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Bioefficacy of different fungicides against *Plasmopara viticola* and *Erysiphe necator* of grapes

Sagar N, Jamadar MM, Shalini NH, Jagginavar SB and Pattar PS

Abstract
Investigations on *in vitro* bioassay of different fungicides for their bioefficacy against *Plasmopara viticola* (downy mildew) and *Erysiphe necator* (powdery mildew) of grapes was carried out at the Department of Plant Pathology, College of Agriculture, Vijayapur during 2020-22. The study revealed that among the different systemic fungicides across different concentrations maximum inhibition of sporangial germination (zoospore release) (95.00%) and zoospore germination (99.00%) of *P. viticola* was observed in famoxadone 16.6% + cymoxanil 22.1% (Equation Pro). Among the different combi-fungicides; significantly highest inhibition (91.33%) of sporangial germination (zoospore release) was recorded in fenamidone 10% + mancozeb 50% (Sectin) while highest zoospore inhibition (99.66%) was observed in fluopicolide 4.44% + fosetyl-Al 66.67% (Profiler). Similarly, maximum inhibition 99.66 and 98.04 per cent of conidial germination of *E. necator* was noticed in azoxystrobin 11% + tebuconazole 18.3% (Custodia) and tricyclazole 18% + mancozeb 62% (Merger) respectively.

Keywords: Grapes, downy mildew, powdery mildew, fungicides

Introduction
Grape (*Vitis vinifera* L.) is one of the most delicious, refreshing and nourishing sub-tropical fruit and its cultivation is one of the most remunerative farming enterprises in India. It is grown in a variety of soil. The fruit are rich in minerals and vitamins, *viz.* A, B1, B2, C and K. Grape occupies the fifth position amongst fruit crops in India with a production of 2,920.09 thousand metric tonnes from an area of 138.91 thousand hectares. The area under grape is 1.23 per cent of the total area of fruit crops and production is 2.8 per cent of total fruits produced in the country. About 80 per cent of the production comes from Maharashtra (2,286.44 thousand MT) followed by Karnataka (524.20 thousand MT and Tamil Nadu (58.93 thousand MT). The production of grapevine is threatened by biotic (viruses, bacteria, fungi and insects) and abiotic stresses (*i.e.* drought, winter cold).

Downy mildew is a severe disease of grapevine, one of the most cultivated plants in India. *P. viticola* is a bio trophic oomycete that causes downy mildew. This devastating disease occurs worldwide, particularly in regions with warm and wet conditions during the growing season. *P. viticola* mainly infects leaves and clusters of young berries and produces oil spot lesions on the adaxial leaf surface accompanied by massive sporulation on the abaxial surface (Perazzolli *et al.*, 2012) [9].

Powdery mildew (*Erysiphe necator*) is the most widespread and destructive disease of grapevines worldwide. All green tissues of the grapevine are susceptible to powdery mildew infection. The disease appears as a whitish-grey powdery coating on the leaves or fruit caused by fungal mycelium and conidia on the surface of the plant. On leaves, initial symptoms appear as chlorotic spots on the upper leaf surface that soon become whitish lesions. Late in the season, small black round structures (chasmothecia) begin to appear on the white powdery lesions. On shoots, infected areas have the appearance of brown/black diffuse patches; on dormant canes, these patches are reddish brown. Severe leaf infections can cause distortion, drying and premature drop. Infected berries can become covered with the fungus, may turn dark brown, shrivel and split or may not ripen properly (Berkett and Cromwell 2015) [3].

Downy mildew and powdery mildew are the most devastating diseases of grapes around the world especially they cause yield and quality loss in humid regions. Not only table grapes but also most of the wine grapes are damaged by both diseases. Envisaging the problem of fungicide resistance an effort was made to evaluate the bio efficacy of different fungicides against *P. viticola* and *E. necator* under *in vitro* conditions.

Material and Methods

The sensitivity of grape downy mildew (*P. viticola*) and powdery mildew (*E. necator*) to different fungicidal formulations, at different concentrations was assessed under *in vitro* conditions by spore germination technique.

Bio efficacy of different fungicides against *P. viticola* of grapes

The sporangial suspension was prepared separately in sterile distilled water. A drop of spore suspension was mixed with one drop of commercially available fungicides as mentioned in (Table 1) in a cavity slide to achieve the required concentration in each treatment with three replications were maintained.

