*Department of Botany, * Department of Biochemistry,*
University of Karachi, Karachi-75270, Pakistan

USE OF MANGROVE FOR THE CONTROL OF *MELOIDOGYNE JAVANICA* IN TOMATO

by
F. S. Mehdi, I. A. Siddiqui, T. Zia* and N. I. Ali

Summary. Aqueous, methanolic and chloroform extracts of *Avicennia marina* and *Rhizophora mucronata* (mangrove) caused significant ($p<0.05$) mortality of *Meloidogyne javanica* juveniles. Methanol soluble fractions showed greater nematicidal activity than chloroform fractions indicating that active nematicidal compound(s) were polar in nature. Soil amendment with *A. marina* and *R. mucronata*, with or without the biocontrol bacterium *Pseudomonas aeruginosa*, significantly ($p<0.05$) reduced root-knot infection due to *M. javanica* in tomato seedlings under glasshouse conditions. The use of *R. mucronata* with *P. aeruginosa* also increased plant height and fresh weight of shoots.

The addition of organic materials to soil has been demonstrated to be a satisfactory control method against many phytoparasitic nematodes, particularly in developing countries because of the cheapness and easy availability of materials (D'Adabbo, 1995). In a previous report (Mehdi *et al.*, 1999), soil organic amendment with *Avicennia marina* (mangrove) was shown to have significant control of soilborne root-infecting fungi such as *Macrophomina phaseolina*, *Fusarium solani* and *Rhizoctonia solani* and the root-knot nematode (*Meloidogyne javanica*) in tomato. Likewise, soil amendment with neem cake and brown seaweed alone or mixed with *Pseudomonas aeruginosa*, a plant growth-promoting rhizobacterium, significantly reduced the number of galls, egg mass production and population of *M. javanica* in soil (Siddiqui *et al.*, 1999). The present report describes the effects of *Avicennia marina* and *Rhizophora mucronata* alone or in conjunction with *P. aeruginosa* in the control of *Meloidogyne javanica* (Treub) Chitw. in tomato, *Lycopersicon esculentum* Mill.

Materials and methods

Leaves of *Avicennia marina* (Forsk). Vierh and *Rhizophora mucronata* Lam collected from Hawk's Bay and Sands Pit near Karachi coast, were washed, dried and powdered. For the extraction of aqueous plant material, 250 g of leaf powder of each plant were immersed in 1 l boiling distilled water for 5 mins. The heated extract was filtered twice, and the filtrate was lyophilized and stored in a refrigerator at 6 °C. For the extraction of plant material with methanol or chloroform, 250 g powdered leaves of mangrove were soaked in 1 l methanol or chloroform for six days. The extract was filtered twice and the filtrate concentrated in a rotary vacuum (EYLA) evaporator. An appropriate amount of the extracts was dissolved in the respective organic solvent to make 10, 1 and 0.1 mg/ml concentrations. Three replicates were prepared for each concentration and 1 ml of each dilution was transferred into a watch glass and left for 48 h to evaporate the organic solvent. A watch glass con-

taining 1 ml of evaporated methanol or chloroform served as a control. A watch glass containing 1 ml sterile distilled water was also kept to compare the efficacy of the aqueous plant extract. One ml of a freshly hatched juvenile suspension of *M. javanica* containing 20-25 juveniles/ml was transferred to each watch glass. After 48 h exposure, the number of juveniles that were killed were recorded using a stereomicroscope. The nematodes were considered to be dead if they did not move when probed with a fine needle (Cayrol *et al.*, 1989).

