

Diseases of Phalaenopsis : Symptoms, Etiology and Management

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Abstract – A survey was conducted in Thrissur district, Kerala to study various diseases of Phalaenopsis. Wilt, collar rot, flower spot, anthracnose and soft rot were the diseases observed. The isolation of pathogens from the infected specimens yielded *Fusarium oxysporum*, *Sclerotium rolfsi*, *Phoma exigua*, *Colletotrichum gloeosporioides* and *Erwinia chrysanthemi* respectively. The symptoms of the diseases were recorded, pathogens were isolated and pathogenicity was proved by standard procedures. The cultural and morphological characters of the pathogens were also studied. Five chemical fungicides and *Pseudomonas fluorescens* were evaluated *in vitro* by poisoned food technique against the fungal pathogens. All fungicides except cymoxanil 8 % + mancozeb 64 % and carbendazim 12 % + mancozeb 63 % showed cent per cent inhibition of all pathogens except *Phoma exigua*. The per cent inhibition of *P. fluorescens* 0.5 % was varied from 86.30 % against *C. gloeosporioides* to 52.97 % against *P. exigua*. The inhibitory effect of Streptocycline 200 ppm, fresh cowdung extract 2%, *P. fluorescens* (0.5%) and fresh cowdung extract + *P. fluorescens* (2% + 0.5%) were evaluated *in vitro* by agar well diffusion method against the bacterial pathogen *E. chrysanthemi* causing soft rot in Phalaenopsis. The maximum inhibition (65.56 %) was recorded in *P. fluorescens* and the minimum inhibition (30.74 %) in Streptocycline. Fresh cowdung extract + *P. fluorescens* and fresh cowdung extract recorded 57.77 and 55.92 per cent inhibition respectively and this was on par with *P. fluorescens*. Streptocycline showed the least inhibition (30.74 %) of the pathogen. Lysis of pathogenic bacteria was observed in Streptocycline where as inhibition of pathogenic bacteria by growth of antagonistic bacteria was observed in other treatments.

Keywords – Anthracnose, Collar Rot, Flower Spot, Fusarium Wilt, *In Vitro* Evaluation, Phalaenopsis Diseases, Soft Rot.

I. INTRODUCTION

Orchids are well known throughout the world for their beautiful, myriad shaped, multi coloured and long lasting quality flowers and constitute an order of royalty in the world of ornamental plants. Orchids are abundant in tropical countries of South East Asia, South and Central America and South Africa. India is blessed with an abundance of orchid flora with Himalayas as their main habitat and others scattered in Eastern and Western Ghats. Based on the growth habit, orchids are classified as monopodials and sympodials. Monopodial orchids have a main stem which continue to grow year after year and sympodials have a main stem which terminate growth at the end of each season. Phalaenopsis or Moth orchid is a major type of monopodial orchid with a short stem and is highly susceptible to diseases. Although orchids are not naturally disease prone, over 100 diseases can attack them and some of them are major production constraints [5],

[13], [14], [3], [15], [9] and [11]. Diseases of orchids are caused by fungi, bacteria, and viruses. They are classified as leaf spots, flower blights and root, stem and pseudobulb rots etc. These diseases affect orchid production in Kerala and cause financial loss to growers. In Kerala, most of the orchid growers are non-conventional agriculturists and are unaware of plant disease and their management. As, literatures are very scanty on diseases of orchids occurring in Kerala, an investigation has been undertaken on symptoms, etiology and management of Phalaenopsis diseases.

II. MATERIALS AND METHODS

i) Symptoms and etiology - A purposive sampling survey was conducted in the orchidarium of AICRP on Floriculture improvement in the Department of Pomology and Floriculture, College of Horticulture, Vellanikkara as well as in the farms of selected orchid growers in Thrissur district to study the various diseases of Phalaenopsis prevalent in these areas. The symptoms of the diseases were recorded, pathogens were isolated and pathogenicity was proved by standard procedures. The cultural and morphological characters of the isolated pathogens were studied in Potato dextrose agar medium for fungi and Nutrient agar medium for bacteria.

ii) In vitro evaluation of chemicals and bio agents - The effectiveness of fungicides viz., potassium phosphonate 40 % + mancozeb 75 % (0.2 + 0.15 %), cymoxanil 8 % + mancozeb 64 % (0.2 %), carbendazim 12 % + mancozeb 63 % (0.15 %), carbendazim 25 % + ipridione 25 % (0.2 %), fenamidone 10 % + mancozeb 50 % (0.1 %) and bio control agent *P. fluorescens* (0.5 % liquid formulation) were tested against fungal pathogens using poisoned food technique [16] and per cent inhibition was worked out. Agar well diffusion method [10] was carried out to evaluate the effectiveness of antibiotic Streptocycline (200 ppm) and fresh cowdung extract (2%) and *P. fluorescens* (0.5%) against the bacterial pathogen *Erwinia chrysanthemi*.