Slides were then incubated at room temperature (20±1 °C) for 24 hrs. The observation on the sporangial germination (zoospore release) and zoospore germination was recorded 24 hrs after incubation under microscope at 40X magnification. A control with only sterile water with sporangial suspension was maintained. Per cent sporangial and zoospore germination was calculated by using the below mentioned formula.

$$\text{Per cent sporangial germination} = \frac{\text{Total empty sporangia observed}}{\text{Total sporangia observed}} \text{ X } 100$$

$$\text{Per cent zoospore germination} = \frac{\text{Total no. of zoospores germinated}}{\text{Total no. of zoospores observed}} \text{ X } 100$$

The per cent inhibition over the control was calculated by using the formula given by Vincent (1947).

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

Where,

I = Per cent inhibition

C = Germination of sporangia/zoospore in control

T = Germination of sporangia/zoospore in treatment

Table 1: List of different fungicides evaluated *in vitro* for inhibition of sporangial (zoospores release) and zoospore germination of *Plasmopara viticola*

Sl. No	Fungicides	Trade Name	Concentrations (ppm)
A. Non- Systemic			
1.	Mancozeb 75% WP	Dithane M-45	1000, 2000, 3000
B. Systemic			
1.	Azoxystrobin 23% SC	Amistar	250, 500, 1000
2.	Kersoxim methyl 44.3% SC	Ergon	250, 500, 1000
3.	Fosetyl AL 80% WP	Aliette	250, 500, 1000
4.	Metalaxyl 35% WS	Bilaxyl	250, 500, 1000
5.	Dimethomorph 50% WP	Acrobat	250, 500, 1000
C. Combi products			
1.	Metalaxyl 8% + Mancozeb 64% WP	Sanchar	1000, 2000, 3000
2.	Metalaxyl 4% + Mancozeb 64% WP	Ridomil Gold	1000, 2000, 3000
3.	Ametoctradin 27% + Dimethomorph 20.27% SC	Zampro	1000, 2000, 3000
4.	Fenamidone 10% + Mancozeb 50% WG	Sectin	1000, 2000, 3000
5.	Famoxadone 16.6% + Cymoxanil 22.1%	Equation Pro	1000, 2000, 3000
6.	Metiram 55% + Pyraclostrobin 5% WG	Cabrio top	1000, 2000, 3000
7.	Fluopicolide 4.44% + Fosetyl-Al 66.67% WG	Profiler	1000, 2000, 3000

Bio efficacy of different fungicides against *E. necator* of grapes

The conidial suspension was prepared separately in sterile distilled water. A drop of spore suspension was mixed with one drop of commercially available fungicides as mentioned in (Table 2) in a cavity slide to achieve the required concentration in each treatment three replications were maintained. The slides were then incubated at room temperature (25±1 °C) for 24 hrs. The observation on the spore germination was recorded 24 hrs. after incubation under microscope at 40X magnification. A control with only sterile water with conidia was maintained. Per cent conidial germination was calculated by using the below mentioned formula.

$$\text{Per cent conidial germination} = \frac{A}{B} \times 100$$

Where,

A - Number of conidia germinated

B - Number of conidia observed

The results obtained were analyzed statistically using arc sine transformations.

Table 2: List of different fungicides evaluated *in vitro* for inhibition of conidial germination of *Erysiphe necator*

Sl. No.	Fungicides	Trade Name	Concentrations (ppm)
A. Non- Systemic			
1.	Wettable sulphur 80% WP	Sulfex	1000, 2000, 3000
2.	Chlorothalonil 75% WP	Kavach	1000, 2000, 3000
3.	Dinocap 48% EC	Karathane	1000, 2000, 3000
4.	Ziram 27% SC	Cuman L	1000, 2000, 3000
B. Systemic			
1.	Azoxystrobin 23% SC	Amistar	250, 500, 1000
2.	Hexaconazole 5% EC	Contaf	250, 500, 1000
3.	Tebuconazole 25.9% EC	Follicur	250, 500, 1000
4.	Myclobutanil 10% WP	Systhane	250, 500, 1000
5.	Tetraconazole 10% EC	Domark	250, 500, 1000
6.	Difenoconazole 25% EC	Score	250, 500, 1000
C. Combi products			
1.	Trifloxystrobin 25% + Tebuconazole 50% EC	Nativo	1000, 2000, 3000
2.	Azoxystrobin 11% + Tebuconazole 18.3% EC	Custodia	1000, 2000, 3000
3.	Tricyclazole 18% + Mancozeb 62% WP	Merger	1000, 2000, 3000
4.	Zineb 68% + Hexaconazole 4% WP	Avatar	1000, 2000, 3000
5.	Carbendazim 12% + Mancozeb 62% WP	SAAF	1000, 2000, 3000

Statistical analysis

All the experiments were of completely randomized design (CRD). Data were subjected to analyses of variance and treatment means were compared at ($P < 0.01$). The WASP package version 2.0 was used for analysis and further the values angular transformed in order to analyse the significance.

