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Evaluations of effective molecules against the pathogen complex causing rhizome rot of ginger

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Abstract

Rhizome rot disease of ginger has become a major threat to all ginger growing areas causing huge economical losses. The complex nature of the pathogens involved in the disease makes the management strategies cumbersome. There is still a need to develop integrated disease management strategies by targeting all the pathogens involved in the complex disease development. In the present study, we have isolated five pathogens involved in the disease development such as Pythium, Fusarium, Sclerotium rolfisii, Ralstonea solanacearum and Meloidogyne incognita. A primary in vitro evaluation of seven different antibiotics such as Streptocycline, K-cycline, Plantomycine, Validamycin, Bactinash, Bactinashak as well as nine nonsystemic, eleven systemic, and twelve combi products were evaluated under lab conditions against the pathogen complex. Our results revealed that among the different antibiotics Streptocycline followed K-cycline, Bactinash, as well as COC, found most effective against Ralstonea solanacearum. Among the different non-systemic chemicals evaluated against the pathogen complex COC and propineb found effective against Pythium, mancozeb, captan, kavach were effective against Fusarium and all the non-systemic fungicides showed high inhibition against Sclerotium rolfisii. Systemic fungicides such as carbendazim, Tricyclazole, Tebuconazole, Alliete were found more effective against Pythium. Tricyclazole, Tebuconazole found effective against Fusarium, and fungicides such as Hexaconazole, Propiconazole, Tricyclazole, Myclobutanil, Azoxystrobin, Tebuconazole, Diniconazole were found effective against Sclerotium rolfsii. As compared to systemic and non-systemic fungicides, combi products were found more effective against all the pathogens involved in the disease complex.

Keywords: Anti fungal, anti bacterial, percent inhibition

Introduction

Ginger is one of the earliest known oriental spices cultivated in India for both fresh vegetables and as a dried spice. It is used as a condiment, flavoring agent, in the preparation of nonalcoholic beverages and also known to have numerous medicinal properties. Ginger is cultivated in most of the states in India. However, states namely Karnataka, Kerala, Orissa, Sikkim, Assam, Meghalaya, Arunachal Pradesh, and Gujarat are the major ginger growing states in India.

The crop is affected by a variety of diseases like soft rot or rhizome rot, leaf spot, and bacterial wilt diseases. Among the major constraints of ginger production, rhizome rot is very important because of severe crop losses. It occurs in several parts of India wherever the crop is grown. The term rhizome rot is commonly used for all the diseases affecting the rhizome irrespective of pathogens involved since the ultimate result is the partial or total loss of rhizome.

This particular disease is caused by the interaction of several plant pathogenic agents such as fungi, Bacteria, and Nematodes. The main pathogens associated with this disease include *Fusarium* spp, *Pythium* spp, *Sclerotium rolfsii*, *Rhizoctonia solani*, *Pseudomonas spp*,

The infection starts at the collar region of the pseudostem and progresses upwards as well as downwards. The affected pseudo stem becomes water-soaked and the rotting spreads to the rhizome resulting in soft rot. Foliar symptoms appear as light yellowing of the tips of lower leaves which gradually spreads to the leaf blades. In the early stages, the middle portion of the leaves remains green while the margins become yellow. The yellowing spreads to all leaves of the plant from the lower region upwards and is followed by drooping, withering, and drying of pseudostems. Infected rhizomes can be pulled out easily appear black in color and emit foul smell (Dohroo, 2015) ^[8].

Several cultivars of ginger are grown in India and they are generally named after the localities where they are grown. However, none of the varieties are completely resistant to the pathogen complex (Pattnaik *et al.* 2015) ^[13]. Rhizome rot disease has now become a major threat to all ginger growing areas causing huge economical losses. There are no curative effective methods for the management of rhizome rot and also preventive options are cumbersome and are not fully encouraging with any chemicals and biological for the effective disease management schedule.

