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A METHODOLOGY FOR EVALUATION OF RESISTANCE IN CARDAMOM TO ROOT-KNOT NEMATODE *(MELOIDOGYNE INCOGNITA)*

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Abstract : Vegetative suckers of a susceptible line of cardamom were inoculated with 100, 500, 1000 and 5000 *Meloidogyne incognita*. The nematode multiplication and gall formation were assessed at 2, 3, 4, 5 and 6 months after inoculation. Nematode multiplication was inversely proportional to the initial inoculum level. Initial population of 100 and 500 nematodes consistently produced susceptible reactions, GI>2 and R>1. GI and Pf at these levels were significantly different from the rest, 3 months after inoculation. Therefore, an initial inoculum of 500 nematodes and an exposure period of 3 months are recommended for evaluation of resistance in cardamom.

Key words : Resistance, Cardamom, Root-knot nematode, Meloidogyne incognita

Root-knot nematodes (*Meloidogyne* spp.) are widely distributed in the cardamom growing tracts of India and are potentially damaging to cardamom (Eapen, 1991). Several pre and post-sowing chemical control measures are recommended for the control of these nematodes. But the high cost and the risk of nematicide residues necessitate a safer, economic and effective means of management like growing resistant plants. For this, numerous cultivable and wild germplasm collections of cardamom have to be screened. This requires a standard screening method, ideal for quantitative assessment of host suitability of cardamom. Several parameters like reproduction factor, dry shoot weight reduction, gall and egg mass indices were proposed for this purpose. Sasser *et al.* (1984) reported a new rating system based on gall index and reproduction factor for screening crop germplasm against root-knot nematodes. This study was undertaken to test the suitability of these parameters for estimating degree of susceptibility in cardamom.

MATERIALS AND METHODS

Nematode free and uniform adult suckers of a known susceptible accession of cardamom (*Elettaria cardamomum.*, Maton), namely Pi, were raised in sterile conditions. Later, one sucker each was transplanted to 20 cm -diameter polythene bags containing about 1500 cm^3 methyl bromide-fumigated soil. They were arranged on screen house benches in a completely randomised block design. Forty five days after transplanting, 25 plants each were infested with 100, 500, 1000 and 5000 juveniles of *Meloidogyne incognita*. The population of *M. incognita* was originally collected from infested carda-

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mom plants in Block 13 of the research centre and had been multiplied on tomato (local variety) in the screen house (average temperature 11.6° - 26.2°C). The nematodes were extracted by a combination of maceration and filteration (Hooper, 1970). Aqueous suspensions (50 ml) of nematodes of each density were poured to the immediate vicinity of roots after carefully removing the top soil in each polybag. After inoculation, sterile sand was sprinkled on top.

Five plants of each treatment were removed from polybags after 2, 3, 4, 5 and 6 months of inoculation. The roots were excised, washed thoroughly and observed under X10 magnification for recording the number of galls per plant. Gall index (GI) was recorded on a 0 - 5 scale (Taylor & Sasser, 1978). Subsequently these roots were pressed to uniform dryness and the dry weights were taken. The nematodes were extracted by chopping the roots in a blender with 1% NaOCl solution. The extracted eggs were stained red with acid fuchsin - acetic acid solution (Byrd *et al.*, 1983). Each nematode suspension was made upto 200 ml with tap water and the population was estimated by drawing three one ml aliquots. This count was used for calculating number of nematodes per gram root and final population in each plant. Reproduction factor was derived by the formula R = Pf/Pi where Pf represents the final population in the infected plant and Pi the initial population used for inoculation. All nematode counts were $\log_{10}$ (X + 1) transformed and the data were subjected to analysis of variance. The means were separated using least significant difference (P, < 0.05).

RESULTS AND DISCUSSION

The mean values of GI, nematodes per g root, Pf and R of each treatment were given in Table 1. The data showed a gradual increase in galling, nematode population and their multiplication with the increase in exposure period. However, the variation was not uniform with the increase in Pi. R values always exceeded 1 at Pi = 100 and 500. But in plants which received 5000 nematodes initially, the Pf seldom exceeded the Pi. Similarly with 1000 nematodes also the Pf was below Pi till the 4th month after inoculation. Therefore the greatest rate of population increase occurred at the lowest Pi, with the rate decreasing as Pi increased. Similar observations are recorded for other perennial crops like black pepper (Ramana & Mohandas, 1989), alfalfa (Noling & Ferris, 1986), and this has been attributed to competition for resources and feeding sites at the higher densities (Seinhorst, 1967).

