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Review

Commercial Biocontrol Agents and Their Mechanism of Action in the Management of Plant Pathogens

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Abstract: Plant diseases are among the main constraints affecting the production and productivity of crops both in terms of quality and quantity. Use of chemicals continues to be the major tactic to mitigate the menace of crop diseases. However, because of the environmental concerns, health conscious attitude of human beings and other hazards associated with the use of chemicals, use of bio agents to suppress the disease causing activity of plant pathogens is gaining importance. The purposeful utilization of living organisms whether introduced or indigenous, other than the disease resistant host plants, to suppress the activities or populations of one or more plant pathogens is referred to as bio control. Biological control involves the use of beneficial organisms, their genes, and/or products, such as metabolites, that reduce the negative effects of plant pathogens and promote positive responses by the plant. In this direction, a number of commercial products have been registered both at national and inter-national levels based on different fungal and bacterial antagonists. These commercial products include, Biocon, Biogaurd, Ecofit, F-Stop, Soilgaurd etc with *Tricoderma* sp. as active ingredient, and Mycostop, Rhizoplus Subilex etc utilizing various Bacillus species as active ingredient. Biological control can achieve the objective of disease suppression through a number of ways such as antibiosis,

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competition, mycoparasitism, cell wall degradation and induced resistance, plant growth promotion and rhizosphere colonization capability. The most effective bio agent studied till date appears to antagonize pathogen using multiple mechanisms as in *Pseudomonas*, utilizing both antibiosis and induction of host resistance to suppress the disease causing microorganisms. Pseudomonas putida strain WCS358r genetically engineered to produce Phenazie and 2, 4- diacetyl-phloroglucinol (DAPG) displays improved capacities to suppress diseases in field grown wheat. Additional DAPG producers aggressively colonize roots that further contribute to increased disease suppression in the rhizosphere of wheat through competition for organic nutrients. As the bioagent represents a living system, it needs to be mass produced and formulated into various commercial products in a way it remains viable for at least two years.

Keywords: Biological control, metabolites, *Tricoderma* sp., 2, 4- diacetyl-phloroglucinol and rhizosphere

1. Introduction

Plant diseases need to be controlled to maintain the quality and abundance of food, feed, and fibre produced by growers around the world. Different approaches may be used to prevent, mitigate or control plant diseases. Beyond good agronomic and horticultural practices, growers often rely heavily on chemical fertilizers and pesticides. Such inputs to agriculture have contributed significantly to the spectacular improvements in crop productivity and quality over the past 100 years. However, the environmental pollution caused by excessive use and misuse of agrochemicals, as well as fearmongering by some opponents of pesticides, has led to considerable changes in people's attitudes towards the use of pesticides in agriculture. Moreover, the health consciousness of the people coupled with the development of resistance to pathogens due to continuous use of the chemicals also contributes to the restricted use of chemicals in crop protection. Under such circumstances exploitation of living organisms to reduce the disease causing activity of pathogenic micro-organisms seems to be the most appropriate alternative to chemicals. This phenomenon of reducing the amount of inoculums or the disease causing activity of plant pathogens accomplished through the use of living organisms other than the man is called as bio-logical control. In other words Biological control refers to the purposeful utilization of introduced or resident living organisms, other than disease resistant host plants, to suppress the activities and populations of one or more plant pathogens (Pal and Gardener, 2006).

2. Historical Back Ground

William Roberts in 1874 demonstrated the antagonistic action of micro-organisms in action of micro-organisms in liquid culture between *Pencillium glaucum* and bacteria and introduced the term antagonism. The term biological control as a feasible preposition of plant disease management was coined for the first time by C. F. Von in 1914. Since then various bio- control products have been found to be very effective in controlling the plant disease. Sanford (1926) observed that the potato scab was suppressed by green manuring antagonistic activities. Weindling (1932) reported the parasitic nature of *Trichoderma lignorumon* several plant pathogens. Grossbard (1948-1952), Wright (1952-1957), and others demonstrated that antibiotics were produced in soil by *pencilium, Aspergillus, Trichoderma, streptomyces* ssp. Kloepper (1980), demonstrated the importance of siderophores produced by *Erwinia carotovora*. Howell (1993) reported P and Q strains of *Trichoderma* sp.