Results

a) Bioefficacy of systemic and combi (S+S) fungicides on inhibition of sporangial germination (zoospore release) and zoospore germination of *P. viticola*

In vitro evaluation of systemic and combi fungicides (Table 3) were conducted with respect to inhibition of sporangial and zoospore germination of *P. viticola* at different concentrations of fungicides. The efficacy of systemic and combi fungicides on inhibition of sporangial and zoospore germination of *P. viticola* differed significantly.

i) Bioefficacy of systemic and combi-fungicides on inhibition of sporangial germination (zoospore release) of *P. viticola*

At 250 ppm concentration, maximum inhibition of sporangial germination was noticed in famoxadone 16.6% + cymoxanil 22.1% (90.00%) followed by fosetyl-Al (85.00%), azoxystrobin (84.00%), dimethomorph (84.00%) and kresoxim methyl (70.00%). There was least inhibition of sporangial germination in metalaxyl (60.00%).

At 500 ppm concentration, similar maximum inhibition of sporangial germination was noticed in famoxadone 16.6% + cymoxanil 22.1% (95.00%) followed by fosetyl Al (88.00%), azoxystrobin (88.00%), dimethomorph (85.00%) and kresoxim methyl (84.00%). The least inhibition of sporangial germination was noticed in metalaxyl (74.00%).

At 1000 ppm concentration complete or the highest inhibition (100%) of sporangial germination was recorded in both famoxadone 16.6% + cymoxanil 22.1% and azoxystrobin fungicides; followed by fosetyl Al (95.00%), kresoxim methyl (95.00%), and dimethomorph (90.00%) while, metalaxyl recorded the least inhibition of sporangial germination (85.00%). Irrespective of fungicide concentrations, famoxadone 16.6% + cymoxanil 22.1% (95.00%) was found to be the best and significantly superior over rest of the fungicides followed by azoxystrobin (90.66%).

ii) Bioefficacy of systemic and combi-fungicides on inhibition of zoospore germination of *P. viticola*

At 250 ppm concentration, maximum inhibition of zoospore germination was noticed in famoxadone 16.6% + cymoxanil 22.1% (99.00%) followed by fosetyl Al (90.00%), azoxystrobin (85.00%), dimethomorph (85.00%) and kresoxim methyl (84.00%). Least inhibition of zoospore germination was noticed in metalaxyl (70.00%).

Table 3: Bioefficacy of systemic and combi (S+S) fungicides on inhibition of sporangial germination (zoospore release) and zoospore germination of *P. viticola*

Sl. No.	Treatments	Sporangial germination (zoospore release) inhibition (%)				Zoospore germination inhibition (%)			
		Concentration (ppm)				Concentration (ppm)			
		250	500	1000	Mean	250	500	1000	Mean
1.	Azoxystrobin 23% SC	84.00 (66.53)*	88.00 (69.88)	100 (90.00)	90.66 (72.20)	85.00 (67.63)*	95.00 (77.63)	100 (90.00)	93.33 (75.03)
2.	Kersoxim methyl 44.3% SC	70.00 (56.85)	84.00 (66.53)	95.00 (77.63)	83.00 (65.65)	84.00 (66.53)	90.00 (71.70)	95.00 (77.63)	89.66 (71.24)
3.	Fosetyl AL 80% WP	85.00 (67.63)	88.00 (69.88)	95.00 (77.63)	89.33 (70.93)	90.00 (71.70)	95.00 (77.63)	99.00 (85.38)	94.66 (76.64)
4.	Metalaxyl 35% WS	60.00 (50.77)	74.00 (59.64)	85.00 (67.63)	73.00 (58.69)	70.00 (56.85)	84.00 (66.53)	88.00 (69.88)	80.66 (63.91)
5.	Dimethomorph 50% WP	84.00 (66.53)	85.00 (67.63)	90.00 (71.70)	86.33 (68.30)	85.00 (67.63)	94.00 (76.13)	95.00 (77.63)	91.33 (72.88)

