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EFFECT OF *TRICHODERMA* ON FUNGI ISOLATED FROM INFECTED TOMATO LEAF

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Abstract : *Trichoderma is* well known for its antifungal and antibacterial activities. This microorganism is having application in agriculture as biofertilizer and biopesticide. During its growth it competes with the plant pathogen for the nutrients and other growth factors, during this process it can inhibit the growth of pathogens thus help in plant protection. Tomato is grown widely in Rayalaseema region, Andhra Pradesh, India. It is economically important crop grown during almost all seasons by local farmers. Tomato crop get infected with various bacteria and fungi resulting in crop loss. Our study was aimed to isolate fungi infecting the tomato crop and evaluate the antimicrobial activity of *Trichoderma.* Three different fungi were isolated from infected tomato plant leaves. The isolated fungi were tested for their susceptibility with bio control agent *Trichoderma.* It was able to inhibit the growth of tested fungi.

IndexTerms **- Trichoderma, Tomato, Fungi, leaf spot.**

I. INTRODUCTION

Fungus *Trichoderma*ins belongs to family Hypocreaceae, more prevalent soil fungi, they have avirulent, symbiotic association with plants. There are many *Trichoderma* species which are endophytes having mutualistic association with many plant species [1]. Several *Trichoderma* species have discovered to have biocontrol ability and can be useful for eliminating soil born diseases. It is being used against many pathogenic fungi such as *Fusarium, Sclerosium, Alternariea* etc. *Trichoderma* is a plant growth promoter that can solubilize phosphates and micronutrients [3]. It make the nutrients available for plant and promotes its growth. The present study was designed to analyse the effect of *Trichoderma* fungus in inhibiting the growth of plant pathogens isolated from infected tomato leaves. Tomato (*Solanum lycopersicum*) is a widely used vegetable crop grown worldwide. It's a main ingredient in various dishes. It is nutritional, being a rich in vitamins A, C, potassium, and antioxidants like lycopene. Tomatoes are consumed fresh, cooked, sauces, ketchup, canned tomatoes and juiced. Tomato plant is with compound leaves, yellow self-pollinating flowers and berry type commonly red fruits. Fruits also found in yellow, green colours. Plant grows in sunlight at varying temperatures requiring regular watering. Pest and Disease Management. Diseases effect the plant growth and production. Common diseases occurring are blight, fusarium wilt, nematodes. Integrated Pest Management (IPM) strategies include crop rotation, resistant varieties, biological control agents and judicious use of pesticides. Tomatoes having high value with demand for commercial and domestic usage. The diseases caused by pathogens including fungi, bacteria, and viruses reduce the plant growth and production. Some important diseases of tomato plant include fungal, bacterial diseases [4]. Early Blight caused by *Alternaria solani* with symptoms of dark, concentric spots on older leaves. Leaves turn yellow and drop prematurely. Septoria Leaf Spot caused by *Septoria lycopersicicum.* With symptoms of small, water-soaked spots which enlarge with time and develop big with gray centers and dark edges. Leaves turn yellow and fall. Bacterial leaf spot disease by *Xanthomonas campestris pv. Vesicatoria* is with a small, water-soaked spots that become dark and raised. Spots can appear on leaves, stems, and fruit. Bacterial Speck by *Pseudomonas syringae pv.* In this infection small numerous and smaller dark spots with yellow halos on leaves, stems, and fruit are seen. Target Spot caused by *Corynespora cassiicola is with* Brown, circular spots with light centers on leaves and stems. Severe infections leads to defoliation. Grey leaf spot caused by *Stemphylium solani* is with small, dark, irregularly shaped spots on matured leaves which expand to entire leaf. The present study is designed to study the effect of *Trichderma* on pathogenic fungi isolated from infected leaves of tomato plant. We have identified the leaf infesting fungi from tomato plant based on the disease symptoms. We have isolated the organisms on suitable media and tested for the inhibitory activity of the *Trichoderma* on plant pathogens. The results indicate that this plant growth promoter can be involved in plant protection by its antimicrobial properties.

