

Trichoderma as a Biocontrol Agent and Biofertilizer in Sustainable Agriculture

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INTRODUCTION

Over the last few decades, agricultural systems have developed to provide a safe, healthy, and high-quality global food supply. Agrochemicals, including synthetic fertilizers and insecticides, play a crucial role in enhancing crop productivity and ensuring food security [1, 2]. However, there are several concerns about the environmental impact of using these agrochemicals. Excessive use of synthetic pesticides and fertilizers has resulted in adverse effects such as resistant plant diseases, pollution, soil degradation, decreased fertility, health risks, and environmental concerns [1, 3, 4]. For solving these issues, it's important to find safe, affordable, and environment friendly solutions. Biocontrol agents and biofertilizers are eco-friendly alternatives to synthetic chemicals that are often used in recent year.

Fungi are commonly used as biological control agents in forestry and industrial agriculture. Recent research has shown that fungi play a crucial role in increasing plant yield and productivity by controlling pest and pathogens different fungi, including *Aspergillus* *Ampelomyces*, *Candida*, *Coniothyriu*, and *Trichoderma* are widely used as biocontrol agents now a day's [5-8]. *Trichoderma* is the most active fungus in controlling harmful fungi and promoting plant growth [9]. The paper aims to highlight the role of *Trichoderma* as a biocontrol agent and biofertilizer, including current advances and mechanisms. It will also provide a brief overview of commercially available *Trichoderma* products and their applications.

TRICHODERMA

Trichoderma is a genus including a diverse collection of fungal species. Anamorphic stage belongs to Hypocreales are characterised by rapid growth, high conidial spore generation, and the ability to form sclerotia [10]. *Trichoderma* species is common saprophytic fungi found in soil and root environments [11-14], as well as on rotting wood and their dead and decaying organic materials [15-19].

Trichoderma strains can be mycoparasites [20] and may be antagonistic to other fungi that use their cell walls and cytoplasmic parts for nourishment. They produce few pigments, like conidia are generally green in colour but sometimes may be colourless, yellowish and shades of brown to grey a greenish-yellow to a reddish tinge, and colourless strains may also be available. Conidia can vary in colour from drab to various tints of green, dim, or earthy [10]. The hyphae of *Trichodрма* are septate and translucent. The conidiophores are short, translucent, and branched, giving them a pyramidal appearance. They bear phialides that can appear singly or in groups. The spores are translucent, ovoid-shaped, and borne in small terminal clusters at the tips of the phialides [21, 22].

Trichoderma is not often considered as harmful fungi as well as harmful to plant growth. Their presence as biofertilizers in the rhizosphere can help plants grow and flourish [10]. In addition, numerous studies have shown that *Trichoderma* species can colonise roots as symbionts and initiate systemic and localised resistance in plants, protecting them from a variety of pathogens such as fungi, bacteria, viruses, and even insects [10,11,18,23], as well as promote plant development and growth [24]. Additionally, *Trichoderma* may grow well in highly competitive environments because to their innate resistance to antibiotics generated by competing microbes [25]. In economic terms, they have a wide range of advantages for a variety of biotechnological applications, particularly in industrial, agricultural, and the natural environment [10].

TRICHODERMA AS BIOCONTROL AGENT

Plant diseases induced by infections have resulted in lower agricultural output reduces growth and developme-

nt. Pathogens, especially some fungal strains, are primarily responsible for developing plant diseases that result in a significant loss of yield [14, 26-28]. The use of synthetic fungicides is one of the most common methods for preventing yield losses caused by pathogenic fungi [14, 27, 29]. However, they may have adverse effects for the environment and human health [30-32], and they may also result in the development of resistance to pathogenic fungi attacking plants [33]. Over time, as the pathogen develops resistance, current synthetic fungicides may become ineffective, therefore it is need to the introduction of different fungicides for future successful disease management [14]. As an alternative agro-practice, microorganisms can be used as because they have biocontrol agent the potential to be an effective and strong approach for controlling plant diseases [14, 34]. This strategy is based on the antagonistic connection between such organisms and the target pathogens [35]. For example, soil-borne plant diseases may be treated by using different microorganisms present in it is a ecofriendly the soil method of plant disease control [36].