6.	Famoxadone 16.6%+ Cymoxanil 22.1%	90.00 (71.70)	95.00 (77.63)	100 (90.00)	95.00 (77.08)	99.00 (85.38)	100 (90.00)	100 (90.00)	99.66 (86.66)
Mean		78.83 (62.61)	85.66 (67.75)	93.66 (75.42)	-	85.50 (67.72)	93.00 (74.66)	96.10 (78.61)	-
Sources of Variation		Treatments	Concen.	T x C	-	Treatments	Concen.	T x C	-
S. Em±		1.05	0.65	1.89	-	1.15	0.85	2.01	-
C. D. @ 1%		3.18	1.86	5.48	-	3.28	2.04	5.88	-
C. V. (%)		3.02				3.26			

* Figures in parentheses indicate angular transformations

Table 4: Bio efficacy of non-systemic and combi (NS+S) fungicides on inhibition of sporangial germination (zoospore release) and zoospore germination of *P. viticola*

Sl. No.	Treatments	Sporangial germination (zoospore release) inhibition%				Zoospore germination inhibition (%)			
		Concentration (ppm)				Concentration (ppm)			
		1000	2000	3000	Mean	1000	2000	3000	Mean
1.	Mancozeb 75% WP	46.00 (42.70)	50.00 (45.00)	70.00 (56.85)	55.33 (48.06)	74.48 (59.84)	82.41 (65.17)	90.74 (72.21)	82.54 (65.30)
2.	Metalaxyl 8% + Mancozeb 64% WP	60.00 (50.77)	74.00 (59.34)	85.00 (67.63)	73.00 (58.69)	70.00 (56.85)	84.00 (66.53)	88.00 (69.88)	80.66 (63.91)
3.	Metalaxyl 4% + Mancozeb 64% WP	60.00 (50.77)	74.00 (59.34)	85.00 (67.63)	73.00 (58.69)	72.00 (58.05)	84.00 (66.42)	90.00 (71.57)	82.00 (64.90)
4.	Ametoctradin 27% + Dimethomorph 20.27% SC	50.00 (45.00)	70.00 (56.85)	75.00 (60.00)	65.00 (71.70)	74.48 (59.84)	82.41 (65.17)	84.00 (66.53)	80.30 (63.65)
5.	Fenamidone 10% + Mancozeb 50% WG	85.00 (67.63)	94.00 (76.13)	95.00 (77.63)	91.33 (72.88)	88.00 (69.88)	99.00 (85.38)	99.00 (85.38)	95.33 (77.52)
6.	Flupicolide 4.44% + Fosetyl-Al 66.67% WG	85.00 (67.63)	88.00 (69.88)	95.00 (77.63)	89.03 (70.66)	99.00 (85.38)	100 (90.00)	100 (90.00)	99.66 (86.66)
7.	Metiram 55% +Pyraclostrobin 5% WG	70.00 (56.85)	84.00 (66.53)	90.74 (72.21)	81.58 (64.58)	84.00 (66.53)	90.00 (71.70)	95.00 (77.63)	89.66 71.24()
Mean		65.14 (53.81)	76.28 (60.85)	85.11 (67.30)	-	80.28 (63.64)	88.83 (70.48)	92.40 (74.00)	-
Sources of Variation		Treatments	Concen.	T x C	-	Treatments	Concen.	T x C	-
S. Em±		1.25	0.95	2.15	-	1.15	0.75	1.99	-
C. D. @ 1%		3.48	2.64	6.34	-	3.28	2.14	5.68	-
C. V. (%)		2.96				3.08			

* Figures in parentheses indicate angular transformations

Table 5: Effect of systemic and combi (S+S) fungicides on inhibition of conidial germination of *E. necator*

