

Harnessing microbial antagonists for biological control of plant pathogens: A global perspective

Abstract

Reducing agricultural production inputs while maintaining a lucrative yield of high-quality goods is becoming more and more necessary as a result of the global sustainability agenda. Plant diseases pose a significant threat to productivity and product quality, yet many times there are no adequate measures available to control them. Consequently, research on substitute methods of crop protection has been mandated and has garnered significant interest from scholars around. A number of biological control agents (BCAs), including *Bacillus*, *Pantoea*, *Streptomyces*, *Trichoderma*, *Clonostachys*, *Pseudomonas*, *Burkholderia*, and specific yeasts, have been screened. Of these alternatives, biological controls through beneficial microorganisms have gained significant importance. BCAs, at the very least, support other sustainable disease management strategies like disease resistance and offer chances to control illnesses for whom alternative strategies are unfeasible or unobtainable. It is reasonable to anticipate that BCAs will be used more often to manage agricultural diseases in environmentally friendly ways.

Keywords: plant diseases, biological control agents, antagonist, sustainable disease control, augmentation

1. Introduction

The shifting agro-climatic conditions and occurrence of several insect pests and the losses that go along with them significantly increased the threat to the food security of the nation. Insects, diseases, and weeds in particular pose a hazard to the yield of crops grown for human consumption. Chemical pesticide misuse has resulted in a number of unfavourable outcomes throughout the world (Devi *et al.*, 2023), including resistance, residue, revival, and secondary pest outbreaks (Coll & Wajnberg, 2017). However, in order for these cultures to thrive and function well, farmers must continue to manage infections with parasites below the threshold of harmfulness (Köhl *et al.*, 2019). Furthermore, increased productivity and global commerce raise

the prevalence of some illnesses, necessitating the use of additional pesticides. These pesticides therefore worsen environmental contamination and accumulate chemical residues in the ecosystem they have treated (Rauscher & Rauscher, 1997). In intensive conventional agriculture, nitrogen fertilisers play a crucial role. When it comes to environmental contamination, however, their use proves to be a significant reason for worry as they can attract unwanted pest (Devi *et al.*, 2023).

The environmental effect of pest control techniques has been reduced by a variety of advances and breakthroughs over time. A variety of insecticides are being applied to limit their infestation in an effort to safeguard the crops. However, conventional methods of controlling insects, such as chemical applications, are costly, labor-intensive, and need frequent application. From a pragmatic perspective, alternatives like genetic pathways provide intriguing management strategies, but they also increase the likelihood that the pathogen may develop resistance genes (Pilet-Nayel *et al.*, 2017). Other options, including the employment of microorganisms in biological controls, may be able to significantly lessen the harmful effects of synthetic chemical use on the environment while also minimising the pollutants and annoyances that come with it (Usta, 2013; Compant *et al.*, 2015). By introducing parasitoids, predators, antagonist populations, or microbial diseases into the field to maintain pest numbers below the threshold level, biological management is, in this sense, a tried-and-true method of managing pest populations (Heimpel & Mills, 2017).

In terms of plant diseases, biological control is typically understood to be the suppression of a disease or its causative pathogen by an antagonist, or collection of antagonists, either directly or indirectly (Cook & Baker, 1983). In an effort to create sustainable agriculture at a reduced ecological cost (Laishram *et al.*, 2020), the idea of biocontrol has sparked a significant scientific, economic, and political discussion (O'Brien, 2017). As a result, some nations have put in place preventative measures that can cut the usage of pesticides by around 50% (Macfadyen *et al.*, 2014). Biocontrol advances have shown promise, particularly with the application of antagonistic biocontrol agents (BCAs) such as *Pseudomonas* spp., *Bacillus* spp., *Burkholderia* spp., and *Trichoderma* sp. against pathogens that cause foliar and soilborne diseases such as *Agrobacterium radiobacter* var. *radiobacter*, *Erwinia* spp., *Fusarium* spp., *Rhizoctonia solani*, *Phytophthora* spp., and *Pythium* spp. diseases (Pérez-García, 2011). An antagonistic effect

against a broad range of diseases has been demonstrated by additional BCAs. These include fungal species like *Aspergillus* spp., *Beauveria* spp., *Fusarium* spp., *Penicillium* spp., and *Phoma* spp. and bacterial species like *Burkholderia* spp., *Paenibacillus* spp., *Pantoea* spp., *Serratia* spp., and *Streptomyces* spp. Additionally, several of these BCAs can directly stimulate plant growth in addition to stopping the spread of the disease (Köhl *et al.*, 2019).

