

Efficacy of Different Fungicides and Bio Control Agents Against *Fusarium oxysporum*, Causal Agent of Potato Dry Rot

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Abstract

Objectives: To study the prevalence of potato dry rot in different vegetables markets. To evaluate the effect of *Fusarium oxysporum* on potato. To study the efficacy of different antagonistic agents and fungicides against mycelia growth of *F. oxysporum*. **Methods/Statistical Analysis:** Survey was done from different vegetable markets of Pakistan. Potatoes showing dry rot symptoms were collected and brought to Plant Pathology laboratory. **Findings:** Antagonistic organisms cause highly significant inhibition in the growth of *F. oxysporum* which was higher than 60%. Lowest growth of *F. oxysporum* was found because of an interaction of *P. varioti* (15.5 mm) and *Paecilomyces lilacinus* (16.75 mm). Both them cause 82.39% and 80.96% inhibition in the growth of targeted pathogen respectively. Whereas in case of interaction with *T. harzianum* and *Trichoderma polysporum* the growth of *F. oxysporum* was 22.00 mm and 27.75 mm, which is still significantly low as compared to the growth of *F. oxysporum* 88.00 mm in separate control plates. The growth of pathogen was inhibited by *Paecilomyces* spp. and mutual inhibition of both antagonist and pathogen at few mm was observed. Whereas, in the case of *Trichoderma* spp. pathogen and antagonist produce intermingled growth, the growth of the *F. oxysporum* was ceased and overgrown by antagonist. In-vitro amendment of fungicide in culture media inhibits the colony growth of *F. oxysporum*. Reduction in colony diameter of *F. oxysporum* was observed with the application of used antagonistic fungi. **Application/Improvements:** These results can be used in the analysis and bio-control methods of Potato dry rot.

Keywords: Bio-Control, Dry Rot, Fungicides, *Fusarium oxysporum*, Potato

1. Introduction

Potato (*Solanum tuberosum* L.) belongs to the family solanaceae that includes other 2000 plant species. Tomato (*Lycopersicon esculentum* L.), sweet pepper (*Capsicum annuum* L.), eggplant (*Solanum melongena* var. *esculentum* L.), tobacco (*Nicotiana tabacum* L.), and petunia (*Petunia hybrid* L.) also belongs to this family. Potato ranks fourth

after wheat, rice and maize in the list of most important staple crop of the world.

It is a good source of iron while their vitamin C contents help in iron absorption. Its consumption is increasing because it is not only cheap but also a rich source of carbohydrates, starch and contributed a lot in the reduction of food shortage globally¹.

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The losses caused by diseases and insects constitute the major constraints that faced by the potato growers worldwide. Apart from the field diseases, postharvest diseases caused considerable economic losses in the quality and quantity of the produced during transport, storage and marketing². Among the diseases the most important wide spread and important caused by pathogenic fungi, affecting tubers and vegetative parts³. A number of field and storage disease of potatoes that reduces the quantity and market value are Black scurf⁴, Late blight (*Phytophthora infestans*)⁵, Early blight (*Alternaria solani*)⁶, Powdery scab (*Spongospora subterranea*)⁷, Wilt disease (*Verticillium albo-atrum*)⁸, Fusarium dry rot (*Fusarium* spp.), Silver scurf (*Helminthosporium solani*), Gangerene (*Phoma exigua*)⁹, Pink rot (*Phytophthora erythroseptica*) and Watery wound rot (*Pythium ultimum* and *P. debaryanum*)¹⁰.

One of the main fungal diseases that attack potato is Fusarium dry rot. This problem persists throughout the world. There are many species of Fusarium reported to cause dry rot of potato worldwide³. Fusarium dry rot caused by many Fusarium species like *F. coeruleum*, *F. eumartii*, *F. oxysporum*, *F. sulphureum* and *F. sambucinum*¹¹. It has been estimated that dry rot caused by Fusarium spp. caused 6 to 25% yield losses¹². Adding to this yield loss, *Fusarium* spp. are also well known for the toxins production in attacked host and responsible for mycotoxicoses of humans and animals^{13,14}. One of the toxins produced by the Fusarium spp. which because dry rots is trichothecene. This toxin is a serious inhibitor of protein synthesis in prokaryote and can cause serious health troubles¹⁵.

In Turkey, four Fusarium spp. namely *F. sambucinum*, *F. solani*, *F. culmorum* and *F. oxysporum* are found to be common causes of dry rot of potatoes and have the potential for complete destruction of potatoes in storage². Eighteen percent of tubers arriving at New York markets from 1972-1980 showed symptoms of Fusarium dry rot¹⁶. While as high as 60% of graded tubers in Scotland was affected by dry rot¹⁷.

Symptoms of Fusarium dry rot include minute brown areas on the surface of tubers^{18,19}, the infected tubers appear wrinkled and rolled tissues from the surface, and rot also creates depressions/ cavity in the surface of the tuber. These affected tissues turn brown, grey or black. When symptoms advances spore masses of blue, white, yellow, purple, black or pink colour may also observe. Seed tubers and potatoes for consumption may not com-

pletely. Storage tuber mummifies and ultimately only the dry shell persists^{20,21}. Mode of spread is by planting infected tubers or by contaminated soil, as the pathogen is soil borne, airborne or carried in plant residue²².

