

Original Research Article

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## Management of Storage Diseases of Onion by Using Different Fungicides and Antibiotics

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### A B S T R A C T

Black mold and *Fusarium* base rot of onion is caused by *Aspergillus niger* and *Fusarium oxysporum f.sp. cepae* are the most destructive diseases of onion in storage causing accountable losses of about 80 per cent and more than 50 per cent respectively. The fungicides, evaluated *in vitro* were found fungistatic/antifungal against both pathogen. In *A. niger* the most effective fungicides with significantly highest mycelial growth inhibition were SAFF (94.55 per cent), carbendazim (92.22 per cent) and mancozeb (90.98 per cent). The bioagents viz., *T. viride* (85.61 per cent) and *T. harzianum* (82.19 per cent) were found potential antagonists against *A. niger*. In *Fusarium oxysporum f.sp. cepae* fungicides viz., hexaconazole, SAFF, mancozeb and carbendazim were found most effective with significantly highest mycelial growth inhibition of 91.73 per cent, 91.39 per cent, 90.82 per cent and 88.42 per cent respectively. The bioagents viz., *T. viride* (88.1 per cent) and *T. harzianum* (81.55 per cent) recorded significantly highest mycelial growth inhibition.

#### Keywords

Fungicides, Bioagents, *A. niger* and *Fusarium oxysporum f.sp. cepae* culture

#### Article Info

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### Introduction

Onion (*Allium cepa* L.), is a member of class Liliaceae and family Alliaceae, widely distributed and grown in tropical, sub-tropical and temperate climatic zones (Fritsch and Friesen, 2002). It is biennial crop (Bohanec *et al.*, 2003), and in India it is grown twice (Rabi and Kharif) in a year (NHRDF, 2009). The name “ wild onion” is applied to a number of *Allium* species but *A. cepa* is exclusively

known from cultivation and its wild original form is not known. The onion is most frequently a biennial or a perennial plant, but is usually treated as an annual and harvested in its first growing season.

Onion rightly called as “Queen of kitchen” is one of the oldest known and an important vegetable crop grown in India. Onion is supposed to have its origin in the Middle East Asian Countries and introduced in India from

Palestine. It belongs to family Alliaceae, and genus *Allium* with about 300 species. Onion is grown in three season i.e. Kharif, Rabi, and summer. In Kharif season, during May-June seeds are sown and transplanted during July-Aug months and onion become ready for harvesting during Oct-Nov month.

The post – harvest losses, viz. sprouting, rotting, and physiological loss in weight pose a great problem. It is reported that annual storage losses were over 40 per cent (Bhagachandani *et al.*, 1980) and between 40-60 per cent (Maini *et al.*, 1984) in India. The *rabi* crop produce having more ability in storage and used for domestic, export and seed bulbs purposes from June to November.

Losses of onion during storage are considerable mainly due to sprouting and contamination by several microorganisms. Nearly 40% of the production is lost during post-harvest handling and sprouting. Microbial spoilage alone contributes approximately 15-20% of the total loss (Pantastico and Bantista, 1976; Bhagchandani *et al.*, 1980). Earlier, (Quadri *et al.*, 1982) stated that the spoilage caused by *Aspergillus niger* was as high as 80%.

## Materials and Methods

### Experimental site

All the experiments (*In vitro*) were conducted at the Department of Plant Pathology, College of Agriculture, Latur.

### Collection of disease samples

Onion bulbs showing symptoms of rot, black, brown and discolouration were randomly collected in the bags from the various markets and fields in the Osmanabad and Latur district. These collected bulbs were brought to the Plant Pathology, Laboratory, College of

Agriculture, Latur and subjected for further studies.

### Culture media

Potato dextrose agar (PDA), the common laboratory culture medium was used as basal medium for isolation, purification, multiplication and maintenance of the pure culture of diseases.

### Chemicals

Standard chemicals, reagents, fungicides, culture media etc. required for the experimentation were obtained from the department of Plant Pathology, college of Agriculture, Latur.

