Studies on factors affecting Rhizoctonia bataticola. VII. Plant extract

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Aqueous autoclaved leaf extracts of eighty three plant species were screened *in vitro* for their fungicidal activity against isolates of *Rhizoctonia bataticola*. Paper disc diffusion assay technique revealed that only a few plant extracts viz., *Rannunculus scleratus*, *Xanthium strumarium*, *Ipomoea carnea*, *Ocimum basilicum* and *Eclipta alba* showed varied antifungal activities. The extract of *R. scleretus* was highly effective in inhibiting both the mycelial growth and sclerotia. In poisoned food technique only, growth inhibition at 30% concentration of *O. basilicum* was observed among *X. strumarium*, *O. basilicum* and *I. carnea*, although all the three extracts had their effects on sclerotia.

Key words: Rhizoctonia bataticola, plant extracts, antifungal activities

INTRODUCTION

Chemical control of the diseases causes severe environmental pollution, development of pathogen resistance, biomagnification of pesticides and non-target effects. Consequently, these adverse effects pave the way to develop eco-friendly means to manage diseases (Gautam et al., 2003). The presence of active principles in higher plants and wild vegetation has long been recognized as an important factor to disease resistance (Bambode and Shukla, 1973). Such compounds being biodegradable and selective in their toxicity are considered valuable for controlling different plant diseases (Misra and Dixit, 1979). The toxic chemicals are present in the form of water/solvent soluble, nonvolatile and/or volatile, thermostable add/or thermolabile constituents. The present study has been undertaken to identify the fungitoxicity of the locally available plants belonging to different families against isolates of R. bataticola.

MATERIALS AND METHODS

Fresh leaves of herbs, shrubs and trees belonging to different families which included weeds, ornamentals and some plants known for their medical properties (Table 1), which were abundantly occurring in Jabalpur district, were collected. Leaves were then thoroughly washed with running tap water followed by distilled water. Extracts from washed leaves were made by crushing fresh leaves with sterile distilled water at the rate of one g tissue in

one ml of sterilized distilled water (w/v) separately. The pulverized mass was squeezed through a quadrifold muslin cloth. This formed the standard plant extract solution (100%). The expressed extracts were autoclaved and used for investigation against R. bataticola. Inhibitory properties of plant extracts against R. bataticola were determined by disc diffusion technique. Discs of 10 mm diameter of Whatman's filter paper No. 42 were soaked in autoclaved plant extracts (121°C for 20 minutes). Four soaked discs, after draining excess extracts, were placed opposite to each other on the surface of plates containing potato dextrose agar and seeded centrally with an 8 mm disc cut out aseptically by a sterile cork borer from the margin of a seven days old culture of R. bataticola. Inoculated plates were incubated at 29±1°C and the observations for growth inhibition were recorded after 72 hrs of incubation.

The effective plant extracts were further studied against seven isolates of R. bataticola. Desired concentrations (0, 10, 20 & 30%) were made by adding appropriate amount of standard solution of plant extracts to Asthana & Hawker's medium. Amended medium was autoclaved and poured into sterilized Petri-plates in triplicate. The poured plates were inoculated after 24 hrs with 8 mm discs cut out aseptically by a sterile cork borer from margins of seven days old cultures of R. bataticola isolates. The inoculated plates were incubated at $29\pm1^{\circ}\mathrm{C}$ and observation for growth and morphological characters were recorded on 3rd and 5th day of incubation.

RESULTS AND DISCUSSION

From the results of the investigation shown in the Tables 1 & 2, it was evident from the preliminary

screening of the aqueous autoclaved extracts by paper disc diffusion technique that, in most cases the antifungal activity was thermolabile and was lost on autoclaving. But in the case of *Ipomoea carnea*,

Table 1 : Effect of plant extracts on the mycellal growth and sclerotia formation of Rhizoctonia bataticola (Disc Diffusion Technique)

