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EFFICACY OF SOME BIOAGENTS AGAINST *SCLEROTIUM ROLFSII* SACC. CAUSING ROOT-ROT IN *ZEA MAYS* L. CROP

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ABSTRACT

Sclerotium rolfsii is a saprotroph fungus with living plants. *Sclerotium rolfsii* is a soil-borne fungus which causes root-rot in Zea mays crop. Zea mays is a staple food in many regions of the world. It is a good nutritional crop which contains proteins, carbohydrates, fats and vitamins (B-complex, A, C & K). It maintains various organs of the body like hair, skin, digestive system, heart, liver, kidney and brain. Zea mays contain selenium element that helps proper function of the thyroid gland which maintains immune system of human body. *Sclerotium rolfsii* was isolated from infected maize from Barabanki district of Uttar-Pradesh. *Penicillium* spp., *Trichoderma viride, Aspergillus niger* and *Aspergillus flavus* were isolated from rhizospheric soil of maize. Bio-agents *Penicillium* sp. (30.20%, 35.82%, 44.22%), *Trichoderma viride* (3.21%, 23.48%, 26%), *Aspergillus niger* (22.43%, 40.98%, 49.02%) and *Aspergillus flavus* (25.18%, 27.96%, 34.39%) on 3rd, 5th, 7th day inhibited the mycelial growth of *Sclerotium rolfsii* using under dual culture technique. *Penicillium* sp. and *A. niger* were more effective antagonists than *Trichoderma viride* and *Aspergillus flavus* against mycelial growth of *S. rolfsii* under *in vitro* condition.

KEYWORD: *Zea mays, Sclerotium rolfsii, Penicillium* spp., *Trichoderma viride, Aspergillus niger* and *Aspergillus flavus*.

1. INTRODUCTION

Zea mays is commonly called Makka, which is facultative short-day and large grain plant. It is mostly 2 to 3 meters in height, internodes occur every 15 to 20 centimeters, each leaf grows 70 to 100 cm in height every one internode. Male flower and female inflorescence are separate in the same plant. Maize is a C_4 plants grow in the temperate zone. *Zea mays* are a fastener food in many countries of the world. It is high-quality nutritional crop which includes proteins, carbohydrates, fats and vitamins (B-complex, A, C & K). It sustains various organs of the body like hair, skin, digestive system, heart, liver, kidney and brain. *Zea mays* restrain selenium element that helps proper function of the thyroid gland which maintains immune system of human body. *Zea mays* productivity inhibited through bacteria, pest and fungal diseases. The fungal diseases like root-rot caused by *Sclerotium rolfsii* at the lower portion of *Zea mays* crop. *Sclerotium rolfsii* is a saprotroph fungus with living plants. *Sclerotium rolfsii* firstly attacks host plants like corn at the soil line and rapidly moves to the root as it destroys plant tissues with oxalic acid and pectolytic enzymes. *Sclerotium rolfsii* is a soil-borne which occurs worldwide and infects more than 500 plant species.[1,12] *Sclerotium rolfsii* grow at 70% soil moisture of field capacity and at a temperature range between 25^{0}C to 30^{0}C.[11] In recent year, farmers have used chemical fungicides in the field against soil-borne fungus which is more advantageous but the chemical fungicides are more dangerous for environmental pollution and threaten human being. Biological control of the pathogen neither creates environmental pollution nor human health. Use of fungal antagonists like *Aspergillus niger, Aspergillus flavus, Trichoderma viride* and *Penicillium* sp. are bio-control agents against several phytopathogens.

Nawar (2013)[9] has reported that *Aspergillus niger* and *Penicillium* sp. were most effective and inhibited the radial growth of *Sclerotium rolfsii* under *in vitro* condition. Parmar et al. (2015)[10] have concluded that *S. rolfsii* is a phytopathogenic fungus which infects crops and was controlled by *T. harzianum, T. hamantum, T. virens, T. viride, T. koningii, T. pseudokoningii* but *T. viride* (61%) inhibited maximum growth of *S. rolfsii* under *in vitro* condition.

2. MATERIALS AND METHODS

2.1 Sample collection

The samples were collected from infected agriculture field of maize crop from district Barabanki. The collection of infected maize crop of the rhizospheric region and non-rhizospheric were carried out in sterilized polythene bags. The collected samples (infected part of crops and soil) were preserved for further studies.

2.2 Isolation and identification of pathogen

Infected maize plant parts collected from the agriculture field of district Barabanki were cut into small pieces of about 2-4 mm length and sterilized with 0.5% mercuric chloride solution for 30 seconds and washed in sterilized distilled water and dried at room temperature. The sections were placed onto potato dextrose agar plates and incubated at 27±1 ^{0}C for 7 days. After 7th day full extend mycelia of *Sclerotium rolfsii* on PDA plate was observed and pure culture was maintained on PDA slant. The isolated *Sclerotium rolfsii* was identified on the basis of morphology and available literature.