Agriculture in the modern day is always changing and growing. The development of biotechnologies and novel farming methods is under way (Deviet *al.*, 2023), after the widespread use of chemical fertilisers and pesticides in the 20th century, which contributed to a significant increase in output (Van-Montagu, 2020). Alternative of chemical fertilizer are to be use as a way for conserving the environment (Khumukchamet *al.*, 2020). The provision of food for almost 9 billion people by 2050 represents the next challenge (Godfray, 2014). Expanding food production capacity, especially those generated from plants, while protecting the environment is one of the main problems in this regard (Dwivedi *et al.*, 2017; Kiran *et al.*, 2024). The development of biofungicide products and other traditional methods, including the importation and release of natural enemies, can improve the efficacy and longevity of biocontrol in the modern period, as this review highlights in light of the current situation.

2. Biological Control of Plant Disease Management

2.1 Classical biological control

The deliberate introduction of an exotic natural enemy for long-term establishment and pest control to an area where the pest has invaded is known as classical biological control. In this case, a biocontrol agent is employed when the program's target pest is an exotic pest. The odds of establishing and controlling the target are increased when a traditional biological control agent is used to the same pest throughout several years and nations (Maurya *et al.*, 2022). More than 200 invasive insect pests and more than 50 weeds have been successfully suppressed globally by biological control since the late 1800s. Benefit-cost ratios have been favourable, ranging from 5:1 to >1000:1, with a 33% success rate in establishing exotic agents and a 10% rate of satisfactory control of targeted insect pests (Cock *et al.*, 2016). Biocontrol practitioners have embraced and created novel selection and assessment techniques in response to the dangers associated with the introduction of natural enemies. These tools have the potential to enhance the safety, accuracy, predictability, and efficacy of biocontrol programmes (Mills & Kean, 2010).

Furthermore, in an attempt to make introductions ecologically safer, risk assessment protocols were developed. These protocols centred on the deliberate selection and subsequent introduction of a specialised natural enemy that can effectively address invasive pest issues (Heimpel & Mills, 2017).

Scientific disciplinary breakthroughs have produced "new tools" that complement traditional biological control and have significant implications for a range of long-standing biocontrol initiatives for invasive arthropod pests (Maurya *et al.*, 2022). These "new" methods may be applied retrospectively to "legacy" pests, in addition to improving biocontrol of newly introduced invasive species. These pests are foreign and have been around for a while in different agro-ecosystems. Over time, as crops acquired new pest species, bio-control efforts to manage legacy pests became less prominent as attention shifted to new issues and the redesign of existing integrated pest management (IPM) programmes. Given enough time, legacy pests may ultimately be accepted as "native" as people would become used to seeing them. On the other hand, the creation of new tools like climate matching, ecological niche modelling, DNA-based analyses to identify species identities and areas of origin, modifying sowing date (Shijagurumayum *et al.*, 2022) and microbiome analyses could have a significant impact on the outcome of future initiatives aimed at both legacy and invasive pests.

2.2 Conservation biological control

The technique of improving the effectiveness of natural enemies by altering the surroundings or current pesticide usage is known as conservation biological control, or CBC (Eilenberg *et al.*, 2001). Primarily used in broad fields and perennial crops, this biological management approach targets endemic pests (Tixier, 2018). It addresses safeguarding against already-existing natural adversaries. Beneficial insects always need a different source of food in addition to the pest prey in order to stay in the agricultural area and feed and procreate. It can support safe biological control methods, but it requires thorough understanding of the ecology of ecological communities and natural enemies.

2.3 Augmentation of natural enemies

It is standard procedure to boost the efficacy of natural enemy populations by the release and multiplication of these populations or through the manipulation of environmental factors that

cause pest numbers to temporarily decline below the threshold level. Abiotic and biotic variables can be detrimental, but the ability of natural foes to cause havoc is the primary determinant of success. A number of factors lead to the use of augmentative biological control, including non-target and environmental effects (such as the recent development regarding neonicotinoids; EASAC, 2015), pesticides being removed from the market due to health concerns, pest resistance developing and making pesticides less effective, and the emergence of new pests for which no pesticides are available (such as the *Tuta absoluta* invasion in Europe in 2006, Urbaneja *et al.*, 2012). In the Almeria region of Spain, biological management supplanted chemical control of pests in 2005 after a two-year period (Calvo *et al.*, 2012).

Augmentation is divided into two parts

- **Mass production:** Mass production is a crucial component of biological control programmes because large-scale continuous releases of natural enemies require vast amounts of parasitoid culture. One of the most important factors that might directly affect mass-rearing investments is the cost of the host production. Factitious hosts are typically used to lower the cost and boost the efficiency of mass-reared parasitoids (El-Wakeil, 2007). A tritrophic system of raising involves the host plant, the natural prey (herbivorous pest), and the entomophagous insect. This is the foundation of many of the current techniques. Because both the host plant and the pest must be produced in order to produce the third species, this method doubles the expense of rearing (Van & Bellows, 1996).
- **Release:** In order to control the population of the target pest, natural enemies are introduced into the field after mass multiplication. The effectiveness of releasing natural enemies is contingent upon many factors that bear a remarkable resemblance to the factors that influence the success of using pesticides: the rate of application, the timing (including the time of day), the synchronisation with the pest's susceptible stage, the coverage, and the intensity of rainfall after the treatment (Maurya *et al.*, 2022). The primary differential between biological management and pesticide treatments is that the former should be adapted to the density of pests, while the latter should be aimed towards the complete coverage of surface area. To guarantee their survival and long-term field release, it is essential to not only produce superior natural enemies but also to release them in the field with effectiveness.