3. Commercial Products of Bio-Control Agents

Unlike the bio control of insects, bio control of plant diseases is relatively new. The first bacterium called *agrobacterium radiobacter* strain K 84 was registered with the United States Environmental Protection agency (EPA) for the control of crown gall in 1979. Ten years later the first fungus *Trichoderma harzianumi* ATCC 20476 was registered with the EPA for the control of plant diseases. Currently a total of 14 bacteria and 12 fungi have been registered with the EPA for the control of plant diseases (Fravel, 2005). Most of these are sold commercially as one or more products. The technology of commercialization is still in its initial phase. 65% of the EPA registered organisms have been registered within the past 10 years while the remaining 36% regestered over the past 5 years. Many technological problems were overcome and shifts in tinking occurred for these products to reach the shelves. Some of the commercially available bio control products available in the market are shown in table 1.

Commercialization of Bio Control Products

Although the number of bio control products in plant disease management is increasing, these products still represent only 1% of the agricultural control measures while fungicides account for 15% of total chemicals used in agriculture (Friavel *et al.*, 2005). In recent years many small and large entrepreneurs have entered into the commercial production of bio control agents resulting into the entry of various bio- control products into the world market.

Commercialization of bio-control products is a multi-step process involving a wide range of activities:

- a) Isolation of micro- organism from the natural ecosystem.
- b) Evaluation of bio-agent both in vitro and under glass house conditions

- c) Testing of the best isolate under field conditions
- d) Mass production
- e) Formulation
- f) Delivery
- g) Compatibility
- h) Registration and release

Table 1: List of bio control products

SN	Bio control agent	Product	Target	Crop	Manufacturer
			disease/organism		
1	Ageobacterium radiobacter strain 84	Galtrol	Agrobacterium tumefaciens	Ornamentals, Fruits, Nuts	AgBioChem, USA
2	Ageobacterium radiobacter strain K 1026	Nagol	Agrobacterium tumefaciens	Ornamentals, Fruits, Nuts	Bio-care
3	Bascillus subtillus strain GB34	GB34	Rhizoctonia, Fussarium	Soyabean	Gustafon, USA
4	Bascillus subtillus strain GB03	Kodiac, companion	Rhizoctonia, Aspergillus	Wheat, barley, peas	Growth products,USA
5	Pseudomonas aureofaciens strain TX-1	Bio-jet, spot less	Pythium, Rhizoctonia solani	Vegetables and Ornamentals ingreen houses	EcoSoil system
6	Pseudomonas fluorescence strain A506	Frostban	Fire blight, bunch rot	Fruit crop, Tomato, Potato	Plant Health Technologies
7	Streptomycine griseoviridis	Mycostop	Soil borne pathogens	Ornamentals, Tree seedlings	Kemira Oy, Finland
8	Trichoderma harzianum T- 22	Root shield, plant shield	Soil borne pathogens	Green house nurseries	Bio works, USA
9	<i>Trichoderma harzianum</i> T-39	Trichodex	Botrytis cinerea	Most of the food crops	Bio works, USA
10	Ampelomyces quisquallis isolate M-10	AQ10	Powdery mildew	Fruits, Ornamentals , Vegetables	Ecogen,USA
11	Aspergillus flavus AF36	Alfa guard	Aspergillus flavus	Cotton	Circleone globa,USA
12	Gliocladium catenulatum strain JI446	Prima stop soil guard	Soil borne pathogens	Vegetables, Herbs , Spices	Kemira Agro Oy, Finland
13	Gliocladium virensGL-21		parasitic nematodes	Food, Fibre,	-Do-