Biological control of dry rot of potato using different antagonists only evaluated for experimental purposes²³. Antagonistic organisms like *Trichoderma* spp. and *Pseudomonas aeruginosa* have been found to be effective management strategy²⁴.

Fungicide like Maxim MZ, Tops MZ, and Moncoat MZ may be used for the efficient control of potato dry rot. Many chemicals, including Thiabendazole, may be applied to seed tubers before sowing²⁵. Continuous use of same fungicide is the main factor to develop resistance for it as documented in potato dry rot pathogen against Thiabendazole¹¹.

Indiscriminate use of chemical pesticides to control various pests and pathogenic microorganisms of crops plants is causing health hazard both in terrestrial and aquatic lives through their residual toxicity²⁶. Much attention is being focused on the alternative methods of pest control²⁷.

Keeping in view the importance of potato dry rot recent studies carried out to determine the prevalence of potato dry rot at study area and to evaluate the efficacy of different antagonistic agents and fungicides against *F. oxysporum* causal agent of dry rot of potato.

2. Materials and Methods

2.1 Survey and Sampling of Diseased Specimen

Survey was done from different vegetable markets of Hyderabad region. Potatoes showing dry rot symptoms were collected and brought to Plant Pathology laboratory. Incidence of dry rot of potatoes was also recorded with help of following formula:

$$\text{Incidence (\%)} = \frac{\text{Number of potatoes with dry rot}}{\text{Total number of potatoes observed}} \times 100$$

2.2 Isolation and Identification of Causal Fungus

The isolation and identification of fungi was carried out as described by² as follows; tubers were washed under running tap water to remove the mud and then air dried. A 6

mm diameter and 5 mm deep pieces were excised with a cork borer from the affected area of each tuber. The tubers sections were surface sterilized in 5% commercial bleach solution for 1 min. Tuber sections were dried on sterilized paper and plated on Potato Dextrose Agar (PDA). After 5 days of incubation at room temperature under natural light, predominantly isolated fungal colonies developing from the plant material were identified by microscopic observations with the help of literature.

Pure culture of fungus was maintained by periodical transfer on PDA plates. Small colony from corner of the fungal growth was picked up with help of inoculation needle and placed on the surface of new freshly prepared PDA plates. Only one disc transferred per PDA plate and incubated at 25°C resulting pure culture were multiplied periodically on new media throughout the study.

2.3 Pathogenicity Test of Causal Pathogen

Pathogenicity test of causal pathogen was carried out for the confirmation of disease-causing fungus under *in-vitro* conditions to prove the Koch's postulates. The procedure was performed according to Peters²⁸. The healthy potato tubers of variety "Diamond" were used in this experiment. Tubers appearing uniform in size (100-120 g) were selected for this test. First tubers were washed to remove the surface soil and sterilized by dipping them in 80% solution of ethanol and then air dried. Then the tubers wounded with a cork borer with a diameter of 5 mm to a depth of 5 mm^{28,29}. An agar plug (5 mm diameter) containing active growth of *F. oxysporum* isolates cut from the margin of a 3-day-old cultures grown on PDA and placed into the wound, which was subsequently sealed with the excised plug of tuber tissue. All the wounded potato tubers were wrapped in polyethylene bags and incubated in the dark at 20°C for 3 weeks. As a control, tubers were wounded with the help of cork borer like it's done in treatment then inoculated with only an agar plug without fungus. After three weeks data were recorded on the basis of symptoms development and lesion area were measured in cm with the help of scale.

2.4 Evaluation of Different Bio-Control Agents

Different biocontrol agents were obtained From Agriculture Research Institute, Tandojam to evaluate against Fusarium dry rot of potato by dual culture method. Briefly PDA plates were prepared and

inoculated by *F. oxysporum* and selected biocontrol agent aseptically. Both of them were placed at the periphery of Petri plate at equidistance of 2 to 3 cm in opposite direction. Petri plates inoculated with pathogen only served as control. All the plates were incubated at 25°C in incubators. Plates were observed regularly and data on colony growth in cm were recorded with the help of scale and antagonistic nature of the bio agent was recorded. Resulting data on colony diameter was calculated for percent inhibition over control with the help of formula given below:

$$\text{Inhibition (\%)} = \frac{C-T}{C} \times 100$$

Whereas, C= Growth of pathogen in control plates; T= Growth of pathogen in dual culture plates.

After prolonged storage interactions were assessed using a key based on observations of Dickinson and Boardman³⁰ as given below:

- A. Mutually intermingling growth where both fungi grew into one another without any microscopic signs of interaction.
- Bi. Intermingling growth where the fungus being observed was growing into the opposed fungus either above or below its colony.
- Bii. Intermingling growth where the fungus under observation has ceased growth and is overgrown by another colony.
- C. Slight inhibition where the fungus approach each other until almost in contact and a narrow demarcation line, 0.1-2 mm, between the two colonies clearly visible.
- D. Mutual inhibition at a distance of > 2 mm.

2.5 Efficacy of Different Fungicides

The efficacy of different fungicides for controlling *F. oxysporum* responsible for postharvest infection was carried out with ten different fungicides. For this purpose, Carbendzim, Topsin-M, Mancozeb, Antracol, Gemstar, Scholar, Nativo, Tilt, Score and Radomil were selected and evaluated with four different doses, i.e., 1 ppm, 10 ppm, 100 ppm, 1000 ppm by food poisoning method under *in-vitro* conditions. The details of fungicides with their company name, active ingredients and brand name are given in Table 1.