### Glass-wares

The common glass-wares (Borosil and corning make) viz., Petri dishes, test tubes, conical flasks, volumetric flasks, measuring cylinder, glass rods, beakers, funnel, pipettes etc. were obtained from the Department of Plant Pathology, College of Agriculture, Latur.

### Equipments

The laboratory equipments viz., Autoclave, Hot air oven, Laminar-airflow Cabinet, BOD incubator, Refrigerator, Binocular Research Microscope, Electronic balance, pH meter, Mixer-cum-grinder etc. available at the Department of Plant Pathology, College of Agriculture, Latur were utilized, as and when required.

### Miscellaneous

Inoculation needle, forceps, blotter papers, paper bags, polythene bags, spirit lamp, mercuric chloride, labels, scales, etc. available at the Department of Plant Pathology were used.

## Fungicides

The following ten systemic and non-systemic fungicides were used for the *in vitro* experiments conducted during present studies.

## Antibiotics

Streptocycline and plantomycin these two antibiotics are used for the *in vitro* study.

## Isolation

Naturally infected onion bulbs showing typical symptoms of diseases were collected from the markets and brought to the laboratory. The infected bulbs were stripped of their outer dry scales and small pieces of the infected onion bulbs were removed with a sterile knife and surface sterilized with 0.1% aqueous solution of mercuric chloride ( $HgCl_2$ ) for two minutes and then washed by giving three changes with sterile distilled water to remove traces of mercuric chloride. The surface sterilized diseased scales were then inoculated on the solidified and cooled PDA (Potato Dextrose Agar) medium in 90 mm-diameter sterilized Petri plates under aseptic conditions of Laminar-air-flow cabinet (make: ACS, Bangalore). Inoculated plates were then incubated in BOD incubator (make: MAC, Delhi) at  $27 \pm 1^\circ C$  temperature. After three to four days of incubation, the well-developed mycelial growth, free from any contaminant was obtained. The developing fungal colonies were sub-cultured on fresh PDA plates to obtain pure culture.

## Identification

The fungal isolates were identified, based on their cultural and morphological characteristics including shapes of spores or conidia (Barnett and Hunter, 1998). The frequency of isolation of each fungal species was recorded.

## Disease management Strategies

### *In vitro* evaluation of fungicides

Efficacy of ten fungicides viz., Carbendazim (50 WP), Mancozeb (75 WP), SAAF (75WP), Benomyl (50 WP), Hexaconazole (5 EC), Tebuconazole (25.9 EC), Copper oxychloride (50 WP), Difenconazole (25 EC), Topsin (70 WP), Captan (50 WP), and two antibiotics viz. Streptocyclin and Plantomycin was tested *in vitro* against *Aspergillus niger* and *Fusarium oxysporum f.sp.cepae* applying poisoned food technique (Nene and Thapliyal, 1984). The requisite quantity of each fungicide and antibiotic based on active ingredient was calculated and mixed thoroughly with autoclaved and cooled ( $40^\circ C$ ) Potato dextrose agar medium (PDA) in conical flasks to obtain desired concentrations. Untreated PDA medium without fungicide served as control. Fungicide amended PDA medium was then poured in Petri plates (90 mm dia.).

After solidification of the medium, all the plates were inoculated aseptically with 5 mm culture disc of the test fungus obtained from a week old culture of *A. niger* and *Fusarium oxysporum f.sp.cepae*. The disc was placed on PDA in the centre of the Petri plate and plates were incubated at  $27 \pm 1^\circ C$  in inverted position. Suitable numbers of replication were maintained for each treatment. When medium in the untreated control plates was fully covered with mycelial growth of the test fungus, radial mycelial growth was measured in all the treatment plates. The diameter of the colony was measured in two directions and average was recorded. Per cent inhibition of mycelial growth in treated plates was calculated by applying the formula given by Vincent (1947).

$$\text{Per cent inhibition (I)} = \frac{\text{C} - \text{T}}{\text{C}} \times 100$$

Where,

C = Growth (mm) of test fungus in control plate,

T = Growth (mm) of test fungus in treated plate.

### Experimental details

Design: C.R.D.

