HOST RESISTANCE IN COCONUT LEAVES AGAINST LEAF BLIGHT DISEASE CAUSED BY PESTALOTIOPSIS PALMARUM (COOKE) STEY

A. Karthikeyan and R. Bhaskaran^{1/}

ABSTRACT

Total and Ortho dihydroxy were more in younger coconut leaves as compared to the older leaves. Seven phenolic compounds were detected in younger leaves while the older leaves had only six phenolic compounds except chlorogenic acid Phenolic compounds inhibited the mycelial growth and Sporulation of P. palmarum. The quantity of sugars, starch and cellulose were lesser in younger leaves. Carbohydrates increased the mycelial growth and sporulation of the pathogen.

INTRODUCTION

Coconut is an important oilseed and plantation crop in India which is cultivated in an area of 1.63 million hectares with an annual production of 12,355 million nuts. Various fungal and mycoplasma diseases are crippling this important oil Yielding crop in India. Among them, leaf blight caused by *Pestalotiopsi spalmarum* (Cooke) Stey is an important fungal disease reducing the vigour and yield of coconut. Leaf blight also called as grey leaf spot, is of common occurrence in all coconut growing areas throughout the country. The disease causes serious damage in nursery plants and adult palms. Coconut palms heavily infected with leaf blight flowered relatively late than less affected ones.

The leaf blight incidence is found only in only in older leaves of coconut palms. Young leaves (top ten leaves from spindle leaf) are absolutely free from the disease. To find out the mechanism of resistance to the Pathogen P. *Palmarum* present in younger leaves, some biochemical constituents both in younger and older leaves were estimated and reported in this paper.

MATERIALS AND METHODS

East Coast Tall coconut palms of 20-30 years old were selected at Coconut Research Station Veppankulam, India for this study. Leaf samples were collected from the healthy top most three leaves (young) from spindle leaf and four leaves in the lowest leaf whorl (old) of these pahns and analysed for various; bio chemical constituents. Alcoholic plant extracts were prepared for the estimation of phenols arid sugars (Chandramohan et al., 1967). Total phenols were estimated by the method described by Bray and Thorpe (1954) using Folin clocalteu reagent. *Ortho dihydroxyphenols* (O.D. phenols) were estimated by employing Amows reagent (Johnson and Schaal, 1957). The results were expressed mi terms of catechol. equivalents. Individual phenolic compounds present in leaf tissues were separated and detected according to the method given by Raio (1958). The chromatograrns were sprayed with ferric alum (0.2%) to detect polyphenols like catechol and polygallol grouping, sprayed first with 1% NaN0₂, in 10% acetic acid and then with 1N NaOH, to confinn the presence of chlorogemic acid.

To study the effect of phenolic substances on the mycelial growth and sponilation of *P. palmarum*, seven phenolic compound viz., catechol, pyrogallol, chlorogenic acid, caffeic acid, cinnamic acid, p-coumaric acid and ferulic acid were incorporated separately at 0.01, 0.02, 0.05% concentrations in the Czapek's Dox liquid medium.

¹ Coconut Research Station, Veppankulam - 614 906, India.

To each 250 ml conical flask, 70 ml of liquid medium was dispensed and stenlized. The flasks were inoculated with 1 0 mm Rmgal disc of 5 day old culture of the pathogen and were incubated at room temperature for 10 days. After incubation, mycelial mat was separated by filtration, dried and then weighed. Sporulation of the pathogen was assessed by using Haemocytometer.

RESULTS AND DISCUSSION

The results (Table 1) indicated that younger leaves had more quantity of total phenols and *Ortho dihydroxy phenols* as compared to older leaves. Seven phenolic compound viz., catechol, pyrogallol, chlorogenic acid, caffeic acid, cinamic acid, p-coumaric acid and ferulic acid were detected in younger leaves while the older leaves had only six phenolic compounds excepts chlorogenic acid. All these phenolic compounds were more in younger leaves when compared to the older ones. Among the phenolic compounds detected, chlorogemic acid and caffeic acid were found in abundance (Table 2). High tissue phenol level with disease resistance have been correlated in tobacco. against *Helminthosporium* infection (Chauhan, 1987), chickpea against *Ascochyta blight* and *Botrystis grey* would (Mohanunadi and Dwivedi, 1991), mustard plants to *Altenaria* leaf blight (Gupta et. al., 1995).

