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Evaluation of Azoxystrobin and Difenoconazole Against Certain Crop Diseases*

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Abstract: Efficacy of two new fungicides Amistar and Score, yet to be introduced to farmers and planters in India, was evaluated under laboratory, greenhouse and field conditions. Three common grapevine diseases viz., downy mildew, powdery mildew and anthracnose caused by *Plasmopara viticola, Uncinula necator* and *Gloeosporium ampelophagum* respectively and blister blight disease of tea caused by *Exobasidium vexans* were studied in this investigation. *In vitro* spore germination assay revealed that both Azoxystrobin and Difenoconazole were superior to other fungicides at 0.05% concentration in arresting the germination of the propagules of the pathogens studied. In greenhouse and field evaluations also Azoxystrobin and Difenoconazole proved to be superior over other fungicides in controlling the grapevine diseases and blister blight of tea to the maximum extent at 0.05% concentrations.

Key words: Amistar, score, downy mildew, powdery mildew, anthracnose, blister blight

Introduction

The development of systemic fungicides in the past 50 years is a landmark in the history of plant disease control using fungicides and is comparable to that of weed control with the advent of 2, 4-dichlorophenoxyaceticacid in the 1940s. Potato failure of Ireland (1845), coffee loss of Ceylon (1893-94) and Great famine of Bengal (1942-43) emphasize the need for biological or chemical control against epidemic fungi (Ganguly, 1989).

Blister blight in tea (*Camellia sinensis*) is known as early as 1868 (Peal, 1868). Despite the fact that this disease struck the tea with devastating severity in Assam and Darjeeling over three quarter century ago, field scale protection against the disease became an economic possibility only a quarter century back. The course of blister blight control until 1970 has been reviewed by Venkataram (1970). Later, several chemicals like copperoxychloride (Pfaeltzer, 1951; Knaap, 1956; Jayaraman and Venkataramani, 1957; Visser *et al*., 1958), daconil and difolaton (Venakataram, 1969), nickel chloride (Venkataram, 1962), 1, 4- oxathiin, carboxin and oxycarbonin (Venkataram, 1969) and tridemorph (Venkataram and Chandramouli, 1976) were proved to be effective. Recently, triazole group of chemicals (hexaconazole, propiconazole and bitteranol) in combination with copperoxychloride were proved to be superior in controlling blister blight in southern India (Premkumar and Muraleedharan, 2000; Premkumar *et al*., 2000).

Of all grape diseases, the most serious are powdery mildew (*Uncinula necator*), downy mildew (*Plasmopara viticola*) and anthracnose (*Gloeosporium ampelophagum*). The application of single chemical for the control of both mildews came in 1973 (Malenin and Kobokhidze, 1973) with benomyl, pyrazophos and/ or thiophonate fungicides.

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Amistar (azoxystrobin 25% EC) and Score (difenoconazole 25% EC) are the two broad spectrum, foliar, systemic fungicides, yet to be released to the farmers and planters in India by Syngenta India Limited, Mumbai. The objective of the present study was to evaluate the efficiency of the said fungicides against two important crop diseases.

Materials and Methods

Collection and Maintenance of Pathogens

Source and Maintenance of Plasmopara viticola Inoculum

Grapevine leaves (cv. Thompson seedless) affected by downy mildew and showing the typical symptoms of the disease were collected from Odaipatti, a traditional grapevine growing area in Theni district, Tamil Nadu and were used as the source of sporangial inoculum of the pathogen. These leaves were surface sterilized with 70% ethanol for 30 sec. in order to remove phylloplane microorganisms. The leaves were then rinsed with sterile distilled water and placed in a sterile Petri dish lined with sterilized moist absorbent cotton in such a way that their abaxial surface was facing upwards. Then the plates were incubated in dark for 10 h at 20°C. Sporangia developed on these leaves were dispersed in sterile distilled water using a camel hair brush and the sporangia were washed by centrifuging the sporangial suspension at 3500 rpm for 5 min. The concentration of the sporangia was adjusted to 5×10^4 sporangia mL^{-1} using a heamocytometer. This sporangial suspension was sprayed on 120-day-old highly susceptible grapevine plants (cv. Thompson seedless) grown in pots and maintained in the glass house. Both 24 h prior to and after inoculation, water congestion was given to grapevine plants to achieve 100% RH by profusely spraying the plants with distilled water and covering with polythene bags. The sporangia collected from these plants were used for further studies.