III. RESULTS AND DISCUSSION

i) Symptoms and etiology- Diseases of Phalaenopsis observed during the survey were wilt, collar rot, flower spot, anthracnose and soft rot. The isolation of pathogens from the infected specimens of wilt, collar rot, flower spot, anthracnose and soft rot yielded *Fusarium oxysporum*, *Sclerotium rolfsi*, *Phoma exigua*, *Colletotrichum gloeosporioides* and *Erwinia chrysanthemi* respectively. All these pathogens were purified and maintained on PDA/ NA medium.

a. Fusarium Wilt

Under natural condition, symptoms were initiated as yellowing of leaves. Later the leaf base showed a black discolouration and the leaves got detached from the plant. The leaves appeared shriveled and ultimately the whole plant wilted and died.

Under artificial inoculation, the symptom appeared on the leaf as black spot after five days of inoculation. Even though leaf base and roots were inoculated, symptoms were not observed in these parts. Similar type of symptoms of root rot and basal rot of Phalaenopsis by *Fusarium* was reported earlier [7].

Cultural characters of pathogen in PDA medium were, sparse white mycelial growth followed by slight pink pigmentation. Mycelium was delicate and compact. Pink colour was more prominent on the reverse side of the Petri dish. The fungus completed full growth in Petri dish (nine cm diameter) in six days at room temperature ($26 \pm 2^\circ\text{C}$). The morphological features of the fungus were hyaline, branched, septate hyphae with $3.85 - 5.09 \times 24.01 - 39.33 \mu\text{m}$ size. Macroconidia were hyaline, straight to sickle shaped 3 - 5 septate, pointed ends with size $22.65 - 30.06 \times 3.32 - 4.41 \mu\text{m}$. Microconidia were hyaline, oval or ellipsoid, non septate with $5.71 - 9.22 \times 2.70 - 3.26 \mu\text{m}$ size.

b. Collar Rot

Under natural conditions, first symptom observed was yellowing of leaves. Later white mycelial growth was appeared and rotting took place. In the advanced stage, numerous light brown sclerotia were formed on the infected area and the leaf got detached from the plant. Upon artificial inoculation, the symptom started as water soaked spot and rotting was more prominent than yellowing compared to natural infection and resulted in complete rotting within five days. Light brown sclerotia were appeared on the infected plant within 10 days of inoculation.

Cultural characters of *Sclerotium rolfsii* in PDA medium were white and even sheet of aerial mycelium with clear mycelial strands with tufts of longer hyphae. Colony had a fast growth and attained nine cm growth in five days at room temperature. Sclerotial development was started at seven DAI as a white structure which later changed to light brown and then to dark brown colour. Sclerotium was round, smooth and shiny in appearance. Number of sclerotia produced in the culture after 20 days of incubation was in the range of 60 -100. The morphological features of the fungus were hyaline, septate, branched hyphae with $3.26 - 4.32 \times 74.97 - 92.56 \mu\text{m}$ size.

The symptoms of wilt caused by *Sclerotium rolfsii* in Dendrobium reported were [2], basal rot of pseudobulb, yellowing and detachment of leaves, development of mycelial growth and formation of numerous small brown coloured sclerotia. In the present study, the symptoms observed were similar to that of Dendrobium.

c. Flower Spot

Under natural conditions, flower spot caused by *Phoma exigua* was appeared as minute, brown coloured sunken spots on petals. Symptoms were prominent in white coloured flowers than flowers of other colour. In advanced

stage, these spots coalesced to form moderate sized brown lesion. At the centre of the lesion, pycnidia were appeared as black dots. Under artificial condition, symptoms were produced as minute brown to black coloured spots on flower petal after four days of inoculation.

