



FIELD EVALUATION OF CHEMICAL AND ECO FRIENDLY STRATEGIES AGAINST POWDERY MILDEW OF MULBERRY CAUSED BY *PHYLLACTINIA CORYLEA*

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ABSTRACT

Mulberry powdery mildew caused by *Phyllactinia corylea* is one of the important catastrophic diseases. The efficacy of fungicides, plant extracts and bio-control agents were assessed against powdery mildew of mulberry under field condition. The fungicides tebuconazole 50% + trifloxystrobin 25% WG and hexaconazole 5% EC at 3% concentration were found to be more effective and reduced the disease to 74.76% and 66.28% respectively over control. The plant extracts at all concentrations tested were effective in suppressing the disease and at higher concentration of 15%, *Zinger officinale* reduced the disease to 60.31 per cent and *Lantana camara* to 59.04 per cent disease over control and considered as best for managing powdery mildew disease in mulberry. Among the culture filtrate of different bioagents tested, the *Trichoderma harzianum* reduced the disease to 51.56 per cent and *Pseudomonas fluorescens* to 51.09 per cent over control at 15% concentration and were shown to be potent in fading away the disease.

KEY WORDS: Powdery mildew; *Phyllactinia corylea*; tebuconazole + trifloxystrobin; *Zinger officinale*; *Trichoderma harzianum*; *Pseudomonas fluorescens*

INTRODUCTION

Mulberry (*Morus alba*) is a perennial plant belongs to the family *Moraceae*-the food plant of silkworm (*Bombyx mori* L.). It is cultivated in both tropical and temperate countries of the world located from north of the equator between 28°N and 55°N latitude. Though mulberry cultivation is practiced in various climates in India, it is extensively grown in the tropical zone covering Karnataka, Andhra Pradesh and Tamil Nadu states with about 90 percent of area where, most of the sericulture industry is concentrated. In the sub-tropical zone, West Bengal, Himachal Pradesh and the northeastern states have major areas under mulberry cultivation. Powdery mildew is caused by an obligate biotrophic ascomycete fungus, *Phyllactiniacorylea* (syn. *P. guttata* and *P. moricola*) throughout the world (Chattopadhyay *et al.*, 2011). The disease is characterized by white dust-like mycelia that develop over abaxial (lower) leaf surfaces. The heavily infected tissues develop chlorosis on the adaxial (upper) surface of leaves and senescence prematurely (Gupta, 2001) with drastic yield reduction. Powdery mildews and other obligate biotrophic pathogens are highly adapted to the host mulberry and reduce leaf quality and yield which in turn affect the silkworm health and cocoon quality and weight. Further, the information on the management of powdery mildew and availability of safest and residue free fungicide on mulberry leaves and silkworms is inadequate. Therefore, there is a need to evaluate effective and safe fungicides and eco-friendly methods against mulberry powdery mildew disease.

MATERIALS & METHODS

A field experiment was conducted during 2016 at College of Agriculture, V.C. Farm, Mandya to study the efficacy of fungicides, plant extracts and bio agents against powdery mildew by using the variety V-1 mulberry in Rabi season. The plot size was 37.2m X 4.2m and treatments maintained in three replication, with the statistical design of RCBD. All the agronomic practices were carried out as per the package of practices recommended for mulberry cultivation by UAS, Bangalore. The contact fungicides *viz*: Wettable sulphur 80% WP, Mancozeb 75% WP, systemic fungicides *viz*: Carbendazim 50% WP, Hexaconazole 5% EC and Difenconazole 25% EC and the Combi-products *viz*: Tebuconazole 50% +Trifloxystrobin 25% WG, Carbendazim 12% + Mancozeb 63% WP were tested at 0.1, 0.2 and 0.3 per cent concentrations against the disease by spraying twice before the occurrence of the disease at 20 days interval.