In recent years, fungi have been identified as promising biocontrol agents and alternatives to chemical use. According to Köhl et al. [37], biological control is the process of reducing the quantity of pathogenic microbes by the application of antagonistic microorganisms. Biocontrol systems can act in a variety of circumstances. Unlike other biocontrol agents like as viruses and bacteria, fungi do not require ingesting to infect their hosts, instead, they assault directly via the organisms. *Trichoderma* is frequently used in the creation of biocontrol agents. Biocontrol agents are microorganisms capable of resisting and suppressing plant disease-causing pathogens. [10] They are better to use than chemical insecticides while being as effective. Biocontrol drugs also have no negative effects on other species that promote plant growth, health, and production. An effective biocontrol agent should be genetically stable, mass-produced effectively with low-cost medium, and effective at low concentrations against a variety of diseases [38, 39]. In addition, the biocontrol agent must not be poisonous to people, pesticide-resistant, and pathogenic to the host plant [14, 39, 40].

Trichoderma is unique ability to generate mycolytic enzymes, along with its non-phytopathogenic nature, renders it a very appealing biocontrol agent [41] in relation to other fungal species. Due to their biocontrol capabilities *Trichoderma* perform biocontrol activity through a variety of mechanisms, including mycoparasitism or parasitic antibiosis, resistance promotion, nutrition and space competition with plant pathogens [12, 15, 20, 33, 42]. Furthermore, it has been found that altering the root architecture enhances the plant's resilience to diseases, as well as destroying the root-knot nematode at various stages of its growth phase and particular segments of adult worms [43]. Numerous bioactive chemicals released by *Trichoderma* species and their significance in biological control. The fungal biocontrol mechanism may be divided into two modes viz. direct and indirect.

***Trichoderma*: Direct Biocontrol Activity**

Mycoparasitism

Many studies have shown that *Trichoderma* can prevent various plant diseases by inhibiting plant pathogens, which are mostly present in the rhizosphere, via mycoparasitism or hyperparasitism [45, 46]. It refers to the method by which *Trichoderma* survives at the cost of another plant parasite by colonising it or living inside it [47-49]. For example, hyper-parasitic *Trichoderma harzianum* is used to treat choking disease caused by the fungus *Epichloe typhina* in the grass species *Puccinellia distans* [47]. This approach has been widely used to tackle plant infections like *Sclerotium rolfsii*, *Botrytis cinerea*, and *Rhizoctonia solani* [50]. Trichodex, a commercially available *Trichoderma* formulation, is commonly used to alleviate plant diseases [47, 51]. Mycoparasitism is a frequent mechanism that involves an antagonistic intimate action with a pathogen. *Trichoderma*'s mycoparasitism involves numerous phases, including pathogen detection, binding to the target, enzymatic destruction of the fungal cell wall, and cytoplasmic content absorption [10, 12, 14, 52]. *Trichoderma* may adhere to a target pathogen by attaching its cell wall carbohydrates to the pathogen's lectin. When *Trichoderma* comes into contact with the target fungus, it forms appressoria. *Trichoderma* may generate cell wall degrading enzymes such as amylase, cellulase, chitinase, glucanase, lipase, pectinase, protease, and xylanase [53], as well as volatile chemicals such as 6-pentyl- α -pyrone (6-PAP), toluene, D-limonene, and α - bergamotene [54]. Other metabolites can also be produced. Cell wall degrading enzymes serve an important role in promoting the hydrolytic breakdown of the fungal pathogen's cell wall, which is composed

of chitin and glucan polysaccharides [10]. *Trichoderma* may create large amounts of extracellular enzymes that breakdown cellulose and chitin [10]. According to Howell [15], the prior breakdown of the cell wall of *Botrytis cinerea* and *Fusarium oxysporum* by lytic enzymes facilitated the entry of antibiotics into pathogen cells.