Isolation of Micro-Organisms from Natural Eco System (Discovery)

The ultimate success of bio control depends on how well the searching and screening process is done. There is no single way to search or screen. Both depend on the target pathogen, the crop and the cropping system. It is a result of extensive screening of natural soils for the presence or absence of the isolate with desirable traits, and usually involves selection of healthy plant s from otherwise infested fields with the target pathogen followed by recovery of microbial colonies from leaves roots or rhizosphere, on the assumption that isolate will contain strain antagonistic to the pathogen (Chaube *et al.*, 2003).

These isolates are then screened for their activity against the pathogen in laboratory and green house conditions. Any suitable isolate found are then evaluated for their efficacy under the field conditions. The isolates that fail to perform well in the field are again subjected to the *In vitro* evaluation to ascertain the cause of their failure before they could be rejected.

4. Mechanism

Plant diseases are the result of interactions among the components of disease triangle i.e. host, pathogen and environment. Biological control agents are the organisms that interact with the components of disease triangle to manage the disease. Understanding how the bio control agents work can facilitate optimization of control as well as help to screen for more efficient strains of the agent. Understanding the mechanisms of biological control of plant diseases through the interactions between biocontrol agent and pathogen may allow us to manipulate the soil environment to create conditions conducive for successful biocontrol or to improve biocontrol strategies (Chet, I. 1987.). Mechanism of some bio-control agents are now understood in detail (Zhang *et al.*, 2002). Understanding the mechanism of action of a bio control agent may improve the consistency of control either by improving the mechanism or by using the bio- control agents under conditions where it is predicted to be more successful.

Bio control agents involve a bewildering array of mechanisms in achieving disease control. However the conclusive evidences for the involvement of a particular factor in biological control is determined by the strict correlation between the appearance of factor and the biological control (Handelsman *et al.*, 1989).

Various mechanisms employed by the bio control agents in controlling the plant diseases are broadly classified into:

- a) Direct antagonisms
- b) Indirect antagonism

Direct Antagonism

Direct antagonism results from the physical contact and / or high degree of selectivity for the pathogens by bio control agent. It includes:

Hyperparasitism

Hyper-parasitism is the most considered and the most direct form of antagonism (Pal *et al.*, 2006). Hyper-parasitism involves tropic growth of bio control agent towards the target organism, coiling, final attack and dissolution of target pathogens cell wall or membrane by the activity of enzymes (Tewari, 1996). It is one of the main mechanisms involved in *Trico-derma* (Sharma, 1996). *Trichoderma harzianum* exhibits excellent mycoparasitic activity against *Rhizoctonia solsni* hyphae (Altomare *et al.*, 1999).

Mycoparasitism is under the control of enzymes. Harman (2000) reported the involvement of chitinase and β -1, 3 glucanase in the *Trichoderma* mediated biological control. Since enzymes are the products of genes, slight change in the structure of gene can lead to the production of different enzyme. Gupta et al. (1995) reported that a strain of *Trichoderma* deficient in the ability to produce endo chitinase had reduced ability to control *Botrytris cineria* but shows increased ability to control *Rhizoctonia solani*.

A single fungal pathogen can be attacked by multiple hypo parasites e.g. Acremonium altenatum, Acrodontium crateriforme, Ampelomyces quisqualis and Gliocladium virens are few of the fungi that have the capacity to parasitize powdery mildew pathogens (Kiss, 2003).

Competition

From the microbial perspective, soils and living plant surfaces are frequently nutrient limited environment. So to colonize the phytosphere, a microbe must effectively compete for the available nutrients (Pal *et al.*, 2006). Both the bio control agents and the pathogens compete with one another for the nutrients and space to get established in the environment. This process of competition is considered to be an indirect interaction between the pathogen and the bio control agent whereby the pathogens are excluded by the depletion of food base and by physical occupation of site (Lorito *et al.*, 1994).

