Indian Phytopathology, 49 (2) : 181-184 (1996)

Toxic effect of some plant extracts on *Phytophthora capsici*, the foot rot pathogen of black pepper

M. ANANDARAJ and N.K. LEELA

Indian Institute of Spices Research, PB 1701, Marikunnu P.O., Calicut 673 012

Keywords : Plant extracts, *Azadirachta indica, Chromolaena odorata, fungitoxic effect, Phytophthora capsici*

One of the major constraints in black pepper (*Piper nigrum* L.) production is the crop losses due to foot rot disease caused by *Phytophthora capsici* (1,5). Use of fungicides forms one of the major components in the disease management. (6,7). The scope of regular use of fungicide is limited due to the cost and adverse environmental hazards, besides development of resistance in pathogens (9). During recent years use of plant secondary metabolites for the control of fungi is gaining importance (3,4,8). Therefore, in the present study aqueous leaf extracts of *Azadirachta indica, Chromolaena odorata, Lantana camera, Piper colubrinum* and *Strychnos nuxvomica* were tried against vegetative and reproductive phases of *P. capsici.*

Leaves from *A. indica, C. odorata, L. camera, P. colubrinum* and *S. nuxvomica* were collected, washed with distilled water and dried in oven for 48 h at 50°C, and the dried leaves were powdered. From each species, 50g of leaf powder was suspended in 250 ml distilled water, boiled 2 minutes and filtered through Whatman No. 1 filter paper. The residue was re-extracted twice, each time with 250 ml distilled water and all aqueous portions were mixed together and concentrated to 50 ml using a flash evaporator. From this extract, concentrations ranging from 0.25 to 2.0% were prepared, which were used for the bioassay. In a preliminary study, the extracts were added to carrot agar medium before and after autoclaving, and the growth of *P. capsici* was tested. There was inhibition of growth both in plates inoculated with the fungus where the extracts were added before and after autoclaving, indicating the thermostable nature of the extracts. Hence, all further studies were conducted by adding extracts and then autoclaving the medium.

The effect of plant extracts on growth of *P. capsici,* sporulation, sporangial germination and zoospore germination were studied. For the studies on growth of mycelium poisoned food technique (10) was used. The test extracts in the concentrated form was taken and appropriate volume mixed with 2% carrot agar to obtain concentrations ranging from 0.25 to 2.0% in the final volume of 60 ml of medium. This 60 ml medium was dispensed into four 9 cm Petri plates for replications. *P. capsici* from black pepper was cultured on carrot agar, 1 cm diam. mycelial discs were cut with cork borer and placed in the centre of each plant. In control sets, appropriate quantities of distilled water was mixed in place of plant extracts. The plates were incubated at 25°C (± 1°C), growth of colony was measured after 72 h and 120 h after inoculation. The radial growth of mycelium was measured at two points along the diameter of the plate and the mean of these two readings was

Table 1 : Effect of aqueous plant extracts on growth and sporulation of *Phytophthora capsici*

Leaf extracts from	Conc. (%)	Mycelial Growth inhibition(%)	Sporulation Inhibition (%)	Zoospore release inhibition(%)	Zoospore germination inhibition(%)
Azadirachta	2.00	75.53 (60.35)	100.0 (89.68)	100.0 (89.68)	100.0 (89.68)
indica	1.00	64.11 (53.19)	100.0 (89.68)	100.0 (89.68)	100.0 (89.68)
	0.50	38.17 (38.15)	100.0 (89.68)	56.66 (48.82)	66.99 (54.93)
	0.25	21.66 (27.73)	100.0 (89.68)	7.02 (15.36)	74.86 (59.90)
Chromolaena	2.00	100.0 (89.68)	100.0 (89.68)	100.0 (89.68)	100.0 (89.68)
odorata	1.00	97.65 (81.19)	100.0 (89.68)	58.54 (49.92)	100.0 (89.68)
	0.50	82.03 (64.92)	100.0 (89.68)	46.80 (44.19)	100.0 (89.68)
	0.25	68.71 (56.00)	100.0 (89.68)	18.96 (25.81)	100.0 (89.68)
Lantana	2.00	72.07 (58.09)	76.30 (60.87)	56.06 (48.48)	4.92 (12.82)
camara	1.00	68.16 (55.65)	14.71 (26.36)	51.94 (46.11)	2.57 (9.21)
	0.50	59.03 (50.20)	8.65 (17.10)	22.26 (28.15)	6.44 (14.70)
	0.25	52.62 (46.49)	8.69 (17.14)	17.34 (24.61)	1.38 (6.75)
Piper	2.00	83.78 (66.25)	96.38 (79.02)	23.71 (29.14)	20.42 (26.86)
colubrinum	1.00	56.21 (48.57)	93.63 (75.38)	33.65 (35.46)	18.26 (25.29)
	0.50	34.47 (35.83)	50.91 (45.52)	11.60 (19.91)	8.19 (16.63)
	0.25	20.34 (26.81)	46.26 (42.86)	24.36 (29.57)	10.19 (18.62)
Strychnos	2.00	38.10 (38.11)	100.0 (89.68)	31.47 (34.12)	13.96 (21.93)
nuxvomica	1.00	18.41 (25.41)	100.0 (89.68)	27.68 (31.74)	28.62 (32.34)
	0.50	6.37 (0.29)	100.0 (89.68)	8.97 (17.43)	17.61 (24.81)
	0.25	0.0 (0.29)	100.0 (89.68)	7.48 (15.87)	15.77 (23.34)
CD 5%		5.31	5.88	14.5	8.51

CD = Critical difference. Figures in parentheses are transformed values.

taken as the diameter of the colony. The growth of the colony in control sets was compared with that of various treatments and the difference was converted in to per cent inhibition.

