellular hyphal growth and the formation of plant-induced papilla-like hyphal tips were also observed [136]. Hyphae of *T. koningi* penetrated the epidermis and entered the intercellular inner cortex tissues [89]. Sterile red fungus has been also demonstrated to invade the inner root regions that helped plants derive nutrients from the soil and protected roots from pathogens [137].

9. Formulation of PGPF

PGPF, especially *Trichoderma*, have many success stories as plant growth promoting agents and appear to have much potential as a commercial formulation. Different organic and inorganic carrier materials have been studied for effective delivery of bioinoculants. A talc-based formulation was developed for *T. harzianum* to supply concentrated conidial biomass of the fungus with high colony forming units (CFU) and long shelf life [138]. The concentrated formulation provided an extra advantage of smaller packaging for storage and transportation, and low

product cost as compared to other carriers such as charcoal, vermiculite, sawdust and cow dung. Seed application of the formulation recorded significant increase in growth promotion in chickpea [138]. Corn and sugarcane bagasse were used as potential carriers for *Trichoderma* sp. SL2 inoculants. The corn formulation of SL2 significantly enhanced rice seedlings root length, wet weight and biomass compared to inoculum mixed with sugarcane bagasse and control [139]. A spray-dried flowable powder formulation was developed for biostimulant *Trichoderma* strains using a CO₂ generating dispersant system, based on polyacrylic acid, citric acid and sodium bicarbonate, polyvinyl alcohol as adhesives and lecithin as wetting agent [140]. Hydrolytic amino acids derived from pig corpses were used in the preparation of *T. harzianum* T-E5-containing bioorganic fertilizer. The resulting bioorganic fertilizer supported higher densities of *T. harzianum* T-E5 and substantially enhanced plant growth when applied as a soil amendment [141]. A composted cattle manure-based *Trichoderma* biofertilizer was developed and tested in the field. Plots fertilized with biofertilizer had the greatest aboveground biomass of any treatment and were significantly more productive than non-amended plots and plots fertilized with any rate of organic fertilizer [142]. Effective formulation of *P. indica* was prepared in talcum powder or vermiculite with 20% moisture. The talcum-based formulations performed significantly better as bioinoculant over vermiculite-based formulations in glasshouse experiments [143]. These show the feasibility of commercial level production and applicability of different PGPF formulations for plant growth promotion in the field.

10. Conclusions

Because of current concerns over the adverse effects of agrochemicals, there is a growing interest in improving our understanding of the role and application of beneficial microbes in agriculture. The plant-associated growth promoting fungi show excellent potential for wider use in sustainable agriculture as they improve plant growth and yield in an ecofriendly and cost-effective manner. However, the PGPF continue to be greatly underutilized, primarily due to some practical problems such as the inconsistency in field performance, which appears to be the greatest challenge in the development of microbial inoculants for plant growth until now and well into the future. If our understanding of complex rhizosphere environment, of the mechanisms of action of PGPF and of the practical aspects of mass production, inoculant formulation and delivery increase, more PGPF products will become available. Knowledge of multiple microbial interaction with different or complementary mode of actions is also of extreme value for development of bio-formulation.

Recent advances in biotechnological tools and reliable transformation system could be useful in engineering of the PGPF to confer improved benefits to the crop. Genetic transformation and overexpression of one or more of the plant growth promoting traits that act synergistically may lead to enhanced performance by the inoculant. Research may be required periodically in order to evaluate the genetic stability and ecological persistence of the genetically modified strain. Efforts should be strengthened to foster linkage between investigators and entrepreneurs in facilitating technology transfer, promotion and acceptance by end users.

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Section 4

Plant Protection

Biological Efficacy of *Trichoderma* spp. and *Bacillus* spp. in the Management of Plant Diseases

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Abstract

This chapter will cover topics about the microbial antagonists *Trichoderma* spp. and *Bacillus* spp. from the perspective of use as potential biological control agents on plant diseases. Results obtained in the laboratory about from their isolation, microbial strain collections for both genera, taxonomic identification, antifungal activity in in vitro tests, obtained evaluation of the antifungal effect of secondary metabolites from microbial antagonists will be shown. Besides, results obtained from bioassays in the greenhouse and field are used as biopesticides in the control of diseases in fruit trees and vegetables and their effects on the promotion of plant growth and increased crop yield.

Keywords: inhibition, disease, plant pathogen, incidence, severity, antagonist microorganisms

1. Introduction

The agricultural production systems are generally based on technological dependence of high-yield varieties and the use of agrochemicals, causing an imbalance in different agroecosystems. The crops had turned to be susceptible to plague organisms to which before they were not and have proliferated because their natural enemies have eliminated the selection pressure has been favored by the monoculture or by the excessive use of plaguicides, what has conducted to the rupture of resistance of host and resistance toward the pesticides.

Those, as mentioned above, do rethink the actual technological management of the crops, researching less aggressive options with low or without environmental impact. This focus allows the searching of microbial alternatives as biological control agents of diseases, as a viable option to reduce their impact, enhancing the yield and quality of the agricultural products. The economic production losses caused by pests and diseases worldwide are estimated to be 36.5% on average, where 14.1% is caused by diseases, 10.2% by insects, and 12.2% by weeds, without considering the 6–12% of agricultural products postharvest losses. Although it estimated that in developing countries, these could reach up to 50% of production losses, considering only the disease, it estimates that annual losses worldwide can reach about 220 billion dollars. Overall, the diseases from plants can destroy crops before and after harvest or yield partial losses and cause loss of quality in the products harvested.

For example, the apple scab caused by *Venturia inaequalis* (Cook) Wint. (Anamorph: *Spilocaea did Fr.*) is the most important disease of this fruit at a worldwide level, which can cause significant economic losses until 100% of the production, affecting the commercial quality of fruits [1, 2]. Generally, its control is based on the use of agrochemicals. In vegetables, wilting of chili pepper and tomato crops is one of the main biological limitations in the production of these crops and can be caused by *Phytophthora capsici*, *Rhizoctonia solani*, and *Fusarium oxysporum* [3]; this disease is reported throughout Mexico, estimating losses of up to 80% due to root rot by invading the vascular system of plants. Likewise, chemical control is the most used method for disease management and is common to reduce the inoculum by disinfecting the soil with metam sodium, 2-thiocyanomethyl benzothiazole (TCMTB), metalaxyl, azoxystrobin, and propanocarp fungicide applications to control *P. capsici* [4]. *R. solani* and *Fusarium* spp. are controlled with tebuconazole, carbendazim, thiabendazole, and methyl thiophanate [5]. The use of this control method significantly increases the production costs and the negative impact it causes on the environment and to human health and induces resistance of the pathogens toward the active ingredients. An alternative is the use of biological control by microorganisms antagonistic to fungi and stramenopiles from the soil, which has little or no effect on the environment and human health.

2. Biopesticides market

The worldwide market of biopesticides was of 1213 million dollars in 2010 and 3222 million dollars in 2017; the annual rate increases to 15.8% since 2012 besides 2017. Within this market, bioinsecticides represented 46% in 2011, and biofungicides were of 600.5 million dollars, reaching 1447 million in 2017. The annual rate from 2012 to 2017 grows up at 16.1%. Given that there currently exists a market demand for free products of pesticide waste, huge agrochemical companies are in the market of bioproducts, acquiring biocontrol companies and developing new biotechnological products.

3. Isolation and identification of *Bacillus* spp. and *Trichoderma* spp.

Trichoderma and *Bacillus* are essential genera of antagonistic microorganisms for control of a large number of phytopathogens. *Trichoderma* is a cosmopolitan soil fungus, which is frequently on soil from the plant root system. This fungus is attractive for organic management of diseases because present different action modes against phytopathogens as competition for nutrients, mycoparasitism, and antibiosis by hydrolytic enzymes and metabolites also produce substances that promote plant growth [6, 7]. On the other hand, *Bacillus* spp. is a large and heterogeneous group of Gram-positive, rod-shaped, aerobic and facultative anaerobic, and endospore-forming bacteria; same as *Trichoderma*, *Bacillus* is an alternative of biological control of plant diseases due to its capability to inhibit phytopathogens and growth promotion in plants [8, 9].

Due to the abovementioned and because there is a large number of species from both microorganisms, their isolation and identification for their possible commercial use are necessary; some of the species of *Trichoderma* are *T. virens*, *T. harzianum*, and *T. viride* and of *Bacillus* spp. are reported as antagonists *B. amyloliquefaciens*, *B. licheniformis*, *B. subtilis*, and *B. pumilus* [10, 11]. Thus a correct identification of the species which needs work is necessary.

3.1 Isolation and identification of *Trichoderma* spp.

3.1.1 Isolation and morphological identification of *Trichoderma* spp.

The first step for correct identification of antagonist microorganisms depends on isolation. In the case of *Trichoderma* spp., they are present in a great variety of agricultural and natural soils. The soil sampling for its isolation is relatively simple; using a shovel at 10–20 cm depth, 500 g of soil is taken and deposited in plastic bags; after, the samples will be moved to a laboratory and placed on storage at 4°C until used. Purification of *Trichoderma* spp. it is essential on investigations and present many ways or techniques; nevertheless, the monosporic culture is suggested by *Trichoderma* on culture media as potato dextrose agar (PDA) or *Trichoderma* Specific Medium (TSM), and incubated at $28 \pm 2^\circ\text{C}$ for 96 h [11–13]. Once the monosporic culture is obtained, the identification of *Trichoderma* species can be realized using taxonomic keys through its morphological features or with molecular biology, extracting DNA and utilizing general or specific primers. In case of the use of taxonomic keys, structures as width and length of phialide, length and width of conidia, and presence of chlamydospores will be observed [7, 14].

3.1.2 Molecular identification

This kind of identification has gained acceptance because it presents more precision and reliability among several strains of *Trichoderma*. The phylogeny of this genus has been based in the sequence analysis of the internal transcribed spacers of ribosomal DNA using the universal primers ITS1 and ITS4 with a subsequent sequencing and analysis through databases [15] but also can be identified through specific primers which are a powerful tool that allows to identify a specific species of *Trichoderma* [16] (**Table 1**). In Mexico, diverse species of *Trichoderma* have been isolated and identified [6]; they identified *T. atroviride*, *T. asperellum*, *T. citrinoviride*, *T. ghanense*, *T. harzianum*, *T. inhamatum*, *T. longibrachiatum*, and *T. yunnanense* (**Figure 1**) from samples taken from several agricultural regions. In a similar research, Osorio et al. [23] identified the species as *T. asperellum*, *T. rossicum*, and *T. hamatum* from different localities of Mexican Northeast region.

Species specificity	Primer sequences (5'–3')	Product size (bp)	References
<i>T. harzianum</i>	HAR-1.6F: GTACCTCGCGAATGCATCTA HAR-1.6R: GGCTATGACCATGATTACGC	1600	[17]
<i>T. asperellum</i>	T2AF: CTCTGCCGTTGACTGTGAACG T2AR: CGATAGTGGGGTTGCCGTCAA	507	[18]
<i>T. virens</i>	TvCTT56f: CTTGATGACAAGCCAAAAGG TvCTT56r: GAAGAGAGGACATAGGGTCTGG	289	[19]
<i>T. atroviride</i>	Q01_4F: GCACACCAACTGCTGGAGCTT Q01_4R: CACGCTGACAATGACCGACAC	1017	[20]
<i>T. aggressivum</i>	Th-F: CGGTGACATCTGAAAAGTCGTG Th-R: TGTCACCCGTTCCGATCATCCG	444	[21]
<i>T. pleuroti</i>	FPforw1: CACATTC AATTGTGCCCGACGA PSrev1: GCGACACAGAGCACGTTGAATC	218	[22]

Table 1.
 Examples of species-specific primers for *Trichoderma* spp.

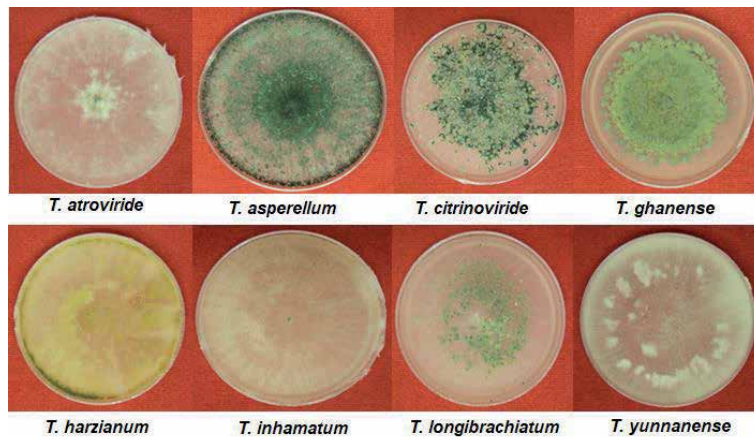


Figure 1.

Morphologic characteristics of different *Trichoderma* spp. isolated from samples of different agricultural systems of Mexico.

3.2 Isolation and identification of *Bacillus* spp.

Bacillus spp. is a genus present in the soil of a considerable amount of crops and naturally is on the rhizosphere; due to this, the traditional tools for determining the soil bacterial community and diversity are used [24]. The first step is to make the collecting of the rhizosphere soil, take 10 g of soil with a sterile spoon, and store the sample at 4°C. Heat or pasteurization treatments are the most commonly used techniques to select spores due to this, the sample is diluted in 90 mL of sterile normal saline and heated at 80°C for 10 min to eliminate vegetative cells; once heated, the sample is serially diluted (10^{-1} – 10^{-4}) and placed on 1 mL nutrient agar (NA) medium with cycloheximide (100 mg mL^{-1}) to prevent fungal growth or carboxymethylcellulose (CMC) agar, and it is incubated at 37°C for 24 h [25, 26].

However, the treatment with heat can be different depending on the species because endospores of some strains are more resistant to heat than others [24]. Due to this, the drying treatment is considered more gentle; this method consists of placing the samples on a dryer at 70°C for 1 h [25]. The considerable variety of physiology of *Bacillus* spp. requires elaborate biochemical and morphological tests for species identification; as colony growth in artificial media, form cell unit, presence, number and orientation of flagella, Gram stain, spore form-position and specific environmental conditions of growth and finally the specific use of carbon sources gave its metabolic diversity [27].

3.2.1 Molecular identification of *Bacillus* spp.

Several molecular approaches are currently utilized for the identification of microorganisms; in this sense, the use of polymerase chain reaction (PCR) in combination with 16S rRNA is a tool frequently used for identification of *Bacillus* spp. from various environments including soil. Using the 16S rRNA sequence, five groups within biological control of root pathogens by plant growth-promoting *Bacillus* spp., the genus *Bacillus* spp., where group 1 comprises species *B. amyloliquefaciens*, *B. subtilis*, *B. pumilus*, and *B. licheniformis*, have been identified [24, 25].

Bacillus spp. can identify through specific primers (Table 2). Such as *Trichoderma* spp., the Mexican agricultural systems are an excellent source to obtain *Bacillus* spp., such as mentioned by Guillén-Cruz et al. [9] and Hernandez-Castillo

Species specificity	Primer sequences (5'-3')	Product size (bp)	References
<i>B. subtilis</i>	p-gyrA-f: CAGTCAGGAAATGCGTACGTCCTT p-gyrA-r: CAAGGTAATGCTCCAGGCATTGCT	741	[28]
<i>B. amyloliquefaciens</i>	trpE(G) F: TTTGAATCCGAGCCCTTATG trpE(G) R: ACATACATTTTCGGGGGATGA	78	[29]
<i>B. pumilus</i>	GC-U968(F): GCAACGCGAAGAACCCTTAC L1401(R): GCGTGTGTACAAGACCC	490	[30]
<i>B. licheniformis</i>	BL8AF: TCACAACCCGTTGACGACAA BL8AR: CGTGTCCGAGTGTGCGTTATAT	247	[31]

Table 2.
 Examples of species-specific primers for *Bacillus* spp.

et al. [32] whom identified several *Bacillus* spp. species as *B. amyloliquefaciens*, *B. pumilus*, *B. licheniformis*, and *B. subtilis* from samples coming from several regions of the center and northern of Mexico.