Cultures of *Phoma exigua* was appeared initially as white mycelial growth which turned to greenish black on the fourth day of incubation and later to greyish black. Reverse side of Petri dish was black in colour. Fungus attained nine cm diameter growth in PDA medium by eight days at room temperature. The morphological features of the fungus were hyaline hyphae turned to brown, septate and branched and $2.58 - 4.83 \times 13.61 - 30.47 \mu\text{m}$ size. Conidia were hyaline, cylindrical with one end round and other end tapering aseptate with $4.27 - 5.80 \times 1.81 - 3.09 \mu\text{m}$ size. Even though *Phoma* sp. was reported as a pathogen of orchids [13], the details on the symptomatology and host range are not available. As per the literature, *Botrytis cinerea* is the common pathogen to cause flower spot in orchids [3].

d. Anthracnose

The symptoms of anthracnose were initiated as water soaked areas on upper surface and chlorotic spot on the corresponding lower surface of the leaf. Later it turned to brown to black irregular sunken spot with brown margin surrounded by prominent yellow halo. In severe cases, these spots coalesced together resulted in the blighting of leaf. Under artificial conditions, the symptom produced was same as that in the natural condition and was visible after nine days of inoculation. The spot appeared on the upper side of the leaf with a yellow halo.

Cultural characters of *Colletotrichum* sp. in PDA medium was cottony white mycelial growth which turned to greyish black colour with pink sporulation. Reverse side of the Petri dish appeared black. Fungus attained nine cm diameter growth in eight days at room temperature. Pinkish spore mass and acervuli appeared after 14 days of incubation. The morphological features of the fungus were hyaline hyphae changed to greyish white colour, branched with $2.53 - 5.01 \mu\text{m}$ width and septate at an interval of $12.53 - 42.66 \mu\text{m}$. Conidia were hyaline, cylindrical with round ends, aseptate with an oil globule and $11.15 - 13.42 \times 2.5 - 3.65 \mu\text{m}$ size. Conidiophores were club shaped, hyaline and non septate. Setae were dark brown, septate with swollen base and tapering apex.

The symptoms of *C. gloeosporioides* infection in Dendrobium are small restricted, black, sunken lesion on leaf base [13] and in Cymbidium are dark brown to black spots on the leaves, blighting of leaves and death of plants [12]. Even though sunken spots and blighting symptoms described in the above reports were similar to the symptoms of anthracnose in the present study, yellow halo of spots observed in Phalaenopsis was not reported by the earlier workers.

e. Soft Rot

Under natural conditions symptoms appeared were greenish water soaked spot on the leaves which later spread to entire leaf within two days and resulted in liquefaction of leaf tissues and rotting of entire leaf. A characteristic foul smell was also associated with rotting.

Upon artificial inoculation, the symptom observed were greenish water soaked spot after 24 h of inoculation and completes rotting within 48 h.

Cultural characters of *Erwinia* sp. in Nutrient Agar medium were, small greyish white to creamy white, smooth, round, glistening, slightly raised translucent colonies. Bacterium was gram-negative, non-spore forming straight rods with round ends and occurred in groups. The symptoms of soft rot in Phalaenopsis and *Dendrobium* reported [1] was water soaked rot which enlarged rapidly without yellow margin. According to [4], the symptoms of bacterial soft rot in Phalaenopsis were water soaking, liquefaction of leaf tissue and rotting of leaf. In the present study also, similar symptoms were observed along with a characteristic foul smell.

ii.a) In vitro evaluation of chemicals and bio agents against fungal pathogens

Five chemical fungicides and *Pseudomonas fluorescens* were evaluated *in vitro* against four fungal pathogens viz., *Fusarium oxysporum* (FO), *Sclerotium rolfsii* (SR), *Phoma exigua* (PE) and *Colletotrichum gloeosporioides* (CG) of Phalaenopsis. The result of the experiment is given in Table 1. All fungicides except cymoxanil 8 % + mancozeb 64 % and carbendazim 12 % + mancozeb 63 % showed cent per cent inhibition of all pathogens except *Phoma exigua*. The per cent inhibition observed in cymoxanil 8 % + mancozeb 64 % and carbendazim 12 % + mancozeb 63 % against *P. exigua* were 79.63 and 87.41 per cent respectively. The per cent inhibition of *P. fluorescens* was varied between 86.30 to 52.97 per cent. The maximum inhibition (86.30 %) was observed against *C. gloeosporioides* and the minimum inhibition (52.97 %) was observed against *P. exigua*. Uchida (1994) proved that *C. gloeosporioides* of orchid is highly sensitive to benomyl. In the present study also, benzimidazole fungicide carbendazim combinations showed cent per cent inhibition of the pathogen. Kotle and Raut (2007) studied the efficacy of fungicides for the management of *S. rolfsii* of orchids and found that mancozeb, difenoconazole and hexaconazole resulted in the total inhibition of the radial growth of the pathogen. In the present study also mancozeb combinations showed complete inhibition of radial growth of *S. rolfsii*.

ii.b) In vitro evaluation of chemicals and bio agents against bacterial pathogen

The inhibitory effect of Streptocycline 200 ppm, fresh cowdung extract 2%, *P. flouescens* (0.5%) and fresh cowdung extract + *P. fluorescens* (2 % + 0.5 %) to the bacterial pathogen *E. chrysanthemi* causing soft rot in Phalaenopsis is given in Table 2.