Replication: Three

Treatment: 13

T1: Carbendazim (1000ppm)

T2: Mancozeb (2500 ppm)

T3: Benomyl (1000 ppm)

T4: Copper oxychloride (3000 ppm)

T5: Topsin (1000 ppm)

T6: Captan (2500 ppm)

T7: Hexaconazole (1000ppm)

T8: Tebuconacozole (1000 ppm)

T9: Difenconazole (1000 ppm)

T10: SAAF (2500 ppm)

T11: Streptocycline (500ppm)

T12: Plantomycin (500ppm)

T13: Control

Observations on radial mycelia growth/ colony diameter were recorded at 24 hrs interval and continued till the untreated control plates were fully covered with mycelial growth of the test fungus.

Per cent mycelial growth inhibition of the test pathogen over untreated control was calculated by applying the formula given by Vincent (1947).

### Results and Discussion

Present studies on the post-harvest and storage diseases of onion were undertaken during Rabi-2014 on the aspects of *in vitro* evaluation of fungicides. The results obtained on these aspects are presented in the following paragraphs.

### *In vitro* evaluation of fungicides and antibiotics against *A. niger*

A total of ten fungicides viz., Carbendazim (50 WP), Mancozeb (75 WP), SAAF (75WP), Benomyl (50 WP), Hexaconazole (5 EC), Tebuconazole (25.9 EC), Copper oxychloride (50 WP), Difenconazole (25 EC), Topsin (70 WP), Captan (50 WP) and also two antibiotics Streptocycline (0.5%) and Plantomycin (0.5%) were evaluated *in vitro* against *Aspergillus niger* applying Poisoned Food Technique (Nene and Thapliyal, 1993), and using Potato dextrose agar (PDA) as basal medium. Effect of these fungicides and antibiotics on radial mycelia growth and inhibition of test pathogen were recorded. All the treatments were replicated thrice and a suitable untreated control (without fungicides) was also maintained.

### Radial mycelial growth

Result revealed that all fungicides and antibiotics tested recorded a wide range of radial mycelial growth (colony diameter) of the test pathogens.

In *Aspergillus niger* the radial mycelial growth recorded with the fungicides tested ranged from 4.90 mm (mancozeb + carbendazim) to 22.56mm (difenconazole). However, highest mean radial mycelial growth was recorded with difenconazole (22.56 mm), which was followed by tebuconazole (13.52 mm), hexaconazole (12.89mm), captan (9.22), topsin (9.00), copper oxychloride (8.89mm), mancozeb (8.11mm), benomyl (8.73mm) and carbendazim (7.00). Significantly least mean radial mycelial growth was recorded with mancozeb+ carbendazim (4.90mm) compared to maximum radial growth (90.00 mm) in untreated control plates. And in antibiotics all of two gives maximum radial growth i.e. Streptocycline (72.99mm) and Plantomycin (80.00mm).

### Systemic fungicides

Sr. No.	Common name	Trade Name	Active Ingredient	Manufacture
1.	Carbendazim	Bavistin	50 WP	Sulphur Mills
2.	Benomyl	Benlate	50 WP	Corom
3.	Topsin	Topsin-M	70 WP	J.U.pest
4.	Tebuconazole	Folicure	25.9 EC	Bayer
5.	Difenaconazole	Score	25 EC	Syngenta
6.	Hexaconazole	Contaf	5 EC	Devidayal limt.

### Combine fungicides

Sr. No.	Common name	Trade Name	Active Ingredient	Manufacture
1	Mancozeb 63 % + Carbendazim 12 %	SAFF	75 WP	Devidayal limited

### Non systemic fungicides

Sr. No.	Common name	Trade Name	Active Ingredient	Manufacture
1	Mancozeb	Dithane M-45	75 WP	INDOFI
2	Captan	Captaf	50 WP	INDOFI
3	COC	Blitox,Blue coper	50 WP	Syngenta

### Antibiotics

Sr. No.	Name	Manufacture
1	Streptocycline	Hindusta Antibiotic Limited
2	Plantomycin	Arise Agro Limited