Sl. No.	Treatments	Per cent germination inhibition (%)			
		Concentration (ppm)			
		250	500	1000	Mean
1.	Azoxystrobin 23% SC	95.00 (77.63)*	99.00 (85.38)	100 (90.00)	98.00 (84.29)
2.	Hexaconazole 5% EC	95.00 (77.51)	99.00 (85.38)	100 (90.00)	98.00 (84.29)
3.	Tebuconazole 25.9% EC	90.00 (71.70)	95.00 (77.63)	99.00 (85.38)	94.67 (78.23)
4.	Myclobutanil 10% WP	70.00 (56.85)	84.00 (66.53)	88.00 (69.88)	80.67 (64.42)
5.	Tetraconazole 10% EC	85.00 (67.63)	94.00 (76.13)	95.00 (77.63)	91.33 (73.71)
6.	Difenoconazole 25% EC	85.00 (67.63)	90.00 (72.01)	94.00 (75.95)	89.33 (71.53)
7.	Trifloxystrobin 25% + Tebuconazole 50% EC	90.00 (71.70)	95.00 (77.63)	99.00 (85.38)	94.67 (78.23)
8.	Azoxystrobin 11% + Tebuconazole 18.3% EC	99.00 (85.38)	100 (90.00)	100 (90.00)	99.66 (88.46)
Mean		88.63 (70.29)	94.50 (76.44)	96.88 (79.83)	-
Sources of Variation		Treatments	Concen.	T x C	-
S. Em±		1.15	0.75	1.99	-
C. D. @ 1%		3.28	2.14	5.68	-
C. V. (%)		2.98			

*Figures in parentheses indicate angular transformations

Table 6: Effect of non-systemic and Combi (NS+S) fungicides on inhibition of conidial germination of *E. necator*

Sl. No.	Treatments	Per cent germination inhibition (%)			
		Concentration (ppm)			
		1000	2000	3000	Mean
1.	Wettable sulphur 80% WP	46.00 (42.70)*	75.00 (60.33)	85.00 (67.40)	68.83 (56.81)
2.	Tricyclazole 18% + Mancozeb 62% WP	94.13 (75.95)	100 (90.00)	100 (90.00)	98.04 (82.62)
3.	Zineb 68% + Hexaconazole 4% WP	90.15 (71.66)	92.57 (74.11)	100 (90.00)	94.24 (77.28)
4.	Carbendazim 12% + Mancozeb 62%WP	74.48 (59.84)	82.41 (65.17)	90.74 (72.21)	82.64 (65.78)
5.	Chlorothalonil 75% WP	35.00 (36.24)	45.00 (42.12)	50.00 (45.00)	43.33 (41.12)
6.	Dinocap 48% EC	80.00 (63.44)	85.00 (67.31)	95.00 (77.51)	86.67 (69.42)
7.	Ziram 27% SC	45.00 (42.13)	55.00 (47.87)	74.33 (59.57)	58.11 (49.86)
Mean		66.40 (54.57)	76.43 (60.96)	85.01 (67.22)	-
Sources of Variation		Treatments	Concen.	T x C	-
S.Em±		0.73	0.57	1.27	-
C. D. @ 1%		2.13	1.65	3.69	-
C. V. (%)		3.12			

*Figures in parentheses indicate angular transformations

At 500 ppm concentration, maximum inhibition of zoospore germination was noticed in famoxadone 16.6% + cymoxanil 22.1% (100.00%) followed by fosetyl Al (95.00%), azoxystrobin (95.00%), dimethomorph (94.00%) and kresoxim methyl (90.00%). Least inhibition of zoospore germination was noticed in metalaxyl (84.00%).

At 1000 ppm concentration complete (100%) inhibition of zoospore germination was recorded in both famoxadone 16.6% + cymoxanil 22.1% and azoxystrobin fungicides followed by fosetyl Al (99.00%), kresoxim methyl (95.00%), and dimethomorph (95.00%) whereas; metalaxyl recorded the least inhibition of zoospore germination (88.00%). Irrespective of fungicide concentrations, famoxadone 16.6% + cymoxanil 22.1% (99.66%) was found to be the best and significantly superior over rest of the fungicides followed by fosetyl Al (94.66%).

b) Bioefficacy of non-systemic and combi (NS+S) fungicides on inhibition of sporangial germination (zoospore release) and zoospore germination of *P. viticola*

In vitro evaluation of non-systemic and combi fungicides were conducted with respect to inhibition of sporangial and zoospore germination of *P. viticola* at different concentrations of fungicides (Table 4). The efficacy of non-systemic and combi fungicides on inhibition of sporangial and zoospore germination of *P. viticola* differed significantly.

i) Bioefficacy of non-systemic and combi-fungicides on inhibition of sporangial germination (zoospore release) of *P. viticola*

At 1000 ppm concentration, both fluopicolide 4.44% + fosetyl-Al 66.67% and fenamidone 10% + mancozeb 50% treatments resulted in maximum inhibition (85.00% each) of sporangial germination followed by metiram 55% + pyraclostrobin 5% (70.00%), metalaxyl 4% + mancozeb 64% (60.00%), metalaxyl 8% + mancozeb 64% (60.00%) and ametoctradin 27% + dimethomorph 20.27% (50.00%). Least inhibition of sporangial germination was noticed in mancozeb (46.00%).