Stanical Name	Family	Inhibition Over Control		
		Mycelial growth	Sclerotia formation	
1	2	3	4	
Abutilon indicum (Linn.) Sweet	Malvaceae		s 	
calypha indica Linn.	Euphorbiaceae	-	_	
chyranthes aspera Linn.	Amaranthaceae	_	10 miles	
denocalyma alliaceum	Bignoniaceae		_	
dhatoda vasica Nees	Acanthaceae	-	_	
geratum conyzoides Linn.	Asteraceae		_	
Ilternanthera sessilis (Linn.) DC.	Amaranthaceae	_	· -	
maranthus viridis Linn.	Amaranthaceae	_	-	
rgemone mexicana Linn.	Papaveraceae	_		
steracanthus longifolia (Linn.) Nees	Acanthaceae	_		
zadirachta indica A. Juss.	Meliaceae		_	
Blumea sp. DC., nom. cons.	Asteraceae	. —		
Bougainvillaea spectabilis Willd.	Nyctaginaceae		_	
Colebrookea oppositifolia J. E. Smith	Lamiaceae	_	_	
Calotropis procera (Ait.)	Asclepiadaceae	_		
Chenopodium album Linn.	Chenopodiaceae			
Cichorium intybus Linn.	Asteraceae			
Clerodendron inerme (Linn.) Gaertn.	Verbenaceae			
Commelina attenuata Koen ex Vahl.	Commelinaceae	-		
	Commelinaceae	_		
Commelina benghalensis Linn.	Apocynaceae	25/12		
Catharanthus roseus (Linn.) G. Don	Euphorbiaceae	K-10		
Croton sp. Linn.	Convolvulaceae			
Cuscuta reflexa Roxb.		_		
Cymbopogon flexuosus (Neesex Steud.) Wats.	Graminae	A	-	
Cynodon dactylon (Linn.) Pers.	Graminae	_		
Cyanotis cristata (Linn.) D. Don	Commelinaceae	_		
Cyperus rotundus Linn.	Cyperaceae			
Dactyloctenium aegyptium (Linn.) Beauv.	Graminae	_		
Datura metel Linn.	Solanaceae			
Delonix regia (Boj ex Hook.) Rafin	Leguminosae	_	_	
Eclipta alba (Linn.) Hassk.	Asteraceae	+	+	
Eichhornia crassipes (Mart.) Solms	Pontederiaceae	_	_	
Eucalyptus globosus Linn. (globulus) Labill.	Myrtaceae	_		
Euphorbia geniculata Orf.	Euphorbiaceae	-	-	
Euphorbia hirta Linn.	Euphorbiaceae	_	_	
Flemingia semialata Roxb.	Fabaceae	-	_	
Gnaphalium indicum Linn.	Asteraceae	-	1000	
Hyptis suaveolens (Linn.) Poit	Labiatae			
nula indica Linn.	Asteraceae	-	(<u></u>)	
pomoea carnea Jacq.	Convolvulaceae	++	+	
xora arborea Roxb. ex. J. E. Smith	Rubiaceae	-	_	
Jatropha sp. Linn.	Euphorbiaceae	-		
latropha gossypifolia Linn.	Euphorbiaceae		-	
Justicia diffusa Willd.	Acanthaceae	-	_	
Kalanchoe pinnata (Lam.) Pers.	Crassulaceae			
antana camara Linn.	Lythraceae			
agerstroemia indica Linn.	Verbenaceae	_	_	
aunaea nudicaulis (Linn.) Hook. F.	Asteraceae	_		
Malvastrum coromandelianum (Linn.) Garcke	Malvaceae		_	
Medicago denticulata Willd	Malvaceae	_	-	
Medicago hispida Gaertn.	Fabaceae	<u> </u>	92 <u>500</u>	
Melilotus alba Medic. ex. Desr. Lam.	Fabaceae	_	_	
Mimosa pudica Linn.	Mimosaceae	-	_	
· · · · · · · · · · · · · · · · · · ·	Rutaceae			
Murraya exotica Linn.	Apocyanaceae		_	
Nerium oleander Linn.	Labiatae	++	+	
Ocimum basilicum Linn.	Labiatae	7.7	(Continue	