2.4 Resistant Varieties

In agriculture, well-established and tested methods such as crop selection (Yambemet *et al.*, 2020) and plant breeding are used to enhance crop variety and generate disease-resistant cultivars. Resistant varieties and lucrative crop can be grown within a short time period and can be beneficial to the soil as well as the environment (Laishram *et al.*, 2023). Abiotic (such as salt, drought, and heavy metals) and biotic (such as insects, pests, and microbial assaults) variables are the major sources of effect on crop development and resilience (Devi *et al.*, 2023). These methods are still in use today and have shown to be effective in the battle against several plant pathogens that cause illness (Ab *et al.*, 2018). In our never-ending quest to boost food production, one of the most popular biotechnological applications is the genetic pathway. In addition to being resistant to disease, genetically modified (GM) cultivators yield higher-quality crops with far lower dependency on expensive chemical inputs, making their cultivation economically feasible (Pandit *et al.*, 2022). Despite these benefits, GM crops are not widely embraced by consumers and require expensive regulatory agency clearance. There are further benefits to managing resistance through gene pyramiding, gene rotation, and multiline variations, among other breeding techniques. In order to control the more recent, aggressive infections, it is essential that newer, more advanced biotechnological technologies be created and used to hasten the generation of enhanced disease-resistant cultivars (Miah *et al.*, 2017). As a result, the primary goal would be to raise production through the use of improved agricultural techniques (Devi *et al.*, 2023).

3. Microbial biocontrol

3.1 Bacteriobiocontrol

Numerous rhizosphere bacteria colonise the majority of plants planted in fields (Sindhu *et al.*, 2014). Based on how they affect plant performance, certain bacteria that are connected with plants are categorised as helpful microorganisms. Plant-growth-promoting rhizobacteria (PGPR) are a type of free-living bacteria that generate a range of antifungal metabolites and plant growth-promoting characteristics (Shaikh *et al.*, 2018). PGPR are found to flourish freely in the rhizosphere soil. PGPRs also function as biopesticides, depending on the capacity or actions of the crops and biocontrol agents (Labuschagne *et al.*, 2010). *Alcaligenes*, *Azospirillum*, *Arthrobacter*, *Acinetobacter*, *Bradyrhizobium*, *Bacillus*, *Burkholderia*, *Enterobacter*, *Erwinia*, *Flavobacterium*, *Pseudomonas*, *Rhizobium*, *Azorhizobium*, *Bradyrhizobium*, *Allorhizobium*,

Sinorhizobium, *Frankia*, *Mesorhizob*, *Azoarcus*, *Exiguobacterium*, *Methylobacterium*, *Paenibacillus*, and *Pantoea* are among the bacterial species that make up PGPRs (Seenivasagan & Babalola, 2021). These advantageous relationships improve plant health and agricultural yield via enhancing nutrient availability (Laishram *et al.*, 2023), producing hormones that stimulate plant growth, reducing illnesses caused by pathogens and pests, or strengthening resilience to environmental stress (Sindhu *et al.*, 2014). In order to stop the development of disease, certain rhizosphere bacteria produce secondary metabolites such as endotoxins, bacteriocins, siderophores, hydrolytic enzymes, hydrogen cyanide (HCN), phenazine-1-carboxylic acid (PCA), 2,4-diacetylphloroglucinol (DAPG), and other antibiotics that kill pathogens (Chauhana *et al.*, 2015). Below is a discussion of certain PGPRs that are used to manage plant diseases and pests.