So far as the competition for nutrients is concerned bio control agents compete for the rare but essential micronutrients, such as iron and manganese especially in highly oxidized and aerated soils. In these soils iron is present in ferric form, which is insoluble in water and where the concentration may be as low as 10^{-8} M, too low to sport the microbial growth (Lindsay, 1979). Competition for micro nutrients exists because Bio control agents have more efficient utilizing uptake system for the substances than the pathogens (Nelson, 1990). This property can be attributed to the production of iron binding ligands called siderophores as in Erwinia caratovora (Kloepper etal.,

1980). Siderophores chealate the Fe (II) ions and the membrane bind protein receptors specifically recognize and take up the Siderophore-Fe-complex (Mukhopadhyay and Mukherjee, 1998). This results in making iron unavailable to the pathogen, which produce less siderophores with lower binding power. The result is less pathogen infection and biological control.

Bio controls agents also compete with the pathogen for physical occupation of site and thereby reduce or delay the root colonization by the pathogen. For example, spray the pine sumps with the spore suspension of infection by *Heterobasidion annosum*. Because the pathogen cannot gain a foothold for establishment on host, bio control can thus reduce the severity of root rot of pine (Maloy, 1993).

Some plant pathogens depend on growth substances or stimulants to overcome their dormancy before they can cause infection and bio control agents are known to exert competition for these stimulants there by reducing their disease causing ability. These substances include fatty acids or peroxidation products of fatty acids (Harman *et al.*, 1994), volatile compounds such as ethanol and acetyldehyde (Paulitz, T. C, 1991)

5. Antibiosis

Refers to the production of low molecular weight compounds or an antibiotic by microorganisms that have a direct effect on the growth of plant pathogen (Weller, 1988). *In situ* production
of antibiotics by several different bio control agents has been measured (Thomashow *et al.*, 2002).
However, the effective quantities are difficult to estimate because of the small quantities produced
relative to the other, less toxic, organic compounds present in the phytosphere. An efficient bio control
agent is one that produces sufficient quantities of antibiotics in the vicinity of the plant pathogen
(Chaube *et al.*, 2003).

List of Antibiotics Produced by Some Biocontrol Agents

Bio agents are known to produce different types of antibiotics which act in different ways to supress the diseases or plant pathogens. Bio agents are known to produce three types of antibiotics viz., nonpolar/volatile, polar/ non-volatile and water soluble. Among all of these the volatile antibiotics are more effective as they can act at the sites away from the site of production. A list of antibiotics produced by the bio agents in supressing the activity if the plant pathogen is given table 2.

Several bio-control strains are known to produce multiple antibiotics which can suppress one or more pathogens. For example, *Bacilluscereus* strain UW85 is known to produce both zwittermycin and kanosamine (Pal and Gardener, 2006). The ability to produce multiple antibiotics probably helps to suppress diverse microbial competitors, some of which are likely to be plant pathogens. The ability to

produce multiple classes of antibiotics, that differentially inhibit different pathogens, is likely to enhance biological control. More recently, *Pseudomonas putida* WCS358r strains genetically engineered to produce phenazine and DAPG displayed improved capacities to suppress plant diseases in field-grown wheat (Glandorf *et al.*, 2001)