4. Antifungal activity in vitro of *Trichoderma* spp. and *Bacillus* spp.

The antifungal activity of *Trichoderma* species has been evaluated in in vitro studies against soilborne and foliar fungi, and there have been acceptable results. The antifungal activity can be determined such a direct manner as indirect manner. In the case of a direct manner, the most used technique is the dual culture where the inhibition percentage, Bell scale, and the days to contact are evaluated to determine the antagonistic activity of *Trichoderma* species. Dual culture consists of Petri dishes with PDA where a disk (5 mm in diameter) with mycelium of the plant pathogen is placed and, on the other side of the Petri dish equidistantly, a disk of mycelium of the same diameter of *Trichoderma* strains under study is placed. The plates inoculated are incubated at $27 \pm 1^\circ\text{C}$ until the growth of control treatment (with only plant pathogen disk) covered the Petri dish. The effect of *Trichoderma* strains on plant pathogens is determined by the percentage of mycelial growth inhibition. The days of contact between plant pathogen antagonistic and antagonistic ability of *Trichoderma* isolates according to the methodology proposed by Bell et al. [33] are also determined. Bell et al. [33] classified the antagonism produced by *Trichoderma* as follows: Class I, *Trichoderma* overgrows completely to pathogen and covers the whole surface of the medium; Class II, *Trichoderma* overgrows two-thirds of the surface of the medium; Class III, *Trichoderma* and pathogen colonized each half of the surface, and nobody seems to dominate the other; Class IV, the pathogen colonizes the two-third parts of the media surface and resists invasion by *Trichoderma*; and Class V, the plant pathogen overgrows completely to *Trichoderma* and covers an area total culture media [6]. In case of the desire to determine the antifungal activity of an indirect manner, the volatile compounds are an option; this method is realized as follows. In the center of a Petri dish having only PDA medium, a disk of 5 mm in diameter with active mycelia of the plant pathogen is placed, and the top of the dish is replaced with another Petri dish in which a disk with mycelia of *Trichoderma* strain is placed; in this case, the lid is pierced with a punch (10 mm in diameter), and the Petri dishes are joined and sealed with parafilm paper and incubated at $26 \pm 1^\circ\text{C}$ until each pathogen control covered the Petri dish. The effect of volatile compounds is measured considering the diameter of pathogen colonies and expressed as percentage of mycelial growth inhibition [6].

Several research were carried out to determine the antifungal activity of *Trichoderma* spp. strains, due to its potential as biocontroller of plant pathogens,

as reported by Hernandez et al. [6] who evaluated several strains of *Trichoderma* spp. against *Sclerotinia sclerotiorum* and *Sclerotium cepivorum* through dual culture and observed rate of inhibition of 45–63.8% and 50.9–81.5 for *S. sclerotiorum* with *T. ghanense* and *T. longibrachiatum*, respectively, and 81.5 and 81.2% of *S. cepivorum* with *T. inhamatum* and *T. asperellum*, respectively. For the Bell scale and contact days for both phytopathogenic fungi, the mean was of 2 days and scale of I, II, and III with all the *Trichoderma* spp. strains. Some research about the inhibition of secondary metabolites, precisely the volatile compounds, present inhibition of *S. sclerotiorum* and *S. cepivorum* against *T. longibrachiatum* with 28.1 and 73.8%, respectively, followed by *T. harzianum* with 12.5 and 62.5%.

Some studies by Osorio et al. [23] mentioned a *Trichoderma* spp. as controller. They reported the overgrowth of *Trichoderma* spp. strains over *Phytophthora capsici*; in total 13 *Trichoderma* spp. strains showed level 1 according to the Bell scale. This effect can be attributed to enzyme production (β -1, 3-glucanase, chitinase, protease, and cellulose) by these *Trichoderma* spp. strains. As too volatile compounds produced by the *Trichoderma* spp. strains, they reported inhibition of *P. capsici* mycelial growth ranged between 4.3 and 48.8, the major effect observed with the *T. asperellum* and the least with *Trichoderma* sp. strain. Some studies mentioned that *Trichoderma* spp. produces volatile compounds, carbon dioxide, oxygen, ethylene, and 6-pentyl- α -pyrone (6PP) which cause adverse effect in the development of phytopathogenic fungus. Furthermore, *Trichoderma* spp. can inhibit the growth of foliar fungus as *Colletotrichum* spp., such as mentioned by Tucuch et al. [34] who determined the antifungal activity *Trichoderma* spp. strains and reported antagonism level 1 in Bell scale by *T. asperellum*, while for volatile compounds, the species *T. lignorum* affected the fungus inhibiting its developments in 24.02% (Figure 2).

Usually, *Trichoderma* spp. inhibit several phytopathogenic fungi due to their capacity to produce enzymes, volatile compounds and compete for nutrients against phytopathogens. Studies realized by Samaniego-Fernández et al. [35] and Kumar et al. [7] showed the antagonistic capacity of *Trichoderma* spp.; in the first case, the species *T. harzianum* and *T. viride* are controlled to *S. rolfsii* y *Fusarium* spp.; in the second study, several *Trichoderma* spp. showed mycelial growth inhibition of *S. rolfsii* more than 50%.

Likewise, then *Trichoderma* spp. strains, the antifungal activity of *Bacillus* spp., can be tested by dual culture; nevertheless, the method is different than with *Trichoderma* spp. PDA disk (5 mm) with active mycelium of the phytopathogen is placed in the center of a Petri dish with PDA; on the same plate, at a distance of 1.5 cm in the four cardinal points, a loopful of antagonistic bacterial isolates is placed. Plates inoculated with the pathogen culture serve as controls. In order to quantify the antagonistic potential of bacterial strains, the size of growth inhibition zones measured after 6 days of incubation at 25–28°C and the percent of radial growth inhibition (PICR) are calculated [36]. In this sense, study also showed the capacity of *Bacillus* spp. to inhibit the growth of phytopathogenic fungi. Thereby

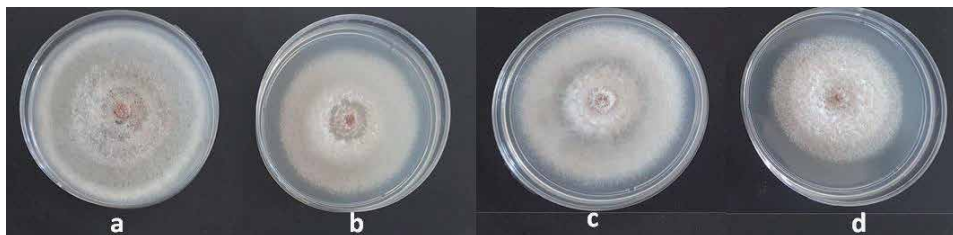


Figure 2. Inhibition of *Colletotrichum* spp. by volatile compounds produced by different *Trichoderma* spp. strains (a) control, (b) *T. asperellum*, (c) *T. yunnanense*, and (d) *T. lignorum*.

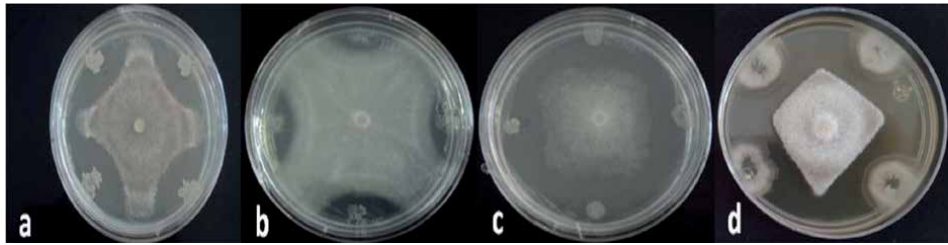


Figure 3. Antagonistic effect of *Bacillus* spp. strains against different phytopathogenic fungus (a) *Rhizoctonia solani*, (b) *Fusarium oxysporum*, (c) *Phytophthora capsici*, and (d) *Colletotrichum* spp.

Jimenez et al. [36] reported the inhibition of *Venturia inaequalis* mycelia by *B. subtilis* and *B. licheniformis* ranged 33.4–41.3%, respectively, and Tucuch et al. [34] observed 50% of inhibition from *B. subtilis* against *Colletotrichum* spp. (Figure 3).

5. Obtaining secondary metabolites of *Trichoderma* spp. and *Bacillus* spp. and their antifungal activity in vitro (PDA methods and microplate dilution)

Generally, the production of the secondary metabolites from biological agents such as *Trichoderma* spp. and *Bacillus* spp. is carried out using liquid media by a fermentation process in a reactor, which can be of different types from a simple bottle to until an automated reactor, where the temperature and shaking rate are the key variables for the emission of secondary metabolites. The liquid medium is integrated by several components such as carbon sources and mineral salts, where the biological microorganism is inoculated [37]. The secondary metabolites obtained from the fermentation of *Bacillus* spp. and *Trichoderma* spp. are filtered with nitrocellulose membrane of 0.22 μm ; after the recovery of these metabolites, it is necessary to perform a screening to determine their ability to inhibit phytopathogenic microorganisms. There are many methods to determine the antifungal activity from secondary metabolites of antagonistic microorganisms, the most common is the poisoned medium, adding the substance to evaluate in the culture medium before solidification, which consists in adding 200 μl of the secondary metabolites on PDA medium in the center of Petri dish and 5 mm mycelium disk of the phytopathogen, then Petri dishes are incubated at $28 \pm 1^\circ\text{C}$ until the control treatment covers the petri dish, after that the percentage inhibition is determined [23].

However, our workgroup has standardized the method in microdilution on plate, which consists in an adaptation of the technique proposed by Masoko et al. [38]; polystyrene microplates of 96 wells are used; in all wells, 100 μl of liquid medium is placed; column 1 is the negative control, column 2 consists of the positive control, and column 3 is a control which consists of the fermentation medium. Starting in column 4, 100 μl of the secondary metabolites from strains is mixed in a pipette with 100 μl of the liquid medium, and then 100 μl of the mixture is transferred to the next column, discarding the last 100 μl from column 12, to get serial microdilutions to 50.00% of the secondary metabolites. Once the microdilutions are carried out, the growth developer 2,3,5-triphenyltetrazolium chloride is added in the whole plate; the concentration of the growth developer is the lowest reported in the literature, as an excess of this indicator can interfere with the growth of the pathogen or react with reagents from the medium; this indicator measures the respiratory activity associated with electron transport chains, and when reduced, it precipitates forming a complex, intense red color; its use is due to its high sensitivity to detect

inhibition of microorganisms with deficient amounts of antimicrobial products; besides that, the red coloration is a visual indicator of the antimicrobial activity of the treatment. Finally, starting in column 2, 10 µl of a spore solution of the fungus at a concentration of 1×10^8 in all wells is added, keeping all the wells a volume of 150 µl in total; each microplate is considered a replicate. The microplates are incubated in agreement with the necessary conditions of the fungus on absorbance realized at 490 nm in a spectrophotometer. The secondary metabolites from the different strains placed in the rows A to F. To calculate the growth and inhibition percentage, the following formulas used: % Growth = $(A - B/C)(100)$; where A is treatment absorbance, B is negative control absorbance, C is positive control absorbance, and % Inhibition = $100 - \% \text{ Growth}$.

In general, the selection principle of strains is the determination of their antagonistic capacity; the method of microdilution in plate is to some extent interesting since it allows determined quickly and efficiently in time and costs its capacity of antifungal inhibition. Several studies demonstrate the effectiveness of secondary metabolites in the control of phytopathogenic fungi with PDA medium; Osorio et al. [23] mentioned that the inhibiting effect by *T. asperellum* and *T. hamatum* against *P. capsici* ranged to 15–20%; this inhibition attributed to the concentration of metabolites like glycotoxins, viridine, trichodermin, furanone, and 6-pentyl- α -pyrone (**Figure 4**).

Likewise, the PDA method or the microdilution in plate method can be an excellent technique to evaluate substance with antifungal activity as the secondary metabolites; in this sense, Jimenez et al. [36] observed that secondary metabolites obtained from *T. yunnanense* and *T. harzianum* at a concentration of 50 and 25% showed an inhibiting total effect of 100% of mycelial growth of *V. inaequalis*, while the metabolites obtained from *T. asperellum* and *T. lignorum* at a concentration of 50% showed an inhibiting effect from 90 to 84%, respectively (**Figure 5**). On the other hand, Jimenez et al. [36] reported that secondary metabolites obtained from *B. licheniformis* at a concentration of 50 and 25% showed an inhibiting effect in 100% against *V. inaequalis*, while the metabolites obtained from *B. subtilis* at a concentration of 50% showed an inhibiting effect near to 78% of the development of this pathogen (**Figure 5**).

In another study, Tucuch-Pérez et al. [39] reported six *Bacillus* spp. strains with antifungal activity against *F. oxysporum*; in this case, the species *B. licheniformis* and *B. subtilis* showed the highest inhibition percentages ranged from 80 to 100%, being the lowest inhibition percentage registered of 50% (**Figure 6**).

The results generated by the microplate dilution method are consistent with the results obtained from indirect test by confrontation or dual test, such as shown in the following results against fungi isolated from crops as melon, pepper, and others, which are produced in different zones from Mexico. In melon crop, the root and stem rot disease is a big problem; in this context, Espinoza-Ahumada et al. [40] studied the in vitro antagonist effects of *Trichoderma* spp. and found that *T. asperellum* have excellent biological activity against *Fusarium* strains, isolated from melon,

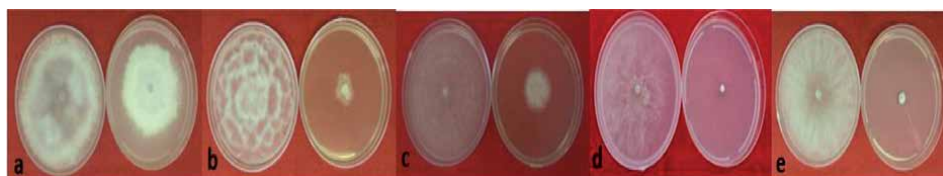


Figure 4. Antifungal activity of secondary metabolites from different *Trichoderma* spp. strains against phytopathogenic fungi; (a) *T. asperellum* vs. *Fusarium oxysporum*, (b) *T. yunnanense* vs. *Phytophthora capsici*, (c) *T. longibrachiatum* vs. *Rhizoctonia solani*, (d) *T. asperellum* vs. *Sclerotinia sclerotiorum* and (e) *T. asperellum* vs. *Sclerotium cepivorum*.

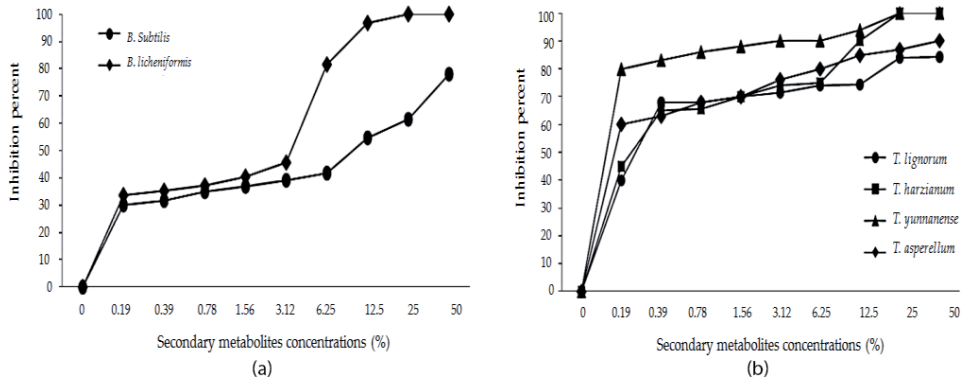


Figure 5. Percentage of inhibition of secondary metabolites obtained from *Bacillus* spp. (a) and *Trichoderma* spp. (b) against *Venturia inaequalis*.