Applications of *Bacillus* species for biocontrol and agricultural growth promotion have a long history (Zhu *et al.*, 2021). The most commercially effective biopesticide available is *Bacillus thuringiensis* (Bt) (Anckaert *et al.*, 2022). Endotoxins, which are produced by Bt and are poisonous, can be used as biopesticides and as a source of genes to create transgenic plants that are resistant to insects (Jisha *et al.*, 2013). Bt strains generate crystalline proteins known as "endotoxins" during their stationary phase of development, which are poisonous to mites, nematodes, protozoa, and flukes in addition to lepidopterous, coleopterous, and dipterous insects (Jisha *et al.*, 2013). Isolates of *Pseudomonas chlororaphis* are utilised as biopesticides in agriculture because they shield plants from nematodes, insects, and a variety of microbiological illnesses. These isolates produce a range of metabolites that directly reduce nematodes, insects, and microbial pathogens (Anderson & Kim, 2018). *P. chlororaphis* PcO6, which was extracted from dryland plant roots, improved plant health and was applied to agriculture as a BCA and biofertilizer. Numerous compounds produced by this bacterium through systemic resistance induction and direct pathogen antagonistic interactions enhance plant development. Studies on PcO6 have shown the methods by which certain bacterial metabolites provide defence against insects, nematodes, and harmful pathogens. *Pseudomonas aeruginosa*, which was isolated from the rhizosphere of banana fields, generated antifungal chemicals such as bacteriocin, HCN, and siderophore. The isolate's bacteriocinogenic, siderophoregenic, and HCN-rich broth inhibited the growth of phytopathogens including *Aspergillus niger*, *Aspergillus flavus*, *Fusarium oxysporum*, and *Alternaria alternata* (Pandit *et al.*, 2022).

3.2 Fungal Biocontrol

Fungi offer biocontrol properties in addition to helping plants absorb nutrients and utilise nitrogen. They can help fight off pests like nematodes and microbial infections that affect the roots, leaves, and fruits of the plant, among other areas. Through mechanisms including mycoparasitism, competition with pathogens for resources, antibiosis, imparting ISR to the host plant, and mycovirus mediated cross-protection, or MMCP, they provide protection against illnesses (Singh, 2017). The fungi *Trichoderma* species, ectomycorrhizas, arbuscular mycorrhizas (AMF), yeasts, and endophytes are a few of the well-known fungal biocontrol agents. Hypovirulence-associated mycoviruses can be used by even nonvirulent strains of certain pathogens to act as biocontrol fungi (Ghorbanpour *et al.*, 2018). Better biotechnological and genetic advancements allow for the introduction of advantageous fungal genes into host plant genomes as well as the interruption or overexpression of these genes to enhance biocontrol capacity.

A thorough biological control fungal agent review by Kasun *et al.* (2020) describes the agents employed against fungal plant diseases. They claim that *Trichoderma*, which has 25 species that have been utilised as biocontrol agents against a variety of plant fungal diseases, is the genus with the most promise for biocontrol. The majority of plant growth-promoting fungi (PGPF), which include *Trichoderma*, *Penicillium*, *Aspergillus*, and *Fusarium* spp., are known to stimulate the plant immune responses in response to enemy attack. They are also regarded as one of the most secure methods for promoting crop plant growth and inducing systemic resistance (ISR) (Jogaiah *et al.*, 2018; Divya *et al.*, 2021).

3.3 Plant Virus Biocontrol

Certain beneficial viruses that are often found and well-characterized are known to improve the aesthetic appeal of ornamental plants. The earliest of these viruses was the lip-breaking kind. But many other valuable ornamentals have viruses that partially or completely destroy their value (Valverde *et al.*, 2012). Additional instances of advantageous plant viruses comprise multiple acute viruses, like the Brome mosaic virus (family Bromoviridae), Cucumber mosaic virus (family Bromoviridae), Tobacco rattle virus (family Virgaviridae), and Tobacco mosaic virus (family Virgaviridae), which endow different crops with resistance to freezing temperatures and drought, and persistent viruses, like the White clover cryptic virus (family Partitiviridae), which

can inhibit legume nodulation when given an adequate amount of nitrogen (Roossinck, 2012). The effects of *Sclerotinia sclerotiorum* hypovirulence-associated DNA virus 1 (SsHADV-1) a single-stranded DNA virus that infects the fungus *Sclerotinia sclerotiarum*, a disease-causing agent of numerous crops, have been clarified by (Qu *et al.*, 2021). Through the use of digital RNA sequencing, Qu *et al.* (2021) have further clarified the changed expression of genes relevant to phenotype following SsHADV-1 infection.

