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3. Biological Control Methods are A Safe and Sustainable Approach to Managing Plant Diseases

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Biological control method, a fundamental aspect of integrated pest management (IPM), is a sustainable and environmentally aware method used in modern agriculture. This approach utilizes natural relationships between organisms to manage the pest populations and create a healthy ecosystem. Beneficial Micro-organisms like *Trichoderma* spp, *Pseudomonas* spp and *Bacillus* spp are used to manage plant diseases incite by phytopathogen, reducing the need for synthetic pesticides and decreasing their potential harm to the environment, non-target species, and human health. By introducing these natural enemies or boosting their numbers in agricultural areas, biological control provides lasting disease/pest control while fostering biodiversity and resilience in ecosystems. Bio-control agents comprise of multiple beneficial characters such as rhizosphere competence, antagonistic potential, and ability to produce antibiotics, lytic enzymes, computation for nutrient and niche. These biological control activities are exerted either directly through antagonism of soil-borne pathogens or indirectly by eliciting a plant-mediated resistance response reported by Zerihun Tsegaye. *et.al.*, (2018).

Agricultural systems globally encounter major obstacles in controlling pest numbers while reducing the adverse environmental effects of conventional chemical pesticides. Continuous and widespread pesticide application has resulted in pest resistance, damage to non-target organisms, and pollution of soil and water. Sustainable agriculture demands efficient pest management methods that prioritize both crop protection and ecological preservation.

The evolving area of disease and insect pest management is experiencing a significant change towards sustainable approaches. Biological Control Agents (BCAs) have become a powerful tool, utilizing predatory insects, parasitoids, and beneficial microorganism to minimize disease incidence and insect pest populations. Biological methods enhance soil health, aiding in the retention of nutrients and water. This promotes a balanced connection between farming and the environment, protecting ecosystems and water quality. Agricultural techniques pave the way for a more sustainable and fertile future.

3.1 What is Biological Control?

Biological control was defined by Baker and Cook (1974) as the "reduction of inoculum density or disease producing activities of a pathogen or parasite in its active or dormant stage, by one or more organisms, accomplished naturally or through manipulation of the environment, host, or antagonists, or by mass introduction of one or more antagonists". Subsequently, they (Cook and Baker 1983) shortened the definition to "biological control is the reduction of the amount of inoculum or disease producing activity of a pathogen accomplished by one or more organisms other than man".

3.2 Biocontrol Agents (BCAs):

Biocontrol agents are microorganisms that harm the population of other microorganisms, such as target pathogens, with which they grow. They typically have the ability to disrupt the life processes of plant pathogens. Biocontrol agents encompass various classes of organisms, including fungi, bacteria, nematodes, protozoa, viruses, and seed plants. Many potential biocontrol agents function by parasitizing target pathogens. For instance, if the biocontrol agent is a fungus, it might act as a mycoparasite, feeding on another fungus, in which case the pathogen serves as its food source. Mycoparasites usually produces either chitinase or cellulose like cell wall degrading enzymes which break down cell wall components of host fungi (Barnett, 1964). Among the fungal antagonists *Trichoderma* have gained maximum popularity as biocontrol agent **(**Mukhopadhyay, 1987).

3.3 Mechanism/ Mode of Action of Biocontrol Agents:

BCAs suppress the growth of plant pathogens by various ways like mycoparasitism, antibiosis, production of enzymes and volatile compounds.

3.3.1 Direct Action of Biocontrol Agents:

A. Parasitism and Mycoparasitism:

Parasitism is an intraction involving physical contact between antagonistic microorganism and plant pathogen that reduces inoculam density of pathogen Mycoparasitsm is an act where one fungus is parasitized on other fungus. The complex process of mycoparasitism consists of several events, including recognition of the host, attack and subsequent penetration and killing (Vinale *et al.* 2008). Fungal bioagents grows tropically toward hyphae of other fungi, coil around them in a lectin mediated reaction, and degrade cell wall of the target fungi by the secretion of different lytic enzymes. Inbar *et al*. (1996) found that once the fungi come into contact, *Trichoderma* sp. attach to the host and can coil around it and form appressoria on the host surface. This attachment is mediated by the binding of carbohydrates in the *Trichoderma* cell wall to lectins on the target fungus.

B. Antibiosis:

Antibiosis is the process to inhibition or reduction of pathogen by a metabolic product of the antagonist such as production of specific toxin, antibiotics or enzymes. This interaction results in suppression of activity of pathogens or pathogen propagules.

Trichoderma are well known biological agents that produce a range of antibiotics which are active against pathogens *in vitro* (Dennis and Webster, 1971a). Howell *et al*. (2000) isolated and described a new antibiotic; "*gliovir*i*din"* from *G. virens* that was strongly inhibitory to *P. ultimum* and *Phytophthora.* Most *Trichoderma* strains used to produce volatile and non-volatile toxic metabolites such as peptaibols, 6-pentyl-α-pyrone, massoilacton and viridin.

a. Siderophore:

Siderophores are low molecular weight ferric iron chelating compounds that are secreted extracellularly under iron limiting conditions and whose main function is to supply iron to the iron starved cells. Some PGPR strains produce siderophores that bind Fe3+, making it less available to certain members of native microflora (Kloepper *et al.,* 1980).

