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Trichoderma spp.: characteristics and applications

Abstract

Trichoderma is the genus of a cosmopolitan fungus, common in different types of environments or agroecosystems. It has different properties that give it beneficial qualities and functions, which is why it is given different uses, for example, as a biocontrol agent, through induced or direct systemic resistance, as a plant growth stimulant, improving the absorption of water and nutrients and producing phytohormones, as a bioremediator, among others. The objective of this work was to compile knowledge of the characteristics of the genre *Trichoderma* and the different applications it can have.

Keywords: antagonist, biocontroller, growth stimulant

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Introduction

The genus *Trichoderma* is taxonomically located in the *Hypocraceae family* (Table 1), it is characterized by not presenting a specific sexual state.[1,2] This genus has more than 30 species, all with beneficial effects for agriculture and other branches.[3]

Table 1 Taxonomy of the genus *Trichoderma*

- **Fungi**
  - Division: Ascomycota
  - Subdivision: Pezizomycotina
  - Order: Hypocreales
  - Class: Sordariomycetes
  - Family: Hypocraceae

The sexual state was described in some isolates, within the genera *Hypocrea, Podostroma, Sarawakus, Aphysiostroma and Protocrea,* with *Trichoderma* being a group of clonal derivatives that lost the ability to complete the sexual cycle. Therefore, *Trichoderma* is asexually reproduced with mycelium and an abundant amount of green conidia (or spores), they are formed from naked cells of its fruiting body, it is fast growing and has extensive production of enzymes.[4] The conidia are ovoid in shape, formed from highly branched and septate conidiophores (Table 2). It also has resistance structures, called chlamydospores, formed on the mycelium with a thick and rough cell wall. The species most used for biological control are: *T. atroviride* ; *T. harmatum* ; *T. asperellum* and *T. harzianum.*[5] *Trichoderma* is a free-living, facultative aerobic, plant-symbiont, avirulent fungus. It is easy to find it in agricultural soils, as well as in other environments and particularly in roots.[3,4] *Hypocrea lixii* was recently identified as the sexual state of *T. harzianum,* a species widely cited in biological control.[5]

Table 2 Morphological characteristics of the genus *Trichoderma* spp

Trichoderma spp.	Morphological characteristics	Asexual reproduction abundant mycelium
		Green conidia (abundant, fast growing).
		Phialides Conidiophores (branched) Chlamydospores
	Physiological characteristics	Enzyme production
		Production of metabolites (volatile and non-volatile)

This genus has a great capacity for colonization and adaptability, it has the possibility of inhabiting different soils (agricultural, forestry, acidic), substrates with different contents of organic matter and a wide range of temperatures. Due to its different capacities, it is placed in the group of hypogean, lignolocultural fungi.[4,6]

The best conditions for the growth and development of this fungus are:

1. **Development temperature:** 15 – 35 °C, the optimal temperature being 25 °C.
2. **Relative Humidity:** grows at 20 to 80%, with an optimum of 70%.
3. **pH:** survives in a wide range since it has the capacity to acidify the environment in which it is found (secretion of organic acids), the optimal range for development being 6 to 6.5.[7]
4. Light is a factor of great importance for development, sporulation and the production of secondary metabolites.[4]

Some *Trichoderma strains* colonize specific sites on the root surface, for example, the entire surface layer (epidermis) or the first or second layers below it.[6]

The correct choice of a promising strain of *Trichoderma* according to the desirable qualities it presents, generates benefits in the modes of action and ecological aptitudes, these being specific to the species or strain in particular to develop its full potential in a successful commercial product.[1,4] Also, *Trichoderma* spp. it is known for its varied application, from seed coating to post-harvest, and from soil to leaves.[1] Its capacity as a biocontroller is due to the multiple mechanisms it possesses: it synthesizes volatile and non-volatile compounds, such as trichodermin, peptide metabolites, gliotoxin and viridin, suzukacillin, dermadin, trhocziamine, gliovinirna and tetracyclic diterpenes, triterpene viridin and sesquiterpenes,[8,9] as antagonistic compounds (proteins, enzymes and antibiotics) and plant growth promoters (hormones and vitamins).[9] It also generates competition for nutrients, space and myco-parasites.[6,10–12]