Table 1: Microbial antagonist and their biocontrol strategies

Pathogen	Host	Biocontrol Strategies	References
<i>Phytophthora sojae</i> , <i>Pythium heterothallic</i> , <i>Pythium irregulare</i> , <i>Pythium sylvaticum</i> , and <i>Pythium ultimum</i>	Glycine max	Pseudomonas water derived strain, 06C 126, effectively inhibited oomycetes	Wagner <i>et al.</i> (2018)
Soil borne fungal pathogens	Pulses, grapes, cotton, onion, carrot, peas, plums, maize, apple, etc.	The fungal genus <i>Trichoderma</i> has biocontrol activity against fungi and nematodes	Aldayel(2019)
<i>Phytophthora</i> spp. and <i>Pythium</i> spp.	Aquaponics	Antagonistic microorganisms	Stouvenakers <i>et al.</i> (2019)
Pathogens in the crop residues	Cereal crops	Microbiome-based biocontrol strategies	Vurukonda& Stefani(2018)
Fungal pathogens	Cereal crops	<i>Streptomyces</i> species produce a range of secondary metabolites that can inhibit the growth of phytopathogens	Newitt <i>et al.</i> (2019)
Diseases caused by fungi, bacteria, viruses, viroids, nematodes, and oomycetes	<i>Citrus</i> sp.	Employment of antagonists produced by <i>Bacillus</i> sp. offers superior capacity to restrict diseases in citrus plants	Chen <i>et al.</i> (2020)
<i>Rhizoctonia solani</i> that induces stem canker, <i>Fusarium solani</i> causes tubers dry rot, and black scurf and <i>Alternaria solani</i> that induces early blight	Potato	Romanian potato tubers isolate 6T4 identified as <i>B. atrophaeus/subtilis</i> revealed promising perspectives for biocontrol strategies	Boiu-sicuiet <i>al.</i> (2017)
<i>Verticillium dahliae</i> soil borne pathogen	Cotton	Endophytic Fungus <i>Fusarium solani</i> CEF559 against <i>Verticillium dahlia</i>	Wei <i>et al.</i> (2019)

Bacterial phytopathogen <i>Pseudomonas syringae</i> pv. Tomato	Tomato	in Cotton Plant <i>Pseudomonas segetis</i> strain P6 isolated from the rhizosphere has the ability to induce plant growth and inhibit quorum sensing abilities of bacterial pathogens	Rodríguez <i>et al.</i> (2020)
Closteroviridae family of plant viruses causing leafroll disease	Grapevine	Case based management, such as use of certified planting material, open field foundation block vineyards on virgin soil etc.	Almeida <i>et al.</i> (2013)
Cucurbit yellow stunting disorder virus, Cucurbit chlorotic yellows virus and Beet pseudo-yellows virus	Vegetable crops	Integrated disease management strategies and using resistant varieties	Abrahamian & Abou-Jawdah (2014)

3.4 Yeast Biocontrol

Currently used as biocontrol agents, yeasts such *Aureobasidium pullulans*, *Cryptococcus albidus*, *Candida oleophila*, *Saccharomyces cerevisiae*, and *Metschnikowiafructicola* are excellent enemies of a variety of plant infections. A class of unicellular fungi known as yeasts may thrive in a variety of conditions, need little in the way of culture, and pose little to no biosafety risks. For plant protection, they use enzymes and poisons, volatiles, competition, mycoparasitism, and the start of immune response processes. They can be used as biocontrol effectors because of these qualities, however there aren't enough research on them to fully understand their function (Freimoser *et al.*, 2019). It is known that yeasts use phage-based competition, enzyme secretion, toxins, volatiles, mycoparasitism, and the generation of resistance activity to carry out their biocontrol function (Freimoser *et al.*, 2019). Several non-conventional yeast species have had their whole genomes sequenced, and their population is steadily increasing (Wendland, 2020). Thus, anticipated novel techniques for the genomic and post-genomic analysis of yeasts before and after further modifications, along with their genetic analysis, will serve as a platform for comprehending the molecular mechanisms underlying both the complex and simple biological features that are expected to be helpful for the creation of new and environmentally friendly applications.

3.5 Algal and Cyanobacterial Biocontrol

Algae and cyanobacteria extracts are rich in bioactive elicitors with antifungal, antiviral, and antibacterial properties (Arunkumaret al., 2010), in addition to being a plentiful source of vitamins, saccharides, enzymes, amino acids, and phytohormones, as well as elements like molybdenum, boron, manganese, iron, iodine, and zinc (Sultana et al., 2011). In order to increase plant life and production, these extracts are often used in agriculture. The symptoms of fungal infections on tomatoes caused by *Alternaria solani* and *Xanthomonas campestris*pv. *vesicatoria* are lessened by the use of extracts from the algae *Sargassum filipendula*, *Ulva lactuca*, *Caulerpa sertularioides*, *Padina gymnospora*, and *Sargassum liebmannii*(Ramkissoon et al., 2017). Research on tomato seedlings with *Macrophominaphaseolina* infection revealed that the treatment of *Kappaphycusalvarezii* resulted in improvement. Increased amounts of phytohormones (abscisic acid, salicylic acid, and indole-3-acetic acid), transcription of the PR-1b1, PR-3, and PR-4 genes, and the cytokinin zeatin all contributed to the algal activity (Agarwal et al., 2016). Utilising extracts from *Cystoseiramyriophylloides*, *Laminaria digitata*, and *Fucus spiralis* against *Verticillium dahliae* wilt was also demonstrated to boost the activity of polyphenol oxidase and peroxidase enzymes, which are vital in plant defence in tomatoes (Essertiet al., 2017). Cyanobacteria, akin to algae, possess the ability to produce large amounts of polysaccharides in reaction to several plant pathogen classifications; nevertheless, their use as biocontrol agents is restricted due to information gaps (Righiniet al., 2019).