All phenolic compound tested inhibited the mycelial growth and sporulation of P. *palmarum*. Of them, chlorogenic acid was very effective in suppressing the growth and sporulation decreased with increase in concentration of resistance of younger leaves of coconut to leaf blight pathogen. Phenolics are known as antiftingal, antibacterial and antiviral compunds and they inhibit fungal germination (Vidhyasekara, 1975) arid mycelial growth (Le Tourneau et al., 1957).

In the present study, younger leaves had lesser quantity of reducing, non reducing and total sugars, starch arid cellulose as compared to older leaves (Table 1). Sugars are the precursors for the synthesis of phenolics, phytoalexis, lignin and callose and they play an important role in the defence mechanism of plants. Plants diseases are classified in to high sugar diseases and low sugar diseases. When the host sugar content is more, some disease will occur severely and they are classified as high sugar diseases (Horsfall and Dimond, 1957). The leaf blight disease was severe in older leaves which contained higher quantity of sugars and hence the leaf blight disease maybe classified as high sugar disease. Mycelial growth and sporulation of *P. palmarum* increased with increase in concentration of sugar under mi vitro (Table 4). This suggests that the high sugar level in the older leaves may support the growth and multiplication of *P. palmarum* more than the younger leaves. Bhat et al. (1992) also reported that various sugar substances responsible for enhancing the mycelial growth and sporulation of *P. palmarum*.

It is concluded that higher level of total phenols, *ortho dihydroxy phenols* and presence of phenolic compound chlorogenic acid may be the reason for disease resistance in younger leaves and higher sugar content in older leaves is responsible for susceptibility against leaf blight disease.

REFERENCES

- Bhat, R.G., Srikanth Kulkarni, Naik, K.S., Bhat, M.N and Subramanyam, K. 1992. C and N requirements in vitro of *Pestalotiopsis palmarum* Cke. Current Research, 21 (3) : 5 8 -5 9.
- Bray, H.G. and Thorpe, W.V. 1954. Analysis of phenolic compounds of interenst in metabolism. Meth. Biochem. Analy., 1: 27 - 59.
- Chandramohan, D., Mahadevan, A. And Rangaswami, G. 1967. Studies on some biochemical properties of le-af tissues of *Amaranthus* tricolor as related to resistance to infection by *Altenaria* sp. India Phytopathology, 20, 109-113.
- Chauhan, M.S. 1987. Amino acids, carbohydrates and phenolics in tobacco varieties susceptible and moderately resistant to *Helminthosporium spiciferum*. Indian J. Mycol. Pl.Pathol., 17(1): 17-23.
- Gupta, S.K., Gupta, P.P. and Kaushik, C.D. 1995. Changes *Mi* leaf peroxidase, polyphenol oxidase, catalase arid total phenols due to *Alternaria* leaf blight in brassica species. Indian J. Mycol. P I.Pathol., 25 (3):175 180.
- Horsfall, J.G. and Dimond, A.E. 1957. Interactions of tissue sugar, growth substances and disease susceptibility Z. Pflkrath PfIschatz. 64: 415 -421.
- Inman, R-E. 1965. Qualitative sugar changes in barley infected with a facultative parasite. Phytopathology. 55 :341 -345
- Johnson, G. and Schall, L.A. 1957. Chlorogenic acid and other *Ortho dihydroxy phenols* in scab resistant Russet Burband. and Scab susceptible Triumph potato tubers of different maturities. Phytopathology, 47 : 25 3 -25 8.
- Le Tourneau, D., Mc Lean, J.G. and Guthrie, J.W. 1957. Effect of some phenols and quinones on in vitro growth of *Verticillium albo-atrum*. Phytopathology, 47: 602 606.
- Mohammadi, A. G. and. Dwivedi, T. S. 1991. Detennination of phenolic acids in chickpea by reversed phase high performance liquid chromatography and their relation. to fungal resistance. Indian J. Plant Pathol., 9 (1 & 2): 42-46.
- Nelson, N. 1944. A photometric adaptation of the somogyi method for determination of glucose. J. Biol. Chem. , 153 :375 380
- Ralo, L. 1958. A method for the paper chromatograpic separation and identification of phenol derivatives in wood metabolism and related. compounds of biochemical interest, using a reference system. J. Chromatog., 1 :338 -373.
- Sandulescu, C. And Raju, T. 1959. Determination of ligriin, cellulose and pectin materials in flax and hemp fibres. Acad. R. Pr. Stud. Cercet., 6: 489 -499.
- Sumner, J.B. and Somers, G.F. 1949. Laboratory Experiments on Biological Chemistry. 2nd ed. Academic press. New York. P. 173.
- Vidhyasekaran, p. 1975. Role of auxin phenol complex in finger millet helmintho sporiose disease resistance. Phyhopath. Z., 82 : 89 99.