Most evidences to support the siderophore theory of biological control by rhizobacteria comes from the work with pyoverdin, a class of siderophores that comprise the fluorescent pigment of fluorescent pseudomonads **(**Demange *et al.,* 1987).

Table 3.1: List of antibiotics produced by BCAs

Sr. No	Antibiotic	Source	Target pathogen	Disease	Reference
1.	Agrocin 84	*Agrobacterium radiobacter*	*Agrobacterium tumefaciens*	Crown gall	Kerr (1980)
2.	Pyoluteorin, pyrrolnitrin	*P. fluorescens* Pf-5	*Pythium ultimum* and *R. solani*	Damping off	Howell and Stipanovic (1980

Sr. No	Antibiotic	Source	Target pathogen	Disease	Reference
3.	2, 4-diacetyl-phloroglucinol	*Pseudomonas fluorescens* F113	*Pythium* spp.	Damping off	Shanahan *et al.* (1992)
4.	Bacillomycin D	*Bacillus subtilis* AU195	*Aspergillus flavus*	Aflatoxin contamination	Moyne *et al.* (2001)
5.	Bacillomycin, fengycin	*Bacillus amyloliquefaciens* F ZB42	*Fusarium oxysporum*	Wilt	Koumoutsi *et al.* (2004)
6.	Gliotoxin	*Trichoderma virens*	*Rhizoctonia solani*	Root rots	Wilhite *et al.* (2001)
7.	Herbicolin	*Pantoea agglomerans* C9-1	*Erwinia amylovora*	Fire blight	Sandra *et al.* (2001)
8.	Mycosubtilin	*B. subtilis* BBG100	*Pythium aphanidermatum*	Damping off	Leclere *et al.* (2005)
9.	Phenazines	*P. fluorescens* 2-79 and 30-84	*Gaeumannomyces graminis* var. *tritici*	Take-all	Thomashow *et al.* (1990)
10.	Pyrrolnitrin, pseudane	*Burkholderia cepacia*	*R. solani* and *Pyricularia oryzae*	Damping off and rice blast	Homma *et al.* (1989)

C. Competition:

Competition is one of the mechanisms of biological control activity of *Trichoderma* sp. against phytopathogenic fungi. It involves competition between antagonist and plant pathogen for space and nutrients (Howell *et al.* 2000; Muslim *et al.* 2003).

3.3.2 In direct action of biocontrol agents:

a. Induced Systemic Resistance (ISR): Indirect toxic effects on pathogens by volatile substances such as ethylene, released by the metabolic activities of the antagonist. Systemic resistance in plants that is triggered by certain strains of non-pathogenic root-colonizing bacteria it signaling requires jasmonic acid and ethylene.

b. Growth Promotion: Biocontrol agents, both fungal and bacterial, are reported to induce the growth of various crops. These responses may be due to-

- suppression of deleterious root micro-flora including those not causing obvious disease,
- production of growth stimulating factors (hormones or growth factors) and/or
- Increased nutrient uptake through solubilization and sequestration of nutrients and/or enhanced root growth.
- Enhanced root development is also helpful in tolerating the biotic and abiotic stresses by the plants**.**

Table 3.2: Plant growth promotion by *Pseudomonas* spp.

Crop	Biocontrol agents	Special features	References
Bean	*Pseudomonas putida*	Increase in shoot and root length and plant weight	Anderson and Guera, 1985
Cauliflower	*P. fluorescens*	Increase in root and shoot growth	Mukherjee *et al.*, 1989
Chickpea	*P. fluorescens*	Increase in root and shoot growth	Pandey, 1996
Cotton	*P. fluorescens*	Increase in plant growth	Sakthivel *et al.*, 1987
Melon	*P. fluorescens*	Increase in germination and dry weight	Ahmed and Baker, 1988
Pigeonpea	*P. fluorescens*	Increase in root and shoot growth	Mehrotra, 1988
Rice	*P. fluorescens*	Increase in plant growth	Mishra and Sinha, 2000.
Tomato	*P. fluorescens*	Promotion of shoot and root length and increase in seed germination	Varshney, 1997
Wheat	*P. fluorescens*	Shoot and root growth promotion	Pierson and Weller, 1994;

3.4 Delivery methods of bio-control agents:

Successful application of biological control requires more knowledge-intensive management (Heydari*et al*., 2004). Biological control agents can be applied using different methods such as seed treatment, root inoculation, cutting and seedling root dip, soil treatment, plant inoculation, furrow application and wound application.

3.4.1 Seed Treatment:

Talc based *Trichoderma viride* formulation used as dry seed treatment @ 4-5 g/ kg seed. Treating seeds with bacterial cultures like *Pseudomonas fluorescens* @ 10 g /kg seeds will improve plant growth and productivity by protecting against plant pathogens.