Source and Maintenance of Uncinula necator Inoculum

Grapevine leaves showing the typical symptoms of powdery mildew were collected from the orchard, Agricultural College and Research Institute, Madurai and the conidia collected from such plants were used for artificial inoculation. From the diseased leaves, conidia were collected by using a camel hair brush and suspended in sterile distilled water containing Tween 20. The conidial suspension was then centrifuged at 3000 rpm for 5 min twice, in order to separate conidia from conidiophores (Kitao and Doazan, 1989). The concentration of the conidia was adjusted to 3×10^4 conidia mL^{-1} using a haemocytometer. This suspension was profusely sprayed on the healthy leaves of potted grapevine plants (120-day-old) using an atomizer and maintained in the glass house. After inoculation, the plants were covered by polythene bags for 24 h to maintain high humidity for disease development.

Isolation of the Gloeosporium ampelophagum

Gloeosporium ampelophagum was isolated from the grapevine leaves showing the typical symptoms of anthracnose using Potato Dextrose Agar (PDA) medium. The fungus was purified by single spore isolation technique and such purified culture was maintained on PDA slants for further studies.

Source and Maintenance of Exobasidium vexans

Basidiospores were collected from naturally infected leaf lesions of the susceptible tea clone TES-34 from the plantations of UPASI Tea Research Institute, Valparai, Coimbatore district. Infected tea shoots were collected from the field in the evenings and the cut ends of the shoots were kept in sterile glass vials containing 1% glucose solution. The leaf with a single actively sporulating lesion was selected from the collected shoots and placed in a sterile beaker and the mouth of the beaker was

covered with a glass plate. The set up was incubated overnight inside a bell jar under 100% relative humidity. The basidiospores liberated into the beaker were made into a suspension (1×10^8 spores mL^{-1}) using sterile distilled water. The spore suspension was used for artificial inoculation and spore germination studies.

The potted plants were gently sprayed with distilled water. Then, the spore suspension with known quantity of basidiospores was painted on the upper surface of first and second leaves, using a sterile camel hair brush. Soon after inoculation, plants were covered by polythene bags and incubated at 20-22°C under 100% relative humidity, for disease development.

In Vitro Evaluation of Fungicides

In Vitro Evaluation of Fungicides Against Grapevine Diseases

Various fungicides viz., Amistar, Score, Ridomil, Calixin, Dithane M-45 and Sulphur (wettable) were evaluated at 0.02, 0.05, 0.1 and 0.25% concentrations under *in vitro* conditions against grapevine pathogens. All sets of measurement were repeated by conducting a separate set of measurements on a separately executed experiment.

Against Sporangial Germination of P. Viticola (Cavity Slide Technique)

The efficacy of fungicides against *P. viticola* was tested by cavity slide technique. One drop of each of the fungicide concentrations was placed separately in a sterile cavity slide and allowed to air dry. One drop of the sporangial suspension of (5×10^4 sporangia mL^{-1}) *P. viticola* was added and mixed thoroughly. Sporangial suspension + sterile distilled water served as control. Then, the slides were kept in a Petri dish where sterilized cotton wool was spread inside and moistened with sterile distilled water. The Petri dish lid was replaced and the plates were incubated at 20°C. Observations on the sporangial germination was recorded 6 h later by counting the total number of sporangia and the number of sporangia germinated in each microscopic filed. Three such microscopic fields were observed and the mean per cent germination and per cent germination inhibition were worked out. Three replicates were maintained for each treatment.