Preparation of extracts of medicinal plants

Different medicinal plants *viz*: Neem leaf, Onion bulb, Ginger rhizome, Sweet sledge dried root, Garlic bulb and Lantana leaf were collected and aqueous extracts prepared. 500g of plant material were taken and cut into small pieces. The sample was put into waring blender containing 500ml sterilized distilled water at a ratio 1:1 (water: plant material). The sample was spun at low speed for 10-15 minutes in a waring blender till the material formed to fine texture. The blended material was then squeezed through a sterilized muslin cloth so as to get a crude liquid extract. The crude extract was filtered through whatman no 1 filter

paper. The filtrate obtained served as a stock solution of 100 per cent concentration and was further diluted to 5, 10 and 15 percent concentrations with water and sprayed twice uniformly at 20 days interval on the mulberry leaves before occurrence of the powdery mildew disease.

Preparations of culture filtrate of bio agents

The cultures of fungal bio control agents *Trichoderma viride* and *Trichoderma harzianum* and bacterial bio agents *Pseudomonas fluorescens* and *Bacillus subtilis* were obtained from the NBAIR, Bengaluru. The fungal and bacterial bio agents were multiplied on potato dextrose and nutrient broth respectively contained in 2 litre capacity flat bottom round flasks incubated at room temperature by regular shaking. Ten day old fungal bio agent and two days old bacterial bio agent cultures from the broth were filtered through Whatman no.1 filter paper. The filtrate obtained served as a stock solution of 100 per

cent concentration and were further diluted to 5, 10 and 15 percent concentrations by adding sterilized distilled water. The different concentrations of culture filtrates of both the bio agents were uniformly sprayed on the surface of mulberry leaves twice at 20 days interval before onset of the disease. The powdery mildew severity was recorded before and after spraying using 0-10 scale. Control treatment was maintained by water spraying. Observations were made one day before and five, ten and fifteen days after spray on disease severity by using scale given by Horsfall and Cowling (1978) for all the treatments (Table 3). From each treatment four plants were selected randomly and from each plant, three branches, each from top, middle and bottom canopy was selected to record disease severity following 0-10 scale. The percent disease control was calculated by the formula:

$$\text{Per cent disease control (\%)} = \frac{\text{PDI in control} - \text{PDI in treatment}}{\text{PDI in control}} \times 100$$

RESULTS & DISCUSSION

In the present field investigation, it was found that tebuconazole 50%+ trifloxystrobin 25% WG at 0.3% showed a significant effect, as it reduced the disease by 74.76% over control. The next best fungicide observed for their efficacy under field conditions was hexaconazole 5% EC at 0.3 % concentration which showed 66.28 % disease reduction over control, followed by tebuconazole 50% + trifloxystrobin 25% WG at 0.2 per cent concentration with the reduction of 62.39 %. Difenconazole 25% EC at 0.3 % concentration with the disease reduction of 60.81 per cent, carbendazim 50% WP at 0.3 % concentration with 60.67 % disease control, wettable sulphur 80% WP with 64.52 % at 0.3 per cent concentration and carbendazim 12%+ mancozeb 63% WP 63.97 per cent at 0.3 % concentration were most effective in managing powdery mildew disease in mulberry (Table 1). The similar results were reported by Shibao *et al.* (2013), Khunti *et al.* (2005) and Govindaiah *et al.* (1994).

Field evaluation of plant extracts against powdery mildew of mulberry

The aqueous extracts of six medicinal plant species viz., *Azadirachta indica* (leaf), *Albizia leppa* (bulb), *Zingiber officinale* (rhizome), *Acorus calamus* (rhizome), *Allium sativum* (clove) and *Lantana camera* (leaf) were tested to assess their efficacy on management of powdery mildew of mulberry (*Morus indica*) caused by *Phyllactinia corylea* under field condition. All the plant extracts tested at three different concentrations reduced the percent powdery mildew disease of mulberry. In all the cases spraying with higher extract concentrations gave better disease control. Among the plant extracts, the highest value of PDC (60.31%) was obtained with spraying of 15 per cent rhizome extract of *Zingiber officinale* by reducing the PDI value from 62.85 to 23.99 per cent at six days after second spray. Leaf, rhizome and clove extracts of *Lantana camera*, *Acorus calamus* and *Allium sativum* at 15 per cent concentrations gave 59.04, 53.32 and 52.19 per cent disease control (PDC) respectively at six days after second