3.4.2 Seedling root dipping:

Trichoderma harzianum powder 4-5 g dissolves in one liter water and dip seedling for 30 min before transplanting in main field.

Talk based formulation of *P. fluorescent* strain by dipping the seedling in water containing mixture of 20g/l for 2h and later transplanting it in the field.

3.4.3 Seed Bio-Priming:

Treating of seeds with bio-control agents and then incubating under warm and moist conditions until just prior to emergence of radical is referred to as bioprimming. This technique has potential advantages over simple coating of seeds as it results in rapid and uniform seedling emergence. Biopriming could also reduce the amount of biocontrol agents that is applied to the seed.

3.4.4 Foliar Application:

The foliar spray of talc-based powder formulation of *P. fluorescent* strain Pf1 (1 kg/ ha) commencing from 45 days after transplanting at 10 days interval for 3 times depending upon disease intensity. If there is no disease incidence, a single spray is sufficient.

3.4.5 Soil Application:

The talc-based product of *Trichodema* spp / *Pseudomonas* spp @ 2.5 Kg/ha mixed with 50 kg well rotten FYM/sand then applied in soil. Soil has a variety of beneficial and harmful microbes. Introducing PGPR strains into the soil will boost the numbers of beneficial bacterial antagonists, thus reducing the growth of harmful microbes in the area of infection.

3.5 Commercially Available BCAs:

Recently, many small and large business owners have entered the commercial manufacturing of biocontrol agents, resulting in the introduction of different biocontrol products to the worldwide market. The commercialization process of these products includes several steps, such as:

1. Isolation of micro- organism from the natural ecosystem.
2. Evaluation of bio-agent both in vitro and in vivo conditions.
3. Testing of the best isolate under field conditions.
4. Mass production
5. Creating Formulation
6. Delivery
7. Compatibility
8. Registration and release.

Table 3.3: List of Commercially available BCAs

Sr. No	Bio control agent	Product	Target disease/pathogen	Crop
1.	*Ageobacterium radiobacter strain 84*	Galtrol	*Agrobacterium tumefaciens*	Ornamentals, Fruits, Nuts
2.	*Ageobacterium radiobacter strain* K 1026	Nagol	*Agrobacterium tumefaciens*	Ornamentals, Fruits, Nuts

Sr. No	Bio control agent	Product	Target disease/pathogen	Crop
3.	*Bascillus subtillus* strain GB34	GB34	*Rhizoctonia, Fussarium*	Soyabean
4.	*Bascillus subtillus* strain GB03	Kodiac, companion	*Rhizoctoni, Aspergillus*	Wheat, barley, peas
5.	*Pseudomonas aureofaciens* strain TX-1	Bio–jet, spot less	*Pythium, Rhizoctonia solani*	Vegetables and Ornamentals ingreen houses
6.	*Pseudomonas fluorescence* strain A506	Frostban	Fire blight, bunch rot	Fruit crop, Tomato, Potato
7.	*Streptomycine griseoviridis*	Mycostop	Soil borne pathogens	Ornamentals, Tree seedlings
8.	*Trichoderma harzianum T 22*	Root shield, plant shield	Soil borne pathogens	Green house nurseries
9.	*Trichoderma harzianum T-39*	Trichodex	*Botrytis cinerea*	Most of the food crops
10.	*Ampelomyces quisquallis* isolate M-10	AQ10	Powdery mildew	Fruits, Vegetables Ornamental
11.	*Aspergillus flavus* AF36	Alfa guard	*Aspergillus flavus*	Cotton

3.6 An ideal Characteristic of BCAs:

1. Broad spectrum activity in controlling many types of diseases.
2. Nonpathogenic to plant, human beings etc.
3. Provides control for extended periods.
4. High degree of host specificity.
5. Genetically stable.
6. Long Shelf life.
7. Fast growth and sporulation capacity.
8. Efficacious under different environmental condition.

3.7 Advantages of Biocontrol agent's over chemical control:

1. It can be combined with bio fertilizers.
2. High degree of host specificity.
3. Cost effective.
4. Safe to handle.
5. Occur naturally and non-phytotoxic.
6. They can multiply easily in soil and don't have residual problem.
7. Improvement in overall crop health and yield.

3.8 Disadvantages/Limitations of Biocontrol Agents:

1. Specific to particular pathogen so different strains are required for different pathogen.
2. It requires expert supervision.
3. Less effective than chemical pesticides.
4. Shelf life is short.
5. Efficiency depends on environmental conditions.
6. Doesn't completely destroy a pathogen.

3.9 Future Prospects:

Biological control is environmentally friendly and active means of decreasing or mitigating of pathogens/ pest. The goal of biocontrol agents is to promote technology and science. The future of biopesticides would fully depend on adoption of application of bioagent. They must able to control /suppress the pathogen and also prevent them relatively in proper manner to conventional methods. Researcher, producers, and farmer should widely explore the uses. It is able to the safe environment and also sustainable crop production.

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