***Trichoderma* Performs Indirect Biocontrol Activity**

Antibiosis

Most fungal species are capable of secreting one or more compounds with antibiotic properties. Many *Trichoderma* species can produce low molecular weight secondary metabolites with antibiotic properties which can interfere with the development of various microorganisms through inhibition [10]. This inhibition action is called antibiosis. It is another mechanism found in *Trichoderma* for controlling plant pathogens, which depends on the secretion of antimicrobial compounds or specific secondary metabolites that exhibit inhibitory properties. There are more than 180 secondary metabolites of *Trichoderma* that has been characterized into various classes of compounds [44, 55]. These compounds have different roles in the biological control of different pathogens. Some secondary metabolites have the capability of modifying plant metabolism and growth. *Trichoderma* strains, for example, *Trichoderma viride*, *T. harzianum*, and *Trichoderma koningii*, are capable of production and secretion of a volatile metabolite, 6-PAP which is responsible for the biocontrol of several pathogenic species such as *Botrytis cinerea*, *Rhizoctonia solani*, and *Fusarium oxysporum* [10]. According to Harman [11], different groups of metabolites might play an important role as plant resistance inducers

Competition

Some studies suggest the existence of a competition between fungi and pathogens as another mechanism of biocontrol. *Trichoderma* may compete for essential nutrients and limit the growth of pathogens [56]. Apart from nutrients, *Trichoderma* can also compete with other pathogens for space including infection sites on plant roots [10]. *Trichoderma* can easily colonize the rhizosphere environments and cause significant changes in plant metabolism [11, 42]. The mechanisms which aid in the colonization of the diverse ecological niches are exceptionally evolved and vary in *Trichoderma* [12, 57, 58]. Moreover, *Trichoderma* is resistant to toxic metabolites that are produced by plants such as flavonoids, terpenoids, phenols, and phytoalexins in response to infection. *Trichoderma* can defend plants against pathogens through a physical interaction between *Trichoderma* and the plants up to the outer layer of the epidermis and the root bark [42]. Acidify their surrounding environment to produce adverse conditions for the growth of pathogens [10, 33]. Furthermore, the wide metabolic versatility of many *Trichoderma* species has enabled them to use different and complex carbon and nitrogen sources, which allow *Trichoderma* to limit competing pathogens from growing and spreading in the environment [25].

Induction of systemic resistance

Trichoderma can induce plant resistance to pathogens through biochemical and physiological modifications caused by complex interactions the microbial biomolecules and plant receptors [11, 59, 60]. This triggers the formation of the defense enhancing chemical, like salicylic acid in the entire plant as a systematically induced defense mechanism and also by inducing the synthesis of regulatory plant proteins that could help in the defense mechanism by recognizing the microbial biomolecules [61]. It is also documented that the induction of systematic and local resistance in a plant is further dependent on the plant species, pathogenic microbial species, environmental factors, and rhizosphere and symbiont relationship [62]. This causes the formation of the defense-enhancing chemical, salicylic acid, throughout the plant as a systematically induced defence mechanism, as well as the synthesis of regulatory plant proteins that aid in the defence mechanism by recognising microbial biomolecules [61]. It has also been established that the induction of systematic and local resistance in plants is influenced by plant species, pathogenic microbial species, environmental variables, and the rhizosphere-symbiont connection [62].