Table 1. List of the fungicides used in the experiments

Brand Name	Company Name	Active ingredient
Topsin-M	Arysta	Thiophanate Methyl 70 WP
Mancozeb	FMC	Dithiocarbamates 75% WP
Antracol	Bayer	Propineb 70% WP
Carbendazim	Clear	Carbendazim 50 WP
Ridomil	Sygenta	Metaxyl 68 WP
Score	Sygenta	Difinaconazole 250 EC
Tilt	Sygenta	Propiconazole 250 EC
Nativo	Bayer	(Tebuconazole+Trifloxystobin)75WG
Scholar	Sygenta	Fludioxonil 230 SC
Gemstar	Sun Crop	Azoxystrobin 250 EC

Concentrations given above were prepared by serial dilution method. The required quantity of fungicide was mixed in the medium after sterilization of media. Medium without fungicide were served as control. Before pouring the media were also amended with streptomycin sulphate at 5 ml/L and penicillin at 10^6 units/L to avoid bacterial contamination. Media without fungicide served as control. Equal volume of media i.e., about 15 ml were poured in each Petri dish and inoculated in the centre with a 5 mm disk of *F. oxysporum* after solidification of media. These plates were incubated as and data were recorded as described above. Growth was recorded daily till any of the plate found full of the growth of *F. oxysporum*. Finally resulting data was calculated for inhibition percent because of fungicide with the help of formula as follows:

$$\text{Inhibition (\%)} = \frac{C-T}{C} \times 100$$

Whereas; C = Colony diameter of pathogen in control plates; T = Colony diameter of pathogen in plates amended with fungicide (treatment plate).

3. Results

3.1 Prevalence of Potato Dry Rot

Potatoes showing dry rot symptoms were collected from different markets (Figure 1). Hundred potatoes were randomly examined from each store. The disease found to prevail in 70% of the stored visited with a very low incidence. Average disease incidences of dry rot of potatoes

were recorded 3.4%. The isolation and identification of fungi was carried out by tissue isolation methods. Isolations were made with affected potatoes. Total 100 sections were placed over the surface of PDA medium. After incubation of 5 days at 25°C fungal colonies identified by microscopic observations with the help of literature as *Fusarium* spp. and *Aspergillus* spp. *F. oxysporum* was appeared as most predominant fungus with 59% frequency (Table 2). Pure cultures of *F. oxysporum* were maintained on PDA medium and were multiplied periodically on new media throughout the study. On PDA medium it produced white aerial mycelia became tinged with light purple. From backside of plates dark purple and produces abounded micro and macro conidia, Microconidia were oval to ellipsoid cylindrical while macroconidia long fusoid to falcate in shape with 3 or 4 septa (Figure 2). These morphological characteristics are similar those described^{19,31}.



Figure 1. Potato tubers showing dry rot disease symptoms.

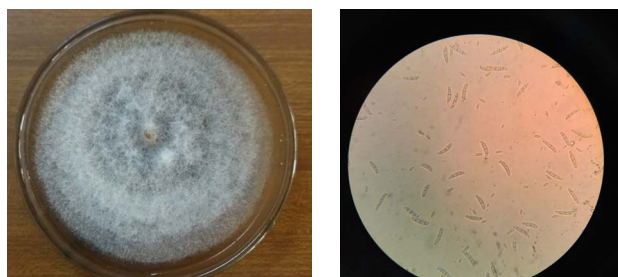


Figure 2. Growth of *F. oxysporum* isolated from potato tubers having dry rot of potato disease on PDA and macro and micro conidia under 400X magnification of compound microscope.

Table 2. Disease incidences of potato dry rot collected from 10 different vegetables market of Hyderabad regions and frequency of isolated fungi

Incidences (%)	= (34/1000) *100 3.4 %
Frequency of isolated fungi	<i>Fusarium oxysporum</i> (58%)
	Another <i>Fusarium</i> spp. (30%)
	<i>Aspergillus</i> spp. (22%)

3.2 Pathogenicity Test *F. oxysporum* on Potato Tubers

Pathogenicity test of *F. oxysporum* was carried out for the confirmation of disease-causing fungus under in-vitro conditions to prove the Koch's postulates. After 3-week incubation, the inoculated potatoes showed wrinkled and rotted symptoms of typical dry rot of potato. The rotted areas of the potatoes were brown, grey, or black and the rot creates depressions in the surface of the tuber. White fungal growth was also apparent on rotted areas. Lesion area after 20 days of inoculation was extended up to 27.10 mm.

Very small lesion were also found to develop in uninoculated control potatoes (1.8 mm), where tubers were wounded with the help of corn borer like it's done in treatment then inoculated with only an agar plug without fungus (Figure 3).