spray (Table 2). Thus, the four plant extracts were most effective in the management of powdery mildew of mulberry although the other extracts also reduced the disease. In untreated control plants the PDI was maximum (75.37 to 60.45%) during 14 days compared to all other treatments. Among the treated plants less infected mulberry leaves was observed and gradually turning into healthy appearance after the spraying with effective plant extracts indicating the curative effect in reducing the disease. The present results are in confirmatory with Tiken *et al.* (2015), Malhotra and Singh, (2003) and Vadivel and Ebenezer (2006).

Field evaluation of bio control agents against powdery mildew of mulberry

Biological control, as a crop protection strategy has emerged as a response to the search for a safe, effective and environmentally friendly approach to replace or supplement the use of chemical pesticides. Biological control of plant diseases involves the use of antagonistic microorganisms to control a pathogen. Over the past three decades, research has repeatedly demonstrated that many microorganisms can act as natural antagonists to plant pathogens (Cook, 2000). Keeping this in background the use of four biocontrol agents viz., *Trichoderma viridae*, *Trichoderma harzianum*, *Bacillus subtilis* and *Pseudomonas fluorescens* were tested against powdery mildew of mulberry.

In the present study it was found that all the bio control agents have reduced the disease at higher concentration of 15 per cent. The foliar sprays of *Trichoderma harzianum* at higher concentration have reduced the maximum disease when compared to other three bio control agents. The maximum per cent disease control of 51.56 per cent was observed in *Trichoderma harzianum* and the next best biocontrol agent observed for their efficacy against powdery mildew disease was *Pseudomonas fluorescens* with 51.09 per cent PDC followed by *Trichoderma viridae* with 45.90 per cent disease control (Table 3).

TABLE 1. Efficacy of fungicides against powdery mildew of mulberry (*P. corylea*)

Tr	Conc. (%)	Per cent disease index			PDC	
		Before spray	At 5 DAS	At 10 DAS		At 15 DAS
T ₁	0.1	53.64 (47.08)	38.49 (38.34)	30.57 (33.56)	22.03 (27.99)	56.53 (48.75)
T ₂	0.2	49.67 (44.81)	37.42 (37.71)	27.82 (31.83)	21.57 (27.67)	57.44 (49.27)
T ₃	0.3	44.17 (41.65)	35.91 (36.81)	26.67 (31.09)	19.93 (26.51)	60.67 (51.15)
T ₄	0.1	44.83 (42.03)	37.56 (37.79)	35.56 (36.60)	23.42 (28.94)	53.79 (47.17)
T ₅	0.2	46.33 (42.89)	35.17 (36.37)	27.42 (31.57)	20.50 (26.92)	59.55 (50.50)
T ₆	0.3	47.93 (43.81)	32.50 (34.75)	31.87 (34.37)	20.11 (26.64)	60.32 (50.95)
T ₇	0.1	41.67 (40.20)	39.90 (39.17)	32.37 (34.67)	24.30 (29.53)	52.05 (46.17)
T ₈	0.2	40.83 (39.71)	33.33 (35.26)	25.10 (30.06)	19.06 (25.88)	62.39 (52.17)
T ₉	0.3	40.83 (39.71)	30.85 (33.74)	23.42 (28.94)	12.79 (20.95)	74.76 (59.83)
T ₁₀	0.1	44.17 (41.65)	40.21 (39.35)	38.17 (38.15)	26.49 (30.97)	47.73 (43.69)
T ₁₁	0.2	44.67 (41.93)	35.44 (36.53)	30.14 (33.29)	21.14 (27.37)	58.29 (49.77)
T ₁₂	0.3	38.33 (38.25)	33.80 (35.54)	30.83 (33.72)	20.42 (26.86)	59.71 (50.59)
T ₁₃	0.1	43.00 (40.97)	40.46 (39.50)	37.54 (37.78)	33.38 (35.29)	34.14 (35.75)
T ₁₄	0.2	38.33 (38.25)	35.38 (36.49)	33.33 (35.26)	24.43 (29.62)	51.80 (46.03)
T ₁₅	0.3	44.17 (41.65)	39.77 (39.09)	32.00 (34.45)	17.09 (24.42)	66.28 (54.49)
T ₁₆	0.1	43.33 (41.16)	40.79 (39.69)	36.32 (37.06)	29.76 (33.06)	41.28 (39.97)
T ₁₇	0.2	38.33 (38.25)	34.41 (35.91)	33.15 (35.15)	22.43 (28.26)	55.74 (48.29)
T ₁₈	0.3	38.33 (38.25)	32.36 (34.67)	32.62 (34.83)	19.86 (26.46)	60.81 (51.24)
T ₁₉	0.1	40.00 (39.23)	37.38 (37.69)	34.70 (36.09)	38.68 (38.45)	23.68 (29.11)
T ₂₀	0.2	48.33 (44.04)	44.28 (41.71)	40.58 (39.57)	26.07 (30.70)	48.56 (44.17)
T ₂₁	0.3	44.17 (41.65)	39.25 (38.79)	37.56 (37.79)	24.12 (29.41)	52.41 (46.38)
T ₂₂	-	60.77 (51.21)	56.48 (48.72)	52.43 (46.39)	50.68 (45.38)	0.00 (0.00)
S.E.m±			1.72	1.00	0.50	1.72
CD @ P=0.05		NS	4.92	2.86	1.53	4.90

