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The Study of Biocontrol Effect of *Trichoderma harzianum* against *Colletotrichum*, *Corynespora*, *Fusarium graminearum* and *Fusarium napiforme*: A Comprehensive Analysis

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Abstract

Trichoderma harzianum, a ubiquitous filamentous fungus, has gained significant attention as a potent biocontrol agent against various phytopathogens. The present research is a comprehensive study on the biocontrol effect of *Trichoderma harzianum* against four important plant pathogens, namely *Colletotrichum, Corynespora, Fusarium graminearum,* and *Fusarium napiforme* isolated from *Luffa acutangula* (ridge gourd) and *Citrullus lanatus* (watermelon) by agar plate method. The study evaluates the antagonistic potential of *Trichoderma harzianum* against the four plant pathogens by dual culture assay. The results revealed that *Trichoderma harzianum* was very effective in controlling the growth of *Colletotrichum*, partially controlled *Fusarium napiforme* and was compatible with *Corynespora* and *Fusarium graminearum*. The findings of this study contribute to a better understanding of *T. harzianum* as a sustainable and eco-friendly approach for plant diseases caused by *Colletotrichum* and *Fusarium napiforme* and their management.

Keywords: *Trichoderma harzianum*, Biocontrol, *Colletotrichum*, *Corynespora*, *Fusarium graminearum*, *Fusarium napiforme*, mode of action, plant disease management

Introduction

1.1 Background:

Plant diseases caused by phytopathogens pose a significant threat to global agriculture, leading to yield losses and economic burdens. The excessive use of synthetic pesticides has raised concerns about environmental contamination and human health risks. Thus, there is an urgent need to explore alternative strategies for plant disease management. Biocontrol agents, such as *Trichoderma* species, have emerged as promising alternatives due to their efficiency, safety, and eco-friendly nature. *Trichoderma harzianum* is a species of filamentous fungus that has

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gained significant attention in the field of biocontrol due to its potential as a biological agent against plant pathogens. As a biocontrol agent, *T. harzianum* offers several advantages over chemical pesticides, making it an environmentally friendly and sustainable alternative.[1,2,3]

T. harzianum exhibits various mechanisms that contribute to its biocontrol activity. It can competitively colonize plant roots, thereby depriving pathogenic fungi of essential nutrients and space. Additionally, it produces a range of antifungal metabolites, including enzymes such as chitinases and β-1,3-glucanases, which can directly degrade the cell walls of target pathogens. Moreover, *T. harzianum* induces systemic resistance in plants, activating their defense mechanisms and making them more resistant to pathogenic attacks. The effectiveness of *T. harzianum* as a biocontrol agent has been demonstrated against several plant pathogens, including *Fusarium, Rhizoctonia*, and *Botrytis*. It has been used in various agricultural systems, including field crops, horticultural crops, and even in nurseries. *T. harzianum*-based biocontrol products are commercially available and have been successfully integrated into integrated pest management programs Furthermore, *T. harzianum* is non-toxic to humans and beneficial organisms, reducing the risks associated with chemical pesticide use. It is also known to enhance plant growth and improve nutrient uptake, further contributing to its positive impact on crop production. *Trichoderma harzianum* is a promising biocontrol agent with its ability to suppress plant pathogens through multiple mechanisms. Its environmentally friendly nature, efficacy, and potential for integration into sustainable agriculture systems make it a valuable tool in the management of plant diseases.[4,5,6,7,8]

Colletotrichum is a devastating plant pathogen responsible for various diseases worldwide. This fungal genus infects a wide range of plant species, causing significant economic losses in agriculture. It primarily targets fruits, vegetables, and ornamental plants, leading to symptoms such as leaf spots, fruit rot, and cankers. *Colletotrichum* spreads through spores that are dispersed by wind, water, and human activities. Once inside the host, it invades plant tissues, colonizing and destroying cells. Effective management strategies include cultural practices, fungicides, and resistant plant varieties. Understanding *Colletotrichum*'s lifecycle and its interactions with host plants is crucial for developing sustainable disease control measures.[9,10]

Corynespora is a genus of filamentous fungi that includes several species known to cause plant diseases. One of the most well-known plant pathogens in this genus is *Corynespora cassiicola*, which infects a wide range of plants worldwide. The symptoms of *Corynespora cassiicola* infection can vary depending on the host plant and environmental conditions. Common symptoms include leaf spots, stem cankers, blights, and fruit rot. The lesions caused by the pathogen are typically brown to dark brown, with irregular shapes.[11,12]