### *In vitro* effect of different fungicides on mycelial growth and inhibition of *A. niger*

Tr. No	Treatments	Conc. (%)	Colony diameter *(mm)	Per cent inhibition
1.	Carbendazim	0.1	7.00	92.22 (67.25)*
2.	Mancozeb	0.25	8.11	90.98 (65.49)
3.	Benomyl	0.1	8.73	90.29 (64.56)
4.	Copper oxychloride	0.3	8.89	90.12 (64.31)
5.	Topsin	0.1	9.00	89.99 (64.17)
6.	Captan	0.25	9.22	89.75 (64.01)
7.	Hexaconazole	0.1	12.89	85.67 (59.42)
8.	Tebuconazole	0.1	13.52	84.97 (58.30)
9.	Difenconazole	0.1	22.56	74.93 (48.55)
10.	SAFF	0.25	4.90	94.55 (71.20)
11.	Streptocycline	0.5	72.99	18.89 (10.89)
12.	Plantomycin	0.5	80.00	11.11 (6.37)
13	Control		90.00	00.00
	S.E. ±		1.28	1.70
	C.D. (P= 0.01)		3.74	4.98

\*: Figures in parenthesis are sin transformed values Mean of three replications

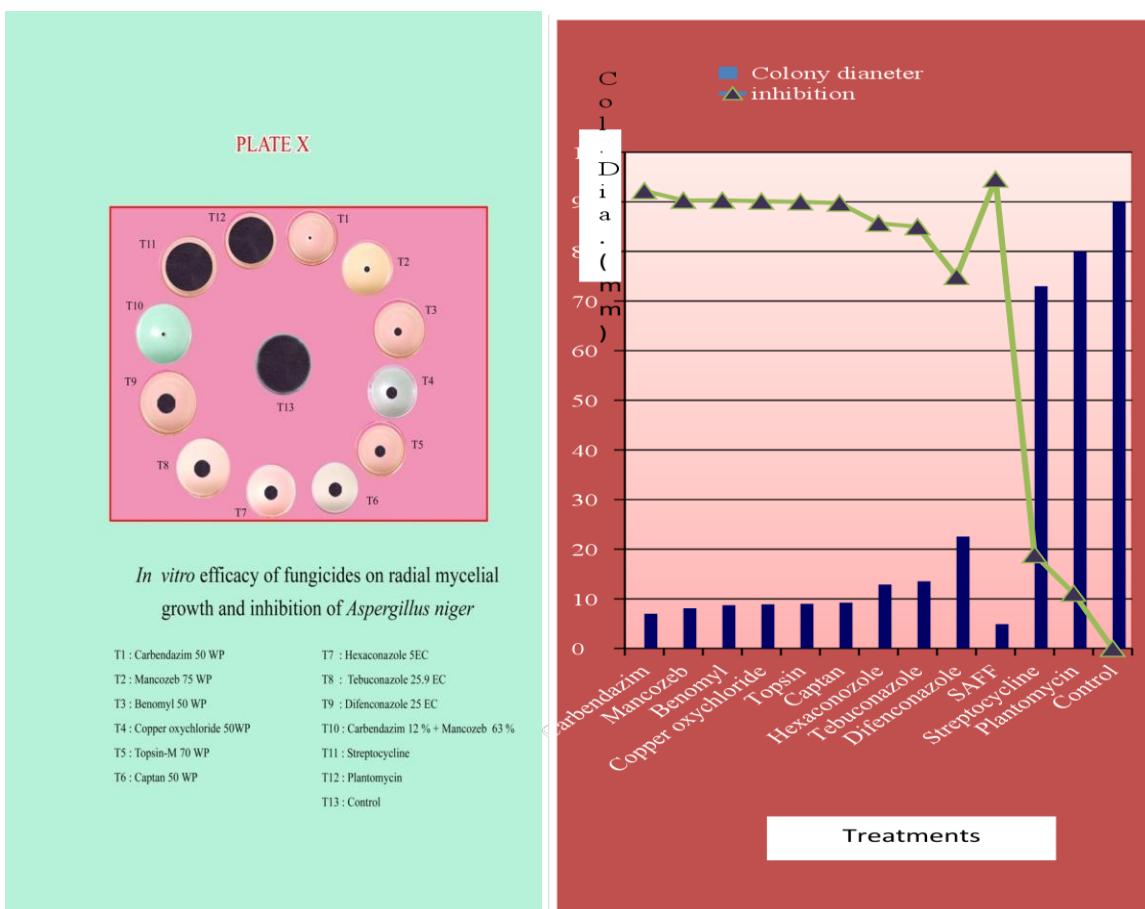
***In vitro* effect of different fungicides on growth and inhibition of *Fusarium oxysporum f. sp. cepae***

