

Control Measures for Leaf Fall Disease in Clove (*Syzygium aromaticum* L.) in Southern Sri Lanka

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Introduction

Among several diseases recorded in Clove plantations, leaf fall disease is severe. Although the disease has been recorded in mid country region (Jayasinghe and Liyanage, 1982) in earlier and currently, it is distributed throughout the southern region. Identical purplish brown color spots are noticed when fungus is infected and gradually turn them into brown color. Severely infected leaves are withered, dropped and dried up and cause sudden death of plants finally. There are no proper cultural or agronomic practices to manage the situation. Therefore, this study was carried out to confirm the causal agent of disease and find out most suitable fungicide and application method to manage the disease in field plants.

Methodology

Leaves with identical lesions collected from Urubokka and Thihagoda areas in Southern Sri Lanka were used to identification of causal organism of leaf fall disease of clove at the Plant Pathology Laboratory, Cinnamon Research Station, Matara. Infected leaves were surface sterilized by submerging in 10% Chlorox for thirty seconds and rinsed in five changes of sterilized distilled water and placed on the Water Agar medium. The plates were incubated at 30 °C until sporulation occurred and sub cultured on Potato Dextrose Agar (PDA) medium. Isolated fungal colonies were used to examine colony morphology and microscopic characteristics for identification of organism. Field experiments were under taken in clove (*S. aromaticum* L.) field at Urubokka, Matara. Mature plants naturally infested with disease were selected and level of infection was more or less similar in all selected plants. Three common fungicides namely Tebuconazole 200 mg/ml, Thiophanate methyl 70% and Hexaconazole 50 g/l were tested with using water as control and three different application methods namely foliar application, bark application and root application with five replicates. Treatment procedure was carried out three times with two weeks interval. Foliar applications were done by using modified power sprayer. Bark applications of liquid fungicides was done through a hole made in stem by using saline bottles and solid fungicide (Thiophanate methyl) was directly applied to xylem tissue via 2 x 3 cm cuts made on bark of stem. Watering can was used for root application method. Disease incidences and severity were recorded in existing and newly emerging leaves.

Results and Discussion

On Bacteriological Agar (BA), two weeks old fungal cultures were grown about 50 mm diameter with submerged mycelia growth. Centre of colony was dark brown in color and becomes slightly light brown on periphery. Cultures were gelatinous, flattened with

submerged profusely branched mycelia strands with numerous microsclerotia. Microscope observations showed that the dimensions of mycelia, microsclerotia were agreed with original description of *Cylindrocladium* spp (Boedijn and Reitsma, 1950). Conidiophore branches were arisen laterally from a stipe (Jayasinghe *et al.*, 2009). Sterile filaments terminate in a narrowly clavate vesicle. Leaves inoculated with *Cylindrocladium* spp developed lesions few days after inoculation (Plate 01).

Minimum number of infected leaves was observed in treatment A₃ (Tebuconazole 200 mg/ml) and it was statistically difference from other treatments A₁ (Hexaconazole 50 g/l), A₂ (Thiophanate-methyl 70%) and A₄ (Control) and same trend was observed in development of lesions (surface area) in initially infected leaves and initially uninfected. Maximum percentages of leaf fall were observed in treatment A₃ (Tebuconazole 200 mg/ml) and A₁ (Hexaconazole 50 g/l) whereas minimum leaf fall were observed in A₂ (Thiophanate-methyl 70%) and A₄ (control), but they were significantly different from A₃ (Table 01). There was no clear relationship between leaf fall and disease incidences (Table 01) because some physical factors such as wind and rain fall that also caused leaf fall during the study period. There was significant difference in growth of disease over the time and minimum rate of disease development was observed in A₃ (Tebuconazole 200 mg/ml) treatment.



Plate 01: Pathogen grown on BA media (A) and microscopic view of pathogen (B)

Table 01: Mean number of infected leaves remaining (percentage), lesion developed (mm²) in initially infected leaves and initially uninfected leaves with percentage of leaf fall under different fungicide applications after 3 months (A₁:Hexaconazole, A₂: Thiophanate-methyl, A₃:Tebuconazole, A₄: Control).

Treatment (A)	Number of infected leaves remaining (%)	Surface area of lesions in initially infected leaves (mm ²)	Leaf fall percentage	Surface area of dark lesions in initially Uninfected leaves (mm ²)
A ₁	82.513 ^a	35.278 ^a	34.995 ^{ab}	13.359 ^a
A ₂	82.733 ^a	38.080 ^a	30.040 ^b	9.411 ^{ab}
A ₃	66.113 ^b	24.600 ^b	44.235 ^a	6.133 ^b
A ₄	90.980 ^a	31.978 ^a	25.624 ^b	12.080 ^a
CV	17.97	36.01	41.18	59.13
F value	7.76	3.71	4.96	4.18
P	0.0003	0.0177	0.0044	0.0105
MSE	14.487	11.698	13.886	6.058
LSD	10.637	8.589	10.196	4.448

(Mean values in each column superscripted by the same letters are not significantly different at $p < 0.05$)

Minimum numbers of infected leaves and leaf fall were observed in B₁ (foliar application) and B₂ (bark application) treatments than B₃ (root application). Development of disease lesion in initially infected leaves was lower B₂ (Bark application) and B₃ (root application)

than B₁ (foliar application) significantly while development of disease lesion in initially uninfected leaves was lowest in B₁ (foliar application) (Table 02). It was statistically different from B₃ (root application), but on par with B₂ (bark application) (Table 02). There were no significant difference was observed in interactions between two factors (fungicide and its application method) tested to control disease. But treatment B₁ (foliar application) of treatment A₃ (Tebuconazole 200 mg/ml) and treatment B₂ (bark application) of treatment A₃ (Tebuconazole 200 mg/ml) were the most effective combinations of fungicide and its application method to manage leaf fall disease in *S. aromaticum* (Figure 01).