Materials and methods

Evaluation of antibiotics, bioagents and fungicides under *In vitro* conditions against pathogens

Antibiotics, Bio agents, fungicides were evaluated at different concentrations to test the efficacy under the laboratory condition. Antibiotics were evaluated at 100 ppm, 200 ppm, 300 ppm, 400 ppm, and 500 ppm. The systemic fungicides were evaluated at the concentration of 500 ppm, 1000 ppm, 1500, and 2000 ppm. The contact and combi products were evaluated at a concentration of 1500, 2000 ppm, 2500 ppm. The fungicides were evaluated using potato dextrose agar (PDA) as the basal medium and by following the poison food

technique (PFT). Antibiotics were evaluated by the agar well diffusion method.

Poison food technique

The requisite quantity of fungicides was weighed by using the formula and mixed properly with the autoclaved and cooled (40-45 °C) PDA in conical flasks to obtain desired concentrations. The 15-18 ml PDA amended with the fungicides was poured into 90 mm sterilized Petri plates under aseptic condition and allowed for solidification under room temperature. On solidification of PDA each treatment plate was inoculated or seeded in the center with the 5 mm mycelial disc obtained from a one-week old pure culture of fungus under aseptic condition. Each treatment replicated thrice with respective concentrations. Petri plates containing PDA without any fungicides were inoculated with 5 mm disc of the test pathogen and maintained as the suitable untreated control. All the inoculated and control Petri plates were incubated at $28 \pm 2^{\circ}$ C in incubator till the mycelial growth of the test pathogen in control covers the entire Petri plate. The antibiotics and fungicides used for In vitro studies were listed in table 1, 2.

CL NL	Energy (1) and	T
Sl. No.	Fungicides Non-systemic Fugicides	Trade name
1	Copper oxychloride 50 % WP	Blitox
1 2	Mancozeb 75 % WP	Indofil M-45
3	Captan 50 % WP	Captaf
4	Propineb 70% WP	Antracol
5	Zineb 75% WP	Indofil Z- 78
6	Chlorothalonil 78.12 % WP	Kavach
7	Krezoxim methyl 44.3 % SC	Ergon
8	Copper hydroxide 53.8% WP	Kocide
	Systemic Fugicides	
9	Carbendazim 50 % WP	Bavistin
10	Hexaconazole 5 % EC	Contaf
11	Propiconazole 25 % EC	Tilt
12	Tricyclazole 75 % WP	Baan
13	Myclobutanil 10% WP	Myclowin
14	Azoxystrobin 23.1 % w/w SC	Amistar
15	Tebuconazole 25.9 % EC	Folicure
16	Dimethomorph 50% WP	Acrobat
17	Difenconazole 25 % EC	Score
18	Fosetyl 80% WP	Alliette
19	Metalaxyl 35% WS	Superior
•	Combi products	· •
20	Carbendazim 12 % + Mancozeb 63 %	SAAF
21	Metalaxyl 4% + mancozeb 64 %	Ridomyl Gold
22	Cymoxanil 8% +Mancozeb 64%	Curzate M8
23	Tebucoazole 50 % + Trifloxystrobin 25 %	Nativo
24	Tebuconazole 50% +Trifloxystrobin 25%	Vitavax
25	Iprovalicarb 5.5% + Propineb 61.25%	Melody
26	Tricyclazole 45% + Hexaconazole10%	Impression
27	Hexaconazole 5%+ Captan70%	Taqat
28	Tricyclazole 18%+ mancozeb 62%	Merger
29	Fenamidone 10%+Mancozeb 50%	Sectin
30	Metiram 55% + Pyraclostrobin 5%	Cabrio top
31	Fluxapyroxad 250 g/l + Pyraclostrobin 250 g/l	Merivon

Table 1: List of fungicides used for In vitro studies

The observations on radial mycelial growth were recorded in each treatment and replications and mean colony diameter and percent inhibition of the test pathogen was calculated by applying the formula given by Vincent (1947).

