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In vitro management of *Fusarium oxysporum* of sesamum (*Sesamum indicum*)

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Abstract
Sesamum is an important oilseed crop known as "Queen of oilseed". Sesamum seeds infected with different seed borne mycoflora which make them unfit for consumption, reduce germination and proper Crop stand in field ultimately reduction in yield. Therefore it is essential to find out certain solution to minimise these losses this topic was designed.
Five varities were tested to detect the seed borne mycoflora by standard blotter paper method, pre-treatment blotter method and agar plate method. It was revealed that six fungal species associated *viz.*, *Fusarium oxysporum, Aspergillus niger, Aspergillus flavus, Alternaria alternata, Curvularia lunata and Cladosporium* spp. *Fusarium oxysporum* found major fungus associate with all sesamum varieties.
Three fungicides and two botanicals were tested against *Fusarium oxysporum* under *in vitro* condition. Among the three fungicides carbendazim + mancozeb and hexaconazole completely inhibited the mycelial growth (100%) followed by tebuconazole (88.82%). Among botanicals neem leaf extract inhibited maximum mycelial growth (77.34%) *Allium sativum* (72.20%). Four bioagents *viz.*, *Trichoderma viride, Trichoderma harzianum, Pseudomonas fluorescens* and *Bacillus subtilis* were tested against *Fusarium oxysporum* by dual culture method. Maximum mycelium growth inhibition observed in *Trichoderma harzianum* (75.85%) followed by *Trichoderma viride* (73.68%) and *Bacillus subtilis* (72.44%).

Keywords: Sesamum seeds, seed mycoflora, *Fusarium oxysporum*, chemicals control, botanical control, bioagents control

Introduction
Sesamum (*Sesamum indicum*) known variously as sesame, til, Gingelly, Simsin, Gergelin etc. belong to family pedaliaceae. It is an important oil yielding crop cultivated in India, Myanmar, Indo-China, China and Japan. Sesamum is regarded as oldest oil yielding plant known to man. Sesamum is called as "Queen of edible oils" in view of the rich 1oil content (40-50%), seed protein (20%), carbohydrates and minerals such as calcium (1%), and phosphorous (0.7%). It is rich source of vitamin E. In India crop is grown in all Seasons *viz.*, kharif, semi-rabi and summer. Uttar Pradesh leads in area and production followed by Rajasthan, Gujrat, Orissa and Karnataka. It has become a popular summer crop in West Bengal and Bihar.
Recently, biological control using bio-agents and Phyto extract has received much attention in both conventional and organic farming to suppress plant disease and to overcome some extent the public concern regarding chemical fungicides (Samnels, 2006) [8]. Fungicides and botanicals inhibits the growth of seed borne mycoflora of sesame *in vitro* (Hosen and Shamsi 2017) [6]. Fungicides have been reported for many years to control plant pathogens and the use of fungicide seed dressing chemicals and bio-agents has become an inevitable method of disease and pest control in Sesame (Anandu *et al.*, 2010) [2].
Mycoflora associated with seeds reduce the seed germination, final stand of crop in the field and overall growth of plant. Infected seeds play an important role in dessimination of pathogens and establishment of disease take place which will lead to increase a cost of cultivation as well as reduce a quality of product, to avoid all these problems and losses it felt necessary to detect and manage a seed borne mycoflora associate with seed.

Materials and Methods
The present investigations were carried out in the plant pathology section of College of Agriculture, Nagpur. The details of materials used and the methodology followed in conducting the experiments are presented here under.

Isolation of *Fusarium oxysporum*

This study was carried out on seeds of 5 cultivars of sesamum including PKV-NT-11, AKT-101, N-8, JLT-408, GT-10 were collected from oilseed research unit Dr. P.D.K.V. Akola. Varieties were analyzed for their association of seed-borne mycoflora by standard blotter paper, Pre treatment and agar plate method. Isolate a *Fusarium oxysporum* associate with seeds of sesamum.

***In vitro* evaluation of fungicides and botanicals against *Fusarium oxysporum* by poisoned food technique**

Three different fungicides and two botanicals were evaluated under *in vitro* against test pathogen by adopting poison food technique. Requisite quantity of each of the fungicides (as per concentration) was added in sterilized melted PDA separately so as to obtain desired concentration. PDA was poured in sterilized petri plate and allow to solidify. Five mm disc of fungus culture was transferred aseptically in the center of petri plate containing the poisoned media with test fungicide. The control plates were kept the culture disc grown in same condition on PDA without fungicides. Treated plates were incubated at room temperature (26±2 °C) for a period of seven days. Colony diameter was recorded in mm and per cent mycelial growth inhibition was calculated as per Vincent's formula based on the average colony diameter. The data was subjected to statistical analysis wherever necessary.

$$I = \frac{C - T}{C} X\ 100$$

Where,
I = Percent growth inhibition
C = Fungal growth in control plate (mm)
T = Fungal growth in treatments (mm)