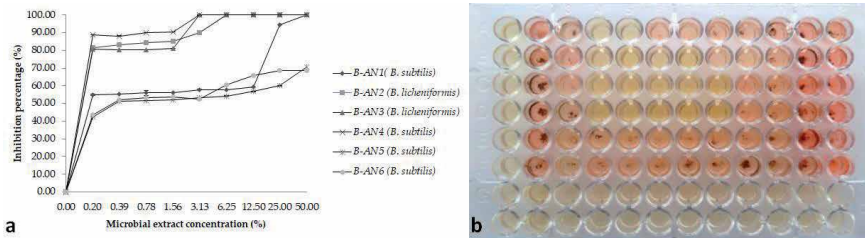


Figure 6. (a) Percentage inhibition of microbial extracts from *Bacillus* spp. metabolite dilutions against *F. oxysporum*. (b) Microplate with treatments to several concentrations, and the pathogen elapsed 48 h after incubation. Row A = B-AN₁, B = B-AN₂, C = B-AN₃, D = B-AN₄, E = B-AN₅, F = B-AN₆; column 1 = negative witness, column 2 = positive witness, 3 = growth medium of *Bacillus* spp., 5 = 50%, 6 = 25%, 7 = 12.50%, 8 = 6.25%, 9 = 3.13%, 10 = 1.56%, 11 = 0.78%, 12 = 0.39%, and 12 = 0.20%.

shown in **Table 3**. In general, these authors report that the inhibition of *Fusarium* spp. is higher when *Trichoderma* spp. are used (62.4–54.8%), in contrast when *Bacillus* spp. (44.5–36.9%) is used (**Figure 7**).

In a work carried out by Francisco et al. [41], where the behavior of *Bacillus* spp. against *Fusarium* species was studied, it showed low inhibition values. However, they report that the species *B. pumilus* and *B. liquefaciens* can be used effectively against many *Fusarium* species. On the other hand, higher effectiveness of *Bacillus*

Antagonistic agent	<i>Fusarium</i> strain					
	FRR-1	FRG-2	FAF-3	FRE-4	FCA-5	FHA-6
<i>B. liquefaciens</i>	46.9 ^{A,ab}	38.4 ^{B,abc}	31.4 ^{BC,bc}	34.8 ^{CD,bc}	28.2 ^{CD,c}	41.6 ^{DE,ab}
<i>B. amyloliquefaciens</i>	51.4 ^{AB,ab}	38.9 ^{B,c}	37.8 ^{B,c}	42.9 ^{BC,bc}	41.2 ^{BC,bc}	55.2 ^{C,a}
<i>B. subtilis</i>	45.7 ^{B,a}	38.6 ^{B,a}	37.2 ^{B,a}	42.4 ^{B,ca}	34.6 ^{C,a}	46.4 ^{D,a}
<i>T. asperellum</i>	57.2 ^{A,b}	61.5 ^{A,ab}	64.3 ^{A,ab}	62.3 ^{A,ab}	54.9 ^{A,b}	74.1 ^{A,a}
<i>T. harzianum</i>	54.9 ^{AB,b}	55.4 ^{A,b}	56.1 ^{A,ab}	60.5 ^{A,ab}	57.9 ^{A,b}	64.7 ^{B,a}
<i>T. viride</i>	49.8 ^{A,ab}	58.3 ^{A,a}	54.9 ^{A,a}	51.1 ^{A,ab}	50.3 ^{BC,a}	64.7 ^{B,a}

High letters indicate comparison between columns; low letters indicate comparison between rows. Percentages of inhibition with different letters are significantly different ($p \leq 0.5$).

Table 3. Percentage of antagonism of different biological agents against *Fusarium* spp. strains.

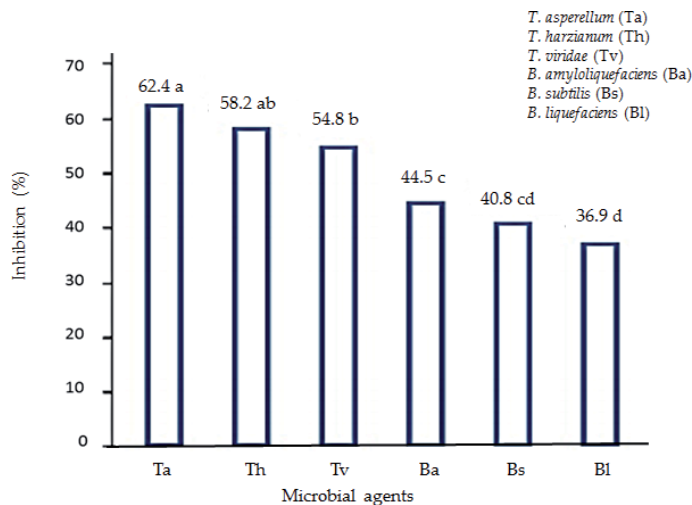


Figure 7. Microbial agents antagonist to *Fusarium oxysporum* (FAF-3, FRE-4, FHA-6) and *Fusarium solani* (FRR-1, FRG-2, FCA-5).

spp. was observed when applied in the early stage of growth [42], showing that *B. cereus* was most effective against *Fusarium* dry rot when applied as young cultures (24 h), however *B. thuringiensis* strains was most effective when applied as older cultures (48–72 h). Nevertheless, different studies revealed that *B. pumilus* produced different antifungal compounds as “iturin” which inhibits the growth of *Aspergillus* sp. and their production of aflatoxins [30]. Osorio et al. [23] found an inhibition ranged between 4.3 and 48.8% of *P. capsici* mycelial growth induced by the volatile compounds produced by *Trichoderma* spp. strains. The Tukey test indicated that 21 *Trichoderma* spp. strains showed the highest percentage inhibition. *T. asperellum* (T25) strain present the best result for activity inhibition, strain (T9) being the one with the least inhibition activity. It observed that the 31 *Trichoderma* spp. strains were able to produce volatile compounds with inhibitory properties against *P. capsici*.

6. Antifungal activity bioassay under greenhouse conditions

From different research projects under greenhouse conditions, we have found satisfactory results both in the disease suppression and in the promotion of growth and quality in crops. Hernández-Castillo et al. [24], made an experiment under greenhouse conditions using silty clay soil from an experimental batch previously plot with chile crop and were symptoms of wilting incidence were express. The experiment included three bacterial strains of the genus *Bacillus* spp. (B1, B3, and B13), a chemical control (thiabendazole, T), and control (TA) without fungicide. Before the application of the suspension, an initial colony-forming units (CFU) count of the pathogen involved by the dilution method is performed. The application of spores of the bacterial strains is performed at the time of the transplant. The seedlings are immersed in a spore suspension at a concentration of 10^8 (CFU/mL for 15 min). Subsequently, at 20 and 40 days after transplantation, the same spore suspension was applied to the stem base. In the final evaluation of each treatment (10 adult plants), plant height, fresh fruit weight per cut, root length, dry root weight, and incidence and severity of wilt are measured. The determination of the severity of the disease was with the scale reported by Copes and Stevenson [43]. As a result of this work, a very low wilt incidence found for plants

is inoculated with biological strains (B13 and B3) with an incidence of less than 10%, while values of 60 and 40% for TA and T, respectively, were observed (**Figure 8A**). Likewise, the wilting showed a reduction in severity in those treatments where three bacterial strains were applied (**Figure 8B**), in contrast to the control treatments where the severity of the damage was more considerable.

In **Figure 9**, we can see that the harmful microbiological population rate also reduced with the use of organisms considered as beneficial, according to the final count at the end of the experiment; that could be because antagonistic bacteria are capable of influencing biocontrol mechanisms against phytopathogenic fungi such as antibiosis, siderophores, competition for nutrients, and production of hydrolytic enzymes. Similarly, Ulacio et al. [44] evaluated organic matter and antagonistic microorganisms as management strategies against white rot in garlic cultivation. These authors reported that the fungus *Sclerotium cepivorum* is significantly reduced and there was a lower incidence of the disease in the treatments where the fungus *T. harzianum*, the bacteria *B. firmus*, and vermicompost were combined.

Some microorganisms possess the ability by several ways to reduce the incidence and severity of diseases in crops, and also can participate in the stimulation of plant growth, yield, and crop quality. **Figure 10A** and **B** shows the values related to the promotion of root length (A) and its weight (B), where this effect is clearly observed. In **Figure 10C** and **D**, it was observed that *Bacillus* spp. strains increase the height of the plant by 28% compared to treatment T, and 34.5% concerning the TA. These results coincide with previous work where the biological effectiveness

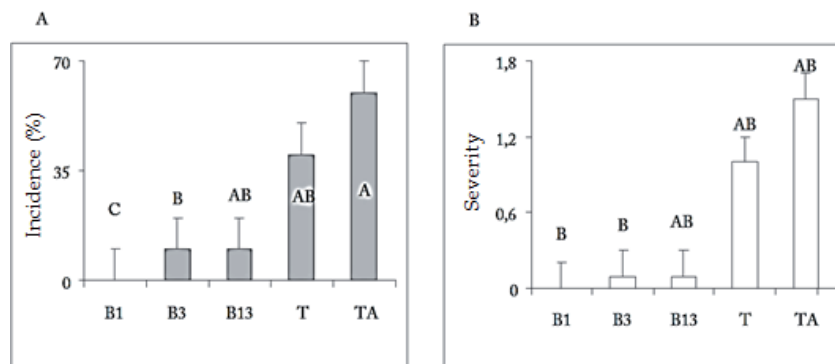


Figure 8. Incidence (A) and severity (B) in plant traits with *Bacillus* spp. strains (B1, B2, B3) in contrast with control (TA) and chemical control (T = thiabendazole).

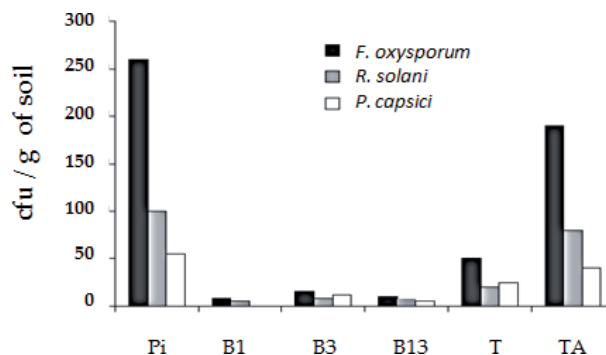


Figure 9. Colony-forming units from initial (Pi) and final populations of phytopathogenic soil fungi after applying *Bacillus* spp. strains (B1, B2, B3) against chemical (T = thiabendazole) and control (TA).

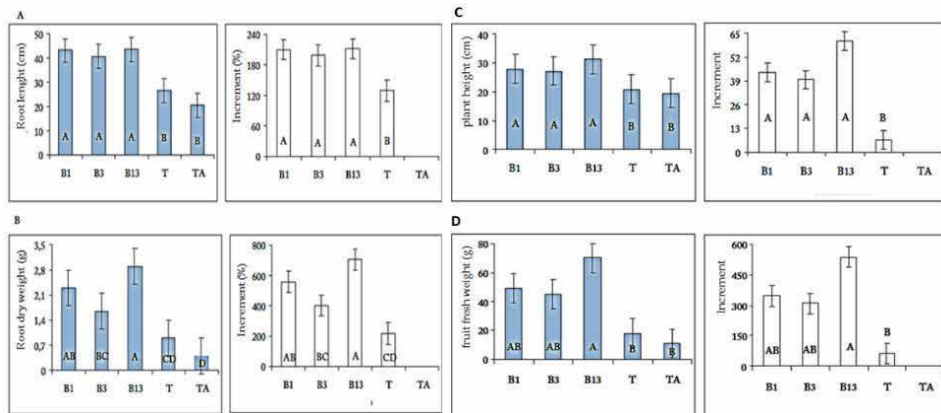


Figure 10. Root length (A), dry rot weight (B), height (C), fresh fruit weight (D), and increments in chili pepper plant by effect *Bacillus* strains (B₁, B₃, B₁₃) against chemical (T = thiabendazole) and control (TA). Different letters with bars indicate significant differences among treatments ($p \leq 0.05$).

of 57 strains of the genus *Bacillus* spp. isolated from the rhizosphere of commercial sowing chili plants in Northeast Mexico was analyzed, which showed an apparent antagonistic effect against *P. capsici*, *F. oxysporum*, and *R. solani* fungi. The plants inoculated with *Bacillus* spp. strains significantly increased height and dry weight in 191 and 60.2%, respectively [12]. The application of native *Bacillus* spp. strains shows a clear tendency to produce more biomass compared to chemical (T) and control (TA) treatments.

Likewise, del Ángel et al. [45] found a decrease in the incidence and severity of the disease caused by *Rhizoctonia solani* and *Fusarium oxysporum* with formulated endophytic bacteria, which induce a positive effect on the promotion of growth in the bean crop, increasing height and stem diameter in the treatments. Those formulated with bacteria in the absence of the phytopathogen stood out for their stimulating effect on the growth of the plants under study. This stimulating growth effect is observed in

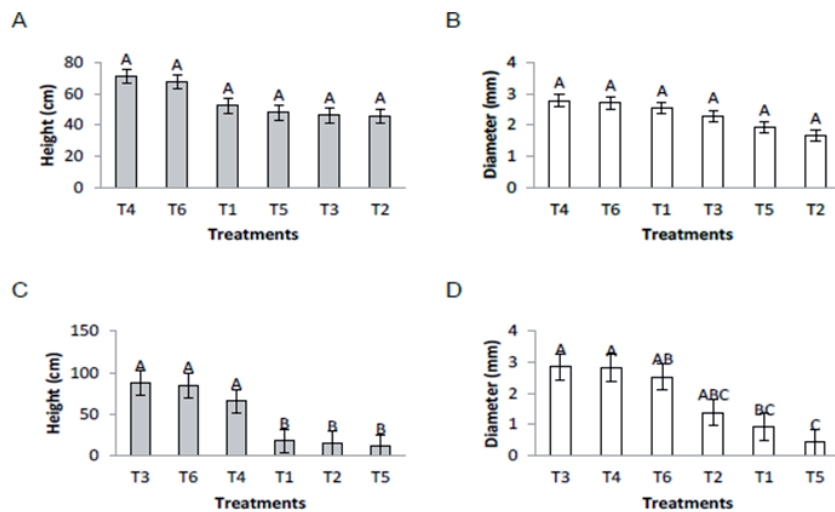


Figure 11. Effect of endophytic bacteria on plant height and stem diameter in bean crop under greenhouse condition. *Fusarium solani*: height (A), diameter (B), and *Rhizoctonia solani*: height (C), diameter (D). Means with the same letter are not significantly different according to the Tukey test ($p \leq 0.05$). Error bars are a standard error of the mean.

plants treated with those formulated and inoculated at the same time with pathogens. It is essential to mention that the plants grew under no chemical treatment. Therefore, they did not receive fertilization by any chemical source (**Figure 11**).

7. Bioassays of antifungal activity under field conditions

7.1 Fruits results

Jimenez et al. [36] report results obtained on apple fruit and trees under the direct influence of the application of CFU from *Bacillus* spp., and *Trichoderma* spp., as control agents against the incidence and severity of *Venturia inaequalis* under field conditions in commercial apple cultivar. **Table 4** shows the incidence of fungus *Venturia inaequalis* in fruit, and this incidence varied from 5.6 to 6.25 when biological agents (*Trichoderma* spp. and *Bacillus* spp.) were used in maxima doses (2 L ha⁻¹) to 19.3% for the control, respectively, after 15 days of a first application. After 60 days from the start of the applications, the incidence is expressed in a range of 42.5–46.62% for *Bacillus* spp. and *Trichoderma* spp. at doses of 2 L ha⁻¹ and for the control observed a 91.2%. The range of severity is observed between 1.8 and 2.6 of lesions per fruit by treatment *Trichoderma* spp. 2 L ha⁻¹ and control, respectively, after 15 days of application initiation. After 60 days of treatment application appears first symptoms, so it was evaluated on a range of the number of lesions per fruit (severity) from 5.3 to 14.5 corresponding to *Bacillus* spp., 2 L ha⁻¹, and control, respectively (**Table 4**). The treatment with the best antagonism effect under field conditions was *Bacillus* spp., 2 L ha⁻¹, who expressed 42.5% by incidence and five lesions per fruit in contrast to the control, which showed 91.2% incidence and 14.5 lesions per fruit (**Figure 12**).