Tr= Treatment; PDC= Per cent disease control; NS= Non-significant; Figures in the parenthesis are Arc sine transformed values;

Treatment details: T₁, T₂ and T₃=Carbendazim50% WP ; T₄, T₅ and T₆= Wettable sulphur 80% WP; T₇, T₈ and T₉= Tebuconazole 50% + Trifloxystrobin 25% WG
 T₁₀, T₁₁ and T₁₂= Carbendazim 12% + Mancozeb 63% WPT₁₃, T₁₄ and T₁₅= Hexaconazole 5% EC T₁₆, T₁₇ and T₁₈= Difenconazole 25% EC
 T₁₉, T₂₀ and T₂₁= Mancozeb 75% WP ;T₂₂= Control (Water spray)

TABLE 2. Efficacy of plant extracts against powdery mildew of mulberry (*P. corylea*)

Tr	Conc. (%)	Before spray	PDI after 1 st spray			PDI after 2 nd spray			PDC
			At 2 days	At 4 days	At 6 days	At 2 days	At 4 days	At 6 days	
T1	5	56.78(48.89)	56.57(48.77)	53.88(47.22)	51.18(45.67)	48.28(44.01)	43.01(40.98)	36.43(37.12)	39.74(39.07)
T2	10	62.56(52.27)	55.70(48.27)	53.33(46.90)	50.26(45.14)	45.26(42.27)	38.95(38.61)	32.77(34.92)	45.79(42.58)
T3	15	60.67(51.15)	57.41(49.25)	49.00(44.42)	46.45(42.96)	40.05(39.26)	32.67(34.86)	30.79(33.70)	49.07(44.46)
T4	5	66.67(54.73)	61.57(51.68)	57.37(49.23)	54.78(47.74)	50.77(45.44)	43.07(41.01)	36.05(36.89)	40.36(39.44)
T5	10	63.33(52.72)	59.47(50.45)	56.61(48.79)	49.25(44.56)	46.35(42.90)	40.96(39.79)	33.99(35.66)	43.77(41.42)
T6	15	69.89(56.71)	57.88(49.53)	54.45(47.55)	47.54(43.58)	42.83(40.87)	36.12(36.94)	31.95(34.41)	47.15(43.36)
T7	5	56.67(48.83)	54.85(47.78)	52.11(46.20)	48.36(44.05)	44.33(41.74)	40.07(39.27)	33.71(35.49)	44.23(41.68)
T8	10	57.71(49.43)	50.66(45.37)	47.39(43.50)	43.47(41.24)	39.76(39.09)	32.11(34.51)	25.22(30.14)	58.28(49.76)
T9	15	62.85(52.44)	50.67(45.38)	46.78(43.15)	40.40(39.46)	34.08(35.71)	29.72(33.03)	23.99(29.32)	60.31(50.94)