Against the Conidial Germination of U. necator (Detached Leaf Technique)

The efficacy of fungicides against the conidial germination of *U. necator* was assessed by detached leaf technique (Varalakshmi *et al*., 1999). Grapevine leaves were washed in sterile distilled water and air dried. Different concentrations of the fungicides taken for study were placed individually on the adaxial surface of the leaf, the droplets were evenly spread with a fine camel hair brush and allowed to air dry. The treated leaves were inoculated with the conidia of *U. necator* (3×10^4 conidia mL^{-1}). The leaves sprayed with the conidial suspension alone served as the control. Three leaves from each treatment were transferred to a Petri dish with their petioles dipped in water and incubated at 20°C. Each treatment was replicated thrice. After 72 h, the leaves were observed under the microscope (equipped with fine light arrangement) for conidial germination. The total and germinated conidia were counted in three microscopic fields and per cent inhibition of conidial germination was worked out.

Against the Conidial Germination of G. ampelophagum

The efficacy of fungicides against the conidial germination of *G. ampelophagum* was assessed by cavity slide technique as described earlier.

In Vitro Screening of Fungicides Against Blister Blight of Tea

Various fungicides namely Amistar, Score, Tilt, Copper oxychloride, Tridemorph, and Nickel chloride were evaluated against *E. vexans* by cavity slide method as described earlier.

Greenhouse Evaluation of Fungicides

All sets of measurement were repeated by conducting a separate set of measurements on a separately executed experiment.

Against Grapevine Diseases

Grapevine stem cuttings (cv. Thompson seedless) were planted in earthen pots (30 cm diameter) and maintained in the glass house. When the grapevine plants were 120-day-old, the fungicides listed earlier were applied on the plants in three separate sets of experiments. Artificial inoculation of grapevine downy mildew, powdery mildew and anthracnose pathogens were made by spraying sporangial suspension (5×10^4 sporangia mL^{-1}), conidial suspension (3×10^4 conidia mL^{-1}) and conidial suspension (5×10^4 conidia mL^{-1}), respectively as described in the earlier section. Water congestion was provided 24 h before and after inoculation by profusely spraying the plants with sterile distilled water and covering with polythene bags.

The experiments were conducted for the downy mildew, powdery mildew and anthracnose diseases in Randomized block design with the similar sets of treatments except for the chemical check. The inoculated and uninoculated controls were also maintained for all the three diseases. Three replicates were maintained for each treatment.

Observations on disease incidence were recorded 15 days after spraying using the score charts given below. The results were expressed as percent disease index.

Score chart for downy mildew (Brown *et al.*, 1999)

Disease grade	Percent leaf area affected
0	No infection
1	0-10
2	10.1-30
3	30.1-60
4	60.1-80
5	80.1-100

Score chart for powdery mildew:

Disease grade	Percent leaf area affected
0	No infection
1	1-10
2	10.1-25
3	25.1-50
4	50.1-75
5	75.1-100

Score chart for anthracnose (Chandrasekararao, 1989):

Disease grade	Percent leaf area affected
0	No infection
1	A few spots on leaves covering upto five per cent leaf area
2	Spots covering 5.1-35% leaf area and few lesions of 2-5 mm size on petiole and stem
3	Severe leaf lesions covering 35.1-70% leaf area and moderate lesions of 5.1-10 mm size on petiole and stem
4	Many leaves dried; Severe lesions of above 10 mm size on petioles and stem portion
5	All the leaves and twigs of new growth killed

Against Blister Blight Disease in Tea

The efficacy of fungicides was studied under greenhouse conditions with potted plants. Tender shoots of tea plants were sprayed lightly with distilled water and inoculated with basidiospores on the upper surface of the first and second leaves. Fungicide treatment was given by spraying the respective fungicides five days before or five days after the inoculation with basidiospores. Soon after inoculation, the plants were covered with bell jars and incubated at 20-22°C under 100% relative humidity. Blister

blight development was recorded up to 15 days after inoculation. In control, the plants were sprayed with distilled water instead of fungicide. Each treatment had five replicates. Blister blight incidence was quantified on per cent of twigs infected. A twig was considered as infected if a single lesion was noted.