Powdered leaves of each mangrove species alone and one application of *P. aeruginosa* alone were given. A further four treatments combined *P. aeruginosa* with each mangrove treatment. Powdered leaves were mixed with sandy-loam soil to give the concentration of 0.5 and 1% w/w and potted into 21 cm diam clay pots, 2 kg of sandy loam per pot. The soil was watered daily to promote decomposition. After three weeks, the soil in each pot was excavated to a depth of 3 cm and 300 ml aqueous cell suspension of *Pseudomonas aeruginosa* (Schroeter) Migula strain IE-6 (2.3×10^8 cfu ml^{-1}) maintained on King's B medium was drenched into each pot. Pots containing soil without the mangrove or biocontrol agent served as control. After soil treatment three tomato seedlings that had been raised in sterile soil were planted in each pot. One week after the seedling transplanting, the soil in each pot was infested with 2000 freshly hatched second stage juveniles of *M. javanica*. There were three replicates of each treatment and the pots were randomized on a glasshouse bench. The experiment was terminated 45 days after the addition of nematode and plant height and fresh weight of shoot were recorded. The number of galls produced on the entire root system were counted with the aid of a low power microscope (x6). Data were analysed and subjected to one way Analysis of Variance (ANOVA) followed by Least Significant Difference (LSD) according to Gomez and Gomez (1984).

Results and discussion

Aqueous, methanol and chloroform extracts of *A. marina* and *R. mucronata* caused significant ($p<0.05$) mortality of *M. javanica* juveniles. With an increased concentration, mangrove activity increased. A methanolic extract of *R. mucronata* at 10 mg/ml concentration produced maximum (78%) juvenile mortality followed by a methanol extract of *A. marina* (72% juvenile mortality). Aqueous and chloroform extracts respectively of *R. mucronata* ad 10 mg/ml caused 55 and 32% mortality of *M. javanica* juveniles (Table I).

TABLE I - *Effect of different concentrations of aqueous, methanol and chloroform extracts of* Avicennia marina *and* Rhizophora mucronata *on the mortality of* Meloidogyne javanica *juveniles.*

Treatments	Concentration	Mortality %		
		Aqueous	Methanolic	Chloroform
Control		0	4	6
Avicennia marina	10 mg/ml	50	72	43
	1 mg/ml	17	32	17
	0.1 mg/ml	8	13	13
Rhizophora mucronata	10 mg/ml	55	78	32
	1 mg/ml	34	42	30
	0.1 mg/ml	14	12	18
LSD 0.05			14.97	

TABLE II - *Effect of mangroves and* Pseudomonas aeruginosa *in the control of* M. javanica *in tomato.*

Treatment	Fresh weight of shoot (g)	Height of shoot (cm)	Galls per root system
Avicennia marina 0.5%	2.2±0.25	21.9±2.40	63±1.34
Avicennia marina 1.0%	3.0±0.47	20.6±2.73	59±10.09
Rhizophora mucronata 0.5%	2.5±0.26	21.7±0.83	58±3.56
Rhizophora mucronata 1.0%	2.7±0.49	21.7±1.83	50±6.38
Pseudomonas aeruginosa (IE-6)	2.4±0.32	21.8±2.69	51±2.02
Avicennia marina 0.5% + Ps. a. IE-6	3.8±0.86	21.1±2.50	51±4.82
Avicennia marina 1.0% + Ps. a. IE-6	2.7±0.19	20.3±0.33	49±6.66
Rhizophora mucronata 0.5% + Ps. a. IE-6	2.8±0.33	21.8±2.71	47±8.38
Rhizophora mucronata 1.0% + Ps. a. IE-6	4.1±0.92	26.4±1.68	45±5.49
Control	1.9±0.44	19.1±2.50	75±2.45
LSD 0.05	0.8	3.1	9

± = Standard deviation.

Soil amendment with mangrove significantly ($p<0.05$) suppressed root-knot development in tomato. *R. mucronata* was more effective than *A. marina* in the control of root-knot infection. *P. aeruginosa* also significantly reduced root-knot development and increased plant growth. Soil amendment with mangrove increased the biocontrol and growth promoting potential of *P. aeruginosa*. *R. mucronata* at 1% mixed with *P. aeruginosa* resulted in the greatest (40%) reduction in gall formation in tomato. Similarly, the maximum plant height and fresh weight of shoots was recorded in the treatment where *R. mucronata* at 1% was used with *P. aeruginosa* (Table II).

The results of the present study indicate that active compounds responsible for the nematicidal activity are methanol soluble and hence may be polar in nature. However, it is not known if the nematicidal activity demonstrated by *A. marina* or *R. mucronata* was due to a single compound or a number of compounds since all the extracts showed significant juvenile mortality at 10 mg/ml concentration. Further investigation is needed to isolate and characterize the nematicidal compounds.

Literature cited

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