Fusarium graminearum is a destructive plant pathogen that poses a significant threat to various crops worldwide. It primarily infects cereals, such as wheat, barley, and maize, causing a devastating disease known as *Fusarium* head blight. This fungus produces mycotoxins, including deoxynivalenol (DON), which not only reduces crop yield but also poses health risks to humans and animals if consumed. *Fusarium graminearum* spreads through spores and can survive in plant debris, soil, and seeds. Effective management strategies involve crop rotation, resistant cultivars, and cultural practices, along with strict monitoring and early detection to minimize economic losses and protect agricultural systems.[13,14]

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Fusarium napiforme is a devastating plant pathogen that causes wilt disease in a wide range of plants. This fungal pathogen invades the vascular system of the host plant, obstructing water and nutrient transport. It secretes toxins and enzymes that further damage plant tissues, leading to wilting, stunted growth, and ultimately plant death. *Fusarium napiforme* has a remarkable ability to survive in soil for extended periods, making it difficult to control. It spreads through contaminated soil, water, seeds, and plant debris. Integrated management strategies, including crop rotation, resistant cultivars, and fungicides, are essential for minimizing the impact of this destructive plant pathogen.[15,16]

1.2 Objectives:

- Investigate the biocontrol potential of *Trichoderma harzianum* against *Colletotrichum*, *Corynespora*, *Fusarium graminearum* and *Fusarium napiforme*.
- Assess the impact of environmental factors on the biocontrol efficacy of *T. harzianum*.
- Discuss the potential applications and limitations of *T. harzianum* in plant disease management.

Methodology

- Collection of Sample

Untreated seeds of ridge gourd and watermelon were collected from Indian Institute of Horticultural Research, Bengaluru, packed in air tight bad and bought to the laboratory and stored in sterile condition till further use.

Seeds of both the types were processed using 0.2% sodium hypochlorite and inoculated on agar plate. After 48 hours, fungal infections was visible on the seeds and the fungal pathogens were mounted using lactophenol cotton blue method and identified as *Colletotrichum*, *Corynespora*, *Fusarium graminearum* and *Fusarium napiforme* by direct microscopy with the help of sporulation characteristics and colony characteristics. *Colletotrichum*, *Fusarium graminearum* and *Fusarium napiforme* were isolated from ridge gourd and *Corynespora* was isolated from watermelon.

Trichoderma harzianum was collected from laboratory of Department of Microbiology, Maharani Cluster University, Bengaluru, Karnataka and was subcultured on potato dextrose agar plate for further use.

- Biocontrol Assays

In vitro dual culture assays was performed to evaluate the antagonistic potential of *T. harzianum* against the target pathogens. Potato dextrose agar was prepared and autoclaved. Amoxicillin was added to avoid bacterial contamination. *Trichoderma* was inoculated on the plate at 1 cm far from edge and the fungus was also inoculated at 1 cm far from edge opposite to *Trichoderma*. After inoculation plates were sealed and incubated at 27°C for seven days. After seven days plates were observed for biocontrol activity and zone of inhibition was calculated and recorded. Experiments were performed in triplicates and the results obtained in the present study is the arithmetic mean of the triplicates.

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Results

Table 1: Biocontrol Efficacy of *T. harzianum*

Fungi	*Colletotrichum*	*Corynespora*	*Fusarium graminearum*	*Fusarium napiforme*
Zone of inhibition (in mm)	2 mm	No inhibition	No inhibition	1 mm

In vitro biocontrol assays demonstrated the significant antagonistic activity of *T. harzianum* against *Colletotrichum* and *Fusarium oxysporum* while *corynespora* and *Fusarium graminearum* were compatible with *Trichoderma harzianum*. The growth inhibition revealed the efficacy of *T. harzianum* in controlling the targeted phytopathogens.