II. MATERIALS AND METHODS

2.1 Isolation of Fungi from tomato

The infected leafs were collected from the fields in vellatur village near yogi Vemana university, Kadapa, Andhra Pradesh, India.A randomly selected diseased leaf was carefully examined and cut in to small pieces (3 mm in diameter) with a sterilized blade, surface sterilized in 1% hypochlorite for 2 min, plated on Potato dextrose agar (PDA) aseptically and then incubated at 28°C for 5 days. A pure culture was obtained and maintained by sub-culturing each of the different colonies that emerged onto the PDA plates and incubating at 28°C for 5 days. As a control, each of the healthy leaves were sterilized with 75% ethanol. The leaves were cut into small segments (3 mm in diameter) with a sterile blade, placed on PDA and then incubated at 28°C for 5 days.

2.2 Preparation of fungi for simple microscopic observation:

Trichoderma culture of fungus from PDA plate was taken with help of a needle along with the mycelium and fungal spores. It was carefully spread on a glass slide Lacto phenol cotton blue stain was added and cover slip was placed. The prepared slide was observed under 45x magnification.

2.3 Inhibitory effect of *Trichoderma* on fungi:

The inhibitory effect of *Trichoderma* was tested by dual culture method. The *Trichoderma* was inoculated at one side and test organism was inoculated on other side of the PDA plate and incubated for 4days. Growth of the *Trichoderma* and the pathogen was measured.

III. RESULTS AND DISCUSSION

3.1 Isolation of Fungi from tomato:

The infected leaf cut in to small pieces and plated on Potato dextrose agar incubated at 28°C for 5 days. After 5 days of incubation fungal colony growth was observed from the edges of the infected leaf. A pure culture was obtained and maintained by sub-culturing each of the different colonies that emerged onto the PDA plates and incubating at 28°C for 5 days (Fig 1). Pure cultures of the fungi were used for further experiments.

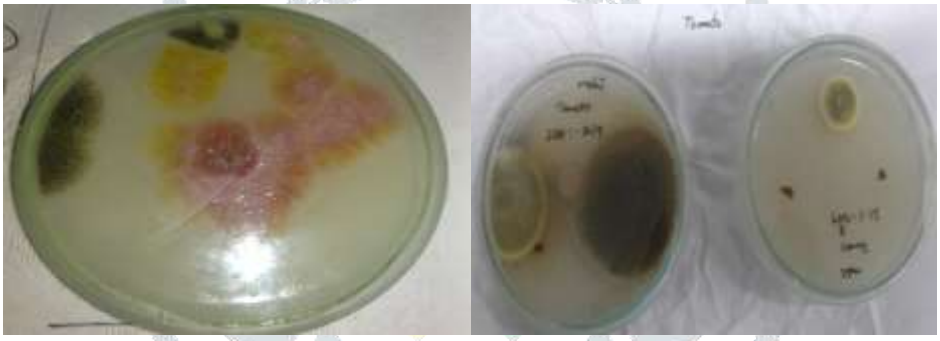

Fig 1: Isolated Fungi from infected tomato leaf

3.2 Preparation of fungi for simple microscopic observation:

Trichoderma culture of fungus from PDA plate was taken with help of a needle along with the mycelium and fungal spores. It was carefully spread on a glass slide Lacto phenol cotton blue stain was added and cover slip was placed. The prepared slide was observed under 45x magnification (Fig 2).