Efficacy of Fungicides Tested under Field Condition

All sets of measurement were repeated by conducting a separate set of measurements on a separately executed experiment.

Against Grapevine Diseases

Field experiments were conducted in a randomized block design with three replicates at Odaipatti in Theni district during monsoon season of 2004 to evaluate the efficacy of various fungicides against downy mildew, powdery mildew and anthracnose diseases. Spraying was given with the fungicides on grapevine plants one month after pruning (most vulnerable stage to all the three diseases) when the disease symptoms usually appear and fifteen days after first spraying. Observations on the disease incidence was recorded 15 days after second spraying by following the score charts of the respective diseases as already furnished. The per cent disease index (PDI) was worked out by using Mckinney's (1923) formula:

$$PDI = \frac{\text{Sum of all number rating}}{\text{Total number of leave observed}} \times \frac{100}{\text{Maximum grade in the score chart}}$$

Against Blister Blight Disease in Tea

Various fungicides were tested for their efficacy against blister blight in the plantation of UPASI Tea Research Institute, Valparai, Coimbatore district, during the south-west monsoon season, 2003. The experiments were carried out in randomized block design with adequate number of bushes (50-60 per plot) and replicates (3 plots). Fungicide sprays were imposed on the 1st, 3rd, 5th, 7th, 10th and 14th of September, 2003.

On the third week after the first fungicide application, disease incidence in the experimental plots was assessed. One hundred shoots of the same age (three leaves and a bud) of uniform size were collected randomly from the harvest and each shoot was examined. A shoot was counted as infected even if a single lesion was noted. Right from oil spots to mature sporulating lesion were taken into account. A partially killed blister lesion was always considered as viable. Fully necrotized lesions alone were considered as controlled. Disease incidence was quantified on percentage basis.

Results

In Vitro Evaluation of Fungicides

Against Grapevine Diseases

Efficacy of the systemic fungicides Azoxystrobin and Difenoconazole were evaluated against *P. viticola*, *U. necator* and *G. ampelophagum* by cavity slide method, detached leaf technique and cavity slide method respectively. Other commonly used fungicides, viz., Ridomil, Calixin, Dithane M-45 and Sulphur were used for comparison. The results are presented in Table 1 to 3. Among the treatments, Azoxystrobin and Difenoconazole at 0.05% recorded the maximum spore inhibition of 95.7 and 92.0% (*P. viticola)*, 80.0 and 86.0% (*U. necator*) and 89.7 and 85.3% (*G. ampelophagum*), respectively. This was followed by Ridomil and Calixin, where per cent spore inhibition was recorded as 76.0 and 74.7% (*P. viticola)*, 37.0 and 82.7% (*U. necator*) and 36.3 and 76.7% (*G. ampelophagum*), respectively.

Table 1: *In vitro* screening of fungicides against sporangial germination of *Plasmopara viticola*

	Concentration of the fungicides (%)							
	0.02		0.05		0.1		0.25	
Fungicide	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)
Amistar	61.5	38.33a	4.5	95.67a	0	100.00a	0	100.00a
Score	65.0	35.00a	8.0	92.00a	0	100.00a	0	100.00a
Ridomil	64.5	35.33a	24.0	76.00b	11.5	88.67b	0	100.00a
Calixin	84.2	16.00b	25.5	74.67b	20.0	80.00c	0	100.00a
Dithane M-45	95.0	5.00cd	85.0	15.00c	63.5	36.67d	52.0	48.00b
Sulphur	91.6	8.33c	81.5	18.67c	68.5	31.67d	64.0	36.00c
Control	100.0	0.00	100.0	0.00	100	0.00	100.0	0.00