At 2000 ppm concentration, maximum inhibition of sporangial germination was noticed in fenamidone 10% + mancozeb 50% (94.00%), followed by fluopicolide 4.44% + fosetyl-Al 66.67% (88.00%), metiram 55% + pyraclostrobin 5% (84.00%), metalaxyl 4% + mancozeb 64% (74.00%), metalaxyl 8% + mancozeb 64% (74.00%) and ametoctradin 27% dimethomorph 20.27% (70.00%). Least inhibition of sporangial germination was noticed in mancozeb (50.00%).

At 3000 ppm concentration, maximum inhibition of sporangial germination was noticed in fluopicolide 4.44% + fosetyl-Al 66.67% (95.00%) and fenamidone 10% + mancozeb 50% (95.00%) followed by metiram 55% + pyraclostrobin 5% (90.74%), metalaxyl 4% + mancozeb 64% (85.00%), metalaxyl 8% + mancozeb 64% (85.00%) and ametoctradin 27% + dimethomorph 20.27% (75.00%). Least inhibition of sporangial germination was noticed in mancozeb (70.00%). Irrespective of fungicide concentrations, fenamidone 10% + mancozeb 50% (91.33%) was found to be the best and significantly superior over rest of the fungicides followed by fluopicolide 4.44% + fosetyl-Al 66.67% (89.03%).

ii) Bioefficacy of non-systemic and combi-fungicides on inhibition of zoospore germination of *P. viticola*

At 1000 ppm concentration, maximum inhibition of zoospore germination was noticed in fluopicolide 4.44% + fosetyl-Al 66.67% (99.00%), followed by fenamidone 10% + mancozeb 50% (88.00%), metiram 55% + pyraclostrobin 5% (84.00%), ametoctradin 27% + dimethomorph 20.27% (74.48%), metalaxyl 4% + mancozeb 64% (72.00%) and metalaxyl 8% + mancozeb 64% (70.00%). Least inhibition of zoospore germination was noticed in mancozeb (64.48%).

At 2000 ppm concentration, complete inhibition of zoospore germination was noticed in fluopicolide 4.44% + fosetyl- Al 66.67% (100.00%), followed by fenamidone 10% + mancozeb 50% (99.00%), metiram 55% + pyraclostrobin 5% (90.00%), metalaxyl 4% + mancozeb 64% (84.00%), metalaxyl 8% + mancozeb 64% (84.00%) and ametoctradin 27% + dimethomorph 20.27% (82.41%). Least inhibition of zoospore germination was noticed in mancozeb (72.41%).

At 3000 ppm concentration, maximum inhibition of zoospore germination was noticed in fluopicolide 4.44% + fosetyl-Al 66.67% (100.00%), followed by fenamidone 10% + mancozeb 50% (99.00%), metiram 55% + pyraclostrobin 5% (95.00%), metalaxyl 4% + mancozeb 64% (90.00%), metalaxyl 8% + mancozeb 64% (88.00%) and ametoctradin 27% + dimethomorph 20.27% (84.00%). Least inhibition of zoospore germination was noticed in mancozeb (80.74%). Irrespective of fungicide concentrations, flupicolide 4.44% + fosetyl- Al 66.67% (99.66%) was found to be the best and significantly superior over rest of the fungicides followed by fenamidone 10% + mancozeb 50% (95.33%).

c) Bioefficacy of systemic and combi (S+S) fungicides on inhibition of conidial germination of *E. necator*

In vitro evaluation (Table 5) of systemic fungicides was conducted with respect to inhibition of conidial germination of *E. necator* at different concentrations of fungicides. The efficacy of systemic and combi fungicides on inhibition of conidial germination of *E. necator* differed significantly.