T10	5	67.33(55.13)	60.13(50.84)	57.16(49.11)	51.81(46.03)	48.25(43.99)	40.13(39.30)	34.49(35.96)	42.94(40.94)
T11	10	63.33(52.72)	60.48(51.04)	53.00(46.71)	50.39(45.22)	46.69(43.10)	37.05(37.49)	30.94(33.79)	48.82(44.32)
T12	15	62.19(52.05)	58.37(49.81)	50.93(45.53)	46.72(43.11)	40.06(39.26)	33.11(35.12)	28.22(32.08)	53.32(46.90)
T13	5	65.20(53.84)	56.85(48.93)	53.25(46.86)	49.18(44.52)	45.13(42.20)	39.32(38.83)	34.48(35.95)	42.96(40.95)
T14	10	64.09(53.18)	62.71(52.36)	56.19(48.55)	50.33(45.18)	45.86(42.62)	36.52(37.17)	30.03(33.23)	50.32(45.18)
T15	15	70.00(56.78)	66.66(54.72)	54.50(47.58)	46.64(43.07)	41.64(40.18)	32.77(34.92)	28.90(32.52)	52.19(46.25)
T16	5	64.39(53.36)	61.17(51.45)	55.83(48.34)	49.15(44.51)	45.44(42.38)	39.36(38.85)	30.42(33.47)	49.68(44.81)
T17	10	52.12(46.21)	50.56(45.31)	52.38(46.36)	46.23(42.83)	41.17(39.91)	35.57(36.61)	26.89(31.23)	55.52(48.16)
T18	15	63.93(53.08)	50.84(45.48)	52.24(46.28)	44.87(42.05)	38.83(38.54)	30.07(33.25)	24.76(29.84)	59.04(50.20)
T19	-	75.37(60.24)	73.25(58.85)	71.83(57.94)	71.02(57.42)	68.53(55.87)	63.39(52.76)	60.45(51.03)	0.00 (0.00)
S.E _{mt}			1.75	1.59	1.45	1.32	1.34	1.12	1.82
CD @ P=0.05		NS	5.01	4.55	4.16	3.80	3.85	3.20	5.22

Tr= Treatment; PDI= per cent disease index; PDC= Per cent disease control;

NS= Non-significant; Figures in the parenthesis are Arcsine transformed values

Treatment details: T₁, T₂ and T₃ = *Azadirachta indica* T₄, T₅ and T₆ = *Allium cepa*; T₇, T₈, and T₉ = *Zingiber officinale* T₁₀, T₁₁ and T₁₂= *Acorus calamus*;

T₁₃, T₁₄ and T₁₅ = *Allium sativum*, T₁₆, T₁₇ and T₁₈ = *Lantana camara*; T₁₉ = Control (Water spray)

TABLE 3. Efficacy of biocontrol agents/filtrate against powdery mildew of mulberry (*P. corylea*)