In a column, means followed by a common letter(s) are not significantly different at the 5% level by DMRT

Comparison	SED	LSD (5%)	LSD (1%)
Treatment vs concentration means	2.69	5.40	7.19

Table 2: *In vitro* screening of fungicides against conidial germination of *Uncinula necator*

	Concentration of the fungicides (%)							
	0.02		0.05		0.1		0.25	
Fungicide	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)
Amistar	51.5	48.67a	20.0	80.00a	0.0	100.00a	0.0	100.00a
Score	58.6	41.67ab	14.2	86.00a	0.0	100.00a	0.0	100.00a
Ridomil	81.5	18.67c	63.0	37.00c	51.4	48.67b	18.8	81.33b
Calixin	59.4	40.67ab	17.5	82.67a	0.0	100.00a	0.0	100.00a
Dithane M-45	85.0	15.00c	74.2	26.00d	63.5	36.67c	39.6	60.67c
Sulphur	65.0	35.00b	55.0	45.33b	43.5	56.33b	25.0	75.00b
Control	100.0	0.00	100.0	0.00	100.0	0.00	100.0	0.00

In a column, means followed by a common letter(s) are not significantly different at the 5% level by DMRT

Comparison	SED	LSD (5%)	LSD (1%)
Treatment vs concentration means	3.90	7.82	10.42

Table 3: *In vitro* screening of fungicides against conidial germination of Gloeosporium ampelogphagum

	Concentration of the fungicides (%)							
	0.02		0.05		0.1		0.25	
Fungicide	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)
Amistar	34.6	65.67a	10.5	89.67a	0.0	100.00a	0.0	100.00a
Score	32.0	68.00a	14.8	85.33a	0.0	100.00a	0.0	100.00a
Ridomil	78.5	21.67c	63.6	36.33c	52.0	48.00b	39.5	60.67b
Calixin	44.5	55.67b	23.2	76.67b	0.0	100.00a	0.0	100.00a
Dithane M-45	84.4	15.67c	77.5	22.67d	63.2	36.67c	52.0	48.00c
Sulphur	81.5	18.33c	75.0	25.33d	58.4	41.33bc	48.0	52.00c
Control	100.0	0.00	100.0	0.00	100.0	0.00	100.0	0.00

In a column, means followed by a common letter (s) are not significantly different at the 5% level by DMRT

Comparison	SED	LSD (5%)	LSD (1%)
Treatment vs concentration means	3.64	7.30	9.73

Against Blister Blight of Tea

The fungicides Azoxystrobin and Difenoconazole were tested *in vitro* against *E. vexans* by cavity slide technique. The fungicides Tilt, Copper oxychloride (COC), Tridemorph and Nickel chloride were used for comparison and the results are presented in Table 4. Among the fungicides, Difenoconazole and Azoxystrobin at 0.05% concentration recorded, respectively 92.7 and 89.0% inhibition of basidiospore germination. This was followed by Calixin where 61.0% inhibition of spore germination was recorded.

Green House Evaluation of the Fungicides Against Grapevine Diseases

Based on the results of previous *in vitro* experiments, the effective concentration of fungicides was fixed as 0.05% in the case of Azoxystrobin and Difenoconazole. For other fungicides, the recommended doses were adopted. The efficacy of these fungicides was tested under greenhouse condition and the results are presented in Table 5. Among the treatments, Azoxystrobin and Difenoconazole recorded the maximum disease reduction of 69.71 and 67.77% (*P. viticola)*, 61.80 and 60.38% (*U. necator)* and 65.73 and 59.09% (*G. ampelophagum*), pectively over control under green house conditions.