At 250 ppm concentration, maximum inhibition of conidial germination was noticed in azoxystrobin 11% + tebuconazole 18.3% (99.00%) followed by hexaconazole and azoxystrobin (95.00% each), tebuconazole and trifloxystrobin 25% + tebuconazole 50% (90.00% each), tetraconazole and difenoconazole (85.00% each). Least inhibition of conidial germination was noticed in myclobutanil (70.00%).

At 500 ppm concentration complete inhibition (100.00%) of conidial germination was noticed in azoxystrobin 11% + tebuconazole 18.3% followed by hexaconazole and azoxystrobin (99.00% each), tebuconazole and trifloxystrobin 25% + tebuconazole 50% (95.00% each), tetraconazole (94.00%) and difenoconazole (90.00%). Least inhibition of conidial germination was noticed in myclobutanil (84.00%).

At 1000 ppm concentration both azoxystrobin 11% + tebuconazole 18.3%, azoxystrobin and hexaconazole inhibited 100 per cent conidial germination followed by tebuconazole and trifloxystrobin 25% + tebuconazole 50% (99.00% each), tetraconazole (95.00%) and difenoconazole (94.00%) whereas, myclobutanil recorded the least inhibition of conidial germination (88.00%). Irrespective of fungicide concentrations, azoxystrobin 11% + tebuconazole 18.3% (99.66%) was found to be the best and significantly superior over rest of the fungicides followed by hexaconazole and azoxystrobin (98.00% each).

d) Bioefficacy of non-systemic and combi (NS+S) fungicides on inhibition of conidial germination of *E. necator*

Efficacy of different non systemic and combi fungicides at 1000, 2000 and 3000 ppm concentrations were evaluated *in vitro* (Table 6) on inhibition of conidial germination of *E. necator*.

Tricyclazole 18% + mancozeb 62% at 1000 ppm was found to inhibit (94.13%) conidial germination and proved significantly superior over other fungitoxicants. This was followed by zineb 68% + hexaconazole 4% (90.15%), dinocap (80.00%), carbendazim 12% + mancozeb 62% (74.48%), wettable sulphur (46.00%) and ziram (45.00%). Least conidial inhibition was observed in chlorothalonil (35.00%).

At 2000 ppm concentration, tricyclazole 18% + mancozeb 62% inhibited 100 per cent conidial germination followed by zineb 68% + hexaconazole 4% (92.57%) and least was observed in chlorothalonil (45.00%).

At 3000 ppm concentration, tricyclazole 18% + mancozeb 62% and zineb 68% + hexaconazole 4% showed 100 per cent inhibition of conidial germination. The least inhibition of germination was found in chlorothalonil (50.00%). However, irrespective of the concentrations maximum conidial inhibition was noticed in tricyclazole 18% + mancozeb 62% (98.04%) and was significantly superior over rest of the treatments. Chlorothalonil (43.33%) was found to be the least effective among the fungitoxicants. All the concentrations differed significantly and maximum inhibition was recorded in higher concentration (3000 ppm) compared to lower concentrations.

Discussions

a) Bioefficacy of different fungicides against *P. viticola* of grapes

Among the systemic and its combi-fungicides (S+S); the highest inhibition (95.00%) across different concentrations evaluated; famoxadone 16.6% + cymoxanil 22.1% was superior over azoxystrobin (90.66%) fosetyl Al (89.33%) dimethomorph (86.33%) and kresoxim methyl (83.00%); while metalaxyl (73.00%) was the least effective fungicide in inhibition of sporangial (zoospore release) germination of *P. viticola*.

Similarly studies with respect to zoospore germination inhibition showed that famoxadone 16.6% + cymoxanil 22.1% (99.66%) was superior over fosetyl Al (94.66%) followed by azoxystrobin (93.33%) dimethomorph (91.33%) and kresoxim methyl (89.66%) across different concentrations evaluated. Metalaxyl (80.66%) was least effective in inhibiting the zoospore germination of *P. viticola*.

On similar lines Trajeevskii (2003) observed 100 per cent control with Ridomil Plus 48WP (8% metalaxyl +40% copper oxy-chloride) and Mikal (50% fosetyl- Al +25% folpet). He further observed good results (more than 90%) with Acrobat MZ (9% dimethomorph + 60% mancozeb), Sandofan C (10% oxadixyl +40% copper oxy-chloride), Folicur-E 50WP (10% tebuconazole + 40% dichlofluanid) and Caltan (6% ofurace +45% folpet).