Effect of different fungicides on colony growth of *F. oxysporum*:

The efficacy of different fungicides against the colony growth of *F. oxysporum* was checked under laboratory conditions. For this purpose, Radomil, Topsin-M, Antracol, Mancozeb, Carbendzim, Score, Tilt, Scholar, Nativo, and Gemstar, were selected and evaluated with

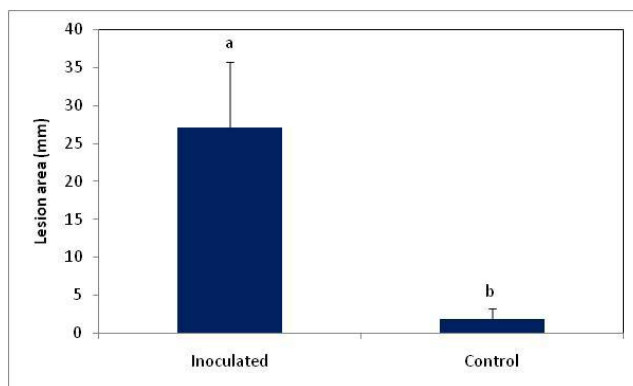


Figure 3. Effect of artificial inoculation of *F. oxysporum* on lesion development. Data was recorded after 22 days of incubation at 25°C.

five different doses, i.e., 1, 10, 100, 1000 and 10000 ppm by Food poisoning method. All concentrations of fungicides reduced the growth of *F. oxysporum* as compared to control. However, higher concentrations were more effective than the lower ones. The growth of the test pathogen gradually decreased with increasing concentrations. All the fungicide at 10000 ppm completely stops the growth of test pathogen.

Score (63.80 mm), followed by Carbendazim (69.33 mm), Gemstar (70.5 mm) and Mancozeb (70.80 mm) at produces the lowest growth as compare to other fungicides at the same dose. Topsin-M, followed by Scholar and Score at 10 ppm produces 29.16 mm, 45.66 mm and 53.83 mm respectively, after seven days of incubation at 28°C as compared to the 89.66 mm growth at control. Topsin-M (9.83 mm and 2.16 mm) followed by Mancozeb (21.50 mm and 5.33 mm) and Scholar (25.00 mm and 6.00 mm) produces lowest growth respectively at 100 and 100 ppm as compare to other fungicide at the same dose (Figure 4).

IC50 values for each fungicide were also calculated. It greatly varied for each fungicide. Lowest IC50 value was found in case of Topsin-M, Scholar, Antracol and Radomil i.e., 8.65 ppm, 16.99 ppm, 36.45 ppm and 46.89 ppm, respectively. Whereas, highest IC50 value was found in Nativo followed by Tilt and Gemstar i.e., 240.36, 206.32 and 109.96, respectively (Figure 5 and 6).

3.3 Effect of Different Bio-Control Agents on the Growth of *F. oxysporum*

Four different bio-control agents i.e. *Paecilomyces lilacinus*, *Trichoderma harzianum*, *T. polysporum* and *P. varioti* were obtained from ARI Tandojam and evaluated against

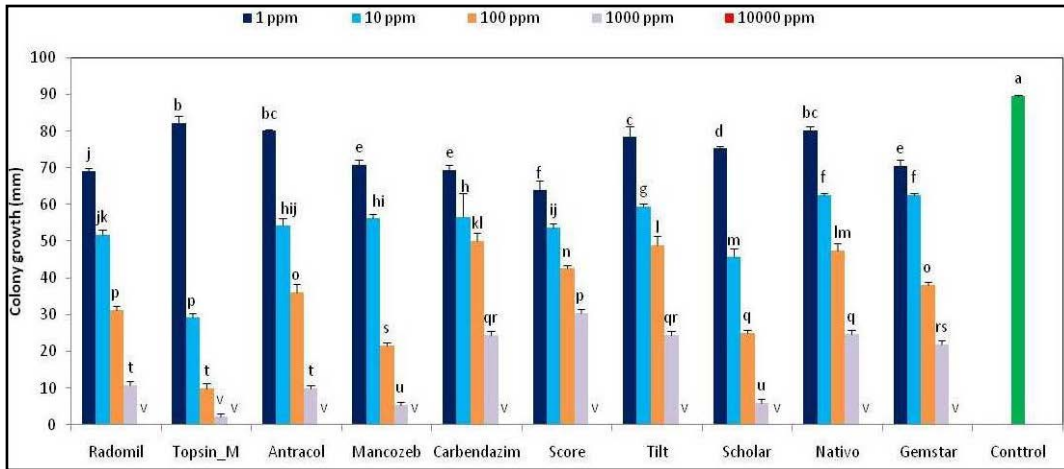


Figure 4. Effect of different concentrations of various fungicides on the colony growth of the *F. oxysporum*. Bar with different letters show significant difference ($P \leq 0.05$) as determined by LSD.