TRICHODERMA AS BIOFERTILIZER

Biofertilizers are defined as substances containing beneficial living microorganisms that colonize the soil ecosystems and promote plant growth by increasing the supply of primary nutrients to the host plants [1, 63, 64], improve soil chemical and biological properties, phosphates solubilization, and agricultural productivity [65]. Biofertilizers have many benefits compared to synthetic fertilizers such as they increase absorption of mineral nutrients in plants, minimize leaching, and are involved in composting of solid wastes [14, 63, 64, 66]. One of the main sources of biofertilizers is fungi that are used for the inoculation of seed and soil to increase nutrient availability for plants [67]. They can restore the nutrient cycle in the soil, form soil organic matter by releasing growth-promoting hormones and improve root proliferation [1]. These biofertilizers are capable of fixing atmospheric nitrogen, decomposing organic materials, and solubilizing soil phosphates, which are beneficial in improving soil fertility as well as enhancing plant growth through the supply of nutrients [14, 68]. Furthermore, they are environmentally friendly and are safe for crops and other plants [69].

Trichoderma species are very well known biofertilizers for promoting plant growth and development, and are regularly utilized by most crop types with or without amendments [19]. They are well known to improve plant nutrient uptake, production of growth hormones and provide plant protection from pathogens [70, 71]. According to Chang and Baker [72], when *Trichoderma* are applied regularly to plant soil, they promote plant growth and seed germination, development of roots, and improved the in height and weight of the plant. *Trichoderma* are known to improve overall plant health and produce different types of secondary metabolites including growth hormones and certain enzymes, and to benefit plants through *Trichoderma*-plant interactions [19, 33]. From these interactions, it is evident that plant growth is stimulated from the production of vitamins, increased availability of biogenic elements such as nitrogen and phosphorus, mobilization of nutrients in soil and plants, and improved mineral uptake and transport [10].

Mastouri *et al.* [78] stated that these fungi mobilize nutrients in plants to increase crop yield. Some *Trichoderma* strains can produce organic acids such as gluconic acid, citric acid, and coumaric acid and acidify the surrounding environment to solubilize phosphorus ions, micronutrients, and mineral cations, for example, Fe^{3+} , Mn^{4+} , and Mg^{2+} , which then become available in the soil for plants utilization [11, 19, 59]. *Trichoderma* strains are also known to produce phytohormones to promote root and shoot growth in plants [11, 80]. As an example, *Trichoderma* can produce zeaxanthin and gibberellin compounds that help in rapid seed germination [10]. The fungal biofertilizer can be used as a soil conditioning agent which helps in improving the diversity and concentration of plant beneficial microorganisms thus helping in the mitigation of greenhouse gases (carbon dioxide and methane) that cause climate change [19]. *Trichoderma* strains can also eliminate minor pathogens in the rhizosphere to maximize the potential of plant growth [10, 81]. Further, their ecofriendly and human friendly nature has attracted various researchers and farmers worldwide, increasing their usage and application [76]. The phytohormones and enzymes produced by *Trichoderma* eventually help in promoting plant growth, yield, nutrient absorption, phosphate solubilization, mineral absorption, antioxidant activity, and soil conditioning [82, 89]. They are also reported to improve the nutrient absorption quality and yield of many vegetables that include brinjal, tomato, sugar beet, chillies, potato, cauliflower, onion, peas, and also other plants such as groundnut, cotton, wheat, tobacco, Bengal gram, sugarcane, red gram, banana, soybean, citrus, and sunflower [19, 74, 90]. Phytohormones play a key role in the plant growth, especially during the shoot and root elongation. Appropriate levels of phytohormones in the requisite time of support would enable the increased and healthy plant growth which generally leads to the increased crop yield [88]. The presence of *Trichoderma* in the germination stage of a seed helps in the improved production (balanced quantities) of growth hormones such as gibberellic acid and indole-3-acetic acid (IAA) which improves the germination rate and seedling vigor [73]. Similarly, phosphate being an important nutrient that plays an imperative role in the plant growth and development is always inadequately absorbed by the plants or inadequately available for plant's absorption especially in the acidic soil [77]. Many *Trichoderma* species, specifically *T. harzianum* and *Trichoderma reesei*, are well known for their ability to solubilize phosphorous with the production of a specific enzyme phytase which on reaction with insoluble tricalcium phosphate solubilizes it and makes it available for plants absorption [74, 86, 91]. On the other hand, *Trichoderma koningiopsis* produces alkaline phosphatase enzyme for the effective solubilization of the insoluble phosphate available in the soil [87]. As nutrients are the essential compound promoting the plant growth and development, an altered uptake of

