



Review Article

Biological Control of Plant Diseases

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Abstract:

In recent years, due to the ever-increasing population, the supply of quality food has become a major concern worldwide. Phytopathogenic fungi cause severe damage to crops worldwide and thus significantly reduce the quality and quantity of agricultural products. On the other hand, reducing the use of chemical pesticides in agriculture is a global goal. Meanwhile, different biological control methods, strategies and approaches are used in the management of plant diseases. Biological control is one of these critical techniques that is currently in priority. Biological control is an environmentally friendly and effective way to reduce the effects of plant diseases. In fact, the rhizosphere of plants supports the development and activity of a large and diverse community of microbial organisms, including plant growth promoting organisms. That these organisms can be used in the biological control of plant diseases. Fungi can be mentioned among these growth enhancing factors. Antagonists of fungi play an important role in the control of pathogenic and plant diseases and are used as biological control agents (BCAs) worldwide. These biological agents use various mechanisms such as activating defense reactions, antibiosis, plant growth stimulation, antibiotic production, and increasing the absorption of nutrients to control plant diseases. Finally, it can be said that the use of these factors significantly reduces the use of chemical fertilizers and pesticides.

Keywords: Pathogenic fungi, biological control, chemical control,

Introduction:

Today, many problems have arisen due to the use of chemical fertilizers and pesticides in agricultural products. In addition to increasing production costs, they lead to irreparable environmental damage in the natural ecosystem [1]. Also, the residues of toxins in agricultural products and their products both directly and indirectly enter the human food chain and cause an increase in diseases such as cancer [2]. On the other hand, every year, a large amount of agricultural products are lost in both pre-harvest and post-harvest stages due to contamination by pathogenic agents. These plant pathogens are very

persistent in their attack and cause direct and indirect damage of 40 billion dollars worldwide [3].

Biological control is one of the best ways to control plant pathogens that are aimed at protecting the environment and pose less threat to human health. The use of biological control to prevent pathogenic agents has been used as an alternative to the use of poisons and chemical fertilizers (Pesticides) [4]. In this regard, the main goal of biological control is to provide management tools in agriculture to control the disease and its agents in plants. Adding different antagonists such as fungi to the soil can reduce the

disease damage to below the threshold of economic loss [5].

Plant pathogenic fungi

The first scientific research on fungal diseases in plants was done by Theophrastus (288-370 BC), a student of Aristotle. In his botanical study, he describes the occurrence of rust diseases on different host species. Plant pathogenic fungi are species that take nutrients from plants and endanger the health of plants. The nutritional needs of each of these fungi are different and depend on their host, these species of fungi are called obligate pathogenic fungi. Of course, there are other types of pathogenic fungi that are closely related to their host and complete the life cycle of the plant, these types of fungi are called facultative pathogenic. Facultative pathogenic fungi grow in different environments than the organisms in which they cause disease. Plant fungi will be divided into three groups, necrotrophs, hemibiotrophs, and biotrophs, according to how they receive energy from plants [6].

A- Biotrophic fungi

Fungi have a large number of hosts that obtain their nutrition from the living tissues of the host plant. *Euromyces faba*, *Botrytis*, *Sclerotinia spp* and *Blumeria (Erysiphe)* are among the most important biotrophic species. That *Uromyces fabae* species cause rust (*Erysiphe*) species of plant pathogenic fungi causing powdery mildew in plants [7]. *Ustilago maydis* and *Cladosporium fulvum* are two other species of, the first species causes downy mildew of corn and the second Causes downy mildew of tomato leaves [8]. *Ustilago maydis* is a plant pathogenic fungus that includes more than 1,500 species and infects a large number of monocots and dicots. It is not economically important, but it is considered one of the most important biotrophic fungi [9].

B- Necrotrophic fungi

Unlike biotrophs, necrotrophic pathogenic fungi are unspecialized and quickly cause the death of the host [10, 11]. This fungus enters the plant through natural holes on the plant and secretes

lytic enzymes and phytotoxins. They are also easily cultivated in unnatural environments. These fungi mostly attack young, weak and damaged plants and can become saprotrophic [12]. Also, these pathogenic fungi act like infections, which include adhesion, host penetration and subsequent necrosis and rotting of plant tissues. Some necrotrophic fungal species include *Cochliobolus*, which causes leaf blight of corn, *Alternaria*, which causes early blight of potatoes, and *Botrytis*, which causes gray mold [13]. Plants resist these fungi in the form of phytohormones, pathogenesis proteins and secondary metabolites [14].

Zymoseptoria tritici is a necrotrophic fungus that infects wheat plants. Which leads to spotting of the wheat plant [15].