2.3 Isolation and identification of antagonistic mycoflora

Antagonistic mycoflora have been isolated from rhizospheric soil of infected agriculture field of maize using serial dilution technique. The isolated antagonistic soil mycoflora were maintained on Potato dextrose agar Petriplate. Isolated antagonistic mycoflora were identified on the basis of morphology and available literature.

2.4 Dual culture technique

The isolated four antagonistic mycoflora viz. *Trichoderma viride, Aspergillus flavus, A. niger* and *Penicillium* sp., against the radial growth of *S. rolfsii* using dual culture technique under *in vitro* condition was studied. In dual culture technique, the PDA medium was prepared with standard protocol and autoclaved at 121^{0}C on 15 lb for 15 minutes. After autoclaving PDA, cooled down PDA was poured in sterilized Petriplates. Five mm diameter block of *S. rolfsii* was inoculated one side and antagonist on opposite side of the same Petriplate as a treatment and one block of *S. rolfsii* was inoculated in the center of another Petriplate as control. The treatment and control plates were incubated at 26±1^{0}C temperatures in an incubator. All treatments and control plates were set in triplicates. After incubation, on 3rd, 5th and 7th day the mycelial growth of *S. rolfsii* with control plate and mycelial growth of *S. rolfsii* with treatment plate was measured. The percent inhibition of *S. rolfsii* was calculated using following formula.

$$\text{Percent Inhibition} = \frac{\text{Control} - \text{Treatment}}{\text{Control}} \times 100$$

2.5 Data analysis

The data were analyzed statistically.

3. RESULTS AND DISCUSSION

3.1 Isolation and identification of pathogen

Sclerotium rolfsii was isolated from infected maize plant on potato dextrose agar Petriplate within seven days after inoculation. The isolated pathogen produced cottony white mycelia on potato dextrose agar plate. After five days white mycelia changed into light brown and after seven days light brown mycelia changed into dark brown and become hard; and mustard seed like "sclerotia" are formed which is a characteristic feature of *S. rolfsii*.[4] *Sclerotium rolfsii* was identified on the basis of available literature.

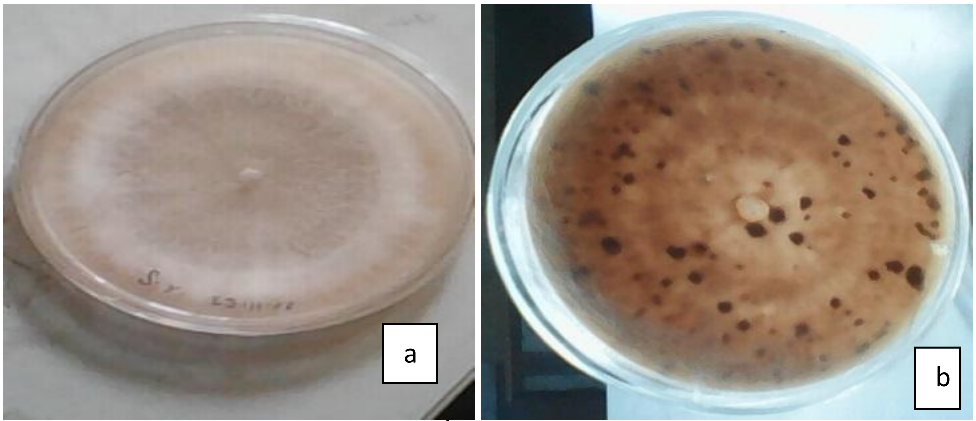

Fig. 1: a. Pure culture of *Sclerotium rolfsii* b. After 7th day old *Sclerotium rolfsii* formed sclerotia.

3.2 Isolation and identification of antagonistic mycoflora

The antagonistic mycoflora were isolated from various collected soil samples using serial dilution technique. These were isolated and identified as *Trichoderma viride, Aspergillus flavus, A. niger, Penicillium* sp.

3.3 Dual culture technique

The isolated four fungal species viz. *Trichoderma viride, Aspergillus flavus, A. niger* and *Penicillium* sp. were tested under dual culture method to find out their antagonistic activity against *S. rolfsii in vitro*. It was observed that among the test fungi, *Penicillium* sp. and *Aspergillus niger* were most effective bioagent. *Penicillium* sp. inhibited the growth of pathogen by overgrowing the mycelium of *S. rolfsii*. It showed that *Penicillium* sp. has antagonistic effect against *S. rolfsii* and percentage inhibition was recorded as 30.20%, 35.82%, 44.22% at 3rd, 5th, 7th day respectively (Table 1). In case of *A. niger* the percentage inhibition was found to be 22.43%, 40.98%, 49.03% (Table 1) followed by *T. viride* 3.21%, 23.48%, 26% (Table 1) and *A. flavus*