The field experiment is carried out to test biocontrol agents for control *V. inaequalis* in commercial apple cultivar; the statistical analysis showed highly significant differences between treatments ($p \leq 0.5$), the incidence in foliage treated with *Trichoderma* spp. 2 L ha⁻¹ was lower in first evaluation (after 15 days of first application) and until harvest. This treatment expressed 10.6% incidence and two lesions per leaf, in contrast to the control which showed 31.8% and three lesions per leaf (**Table 5**). On the other hand, severity did not show significant differences among treatments.

7.2 Vegetable results

Espinoza-Ahumada et al. [40] aimed to find more environmentally friendly alternatives to the wilting of chile pepper; they evaluated the application of

Treatment	Incidence (%) in fruit		Severity (lesions) in fruit	
	15 days	60 days	15 days	60 days
<i>Bacillus</i> spp. 1 L ha ⁻¹	18.12 ± 2.4a	51.87 ± 5.5b	1.82 ± 0.6ab	8.02 ± 0.7b
<i>Bacillus</i> spp. 2 L ha ⁻¹	6.25 ± 5.2b	42.50 ± 6.5b	1.07 ± 0.8b	5.30 ± 0.5b
<i>Trichoderma</i> spp. 1 L ha ⁻¹	15.00 ± 3.5a	55.00 ± 5.4b	1.77 ± 0.5ab	7.62 ± 0.2b
<i>Trichoderma</i> spp. 2 L ha ⁻¹	5.62 ± 4.7b	45.62 ± 5.2b	1.00 ± 0.0b	6.32 ± 0.7b
Control	19.37 ± 4.7a	91.25 ± 4.3a	2.65 ± 0.5a	14.57 ± 0.3a

Treatments with the same letter are statistically equal to each other (p < 0.05).

Table 4.
 The incidence in apple fruits by *Venturia inaequalis*.



Figure 12.

Expression of symptoms caused by *Venturia inaequalis* in apple trees. (a) Without treatment, (b) *Bacillus* spp. effect, and (c) *Trichoderma* spp. effect.

Treatments	Incidence (%) in leaves			
	15 days	30 days	45 days	60 days
<i>Bacillus</i> spp. 1 L ha ⁻¹	8.12 ± 6.9ab	11.25 ± 1.2bc	13.12 ± 2.1bc	22.50 ± 4.8b
<i>Bacillus</i> spp. 2 L ha ⁻¹	6.25 ± 2.1ab	11.25 ± 2.5bc	11.87 ± 2.4bc	17.50 ± 4.6b
<i>Trichoderma</i> spp. 1 L ha ⁻¹	8.13 ± 3.8ab	13.12 ± 5.5bc	13.75 ± 4.7bc	20.62 ± 2.4b
<i>Trichoderma</i> spp. 2 L ha ⁻¹	2.50 ± 1.7b	6.25 ± 1.4c	6.25 ± 1.4c	10.62 ± 1.3c
Control	18.12 ± 4.1a	21.87 ± 3.1a	23.75 ± 4.3a	31.87 ± 3.8a

Treatments with the same letter are statistically equal to each other ($p < 0.05$).

Table 5.

The incidence in apple leaves by *Venturia inaequalis*.

biological agents for this purpose under field conditions. For this, an experiment is established where different genotypes of chile pepper are evaluated (Serrano, HS-52, Coloso, HS-44, Centauro, Paraíso and Tampiqueño 74 cv.) generated by INIFAP-Mexico. In this experiment, the microbial agents *T. asperellum*, *T. harzianum*, *T. yunnanense* [23, 29], *B. amyloliquefaciens*, *B. licheniformis*, and *B. subtilis* [24] under a mixture of microbial propagative ferment (consortium ferment) are based on *Trichoderma* spp. and *Bacillus* spp. Treatments of bioassay by *Trichoderma* spp. were different: consortium treatment one consists of a *Trichoderma* spp. at 1×10^8 CFU; treatment two consists of ferment consortium; treatment three consists of a *B. consortium* at 1×10^8 CFU; treatment four consists of a chemical control by thiabendazole prepared at 60% W/V; and the treatment five consists of an absolute control. A dose of 1 L.ha⁻¹ was applied for treatments one, two, and three, while the dose applied for thiabendazole was 0.5 kg.ha⁻¹. Field sowing is done with chile seedlings (10 cm), transplanted in 1.5 m double row beds. The application is made to drench with a manual sprinkler at 7, 28, and 49 days after the transplant (DDT). After 85, 105, 125, and 145 DDT, the yield per block (4.5 m²) is determined and transformed to t ha. To determine yields and improvements of treatments, ten fruits were evaluated, where the weight (g) and size (mm) per fruit were determined. In the first and last harvest, the incidence assessed and transformed into a percentage. The severity is evaluated through the visual scale, where 0 = no visible symptoms; 1 = initial light chlorosis and presence of flowers and fruits; 2 = intermediate, partial wilt, severe chlorosis, and premature ripening of fruits; and 3 = advanced. For total wilt without recovery, the leaves and fruits remain stuck to the stem. The field results observed as the effects of biological agents are shown in **Table 6**. The disease incidence values between HS-52 and Coloso treatments were statistically different ($p \leq 0.05$); in the other varieties, there were no differences between treatments. The treatment based on *Trichoderma* is the biological one that suppresses in higher

Microbial agents	Serrano chile pepper varieties					
	HS-52	Coloso	HS-44	Centauro	Tampiqueño 74	Paraíso
<i>Trichoderma</i> spp.	10.67a	18.17ab	16.84a	19.17a	12.5a	10.00a
Consortium	26.67ab	15.5ab	10.50a	15.33a	19.83a	10.67a
<i>Bacillus</i> spp.	29.17ab	29.67b	20.07a	20.5a	21.83a	19.5a
Thiabendazole	21.00ab	6.83a	19.51a	24.17a	10.33a	16.17a
Control	31.83b	21.33ab	21.51a	23.33a	24.67a	22.33a

Mean values on the same column indicated by different letters are statistically different ($p < 0.05$) according to the LSD test.

Table 6.
 Incidence of the disease (%) in serrano chile pepper varieties inoculated with microbial agents in the field.

percentage the incidence of wilting disease in chile pepper crops; in this case, the lowest incidence was in the HS-52 variety which showed a value of 10.67%, while that in the witness it was 31.87%, which represents a decrease of 71% concerning the latter.

Disease evaluation in the presence of treatments of consortium and *Trichoderma* demonstrates the lowest incidence percentage with values between 14.39 and 16.39%, while the control and *Bacillus* spp. were having high levels of the presence of symptoms (24.08 and 23.36%). In the case of severity, it also behaves differently between treatments. **Table 7** shows the values related to the severity of the disease

Treatment	Serrano chile pepper varieties					
	HS-52	Coloso	HS-44	Centauro	Tampiqueño 74	Paraíso
<i>Trichoderma</i>	11.3ab	18.43a	10.8a	6.83a	7.83ab	6.93a
Consortium	8.33a	16.28a	24.45a	12.76a	14.35ab	6.46a
<i>Bacillus</i> spp.	14.4ab	19.16a	15.09a	11.54a	17.6b	16.22ab
Thiabendazole	20.04b	14.07a	17.97a	15.74a	6.56a	18.24b
Control	19.45ab	24.35a	24.07a	13.65a	17.96b	18.43b

Mean values on same column indicated by different letters are statistically different ($p < 0.05$) according to LSD test.

Table 7.
 Severity of the disease (%) in serrano pepper with respect to treatments.

Microbial agents	Total yield in chile pepper varieties (t ha ⁻¹)			
	HS-52	Centauro	Paraíso	HS-44
<i>Trichoderma</i> spp.	15.67a	13.22a	8.48b	7.55a
Consortium	10.37ab	11.52ab	10.59a	13.04a
<i>Bacillus</i> spp.	7.26b	8.18ab	5.41b	10.3a
Thiabendazole	10.02ab	8.69ab	7.44b	10.62a
Control	5.98b	5.15b	2.59b	6.94a

Mean values on the same column indicated by different letters are statistically different ($p < 0.05$) according to the LSD test.

Table 8.
 Total yield, length, and weight of fruit of the serrano chile pepper crop obtained with the use of microbial agents.



Figure 13. Expression of incidence of coffee rust. (a) Plants with treatment based on bio formulate based on *Bacillus* spp., and (b) plants without treatment, where leaf defoliation is clearly expressed.

as transformed percentages ($p \leq 0.05$). It can be seen that *Trichoderma* spp.-based treatments alone or in combination have lower severity values.

The effects on yield as the weight and size of the fruit showed by the use of microbial agents applied alone or in combination as shown in **Table 8**. When *Trichoderma* is used, the yield increased; for example, its increase in the production was 62% when used alone and up to 76% when used as a mixture in comparison with the control.

This behavior of positive effect has already evidenced with the use of different *Trichoderma* species on habanero pepper plants (*Capsicum chinense*) [46], lettuce (*Lactuca sativa*), and radish (*Raphanus sativus*) [47]. In the same context, Cubillos-Hinojosa et al. [48] tested *T. harzianum* in the passion fruit crops (*Passiflora edulis*) where they were able to determine an antagonist to *F. oxysporum* and *F. solani*, in addition to stimulating germination, increased biomass, and root length.

In other field tests with *Bacillus* spp. bioformulate prototypes, a reduction in incidence and severity of coffee rust (*Hemileia vastatrix*) was observed. It was observed that the control presented 38% of incidence; nevertheless, it showed defoliation compared with the prototype treatments, which present an incidence between 5 and 15%, while with the chemist, the incidence was 9% (**Figure 13**).

The positive interaction between *Trichoderma* spp. and the host plant is attributed to a complex chemical activity of volatile and diffusible secondary metabolites and release of phytohormones and antibiotics in the rhizosphere, which promote root development and increased nutrient absorption, which helps control phytopathogens and increase yield [49]; this effect explains the results produced in this research. Microbial extracts as biofertilizers can generate hormones that stimulate development and increase yield, which are verified with the application of exudates from consortium *Trichoderma* spp. and *Bacillus* spp., which showed an effect on disease control and crop development in the same or better percentage than when using microorganisms.

8. Resistance induction by *Trichoderma* spp. and *Bacillus* spp.

In addition to the above aspects, plants can develop an increase in resistance to pathogen infection by treatment with a wide variety of biotic and abiotic inducers. Among the biotic inducers, we have the same phytopathogens, the growth-promoting rhizobacteria, and the microbial agents of the species of the genera *Bacillus* spp., *Streptomyces*, *Pseudomonas*, *Burkholderia*, and *Agrobacterium* and nonpathogenic microorganisms such as *Trichoderma* species (antibiotics or siderophores that lead to induction of resistance).

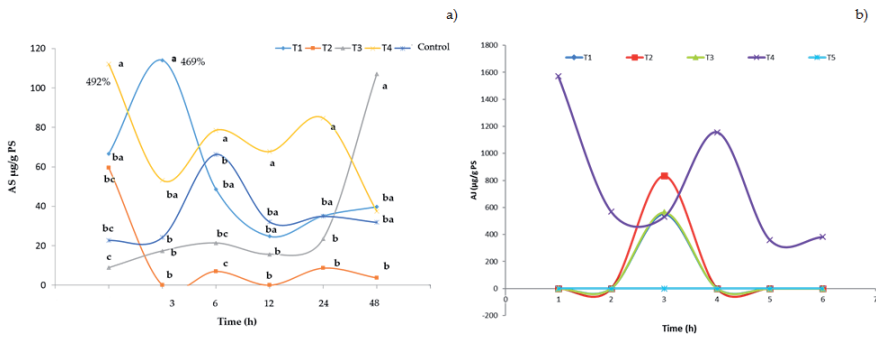


Figure 14. (a) Salicylic acid production on potato leaves in a different time. T1 = *Bacillus* spp. and *Pseudomonas fluorescens*, T2 = jasmonic ac. 1500 ppm, T3 = mezcla T1 0.5% + T2 0.1%, T4 = Milor®, and T5 = control (agua). Different letters indicate significant difference. (b) Jasmonic acid production on potato leaves in different time. T1 = *Bacillus* spp. and *Pseudomonas fluorescens*, T2 = jasmonic ac. 1500 ppm, T3 = mezcla T1 0.5% + T2 0.1%, T4 = Milor®, and T5 = control.

Among the abiotic inducers are salicylic acid (SA), jasmonic acid (JAS), β -aminobutyric acid, ethylene, chitosan, potassium, sodium or magnesium phosphate, acibenzolar-S-methyl (ASM), menadione, sodium bisulfite, and phosphites. The application of these inducers causes specific biochemical changes that occur after their application such as expression of genes that code for PR proteins; the increase of certain defense-related enzymes such as polyphenol oxidase, lipoxygenase, peroxidase, superoxide dismutase, and phenylalanine ammonia-lyase (PAL); the accumulation of phytoalexins and phenolic compounds; and the reinforcement of the cell wall with lignin deposition.

In this regard, we have observed changes in the endogenous levels of salicylic acid and jasmonic acid in potato plants in response to foliar application of microbial consortiums based on *Bacillus* spp. and *Pseudomonas fluorescens*. The microbial consortium of *Bacillus* spp. significantly increased the production of SA 3 h after spraying raising to 114.02 $\mu\text{g/g}$ DW. This is 496% more than the control (Figure 14a). Jasmonic acid is not detected in control plants but detected in plants treated with the microbial consortium. The level of jasmonic acid, 6 h later, reached a level of 550 $\mu\text{g/g}$ DW (Figure 14b).

The resistance induction is associated with some defense gene expression as encoding pathogenicity-related proteins (PR), for example, phenylalanine ammonia-lyase, which is crucial in the synthesis of phytoalexins, because these constitute highly toxic compounds to the pathogen. On the other hand, PAL is part of the synthesis of salicylic acid and phenolic compounds that reduce the incidence of diseases in plants. It has also shown that *B. amylolicheniformis*, *B. subtilis*, *B. pumilus*, and *B. cereus* are capable of eliciting and activating the induced systemic resistance by increasing the levels of biochemical compounds related to resistance induction. Besides, it reported that some *Pseudomonas* species could induce systemic resistance in plants.

9. Conclusions

The results shown in this chapter allow to demonstrate the efficacy of *Bacillus* and *Trichoderma*, as agents of biological control of fungi and stramenopiles that are causatives of plant diseases; these beneficial microorganisms can be used under a sustainable agriculture program or under integrate management pest program in a conventional agriculture. The microbial agents also express other advantages due

to their beneficial effects on the increase of the yields, growth, and development of plants, as well as the induction of systemic resistance in plants to phytopathogens. Currently our workgroup has any projects on the development of prototypes based on these microbial agents, alone or in consortium, as well as micro- and nanoencapsulated formulations.

Acknowledgements

The authors acknowledge the financial support for the development of research from the National Council of Science and Technology of Mexico (CONACYT), University Autonomy Agrarian Antonio Narro and Greencorp Biorganiks of Mexico, S.A. de C.V corporation.

Conflict of interest

The authors declare no conflict of interest.