In turn, *Trichoderma* has several advantages as a biological control agent. It takes nutrients from the fungi it parasitizes, which it degrades, and from organic matter, helping its decomposition, which is why the incorporation of organic matter and composting favor it.[3] Among the nutritional requirements of *Trichoderma* are complex substrates such as: starch, pectin and cellulose among others that can be degraded, and it can also assimilate nitrogenous compounds such as: nitrites, urea, amino acids and ammonium sulfate, while the trace elements required for its growth are: iron, zinc, copper, molybdenum, manganese and vitamins, being required in concentrations of around 10^{-9} M.[13]

Mode of action as antagonist

Antagonists can present more than one way of acting, this is of great interest when choosing the biological control agent,[14,15] in this way the resistance of the strains.[16] As for the mechanisms, it depends on each strain of *Trichoderma* and the environmental conditions. The antagonistic mechanism of action of *Trichoderma* was recorded by Weindling in 1932. Currently, indirect and direct actions are known, the latter regulate the development of phytopathogenic fungi.[4,8,15] Among the direct actions are known: mycoparasitism, antibiosis and competition for nutrients and space. Guedez et al.,[17] consider that mycoparasitism is the main mechanism of action of these fungi and the best known.[10]

Mycoparasitism

It is the direct impact of *Trichoderma* on another species of fungus. Currently, 75 species of *Hypocrea / Trichoderma* are known to have this capacity. The processes described below occur sequentially and continuously.[8,12,18,19]

Chemotrophic growth: It is direct growth towards a chemical stimulus. In the host localization stage, *Trichoderma* can detect the phytopathogen from a distance and its hyphae grow in the direction of it.

Recognition: Recognition is carried out through lectin-carbohydrate interactions, with carbohydrates present in the cell wall of *Trichoderma*, while lectins are found in phytopathogens. Then the process continues with the development of hyphae and appressoria. These are considered effective only against specific phytopathogens, so molecular recognition between *Trichoderma* and the phytopathogen (host) is essential for antagonism.

Adhesion and coiling: After recognition, *Trichoderma* hyphae adhere to the host's hyphae through hyphae and appressoria that coil around the host. The adhesion of *Trichoderma hyphae* occurs through the carbohydrate-lectin association.

Lytic activity: At this stage there is production of extracellular lytic enzymes (chitinase, cellulase, glucanase and proteases), which degrade the cell wall of the phytopathogen and resistance structures that enable the penetration of the hyphae of the antagonist, as well as facilitate the insertion of specialized structures (hyphae, haustoria) for the absorption of nutrients from the interior of the phytopathogen. Finally, mycoparasitism ends with the loss of the cytoplasmic content of the phytopathogen cell. The remaining cytoplasm is found surrounding the invading hyphae, with symptoms of disintegration, retraction of the plasma membrane and disorganization of the cytoplasm.[6,8,15,17]

Some of the research shows the action of *Trichoderma* against *Rhizoctonia solani*, *Alternaria alternata, Sclerotinia sclerotiorum*, *Fusarium* spp., *Botrytis cinerea, Pythium* spp. and *Ustilago maydis,* where the deterioration of phytopathogens is confirmed.[12] Studies through microscopic observations highlight that it is not always feasible to visualize these interactions, since it depends on the *Trichoderma isolate* and the phytopathogenic agent in question.[15]

Antibiosis

It is the direct action of antibiotics or toxic metabolites produced by one microorganism on another. Many strains of *Trichoderma* produce volatile and non-volatile secondary metabolites, some of which inhibit the development of other microorganisms, without reaching physical contact; such inhibitory substances are considered antibiotics. More than 180 secondary metabolites are known to be produced by the genus *Trichoderma,* which they are grouped into peptaibol, polyketide and terpene according to their biosynthetic origin. The ability of a *Trichoderma strain* to simultaneously produce different antifungal compounds reduces the risk of resistance in microorganisms.[15] As in the case of *T. viride,* which produces a wide variety of antibiotics, including trichotoxins A and B, tricodecenins, tricorovines and trichocelins.[12] Howell[10] determined that *T. virens* inhibited the growth of *Pythium ultimum* and a species of *Phytophthora*, but the same did not occur with *Rhizoctonia solani, Thielaviopsis basicola, Phymatotrichum omnivorum, Rhizopus arrhizus* or *V. dahliae*. It was also proven that the enzyme produced by *Botrytis cinerea*, to degrade the plant cell wall, is degraded by an enzyme secreted by *T. harzianum,* as is the phytotoxin produced by *R. solani*, which is degraded by α-glocosidase excreted by *T. viride.*[15]