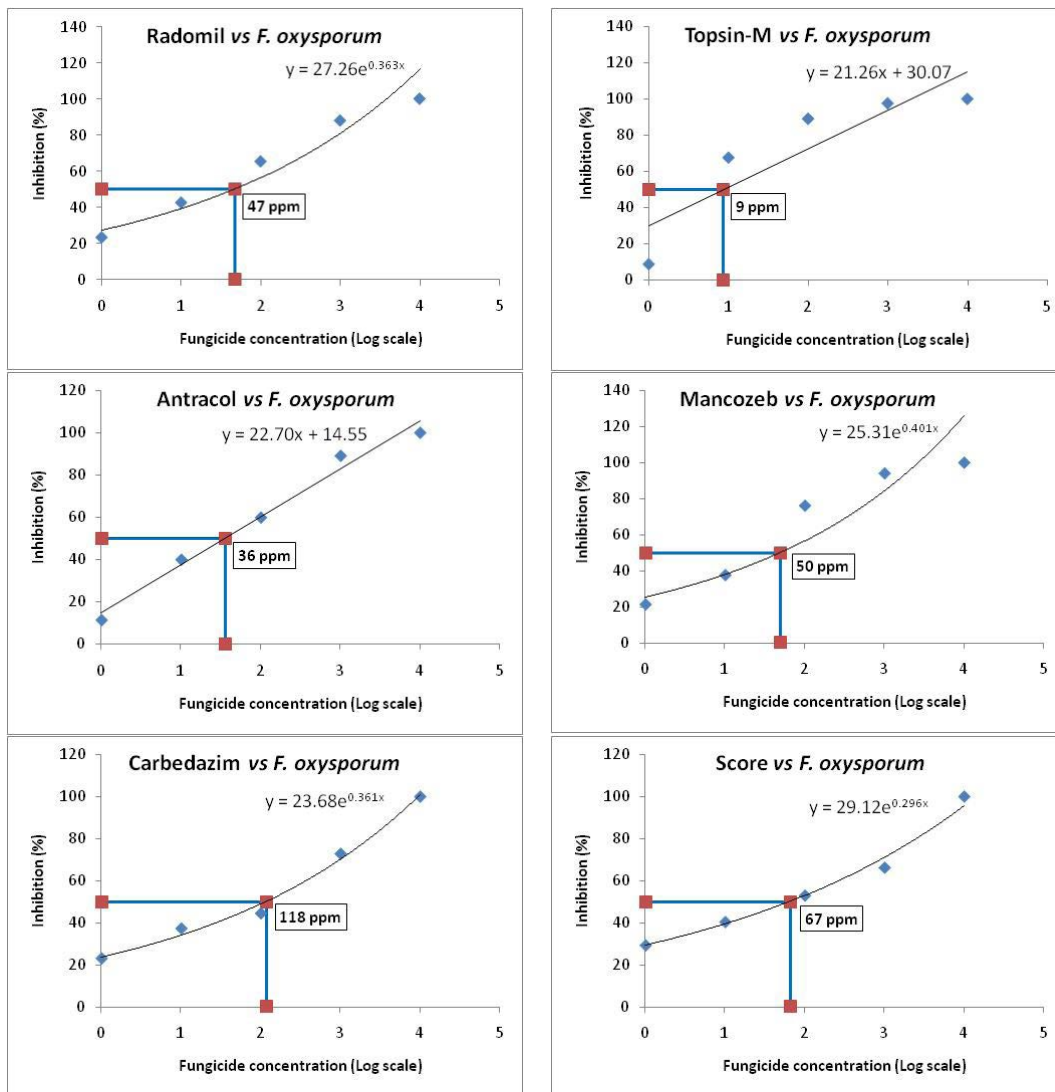


Figure 5. IC50 value of Radomil, Topsin-M, Antracol, Mancozeb, Carbendazim and Score against *F. Oxysporum*.

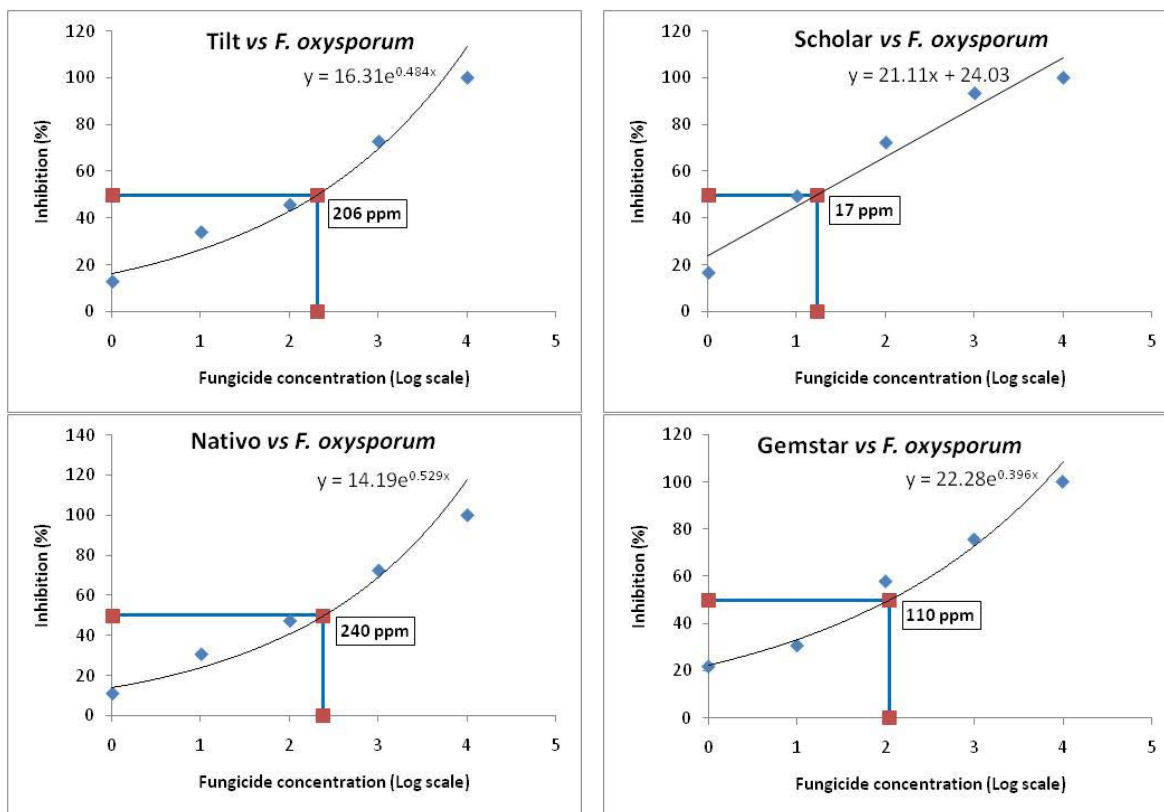


Figure 6. IC50 value of Tilt, Scholar, Nativo and Gemstar against *F. Oxysporum*.