Tr. No	Treatments	Conc. (%)	Colony diameter *(mm)	Per cent inhibition
1.	Carbendazim	0.1	10.42	88.42 (62.30)*
2.	Mancozeb	0.25	8.26	90.82 (65.36)
3.	Benomyl	0.1	15.54	82.73 (55.83)
4.	Copper oxychloride	0.3	9.99	88.9 (62.75)
5.	Topsin	0.1	15.44	82.84 (55.99)
6.	Captan	0.25	16.43	81.74 (54.84)
7.	Hexaconazole	0.1	7.44	91.73 (66.06)
8.	Tebuconazole	0.1	20.23	77.52 (50.83)
9.	Difenconazole	0.1	13.19	85.34 (58.65)
10.	SAFF	0.25	7.74	91.39 (66.56)
11.	Streptocycline	0.5	78.84	12.39 (7.11)
12.	Plantomycin	0.5	78.94	12.28 (7.05)
13	Control		90.00	00.00
	S.E.±		<b>0.95</b>	<b>1.21</b>
	C.D.(P= 0.01)		<b>2.76</b>	<b>3.153</b>

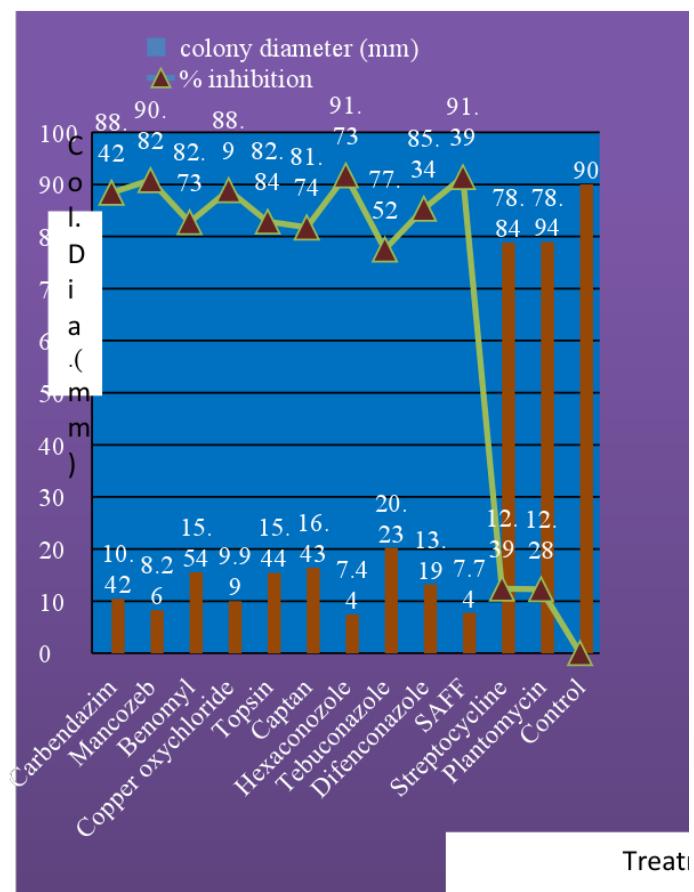
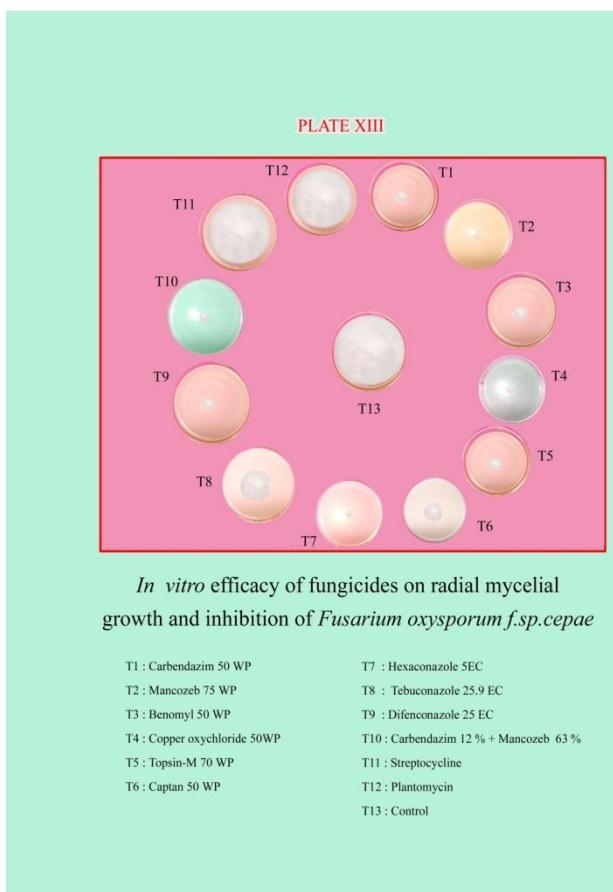
\*: Figures in parenthesis are sin transformed values