Where,

C= growth of the test fungus in untreated control plates T= growth of the test fungus in treated plates

Per cent inhibition=
$$\frac{C-T}{C} \ge 100$$

Sl. No.	Antibiotic	Trade name
1	Streptomycin sulphate 90% + tetracycline 10 %	Streptocycline
2	Streptomycin sulphate 90% + tetracycline 10 %	K-cycline
3	Streptomycin sulphate 90% + tetracycline 10 %	Plantomycin
4	Validamycin 3%	Validamycin
5	2 Bromo 2 nitro- propane-1,3 diol 95%	Bactinash
6	2 Bromo 2 nitro- propane-1,3 diol 95%	Bactinashak
7	Copper oxy chloride 50 % WP	Blitox
8	Copper hydroxide 53.8 %	Kocide

Table 2: List of antibiotics used for In vitro studies

Results and Discussion

Evaluation of antibiotics against Ralstonea solanacearum

In vitro evaluation of available antibiotics and fungicides revealed that among the different antibiotics screened against the bacteria, Streptocycline found more effective with inhibition of 22mm, at 100 ppm, 24.33 mm at 200 ppm, 25.66 mm at 300 ppm, 28.00 mm at 400 ppm, and 30.66 mm at 500 ppm concentration Table 3. On average Streptocycline

showed 26.13 mm inhibition as compared with other antibiotics. Validamycin was found least effective in all the concentrations. Effective antibiotics are also screened as individuals as well as in combination with an anti-bacterial compound such as COC and Kocide. However, under *In vitro* conditions, a combination of antibiotics with COC and Kocide does not produce significant inhibition against the bacteria.

Table 3: In vitro evaluation of antibiotics against Ralstonea solanacearum

Tuestment no	Antibiotics	In	hibition zon	e (mm)* Co	ncentration	at	A
Treatment no	Antibiotics	100 ppm	200 ppm	300 ppm	400 ppm	500 ppm	Average
T_1	Streptocycline	22	24.33	25.66	28.00	30.66	26.13
T_2	K-cycline	17.86	21.33	23.00	24.33	25.66	22.44
T3	Plantomycine	0	11.66	12.66	15.33	17.00	11.33
T_4	Validamycin	0	0	0	0	0	0
T5	Bactinash	16.33	20.33	21.33	23	25.33	21.26
T ₆	Bactinashak	8.66	12.33	14.00	15.667	19.33	14.00
		1000 ppm	1500 ppm	2000 ppm	2500 ppm	3000 ppm	
T 7	Kocide	15.66	16.33	18.66	18.00	19.66	17.66
T_8	COC	19.33	20.66	22.33	21.33	22.66	21.26
T9	Streptocycline+COC	17.66	18.00	21.33	22.66	23.66	20.66
T ₁₀	Streptocycline+Kocide	11.66	14.66	18.33	18.33	19.66	16.53
T11	K-cycline+COC	14.66	16.33	19.33	22.00	22.66	19.00
T ₁₂	K-cycline+Kocide	15.667	16.667	17.667	21.667	22.00	18.73
T ₁₃	Bactinash+COC	14.667	16.00	18.00	18.333	20.333	17.46
T ₁₄	Bactinashak+ Kocide	11.667	12.00	15.00	15.00	19.66	14.66
T ₁₅	Control	0	0	0	0	0	0
	SE(m)	0.44	0.45	0.53	0.53	0.43	
	C.D.	1.29	1.32	1.54	1.54	1.27	

Evaluation of fungicides against the pathogen complex *In vitro* evaluation of non-systemic fungicides against the pathogen complex

Eight different non-systemic fungicides were evaluated against *Pythium, Fusarium, Sclerotium rolfisii* at 1500 ppm,

2000ppm, and 2500 ppm concentrations. Among the different nonsystemic fungicides, Copper oxychloride and Propineb found most effective against *Pythium* and percent inhibition of the pathogen has been increased when the concentration of the fungicides increased.

 Table 4: Among the different nonsystemic fungicides, Copper oxychloride and Propineb found most effective against *Pythium* and percent inhibition of the pathogen has been increased when the concentration of the fungicides increased