Competition for substrates

It occurs when two or more organisms have the same nutritional requirements, but one of them is more competitive, which reduces the amount available for the others. *Trichoderma* due to the speed of growth and development it has, it has the capacity to compete for: substrate, nutrients, space and oxygen.[8,12,15,20] Competition for carbon (C) between *Trichoderma* and phytopathogenic fungi such as *Rhizoctonia solani and Fusarium oxysporium*, as well as for points of infection with rhizosphere phytopathogens.[12] In turn, *Trichoderma,* compared to other microorganisms, has great efficiency in mobilizing and absorbing nutrients from the soil. It can also reduce soil pH through biosynthesis and the ability to release organic acids that facilitate the solubilization of micronutrients. Likewise, *Trichoderma* has the ability to generate siderophores that sequester Fe (Fe^{2+}) from its environment or can insolubilize it by acting as a chelating agent, preventing the availability of this element for phytopathogens.[12,20]

This fungal genus can be added to soil or applied as seed treatments and grows easily along with the root system of the treated seed,[10] this is very important for a biological control agent since it would not otherwise be able to compete. If i didn't have that ability. Competition is also influenced by abiotic factors such as: type of substrate, pH and temperature, among others, with *Trichoderma* being adapted to take available nutrients or survive (as chlamydospores) when they are scarce.[15]

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Uses of *Trichoderma*

Use as a bioremediator

Trichoderma has the capacity to generate enzymes capable of degrading organic matter as well as substances toxic to the environment, important for the bioremediation process.[21] It is known that *Trichoderma* acts as a biodegrading agent for phytosanitary products and degrades groups of highly persistent pesticides in the environment, serving as a soil decontaminant.[3] In a biodegradation study of chloroacetanilide herbicides, such as alachlor and metolachlor, using eight strains of *Trichoderm* a spp., 80 to 99% of alachlor and 40 to 79% of metolachlor were biodegraded after 7 incubation days.[22] Likewise, Sood and collaborators,[12] in research carried out with different strains of *Trichoderma*, found effectiveness as decontaminants, such as *T. harzianum* in the degradation of phenols, cyanide and nitrate, as well as in the detoxification of soils with cadmium. (Cd), while *T. inhamtum* reduced the concentration of chromium (Cr).

Agricultural use

In agriculture, *Trichoderma* is widely used for the various capacities it has, including as a biocontrol agent against phytopathogenic organisms, such as bacteria, nematodes and fungi in diseased plants. It acts indirectly, improving resistance in plants attacked by biological agents, and it also acts as a stimulant for plant growth and development.[12,23] This fungus is also present in organic waste decomposition processes, cooperates in the mineralization and reuse of waste as well as promotes the absorption and efficient use of nutrients and fertilizers.[9,12,21]

Industrial procurement

Trichoderma has the great capacity to grow in different agro-industrial wastes, thus generating byproducts of great value for biological control processes. Currently, it is possible to obtain these byproducts of quality and in sufficient quantity for massive applications, through simple, economical and effective technologies. This generated a line of research with significant potential, due to the diversity and volume of agroindustrial waste generated in productive areas.

There are different studies that evaluate the production of viable spores and in quantity of *Trichoderma.* They generally use the husk or straw of rice, wheat and products from the sugar industry. For the multiplication of *Trichoderma* through liquid and/or solid fermentation. The use of agricultural products with high cellulosic content is a possibility to replace the use of rice and wheat since these have high costs[24] which is why it is presented as alternatively the use of tomato peel (ripe calyx); rice (glumes and lemmas); garlic (leathery cataphylls); cocoa (seed kernel); peanuts (fruit pericarp), grains of rice, sorghum, canary seed and corn; soybean and corn stubble,[24] as well as tree seeds (*Artocarpus incisa*), which can be used to enrich substrates given their high nutritional power (rich in sugars, with 20 to 37%). of carbohydrates, calcium, iron, phosphorus and niacin, etc.).[13] The effect of inoculation of the *T. viride* 137 strain on sugarcane bagasse fermented with its own microbiota was also studied.[25] Allori Stasonelli et al.,[26] concluded that it is feasible to use waste from the sugar industry (bagasse and molasses) for the artisanal production of *Trichoderma*, through solid fermentation.