nutrients can result in the malnourished crops with stunted growth and reduced crop yield. *Trichoderma* application as biofertilizer improves the uptake of nutrients in crops and also the availability of nutrients in soil for crop's absorption. Yadav *et al.* [75] have suggested that the presence of *T. viride* in soil improved the uptake of nitrogen, phosphorus, potassium, and organic carbon in the sugarcane crops. Another report stated that the presence of *Trichoderma* in rhizosphere can lead to the colonization of crop roots as endophytes, which in turn enabling the solubilization, better availability, and improved absorption of nutrients by the crop [92].

COMMERCIALLY AVAILABLE *TRICHODERMA* BASED BIOACTIVE COMPOUNDS AND BIOFERTILIZERS

Several commercial formulations of *Trichoderma* as bioactive compounds and biofertilizers are known and are used worldwide. Moreover, many initiatives and studies were carried out by various researchers and commercial agencies for the development and usage of *Trichoderma* as an agent controlling plant diseases and enhancing plant growth [93]. As these compounds are prepared to support the plants during adverse environmental conditions, it is highly essential to stipulate standard norms for the production and the maintenance of the compound as altered environmental conditions such as pH, temperature, and so on could influence the formulation's activity. Hence, it has been stipulated that the standard formulation should have a shelf-life period of 2 years, easy handling procedures, and should have an endurance to the temperature ranging from 5°C to 35°C [94]. It should also not be phyto-toxic, cost effective, easily get dissolved in water, easy, and cheap availability of carrier materials, and should be compatible with agrochemicals [95]. Various formulations available worldwide with its applications. Further, these formulations are available in two different forms namely solid and liquid forms. Kumar [95] have suggested that the solid formulations are available as dry dust, wet dust, capsules, and granules at the concentration of 10⁸ to 10⁹ propagules available in a gram of the carriers such as clay, compost, and adhesives such as arabic gums and carboxymethylcellulose. Further, they are formulated by pulverizing and grinding the dried *Trichoderma* mats which are further mixed with the appropriate carriers for different applications. Similarly, liquid formulation is limited in production due to difficulty in handling and maintenance. They are generally applied through irrigation of the crops [94].

CONCLUSION

This report reviews the importance of the application of *Trichoderma* as a biocontrol agent to control diseases in plants by suppressing the growth of the fungal pathogens and as a biofertilizer to improve plant growth and development in industrial agriculture. These eco-friendly alternatives can substitute the excessive use of synthetic products that can cause problems in the long term. There are many commercial frequently used mycofungicides and biofertilizers available that are produced from fungi such as *Trichoderma*. The biotechnological advances from these microorganisms, such as fungi, are immense and yet to be explored. Thus, more studies need to be conducted on the development of sustainable biotechnological applications of the products developed on the biocontrol agents and biofertilizers.

FUTURE NEEDS AND LIMITATIONS

Although several studies have been carried out on the beneficial role of *Trichoderma* spp. as a biocontrol agent and biofertiliser, it is essential to well define the mode and mechanism of the species-specific and pathogen-specific antagonistic behavior expressed by the fungi. Also, it is imperative to ascertain the dose concentration of the plant growth-stimulating metabolites and their activity specific to each plant species.

AUTHOR'S CONTRIBUTION

All authors of the paper have made substantial contributions in the conceptualization and designing of the manuscript, data acquisition.

CONFLICT OF INTEREST

No conflict of Interest

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