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Section 5

Weed Management

Nonchemical Weed Control in Winter Oilseed Rape Crop in the Organic Farming System

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Rita Pupalienė, Rimantas Velička, Zita Kriauciūnienė,
Lina Marija Butkevičienė and Robertas Kosteckas*

Abstract

A field experiment was conducted during the 2014–2017 period at Aleksandras Stulginskis University (now—Vytautas Magnus University Agriculture Academy) on a *Endocalcaric Endogleyic Luvisol (LV-can.gln)* according to the WRB 2014. The three nonchemical weed control methods were explored: (1) thermal (using wet water steam), (2) mechanical (interrow loosening), and (3) self-regulation (smothering). In the thermal and mechanical weed control treatments, winter oilseed rape was grown with an interrow spacing of 48.0 cm and in weed smothering (self-regulation) treatment with an interrow spacing of 12.0 cm. Winter oilseed rape was grown in the soil with a regular humus layer (23–25 cm) and with a thickened humus layer (45–50 cm). Annual weeds predominated in the winter oilseed rape crop. In the soil with both humus layers, regular and thickened, the most efficient weed control method was mechanical weed management both during the autumn (efficacy 26.7–75.1%) and spring (efficacy 37.1–76.7%) growing seasons. Thermal and mechanical weed control in combination with the bio-preparations in droughty years significantly reduced the number of weed seedlings. Dry matter mass of weeds most markedly decreased through the application of the mechanical weed management method.

Keywords: winter oilseed rape, weed control methods, organic farming system, bio-preparations, soil humus layer

1. Introduction

The development of organic farming was prompted by the environmental concerns, health-related issues, and the search for solutions to social problems.

In Lithuania, organic farms account for more than 5% of the area under cultivation and are in line with the EU average. According to the popularity of organic farming, Lithuania surpasses the neighboring Poland but is far behind the other Baltic States. Although organic farms represent a small proportion of the total number of farms, their number has increased rapidly over the past decade. In 2017, 2448 organic farms were certified in Lithuania; they cover about 244,000 hectares of the agricultural land [1]. Most of the organic production farms are 10–30 ha in size.

Oilseed rape is one of the world's most important oil crops [2, 3]. The cultivation of oilseed rape on organically managed farms was encouraged by a search for healthy, high-quality, and safe food. According to the data from the public organization "Ekoagros," in 2017, the total area devoted to oilseed rape production on organically managed farms in Lithuania amounted to 3962.2 ha, including 3250.98 ha of winter oilseed rape and 711.22 ha of spring oilseed rape. The main reasons why winter oilseed rape production area on organically managed farms is not increasing are the problems associated with plant nutrition [4, 5], weed, disease and pest control, and unstable plant overwinter survival, and all these factors result in low rapeseed yields [6, 7]. Many organic farms in Lithuania are located on infertile soils, and the erosion and productivity problems are relevant there. Organic crop production farms are prevalent in this region; therefore the problem of crop rotation, nutrient, and humus balance is highly relevant. In the organic production farms, the inclusion of oilseed rape in the crop rotation is very important because it is characterized by phytosanitary properties, is a good pre-crop for other crops, and improves soil properties [8].

In the organic production farms, in the absence of the possibility of controlling weeds with herbicides, a great deal of attention is paid to nonchemical methods of weed control—mechanical, thermal, and natural crop-weed competition/self-regulation. Weed control by using wet water steam has not been extensively studied in the world. More comprehensive studies on the thermal weed control by water steam have been carried out by Lithuanian scientists [9–13].

2. Material and methods

Field experiments were conducted in 2014–2017 at the Experimental Station (54°53' N, 23°50' E) of Aleksandras Stulginskis University on an *Endocalcaric Endogleyic Luvisol (LV-can.gln)* according to the WRB 2014. Agrochemical properties of the experimental soil with a regular humus layer (averaged data of 2014, 2015, and 2016) were as follows: pH, 7.30; humus, 1.79%; contents of available nutrients in the soil: P₂O₅, 199.0 mg kg⁻¹; K₂O, 97.7 mg kg⁻¹; total nitrogen, 0.079%. Agrochemical properties of the experimental soil with a thickened humus layer were as follows: pH, 7.20; humus, 2.19%; contents of available nutrients in the soil: P₂O₅, 277.7 mg kg⁻¹; K₂O, 123.0 mg kg⁻¹; total nitrogen, 0.115%.

Two-factor field experiments were established using a split-plot design. Winter oilseed rape was grown in the soil with a regular humus layer (23–25 cm) (experiment I) and in the soil with a thickened humus layer (45–50 cm) (in 1988, a thickened humus-rich layer was artificially formed using fertile soil delivered from elsewhere) (experiment II). The length of initial plots was 14 m, width 6 m, and the area 84 m². The length of the harvested plots was 10 m, width 2 m, and the area 20 m². The experiments included four replications. The winter oilseed rape crop was preceded by black fallow.

The study object was winter oilseed rape (*Brassica napus* L. spp. *oleifera biennis* Metzg.) agrocenosis.

Experimental treatments:

Factor A: nonchemical weed control methods:

1. Thermal (water steam)
2. Mechanical (interrow cultivation)
3. Self-regulation/smothering (natural weed-crop competition, sowing with narrow interrows)

Factor B: biological preparations (bio-preparations):

1. Without bio-preparations

2. With bio-preparations

A winter rape cultivar “Cult” (Sweden, SW Seed) was grown in the experiment. The crop was sown at a seed rate of 3 kg ha⁻¹ with a Multidrill M300 sowing machine. In 2014, winter rape was sown on September 1, in 2015 on August 27, and in 2016 on August 29. In the thermal weed control treatment, the oilseed rape crop was grown with interrow spacing of 48 cm, and weeds were killed using a tractor-mounted wet water steam unit at a 3–4 leaf growth stage (BBCH 13–14) of winter rape. The thermal power of the device is 90 kW, with a capacity of 120 kg h⁻¹ steam; the device is run on liquefied gas. The temperature of steam is 99°C, with thermal treatment time of 2 s (Sirvydas, Kerpauskas, 2012).

The principal scheme of the tractor-mounted thermal weed control unit using wet water steam is presented in **Figure 1**.

2.1 Description of operation of the mobile thermal weed control unit

Liquefied gas is fed through tube 6 into the combustion chamber 4 of demountable steam boiler 8. There the burning gas heats water present in the steam boiler 8. The wet water steam which has formed in steam boiler 8 gets into the steam separator, in which steam dampness is reduced. Then the wet water steam which has passed through the steam overheater 11 is fed through the tube 5 into steam diffusers 2, which spread/distribute steam in the environment of target weeds.

The height of the steam diffusers is adjusted by the height adjustment mechanism 3. To prevent the liquefied gas from cooling, the gas cylinder 9 is placed into the heating tank 10. Hot water from steam boiler 8 is fed into the gas cylinder's heating tank 10 through tube 7. The mobile thermal weed control unit is mounted on a tractor 12 with a mounting device 11.

In the mechanical weed control treatment, the interrows were loosened with an interrow cultivator (KOR-4.2-01, Ukraine) using two passes. In the weed control treatment involving weed smothering (self-regulation), the winter oilseed rape was

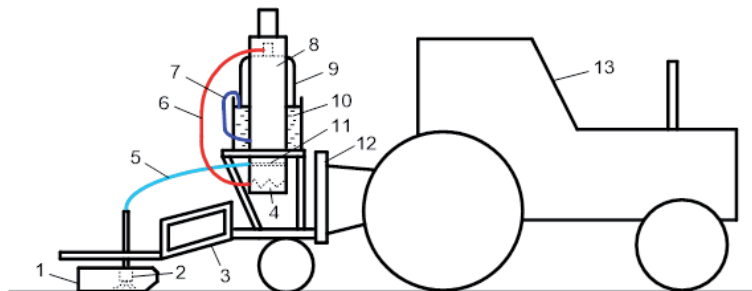


Figure 1.

The principal technological scheme of the tractor-mounted thermal weed control unit: (1) protectors of steam diffusers, (2) steam diffusers, (3) diffusers' height adjustment mechanism, (4) combustion chamber, (5) tube by which wet water steam is fed to steam diffusers and discharges, (6) tube by which gas is fed to combustion chamber, (7) tube by which hot water is fed to the heating tank of gas cylinder, (8) demountable steam boiler, (9) gas cylinder, (10) heating tank of gas cylinder, (11) steam overheater, (12) mounting device of thermal weed control unit, and (13) tractor.

grown with 12.0 cm interrow spacing. Winter rape was not fertilized, and no chemical plant protection products were applied.

In the treatments with the use of the bio-preparations, pre-sowing, the seeds of winter rape were coated with the bio-organic fertilizer Nagro (BioPlant) (0.5 l per ton of seeds and 10 l of water), and during the growing season, the winter rape crop was sprayed twice with the bio-preparations (in the autumn with Terra Sorb Foliar (BioIberica) (2 l ha⁻¹), in the spring with Terra Sorb Foliar (1 l ha⁻¹) and 0.3% Conflic (Atlantica Agricola)).

2.2 Assessment of weed incidence in the crop

The analysis of weed seedlings was carried out at winter rape 3–4 leaf growth stage (BBCH 13–14) in the autumn and after resumption of vegetation in the spring (BBCH 50) before the application of thermal and mechanical weed management methods. In each experimental plot, in four randomly selected 0.10 m² record plots, the number of weed seedlings and weed species composition were established. This analysis was done for the second time 5–7 days after application of the weed control methods in the marked record plots. The number of weed seedlings was recalculated as per m². The efficacy (E) of the weed management methods for the change in the number of weed seedlings was calculated according to the formula:

$$E = (S1 - S2) / S1 \times 100 \%, \quad (1)$$

where S1 is the number of weed seedlings per m² before application of the weed control methods and S2 is the number of weed seedlings per m² after application of the weed control methods.

At winter rape green silique stage (BBCH 79), the number of weeds and weed species composition were determined in each plot in four 0.25 m² record plots; the weeds were dried in a drying chamber at 60°C temperature and weighed [14].

2.3 Statistical analysis

The significance of the differences between the means was estimated using the *t* criterion; the interplay between the traits was determined by the correlation regression methods. The statistical analysis of the experimental data was performed using software STAT from the software package SELEKCIJA [15]. The experimental data that did not fit the normal distribution law, prior to the statistical evaluation, were transformed using the function $y = \ln x + 1$.

2.4 Meteorological conditions

In 2014 autumn was warm and long, so conditions for rape growing were favorable. In winter, meteorological conditions were favorable for rape over-wintering. In 2015, autumn was warm and humid. During the first decade of January 2016 of very cold weather and the absence of snow, over-wintering of rape was not successful. In 2016, the conditions for rape preparation for wintering and for over-wintering were favorable. In 2017, rape vegetation renewed on March 31. April was cold and humid with 35.3 mm more rainfall than usual. As a result, some winter rape has not over-wintered.

3. Experimental results

3.1 Weed incidence in the winter rape crop in the autumn and spring growing seasons

The following annual weed species predominated in the winter rape crop: *Chenopodium album* L., *Tripleurospermum perforatum* (Merat) M. Lainz, *Stellaria media* (L.) Vill., *Viola arvensis* Murray, *Veronica arvensis* L., *Sinapis arvensis* L., *Capsella bursa-pastoris* (L.) Medik, and *Poa annua* L.

In the soil with both regular and thickened humus layers, in the treatments where winter rape was grown with wide interrow spacings (48 cm), when the light and moisture conditions were favorable, the number of emerged weeds was higher than in the treatments with narrow interrows (12 cm), except for the spring growing season of 2017. The application of the bio-preparations in most cases reduced the number of weed seedlings in the winter rape crop both during the autumn and spring growing seasons.

Experiment I: in the soil with a regular humus layer. The most effective weed control method in organic winter rape crop was mechanical: efficiency 26.7–71.5% without biological preparations and 54.2–71.7% with biological preparations (**Figure 2**). The efficiency of the thermal weed control method was lower than the mechanical one. In Ref. [16], it was stated that the efficiency of mechanical and thermal weed control was 50–100%. Biological preparations enhanced the effectiveness of thermal and mechanical weed control techniques only in 2014. The effectiveness of the self-regulation method for the change of weed sprouts was negative throughout the study years.

Experiment II: in the soil with a thickened humus layer. The most effective weed control method in rapeseed crop was mechanical: efficiency was 39.8–75.1% without biological preparations and 53.0–68.9% with biological preparations (**Figure 3**). The efficiency of the thermal weed control method was lower than the mechanical one. Bio-preparations enhanced the effectiveness of mechanical weed control only in 2014. The method of self-regulation did not reduce the number of weed sprouts.

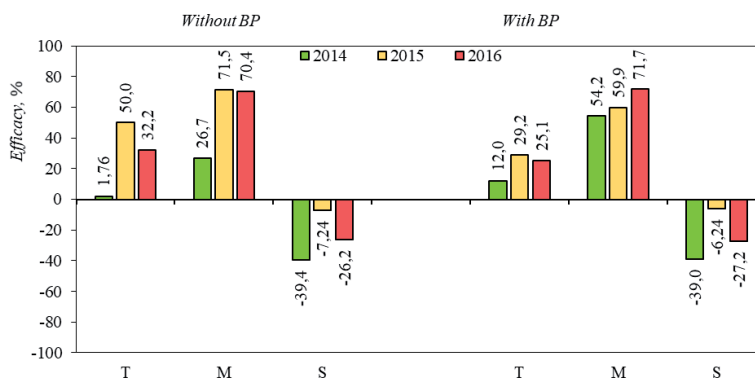


Figure 2.

The efficacy of the nonchemical weed control methods for the change in the number of weed seedlings in the winter oilseed rape crop, grown in the soil with a regular humus layer in the autumn (2014–2016). Note. T, thermal; M, mechanical; S, self-regulation; BP, bio-preparations.

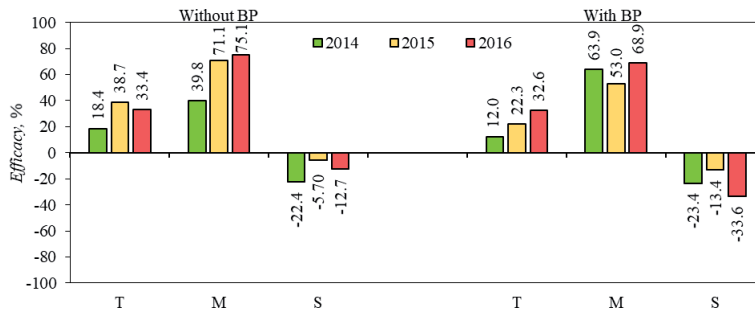


Figure 3.

The efficacy of the nonchemical weed control methods for the change in the number of weed seedlings in the winter oilseed rape crop, grown in the soil with a thickened humus layer in the autumn (2014–2016). Note. T, thermal; M, mechanical; S, self-regulation; BP, bio-preparations.

3.2 Weed incidence in the winter rape crop before harvesting

Experiment I: in the soil with a regular humus layer, the nonchemical weed management methods did not exert any pronounced effect on the weed number in the winter rape crop before harvesting throughout study years (**Figure 4**). Using bio-preparations in 2015 in experimental plots with mechanical weed control method, weed number was significantly 42.9% lower than in experimental plots with self-regulation. In 2016 and 2017, there were no significant differences in weed number between the different weed control methods.

The use of bio-preparations did not significantly affect the number of weeds in the oilseed rape crop during all study years compared to the treatment where they were not used.

There was no significant difference in weed dry matter mass between different weed control methods in plots with bio-preparations or without bio-preparations in 2015 (**Figure 5**). In 2016 and 2017, weed killing with interrow loosening and without biological preparations resulted in significantly lower weed dry mass than this in plots where thermal weed control and self-regulation methods were used, respectively, 2.0 and 2.5 times and 3.0 and 4.5 times. In 2017, the weed dry matter mass in plots with thermal weed control was significantly 32.3% lower than that of the self-regulation plots. In 2016 in plots with bio-preparations and mechanical

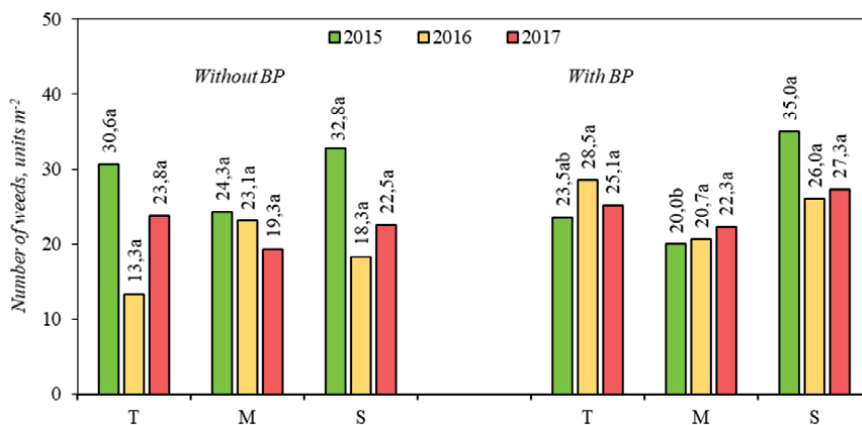


Figure 4.