Table 2: List of antibiotics

SN	Antibiotic	Source	Target pathogen	Disease	Reference
1	2,4 - Diacetyl- pholoroglucinol	Pseudomonas fluorescence F113	Pythium	Damping off	(Shanahan <i>et al.</i> , 1992)
2	Agrocin 84	Agrobacterium radiobacter	Agrobacterium tumefaciens	Crown gall	(Kerr, 1980)
3	Bacillomycin D	Bacillus subtillus AU195	Aspergillus flavus	Aflatoxin contamination	(Moyne et al., 2001)
4	Bacillomycin D	Bacillus amylo liquefaciens strain FZB42	Fussorium oxysporium	Wilt	(Koumoutsi et al., 2004)
5	Xanthobacin A	Lycobacter sp. Strain K88	Aphanomyces cochlioides	Damping off	(Islam <i>et al.</i> , 2005)
6	Gliotoxin	Trichoderma virens	Rhizoctonia solani	Root rot	(Wilhite <i>et al.</i> , 2001)
7	Zwitermycin A	Bascillus cereus UW85	Pythium aphanidermatum	Damping off	(Smith <i>et al.</i> , 1993)
8	Mycostubilin	Bascillus BBG100	Pythium aphanidermatum	Damping off	(Leclere <i>et al.</i> , 2005)
9	Herbicolin	Pantoea agglomerans C91	Erwinia amylovora	Fire blight	(Sandra <i>et al.</i> , 2001)
10	Iturin	Bascillus subtillus QST713	Botrytis, Rhizoctonia solani	Damping off	(Paulitz and Blanger, 2001)

Secreation of Lytic Enzymes

Diverse microorganisms secrete and excrete other metabolites that can interfere with pathogen growth and/or activities. Many microorganisms produce and release lytic enzymes that can hydrolyze a wide variety of polymeric compounds, including chitin, proteins, cellulose, hemicellulose, and DNA. Expression and secretion of these enzymes by different microbes can sometimes result in the suppression of plant pathogen activities directly. For example, control of *Sclerotium rolfsii* by *Serratia marcescens* appeared to be mediated by chitinase expression (Ordentlich *et al.*, 1988). And, a b-1, 3-

glucanase contributes significantly to biocontrol activities of *Lysobacter enzymogenes* strain C3 (Palumbo *et al.*, 2005).

Other microbial by products may also contribute to pathogen suppression. Hydrogen cyanide (HCN) effectively blocks the cytochrome oxidase pathway and is highly toxic to all aerobic microorganisms at pico molar concentrations. The production of HCN by certain *fluorescent pseudomonads* is believed to be involved in the suppression of root pathogens. *P.fluorescens* CHA0 produces antibiotics, siderophores and HCN, but suppression of black rot of tobacco caused by *Thielaviopsis basicola* appeared primarily to be due to HCN production (Voisard *et al.*, 1989). Howell et al. (1988) reported that volatile compounds such as ammonia produced by *Enterobacter cloacae* were involved in the suppression of *Pythium ultimum*-induced damping-off of cotton.

Plant Growth Promotion

Bio agents can reduce the disease incidence of crops by increasing their growth at least during the early stages of the life cycle by the way of disease escape. The best example of this is the resistance of damping off of *Solanaceous* crops with advance of age. Chaube *et al.*, (2003) reported that bio agents both fungal and bacterial help in managing the plant diseases by promoting the growth of plants through increased solubilisation of nutrients, increased nutrient uptake through enhanced root growth and sequestration of nutrients.

Aspergillus niger strain AN-27 was reported to produce growth promoting compounds, 2-carboxy-methyl- 3 –hexyl-maleic anhydride and 2 methyl ene-3- hexyl-butanedioic acid (hexylitaconic acid)that were directly responsible for increased root and shoot length and biomass of crop plants (Selvakumar and Srivastava, 2000). *Trichoderma* preaprations have been reported to increase vigour and emergence of oat seedlings (Weindling, 1932). Seeds coated with *T. viride* increased fresh and dry weight of shoot, root and nodules of broadbeans (Woo *et al.*, 2006).

Induced Systemic Host Resistance

Induced resistance is the most indirect form of antagonism. Induced resistance can be local or systemic. Salicylic acid (SA) and non-expressor of pathogenesis-related genes1 (NPR1) are key players in systemic acquired resistance *Trichoderma harzianum* when inoculated on to roots or on to leaves of grapes provides control of diseases caused by *Botrytis cineria* on leaves spatially separated from the site of application of the bio control agent (Desmukh *et al.*, 2006). Many classes of compounds are released by the *Trichoderma* sp. into the zone of interaction and induce resistance in plants. The first class is proteins with enzymztic or other activity. Fungal proteins such as xylanase, cellulases and swollenins are secreated by *Trichoderma* species (Martine *et al.*, 2001). *Trichoderma* endochitinase can also enhance defense, probably through induction of plant defense related proteins.