SI.	Fungicides	Perce	nt myce inhibiti		nibition ainst <i>Py</i> i		ercent	Per	cent myo inhibi		hibition inst <i>Fus</i>		ercent	Percent mycelial inhibition and percent inhibition against <i>Sclerotium</i>							
No	Fungiciaes	1500 ppm	PI	2000 ppm	PI	2500 ppm	PI	1500 ppm	PI	2000 ppm	PI	2500 ppm	PI	1500 ppm	PI	2000 ррт	PI	2500 ppm	PI		
1	Copper oxychloride	21.33	76.30 (60.84)	11.67	87.04 (68.87)	0.00	100 (90)	21.33	68.617 (55.91)	12.67	81.37 (64.41)	9.33	86.26 (68.22)	21.67	75.93 (60.59)	0	100 (90)	0	100 (90)		
2	Mancozeb	79.67	11.48 (19.79)	61.33	31.85 (34.34)	50.33	44.07 (41.57)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0	100 (90)	0	100 (90)		
3	Captan	41.67	53.70 (47.10)	40.33	55.18 (47.95)	39.00	56.67 (48.81)	21.00	69.107 (56.21)	17.67	74.00 (59.32)	0.00	100 (90)	0.00	100 (90)	0	100 (90)	0	100 (90)		
4	Propineb	47.67	47.04 (43.28)	28.67	68.15 (55.62)	0.00	100 (90)	5.33	92.15 (73.72)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0	100 (90)	0	100 (90)		
5	Zineb	90.00	0 (0)	87.33	2.96 (9.86)	57.33	36.3 (37.03)	40.67	40.19 (39.33)	39.33	42.14 (40.46)	37.33	45.08 (42.16)	0.00	100 (90)	0	100 (90)	0	100 (90)		
6	Kavach	54.67	39.26 (38.78)	52.33	41.85 (40.29)	31.67	64.813 (53.59)	24.67	63.72 (52.94)	20.67	69.603 (56.52)	0.00	100 (90)	0.00	100 (90)	0	100 (90)	0	100 (90)		
7	Kresoxim methyl	35.00	61.113 (51.42)	30.00	66.67 (54.71)	23.33	74.073 (59.39)	34.33	49.5 (44.69)	28.33	58.32 (49.77)	27.33	59.79 (50.63)	17.33	80.74 (63.94)	0	100 (90)	0	100 (90)		
8	Copper	89.00	1.11	86.67	3.7	85.00	5.56	61.00	10.29	54.33	20.08	44.67	34.30	90.00	0				0		

	hydroxide		(6.04)		(11.06)		(13.63)		(18.70)		(26.60)		(35.83)			90	0	90	(90)
9	Control	90.00	0 (0)	90.00	0 0	90.00	0 0	68.00	0	68.00	0	0	0	90	0	90	0	90	0 (90)
	SE(m)	1.0)83	0.	327	0.	699	0.	543	0.	63	0.	487						
	C.D at 0.5 %	3.2	.43	0.	0.978		2.092		626	1.8	888	1.	458						