The GI among various Pi were on par at 2, 5 and 6 months while Pf were on par 2, 4, 5 and 6 months after inoculation. Nematodes per g root were at par at all months except at the 2nd month. However, all variates increased over time. Susceptible reactions (GI>2 and R>1) were consistently observed with 100 and 500 initial populations, at all intervals. Moreover they showed significant difference in GI and Pf from the rest of the Pi at the 3 - month interval. Among these, Pi = 500 had given higher values for GI and per g nematodes. Therefore, an inoculum level of 500 nematodes and an exposure period of 3 months were found to be optimum for screening of cardamom germplasm suckers. As reported elsewhere (Sasser *et al.*, 1984; Shepherd, 1978), GI and Pi were found to be sufficient for estimating host suitability in cardamom also. According to Canto-Saenz (1983), hosts suitability is based jointly on host efficiency and plant damage. Galling (Sasser *et al.*, 1984) and dry shoot weight reduction (Husain, 1986; Abd-Elgawad & Auter, 1989) are the measures of plant damage. Eventhough the gall size is quite small in cardamom suckers (Eapen, 1991) it is the best available indicator of plant damage. Staining with Phloxine B was reported to make the rating relatively easy (Hartman, 1983). The growth reduction consequent to nematode infestation was also not visible within six months. Therefore plants with GI<2 in the initial screening should be subjected to further rigorous testing for confirming their resistant behaviour. Egg mass index (Araujo *et al*., 1982; Taylor & Sasser, 1978), rates of penetration, maturation and reproduction (Abd - Elgawad & Auter, 1989) can also be used in this second round to investigate further the response of plants.

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TABLE 1: Mean gall indices, nematodes per gram root, final population and reproduction factors of plants inoculated with various levels of *M. incognita* (Mean of five replications).

a) *Gall index (GI)*

Pi	*Observation* intervals 2	3	4	5	6	Mean for Pi
100	2.2	3.6	2.2	3.2	3.8	3.00
500	3.4	3.8	1.8	3.6	3.5	3.22
1000	3.5	3.2	3.6	2.5	3.4	3.24
5000	2.0	1.8	2.0	4.4	4.0	2.84
Mean for intervals	2.76	3.10	2.40	3.43	3.68	

LSD_{05} for Pi = N.S.

LSD_{05} for intervals = 0.89

LSD_{05} for interaction = 1.79

b) *Nematodes per gram root*

Pi	Observation intervals 2	3	4	5	6	Mean for Pi
100	109.92*	96.95	151.05	212.30	401.72	169.22
	(2.045)	(1.991)	(2.182)	(2.329)	(2.605)	(2.231)
500	189.11	104.93	217.78	735.21	533.56	279.54
	(2.279)	(2.025)	(2.340)	(2.867)	(2.728)	(2.448)
1000	269.40	40.88	244.47	503.66	489.91	232.35
	(2.432)	1.622)	(2.390)	(2.702)	(2.691)	(2.368)
5000	61.63	36.24	223.39	445.68	237.23	139.93
	(1.794)	(1.571)	(2.351)	(2.650)	(2.377)	(2.149)
Mean for intervals	136.40	62.39	206.01	432.51	397.11	
	(2.138)	(1.802)	(2.316)	(2.637)	(2.600)	

LSD_{05} for Pi = (N.S) LSD_{05} for intervals = (0.281)

LSD_{05} for interaction = (0.562)

* Weighted mean obtained by detransforming
Figures in parentheses are $\log_{10}$ transformed values

c) *Final population* (Pf)

Pi	Observation intervals 2	3	4	5	6	Mean for Pi
100	308.74*	1464.55	1547.82	3705.81	4445.31	1632.05
	(2.491)	(3.166)	(3.190)	(3.569)	(3.648)	(3.213)
500	425.58	1201.26	1427.89	7361.07	4829.59	1917.67
	(2.630)	(3.080)	(3.155)	(3.867)	(3.684)	(3.283)
1000	537.27	260.82	2522.48	2998.16	8789.23	1562.15
	(2.732)	(2.418)	(3.402)	(3.477)	(3.944)	(3.194)
5000	502.50	259.62	1467.93	7584.78	1998.86	1237.80
	(2.702)	(2.416)	(3.167)	(3.880)	(3.301)	(3.093)
Mean for intervals	434.51	587.84	1693.34	4987.84	4404.55	
	(2.639)	(2.770)	(3.229)	(3.698)	(3.644)	

LSD_{05} for Pi = (N.S.) LSD_{05} for intervals = (0.322)

LSD_{05} for interaction = (0.643)

* Weighted mean obtained by detransforming
Figures in parentheses are $\log_{10}$ transformed values

d) *Reproduction factor* (R)

Pi	Observation intervals 2	3	4	5	6	Mean for Pi
100	5.60	18.00	19.33	65.16	66.85	34.96
500	1.56	3.86	3.33	24.39	26.22	11.87
1000	0.60	0.90	4.82	6.00	15.20	5.50
5000	0.13	0.36	0.42	1.32	0.87	0.62
Meari for intervals	1.97	5.78	6.98	24.18	27.28	

LSD_{05} for Pi = 10.04 LSD_{05} for intervals = 11.22

LSD_{05} for interaction = 22.45

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