1	2	3	4	
Opuntia dillenii (Kol. Gawl.) Haw.	Cactaceae	_	_	
Parthenium hysterophorus Linn.	Asteraceae		_	
Peltophorum pterocarpum (DC.) Backer ex K. Heyne.	Cesalpiniaceae	<u> </u>		
Phalaris minor Retz.	Graminae		_	
Phyllanthus urinaria Linn.	Euphorbiaceae			
Physalis minima Linn.	Solanaceae	200		
Polyalthia longifolia (Sonn.) Thw.	Annonaceae	_	_	
Pongamia glabra Vent.	Leguminosae	-	· ·	
Punica granatum Linn.	Punicaceae			
Raneunculus sceleratus Linn.	Rannunculaceae	+++	++++	
Rumex dentatus Linn.	Polygonaceae			
Saponaria vaccaria Linn.	Caryophyllaceae		<u></u>	
Sansevieria trifasciata Hort. ex. Prain	Agavaceae			
Bida spinosa Linn.	Malvaceae	_	_	
Solanum nigrum Linn.	Solanaceae	10 <u>-1-10</u>	<u></u>	
Solanum xanthocarpum Schrad & Wendl.	Solanaceae			
Sonchus arvensis Linn.	Asteraceae	_	-	
Sphaeranthus indicus Linn.	Asteraceae		-	
Tabernaemontana coronaria (Jacq.) Willd.	Apocyanaceae	_		
Tagetes minuta Linn.	Asteraceae			
Thevetia nerifolia Juss. ex Steud.	Apocyanaceae	_	-	
Trianthema portulacastrum Linn.	Aizoaceae	_	_	
ridax procumbens Linn.	Asteraceae	_		
ypha latifolia Linn	Typhaceae		-	
Jrena lobata Linn.	Malvaceae			
/ernonia cinerea (Linn.) Less.	Asteraceae			
Kanthium strumarium Linn.	Asteraceae	++	+	

⁻ = No inhibition; + = 25 % inhibition; + + = 50 % inhibition; + + + = 75 % inhibition; + + + + = 100 % inhibition.

Table 2: Effect of plant extracts on the morphological characters of different isolates of R. bactaticola

Plant	Isolate	Conc. (%)	Pattern / P	Hyphae		Sclerotia		73+
				Pattern / Colour	L×W μ	Size / Shape	Pattern / Initiation	Colour
1	2	3	4	5	6	7	8	9
Control	Rb1 Rb2 Rb3 Rb4 Rb5 Rb6 Rb7	Nil	Appr./Even Flocc/Wavy Flocc/Wavy Appr./Even Flocc/Wavy Appr./Even Flocc/Wavy	Dn/LB to B Dn/LB to B Dn/B. Sp/H to LB Dn/LB Dn/LB to B Dn/LB to B	105.62 × 88.25 112.22 × 95.81 79.63 × 73.24 124.4 × 110.31 96.29 × 86.54 107.86× 95.37 92.68 × 84.08	Md/R to O Md/R to O Small/R to O Large/R to O Md/R to O Md/R to O Md/Irre Md/R to O	Dn/Ely Dn/Ely Sp/Ely Dn/Ely Dn/Ely Dn/Ely Sp/Dly	BI BI BI DB BI B
Ipomoea carnea	Rb1 Rb2 Rb3 Rb4 Rb5 Rb6 Rb7	10	Appr./Even Flocc/Wavy Flocc/Wavy Appr./Wavy Flocc/Even Appr./Even Flocc/Even	Sp/H to LB Sp/H to LB Dn/LB. Sp/H Dn/B Dn/B Dn/B	95.33 × 83.41 105.62 × 97.50 49.59 × 46.93 103.75 × 88.75 75.34 × 62.04 67.50 × 61.25 62.00 × 52.25	Md/R to O Md/R to O V. Small/D Md/R to O Small/R to O Small/Irre Small/R to O	Dn/Ely Dn/Ely Sp/Dly Dn/Ely Dn/Ely Sp/Dly	BI BI DB BI BI DB
	Rb1 Rb2 Rb3 Rb4 Rb5 Rb6 Rb7	20	Flocc/Even Flocc/Wavy Flocc/Wavy Appr./Wavy Flocc/Even Appr./Even Flocc/Even	Sp/H to LB Sp/H to LB Dn/LB. Sp/H Dn/B Dn/B Dn/B	95.44 × 85.54 86.66 × 74.47 46.50 × 42.45 79.62 × 65.00 60.06 × 53.75 64.44 × 57.79 53.25 × 44.94	Md/R to O Small/R to O V. Small/D Small/R to O Small/R to O Small/Irre Small/R to O	Dn/Ely Dn/Ely Sp/Dly Dn/Ely Dn/Ely Dn/Ely Sp/Dly	BI BI DB BI BI DB
	Rb1 Rb2 Rb3 Rb4 Rb5 Rb6 Rb7	30	Flocc/Even Flocc/Wavy Flocc/Wavy Appr./Wavy Flocc/Even Appr./Even Flocc/Even	Sp/H to LB Sp/H to LB Dn/LB. Sp/H Dn/B Dn/B	97.91 × 88.36 83.95 × 75.83 43.29 × 40.34 75.88 × 65.22 62.63 × 55.81 64.54 × 58.35 52.31 × 44.75	Md/R to O Small/R to O V. Small/D Small/R to O Small/I to O Small/I to O Small/I to O	Dn/Ely Dn/Ely Sp/Dly Dn/Ely Dn/Ely Dn/Ely Sp/Dly	BI BI DB BI BI DB (Continued)