Table 4: *In vitro* screening of fungicides against basidiospore germination of *Exobasidium vexans*

	Concentration of the fungicides (%)							
	0.02		0.05		0.1		0.25	
Fungicide	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)	Germination (%)	Inhibition (%)
Amistar	70.2	30.00a	11.0	89.00a	0.0	100.00a	0.0	100.00a
Score	67.5	32.33a	7.5	92.67a	0.0	100.00a	0.0	100.00a
Tilt	82.0	18.00b	60.5	39.67c	31.8	68.33c	15.0	85.00b
COC	91.5	8.67c	84.4	15.67d	67.2	33.00e	43.5	56.33d
Tridemorph	74.4	25.67a	39.0	61.00d	10.5	89.33b	0.0	100.00a
Nickel chloride	81.5	18.67b	65.0	35.00c	42.0	58.00d	35.0	65.00c
Control	100.0	0.00	100.0	0.00	100.0	0.00	100.0	0.00

In a column, means followed by a common letter(s) are not significantly different at the 5% level by DMRT

Comparison	SED	LSD (5%)	LSD (1%)
Treatment vs concentration means	3.38	6.77	9.01

Table 5: Greenhouse evaluation of fungicides against grapevine diseases

		Downy mildew		Powdery mildew		Anthracnose	
Fungicide	Concentration (%)	PDI	Disease reduction over control (%)	PDI	Disease reduction over control (%)	PDI	Disease reduction over control (%)
Amistar	0.05	27.40d	69.71	34.57d	61.80	31.60d	65.73
Score	0.05	29.13d	67.77	35.85d	60.38	37.73d	59.09
Ridomil	0.25	39.13d	56.71	62.27b	31.19	51.40b	44.26
Calixin	0.1	37.33d	58.70	44.13c	51.23	39.50c	57.17
Dithane M-45	0.25	64.20c	29.01	78.30a	13.48	54.50b	40.90
Sulphur	0.25	77.07b	14.71	50.63b	44.05	61.40b	33.42
Control	-	90.40a	-	90.50a	-	92.23a	-

In a column, means followed by a common letter(s) are not significantly different at the 5% level by DMRT

Comparison	SED	LSD (5%)	LSD (1%)
Treatment vs disease means	6.19	12.51	16.74

Table 6: Greenhouse evaluation of fungicides against blister blight of tea

Fungicide	Concentration (%)	PDI	Disease reduction over control (%)
Amistar	0.05	25.50a	72.10
Score	0.05	21.80a	76.14
Tilt	0.25	67.00c	26.69
COC	0.1	85.50d	6.45
Tridemorph	0.25	53.20b	41.79
Nickel chloride	0.25	69.60c	23.85
Control	-	91.40d	-

Note: PDI- plant disease index Means followed by a common letter are not significantly different at the 5% level by DMRT

Comparison	SED	LSD (5%)	LSD (1%)
Treatment means	4.18	9.11	12.78

Table 7: Field evaluation of fungicides against grapevine diseases

		Downy mildew		Powdery mildew		Anthracnose	
Fungicide	Concentration (%)	PDI	Disease reduction over control (%)	PDI	Disease reduction over control (%)	PDI	Disease reduction over control (%)
Amistar	0.05	28.80 f	68.89	36.80 e	60.48	34.70 e	63.38
Score	0.05	32.40 ef	65.01	38.73 e	58.41	39.87 de	57.92
Ridomil	0.25	42.50 d	54.10	65.00 c	30.20	55.63 c	41.29
Calixin	0.1	39.00 de	57.88	46.80 d	49.74	42.63 d	55.01
Dithane M-45	0.25	66.50 c	28.18	80.20 b	13.88	59.00 bc	37.74
Sulphur	0.25	82.40 b	11.01	53.70 d	42.33	65.13 a	31.27
Control	-	92.60 a	-	93.13 a	-	94.77 a	-

Note: PDI - plant disease index In a column, means followed by a common letter are not significantly different at the 5% level by DMRT