The number of weeds in the winter oilseed crop, grown in the soil with a regular humus layer, before harvesting (2015–2017). Note. The differences of factor a, marked by not the same letter (a, b), are significant ($P < 0.05$). T, thermal; M, mechanical; S, self-regulation; BP, bio-preparations.

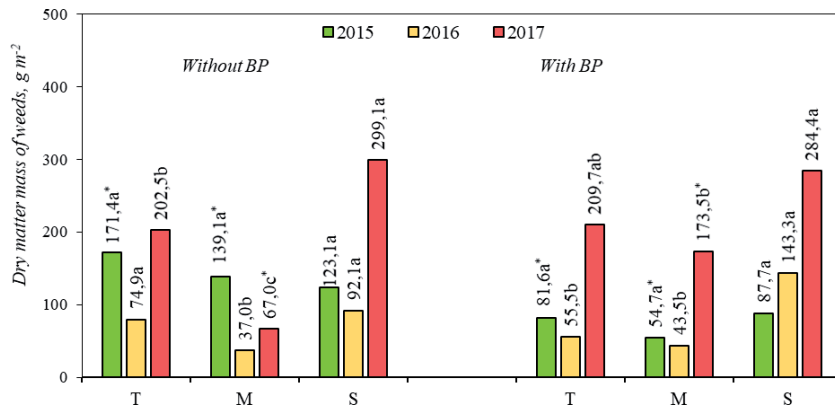


Figure 5. The dry matter mass of weeds in the winter oilseed crop, grown in the soil with a regular humus layer, before harvesting (2015–2017). Note. The differences between the averages of treatments of factor A, marked by not the same letter (a, b, c), and between the averages of treatments of factor B, marked by an asterisk, are significant ($P < 0.05$). T, thermal; M, mechanical; S, self-regulation; BP, bio-preparations.

weed control method, the weed dry matter mass was significantly 2.6 and 3.3 times lower than that in the self-regulation weed control method plots. In 2017, mechanical weed control resulted in a significantly 39.0% lower dry mass of weeds than self-regulation method. It was found that thermal weed control resulted in a 44.0% reduction in dry mass of weeds using thermal weed control with water steam [16]. The similar results were obtained in the experiments in Lithuania—weed numbers were 3.2–4.4 times lower in plots with mechanical weed control method than this in plots with self-regulation weed control method and respectively weed dry mass by 2.2–3 times [17, 18].

With the use of biological preparations, the dry matter mass of weeds decreased significantly by 2.1 and 2.5 times in the fields of thermal and mechanical weed control methods only in the droughty 2015, as the more fertile rapeseed crop using biological preparations better suppressed weeds. K. Różyło and E. Pałys [19] found that as the assimilation leaf area of rape increased, the dry matter mass of weeds decreased. In 2017 with the use of biological agents, the dry weight of weeds increased significantly 2.6 times in the fields of mechanical weed control.

Experiment II: in 2015 in the soil with a thickened humus layer, interrow loosening without biological preparations significantly decreased weed number by 38.8 and 36.8% compared to thermal weed control and self-regulation (Figure 6). In 2016 and 2017, there were no significant differences in the number of weeds in the plots with various weed control methods without biological preparations. Using bio-preparations in 2015 in plots with mechanical weed control method, and in 2017 in plots with thermal weed control method, the weed numbers were significantly lower than in the plots of self-regulation weed control method by 42.9 and 34.7%, respectively. In 2016, there was no significant difference in the number of weeds in the plots with different weed control methods and with biological preparations.

Without biological preparations in 2015 and 2016, there were no significant differences in weed dry mass between the different weed control methods (Figure 7). Different weed control techniques used in plots with biological preparations in 2015 had no significant effect on the dry matter mass of the weeds. In 2016 in plots with mechanical weed control and with biological preparations, weed dry matter mass was significantly 4.1 and 5.1 times lower than that in plots with thermal weed control and self-regulation. In 2017 the dry matter mass of weeds was 6.7 and 5.7

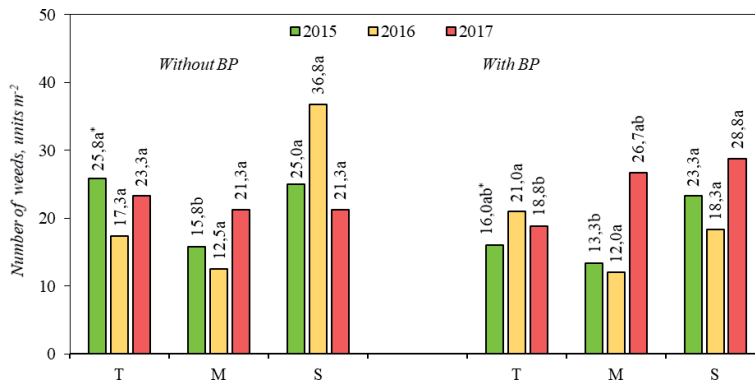


Figure 6.

The number of weeds in the winter oilseed crop, grown in the soil with a thickened humus layer, before harvesting (2015–2017). Note. The differences between the averages of treatments of factor a, marked by not the same letter (a, b), and between the averages of treatments of factor B, marked by an asterisk, are significant ($P < 0.05$). T, thermal; M, mechanical; S, self-regulation; BP, bio-preparations.



Figure 7.

The dry matter mass of weeds in the winter oilseed crop, grown in the soil with a thickened humus layer, before harvesting (2015–2017). Note. The differences between the averages of treatments of factor a, marked by not the same letter (a, b), and between the averages of treatments of factor B, marked by an asterisk, are significant ($P < 0.05$). T, thermal; M, mechanical; S, self-regulation; BP, bio-preparations.

times higher, respectively, without the use of biological preparations and 11.1 and 5.8 times higher with the use of biological preparations than in 2015 and 2016. Apparently, the higher weed dry matter content was caused by the lower rape crop and the humid and cold weather during rape vegetation. The mechanical weed control without biological preparations were found to have a significantly lower mass of weed dry mass than the thermal weed control and self-regulation, by 28.1 and 40.9% lower. The use of biological preparations and thermal weeds to control using wet water vapor resulted in a significantly by 33.7% lower dry mass of weeds than that in the self-regulation plots.

The use of biological agents significantly reduced weed dry matter in mechanical weed control fields only by 2016.

In 2017, negative, very strong, significant correlations were determined between the winter rape plant population density and weed dry matter mass: in the soil with a regular humus layer $r = -0.95$, $P < 0.01$; in the soil with a thickened humus layer $r = -0.91$, $P < 0.05$. [20]. R. Kosteckas (2011) [20] also found that the dry matter mass of weeds correlates with the density of rape crop.

4. Conclusions

Annual weeds predominated in the winter oilseed rape crop: *Chenopodium album* L., *Tripleurospermum perforatum* (Merat) M. Lainz, *Stellaria media* (L.) Vill., *Viola arvensis* Murray, *Veronica arvensis* L., *Sinapis arvensis* L., *Capsella bursa-pastoris* (L.) Medik., and *Poa annua* L.

In the soil with both humus layers, regular and thickened, the most efficient weed control method was mechanical weed management both during the autumn (efficacy 26.7–75.1%) and spring (efficacy 37.1–76.7%) growing seasons.

Thermal and mechanical weed control in combination with the bio-preparations in droughty years significantly reduced the number of weed seedlings. Dry matter mass of weeds most markedly decreased through the application of the mechanical weed management method.

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Section 6

Socio-Economic Aspects

Socio-Economic Dimensions of Adoption of Conservation Practices: What Is Needed to Be Done?

Nisa Sansel Tandogan and Haluk Gedikoglu

Abstract

Promoting sustainable agricultural production requires farmers to adopt new technologies such as organic farming to increase the agricultural productivity, while conserving the environment. Adoption and diffusion of new technologies need a long process, as experienced in the past. There are social and economic factors, identified in the literature, and those could cause delays in farmers' use of new technologies. Hence, technology adoption and diffusion are important policy issues in agriculture. For that reason, this paper provides a literature review including factors influencing the adoption and diffusion of technology in agriculture and aims to contribute to the future studies and policies, especially focusing on the social capital or the social aspects, which are proven not to be analyzed by the previous studies comprehensively. The results show that interaction with neighbors and relatives, and membership in a group or organization, which represent the social aspects, has a positive influence on adoption and diffusion of new technologies. Hence, policy-makers should incorporate the social aspects when designing the policies, such as cost sharing programmes, to promote adoption and diffusion of new technologies.

Keywords: technology adoption, diffusion of innovations, conservation practices, sustainable agriculture, social capital

1. Introduction

Promoting sustainable agricultural production requires farmers to adopt new technologies to increase the agricultural productivity, while conserving the environment. Since the seminal study by Griliches, adoption of new technologies has been widely analyzed in the agricultural economics literature [1]. Technology adoption in agriculture is also analyzed in the rural sociology literature. The studies in rural sociology mostly focused on diffusion of new technologies in a region, whereas studies in agricultural economics focused on adoption of new technologies by an individual farmer. Initially, the focus of agricultural technology adoption was to increase the productivity of the farmers or the profitability, especially during the Green Revolution. Hence, profitability of the technology was found to be an important factor for adoption [1].

Previous studies incorporated factors related to technology, farm and farmer characteristics into the analysis to explain why a technology is adopted or not. Hence, starting with profitability of the new technology, many factors were identified in the literature influencing the farmers' use. Since the 90's environmental concerns becoming important, and recently the global warming, the focus of technology adoption has shifted mostly from productivity-increasing technologies to sustainable agricultural technologies and the conservation practices. Especially for the developed countries, the focus is more on conservation practices and technologies such as precision agriculture that would both increase profitability and conserve the environment. The 2030 Agenda for Sustainable Development has mainly focused on the global aims, which include demanding unprecedented actions and efforts across multiple interconnected social, economic and environmental issues. In this sense, science, technology and innovation have a big share to realize these aims. The contributions of technology with the innovations to the economies provide the opportunities to improve living standards through rise in productivity, fall in costs and prices, and contributions to

Factor category	+	—	D	A	N	Total	References
Age	3	2	1	0	0	6	[22, 24–27, 49]
Gender	3	0	0	0	0	3	[24, 26, 28]
Income	2	0	1	0	0	3	[17, 26, 29]
Off-farm income	4	0	1	0	0	5	[30–34]
Ownership and wealth	6	0	1	0	0	7	[10, 18, 22, 24, 26, 36]
Education	14	1	0	0	0	15	[7, 11, 18, 23, 24, 27, 37, 38, 42, 43, 50, 68]
Learning	6	0	2	1	0	9	[18, 28, 44–49, 69]
Information	9	0	0	0	0	9	[7, 15, 17, 25, 26, 38, 41, 50]
Social capital	3	0	0	3	0	6	[57, 59, 60, 61, 63, 64]
Norms	1	2	0	0	0	3	[57, 59, 66]
Neighborhoods	4	0	0	2	0	6	[22, 24, 25, 44, 46, 48]
Relatives	4	1	0	0	0	5	[25, 26, 28, 36, 61]
Contacts with extension agents	4	0	0	0	0	4	[27, 43, 60, 68]
Membership in a group and organization	3	0	0	1	0	4	[25, 27, 36, 61]
Experience	4	0	0	0	0	4	[41, 46, 50, 69]
Farm size	5	0	1	0	0	6	[11, 15, 27, 29, 37, 41]
Farm location	3	2	0	0	0	5	[6, 22, 25, 36, 60]
Characteristics of soil and land	3	0	0	2	0	5	[10, 36, 37, 59, 60]
Prices of inputs	3	0	0	0	0	3	[10, 19, 70]
Risk	3	1	1	1	0	6	[7, 11, 29, 45, 50, 61]
Credit accessibility	5	1	1	1	0	8	[17, 19, 26, 27, 29, 59, 60, 61]

+: affect positively.

-: affect negatively.

D: significance of the effect changes depending on situation.

A: the sign of the effect changes based on situation.

N: no information.

Table 1.
Factors influencing technology adoption in agriculture.

the real wages [2]. It also plays a crucial role in the field of agricultural development [3, 4]. Technology decreases the risk of diseases and pest and increases the productivity and developments in agriculture by providing more information about the crop and soil structure for the farmers [5–9]. Additionally, new technologies are shown as a solution to the impeding conditions in agriculture, such as water scarcity, drainage and pollution [10–13]. Not only for the crops and soils but also for communication and information, technology contributes to the agriculture by providing several communication infrastructures and digital portals, reducing human intervention and eliminating technology breaks [14]. All of the effects mentioned above are important factors for sustainable agriculture. However, in this respect, there are two important issues: the adoption and the diffusion of technology. In the field of agriculture, with the well-known study of Feder and Slade, there have been many studies about agricultural innovations and their adoption [15]. As it is understood from the definition of technology for adaptation by UNFCCC (2005), which is stated as ‘the application of technology in order to reduce the vulnerability, or enhance the resilience, of a natural or human system to the impacts of climate change’, technology adoption influencing agricultural productivity significantly is an important issue in the economic sense [16]. In parallel with the adaptation, the diffusion of technology in agricultural field has been approached in many studies [17–19] beginning with Griliches [1]. Some tools have also been used to predict the speed and level of adoption [20]. As a reason, technology should have been used and diffused to benefit from its advantages; however, to do that, profitability needs to be understood by adopters [21]. When analyzing these studies, it is seen that there are many factors affecting technology adoption and diffusion (see **Table 1**). The main object of this paper is to review the factors influencing the adoption and diffusion of technology in agriculture comprehensively and contribute to the future studies.

2. Methodology

While this review handles the factors influencing the adoption and diffusion of technology in agriculture, it specifically focuses on the social capital including norms, neighborhoods, relatives, contacts with extension agents and membership in a group or an organization. Accordingly, the literature from 1974 to 2018 was analyzed by taking the social capital as a base. Our paper consists of 78 reviews, which cover 56 articles in journals, 10 book chapters, 4 selected papers for presentation, 3 working papers, 3 reports, 1 conference and 1 discussion paper. Rather than examining a specific time period or a specific region, 44 empirical studies, which cover various time periods and places, were analyzed to find the factors influencing the adoption and diffusion of technology. The revealing 21 factors were collected under the three main titles: the characteristics of farmers, the characteristics of farms and external incentives or disincentives. The effects of factors analyzed were tabulated depending on their positivity, negativity and variability to ease the future studies.

3. Factors influencing technology adoption in agriculture

3.1 Characteristics of farmers

3.1.1 Age

In the decision-making process, age is another influencing factor because it affects attitudes and perspectives on the new technology. Many studies about the

effect of age on technology adoption indicate different results. The analysis of Case shows that if the age of the additional male household member is between 0 and 15, farmers more adopt the new technology but if it is above the age of 55, the adoption rate decreases [22]. Hua et al. support that idea with their study by showing that while farmers under the age of 50 are more likely to adopt one type of new technology, farmers over 60 have lesser tendency to the adoption [23]. However, for another kind of new technology, the same study also shows that there is no significant difference between farmers under the age of 40 and over 59. Hence its effect can be attributed to the type of technology. From a different perspective, Weir and Knight found that farmers tend to be more influenced by someone in the same age group; however, there is a significant difference in this point [24]. If the farmers are educated, they have been more affected by older people rather than being influenced by people in the same age group. In this sense, younger farmers are more likely to be educated because they have more role models than older farmers. Contrary to most studies, some studies assert that older farmers have more tendency to adopt new technology even earlier than younger farmers due to their extensive experience in farm, and increase in the age shows positive effect on the adoption [24–26]. The study of Abdulai et al. differentiates the effect of age on adoption by determining criteria rather than differentiating it as older and younger [27]. According to their studies, although an increase in age at the younger ages affects adoption positively, increase at the older ages affects adoption negatively.