25.18%, 27.96%, 34.39% (Table 1) at 3rd, 5th and 7th day. The present *in vitro* study the *A. niger* and *Penicillium* sp. is more effective bioagent compared to another bioagent against *S. rolfsii*. *Trichoderma viride* was most effective inhibitor of the mycelial growth of *S. rolfsii* using dual culture technique. Karthikeyan et al. (2006)[8]; Saigan et al. (2008)[13]; Bosah et al. (2010)[3] reported that *Trichoderma viride, Aspergillus niger* and *Penicillium* species were most effective antagonists against the *S. rolfsii* causing stem rot in groundnut under *in vitro* condition. Zape et al. (2014)[14] observed that *Trichoderma viride* was most effective antagonist against inhibition of mycelial growth of *S. rolfsii* under *in vitro* condition using dual culture technique. *Aspergillus flavus* was least effective to inhibit the mycelium growth of *S. rolfsii*. Basumatary et al. (2015)[2] observed the similar result against mycelium growth of *S. rolfsii*. *Trichoderma viride* and *T. harzianum* secreted antibiotics and hydrolytic enzymes which suppresses the plant pathogen.[2] *Trichoderma viride, A. flavus, A. niger* and *Penicillium italicum* were effective bioagent against inhibition of mycelium growth of *S. rolfsii* under *in vitro* condition.[6] *Trichoderma viride* effectively inhibits the growth of *S. rolfsii* at different pH value under *in vitro* condition and 77% inhibit at 37^0 C and pH 4.[7] *Trichoderma viride*, *Penicillium* sp. and *A. niger* were more significant inhibiting the radial growth of *S. rolfsii* causing collar rot, sclerotium wilt, stem-rot, charcoal rot, seedling blight, damping-off, foot-rot, stem blight and root-rot in maize, tomato, chilli, sunflower, cucumber, brinjal, soybean, groundnut, bean, watermelon and other crops.[5]

Table 1: Percent inhibition of *S. rolfsii* by fungal antagonists using dual culture technique under *in vitro* condition

Treatment	Incubation period (Day)					
	3rd		5th		7th	
	* MG	% I	MG	% I***	MG	% I
Trichoderma viride	a35.25±0.25	3.21	47±0.25**	23.48	60.25±0.25	26.00
Aspergillus niger	28.25±0.25	22.43	36.25±0.25	40.98	41.5±0.25	49.03
Aspergillus flavus	27.25±0.25	25.18	44.25±0.25	27.96	53.42±0.14	34.39
Penicillium sp.	25.42±0.38	30.20	39.42±0.38	35.82	45.42±0.38	44.22
Control	36.42±0.38	-	61.42±0.38	-	81.42±0.38	-

*MG = Mycelial growth (in mm), ** Mean value ±SD, aValues are average of triplicate
*** % I = Percent inhibition

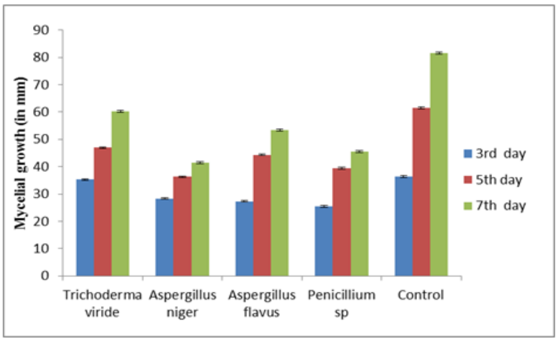

Fig 2: Mycelial growth (Mean ±SD) of *Sclerotium rolfsii* by using dual culture technique under *in vitro* condition.

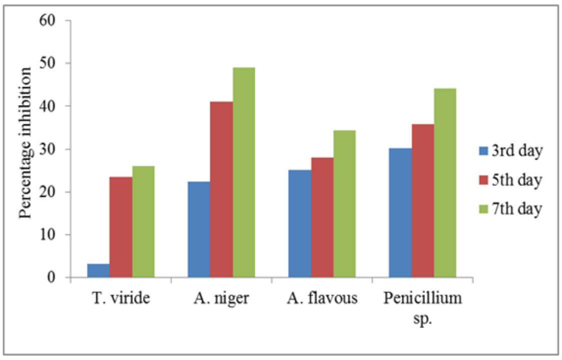

Fig 3: Percent inhibition of *Sclerotium rolfsii* by fungal antagonists using dual culture technique under *in vitro* condition.

4. CONCLUSION

The management of soil borne diseases in maize with chemicals under field condition is price prohibitive, hazardous, loss of soil fertility and cause serious environmental pollution, so, use of biological control as eco-friendly management of *Sclerotium rolfsii* is an alternative best way. The present research article concluded that biological antagonists ie. *Aspergillus niger*, *Aspergillus* flavus, *Trichoderma viride*, and *Penicillium* sp. inhibited mycelial growth of *S. rolfsii* under *in vitro* condition. *Aspergillus niger* and *Penicillium* sp. were more effective compared to *Aspergillus flavus* and *Trichoderma viride*.

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