Table 2: Improved Biocontrol Through Microbial Antagonism

Tolerance	Bio control attributes	References
Climatic tolerance	The National Bureau of Agricultural Important Insects (NBAIL), Bangalore, has created improved strains of <i>Trichogrammachilonis</i> Ishii, an important egg parasitoid of lepidopterans, by use of a selection approach for adaptability to both high and low temperature regimes. This strain has been observed to have a greater parasitism rate, a longer lifespan, and to be effective at 36 °C and 60% relative humidity. During the summer, it works especially well against pests that affect cotton, sugarcane, and vegetable crops. Similar to this, a strain of <i>T. chillonis</i> suited to low temperatures was created in the lab by selection over 30 generations at 18–24 °C. It was discovered to have	Ballal & Singh (2003) Jalali et al. (2016)

Insecticides tolerance	<p>improved host finding efficiency, a greater parasitism rate, and favourable biological characteristics.</p> <p>The phytoseiid mite <i>Metaseiulus occidentalis</i>, Nesbitt, is the primary predator of spider mites. A pesticide-resistant strain of this mite, dubbed the COS strain (carbaryl-OP-sulphur-OP-resistant strain), was enhanced through artificial selection and was eventually found to be resistant to diazinon, azinphos-methyl, and phosmet (like organophosphorus insecticides).</p>	Arora & Shera (2014)
Tolerance to Multiple Traits	<p>Through 341 generations of consecutive exposure of adult <i>T. chilonis</i> parasitoids to varying doses of endosulfan (0.004–0.09%), a resistant strain of <i>T. chilonis</i> known as "endogram" was created. It was discovered that this strain had 56% more <i>Helicoverpa armigera</i> (Hubner) parasitization than the susceptible strain, which had just 3%.</p> <p>A <i>T. chilonis</i> strain known as multiple insecticide and temperature tolerant (MITT) has been created to efficiently manage crops subjected to heavy pesticide pressure. It was discovered that this strain was resistant to high temperatures (32–38 °C), endosulfan (organochlorine), monocrotophos (organophosphate), and fenvalerate (synthetic pyrethroid). After being exposed to three pesticides continuously for six hours, there was also an increase in parasitism from 35 to 90–95% and a drop in mortality from 100 to 57–70%.</p>	Jalali <i>et al.</i> (2006) Kumar <i>et al.</i> (2008)
Altered Biological Traits	<p>It has been demonstrated to be advantageous to increase the lifetime of several parasitoids. Better strains of <i>T. chilonis</i> (Bio SC1: graminaceous tissue borers), (Bio H3: <i>H. armigera</i>), and (Bio C1: cotton bollworms) have been created by NBAII, Bangalore. It was discovered that these strains were 60–100% more prolific than the ones that had been employed before. There have also been attempts to boost the fitness of parasitoids through hybridization and heterosis.</p>	Arora and Shera (2014)
Enhancement of Entomopathogenic Microbes Genetically in the Host Range	<p>Based on "Germany-China Scientific Cooperation" research, GCSC-BtA is another novel biocide that was created by combining the toxin of <i>Streptomyces avermitilis</i>, amamectin, with the delta-endotoxin of <i>B. thuringiensis</i>. This was discovered to be more effective against agricultural pests than either of the protectors by itself, with a wider host spectrum.</p>	Liu & Sengonca (2003)
Improvement in Photostability and Activity	<p>Since melanin, a naturally occurring substance, has been shown to impart UV undegradability, <i>B. thuringiensis</i> mutants produced by ethyl</p>	Mohamed <i>et al.</i> (2010)

<p>Enhanced Activity Using an Alternative Delivery System</p>	<p>methanesulphonate (EMS) have been shown to be UV tolerant and to exhibit greater insecticidal effects against the potato tuber moth, <i>Phthorimaea operculella</i> Zeller</p> <p>By using different delivery methods to reach the poisons to the intended insects, <i>B. thuringiensis</i> has also demonstrated increased insecticidal action. To increase the delivery, residual activity, and durability of Cry protein, Bt genes have been cloned and expressed in eukaryotic plants, endophytic, epiphytic, and/or aquatic bacteria.</p>	<p>Sharma (2008)</p>
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4. Mechanisms of biological control

BCAs are used in the disease management of plant pathogens, where they exert control on the pathogens through a range of distinct mechanisms. Comprehending the processes behind the protective effects of BCAs will aid in control optimisation and enable the deployment of more effective strains in the appropriate setting (Ghorbanpouret *al.*, 2018). To suppress plant disease both directly and indirectly, the BCA may employ these strategies alone or in combination. Researchers have concentrated on characterising the mechanisms functioning in many experimental scenarios since biological control may arise from a wide variety of interactions between organisms. The existence and actions of other organisms that pathogens come into contact with always operate as a source of hostility. In this instance, we contend that the various mechanisms of antagonistic relationships arise along a spectrum of directionality associated with the degree of interspecies contact and interaction specialisation (Table 3).