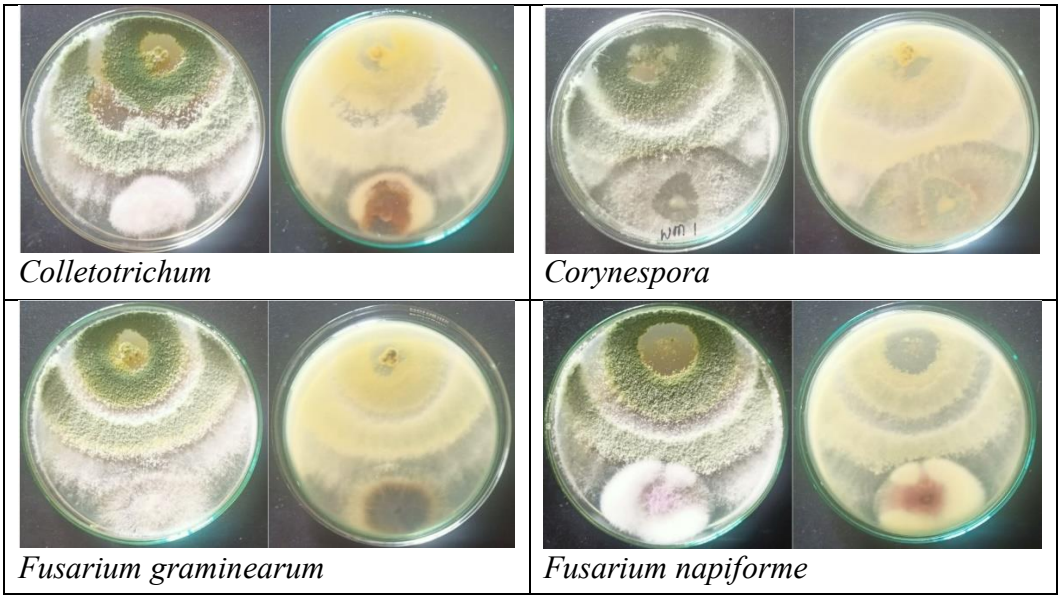

Fig 1: Biocontrol efficacy of *Trichoderma harzianum*

Discussion

Chen Jun et al., had studied the antagonistic activity of *Trichoderma spp*. against *Fusarium oxysporum*. Results revealed that *Trichoderma spp.* inhibited the growth of *Fusarium oxysporum*.[17] Similarly, in the present study *Trichoderma harzianum* has inhibited the growth of *Fusarium napiforme*. But at the same time their results are in contrast with our results where *Trichoderma harzianum* didn't inhibited the growth of *Fusarium graminearum*. Rojan P et al., had studied the antagonistic activity of *Trichoderma viride* against *Fusarium oxysporum*. Results revealed that *Trichoderma viride* inhibited the growth of *Fusarium oxysporum*.[18] Similarly, in the present study *Trichoderma harzianum* has inhibited the growth of *Fusarium napiforme*. But at the same time their results are in contrast with our results where *Trichoderma harzianum* didn't inhibited the growth of *Fusarium graminearum*. Freeman et al., had studied the antagonistic activity of *Trichoderma harzianum* against *Colletotrichum*. Results revealed that *Trichoderma harzianum* inhibited the growth of *Colletotrichum*.[19] Similarly, in the present

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study *Trichoderma harzianum* has inhibited the growth of *Colletotrichum*. Rahman Ahsanur et al., had studied the antagonistic activity of *Trichoderma* against *Colletotrichum*. Results revealed that *Trichoderma harzianum* inhibited the growth of *Colletotrichum*.[20] Similarly, in the present study *Trichoderma harzianum* has inhibited the growth of *Colletotrichum*.

The study highlights the potential applications of *Trichoderma harzianum* as a biocontrol agent in integrated disease management programs. However, certain limitations, such as variable efficacy under different environmental conditions and formulation challenges, need to be addressed for its successful implementation. Further research should focus on addressing the limitations of *Trichoderma harzianum*, including the development of improved formulations and strategies to enhance its efficacy under diverse environmental conditions. Additionally, field studies assessing the long-term effects and compatibility with other management practices would provide valuable insights for its practical implementation.

Conclusion

The biocontrol study of *Trichoderma harzianum* against four plant pathogens was studied by dual culture assay. *Trichoderma harzianum* exhibits a strong biocontrol effect against *Colletotrichum* and *Fusarium napiforme* while it is compatible with *Corynespora* and *Fusarium graminearum*. However, the biocontrol efficacy of *T. harzianum* can be influenced by environmental factors. The present study contributes to a better understanding of *Trichoderma harzianum* as a sustainable and eco-friendly approach for plant disease management, paving the way for its practical applications in agriculture.

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