Comparison	SED	LSD (5%)	LSD (1%)
Treatment means	3.63	7.34	9.82

Table 8: Field evaluation of fungicides against blister blight of tea

Fungicide	Concentratio (%)	PDI	Disease reduction over control (%)
Amistar	0.05	26.70a	71.50
Score	0.05	24.70a	73.63
Tilt	0.25	70.40c	24.86
COC	0.1	89.00d	5.01
Tridemorph	0.25	55.70b	40.55
Nickel chloride	0.25	70.10c	25.18
Control	-	93.70d	-

Note: PDI - plant disease index Means followed by a common letter (S) are not significantly different at the 5% level by DMRT

Comparison	SED	LSD (5%)	LSD (1%)
Treatment means	3.01	6.56	9.19

Against Blister Blight of Tea

Efficacy of Azoxystrobin (0.05%) and Difenoconazole (0.05%) along with four other fungicides (recommended doses) was tested against *E. vexans* under green house conditions and the results are presented in Table 6. Among the treatments, Difenoconazole and Azoxystrobin were highly effective where 76.14 and 72.10% disease reduction was recorded when compared with control.

Field Evaluation of the Fungicides
Against Grapevine Diseases

Field evaluation also confirmed the fact that among all the fungicides tested, sulphur proves to be the least effective against downy mildew (82.4% PDI) and anthracnose (65.13% PDI). But, the best control against all the grapevine diseases tested was achieved using Azoxystrobin and Difenoconazole under field conditions (Table 7).

Against Blister Blight of Tea

Field evaluation carried out against blister blight disease of tea indicated that the best disease control was obtained with Difenoconazole (73.63% PDI) followed by Azoxystrobin (71.50% PDI). On the other hand, COC proved to beless effective against the disease with 89.0% PDI (Table 8).

Discussion

Both Azoxystrobin and Difenoconazole are systemic and broad spectrum fungicides with protective and curative properties. Azoxystrobin belongs to strobilurin-class of fungicides and has azoxystrobin as its active ingredient (25%). The strobilurins are a new class of fungicidal compounds

modeled after natural compounds isolated from several Basidiomycetes species that decay woody substrates (Anke, 1995). They are also known as QoI fungicides because they inhibit mitochondrial respiration in fungi by binding to the Qo (quinone-oxidizing) sites (Gisi *et al*., 2002). Strobilurins were developed in the nineties and among them, azoxystrobin has been registered for use in Israel and other countries (Baldwin *et al*., 1996) and very little information is available on its efficiency in tropical countries. The active ingredient of Score is difenoconazole (25% EC), which was introduced by Syngenta in early 1990's. It is taken by the plant and acts on the fungal pathogen during penetration and haustoria formation by interfering with the biosynthesis of sterols in cell memberane.

Three common grapevine diseases viz., downy mildew, powdery mildew and anthracnose incited by *Plasmopara viticola*, *Uncinula necator* and *Gloeosporium ampelophagum* respectively and blister blight disease of tea caused by *Exobasidium vexans* were taken for the study. Four commonly used fungicides viz., Ridomil, Calixin, Dithane M-45 and Sulphur were used as standard treatments for comparison along with the test fungicides in the control of grapevine diseases. In the case of blister blight of tea, Tilt, Copper oxychloride, Tridemorph and Nickel chloride were used as standards along with Azoxystrobin and Difenoconazole for comparison.

In vitro spore germination assay revealed that both Azoxystrobin and Difenoconazole were superior to other fungicides in arresting the germination of the propagules of the pathogens studied. Both the test fungicides gave 100% inhibition of spore germination of all the pathogens studied at their 0.1% concentration. At this concentration, Calixin g ave similar spore germination inhibition of *U. necator* and *G. ampelophagum* but showed only 80% inhibition in the germination of *P. viticola* sporangia. Dithane M-45 and Sulphur showed poor inhibition of spore germination among grapevine pathogens. In the spore germination assay of *E. vexans* also, all the other fungicides used were inferior to Azoxystrobin and Difenoconazole.