Sectin

0.00

0.00

0.00

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SI.	E I	Mean p	ercent n	nycelial inhib l	oition and Pythium	-	ent inhib	ition a	gainst	P	ercent n	iycelial inh		nd per	rcent inł	hibition ag	ainst	P	ercent m	ycelial inh		nd per otium	rcent inl	nibition ag	ainst	
No	Fungicides	500 ppm	PI	1000 ppm	PI	1500 ppm	PI	2000 ppm	PI	500 ppm	PI	1000 ppm	PI	1500 ppm	PI	2000 ppn	n PI	500 ppm	PI	1000 ppm	PI	1500 ppm	PI	2000 ppr	n PI	
1	Carbendazim	0.00	100 (90)	0.00	100 (90)	90.00	(90)	0.00	100 (90)	14.67	81.66 (64.62)	13.00	83.75 (66.20)	12.00	85 (67.18)	9.67	87.91 (69.63)	44.67	50.37 (45.19)	40.00	55.55 (48.17)	38.67	57.04 (49.02)	20.33	77.41 (61.59)	
2	Hexaconazole	22.67	74.81 (59.85)	20.33	77.41 (61.59)	75.00	83.33 (65.88)	14.33	84.07 (66.45)	38.67	51.66 (45.93)	24.67	69.16 (56.24)	20.33	74.58 (59.70)	18.67	76.66 (61.09)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	
3	Propiconazole	22.67	74.81 (59.85)	20.33	77.41 (61.59)	72.33	80.37 (63.67)	14.67	83.7 (66.16)	12.67	84.16 (66.52)	12.33	84.58 (56.24)	10.67	86.66 (68.56)	10.00	87.5 (69.26)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	
4	Tricyclazole	23.00	74.44 (59.61)	0.00	100 (90)	90.00	100 (90)	0.00	100 (90)	21.00	73.75 (59.16)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	
5	Myclobutanil	34.33	61.85 (51.83)	24.00	73.33 (58.88)	66.33	73.7 (59.12)	20.00	77.78 (61.85)	30.67	61.66 (51.72)	17.67	77.91 (61.94)	16.67	79.16 (62.82)	15.00	81.25 (64.31)	3.33	96.29 (83.5)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	
6	Azoxystrobin	27.33	69.63 (56.53)	24.67	72.59 (58.40)	66.33	73.7 (59.12)	19.33	78.52 (62.36)	24.33	69.58 (56.53)	20.67	74.16 (59.42)	20.00	75 (59.97)	17.67	77.91 (61.94)	34.00	62.22 (52.05)	26.00	71.11 (57.5)	24.67	72.59	0.00	100 (90)	
7	Tebuconazole	0.00	100 (90)	0.00	100 (90)	90.00	100	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	
8	Dimethomorph	46.33	48.52 (44.13)	39.67	55.93 (48.38)	54.33	60.37 (50.96)	34.33	61.85 (51.83)	62.67	21.66 (27.72)	58.00	27.5 (31.61)	49.00	38.75 (38.48)	48.33	39.58 (38.96)	61.67	31.48 (34.11)	54.33	39.63 (38.99)	47.67	47.04 (43.28)	44.67	50.37 (45.19)	
9	Diniconazole	22.33	75.18 (60.10)	21.00	76.67	72.33	80.37 (63.67)	16.33	81.85 (64.76)	22.67	71.66 (57.81)	20.00	75 (59.97)	17.67	77.91 (61.94)	15.33	80.83	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	0.00	100 (90)	
10	Alliete	60.33	32.96 (35.02)	37.00	58.89 (50.10)	50.00	55.56 (48.17)	25.00	72.22 (58.16)	34.33	57.08 (49.05)	32.33	59.58 (50.50)	30.00	62.5 (52.21)	27.67	65.41 (53.95)	66.67	25.92 (30.58)	64.67	28.15 (32.03)	64.67	28.15 (32.03)	44.67	50.37 (45.19)	
11	Metalaxyl	86.67	3.70 (6.48)	78.00	13.333 (21.28)	40.00	44.44	45.00	50	46.33	42.08 (40.42)	33.33	58.33 (49.77)	32.33	59.58 (50.50)	30.00	62.5 (52.21)	81.00	10 (18.41)	73.33	(25.42)	70.00	22.22 (28.11)	65.67	27.04 (31.31)	
12	Control	90.00	0	90.00	0	90.00		90.00	· /	80.00	· /	80.00	0	80.00	· /	80	0	90.00	0	90.00	0	90.00		90.00	0	
	SE(m)	1.1		0.87			0.30		0.23		.78	0.4			0.3	0.4			.17	0.60			0.62		21	
	C.D at 0.5 %	3.5	1	2.56		().88	(0.70	2	.28	1.22	2	0	.99	1.2	22	3	.45	1.70	6	0	0.21	0.	62	
SI.		Mean P	er cent r	nycelial inhil I	oition an Pythium	d perc	ent inhib	ition a	gainst	Pe	r cent n	ycelial inhi	ibition a Fusa		cent inh	nibition ag	ainst	P	er cent n	nycelial inh	lial inhibition and percent inhibition agains Sclerotium					
No	Fungicides	500 ppm	1	PI	1000 p	opm		1500 ppm	PI	500 ppm	P	I 10	00 ppm		PI	1500 ppm	Ы	500 ppm	PI	100)0 ppm		PI	1500 ppm	PI	
1	SAAF	18.33	79	.63 (63.14)	0	1		0.00	100 (90)	0.00	100 ((90)	0.00	10	0 (90)	0 1	00 (90)	80	100 (9	90)	80	10	0 (90)		100 (90)	
2	Ridomyl Gold	0.00		100 (90)	0	1	< /	0.00	100 (90)	0.00	100 (< /	0.00		0 (90)	0 1	00 (90)	80	100 (9	/	80		0 (90)	80	100 (90)	
3	Curzate M8	31.67	64	.81 (53.59)	0		100 (90)	0.00	100 (90)	20.33	(59.	70)	17.33	(6	8.33 2.23)	0	100 (90)	80	100 (90))	80	(100 (90)	80	100 (90)	
4	Nativo	0.00		100 (90)	0		100 (90)	0.00	100 (90)	0.00	10 (90		0.00		100 (90)	0	100 (90)	80	100 (90)		80		100 (90)	80	100 (90)	
5	Vitavax	0.00		100 (90)	0		100 (90)	0.00	100 (90)	7.67	90.4 (71.9		0.00		100 (90)	0	100 (90)	80	100 (90)		80		100 (90)	80	100 (90)	
6	Melody	25.00		72.22 (58.16)	19		78.88 (62.62)	5.00	94.44 (76.33)	34.67	56. (48.	· · · ·	24.00		70 6.76)	0	100 (90)	80	100 (90)		80		100 (90)	80	100 (90)	
7	Impression	0.00		100 (90)	0		100 (90)	0.00	100 (90)	0.00	10		0.00		100 (90)	0	100 (90)	80	100		80		100 (90)	80	100 (90)	
8	Taqat	18.67		79.26 (62.88)	18		79.99 (63.41)	16.67	81.48 (64.49)	5.67	92. (74.	91	0.00		100 (90)	0	100 (90)	80	100)	80		100 (90)	80	100 (90)	
9	Merger	9.33		89.63 (71.20)	0		100	0.00	100 (90)	5.00	93.' (75.4	75	3.33	9	5.83 8.21)	0	100 (90)	80	100)	80		100 (90)	80	100 (90)	
10	Contin	0.00	1	100	0		100	0.00	100	0.00	10		0.00		100	0	100	80	100	,	80		100	80	100	