Hemitrophic fungi

Hemitrophic fungi display characteristics of both biotrophs and necrotrophs. Examples of hemibiotrophic fungi include *Magnaporthe grisea*, *Phytophthora*, *Pythium*, *Fusarium*, *Colletotrichum*, and *Venturia* [16].

Application of fungi in biological control:

Among fungi, microbes, bacteria and other microorganisms, these fungi are the most studied and used in biological control. The use of fungi in the biological control of plant diseases has several properties. First, fungi improve nutrient uptake and nitrogen utilization in plants. Second, they can help fight against pests such as nematodes and microbial pathogens that infect different parts of the plant such as roots, leaves, and fruit [17]. According to Thomas and Reed [18], the main reasons for the widespread use of fungi in biological control are their efficiency in killing the host, as well as their high metabolic diversity, since fungi play a role as decomposers. As a result, it leads to an increase in the relative safety of the environment and finding suitable isolates. Fungi protect the plant against diseases by processes such as mycocystism, competition for resources with pathogens, antibiosis, conferring induced systemic resistance (ISR) to the host plant, and mycovirus-mediated cross-protection [19]. Due to the improved biotechnological and genetic developments in this field, useful genes in

fungi can be isolated and transferred to the genome of the host plant. It is also possible to increase or control the expression of these genes to improve the ability of biological control [20, 21]. Basal and top fungi are two species of fungi used in biological control. The fungi that are included in the group of basal fungi include *Conidiobolus*, *Entomophthora*, *Erynia* and *Entomophaga* genera, which belong to the Entomophthoromycotina and Blastocladiomycota phylum. Also, species of *Purpureocillium*, *Metarhizium*, *Beauveria*, *Cordyceps*, *Fusarium*, *Trichoderma*, etc., which belong to the Phylum of Ascomycota (Subkingdom Dikarya), are among the above fungi that participate in biological control. Fungal species that are used in biological control include both lower and higher fungi. Among the basal fungi, there are representatives of the phylum Blastocladiomycota, such as *Coelomomyces*, and of the subphylum Entomophthoromycotina, such as species of the genera *Conidiobolus*, *Entomophthora*, *Erynia* and *Entomophaga*. Among the higher fungi, several species of the phylum Ascomycota (Subkingdom Dikarya) have reported potential and are commercialized and applied in the field, such as *Purpureocillium*, *Metarhizium*, *Beauveria*, *Cordyceps*, *Fusarium*, *Trichoderma*, and others [22]

Mbugala et al. [23] provides a complete catalog of fungal biological control agents used against fungal plant pathogens based on modern taxonomic concepts. These researchers listed about 300 fungal antagonists belonging to 13 classes and 113 genera along with target pathogens and relevant plant diseases. According to them, *Trichoderma*, which includes 25 species, is one of the most well-known genera with the greatest biological control potential. A number of fungi such as *Trichoderma*, *Penicillium*, *Aspergillus* and *Fusarium* species are known as plant growth promoters because they stimulate the plant's immune responses when the plant is attacked by pathogens. Also, these fungi are considered as the safest modes for induced systemic resistance (ISR) and growth promotion in crop plants [24].

In another study, it has been stated that *Trichoderma* species are of special importance due to the beneficial uses of this fungi in plant health. They are filamentous fungi and through complex mechanisms such as soil and host root colonization, establishment in a physical space and escape from the reproduction of plant pathogens and simultaneous production of cell wall destroying enzymes, antimicrobial metabolites to destroy the pathogen, inducing plant defense, increasing plant growth and improving plant tolerance to biotic and abiotic stressors play a role in controlling pathogens [25].

Management of plant fungal diseases:

1- Chemical control

To control fungal diseases in agricultural plants, more chemical compounds are used. Despite the increasing use of fungicides since the 1960s, this type of chemical control remains one of the important methods [26]. Although this method of management is important for disease control in a large scale of products, it also has disadvantages, for example, polluting the environment with toxins, bioaccumulation, harmful effects on plants and animals, and human health. Also, the use of these chemical compounds lead to various cancers, respiratory disorders and hormonal imbalance in humans [27]. Most importantly, data from FAO-WHO and the US Food and Drug Administration (FDA) reported that persistent organic pollutants (POPs) are not easily degraded and end up in livestock products such as dairy products, poultry and meat [28]. On the other hand, the high use of these chemical pesticides makes pathogens resistant to these compounds, and as a result, reduces the effectiveness of many chemical control methods [26].