The efficacy of Azoxystrobin (azoxystrobin) in inhibiting the sporangial germination of the grapevine downy mildew pathogen *Plasmopara viticola* at the lowest concentration of 250 ppm has been reported (Sendhilvel *et al*., 2004). Trifloxystrobin, another strobilurin fungicide, strongly inhibited spore germination of *Uncinula necator* at low concentrations (Reuveni, 2000). Strong antisporulant action of triazole class of fungicides (bitertanol, hexaconazole and propiconazole) on the blister blight of tea pathogen *Exobasidium vexans* was observed by Premkumar (2001). They also affected the viability of the basidiospores of the pathogen significantly. Inhibition of spore germination of *E. vexans* by triazole fungicides on the leaf surface of tea was reported by Baby *et al*. (2000).

In greenhouse and field evaluations also Azoxystrobin and Difenoconazole proved to be superior over other fungicides in controlling the grapevine diseases and blister blight of tea to the maximum extent at 0.05% concentrations. In the control of grapevine diseases, Calixin was the next preferred fungicide. On the other hand, in blister blight control, no other fungicide gave better disease control as Azoxystrobin and Difenoconazole.

Effective control of downy and powdery mildews in grapevine at very low dosage level of Azoxystrobin (125 g (a.i.,) ha^{-1}) has been reported by earlier workers (Godwin *et al*., 1992; Baldwin *et al*., 1996; Wilcox and Riegel, 1998; Ranganatha *et al*., 2001; Wong and Wilcox, 2001; Wicks and Hitch, 2002; Kannan and Ganesh, 2003 and Sendhilvel *et al*., 2004). The efficiency of azoxystrobin against *Pythium aphanidermatum* in cucumber root rot (Utkhede and Bogdanoff, 2003), *Claviceps africana* in sorghum ergot (Prom and Isakeit, 2003) and *Fusarium oxysporium* f.sp. *dianthi* in carnation wilt (Gullino *et al*., 2002) were also reported. The compound also appeared to be effective against *Fusarium moniliforme* (sheath rot of rice), *Drechslera oryzae* (brown leaf spot) and *Aspergillus niger* (collar rot of groundnut) (Thind *et al*., 2002).

Like azoxystrobin, difenoconazole was also reported to be highly effective at lower doses (0.05%) as it reduced the disease index of powdery mildew of mango (97.9% over control) when compared with other conventional fungicides (Raj and Badiyala, 2000). Difenoconazole at 0.015% and hexaconazole

at 0.10% were also reported effective against powdery mildew of pea incited by *Erysiphe polygoni* (Gupta and Shayam, 1998). Generally all the triazole fungicides performed better as compared to conventional fungicides (Dhruj *et al*., 2000; Khunti *et al*., 2002). Evaluation of a number of triazole fungicides in recent years revealed that formulations containing cyproconazole, bitertanol, hexaconazole, propiconazole and tebuconazole were excellent in controlling blister blight of tea (Premkumar *et al*., 1998). The efficacy was improved when triazoles were used in combination with copper oxychloride (Chandramouli and Agnihothrudu, 1989).

Strobilurin fungicides were reported to be effective at very low concentrations, say 12-20 μg mL^{-1} (Wong and Wilcox, 2002; Miller and Gubler, 2004). However, in the present study, concentrations of azoxystrobin as high as 250 mg mL^{-1} were used for control of diseases. This may be attributed to the possible development of resistance of this fungicide in the grapevine and tea pathogens, which were receiving other strobilurin fungicides for the past four years. Cross resistance for a number of QoI fungicides by pathogens is already known in literature (Chen *et al*., 2001; Steinfeld *et al*., 2001; Bartlett *et al*., 2002; Vincelli and Dixon, 2002; Avila-Adame and Koller, 2003; Karaoglanidis and Thanassoulopoulos, 2003; McGrath and Shishkoff, 2003a, b). Hence, close monitoring of the potential development of resistance is prudent to prevent disease control failure.

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