Table 5: In vitro evaluation of systemic fungicides against the pathogen complex

0.00

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			(90)		(90)		(90)		(90)		(90)		(90)		(90)		(90)		(90)
11	Cabriotop	0.00	100 (90)	0	100 (90)	0.00	100 (90)	13.67	82.91 (65.56)	0.00	100 (90)	0	100 (90)	80	100 (90)	80	100 (90)	80	100 (90)
12	Merivon	0.00	100 (90)	0	100 (90)	0.00	100 (90)	12.00	85 (67.18)	1.00	98.75 (83.54)	0	100 (90)	80	100 (90)	80	100 (90)	80	100 (90)
13	Control	90.00	0	0	0	90.00	0	80.00	0	80.00	0	80	0	0	0	0	0	0	0
	SE(m)	0.272		0.252		0.272		0.258		0.16	3								
	C.D at 0.5 %	0.794				0	0.794 0).755	0.47	8								

In vitro evaluation of systemic fungicides against the pathogen complex

Eleven different systemic fungicides were evaluated against pathogen complex. Carbendazim, the Tricyclazole, Tebuconazole, and Alliete found effective against Pythium. Carbendazim significantly inhibited the pathogen growth at all concentrations. Tricyclazole, Tebuconazole showed 100 percent inhibition at 1000ppm, 1500 ppm, and 2000 ppm concentrations. Tricyclazole, Tebuconazole also showed 100 percent inhibition against Fusarium from 1000 ppm concentration onwards. In the case of Sclerotium rolfsii, Hexaconazole, Propiconazole, Tricyclazole, Myclobutanil, Azoxystrobin, Tebuconazole, Diniconazole resulted in higher percent inhibition effectively at all the concentrations (Table 5).

In vitro evaluation of combi products against the pathogen complex

In the case of *Pythium*, all the combi products except Melody showed 100 percent inhibition at 1000 ppm concentration onwards. In the case of *Fusarium* combi products such as SAAF, Ridomyl Gold, Nativo, Vitavax, Impression showed 100 percent inhibition at 1000 ppm concentration onwards (Table 6). As compared to nonsystemic and systemic fungicides, combi products were found more effective against all the pathogens involved in the complex.

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