2-Use of resistant varieties

Today, by using breeding programs in products, they create cultivars that are resistant to many diseases. This method has been proven and used and they have shown that they are resistant to all kinds of plant pathogenic pathogens [29]. In addition to the production of resistant crops, genetic programs have led to the production of quality products and reduced costs. For this

reason, this management method is more cost-effective from an economic point of view to control diseases [30]. Using breeding programs in many crops, it was shown that plant diseases such as rice bursting, cotton leaf curl, grape downy mildew and wheat yellow rust were significantly reduced [31, 32].

3- Biological Control

Among the non-chemical methods, biological control has been more appropriate and better for controlling plant diseases. As it is environmentally safe, it is also more economical. Microbial biological control agents for plant diseases are usually fungal or bacterial species isolated from the phyllosphere, endosphere or rhizosphere, which will play an important role in controlling plant pathogens. Among biological control methods, the use of fungal strains has been used more [33]. Before applying this method, a deeper understanding of the relationship between plants and pathogens with environmental factors common in a particular region, especially in the context of many diseases, must be understood.; Biological control in plant pathology is defined as "the interaction of multiple environmental elements aimed at reducing the negative effects of harmful species while promoting the growth of beneficial crops, beneficial insects and microorganismes" [34]. Before applying this method, a deeper understanding of the relationship between plants and pathogens with environmental factors common in a particular region, especially in the context of many diseases, must be understood.; Biological control in plant pathology is defined as "the interaction of multiple environmental elements aimed at reducing the negative effects of harmful species while promoting the growth of beneficial crops, beneficial insects and microorganismes" [35]. Biological control depends on factors such as multiple agonistic and antagonistic interactions between plants and microorganismes living in the rhizosphere and phyllosphere [36] and their use to minimize plant diseases. In general, it can be said that biological control means the use of parasitoids, predators, pathogens, antagonists or competing microorganisms to reduce the disease

population, so that the disease is less prevalent and causes less damage than possible. The main mechanisms for biological control are those that have the most effect on pathogens. Antagonists are effective in the biological control of the disease through different mechanisms directly and indirectly. These mechanisms are explained below.

- 1) Antibiotics: compounds in which an inhibitory or antibiotic metabolite is formed by an antagonist.
- 2) Mycoparasitism: to compounds in which the antagonist receives some or all of its nutrients from the fungal host.
- 3) Induced resistance: induction of plant defense response against plant pathogens
- 4) Increasing growth: microbial biological control while the effects of the disease are reduced, they can increase plant growth through microbial hormones such as indole acetic acid and gibberellic acid.

Other methods involved in the biological control of the disease include the release of extracellular hydrolytic enzymes by antagonists, competition for space and nutrients between organisms, and detoxification of pathogenic agents [37]. The effects of systemic or local resistance caused by microbial biological control on plants are important. So that these fungi or bacteria parts can release bioactive molecules by occupying the root epidermis and outer cortex layers, which causes the separation of the fungal thallus or bacterial colonies [38]. Finally, they change the transcriptome and proteome mechanisms of plants, these changes bring many benefits to the host plant, such as increasing plant growth and absorption of nutrients, as well as creating resistance pathways in plants.

History of Fungal Biological Control Applications

Since ancient times, humans have tried to produce more crops with less disease severity. They have done this by changing cultivation methods and reducing pollution [39]. While today, with the discovery of microorganisms and their

interactions, there are many methods to control pathogens through the use of fungal antagonists. Hartley [40] studied the direct application of biological control of plant pathogens, this researcher inoculated 13 microorganisms that had antagonistic properties to soil contaminated with *Pythium debaryaum* and showed that these microorganisms reduced the root of the plant rots. In 1928, with the discovery of penicillin and its purification in drug production, many studies were conducted on antagonists of plant pathogens and their importance was expressed [41].

The use of fungi in the biological control of plant pathogens:

As mentioned, due to the direct interaction between fungi and plants, fungi can be easily used to control plant diseases. In this case, fungi act as antagonists of plant pathogens, in other words, fungi through several different mechanisms such as the production of metabolites (Antibiotics, Volatile compounds - Ammonia, Cyanide, Alcohols, Esters, Ketones and Enzymes), competition (for space, C, N or mineral resources), parasitism, or induction of systemic resistance in the plant or enhancement of its growth response, control diseases in plants [42]. One of the most well-known fungal genera in plant disease control is *Trichoderma* (Hypocreales), because it has many activities against plant pathogens. This genus includes a large number of species that are usually found in the soil. They also have fast growth and their main role in nature is known as primary decomposers. Also, these genera have been investigated with regard to the ability to produce antibiotics, special enzymes and their potential as biological control agents for commercial exploitation [43].