F. oxysporum by dual culture method. Both of them were placed at the periphery of Petri plate at equidistance in opposite direction. All four antagonistic organisms cause highly significant inhibition in the growth of *F. oxysporum* which was higher than 60%. Lowest growth of *F. oxysporum* was found as a result of interaction of *P. varioti* (15.5 mm) and *P. lilacinus* (16.75 mm). Both of them cause 82.39% and 80.96% inhibition in the growth of targeted pathogen, respectively. Whereas in case of interaction with *T. harzianum* and *T. polysporum* the growth of *F. oxysporum* was 22.00 mm and 27.75 mm, which is still significantly low as compare to the growth of *F. oxysporum* 88.00 mm in separate control plates. *T. harzianum* and *T. polysporum* cause 75.00% and 68.46% inhibition in the growth of *F. oxysporum* (Figure 7). Antagonistic nature of the bio-agents was recorded with prolonged incubation. Both specie Paecilomyces of shows D type interaction i.e. mutual inhibition of both at a distance of few mm while Trichoderma spp. shows Bii type interaction i.e., *F. oxysporum* and *Trichoderma* spp. produces intermingled growth; growth of the *F. oxysporum* was ceased and overgrown by antagonist (Figure 8).

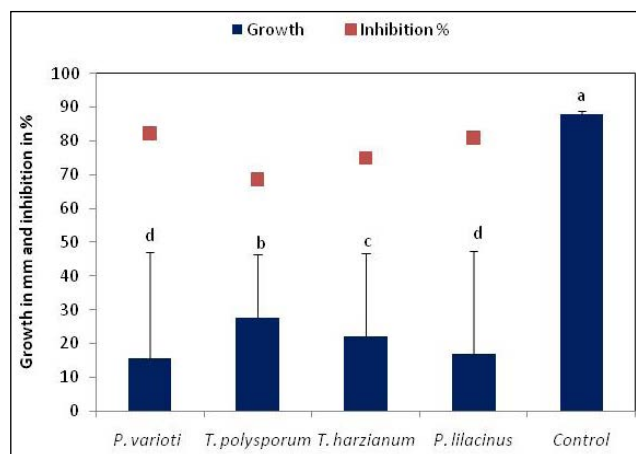


Figure 7. Effect of *Paecilomyces varioti*, *Trichoderma polysporum*, *T. harzianum* and *P. lilacinus* on the growth/inhibition of *F. oxysporum*. Bar with different letters show significant difference ($P \leq 0.05$) as determined by LSD.

4. Discussion

Dry rot potato is one of the main fungal pathogens that attack potato throughout the world. The disease found to

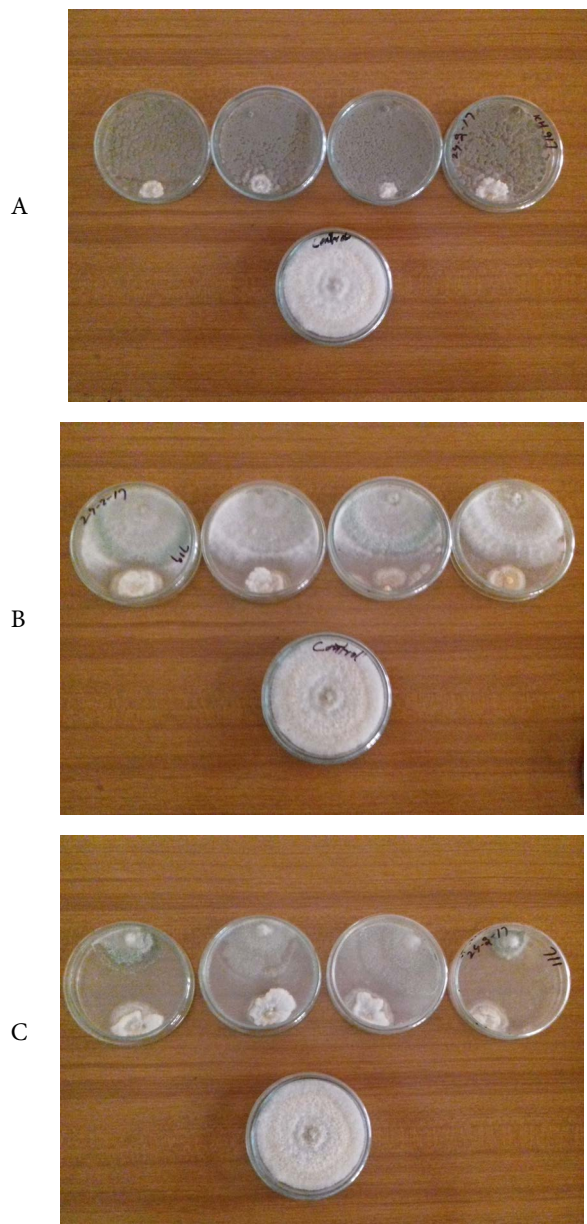


Figure 8. Effect of different biocontrol fungi on the growth/inhibition of *F. oxysporum* A=*Paecilomyces varioti*; B=*Trichoderma harzianum* and C= *T. Polysporum*.

prevail in 70% of the stored visited in Hyderabad region with a very low incidence. Average disease incidences of dry rot of potatoes were recorded 3.4%. It has been estimated dry rot caused by *Fusarium* spp. causes 6% to 25% yield losses¹².