Use as a biocontroller

The genus *Trichoderma* is part of 60% of the biofungicides registered in the world,[12] where 67% is made up of a single species of *Trichoderma* and the rest are combinations between two or more species of the same genus or with mycorrhiza-forming fungi, bacteria or other biological compounds. Biofungicide formulations can be both liquid and solid, formulated in talc, vermiculite, wheat bran, coffee peel, vegetable or mineral oil, and banana waste. The most used species are *T. viride*, *T. virens* and to a greater extent *T. harzianum*.[5,9,27,28]

Trichoderma not only produces antibiotic substances, but also stimulates plants to produce their own antimicrobial compounds.[6] In studies conducted by Sood et al.,[12] with *T. harzianum* the assimilation of chitinase was evaluated in potato and tobacco plants , and obtained transgenic plants with resistance to both soil and foliar diseases, produced for example by *Rhizoctonia solani* and *Alternaria alternata,* respectively.

There are several investigations on the use of *Trichoderma,* such as, in the post-harvest stage for the protection of fruits and vegetables during storage, there are investigations where 10 isolates of *T. harzianum* and *T. viride were used* to control *Lasiodiplodia theobromae* (banana fruit rot pathogen), as well as an emulsion of *T. harzianum was applied* to apples to prevent decay caused by blue mold after harvest.[12] There are experiences in greenhouses with the *T. harzianum strain* (strain T-22) for disease control, which lasts longer than agrochemicals and is less expensive.[6] A study carried out by Santamaria and Roselló[29] with *T. harzianum* showed the antagonistic capacity against *V. dahliae and Rhizoctonia solani,* inhibiting mycelial growth.[30] Furthermore, Benouzza et al.,[31] evaluated the antagonistic capacity of different strains of *Trichoderma* native to Algeria *as* antagonists against two strains of *V. dahliae*, the strains of *T. artroviride* and those of *T. harzianum* are the ones that presented the greatest antagonistic capacity against the phytopathogen. Thus they also controlled wilting in tomato plants. While other antagonism tests between *Trichoderma strains* against *V. dahliae* resulted with antagonistic potential and the emission of several compounds was evident.[32]

Studies carried out on the growth, severity of the disease and defense of eggplant plants with verticillosis, treated with strains of *T. artroviride* and *T. virens*, also combined with growth-promoting strains such as rhizobacteria, showed that they reduced the severity of the disease and increased growth parameters.[33] Meanwhile, in biological preparations with Harpin protein (Messenger Gold), Mycorrhiza ERS - *Trichoderma harzianum* (T22 Planter Box) and together with Bordeaux broth in different combinations, they achieved efficiency in the control of wilting caused by *V. dahliae.* Among other antagonists, *T. flavus* is one of the most used to control *Verticillium wilt* in eggplant, potato, cotton and tomato.[34]

Use as a growth stimulant

Trichoderma in recent years has also been used as a biofertilizer, due to its ability to stimulate the growth, development and performance of plants, especially the root system due to the secretion of phytohormones that induce the growth of primary meristematic tissue,[15] as well as for its ability to increase the availability and absorption of nutrients (soluble or insoluble) and improve tolerance to biotic and abiotic stress.[6,8,12,35–38]

There is reference to the ability of *T. virens* and *T. atroviride* to increase biomass production and the development of lateral roots in *Arabidopsis silvestre*. As well, there is reference that *T. atroviride* promotes the growth *of Capsicum annuum* and *T. hamatum* promotes both root and height growth of *Pinus radiata.* This effect that *Trichoderma produces* on plants lasts over time; in annual plants it is applied in small quantities or in seeds.[23]

Use as a resistance inductor

The production of inhibitory compounds secreted by *Trichoderma* also has a bioregulatory action, inducing physiological and biochemical defense mechanisms in plants, activating compounds and achieving resistance against the attack of harmful microorganisms. This process can be: systemic, produced by xylanase and cellulase, or localized and transient related to the synthesis of ethylene in plants.[3,6,8] As an example, *T. virens, T. asperellum, T. atroviride* and *T. harzianum* inoculated into plants induce metabolic changes that increase resistance to a wide range of phytopathogens and viruses, producing an effective response.

The spores or other propagative structures of *Trichoderma* are added to the soil and come into contact with the roots of the plants, germinate and grow on the surface of these and lead to the production of at least three classes of substance as defense responses of the plants, preventing infections caused by phytopathogens in the roots. These inducers include peptides, proteins and low molecular weight compounds.[6] The induction of resistance produced by *Trichoderma* is an important component of plant disease control, however, different mechanisms could be responsible for the biocontrol caused by different strains and in different plants. Studies have shown that colonization of *Trichoderma* strains in roots results in higher than normal levels of defense-related plant enzymes, including several peroxidases, chitinase, β-1,3-glucanase, and those of the lipoxygenase pathway. hydroperoxide lyase.[6]

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Conflicts of interest

Authors declare that there is no conflict of interest.

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