These fungi can inhibit the growth of pathogenic fungi by inducing plant resistance or as an antagonist, competitor or mycophagy by acting directly against pathogens [23]. Of course, to improve the use of these microorganisms, it is necessary to know how the fungi behaves in the presence of other plant parasitic fungi. As mycoparasites, *Trichoderma* species produce helical hyphae around the hyphae of a pathogenic fungi, forming appressoria, where degrading lytic

enzymes are produced and released, allow penetration of the pathogen hyphae [43]. When these types of fungi act as antagonists, they produce compounds with antimicrobial activities [44]. The most important application of *Trichoderma* spp is resistance to the basidiomycete *Rhizoctonia solani*, this basidiomycete is a well-known plant pathogen that causes disease in the roots of a number of plants.

The fungus *Beauveria bassiana* is a microorganism widely utilized in the field of biological control for insect pests. Studies, as indicated by Sinno et al. [45], have demonstrated that direct interactions between *B. bassiana* and plants exhibit a dual nature. For instance, it has been described how *B. bassiana* provides protection to tomato seedlings against pathogenic attacks caused by fungi such as *Rhizoctonia solani* and the oomycete *Pythium myriotylum*, which in turn lead to plant root rot.

The present era witnesses a growing interest in studies addressing the use of fungi in biological control, a practice that is already widespread. For example, recent research has revealed the existence of complex and important relationships that may exist between plants and fungi such as two species of *Metarhizium* and *Beauveria*, which were previously primarily used to control insect diseases [46, 47].

In study aimed at determining the antagonistic potential of different *Trichoderma* species, isolated from forest soils of central India, against the fungus *Rhizoctonia solani* under in vitro conditions, Parihar et al. [48] reasoned that all *Trichoderma* isolates are characterized by rapid growth and possess the ability to inhibit the growth of *Rhizoctonia solani*. One of the isolates, tentatively identified as *Trichoderma* cf. *longibrachiatum* (isolate number TD14), exhibited the highest inhibition rate of 56.25%."The research by Liu et al.[49] investigated the potential of three *Beauveria bassiana* strains (CBM1, CBM2, and CBM3) as biological control agents against lepidopteran pests infesting wheat and their ability to colonize wheat plants.

Pathogenicity assays revealed that all strains exhibited insecticidal activity against the larvae of *Helicoverpa armigera*, *Spodoptera frugiperda*, *Mythimna separata*, and *Plutella xylostella*, with strain CBM1 demonstrating the highest virulence. Subsequent experiments explored the colonization of wheat plants via seed immersion and soil drenching. *B. bassiana* CBM1 displayed superior colonization rates, particularly when applied through soil drenching. Notably, soil inoculation with *B. bassiana* CBM1 promoted wheat growth, as evidenced by increased plant height at 30 days after sowing and enhanced root length at 15 days after sowing. Furthermore, wheat plants colonized by *B. bassiana* CBM1 exhibited increased mortality rates against the fall armyworm

(*Spodoptera frugiperda*). These findings underscore the promise of *B. bassiana*, especially strain CBM1, as a valuable component of integrated pest management strategies for wheat, offering dual benefits of pest control and plant growth promotion.

Fungal Antagonists:

Considering that mushrooms have a relatively high reproduction rate, they have a special purpose. So that in the absence of the host, they can stay in the affected environment and change from parasitic to saprotrophic to maintain their stable state. The figure shows the key sub-mechanisms in the biological control of fungal diseases of plants by fungal antagonists (Figure 1)