Table 1: Phenolics and carbohydrates (mg g ¹) in younger and older Leaves of coconut palms				
Constituent	Younger*	Older*	CD	
	Leaves	leaves	(P=0.05)	
Total phenols	2.98	2.46	0.09	
O.D. phenols	0.78	0.44	0.05	
Reducing sugars	3.26	3.98	0.12	
Non reducing sugars	3.03	3.74	0.10	
Total sugars			0.20	
Strarch	0.20	0.33	0.03	
Cellulose	89.4	95.2	0.86	

*Mean of 20 replications

Table 2. Phenolic compounds* in younger and older leaves of coconut Palm				
Phenols	Younger leaves	Older leaves		
Catechol	3	2		
Pyrogal o1	3	2		
Chlorogenic acid	5	0		
Caffeic acid	4	2		
Cinnamic acid	2	1		
P-coumaric acid	3	1		
Ferulic acid	2	1		

*Mean of three chromatograms.

The spots in the chromatogram were categorised as 0 - absent; 1 - traces; 2 - fair; 3 - moderate; 4 - good and 5 - abundant according to the size and intensity of the colour of the spot.

Table 3: Effect of phenols on the mycellial growth and sporulation of P. palmarum						
Phenolic	0.02%		0.02%		0.05%	
substances	ances M.wt* S*		M.wt*	S*	M.wt*	S*
	(mg)	(10 ₆ ml ⁻¹)	(mg)	(10 ₆ ml ⁻¹)	(mg)	(10 ₆ ml ⁻¹)
Catechol	265.7	8.2	223.5	7.9	170.7	6.0
Pyrogallol	283.4	8.4	245.3	8.1	196.3	6.3
Chlorogenic acid	90.3	0.0	63.4	0/0	28.0	0.0
Caffeic acid	105.0	4.7	75.0	0.0	44.5	0.0
Cinamic acid	312.0	9.8	297.3	8.8	216.3	7.6
P. coumaric acid	305.0	9.5	280.0	8.4	212.3	7.6
Ferulic acid	304.5	9.7	280.5	8.4	198.7	6.5
Control (with cut	480.0	19.8	480.0	19.8	480.0	19.8
phenols)						
C.D. (P=0.05)	9.47	0.13	18.50	0.10	29.80	0.10

Table 4: Effect of carbohydrates on the nycelial growth and sporulation of P. Palmarum									
	0.	0.5%		1.0%		3.0%		5.0%	
Carbohydrate	M.wt* (mg)	S* 10 ₆ (ml-1)	M.wt* (mg)	S* 10₀(ml-1)	M.wt* (mg)	S* 10 ₆ (ml ⁻¹)	M.wt* (mg)	S* 10₀(ml⁻¹)	
Glucose	100.0	2.3	124.0	4.5	382.5	11.5	540.3	20.0	
Maltose	104.3	3.7	127.7	4.5	460.0	19.7	570.3	22.5	
Sucrose	109.4	4.0	136.5	4.9	480.3	20.0	590.0	22.7	
Lactose	105.0	3.7	124.0	4.5	385.0	11.5	547.0	21.8	
Mannitol	92.0	0.0	106.5	3.7	298.3	9.2	478.7	19.7	
Strarch	56.3	0.0	90.0	0.0	310.0	10.6	443.3	19.3	
Cellulose	40.5	0.0	64.3	0.0	142.0	4.9	225.3	7.5	
C.D. (P=0.05)	5.77	0.03	6.45	0.07	6.84	2.41	7.20	2.12	

*Mean of three replications M.wt Mycelial weight S: Sporulation