Table 3: Types of plant pathogens under biological control due to interspecies antagonists

Type	Mechanism	Examples
Direct antagonism	Hyperparasitism/predation	<i>Lytic/some nonlytic mycoviruses</i> <i>Ampelomyces quisqualis</i> <i>Lysobacterenzymogenes</i> <i>Pasteuria penetrans</i> <i>Trichoderma virens</i>
Mixed-path antagonism	Antibiotics	2,4-diacetylphloroglucinol Phenazines Cyclic lipopeptides
	Lytic enzymes	Chitinases Glucanases Proteases

	Unregulated waste products	Ammonia Carbon dioxide Hydrogen cyanide
	Physical/chemical interference	Blockage of soil pores Germination signals consumption Molecular cross-talk confused
Indirect antagonism	Competition	Exudates/leachates consumption Siderophore scavenging Physical niche occupation
	Induction of host resistance	Contact with fungal cell walls Detection of pathogen-associated, molecular patterns Phytohormone-mediated induction

(Source adapted from Pal & Gardener, 2006)

5. Factors Influencing the Biological Control of Plant Pathogens: Success or Failure

The growing need for alternatives to chemical control has made biological control in plant health management seem insignificant, despite decades of study to the contrary. Reports of a meaningful impact were far more prevalent in laboratory settings (in vitro or planta) than in field experiments. Moving from the controlled settings of a laboratory experiment to the harsh conditions faced in the field has shown to be more challenging for biopesticides than for chemicals, as is well known. Although the field efficacy of BCAs can be as high as or higher than that of chemical pesticides, this can change over time and between different locations (Nicotet *al.*, 2012). To put it another way, an adversary that suppresses or manages diseases in the lab is seldom useful in the real world. The host, the disease, the antagonist, the environment, and all the complex interactions that take place between them are to blame for this. For instance, the host undergoes a number of evolutions or mutations that alter its chemical, biological, and physical characteristics. Pathogenic characteristics also influence the antagonist's effectiveness. The opponent may adapt to shifts in the population, the environment, or the existence of microbial colonists inside the biological system (Umeret *al.*, 2021).

5.1 Effect of the Plant on Biocontrol Activity

Plant species and genotypes have the potential to significantly impact the results of biological control. In biocontrol, the plant itself has two functions. Plant genotype can affect the level of rhizosphere colonisation, antagonists' synthesis of antibiotics, and plants' development of

induced resistance. For example, a plant's vulnerability to a particular nematode species may affect how well control works; excellent hosts would need more suppression than bad hosts (Timper, 2014). Additionally, the plant acts as a site of interaction between antagonists and diseases. Surface temperature, gaseous exchange, ion and water absorption, and host exudate excretion all affect the interactions between the antagonist and the pathogen (Umeret *al.*, 2021). The microbial population that exists on the surface of plants and in their immediate surroundings is influenced by the expression of plant genes.

5.2 Effect of the Pathogen on Biocontrol Activity

One of the most important things to take into account while selecting BCAs is pathogen behaviour; because of genetic variety and ecological fitness variation, every pathogen interacts with hosts in a different way. It is crucial to emphasise that while pathogens are pathogenic and susceptible to antagonist action, their behaviour is different from that of antagonists (Umeret *al.*, 2021). Durability is the term used to describe a plant protection control system's capacity to remain effective throughout time and space. It is dependent upon two things: (i) the selection pressure that is applied to populations of plant-pathogens, and (ii) the pathogen's capacity to adjust to the control method. Put another way, plant pathogens can exhibit a broad range of BCA sensitivity, including very low sensitivity, depending on how complicated their method of action is. In a few generations, certain diseases can adjust to the selection pressure that BCAs apply (Bardinet *al.*, 2015).

It is often believed that biological control outlasts chemical control. Research on pest management in agricultural settings suggests that this assumption may not always be valid. It has been shown that a number of pests are resistant to one or more Bt toxins, and that the codling moth *Cydia pomonella* is resistant to the *C. pomonella* granule virus. Biological control of plant diseases has been around for a while, but it hasn't gotten as much attention as pest control. No research studies showing that BCAs are no longer effective against plant pathogens have been published as of yet. BCAs may not be as effective against plant diseases as they formerly were since pathogen populations have the potential to become resistant to them in a manner akin to that of single-mode chemical fungicides. Finding a window of opportunity in the pathogen life cycle that is a weak point is essential for effective biocontrol. The goal of the antagonist should be to break the illness cycle and enter the window of opportunity. For instance, the window of

opportunity for certain unspecialized necrotrophic diseases is to obstruct the absorption of nutrients required for development. This phase will be repressed when the antagonist competes for nutrients and stops the saprophytic phase from establishing. When hostile organisms are sufficiently prevalent in the vicinity of the pathogen spores, the loss of endogenous nutrients from the disease spore may restrict or completely stop germination. The antagonist's ability to synthesise enzymes or antibiotics may also be useful in the fight against these illnesses (Umeret *al.*, 2021).

