

SEPARATE AND COMBINED EFFECTS OF *RADOPHOLUS SIMILIS* AND *CYLINDROCARPON OBTUSISPORUM* ON ARECANUT SEEDLINGS

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Abstract : A direct correlation between levels of *Radopholus similis* population present in soil and growth and intensity of root lesions in arecanut (*Areca catechu*) seedlings was recorded. The nematode in combination with *Cylindrocarpon obtusisporum* caused more damage in inoculations where nematode was introduced three weeks prior to fungus. The fungus alone did not cause any appreciable damage. Nematode multiplication was inhibited in the presence of fungus.

Key words : *Radopholus similis*, *Cylindrocarpon obtusisporum*, Pathogenicity, *Arecanut*

Association of burrowing nematode, *Radopholus similis*, with arecanut was first reported by Kumar *et al.*, (1971). Subsequently, it was reported to cause lesions and severe root rotting of arecanut (Koshy *et al.*, 1976, 1978). Besides *R. similis*, a fungus, *Cylindrocarpon obtusisporum* was also isolated from the lesioned roots (Sundararaju and Koshy, 1984) and though the nematode has been reported as a potential pathogen to arecanut (Koshy and Sundararaju, 1983), its pathogenic effect along with *C. obtusisporum* has not been studied so far. This aspect was investigated, therefore, under pot conditions and the results are reported here.

MATERIALS AND METHODS

Arecanut seedlings cv, 'South Kanara' were raised in earthen pots (35 × 35 cm), containing methyl bromide-fumigated sandy loam soil in March, 1982. After 6 months, 60 seedlings of uniform growth were selected. Sixty cement tubs

(70 × 80 cm) were placed in soil pits, leaving the top 15 cm above the soil surface, at 90 cm distance in field. Each tub was filled with 500 kg (270 litre) of methyl bromide-fumigated sandy loam soil in September, 1982. Arecanut seedlings were transplanted in the tubs and at the same time two sterile PVC tubes (25 mm diam. and 30 cm long) were placed on to the root system of each plant.

There were ten treatments replicated 6 times :

T₁-0 (control),

T₂-10,

T₃-100,

T₄-1,000,

T₅-10,000,

T₆-1,00,000 (T₂-T₆ Nematodes alone)

T₇-Fungus alone

T₈-Nematode (10,000) and Fungus inoculation simultaneously

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T₉-Nematode (10,000) inoculation 3 weeks prior to fungal inoculation.

T₁₀-Nematode (10,000) inoculation 3 weeks after fungal inoculation.

The nematode population was multiplied axenically on carrot discs and the fungus was multiplied in 100 ml flasks, containing 25 ml of potato dextrose agar medium. One month after transplanting, six plants each were inoculated with nematodes suspended in sterile water using different levels of inoculum. Fungus inoculations were done using 100 ml aqueous suspension (100,000 spores/0.5 ml) per plant. Water alone was added to the control plants. All inoculations were made through plastic tubes placed *in situ* before. The plastic tubes were removed after inoculations and the holes were covered with sterilized sandy loam soil. *Pueraria phaseoloides* was grown in the interspace as a cover crop to reduce heat and soil splashing. Observations on growth characters were recorded at 6 monthly intervals. One seedling each of treatment T₆ and T₉ died during the experiment period.

The experiment was terminated in December, 1985. Soil samples were drawn with the help of 45 cm diam. galvanised iron tube, at 25, 50 and 75 cm depths around the plant from each tub and then mixed together for analysis. Seedlings were displaced from the tubs using a strong jet of water to displace soil particles and plant growth parameters were recorded. Lamina length was taken by measuring the distance from the leaf tip to the point from where the leaflets emerged. Leaflet length was recorded by measuring the longest leaflet in the middle of the leaf. The roots were indexed visually for lesions and rotting on a scale of 1 to 5 (1=no infection, 2=small, elongate lesions on white roots with necrotic root tips, 3=prominent lesions of dark brown to black with necrotic root tips, 4=coalescing lesions with necrotic root tips and partial decay

of roots, 5=severe decay of roots with necrotic root tips). After indexing, roots were cut into small pieces, mixed thoroughly and stained in boiling acid fuchsin-lactophenol for three minutes, cleared and churned for 40 seconds using a Waring blender for population assessment. Re-isolation of fungus from roots of inoculated plants was carried out and the fungus was identified:

RESULTS AND DISCUSSION

Plants inoculated with different levels of nematode population and in combination with fungus exhibited general yellowing with visible reduction in growth compared to control. Generally there was a direct correlation between nematode levels and plant growth, with significant reductions, except shoot weight and leaf production, in all cases (Table 1). An initial inoculum level of 100 nematodes per plant caused significant also reduction in plant growth as reported by Koshy and Sundararaju (1983). No appreciable damage was noticed in plants inoculated with fungus alone. Root colonization by the fungus was poor when inoculated alone. However, the presence of the fungus inhibited the multiplication of the nematode. Consequently root lesion indices and reduction in growth parameters were less in the presence of fungus except in inoculations where nematode was introduced three weeks prior to fungus.