Other proteins and peptides that is active in inducing terpenoid phytoalexin biosynthesis and peroxidase activity in cotton, e.g., the small protein, SM1, which has hydrophobin-like properties, were found to be produced by strains of *T. virens* (Dreuge *et al.*, 2007). Another hydrophobin-like protein produced by T22 that induces both enhanced root development and disease resistance was identified (Ruocco *et al.*, 2007). Another group of proteins that induce defense mechanisms in plants are the products of avirulence-like (Avr) genes (woo *et al.*, 2006). These are produced not only by a variety of fungal and bacterial plant pathogens but also by BCF. They usually function as race- or pathovar-specific elicitors of hypersensitive and other defense-related responses in plant species that contain the corresponding resistance (R) gene.

Saksirirat *et al.*, 2009, evaluated the efficacy of *Trichoderma* strains in inducing resistance in tomato (Fig. 1). The strains of *Trichoderma* were inoculated in soil and the activity of chitinolytic and glucanasese in the leaves was examined at 0, 5, 8, 11, 14 days after inoculation. It was found that the activity of the enzymes in the leaves of tomato increased up to 14th day. This indicates that trichoderma was effective in inducing systemic resistance in tomato plant.

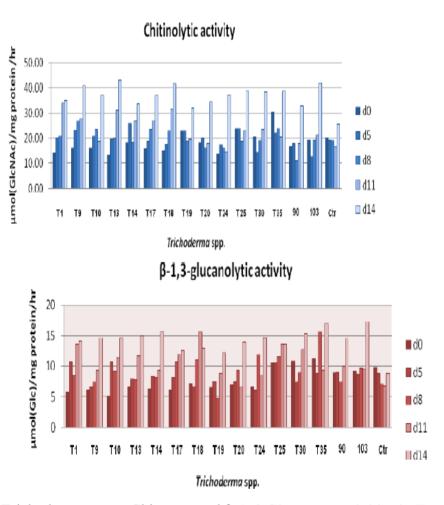


Fig. 1: Effect of Trichoderma sp. on Chitanase and β -1, 3 Glucanase activities in Tomato Leaves

6. Mass Production

One of the greatest obstacles to the biological control by introduced micro- organisms is the lack of the methods for the mass production of the bio agent. The most widely used fungal antagonist *Trichoderma* spp. have been grown on solid substrates like Sorghum grain, wheat straw, wheat bran, spent tea leaf waste, coffee husk, saw dustetc. For their mass multiplication Selvakumar and Srivastava (2000), Zaidi and Singh (2004) multiplied *Trichoderma harzianum* on pre soaked and autoclaved Jhangora seeds for 12 days at 28°C, air dried, ground and passed through 50 and 80 mesh sieves simultaneously to obtain the spore powder. The commercial formulation was prepared by diluting this powder with the talcum powder containing 1% carboxymethyl-cellulose to get the desirable concentration of biocontrol agent.

Formulations

Formulation is blending of active ingredients such as fungal spores with the inert material such as diluents and surfactants in order to alter the physical characteristics of to a more desirable form. A final formulation must:

- o Be easy to handle.
- o Be stable over a range of -5 to 35°C.
- o Have a minimum shelf- life of two years at room temperature.

One of the critical obstacles in the commercialization of bio agents is the loss of viability of the propagulues over the time. Shelf life of the bio control product is dependent on the storage temperature and the carriers used in the formulation of bio control product. A talc based preparation of the *T. virens* conidia retain 82% viability at 5 °C in refrigerator after 6 months while at room temperature same level of viability was observed for a period of 3 months (chaube *et al.*, 2003).