For testing the effect of plant extracts on sporulation, *P. capsici* was grown on carrot agar in dark for 48 h at 20°C and 1cm diam. discs of mycelium were cut and placed in Petri plates. Aqueous plant extracts of different concentrations were placed on these discs and incubated under light for 24 h. In control, the discs were covered with distilled water. Number of sporangia produced per microscopic field were counted. The average for 6 microscopic field for each replication was counted and compared with that of control. To study the effect of plant extracts on zoospore release (indirect germination of sporangium), *P. capsici* culture grown on carrot agar medium for 48 h were taken, 1 cm diameter discs were cut and allowed to sporulate by incubating in light as described above. Such sporulating discs were taken in Petri plates, plant extracts of concentrations ranging from 0.25 to 2.0% placed over them and incubated at 4°C for 10 minutes. These plates were then taken out and incubated at laboratory

temperature for 30 minutes before observation. For control sets, distilled water was used in place of plant extracts. Due to cold shock, zoospores formed inside sporangia were released.The number of sporangia which released zoospores were counted. Six microscopic fields were observed for each replication and per cent inhibition was calculated by comparing with control plates.

To evaluate the effect of plant extracts on germination of zoospores, sporulating discs were subjected to cold shock at 4°C for 10 minutes as described above, and zoospores released were collected in test tubes and vortexed. The zoospores which settled at bottom were collected and 0.5 ml of the water containing zoospores were placed on clean microscopic slides and 0.5 ml of plant extracts were poured and mixed so that the final concentration ranged from 0.25 to 2.0%. In control, distilled water was used. All the slides were incubated at room temperature inside Petri plates lined with moist filter paper for 12 h. The treatment was replicated four times. In each slide, four microscopic fields were observed for the number of zoospores present and number germinated were counted. Inhibition of zoospore germination was calculated by comparing with control.

Among the five plant species whose extracts were tried on *P.capsici, P.colubrinum* is resistant to this fungus and other species are the non-hosts of this fungus. The effect of these extracts on different growth phases of *P. capsici* is presented in Table I. Mycelial growth, sporangial production, zoospore production and release, and zoospore germination were completely inhibited by *C. odorata* extracts at 2.0% concentration. *A. indica* extracts also acted similarly but mycelial growth was inhibited only by 75.5%. *P. colubrinum* extracts inhibited mycelial growth and sporangial production whereas, sporangial germination and zoospore germination were inhibited by 23.71% and 20.42% even at 2% concentration. The extracts from *S. nuxvomica* had inhibitory effect on sporangial production at 0.25% concentration whereas, on other phases of the fungus, it was not very effective. The extracts from *L. camera* was effective only at 2% concentration but it is not as effective as other extracts. *P. capsici* is a wet weather pathogen and maximum population build up occur through asexual phase by production of sporangia and zoospores. This phase is epidemiologically significant as it contributes to the course of the epidemics caused by *Phytophthora.* For the control of *pythiaceous* fungi, solubility of the chemical in water is an added advantage since free water is required to complete the life cycle of these organisms (2). The water soluble nature of the toxic principle in *A. indica* and *C. odorata* is ideal for developing into a botanical pesticide. Since the aqueous extracts of these plants were found to affect the critical phases of *P. capsici,* it is encouraging to identify and characterise the active principle.

REFERENCES

1. **Anandaraj, M., Ramachandran, N. and Sarma, Y.R.** (1991). In *Black Pepper Diseases* (Eds., Sarma, Y.R. and Premkumar, T.). National Research Centre for Spices, Calicut. pp. 114-135.
2. **Bruin, G.C.A. and Edgington, L.V.** (1988). In *Zoosporic Plant Pathogens - A Modern Perspective* (Ed. Buczacki, S.T.). Academic Press. pp. 352.
3. **Dixit, S.N., Dubey, N.K. and Tripathi, N.N.** (1983). In *Recent Advances in Plant Pathology.* (Eds., Husain, A., Singh, B.P. and Agnihotri, V.P.). CBS, New Delhi, pp. 248-252.
4. **Fawcett and Spencer, D.M.** (1990). *Annu. Rev. Phytopathol.,* **8** : 403-408.
5. **Holliday, P. and Mowat, W.P.** (1963) Phytopathological Paper no. 5, Commonwealth Mycol. Institute, Kew, Surrey. pp. 62.
6. **Ramachandran, N., Sarma, Y.R. and Anandaraj, M.** (1991) In *Black Pepper Diseases* (Eds., Sarma, Y.R. and Premkumar, T.). National Research Centre for Spices, Calicut. pp 158-174.
7. **Sarma, Y.R., Ramachandran, N. and Anandaraj, M.** (1991) In *Black Pepper Diseases* (Eds., Sarma, Y.R. and Premkumar, T.). National Research Centre for Spices, Calicut. pp 55-109.
8. **Singh, J. and Tripathi, N.N.** (1993) *J. Ind. Bot. Soc.* 73 : 1-3.

9. **Wellman** (1977) *Annu. Rev. Phytopathol.* **15** : 153-163.

10. **Zentmyer, G.A.** (1955) *Phytopathology,* **45** : 398-404.

Received for publication January 16, 1995