Symptoms of *Fusarium* dry rot include minute brown areas on the surface of tubers¹⁸, the infected tubers appear wrinkled and rolled tissues from the surface, and rot also creates depressions/cavity in the surface of the tuber.

These affected tissues turn brown, grey or black. When symptoms advances spore masses of blue, white, yellow, purple, black or pink colour may also observe.

Seed tubers and potatoes for consumption may rot completely. Storage tuber mummifies and ultimately only the dry shell persists^{14,20,21}.

The isolation from diseases tissues revealed the association of *Fusarium* spp. and *Aspergillus* spp. *F. oxysporum* was appeared as most predominant fungus. There are many species of *Fusarium* reported to cause dry rot of potato worldwide³. *Fusarium* dry rot caused by many *Fusarium* species like *F. coeruleum*, *F. eumartii*, *F. oxysporum*, *F. sulphureum* and *F. sambucinum* Fuckel¹¹. Mejdoub-Trabelsi²⁴ isolated four *Fusarium* spp. predominantly associated with dry rot of potato. i.e., *F. sambucinum*, *F. oxysporum*, *F. solani* and *F. graminearum*. On PDA medium it produced morphological characteristic that are similar those described by^{19,31}.

Pathogenicity test of *F. oxysporum* confirmed the virulent nature of the pathogen and produces the similar symptoms of potato dry rot. Author in³² confirmed the pathogenicity of two *F. oxysporum* isolates associated with potato dry rot in Colombia. Inoculation with both induced moderate dry rot. Mode of spread is by planting infected tubers or by contaminated, as the pathogen is soil borne, airborne or carried in plant residue²². Therefore, emphasis must be given to control dry rot of potato. So far, this disease is managed with the application of fungicides.

Continuous use of same fungicide is the main factor to develop resistance for it as documented in potato dry rot pathogen against Thiabendazole¹¹. Therefore, in present study different fungicide determined against *F. oxysporum* causal agent of dry rot of potato. Radomil, Topsin-M, Antracol, Mancozeb, Carbendzim, Score, Tilt, Scholar, Nativo, and Gemstar, were selected and evaluated with five different doses, i.e., 1, 10, 100, 1000 and 10000 ppm by food poisoning method. All concentrations of fungicides reduced the growth of *F. oxysporum* as compared to control. However, higher concentrations were more effective than the lower ones. The growth of the test pathogen gradually decreased with increasing concentrations. All the fungicide at 10000 ppm completely stops the growth of test pathogen. Similarly, Piwoni³³ found sixty isolates of *F. avenaceum* and forty isolates of *F. coeruleum* sensitive to used fungicide, while eighty-five isolates of sixty eight percent of *F. sulphureum* and one isolate of *F. culmorum* were found not sensitive to Thiabendazole. All

the *Fusarium* spp. were sensitive to Imazalil and were pathogenic when inoculated into potato tubers. Yasmin³⁴ also found Azoxystrobin, Quinoline, Hymexazol and Fludioxonil with inhibitory effect on mycelial growth of *F. oxysporum* f. sp. *tuberosi*.

IC50 values for each fungicide were also calculated. It greatly varied for each fungicide. Lowest IC50 value was found in case of Topsin-M, Scholar, Antracol and Radomil. Whereas, highest IC50 value was found in Nativo followed³⁵ tested six fungicides; Carbendazim, Benomyl, Prochloraz, Azoxystrobin, Fludioxonil and Bromuconazole, against *F. oxysporum* f. sp. *lycopersici* with seven different concentrations. Prochloraz and Bromuconazole were the most effective fungicides against the pathogen both in vitro and in vivo, followed by Benomyl and Carbendazim. Fungal radial growth was measured and median effective concentration (EC50) values ($\mu\text{g/ml}$) determined. Biological controls of dry rot with different bio-control agents such as fungi, bacteria, and yeasts have been reported as effective under experimental conditions^{23,36}. Four different bio-control agents' i.e., *Paecilomyces lilacinus*, *P. varioti*, *Trichoderma harzianum* and *T. polysporum*. All four antagonistic organisms cause highly significant inhibition in the growth of *F. oxysporum* which was higher than 60%. Antagonistic organisms like *Trichoderma* spp. and *Pseudomonas aeruginosa* have been found to be effective management strategy²⁴. Author in³⁴ found reduction in colony diameter of *F. oxysporum* isolated from potato dry rot by *T. harzianum*, *T. viride* and *T. virens*.

Antagonistic nature of the bio-agents was recorded with prolonged incubation. Growth of pathogen was inhibited by *Paecilomyces* spp. and mutual inhibition of both antagonist and pathogen at a distance of few mm was observed. Whereas in case of *Trichoderma* spp. pathogen and antagonist produces intermingled growth, growth of the *F. oxysporum* was ceased and overgrown by antagonist. Similar interaction was also reported³⁷. Author in³⁴ evaluated different *Trichoderma* spp. against *F. oxysporum* isolate from potato dry rot i.e., *T. harzianum*, *T. viride* and *T. virens* and studied interaction mechanisms which include disintegration of host cytoplasm and/or alteration into cords and/or coiling around pathogen hyphae.