Sabaratnam and Traquair (2002) evaluated the shelf life of the three formulations of Streptomyces at two different temperatures of 4 °C and 24 °C and found that in general the shelf life was greater at 4 °C as compared to 24 °C (Fig. 2). This is because low storage temperature and moisture free environment slows the metabolic growth of viable propagules and prevents the accumulation of metabolic toxins and depletion of nutrients. Among the three formulations of bio agents the kaolin- talcum powder was found to retain the viability of propagules for the longest time and retained the viability of about 100% for 12 months which decreased to 80% after 24 months, drum flour retained 40% viability after 24 months.

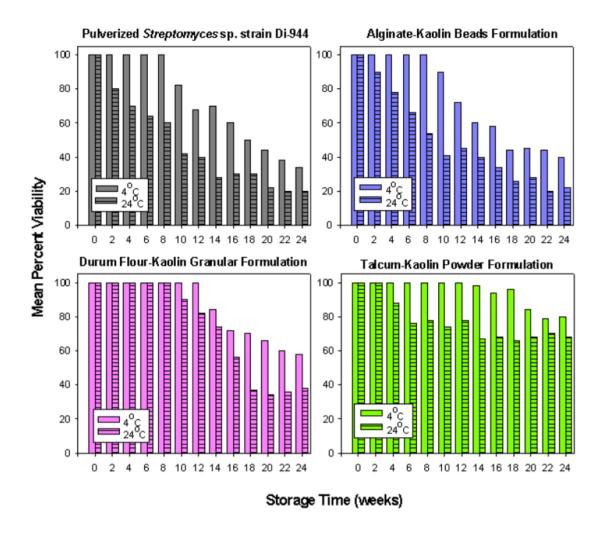


Fig. 2: Viability of Streptomyces at two different temperatures

Registration of Bioagents

Two important factors in the registration of bio agents are the toxicity and the environmental fate. Under the section 9 (3) of pesticide act of India 1968 information required for the registration of any bio pesticide are:

- o Systemic name and common name of the bio control agent
- Natural occurrence
- Morphological description of the of the bio agent
- Details of manufacturing process
- o Mammalian toxicity
- o Environmental toxicity
- Residual analysis.

7. Commercial Bio- Control Products and Pathogen Management

Effect of Mycostop on the Groth of Ceratocystis Radicicola

The effect of Mycostop on the growth of *C. radicicola* is shown as percent reduction of growth and rate of growth of mycelium was evaluated by Suliman *et al.*, 2002 (Fig. 3) and found With increase in the rate of Mycostop, there was a significant decline in both mycelial radial extension and growth rate. At the rates of 0.6 and 0.8 g/l the radial growth of *C. radicicola* appeared to have stopped after 3 days, but some fluffy vertical growth was visible. The margins of colonies were notched or lobbed with a clear zone of inhibition in the vicinity of *S. griseoviridis* colonies.

The reason for the inhibition of mycelial growth and growth rate can be attributed to the antibiosis effect of the mycostop. The ability of *S. griseoviridis* (a spore suspension of Mycostop) to produce metabolites which adversely affected sporulation and spore germination, produced deformed germlings and inhibition of mycelial growth as observed by the presence of clear inhibition zones is also indicative of antibiosis of *S. griseoviridis*.