3.1.2 Gender

Differences stemming from gender have been observed in many subjects, the adoption of new technology in the agricultural field also shows different results depending on the gender. The study of Weir and Knight demonstrates that people are influenced by the people of the same gender while trying the new inputs [24]. Although the percentage being influenced is different among males and females, it is clear that they have a tendency to copy what people of the same gender do. The findings in their study show that female-headed households and male-headed households are influenced by the households of the same gender by 53% and 94% respectively. The adoption rate of female and male households differs from each other. Bandiera and Rasul studied on the social networks and technology adoption in Mozambique, which is mentioned as sunflower adoption in the study, and concluded that female-headed households have more tendency to adopt [28]. Other findings by Deressa et al. support the effect of gender differences on the decisions of the farmer by showing more adoption of male-headed households to climate change such as conserving soil, changing crop varieties and planting trees [26].

3.1.3 Income

The rises and falls in income have an effect on the decisions of adoption because farmers may change their preferences according to their new income level. Using new agricultural products causes changes in income, and income is doubled when modern crop yields per acre are used rather than traditional technology [17]. However, the use of new agricultural products or technology also depends on income because it needs financial facilities. In regard to this issue, the study of Feder and O'Mara indicates that the larger landowners benefit from the innovations more than smaller farmers because of their different income levels [17]. While experiencing the innovation is too risky for smaller farmers and hence

they abstain from the adoption, when there is a fall or stability in the relative risk aversion with income, larger farmers allocate more land for new yields. They also suggest that income distribution that occurred with the different income levels is improved as the smaller farms switch to new technology. Correspondingly, the diffusion process is completed when the incomes of all farmers increase and thus it is possible to worsen income distribution in the initial stages until the smaller farmers participate in the adopters' ranks. From a different perspective, Feder shows that the larger farmers allocate relatively less land for the risky activity and associate income with risk aversion [29]. He asserts that, in the case of binding credit constraint, increasing farm size leads to decrease in the allocated land for the modern crop in the situation of increasing relative risk aversion with income. Then, the rise of relative risk aversion with income decreases the expected income per acre and this causes the improvement of income distribution via the introduction of modern crop. On this issue, Deressa et al. analyze the influence of farm income and non-farm income on adoption [26]. It is shown that when farming is the main source for the income, farmers have the tendency to make an investment on productivity, and farm income affects conserving soil and using different crop varieties. On the other hand, although non-farm income has a negative effect on these modern agricultural methods even if it is not significant, it has a positive effect on the possibility of planting trees and using irrigation as an adoption option. Both farm income and non-farm income also increase the possibility of changing planting dates.

3.1.4 Off-farm income

Off-farm income, as in income provided by farm, is also a significant factor for the adoption of technology because it contributes to the economic performance of the farm household [30]. It helps farmers to increase capital availability and financial resources to invest in new inputs, practices or technology [31, 32]. Hence, various studies indicate that off-farm income has a positive impact on the adoption of new technology [30, 31, 33]. However, this impact has shown differences depending on the technologies. Gedikoglu et al. assert that although there is a positive relationship between the adoption of capital-intensive technology and operator's off-farm employment, it cannot be supported for the relatively labour-intensive practice [32]. Fernandez-Cornejo et al. confirm that farms with labour-intensive enterprises less prefer off-farm work [30]. Moreover, the decisions taken by the operators to work off-farm have an influence on the decisions of their spouses and that decision for off-farm work has also a positive relationship with adoption. This relationship is not one-sided, there is a correlation because adoption also enables the off-farm work [34]. The study of Fernandez-Cornejo et al. indicates that the more off-farm income increases, the more the probability of adoption of technologies increases for better time management [30]. For lower off-farm income, it is seen that fall in the off-farm income directs farmers to adopt yield monitors. From another perspective, this study emphasizes that the increase in off-farm work activities leads to a decrease in the farm-level efficiency because of allocating less time for farm management. This situation affects the adoption of management-intensive technologies negatively. In this sense, the study by Goodwin and Bruer provides an explanation that because crop producers make seasonal production, they are more advantageous than livestock product producers who have to work year-round [35]. Although there are some factors influencing off-farm employment stated in the literature such as farm size and wages, the positive effect of off-farm work on the adoption of technologies is apparent [30, 32, 35].

3.1.5 Ownership and wealth

Incomes and livestock owned represent wealth in agriculture [26]. Therefore, most of the studies taking livestock and machinery as a measure for wealth show that as the household wealth increases, the farmers have more tendency to experience new agricultural products and adopt them [22, 36]. However, this relationship is not stable, it changes as less wealthy households adopt in time [18]. The study of Weir and Knight on adoption and diffusion of agricultural innovations draws a conclusion that a great majority of households are influenced more by richer households in the process of adoption decision and this factor is valid also for the adoption decisions of educated households [24]. In parallel with the effect of wealth, ownership has an impact on adoption. While the study of Deressa et al. shows the positive effect of livestock ownership on adoption methods [26], Kassie et al. emphasize its positive and significant effects on the adoption of improved seeds [36]. Land tenure is also evaluated as a factor within the scope of ownership. Kassie et al. who examined this issue in detail indicate that land tenure has an influence on adoption in terms of security because while better tenure security increases the probability on farmers' investments, worse security decreases the adoption of some agronomic practices on rented plots [36]. Additionally, being a tenant is also effective in the decision process because while tenants focus on short-term soil fertility by overusing chemical fertilizers, owners take a long-term decision about soil fertility on their plots, thus its effect in the short term is stated as ambiguous.

3.1.6 Education

As in most of other fields, education has an impact on technology adoption in agriculture. There have been many studies that show the positive effects and contributions of the education on technology adoption from different perspectives. First of all, education increases the decision-making efficiency of adoption [37] and improves the systematic and creative thinking skill for making innovative decisions [38, 39]. It helps the understanding of the effects and the results of the technology adoption [38]. Cotlear indicates that all three types of education, which are formal, nonformal and informal education, play a great role in the diffusion of innovations through the rise in farm productivity, explanation of the information and shaping of behaviors, beliefs and habits [40]. From a different perspective, education has a significant effect on the initial adoption of innovations [24] because it leads to decrease in adoption costs and uncertainty and hence the timing of adoption becomes shorter and the probability of early adoption increases [41]. Rise in productivity also has been seen as being related with high education because of its contribution on the information acquisition and the accessibility of improved technologies [42]. The study of Huffman indicates that benefiting from the available information totally depends on the education level of decision-makers [43]. He also states that education improves the skill to obtain and process information and accelerates the changes, and the production increases at the end of the process.

3.1.7 Learning

Learning is another element influencing technology adoption. Social learning has an important role in the farmers' decisions and knowledge diffusion, and the investments in learning about technology are related with the technology adoption [44]. Besley and Case explain the cause of the slow technology diffusion as the lack of learning its profitability [18]. In line with this idea, learning is explained as the fall in the likelihood of allocative error by obtaining more information about the

likelihood of output [45]. There have been many studies about the understanding of how learning comes true. A study conducted by Foster and Rosenzweig examines both learning by doing and learning from others [46]. Their study emphasizes that if the learning realizes with both neighbors' and own experiences, profitability occurs more rapidly. In this sense, while Krishnan and Patnam support the power of social learning in adoption, they claim that the effect of learning from farmers' own and farmers' neighbors' experiences on adoption is greater than the effect of learning from extension services, in their study [47]. Conley and Udry also assert that neighbors influence the behaviors of individuals in the lack of learning, and individuals may behave according to their preferences because of being subjected to an unknown thing [49]. Munshi states that when the technology performance depends on the latent characteristics of neighbors, social learning will be weaker [48]. As a reason, farmers take into account not only their own direct observations of realizations but also those learned by neighbors [49]. Even, Besley and Case provide the reason of early adoption as being forward looking by learning more about the new technology [18].

3.1.8 Information

There is an extensive literature about the effect of information on the adoption in agriculture because it is an important stage for the adoption and diffusion of technology. It has been considered that the resource allocation skills and efficiency of adoption decisions can be increased via information gathering [7]. However, this information-gathering process can be affected by the adopters' specific attitudes [50] and hence the differences in the interpretations of information lead to different adoption decisions [38]. In this sense, especially, uncertainty and the lack of information are the subjects, which are mostly touched on. Imperfect information and uncertainty have an influence on the adoption of the decision and information diffusion affects the adoption positively by reducing the uncertainty [50]. Fall in uncertainty also decreases the cost over time [17] and hence the farmers who have more information tend to adopt earlier more than other farmers who do not [15, 41] because they consider other options less valuable to wait [7]. However, information access is different for each farmer and adoption changes accordingly. Feder and Slade indicate that the resources used for obtaining information by larger farmers are more than those used by smaller farmers and this leads larger farmers to have more knowledge and adopt earlier [15]. From a different perspective, Wozniak states that information increases innovative ability and while having more information makes farmers innovators, having less information makes them operator [38]. As a source of information, agricultural extension services and private agricultural supply firms have an important role in the agricultural sector [41] but information obtained from agricultural extension services has been seen as more valuable than information obtained from private agricultural supply firms because while the private firms can provide service for profit of the firm, agricultural extension services exist for giving technical information [38]. He added that the diffusion of the information has influenced production and welfare positively.

3.1.9 Social capital

The concept of social capital, which was first mentioned by Hanifan, has been defined in many different ways since there is no consensus about the definition [51–54]. One of the most important differences differentiating this term with other kinds of capital has been seen as its existence in social relationship [55]. In this sense, one of the most accepted definitions about the social capital was made by

Smithson as 'social capital is a person's or group's sympathy toward another person or group that may produce a potential benefit, advantage, and preferential treatment for another person or group of persons beyond that expected in an exchange relationship' [56]. Thus, its measures have been generally taken as networks, trust and norms [57, 58]. Many studies about the social capital have shown that it also affects technology adoption in different ways [57, 59]. According to these studies, firstly, social capital enables farmers financially in the lack of credit accessibility [60, 61]. Secondly, social networks which are a part of social capital decrease the transaction costs [62] because the major driver of that costs is the lack of information and contract enforcement assistance [63]. In the same way, it fills the information gap leading to market inefficiency [64]. Social capital also contributes to the well-being of an agent by influencing that person's relationship with others [65]. All these effects cause the rise in production and adoption. On the other side, some studies about the influence of social capital on adoption indicate that social capital may also influence adoption negatively because it depends on technology [60]. However, the studies about its positive sides on the adoption are quite a few than its negative sides. By benefitting from the previous studies, the instruments of social capital, in this paper, have been taken as norms and networks including neighborhoods, relatives, contact with extension agents and membership in a group or organization.

3.1.9.1 Norms

As it is shown many times in the literature, individuals are affected by the opinions and decisions of others. Social norm, in this sense, is another factor that influences the likelihood of adoption. Laple and Kelley examined this issue and suggested that the decision of adoption is made by not only farmers but also others [66]. It is found that belief-based subjective norms are a prominent motivation for farmers to convert their intentions in their study. Hunecke et al. studied on the topic of understanding the role of social capital in adoption decisions and concluded that norms affect the adoption of scheduling significantly and negatively [57].

3.1.9.2 Neighborhoods

One way to obtain information is from neighbors, and the effects of neighbors on adoption should not be overlooked to obtain unbiased and valid results because it influences both decisions and actions of farmers [22]. It is seen that if there is no opportunity for social information, individuals have to experience on their own but individual information does not compensate for the information gained from neighbors [48]. While more educated household heads or novice farmers have more tendency to learn using new inputs or obtain information from their neighbors [24, 44], farmers, who have the lower ability for information obtained from neighbors, have less reaction and slower adoption speed [44]. Also, if there is not a feasible environment, the person under the social pressure may prefer to act like a neighbor to improve the productivity [44, 46]. Neighbors contribute to farmers in terms of teaching input use and they take an active role in providing information with agricultural extension services [24, 46]. Not only for using input but also for changes in the use of inputs, neighbors are an important information source. The study of Conley and Udry (2010) emphasizes the strong relationship between changes in use of inputs and neighbors and indicates that if an information neighbor makes higher profits than expected by using more inputs, the farmers have a tendency to increase their input uses [44]. In this sense, the experience of the neighbors is one of the main points. According to the findings of Munshi, if the experiences of neighbors

cannot be observed well by the individuals, weak social learning and slow diffusion rate will occur [48]. Similarly, with this study, Foster and Rosenzweig indicate that profitability stemming from new technology increases with the individuals' own experiences and those of their neighbors; even an increase in the experience of neighbors approximately doubles the profitability according to the same increase in individuals' own experience [46]. The study also shows that decisions taken by neighbors play an important role in the decisions of farmers, future decisions taken by farmers are influenced by the past decisions of neighbors, and this shapes the planning decisions of farmers. Even this influence may outweigh the influence of extension services; Krishnan and Patnam assert that the influence of adoption by neighbors is approximately three times higher than the influence of extension agents [47]. Being approved by the society is another reason encouraging the farmer to adopt new agricultural technology [25]. In this sense, access to neighbors is an important factor in the adoption of new agricultural technology. Findings indicate that farmers who have access to neighbors have more tendency to adopt the new technology and adopt more quickly with information obtained [48, 59]. Wollni and Andersson state that if the neighbors are adopters, farmers become more disposed to adopt [25]. However, Munshi supports that even if an individual observes the decisions of neighbors well, that person may not obtain the same result with theirs because of the effect of different characteristics on performance [48]. On the other hand, although there are many benefits of the connection with neighbors, some farmers prefer to abstain from adoption because of the idea that the adoption also provides benefit to their neighbors' plots [25]. Nowak, whereas, defends the opinion that collective work is important to solve local resource management problems [67].

3.1.9.3 Relatives

When it is considered that the adoption decisions of farmers are affected by external factors, the effect of family on the decisions taken is inevitable. The findings of Bandiera and Rasul indicate that there is a strong correlation between adoption decisions and family [28]. As the number of relatives increases, the adoption possibility of households increases because of the opportunity for experiencing new technologies with lower risk; however, at the same time, great numbers of relatives lead to decrease in the work efficiency [36]. The factor that increases the probability of adoption can be shown as the labour supply provided by family and lower opportunity costs [25]. Deressa et al. confirm this positive relationship although the coefficients are not significant in their study and show that having more relatives in a local place affects adoption positively [26]. The study of Bandiera and Rasul examines the effect in terms of being the adopters of family members and concludes that increasing the number of adopters in the family increases the adoption probability of farmers [28]. However, this situation is found as valid just up to 10, then the marginal effect of the network is negative, so the relationship is shown in an inverse-U shape. Contrary to these studies, Wossen et al. show that compulsory sharing and strong loyalty among kin members have a negative effect on the adoption of farmers because they cause the free-riding behaviors [61].

3.1.9.4 Contacts with extension agents

As in mentioned in the effect of gathering information on adoption, the sources providing information also have an influence on the adoption; one of them is agricultural extension services. The study of Huffman who addresses the extension activity as an indicator of information availability indicates that there is a positive

and significant effect of the availability of information on the information gathering and processing it if adjustment is needed [43]. Also, agricultural extension services can substitute education in allocative efficiency. The information provided by the extension services helps the farmers in terms of understanding the process and using new technology; hence farmers who are in contact with agricultural extension services adopt the new technologies more [27]. The study of Husen et al. on this issue proves that when it is compared to the farmers without contact with agricultural extension services, the adoption of farmers having contacts with extension services increased by 28.85% for productivity-enhancing technologies [60]. From a different perspective, Huffman examines the role of human capital in the farm and off-farm work decisions in his study by including the effect of agricultural extension as an input [68]. His findings support that there is a positive relationship between the off-farm labour supply and the agricultural extension input because the extension services increase the productivity of farm and shorten the time for gathering information. This situation leads to shift in the demand of farmers for farm work and increases the farmers' days of off-farm work.