Tr	Conc. (%)	Before		PDI after 1 st spray			PDI after 2 nd spray			PDC
		At 2 days	At 4 days	At 6 days	At 2 days	At 4 days	At 6 days			
T1	5	58.89 (50.11)	56.87 (48.94)	54.26 (47.44)	50.05 (45.02)	47.37 (43.49)	41.72 (40.23)	34.40 (35.91)	31.88 (34.37)	
T2	10	56.67 (48.83)	53.55 (47.03)	50.61 (45.34)	45.00 (42.12)	41.08 (39.86)	41.43 (40.06)	34.51 (35.97)	31.66 (34.24)	
T3	15	56.67 (48.83)	51.36 (45.77)	48.30 (44.02)	45.37 (42.34)	48.30 (44.02)	34.52 (35.98)	27.32 (31.51)	45.90 (42.64)	
T4	5	65.56 (54.06)	61.39 (51.58)	58.47 (49.87)	54.10 (47.35)	50.48 (45.27)	43.35 (41.17)	29.36 (32.81)	41.86 (40.31)	
T5	10	53.40 (46.94)	50.67 (45.38)	46.20 (42.82)	42.22 (40.52)	46.40 (42.93)	32.56 (34.79)	24.70 (29.80)	51.09 (45.62)	
T6	15	55.56 (48.19)	49.45 (44.68)	45.33 (42.31)	40.30 (39.40)	38.30 (38.23)	32.38 (34.68)	24.46 (29.64)	51.56 (45.89)	
T7	5	60.00 (50.76)	57.71 (49.43)	55.70 (48.27)	52.41 (46.38)	48.66 (44.23)	46.28 (42.86)	36.43 (37.12)	27.86 (31.85)	
T8	10	55.56 (48.19)	52.45 (46.40)	50.25 (45.14)	46.96 (43.25)	41.23 (39.94)	42.38 (40.61)	28.28 (32.12)	44.00 (41.55)	
T9	15	57.78 (49.47)	51.40 (45.80)	49.61 (44.77)	44.44 (41.80)	40.43 (39.48)	34.93 (36.22)	27.68 (31.74)	45.19 (42.23)	
T10	5	62.22 (52.07)	60.14 (50.84)	58.48 (49.88)	55.13 (47.94)	52.22 (46.27)	40.83 (39.71)	39.60 (38.99)	21.58 (27.68)	
T11	10	60.00 (50.76)	58.22 (49.72)	55.41 (48.10)	51.11 (45.63)	47.86 (43.77)	39.47 (38.92)	33.34 (35.26)	33.98 (35.65)	
T12	15	63.33 (52.72)	57.38 (49.24)	53.36 (46.92)	50.22 (45.12)	45.03 (42.14)	37.63 (37.83)	33.62 (35.43)	33.43 (35.32)	
T13	-	67.78 (55.41)	65.46 (54.00)	63.12 (52.60)	62.60 (52.29)	58.43 (49.85)	52.24 (46.28)	50.50 (45.28)	0.00 (0.00)	
S.E _{mt}		1.19	1.60	1.15	1.60	1.31	1.14	1.72		
CD @ P=0.05		NS	3.48	4.67	3.35	4.67	3.83	3.31	5.02	

Tr= Treatment; PDI= per cent disease index; PDC= Per cent disease control; NS= Non-significant;

Figures in the parenthesis are Arc sine transformed values

Treatment details: T₁, T₂ and T₃ = *Trichoderma viridae*; T₄, T₅ and T₆ = *Trichoderma harzianum*; T₇, T₈ and T₉ = *Pseudomonas fluorescens*

T₁₀, T₁₁ and T₁₂ = *Bacillus subtilis*; T₁₃ = Control (Water spray)

The highest per cent disease control by *Trichoderma harzianum* this might be due to antagonistic ability against the pathogen which inhibit the spore germination by producing hydrolytic enzymes. The present findings were in close conformity with Biswas *et al.* (2000) and Philip *et al.* (2000), who reported that at higher filtrate concentration of *Trichoderma harzianum* and *Trichoderma viridae* had reduced the disease intensity of powdery mildew of mulberry. The lowest per cent disease control of 21.58 per cent was recorded in *Bacillus subtilis* this might be due to less favourable climate to show its antagonistic ability against the pathogen in the prevailing mulberry ecosystem.

The present investigation identified the potential fungicides, botanicals and bio agents which were effective in suppressing the mulberry powdery mildew disease. The finding could be exploited to formulate eco friendly cost effective integrated management strategy for this disease.

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