1	2	3	4	5	6	7	8	9
	Rb1 Rb2 Rb3	3	Appr./Even Flocc/Even Flocc/Wavy	Dn/B Dn/LB Dn/LB	66.13 × 55.05 89.37 × 77.18 48.59 × 44.53	Small/Irre Small/R to O V. Small/D	Dn/Ely Sp/Ely Sp/Dly	BI BI DB
	Rb4 Rb5	10	Appr./Even Flocc/Irre	Sp/LB Dn/B	94.79×78.54 73.12×65.00	Md/R to O Small/R to O	Dn/Ely Dn/Ely	BI
	Rb6 Rb7		Appr./Even Flocc/Wavy	Dn/B Dn/B	76.25×65.29 65.00×56.87	Small/Irre Small/R to O	Dn/Ely Sp/Dly	BI DB
Ocimum basilicum	Rb1 Rb2 Rb3 Rb4 Rb5 Rb6 Rb7	20	Appr./Even Flocc/Even Flocc/Even Appr./Even Flocc/Even Appr./Irre Flocc/Wavy	Dn/B Dn/LB Dn/LB Sp/LB Dn/B Dn/B Dn/B	65.00 × 57.61 81.25 × 73.12 45.59 × 42.05 87.50 × 73.75 67.16 × 62.83 74.75 × 65.25 65.20 × 57.08	Small/R to O Small/R to O V. Small/D Small/R to O Small/R to O Small/Irre Small/R to O	Dn/Ely Dn/Ely Sp/Dly Dn/Ely Dn/Ely Dn/Ely Sp/Dly	BI BI DB BI BI DB
0	Rb1 Rb2 Rb3 Rb4 Rb5 Rb6 Rb7	30	Appr./Even Flocc/Even Flocc/Even Appr./Wavy Flocc/Irre Appr./Even Flocc/Even	Dn/B Dn/B Dn/LB Sp/LB Dn/B Dn/B Dn/B	58.50 × 53.62 85.33 × 75.29 46.99 × 44.34 75.83 × 65.00 65.63 × 57.04 76.44 × 66.32 62.87 × 54.92	Small/R to O Small/R to O V. Small/D Small/R to O Small/R to O Small/Irre Small/R to O	Dn/Ely Dn/Ely Sp/Dly Dn/Ely Dn/Ely Dn/Ely Sp/Dly	BI BI DB BI BI DB
-	Rb1 Rb2 Rb3 Rb4 Rb5 Rb6 Rb7	10	Appr./Wavy Flocc/Even Flocc/Even Appr./Even Flocc/Wavy Appr./Even Flocc/Even	Dn/LB Dn/LB Dn/LB Sp/H to LB Dn/LB Dn/B Dn/B	75.83 × 68.61 66.62 × 61.75 46.62 × 40.11 95.17 × 87.05 88.47 × 81.25 60.56 × 54.65 65.96 × 57.25	Small/R to O Small/R to O V. Small/D Md/R to O Small/R to O Small/Irre Small/R to O	Dn/Ely Sp/Ely Sp/Dly Dn/Ely Sp/Ely Dn/Ely Sp/Dly	BI BI BI BI DB
Xanthium strumarium	Rb1 Rb2 Rb3 Rb4 Rb5 Rb6 Rb7	20	Appr./Wavy Flocc/Even Flocc/Even Appr./Even Flocc/Even Appr./Wavy Flocc/Even	Dn/LB Dn/LB Dn/LB Sp/H to LB Dn/LB Dn/B Dn/B	75.89 × 67.08 67.32 × 60.35 48.50 × 42.05 97.5 × 92.08 85.31 × 79.25 54.65 × 48.75 68.56 × 60.23	Small/R to O Small/R to O V. Small/D Md/R to O Small/R to O Small/R to O Small/R to O	Dn/Ely Sp/Ely Sp/Dly Dn/Ely Sp/Ely Sp/Ely Sp/Dly	BI BI BI BI BI DB
×	Rb1 Rb2 Rb3 Rb4 Rb5 Rb6 Rb7	30	Appr./Even Flocc/Even Appr./Wavy Flocc/Even Appr./Wavy Flocc/Even	Dn/LB Dn/LB Dn/LB Sp/H to LB Dn/LB Dn/B Dn/B	65.00 × 58.22 64.75 × 57.00 46.22 × 41.39 88.37 × 82.83 66.62 × 60.12 55.92 × 48.65 64.37 × 57.73	Small/R to O Small/R to O V. Small/D Small/R to O Small/R to O Small/R to O Small/R to O	Dn/Ely Sp/Ely Sp/Dly Dn/Ely Sp/Ely Sp/Ely Sp/Dly	BI BI BI BI DB