Mean of three replications

***In vitro* efficacy of the fungicides on mycelial growth and inhibition of *Aspergillus niger***



### ***In vitro* efficacy of the fungicides on mycelial growth and inhibition of *Fusarium oxysporum f.sp. cepae***



### **Mycelial growth inhibition**

Results revealed that all the fungicides and antibiotics tested significantly inhibited mycelial growth of the test pathogen over untreated control (00.00%).

In *Aspergillus niger* mycelial growth inhibition percentage recorded with all the fungicides tested ranged from 74.93% (Difenconazole) to 94.55% (Mancozeb+Carbendazim). However, mancozeb+carbendazim were found most fungistatic which recorded significantly highest mycelial growth inhibition of 94.55 per cent over untreated control (00.00%). The second and third best fungicides found were carbendazim (92.22 %) and mancozeb

(90.98%). This was followed by benomyl (90.29%), copper oxychloride (90.12%), topsin (89.99%), captan (89.75%), hexaconazole (85.67%), tebuconazole (84.97%) and difenconazole (74.93%). And in antibiotics two gave minimum mycelial growth inhibition i.e. plantomycin (11.11%) and streptocycline (18.89%).

Thus the all the fungicides and antibiotics tested were found fungistatic/antifungal against *A. niger* and significantly inhibited its mycelial growth, over untreated control (00.00).

However, fungicides found most effective in the order of merit were SAFF, carbendazim, mancozeb, benomyl, copper oxychloride,

topsin, captan, tebuconazole, hexaconazole and difenconazole. These results are similar to the findings of several workers (Suryawanshi and Deokar, 2001; Raju and Naik, 2006; Wani and Taskeen-Nisa, 2011 and Nandeesh et al., 2013).

### ***Fusarium oxysporum F.sp. cepae***

#### **In vitro evaluation of fungicides and antibiotics**

A total of ten fungicides viz., Carbendazim (50 WP), Mancozeb (75 WP), SAAF (75WP), Benomyl (50 WP), Hexaconazole (5 EC), Tebuconazole (25.9 EC), Copper oxychloride (50 WP), Difenconazole (25 EC), Topsin (70 WP), Captan (50 WP) and also two antibiotics Streptocycline (0.5%) and Plantomycin (0.5%) were evaluated *in vitro* against *Fusarium oxysporum f.sp.cepae*. applying Poisoned Food Technique (Nene and Thapliyal, 1993), and using Potato dextrose agar (PDA) as basal medium. Effect of these fungicides and antibiotics on radial mycelial growth and inhibition of test pathogen were recorded. All the treatments were replicated thrice and a suitable untreated control (without fungicides) was also maintained.

#### **Radial mycelial growth**

Result revealed that all fungicides and antibiotics tested recorded a wide range of radial mycelial growth (colony diameter) of the test pathogens.