***In vitro* evaluation of bio agents against *Fusarium oxysporum* by dual culture technique**

PDA was autoclaved and poured into a 90 mm petri plate, where it solidified. Then, on one end of the plate, a 5 mm disc of test species was set, and on the other end, an antagonistic disc and striking bacterial culture. Along with the control, the plates were incubated at room temperature (26+2 ^{0}C) for seven days. The data was subjected to statistical analysis wherever necessary.

$$I = \frac{C - T}{C} X\ 100$$

Where,
I = Percent growth inhibition
C = Fungal growth in control plate (mm)
T = Fungal growth in treatments (mm)

Result and Discussion

Management of *Fusarium oxysporum in vitro*

1. Through poison food technique

Result obtained were presented in Table 1. it was clearly indicated that all the treatments were significantly superior over control in inhibiting the growth of *Fusarium oxysporum.* Among the three fungicides Carbendazim+mancozeb and hexaconazole completely inhibited the mycelial growth upto 100% followed by tebuconazole recording (88.82%). Among the botanicals neem leaf extract inhibited maximum mycelial growth (77.34%) than *Allium sativum* (72.20%). Highest growth of *Fusarium oxysporum* was recorded in control.

In this study it was observed that among all the treatments, carbendazim+mancozeb (T1) and hexaconazole (T3) were best treatment followed by tebuconazole (T2) and neem leaf extract (T4). *Allium sativum* (T5) was less effective among the treatments over a control. Similar result given by Behera (2016) [5].

Table 1: Efficacy of fungicides and botanicals against *Fusarium oxysporum in vitro*

Tr. No.	Treatment detail	Conc. Per cent	Mean colony diameter (mm)	Per cent growth inhibition
1	Carbendazim 12% + Mancozeb 63% WP	0.25	0.00	100
2	Tebuconazole 25% EC	0.1	9.25	88.82
3	Hexaconazole 5% EC	0.1	0.00	100
4	Neem leaf extract	10	18.75	77.34
5	*Allium sativum* (Clove)	10	23.00	72.20
6	Control	--	82.75	--
	SE(m)±	--	0.43	--
	CD (P=0.01)	--	1.79	--

T1: carbendazim + mancozeb, T2: tebuconazole, T3: hexaconazole, T4: Neem leaf extract, T5: *Allium sativum*, T6: Control

Plate 1: Efficacy of fungicides and botanicals against *Fusarium oxysporum in vitro*

2. Dual Culture Technique

Efficacy of *Trichoderma viride*, *Trichoderma harzianum*, *Pseudomonas fluorescens* and *Bacillus subtilis* were tested against *Fusarium oxysporum* by dual culture technique.Result depicted in Table 2, Maximum mycelia growth inhibition of *Fusarium oxysporum* was observed in *Trichoderma harzianum* (75.85%) followed by *Trichoderma viride* (73.68%) *Bacillus subtilis* (72.44%). Minimum mycelial growth inhibition was observed in *Pseudomomas fluorescens* (61.61%).

It was observed that all the treatment were statistically superior over control in reducing the growth of *Fusarium oxysporum*. Minimum growth of *Fusarium oxysporum* was noticed in the treatment of *Trichoderma harzianum* followed by *Trichoderma viride*, *Bacillus subtilis* and *Pseudomonas fluorescens* recording 19.50, 21.25, 22.25 and 31.00 mm respectively. Maximum growth of *Fusarium oxysporum* observed in control 80.75 mm. Similar results were given by Mahmoud and Abdalla (2018) [7], Ahmed and Abdel-Gayed (2017) [5] and, Barhate *et al.*, (2015) [3].

Table 2: Efficacy of bioagents against *Fusarium oxysporum in vitro*

Tr. No.	Treatment detail	Mean colony diameter (mm)	Per cent growth inhibition
1	*Trichoderma viride*	21.25	73.68
2	*Trichoderma harzianum*	19.50	75.85
3	*Pseudomonas fluorescens*	31.00	61.61
4	*Bacillus subtilis*	22.25	72.44
5	Control	80.75	--
	SE(m)±	0.55	--
	CD(P=0.01)	2.36	--

T1: *Trichoderma viride*, T2*: Trichoderma harzianum*
T3*: Pseudomonas fluorescens*, T4*: Bacillus subtilis* T5*: Control*

Plate 2: Efficacy of bioagents against *Fusarium oxysporum in vitro*

Conclusion

Sesamum varities were highly associated with *Fusarium oxysporum*, Maximum inhibition of *Fusarium oxysporum* was recorded in carbendazim + mancozeb and hexaconazole followed by tebuconazole in poison food technique. *Trichoderma harzianum* and *Trichoderma viride* had shown maximum inhibition of *Fusarium oxysporum* by dual culture method.

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