5. Summary

Dry rot is considered the most important post-harvest disease that attack potato throughout the world especially

for seed production where are store for prolonged duration. The disease found to prevail in 70% of the stored visited in Hyderabad region with very low incidences. Average disease incidences of dry rot of potatoes were recorded 3.4%. Symptoms of *Fusarium* dry rot include minute brown areas on the surface of tubers. The infected tubers appear wrinkled and rolled tissues from the surface, rot also creates depressions/ cavity in the surface of the tuber. These affected tissues turn brown, grey or black. When symptoms advance fungal spore masses of varying colour may also observed. The isolation from diseases tissues revealed the association of *Fusarium* spp. and *Aspergillus* spp. *F. oxysporum* was appeared as most predominant fungus. Pathogenicity test of *F. oxysporum* confirmed the virulent nature of the pathogen and produces the similar symptoms of potato dry rot.

So far, this disease is managed with the application of fungicides. Continuous use of same fungicide is the main factor to develop resistance for it as documented in potato dry rot pathogen against Thiabendazole. In present study, different fungicides were tested against causal agent of dry rot of potato. Ten fungicide i.e., Radomil, Topsin-M, Antracol, Mancozeb, Carbendazim, Score, Tilt, Scholar, Nativo, and Gemstar, were evaluated against *F. oxysporum* with five different doses, i.e., 1, 10, 100, 1000 and 10000 ppm by food poisoning method. All concentrations of fungicides reduced the growth of *F. oxysporum* as compared to control. However, higher concentrations were more effective than the lower ones. The growth of the test pathogen gradually decreased with increasing concentrations. All the fungicide at 10000 ppm completely stops the growth of test pathogen. IC50 value for each fungicide was also calculated from fungal radial growth at five different concentrations. It greatly varied for each fungicide. Lowest IC50 value was found in case of Topsin-M, Scholar, Antracol and Radomil. Whereas, highest IC50 value was found in Nativo followed.

Four different bio-control agents' i.e., *Paecilomyces lilacinus*, *P. varioti*, *Trichoderma harzianum* and *T. polysporum*. All four antagonistic organisms cause highly significant inhibition in the growth of *F. oxysporum* which was higher than sixty percent. Antagonistic nature of the bio-agents was recorded with prolonged incubation. Growth of pathogen was inhibited by *Paecilomyces* spp. and mutual inhibition of both antagonist and pathogen at a distance of few mm was observed. Whereas in case of *Trichoderma* spp. pathogen and antagonist produces

intermingled growth, growth of the *F. oxysporum* was ceased and overgrown by antagonist.

6. Conclusions

Prevalence of potato dry rot in Hyderabad, Sindh, efficacy of different antagonistic agents and fungicides against *F. oxysporum* causal agent of dry rot of potato are studied. Different *Fusarium* spp. and *Aspergillus* spp. were found associated with the collected potatoes with varying frequencies. *F. oxysporum* was appeared as most predominant isolated fungus with the maximum frequency of 59%. Typical symptoms of dry rot of potato appeared on artificially inoculated tubers with *F. oxysporum*. Fungal fruiting bodies also appeared on rotted areas after prolonged storage.

In vitro fungal growth test in the presence of different fungicide were performed in order to find out best fungicide. Ten fungicides viz., Radomil, Topsin-M, Antracol, Mancozeb, Carbendzim, Score, Tilt, Scholar, Nativo and Gemstar were evaluated against *F. oxysporum* with five different doses, i.e., 1, 10, 100, 1000 and 10000 ppm by food poisoning method. Fungal diameter in the Petri dishes was recorded every day till any of the treatment find full of fungal growth. All concentrations of fungicides reduced the growth of *F. oxysporum* as compared to control. However, higher concentrations were more effective than the lower ones. The growth of the test pathogen gradually decreased with increasing concentrations. All the fungicide at 10000 ppm completely stops the growth of test pathogen.

IC50 value for each fungicide was also calculated from fungal radial growth at five different concentrations, it greatly varied for each fungicide. Lowest IC50 value was found in case of Topsin-M, Scholar, Antracol and Radomil. Whereas, highest IC50 value was found in Nativo followed.

Four different bio-control agents i.e. *Paecilomyces lilacinus*, *P. varioti*, *Trichoderma harzianum* and *T. polysporum* were tested against dry rot pathogen. All four antagonistic organisms cause highly significant inhibition in the growth of *F. oxysporum* which was higher than sixty percent. Growth of pathogen was inhibited by *Paecilomyces* spp. and mutual inhibition of both antagonist and pathogen at few mm was observed. Whereas in case of *Trichoderma* spp. pathogen and antagonist produces intermingled growth, growth of the *F. oxysporum* was ceased and overgrown by antagonist.

7. Recommendation

The results of the present study shows that dry rot of potato prevail in study area but with low incidences. However, to reduce the present incidences and to reduce the further spread management methods should be evaluated. In-vitro amendment of fungicide in culture media inhibits the colony growth of *F. oxysporum*. Therefore in-vivo application of fungicide for the control of this disease should be evaluated. Reduction in colony diameter of *F. oxysporum* was observed with the application of used antagonistic fungi. In this connection further studies should be carried out to find out the alternative of fungicide for management of this disease.

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