Sawant *et al*. (2011) revealed that use of azoxystrobin 23SC spraying after fruit pruning in vineyards of Maharashtra on Thompson seedless and Krishna varieties was found most effective against downy mildew of grapes.

Among non-systemic and its combi-products (NS+S) the significantly highest inhibition was obtained by fenamidone 10% + mancozeb 50% (91.33%) followed by fluopicolide 4.44% + fosetyl Al 66.67% (89.03%) and mancozeb (55.33%) was least effective in inhibiting the sporangial germination of *P. viticola*. Also the highest zoospore inhibition was observed in fluopicolide 4.44% + fosetyl-Al 66.67% (99.66%) followed by fenamidone 10% + mancozeb 50% (95.33%) while mancozeb (82.54%) was least effective in inhibiting the zoospore germination of *P. viticola*. The resulst obtained were in agreement with; Czermainski and Sonego (2004) reported that cymoxanil + mancozeb, metalaxyl + mancozeb and dithianon were the most effective fungicides in controlling the downy mildew of grapes, whereas copper oxy-chloride alone or in combination with mancozeb and copper sulphate was least effective.

Rekanovic *et al.* (2008) worked on efficacy of new fungicide mixtures (fluopicolide + fosetyl Al and fosetyl Al + folpet) in controlling *P. viticola* and found that both the tested fungicides exhibited high efficacy. There were no significant differences in the bio-efficacy of these combi-fungicides *viz*. Profiler [fluopicolide + fosetyl Al] (9.1-99.7%) and Mikal Flash [fosetyl Al + folpet] (94.9-99.2%).

b) Bioefficacy evaluation of different fungicides against *E. necator* of grapes

Among the systemic and its combi-fungicides (S+S); across different concentrations the highest inhibition was obtained by azoxystrobin 11% + tebuconazole 18.3% (99.66%) followed by azoxystrobin and hexaconazole (98.00% each), tebuconazole (94.67%), trifloxystrobin 25% + tebuconazole

50% (94.67%), tetraconazole (91.33%) and difenoconazole (89.33%) whereas, myclobutanil recorded the least inhibition (80.67%) of conidial germination.

Similarly, several researchers (Shitole *et al.* (2001) [12], Archana (2009) [1] and Divyajyothi (2012) [5] are in agreement with the present evaluation. Nithyameenakshi *et al*. (2010) [8] evaluated six fungicides *viz.* Amistar, Score, Ridomil, Calixin, Dithane M- 45 and wettable sulphur at 0.02, 0.05, 0.1 and 0.25 per cent concentration under *in vitro* conditions against *U. necator*. They reported that azoxystobin and difenoconazole were superior over other fungicides at 0.05 per cent in arresting the germination of *U. necator.*

Among non-systemic and its combi products (NS+S) the significantly highest inhibition was obtained by tricyclazole 18% + mancozeb 62% (98.04%) followed by zineb 68% + hexaconazole 4% (94.24%), across different concentrations and chlorothalonil (43.33%) was found least effective in inhibiting the germination of *E. necator*.

Shithole *et al.* (2001) [12] reported Karathane as the best non-systemic fungicide in inhibiting spore germination of *U. necator*. Ashwathanarayana (2003) [2] also reported karathane and wettable sulphur as effective in complete inhibition of conidial germination at 0.3 per cent against grape powdery mildew.

These results indicated that the maximum inhibition of sporangial and zoospore germination of *P. viticola* was noticed in famoxadone 16.6% + cymoxanil 22.1% (Equation Pro) and fluopicolide 4.44% + fosetyl-Al 66.67% WG (Profiler) and maximum inhibition of conidial germination of *E. necator* was noticed in azoxystrobin 11% + tebuconazole 18.3% (Custodia) and tricyclazole 18% + mancozeb 62% (Merger). The efficacy of fungicides obtained in the present study is in agreement with the earlier workers (Wicks and Lee, 1982; Wicks *et al*. 1987; Magarey *et al*. 1991; Wicks *et al*. 1999 and Wong and Wilcox, 2001) [13, 15, 7, 14, 16]. Efficient use of either of these fungicides will rely on the development of disease forecasting systems that monitor climatic conditions and accurately predict the occurrence of *P. viticola* and *E. necator* on grapes. However, in many viticulture areas accurate disease forecasting systems are not yet available and under this situation the only alternative is to apply the fungicide sprays within a few days before the start of an infection period in order to manage the diseases at earliest.

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