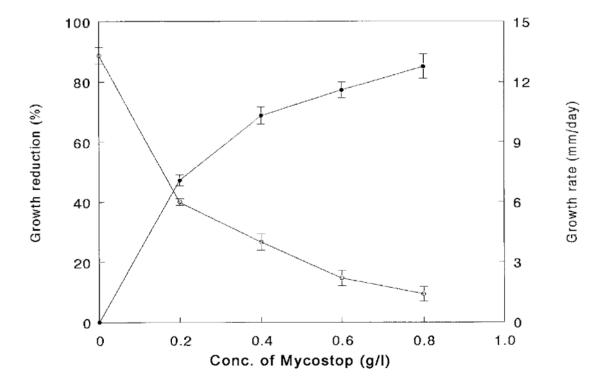
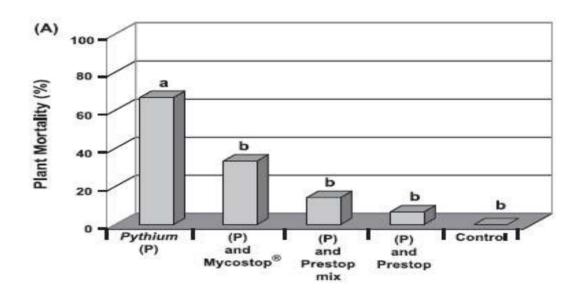


Fig. 3: The effect of Mycostop concentrations, (spore suspension of *Streptomyces griseoviridis*) on the mycelial growth and growth rate of *C. radicicola*, 5 days after treatment.

The percent mycelial growth reduction is represented as (•) and mycelial growth rate per day as (•). Bars represent SE.

Biological Control of Damping-Off and Root Rot Caused by Pythium aphanidermatum on Greenhouse Cucumbers

Acording to Punja and Raymond (2003) (Fig. 4), among the four bio control products evaluated against the damping off and root rot of tomato the bio control products were effective in reducing the mortality of tomatoes, with presstop WP and presstop mix being most effective in both the seasons (fall 2001 and fall 2002). This was followed by Mycostop. The study revealed thay the plant mortality was reduced significantly by pressstop mix andpresstop WP. The reason for the improved activity of both these formulations over the control can be attributed to the mode of action. The modeof action of these formulations is believed to involve mycoparasitism (McQuilken *et al.*, 2001). Also in invitro studies the fungus was shown to produce cell wall degrading enzyme (McQuilken *et al.*, 2001)



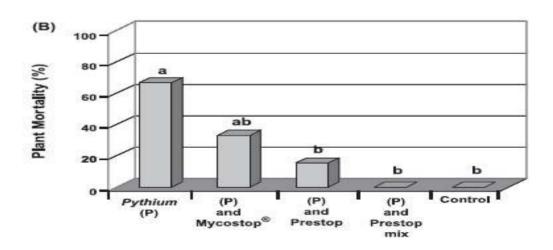


Fig. 4: Green house evaluation of four commercial bio control agents against the damping off and root rot of cucumber during fall 2001(A) and fall 2002 (B.)

8. Future Prospects

In the present crop production scenario, the biocontrol is of utmost importance, but its potential is yet to be exploited fully mainly because the research in this area is still confined to the laboratory and very little attention has been paid to produce the commercial formulations of bio agents. Moreover, whatever has been commercially produced has not been used efficiently by the farmers owing to the lack of information regarding its use. So to popularise the concept of biological control extension at University level in this direction needs to be improved.

Most of the bio agents perform well in the laboratory conditions but fail to perform to their fullest once applied to the soil. This is probably attributed to the physiological and ecological constraints that limit the efficacy of bio agents. To overcome this problem, genetic engineering and other molecular tools offer a new possibility for improving the selection and characterisation of bio control agents. Various methods that can contribute to increase the efficacy of bio agent include mutation or protoplasm fusion utilising poly ethylene glycol. There is also an urgent need to mass produce the bio agents, understand their mechanism of action and to evaluate the environmental factors that favour the rapid growth of bio control agents.

9. Conclusion

With people turning more health conscious Biological control seem to the best alternative to disease suppression. Bio agents bring the disease suppression with no environmental hazards. Research has proved that the bio agents trigger the growth of plants. Bio agents themselves being non-pathogenic to plants need to be formulated in a way that favours the activity and survival of microbe it contains. Moreover the novel concept of bio control needs a space outside the laboratory to see its fruits in present production systems.

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