3.1.9.5 Membership in a group or an organization

Being a member of an organization or an institution is important in terms of accessing information and knowledge about new practices and technologies [25]. As shown in the literature, the better and easier access to information mostly has a positive effect on the adoption; hence the membership in an organization and institution also plays a role in the adoption. The study of Wollni and Anderson proves that membership in a farmer group, which provides assistance and information for farmers, increases the adoption of organic agriculture by 26% [25]. Wossen et al. examined this issue in detail, indicating that being a member of an association is not only important for gathering information but also for providing financial resources and having labour-exchange options [61]. As a reason, while membership in informal credit and saving associations helps farmers by solving liquidity problems, a member in labour-sharing arrangements may benefit from the opportunities of labour resources provided by those arrangements. These opportunities relax farmers and facilitate the adoption. The findings of their study show that these associations increase the adoption; when a farmer becomes a member of a local saving credit association, the likelihood of adoption of land management practices increases by 19.4%. Corresponding to the result of this study, Kassie et al. and Abdulai et al. confirm the positive relationship between being a member in an institution or association and adoption of new agricultural technologies or practices [27, 36]. On the other hand, the study of Wossen et al. emphasizes that the effect of this relationship depends on the type of institution or arrangement because it is found that being a member in funeral insurance arrangements or having a great number of relatives has a negative effect on the adoption of improved land management practices [61]. For the reason, funeral insurance arrangements direct farmers to make their social commitments and make money for funeral expenses, leading to abstaining from the agricultural innovation. The kinship also decreases the expectations from the adoption and so the likelihood of adoption because of the low incentives for collective sharing of benefits from adoption.

3.1.10 Experience

Although there are many beliefs and opinions about the new technology, these change with experience in time [18]. Especially in the adoption process, prior adoption experience has a significantly positive effect on adoption intensity [50].

The study of Wozniak shows that while uncertainty and the fixed costs of adoption obstruct early adoption, gaining experience increases the probability of being the early adopter of farmers because the more farmers gain experience, the more they cope with the difficulties of the adoption process [41]. Experience also affects the productivity positively [69]. Foster and Rosenzweig, who studied on the topic of learning by doing and learning from others, conclude that the experience of farmers and their neighbors allows them to take better decisions about the use of new technologies, and these decisions affect the profitability of adoption [46]. It is shown that experienced neighbors make more profit for farmers than inexperienced neighbors, and moreover, an increase in average experience of a farmer's neighbor affects the profitability of a farmer by approximately twice more than own experience. Conflictingly, although Wozniak supports the positive relationship between experience and the adoption time, he asserts that the relationship between experience and adoption shows differences because experience depreciates in a technological environment in time and hence the result may be biased in case of no any measurement for specific experience of adoption behavior [41].

3.2 Characteristics of farm

3.2.1 Farm size

The likelihood of adoption of new technologies has been also associated to farm size, in the literature, because the probability of adoption has changed significantly depending on farm size [37, 41]. One of the reasons for this difference has been shown as information accumulation. Feder and Slade emphasize that there is a positive relationship between the accumulation of information and farm size. The more farm size becomes large, the faster critical level of information is reached because larger farmers allocate more resources to get information [15]. All of these lead to the earlier adoption. Supportively, Wozniak and Rahm and Huffman confirm that larger producers have a relatively greater incentive to obtain information about innovations; hence they spend more time and more money for better quality information [37, 41]. This more allocation for information makes large-scale producers early adopters by allowing them to adjust the inputs according to the innovations. When profitability and costs of the inputs are taken into consideration, Torkamani and Shajari also support that the larger farms adopt new technologies more rapidly than smaller farms to prevent water cost and derive more profit [11]. In contrast with these studies, the study of Feder suggests that larger farmers tend to allocate relatively less land to the modern crop so as to not endanger their wealth because of the risk factor [29]. Besides all these, Abdulai et al. emphasize that farm size is not the only factor influencing adoption, cropping patterns and physical characteristics also have an effect on the adoption because while farm size has a positive and significant effect on one product, it may not have any significant effect on another product such as onion [27]. However, the effect of farm size on the selection of product for growing is also shown in their study in that while larger farmers tend to grow some kind of agricultural products such as onions and cabbage, smaller farmers tend to grow other products such as lettuce.

3.2.2 Farm location

When obtaining information about the new technology, the location of farm is important in terms of ease of accessibility and availability of information. While opportunities that stem from the farm location make adoption easier and shorten the adoption time, negative things caused by farm location make adoption difficult

and extend the time. The study of Khanna, which is one of the studies on this issue, handles the farm location as a main factor influencing adoption of soil testing [6]. It is shown that the more proximity of farmers to professional services increases, the more likelihood of soil testing adoption increases because most farmers trust the services provided by professional dealers. By confirming this relationship between farm location and adoption, the study of Kassie et al. indicates that the location has an effect on the investment decisions of farmers and hence being far from a village or a household that has more opportunities in terms of input and output decreases the likelihood of adoption of sustainable agricultural projects [36]. Case and Husen et al., who handle the subject in terms of accessibility to agricultural extensions, also emphasize that while proximity to agricultural centre affects adoption positively, parcel distance and being distant from their farm affect their adoption and farm management negatively [22, 60]. Due to these positive effects on adoption, it is shown that proximity to main market center provides better access to organic market outlets and hence their adoption becomes easier [25]. Eventually, when information is considered as positively related with the adoption, it is clearly seen that the accessibility to information, which means the closeness of farm location to the information sources, increases the likelihood of adoption.

3.2.3 Characteristics of soil and land

Many research studies conducted on technology adoption in agriculture prove that soil characteristics have a certain effect on the yield [37]. The study by Rahm and Huffman indicates that reduced tillage practices, which are used to measure adoption, influence the yield positively on the soils having poor characteristics [37]. Even, they expect to be dependent of per acre profitability on soil characteristics [10] because technologies decrease problems, which can stem from climatic conditions and natural events, by providing necessary conditions as required by the soil [37]. Larger and unfavorable fields for agriculture have more tendency to be equipped with modern technologies [10]. The probability of adoption changes depending on the soil characteristics and hence adoption shows the differences among farms [37]. Rahm and Huffman and Isham indicate that the probability of adoption under better soil conditions is higher [37, 59]. The benefits of reducing unfavorable conditions and improving soil characteristics provided by adoption affect the crop production value, input expenditures, productivity and sustainability significantly [36]. However, in this sense, the type of technology also causes differences in adoption. For instance, the study of Husen et al., examining the adoption of soil and water conservation practices (SWCs) and productivity-enhancing technologies (PETs), indicates that although land slope affects the adoption of SWC positively, it has a negative effect on the adoption of PET [60]. The adoption differences in terms of soil fertility, parcel distance and agricultural extension had also been observed in this study.

3.3 External incentives or disincentives

3.3.1 Prices of inputs

In the process of diffusion and adoption, the prices of inputs have an effect on the decisions of farmers. Dinar and Yaron suggest that past and future price expectations for inputs and outputs have importance in investment decisions for new equipment, which is mentioned in their study as irrigation equipment [70]. Moreover, the rise in these prices affects the use of modern technologies positively. In contrast to this study, the findings of Dinar et al. indicate the possibility

that although the price of cotton increases and thus the area allocated for cotton increases, the amount of farms equipped with modern technologies decreases in their study [10]. With these two opposite results, the study of Abdulai and Huffman concludes that the effect of price on the adoption and diffusion depends on the phases [19]. While the expected price of the new technology affects the diffusion process positively and significantly in the early adoption phase, it is shown that the price of new technology in the second phase does not have a significant effect on the adoption. The positive effect of the expected price can be shown by the result of its diminishing time and accelerating adoption effect, in their study.

3.3.2 Risk

Risk is one of the most commonly addressed issues about technology adoption in agriculture, in the literature. The study by Koundouri et al. indicates that risk has an important effect on the adoption decision process for a new technology [7]. They assert that farmers tend to invest in and adopt new technology more to avoid the production risk they encounter with the risk of extreme outcomes. In their study, it is shown that farmers who face adverse climatic conditions adopt new technologies to decrease the risk level. Uncertainty about the profitability, which is a risk factor, also increases the probability of farmers' adoption because the adoption of new technology decreases the production risk, risk premium and relative risk premium [7, 11]. If the producer is decisive in adopting, the adoption degree is also affected by the risk factors [50]. For instance, the larger farmers allocate relatively less land and hence smaller proportion of their incomes to risky activities, which means the higher input of fertilizer per acre [29]. In this sense, Hiebert suggests that while risk-preferring farmers tend to use more land and fertilizer for production than the risk-neutral farmers, risk-neutral farmers tend to use more inputs than risk-averting farmers [45]. Correspondingly, Wossen et al. assert that, in the adoption decision process, risk-averse households trust their social capitals in terms of adoption decision more than risk-loving households [61]. Networks and traditional sharing norms such as the social capital affect the risk-mitigating measures negatively [71]. On the other hand, Wossen et al. suggest that the relationship between social capital and risk aversion changes among households [61]. While some results show a significant and negative effect on risk aversion, some show a positive effect in their study.

3.3.3 Credit accessibility

No doubt that having a financial potential is necessary while experiencing an innovation. Financial assets also have an influence on adoption, and hence credit constraint leads to different adoption rates [17]. In the literature, there are many studies that show the importance of credit accessibility in the adoption. Access to credit facilitates investment because it provides the support for liquidity requirements [61]. Through this support, farmers may buy the inputs such as fertilizer and benefit from the facilities more easily; it also influences the change in planting dates and using irrigation systems positively [26]. By depending on its contributions to the agriculture, most studies indicate that credit availability encourage people to adopt, and increase the adoption by accelerating [19, 27, 59]. However, on the other hand, Husen et al. state that the effect of credit availability on adoption depends on the technology because they show that while credit access has a positive effect on the adoption of one of the technologies, it has a negative effect on the adoption of other technology in their study [60]. This negativity has been explained in that credit access may direct people to non-agricultural sectors for the investment. From

a different perspective, Abdulai and Huffman emphasize that if credit constraint can be substituted with another financial source such as household savings, the constraint does not influence the adoption [19]. Correspondingly, in the literature, traditional community networks, friends and relatives have been also considered as a financial source in case of lack of credit [29, 61]. Husen et al. confirm the positive financial effect of social capital on adoption by supporting this idea [60]. Relaxing effect of credit on the liquidity can be provided to a member in a credit or saving organization, these kinds of organizations relax the farmers in terms of cash constraints [61]. Abdulai et al. show that credit access has an effect not only on the investment but also on the crop choices because the farmers suffering from credit access have a liquidity problem and this directly affects the crop choices [27]. When considered from this aspect, smaller farmers who have limited credit have less advantage than larger farmers having better credit opportunities in terms of adoption [17]. Moreover, the study of Abdulai and Huffman indicates that, in the situation of credit constraint, if farmers just consider the current generation, higher adoption is expected in the middle-aged farmers; but if there is no credit constraint and farmers behave by considering future generations, higher adoption is expected in younger farmers [19].

4. Policy implications and future directions

As shown in the table, when the literature about the adoption and diffusion of technology in the agriculture is reviewed, it is clearly seen that while some factors have an exactly positive effect on adoption, some have negative effects and some are changeable depending on the situation. This review reveals the significant factors and makes the policies that may be implemented to expedite the technology adoption and diffusion more explicit.

When the knowledge of people is taken into consideration, the factors including education, learning and information play a big role in adoption. In this sense, governments should provide a good and an extensive extension service not only in central locations but also in remote locations because as it can be seen in the literature, being closer to opportunities increases the likelihood of adoption and diffusion. Removing the disadvantages of farmers who live in remote areas and providing accessibility to information will contribute to the adoption.

Some organizations and activities that strengthen social relations and facilitate communication among people also need to be set up regularly by governments because social capital is another significant part of the adoption and diffusion of technology. Encouraging farmers to be a member in a group or an organization develops social capital and this helps to obtain information and learning by others. Besides, when the effect of neighbors and relatives is considered, some key persons who can be trained and affect more people can be chosen by the government. These key persons who communicate with their immediate circles such as relatives and neighbors can produce a 'butterfly effect' and increase the adoption and diffusion of technology. In this sense, finding the key person on the farmers is important. For that reason, governments need to prepare specific and different programmes for households having different social identities. These aim-targeted plans will affect the tendency of farmers for adoption positively.

On the other hand, financial aid should be provided for farmers to ease technology use because the literature shows the dominant positive effect of ownership and wealth on adoption and diffusion of technology. Liquidity problem of farmers can be solved with special funds, credit facilities with low interest rate and subsidies provided for new investment and new crops. These opportunities both relax farmers and increase

the tendency of farmers to adopt the new technology. Undoubtedly, all of these are related to good and reliable governance; thus, if it exists, the adoption and diffusion of technology can be expedited.

Although education and learning are very big parts in the adoption and diffusion of technology in agriculture, it is necessary to do more detailed research on the dilemmas that 'Although communication and network are very effective on the adoption and diffusion, does learning by others without official agents cause mislearning and misuse of technology and hence to detract farmers from technology adoption or not?'

Future research may also analyze the effects of communication among relatives or neighbors of farmers on adoption because although they have mostly positive effects on adoption, there are also negative effects because of the idea of refusing to provide benefit for others. In brief, more precise results can be obtained about the effect of social capital on adoption and diffusion of technology by examining social relations in detail.

5. Conclusion

In these times when the competition among firms, sectors and countries is very strong, the importance of technology as a formula of long-run economic growth is great [72]. Technology contributes to the sectors in terms of real wages, increasing productivity and decreasing costs and prices [2]. These positive effects make it a big player in the agricultural sector for development [3]. Thus, the adoption and diffusion of technology in agriculture are very important to maintain continuity in production, increase production and generate more income.


In this paper, the factors influencing the adoption and diffusion of technology in agriculture had been analyzed and the results and policy that can be implemented were presented. The literature review includes 44 analyses and 21 factors on the adoption and diffusion of technology in agriculture. Analyses had been handled on a large scale rather than examining a specific time period or a specific region. For the section of characteristics of farmers, the results show that there is a predominantly positive relationship between income, off-farm income, ownership and wealth, education, learning, information, neighborhoods, relatives, contacts with extension agents, membership in a group or organization and experience with the adoption and diffusion of technology. General inference by looking at the effects of age, gender and norms cannot be made because although they have an effect on the adoption and diffusion of technology, the positivity and negativity vary by situation. For the section of the characteristics of farms, it is seen that both factors which cover the farm size and the closeness to the farm have positive effects on adoption. The following section, on external incentives and disincentives, indicates that prices of inputs and credit accessibility show a predominantly positive effect on the adoption and diffusion of technology in agriculture whereas the effect of risks changes. Although the factors examined reveal the main elements for the adoption and diffusion of technology in agriculture, future research may show more precise results for the uncertain factors depending on the situation.

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Edited by Shaon Kumar Das

Organic crop production is the science and art of growing field crops, fruits, vegetables, and flowers by adopting the essential principles of organic agriculture in soil building and conservation, pest management, and heirloom variety conservation. This book provides detailed insights into organic farming in agriculture, biological efficacy in the management of plant diseases, organic nutrient management, socio-economic dimensions of adoption of conservation practices, nonchemical weed control, plant growth promoting fungi for phytostimulation, nanotechnological approaches, and finally vermicomposting. The book primarily focuses on research and development based organic agriculture and horticulture production technologies, and has attempted to abridge information on organic crop production of the major food grain crops. The book also contains comprehensive information on the various related dimensions of organic crop production.

Published in London, UK

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