Appr. – Appressed, D – Deformed, Flocc. – Floccose, Irre – Irregular, ⋑n – Dense, Sp – Sparse, B – Brown, DB – Dark Brown, Bl – Black, LB – Light Brown, LB1 – Light Black, Md – Medium, R – Round, O – Oval, E – Elongated, Ely – Early, Dly – Delayed, H – Hyaline, V – Very.

Ocimum basilicum, Xanthium strumarium, Eclipta alba and Rannunculus scleratus there was less effect of autoclaving on fungitoxicity. Extract of Rannunculus scleratus was highly toxic in inhibition of mycelial growth and sclerotia formation. The fungitoxicity of Ipomoea carnea, Ocimum basilicum and Xanthium strumarium was almost equal. Eclipta alba showed less inhibition of mycelial growth as compared to others. The results is in conformity with the findings of Bambode and Shukla (1973). Jha et al. (2000) reported antifungal activity of E. alba on M. phaseolina. Anwar et al. (1994) reported antifungal activity of inflorescence of O. basilicum and I. carnea on M. phaseolina. Leaf extracts of

Ipomoea carnea, Ocimum basilicum and Xanthium strumarium were further investigated against seven isolates employing poisoned food technique to know their effect on sclerotia. Results indicated that Ipomoea carnea and Xanthium strumarium did not show any growth inhibition at 10, 20 and 30% concentrations. Only 30% concentration of Ocimum basilicum inhibited growth of isolates at some extent. It was observed that all the leaf extracts reduced the size of sclerotia at all the concentrations. Generally at higher concentration sclerotial size reduction was more pronounced as compared to lower concentration. However, Gautam et al. (2003) showed growth inhibition of R. solani in alcoholic

extracts of Xanthium strumarium.

The inactivity of the plant extracts excluding the effective ones on *R. bataticola* may be due to thermolabile substances which was inactivated on autoclaving (Bambode and Shukla, 1973) or absence of active principles against *R. bataticola*. According to Skinner (1955) presence of antibiotic contributed to the inhibiting activity of the the plant extracts.

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