The root system of all inoculated plants showed lesions, root rotting, blackening of root tips and reduction in number of lateral roots in various intensities (Table 2). Drastic reduction of root mass and volume could be obtained with the nematode inoculum levels of 1000 and above, alone, and in combination with fungus. Similar results have been reported on coconut by Koshy and Sosamma (1983). Significant differences in root lesion indices were recorded in all the treatments compared to control

TABLE 1. Effect of individual and combined inoculation with *Radopholus similis* and *Cylindrocarpon obtusisporum* on the growth of arecanut (Av. of six replications)

Treatments	Shoot		Leaf				Root		Collar girth (cm)	
	Length of the stem (cm)	Length of the plant (cm)	Weight (kg)	No. of leaves (produced)	No. of leaves (retained)	Lamina length (cm)	Leaflet length (cm)	No. of main roots		Weight (kg)
0 (T ₁)	72.2	281.3	5.4	14.5	5.0	132.1	53.1	45.0	3.0	37.5
10 (T ₂)	58.7	262.6	4.4	23.8	4.8	112.4	45.7	43.2	2.6	34.5
100 (T ₃)	48.5	231.5	4.4	13.2	4.5	112.2	44.2	36.3	2.5	32.5
1,000 (T ₄)	46.3	222.0	4.2	13.3	4.3	106.5	43.8	36.0	1.6	31.0
10,000 (T ₅)	36.4	211.5	3.0	12.3	4.2	105.1	43.4	29.7	1.4	27.3
100,000 (T ₆)	33.5	193.0	2.9	11.8	3.2	95.0	36.6	22.8	0.9	25.6
Fungus										
alone (T ₇)	52.2	257.6	4.7	13.8	4.7	122.9	50.8	42.0	2.6	36.8
N + F (T ₈)	41.7	212.0	3.2	13.0	4.3	112.8	42.6	30.0	1.7	27.2
N → F (T ₉)	36.0	168.4	2.1	12.8	4.0	84.4	36.1	27.8	0.9	23.8
F → N (T ₁₀)	45.7	225.8	3.8	14.0	4.7	100.8	45.3	34.0	1.8	30.9
Gen. Mean	47.6	226.5	3.9	13.3	4.4	109.1	44.4	35.0	1.9	30.8
S.E.	16.9	36.0	1.7	1.6	0.8	13.4	5.9	11.5	0.9	5.8
CD at 5% (1)	19.6	42.0	NS	NS	0.9	16.8	6.8	13.4	1.0	6.8
" (2)	21.4	46.0	NS	NS	1.0	18.3	7.5	14.6	1.2	7.4
" (3)	20.6	44.0	NS	NS	1.0	17.6	7.2	14.0	1.1	7.1

N + F = Nematode and fungus simultaneously,

N → F = Nematode 3 weeks prior to fungus

F → N = Fungus 3 weeks prior to nematode

CD at 5% (1) For comparing any treatments, except 6 and 9

" (2) For comparing treatment with 6 and 9

" (3) For comparing any treatments, with treatment 6 or 9.

TABLE 2. *Effect of individual and combined inoculation with Radopholus similis and Cylindrocarpon obtusisporum on host infestation and population build up of arecanut (Av. of 6 replication)*

Treatments	Root lesion index	Nematode population		
		Soil	Root	Total
0 (T ₁)	1.0	—	—	—
10 (T ₂)	1.8	7783.3 (3.80)	37208.3 (4.55)	44991.7 (4.64)
100 (T ₃)	3.0	10150.0 (3.94)	60783.3 (4.74)	70933.3 (4.82)
1,000 (T ₄)	3.8	12091.7 (4.07)	133383.3 (5.06)	145475.0 (5.11)
10,000 (T ₅)	4.7	19850.0 (4.25)	108433.3 (5.02)	128283.3 (9.09)
100,000 (T ₆)	5.0	23596.0 (4.34)	86480.0 (4.88)	110076.0 (5.00)
Fungus alone (T ₇)	1.0	—	—	—
N + F (T ₈)	3.4	12433.3 (4.07)	69875.0 (4.82)	82441.7 (4.90)
N → F (T ₉)	3.8	14350.0 (4.14)	80740.0 (4.90)	95090.0 (4.97)
F → N (T ₁₀)	2.2	8741.7 (3.90)	61016.67 (4.74)	69758.3 (4.81)
Gen. Mean	2.9	13392.0 (4.06)	79571.7 (4.84)	92961.1 (4.91)
S.E	0.76	0.22	0.20	0.17
CD at 5% (1)	0.88	0.26	0.23	0.20
„ 2)	0.97	0.28	0.25	0.22
„ 3)	0.93	0.27	0.24	0.21

Figures in parentheses are log transformed values.

N + F — Nematode and fungus simultaneously

N → F — Nematode 3 weeks prior to fungus

F → N — Fungus 3 weeks prior to Nematode

CD at 5% (1) For comparing any treatments except T₆ and T₇.

„ (2) For comparing treatments with T₆ and T₇.

„ (3) For comparing any treatments, with T₆ or T₇.

and plants inoculated with fungus alone. Maximum root lesion indices (5.0) were observed with 1,00,000 nematodes. The rate of nematode multiplication was inversely proportional to the inoculum level, with maximum at the lowest level and minimum at the highest level. The final population from soil and root was

found to be maximum at 1,00,000 and 1,000 nematodes per plant respectively (Table 2). Such relationships have been established with *R. similis* on turmeric (Sosamma *et al.*, 1979), on ginger (Sundararaju *et al.*, 1979), on coconut (Koshy and Sosamma, 1983) and on arecanut (Koshy and Sundararaju, 1983).

Plants inoculated with fungus and nematode simultaneously or one after the other recorded reduced rate of multiplication of the nematode as in the case of *R. similis* and *C. musae* on banana (Pinochet and Stover, 1980) and *R. similis* and *C. effusum* on coconut (Koshy and Sosamma, 1983).

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