Table 02: Mean number of infected leaves remaining (percentage), lesion developed (mm²) in initially infected leaves and initially uninfected leaves with percentage of leaf fall under different fungicide application methods (B1:Foliar Application, B2: Bark Application, B3:Root Application).

Treatment (A)	Number of infected leaves remaining (%)	Surface area of lesions in initially infected leaves (mm ²)	Leaf fall percentage	Surface area of dark lesions in initially Uninfected leaves (mm ²)
B ₁	75.195 ^b	38.800 ^a	32.492 ^{ab}	7.050 ^b
B ₂	73.735 ^b	28.618 ^b	29.118 ^b	7.401 ^b
B ₃	92.825 ^a	30.033 ^b	39.562 ^a	16.287 ^a
CV	17.98	36.01	41.18	59.13
F value	10.76	4.45	2.95	14.93
P	0.0001	0.0169	0.0621	0.0001
MSE	14.487	11.698	13.886	6.058
LSD	9.2116	7.4381	8.8296	3.8521

(Mean values in each column superscripted by the same letters are not significantly different at p < 0.05)

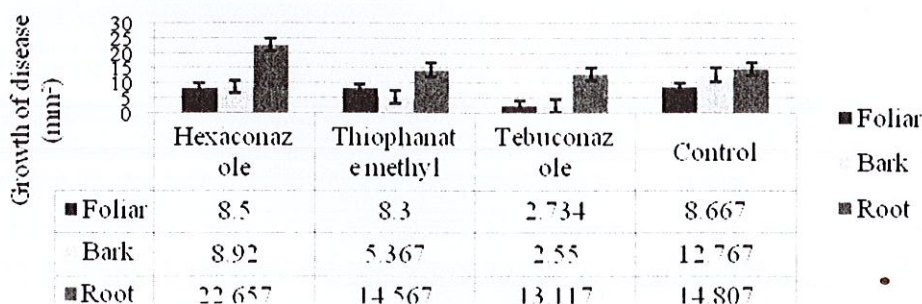


Figure 01: Development of surface area of lesions due to infection with different fungicide and their application methods

Conclusion

According to the morphology of fungus, causal agent of leaf fall disease in *Syzygium aromaticum* in southern Sri Lanka is *Cylindrocladium spp.* but species level identification is remaining. Most effective fungicide and application methods to manage leaf fall disease in *Syzygium aromaticum* at severely and slightly infected stages are foliar application of Tebuconazole 200 mg/ml. Bark application is also effective method to leaf fall disease in tall clove plants.

Acknowledgement

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References

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Treatment (A)	Number of infected leaves remaining (%)	Surface area of lesions in initial (mm ²)	Surface area of lesions in final (mm ²)	Percentage reduction in lesions	Surface area of bark (mm ²)
B ₁	73.192	38.800	32.402	16.02%	7.020
B ₂	73.725	38.618	30.118	21.21%	7.401
B ₃	67.825	30.032	20.502	31.52%	16.282
CV	17.98	16.01	41.18		20.13
F value	10.76	4.42	2.97		14.93
P	0.0001	0.0169	0.0021		0.0001
MSE	11.487	11.098	13.886		6.038
FSD	9.216	7.181	8.296		3.821

(Mean values in each column superscripted by the same letters are not significantly different at p < 0.05)

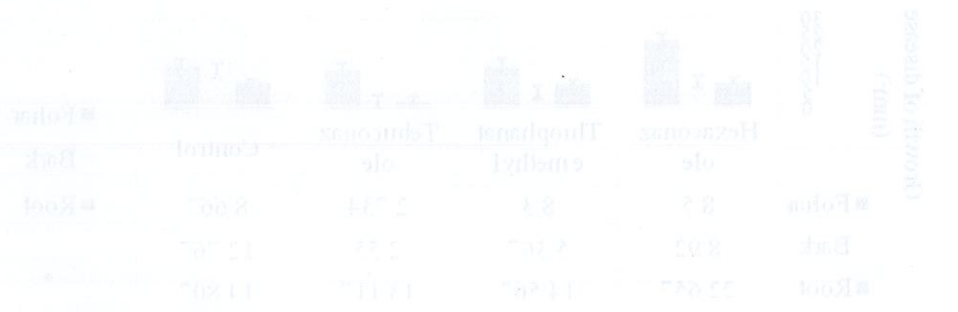


Figure 01: Development of surface area of lesions due to infection with different fungicides and their application methods

Conclusion

According to the morphology of fungus, causal agent of leaf fall disease in *Syzygium cumatum* in southern Sri Lanka is *Cylindrocladium* spp. but species level identification is remaining. Most effective fungicide and application method to manage leaf fall disease in *Syzygium cumatum* at severely and slightly infected stages are foliar application of Tebuconazole 300 mg/ml. Bark application is also effective method to leaf fall disease in all clove plants.