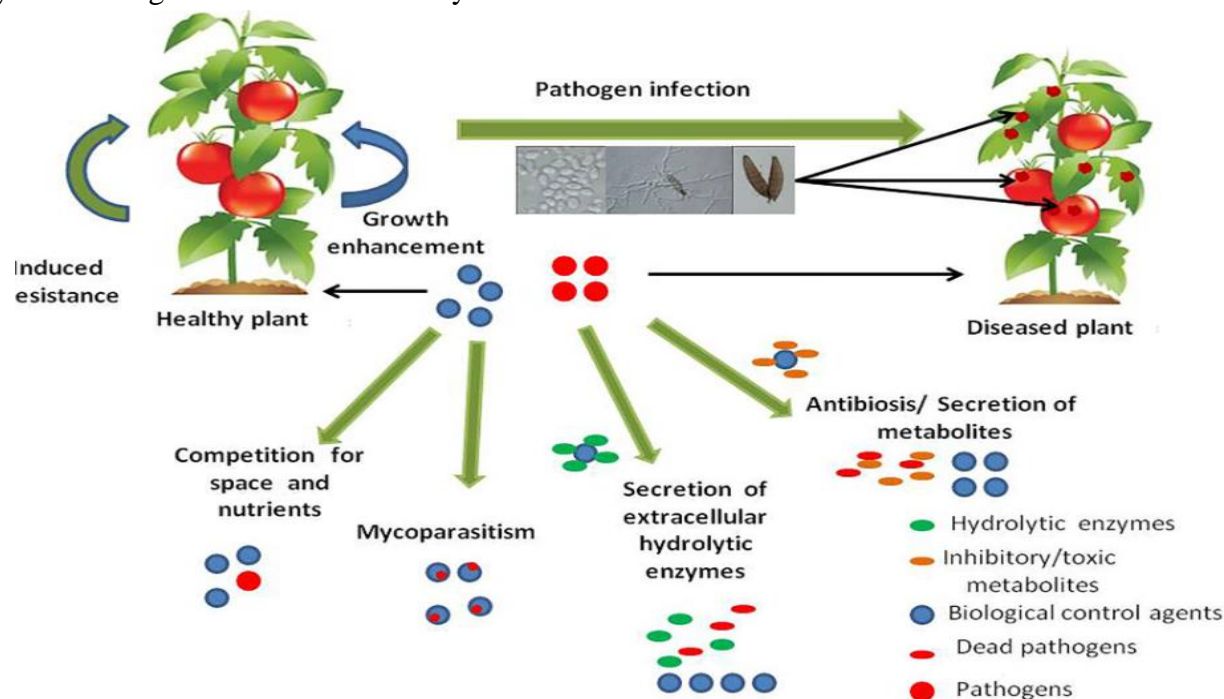


Figure 1: How fungal antagonists work in the biological control of fungal diseases [23].

Nematophagous fungi and their biological control

About 26,000 species of nematode fungi are known, and of these 26,000 species, approximately 4,100 species are plant parasites that live in the soil and cause plant root damage [50]. These plant parasites are economically important because they cause more than 100 billion dollars of damage to crops annually [51].

These parasites do not have specific symptoms, so it is difficult to detect them in crops. Of course,

the symptoms of contamination of products with these parasites include wilting, color change, loss of strength, lack of nutrients, root damage, reduced flowering, fruit loss, low productivity and even death. In the aerial parts of the plant, there is a possibility that chlorotic or necrotic spots are formed and they spread quickly from the infection site, also in the roots, cyst formation and the accumulation of worms occur [51].



Figure 2: Accumulation of nematode in the roots of guava (*Psidium guajava* var. *Paluma*) [51].

Controlling these parasites with nematicides is not cost-effective because, on one hand, they are expensive, and on the other hand, due to the lack of specific symptoms and locations for these parasites, they are non-selective and toxic to humans [52, 53]. Therefore, using biological agents is a more economical and environmentally friendly way to manage nematodes [54].

The use of nematophagous fungi, which are specific parasites of these worms, have been studied in terms of nematode control. To be efficient and improve the performance of these fungi, they must germinate on their host's body, and finally the formed hyphae must penetrate the nematode and make it sick [55].

Purpureocillium lilacinum, one of the most important nematophagous fungi that have the ability to deal with nematodes, can be used as biological control fungi [56]. Most biological nematicides are from *P. lilacinum* species [57]. *Verticillium* species also have this ability, and among these species, *V. (= Pochonia/Metacordyceps) chlamydosporium* and *V. lecanii* are the best representatives for biological control [58].

Other positive aspects of *Purpureocillium* representatives for use in biological control include the production of secondary metabolites (such as Phytohormones (Gibberellins and auxins)

that are able to enhance plant growth. They can also produce substances that protect them against the harmful effects of biotic and abiotic stresses (pH and salinity) [59]. There are several studies that state the importance of these fungi, that these fungi increase and improve plant growth through the dissolution of phosphorus in the plant. Because there are different forms of phosphorus in the soil that are insoluble and not accessible, this fungi converts phosphorus into an absorbable form [60].

Conclusion:

It can be concluded that the microbial biological control of fungal and bacterial strains was of particular importance due to their economic and environmental advantages, surely improvement and expansion of their use will be observed in the future. Fungal biological control is more important than other biological controls, because in addition to having the ability to control plant diseases by special mechanisms, it improves plant performance. Among the antagonistic fungi, the most important species were *Trichoderma spp* and *nematophagous fungi Purpureocillium lilacinum*, which increased plant growth by dissolving nutrients such as phosphorus in the soil. Therefore, studies on the identification and development of biological pesticides are increasing. Finally, by using fungal biological control to reduce diseases, it is possible to provide quality agricultural products without harming human health.

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