The per cent inhibition was varied from 65.56 % to 30.74 %. The maximum inhibition (65.56 per cent) was recorded in *P. fluorescens* (T₃) and the minimum inhibition (30.74%) in Streptocycline (T₁). Fresh cowdung extract 2 % + *P. fluorescens* 0.5 % (T₄) and fresh cowdung extract 2% (T₂) recorded 57.77 and 55.92 per cent inhibition respectively and this was on par with *P. fluorescens* (T₃). Streptocycline (T₁) showed the least inhibition (30.74 %) of the pathogen among the treatments. Streptocycline caused lysis of the bacteria and it was indicated by the

formation of clear zone around the well. In all other treatments, the pathogenic bacteria were inhibited by the growth of antagonistic bacteria.

A detailed investigation was carried out on symptoms and etiology of four fungal diseases viz., Fusarium wilt, Collar rot, Flower spot and Anthracnose and bacterial disease *Erwinia* soft rot of Phalaenopsis. The *in vitro* evaluation of chemicals and bioagents revealed a high level inhibition of the pathogens.

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REFERENCES

- [1] Abdullah, H. and Kadzimin, S. 1993. Etiology of bacterial soft rot of orchids. *Pertanika Journal of Tropical Agricultural Science* **16** (1): 1 – 4.
- [2] Bag, T.K. 2003. Orchid wilt incited by *Sclerotium rolfsii* on some Indian orchids. *Indian Journal of Hill Farming*. **16** (1/2): 97-98.
- [3] Bag, T.K. 2006. Report of orchid wilt (*Sclerotium rolfsii*) on Vanda group of orchids. *Indian Journal of Hill Farming*, **19** (1): 44-45.
- [4] Geowching, W. and Wenhuei, C. 1998. Influence of temperature and relative humidity on the incidence of soft rot of orchid Phalaenopsis. *Reports of the Taiwan Sugar Research Institute*. (159): 43-57.
- [5] Jin, K.S., Lee, S.W., Kim, J.J. and Ryu, H.Y. 1994. Identification of bacterial isolates obtained from diseased orchid and aloe plants caused by *Erwinia chrysanthemi*. *RDA Journal of Agricultural Science Crop Protection* **36**(1): 301 - 306.
- [6] Jones, S. 2003. Botrytis. *American Orchid Society Bulletin*, American orchid society, Florida. 65p.
- [7] Kim, J.W. and Chun, S.C. 2007. Root and basal stem rot of moth orchid (*Phalaenopsis* sp), Pung - nan (*Neotinetia falcata*) and nadopung - nan (*Aerides japonicum*) caused by *Fusarium* sp. *Research in Plant Disease* **13**: 6 - 14.
- [8] Kotle, V. S. and Raut, S.P. 2007. Efficacy of fungicides in management of collar rot of orchids incited by *Sclerotium rolfsii*. *Annals of Plant Protection Science* **15**(2): 519-520.
- [9] Pant, R.P., Gupta, S., Meena, N.K. and Medhi, R.P. 2011. Emerging diseases of orchids in North Eastern states of India and their management. Abstracts, *National Consultation for Production and Utilisation of Orchids*; p.136, 19 - 21 February, 2011, Pakyong, National Research Centre for Orchids, Sikkim
- [10] Perez, C., Paul, M. and Bazerque, P. 1990. An antibiotic assay by the agar well diffusion method. *Acta Biologie Et Medicine Experimental*. **15**: 113 - 115.
- [11] Rajeevan, P.K. 2011. Production technology of tropical orchids. Abstracts, *National Consultation for Production and Utilisation of Orchids*; p.25, 19 - 21 February, 2011, Pakyong, National Research Centre for Orchids, Sikkim.
- [12] SukYoung, P., HeeJeong, C., GaYoung, K. and YoungJin, K. 1996. Characteristics of anthracnose of orchids caused by *Colletotrichum gloeosporioides*. *Korean Journal of Plant Pathology*. **12**(4): 455-458.
- [13] Uchida, J.Y. 1994. Diseases of orchids in Hawaii. *Plant Disease* **78** (3): 220 - 224.
- [14] Wey, G. C. 1988. Occurrence and investigation of important diseases on Phalaenopsis in Taiwan. *Reports of the Taiwan Sugar Research Institute*. (**122**): 31 - 41.
- [15] Yadav, L. B., Tiwari, A. K., Maurya, R. P. and Srivastav, R. M. 2010. A new record of leaf spot caused by *Alternaria alternata* on orchids in Uttarakhand. *Annals of Plant Protection Science* **18** (2): 551-552.
- [16] Zentimeyer, G.A. 1955. A laboratory method for testing soil fungicides with *Phytophthora cinnamomi* as test organism. *Phytopathology* **45**: 398 - 404.