5.3. Effect of the Biocontrol Agent on Biocontrol Activity

The primary cause of BCAs' decreased effectiveness in the field is their capacity to adjust to local biotic and abiotic environmental variables (Deviet *al.*, 2023). For a complete understanding of this phenomena, it is necessary to investigate the spatial distribution pattern of BCAs in the rhizosphere (Nandinet *al.*, 2021). To get pertinent biocontrol outcomes, more appropriate native strains of BCAs should be gathered and evaluated (Yuet *al.*, 2021). The modes of action of BCAs are intimately associated with their efficiency (Hassanet *al.*, 2011), often involving trade-offs with other inherent properties of the agents, including environmental persistence and specificity.

The microbial strain's inherent biological characteristics (such as its "ecological competence"), the calibre of the prepared materials used in the field, or insufficient application time or technique are among the most often noted obstacles in biocontrol (Nicotet *al.*, 2012). Effectiveness of the biocontrol agent will increase in an optimal partnership. As a result, timing the antagonist's administration is essential for effective biocontrols. The biocontrol will be successful if the antagonist is administered prior to the pathogen's establishment. Furthermore, it's essential to comprehend how BCAs function in order to obtain the best possible disease management. Comprehending the method of action is crucial in order to discern any hazards to human health or the environment, in addition to the possibility of resistance developing against the BCA (Esmaeelet *al.*, 2020).

5.4. Effect of the Environment on Biocontrol Activity

The biological makeup of the soil and external conditions greatly influence the effectiveness of biocontrol initiatives. Consequently, by evaluating each organism's proportional contribution to

the biocontrol process, soil biology research should pinpoint the many noteworthy traits of different species, especially in the plant rhizosphere. Likewise, as essential components of plant health, ecological study has to investigate all biotic and abiotic factors that affect the BCAs (Singh & Vimal, 2020). One practical way to manage the reliability of BCAs under practical situations is to incorporate the function of microbial communities in the construction of a BCA that is compatible with other soil microbiomes. A complex network function in the soil maintains a multitude of activities that collectively make up a microbial BCA, a complex metabolic phenotype. Applying BCAs in conjunction with an appropriate complex biocontrol mixture that contains important macronutrients, helpful helper strains, and biocontrol prebiotics may facilitate the development of BCAs in the epiphytic microbial network.

Examples of how agricultural methods may support or undermine the biological control of plant-parasitic nematodes (PPNs) and other soil-borne pests were highlighted by Timper (2014). By offering more food sources, such as when organic soil amendments are added, nematode antagonist efficacy can be increased. However, some organic additives, such as manures and plants with allopathic chemicals, may be detrimental to nematode antagonists. Production techniques like as crop rotation, tillage, pesticide spraying, and fallow periods can all directly disturb populations of hostile organisms. BCAs are being increasingly commonly used as a safer substitute for hazardous chemical nematodes in the management of a range of PPN problems. Despite this, they have only been used in a small portion of the pesticide business because of their ineffectiveness, inconsistent field performance, and/or unfavourable economic conditions. Improved sampling, a better understanding of BCAs' interactions with soil biota and ecology, the cost-effective use of BCAs, genetic manipulation for better PPNs control, grower acceptance and awareness of BCAs techniques, and commercial application are just a few ways that a comprehensive understanding of soil biological and ecological components can improve the efficacy and success of biocontrol (Singh & Vimal, 2020).

6. Conclusion

Resistance, residue, and revival are only a few of the ecological and environmental issues that have arisen from an over-reliance on chemicals. The population of pests has been managed through the use of various biological control agents in an attempt to mitigate these effects; nevertheless, the main obstacles to the effective use of various natural enemies are climate

restrictions and factors linked to persistence. The evidence that is now available challenges the notion that chemical control is necessarily less sustainable than biological control. The efficiency of these biocontrol products can be increased by modifying the environment, using mixtures of beneficial organisms, enhancing the physiological and genetic functions of the biocontrol mechanisms, modifying the formulation, and combining biocontrol with other complementary techniques that have additive effects. To promote plant development in sustainable agriculture, these BCAs might be employed efficiently. Enhancement of the host range, numerical and functional efficiency, broader climate adaptation, resistance to various pesticides, and resistance to hyperparasites is highly promising. These characteristics are thought to be crucial for improving biocontrol agents and raising their effectiveness.

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