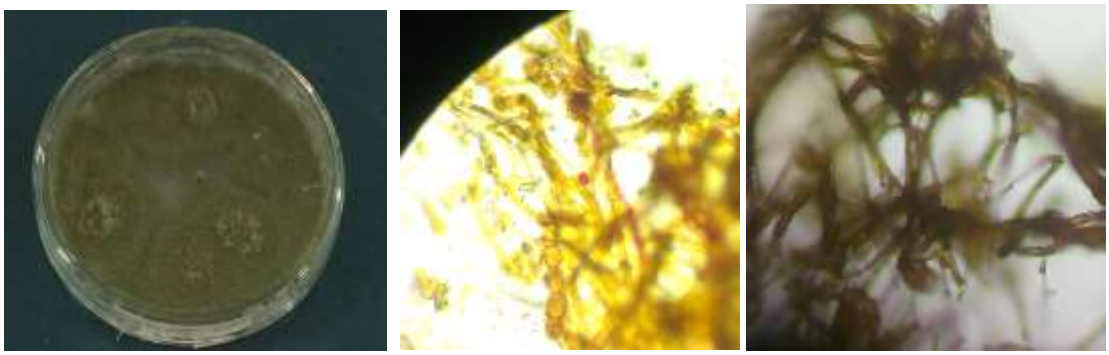

Fig 2: Trichoderma pure culture & Trichoderma under microscope 45 X

3.3 Inhibitory effect of *Trichoderma* on fungi:

The inhibitory effect of *Trichoderma* was tested by dual culture method. The *Trichoderma* was inoculated at one side and test organism was inoculated on other side of the PDA plate and incubated for 4days. Growth of the *Trichoderma* and the pathogen was measured Fig 3. *Trichoderma* exhibited good growth inhibition for Isolate F3 Fungus. The two other fungal isolates F2 and F3 were able to grow in presence of Trichoderma, but the growth was less when compared with the growth of *Trichoderma* (Table 1).

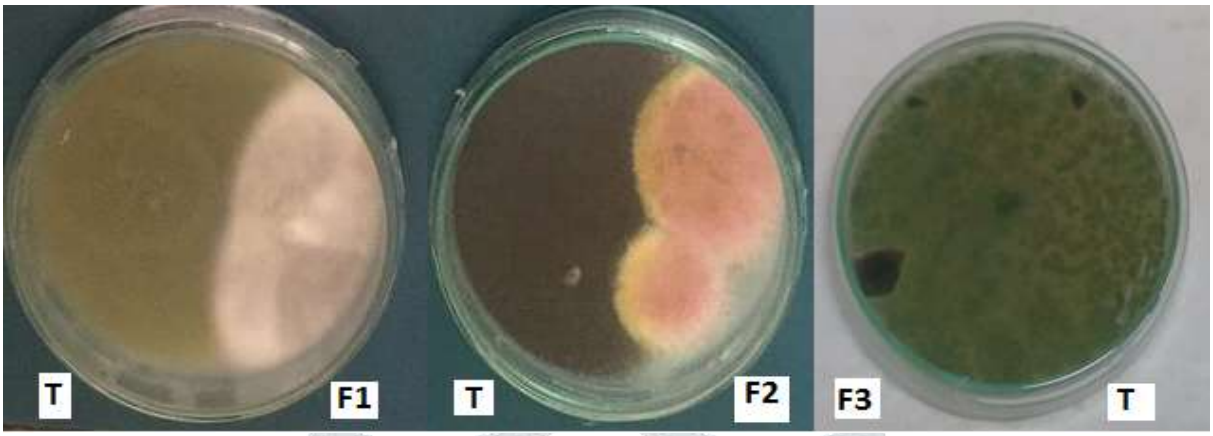

Fig 3: Inhibitory effect of Trichoderma on fungi isolated from tomato
T : Trichoderma F1, F2, F3 : fungal isolates from tomato

Table 1: Anti-fungal activity: Effect of *Trichoderma* on fungal isolates. Results are mean values of triplicates.

Trichoderma Growth in (mm)	Fungal Isolate from Infected Tomato	Fungal Isolates Growth in (mm)	Growth Inhibition effect of *Trichoderma* on fungal isolates
55	F1	35	Less Inhibition
58	F2	23	Less Inhibition
75	F3	15	Effective Inhibition

Several *Trichoderma* species have been identified for their antimicrobial properties [5]. These fungi are present in all types of soils. They are the most prevalent fungi. Many species in this genus can be characterized as avirulent plant symbionts and bio pesticides [6]. In our study we have screened three fungal isolates F1, F2 and F3 from infected tomatoes. *Trichoderma* was able to inhibit isolate F3 effectively. The study can further extended to sequence the isolates and study the effect of *Trichoderma* in controlling the fungal pathogens by field experiments.

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