Table1. Evaluation of fungicides and *Pseudomonas fluorescens* against fungal pathogens of Phalaenopsis

T. No.	Treatment Details	Concentration (%)	Per cent inhibition over control			
			¹ FO	² SR	³ PE	⁴ CG
T ₁	Potassium Phosphonate (40 %) + mancozeb (75 %)	0.20 + 0.15	100	100	100	100
T ₂	Cymoxanil 8 % + mancozeb 64 %	0.20	100	100	79.63	100
T ₃	Carbendazim 12 % + mancozeb 63 %	0.15	100	100	87.41	100
T ₄	Carbendazim 25 % + ipridione 25 %	0.20	100	100	100	100
T ₅	Fenamidone 10 % + Mancozeb 50 %	0.10	100	100	100	100
T ₆	<i>Pseudomonas fluorescens</i> (Liquid)	0.5	61.85	64.80	52.97	86.30
T ₇	Control		-	-	-	-

¹FO - *Fusarium oxysporum*

²SR - *Sclerotium rolfsii*

³PE - *Phoma exigua*

⁴CG - *Colletotrichum gloeosporioides*

Table.2. *In vitro* evaluation of Streptocycline, fresh cowdung extract and *Pseudomonas fluorescens* against *Erwinia chrysanthemi*, the soft rot pathogen of Phalaenopsis

T.No.	Treatment Details	Concentration	Mean diameter of bacterial zone (mm)	Inhibition per cent over control
T ₁	Streptocycline	200 ppm	62.33 ^b	30.74
T ₂	Fresh cowdung extract	2 %	39.67 ^a	55.92
T ₃	<i>Pseudomonas fluorescens</i> (L)	0.5 %	31.00 ^a	65.56
T ₄	Fresh cowdung extract + <i>Pseudomonas fluorescens</i> (L)	2 % + 0.5 %	38.00 ^a	57.77
T ₅	Control		90.00 ^c	0

Plate 1. Symptomatology of diseases of Phalaenopsis

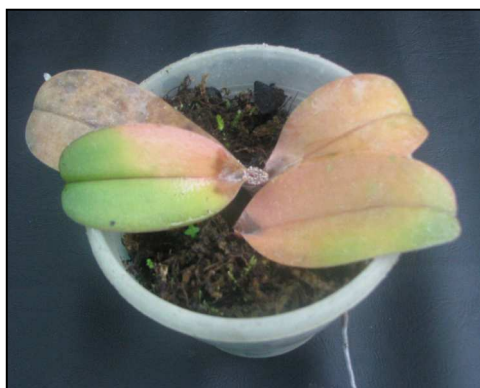


Natural



Artificial

A. *Fusarium wilt*



Natural



Artificial

B. *Collar rot*



Natural



Artificial

C. Flower spot.

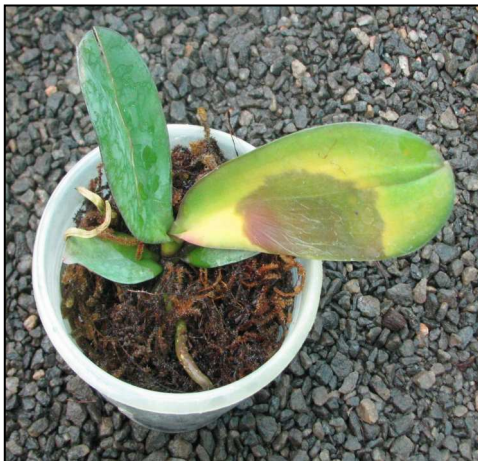


Natural



Artificial

D. Anthracnose



Natural



Artificial

E. Soft rot