In *Fusarium oxysporum f.sp. cepae* the radial mycelial growth recorded with the fungicides tested ranged from 7.44mm (hexaconazole) to 20.23mm (tebuconazole). However, highest mean radial mycelial growth was recorded with tebuconazole (20.23mm), which was followed by captan (16.43 mm), benomyl (15.54), topsin (15.44mm), difenconazole (13.19mm), carbendazim (10.42 mm), copper

oxychloride (9.99mm), mancozeb (8.26 mm) and mancozeb+ carbendazim (7.74 mm). Significantly least mean radial mycelial growth was recorded with hexaconazole (7.44mm) compared to maximum mean radial growth (90.00 mm) in untreated control plates.

#### **Mycelial growth inhibition**

Results revealed that all the fungicides and antibiotics tested significantly inhibited mycelial growth of the test pathogen over untreated control (00.00%). In *Fusarium oxysporum f.sp.cepae* mycelial growth inhibition percentage recorded with all the fungicides tested ranged from 77.52 (tebuconazole) to 91.73 (hexaconazole). However, hexaconazole found most fungistatic which recorded significantly highest mycelial growth inhibition of 91.73 per cent over untreated control (00.00). The second and third best fungicides found were mancozeb+carbendazim (91.39 %) and mancozeb (90.82%). This was followed copper oxychloride (88.90%), carbendazim (88.42%), difenocoazole (85.34%), topsin (82.84%), benomyl (82.73%), captan (81.74%) and tebuconazole (77.52%) and in antibiotics Streptocycline (12.39%) and Plantomycin (12.28%).

Thus all the fungicides and antibiotics tested were found fungistatic/antifungal against *Fusarium oxysporum f.sp.cepae* and significantly inhibited its mycelial growth over untreated control (00.00). However, fungicides found most effective in the order of merit were hexaconazole, SAFF, mancozeb, copper oxychloride, carbendazim, difenconazole, topsin, benomyl, captan and tebuconazole. These results are similar to the findings of several workers (Singh et al., 1997; Patel et al., 2005; Banyal et al., 2008; Barhate et al., 2011; Taskeen-Un-Nisa et al., 2011).

All the 10 fungicides and 2 antibiotics tested *in vitro* were found fungistatic against *Aspergillus niger* and significantly inhibited its mycelial growth, over untreated control. However, mancozeb+carbendazim were found most fungistatic which recorded significantly highest mycelial growth inhibition of 94.55 per cent over untreated control (00.00%). The second and third best fungicides found were carbendazim (92.22 %) and mancozeb (90.98%). This was followed by benomyl (90.29%), copper oxychloride (90.12%), topsin (89.99%), captan (89.75%), hexaconazole (85.67%), tebuconazole (84.97%) and difenconazole (74.93%). In antibiotics two gave minimum mycelial growth inhibition i.e. plantomycin (11.11%) and streptocycline (18.89%).

In *Fusarium oxysporum f.sp. cepae* all the test fungicides significantly inhibited mycelial growth over untreated control. However, hexaconazole found most fungistatic which recorded significantly highest mycelial growth inhibition of 91.73 per cent over untreated control (00.00). The second and third best fungicides found were mancozeb+carbendazim (91.39 %) and mancozeb (90.82%). This was followed by copper oxychloride (88.90%), carbendazim (88.42%), difenconazole (85.34%), topsin (82.84%), benomyl (82.73%), captan (81.74%) and tebuconazole (77.52%). And in antibiotics Streptocycline (12.39%) and Plantomycin (12.28%) was least effective.

Thus from the results obtained on various aspects during investigation on postharvest and storage diseases of onion, following conclusions are being drawn.

All the test fungicides, evaluated *in vitro* found fungistatic / antifungal to both the pathogens (*A.niger* and *Fusarium oxysporum f.sp. cepae*). However, fungicides viz., SAFF, carbendazim, mancozeb copper oxychloride

and benomyl were most effective against *A.niger*, whereas the fungicide viz., hexaconazole, SAFF, mancozeb and carbendazim were most effective against *Fusarium oxysporum f.sp. cepae*.

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