SCREENING MEDICINAL PLANTS FOR ANTIHELMINTHIC PROPERTIES AGAINST DIFFERENT LIFE STAGES OF BANANA BURROWING NEMATODE, Radopholus similis [Cobb, 1893] Thorne 1949

BY

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THESIS

submitted in partial fulfilment of the requirement for the degree MASTER OF SCIENCE IN AGRICULTURE Faculty of Agriculture

Kerala Agricultural University

DEPARTMENT OF AGRICULTURAL ENTOMOLOGY COLLEGE OF HORTICULTURE VELLANIKKARA, THRISSUR KERALA, INDIA

1996



DECLARATION

I hereby declare that this thesis entitled "Screening medicinal plants for antihelminthic properties against different life stages of banana burrowing nematode *Radopholus similis* (Cobb, 1893) Thorne 1949" is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title of any other University or Society.

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CERTIFICATE

Certified that this thesis entitled "Screening medicinal plants for antihelminthic properties against different life stages of banana burrowing nematode *Radopholus similis* (Cobb, 1893) Thorne 1949" is a record of research work done independently by Ms. P. Sreeja, under my guidence and supervision and that it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to her.

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CERTIFICATE

We, the undersigned members of the Advisory Committee of Ms. P.Sreeja, a candidate for the Degree of Master of Science in Agriculture with major in Agricultural Entomology, agree that this thesis entitled "Screening medicinal plants for antihelminthic properties against different life stages of banana burrowing nematode *Radopholus similis* (Cobb, 1893) Thorne 1949" may be submitted by Ms. P. Sreeja, in partial fulfilment of the requirement for the degree.

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ABBREVIATIONS USED

.

AICRP	-	All India coordinated Research Project
BOD	-	Biological Oxygen Demand
°C	-	Degree Celcious
cm	-	Centemetre
g	-	grams
h	-	hour
ha	-	hectare
Kg	-	kilogram
μ	-	Micron
ml	-	Milli litre
ppm	-	parts per million
r pm	-	Revolutions per minute
spp	-	Species

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INTRODUCTION-

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INTRODUCTION

Banana is one of the most important fruit crops grown in India. In respect of area and production it ranks second only to mango in the country. The area under banana in the country as per 1993-94 statistics is 3,69,400 ha with a production of 1,50,000 tonnes. The area under banana in Kerala is 49.561 ha with a production of 3,15,897 tonnes. This constitutes 17.84% and 6.6% of national area and production respectively. The productivity in Kerala is only 13,410 kg/ha compared to the national average of 19,350 kg/ha.

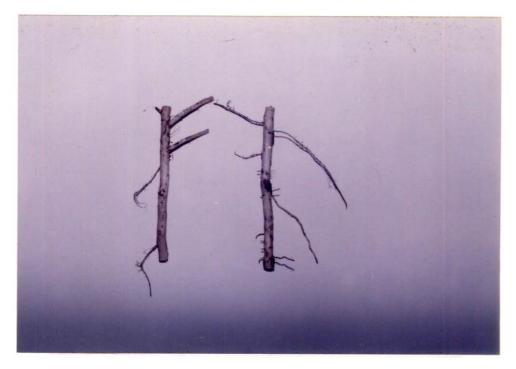
It is a recognised fact that nematode infestation constitutes one of the major limiting factors to banana production. The burrowing nematode *Radopholus similis* (cobb 1893) Thorne 1949 is the economically important pest which damage large number of roots and cause severe decline in growth of banana plant and yield reduction. The disease of banana caused by R. *similis* is known through out the world by different names, the most common are black head disease and toppling disease. R. *similis* may cause toppling of the plant and thereby cause complete loss of the bunches.

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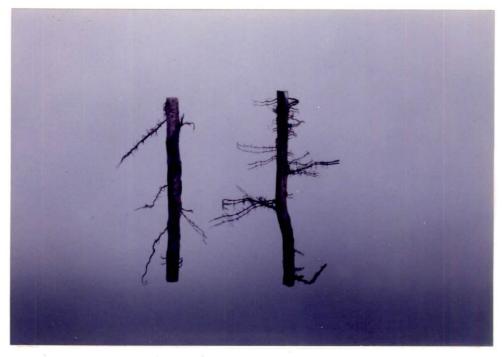
The first authentic record of *R. similis* infestation in banana from India was in the Palakkad district of Kerala (Nair *et al.*, 1966). It is now widely distributed in Kerala causing heavy root damage and yield loss (Charles and Venkitesan, 1993).

The burrowing nematode attack has been reported from the major banana growing states in our country, which include Kerala, Tamil Nadu, Maharashtra, Andhra Pradesh, Bihar and Tripura. Dipping pared suckers in pesticides or pralinage with granular nematicides are commonly followed by farmers for the control of *R. similis*. Placing together the problem of environmental pollution and cost benefit ratio for better cropping system there is a need for further advancement in nematode control systems. One of the promising alternatives is the use of plant extracts and plant products for the control of these nematode pests. The plants having nematicidal properties involves little cost, easy to apply, free from pollution hazards and have capacity to improve soil health structurally and nutritionally.

Though extracts of various plants are reported to control several plant parasitic nematodes, informations on the control of R. *similis* on banana are scanty. Hence investigations were carried out to screen extracts of plant parts of 20 species of medicinal and aromatic plants for their antihelminthic properties against different life stages of R. *similis*.



1. Normal banana root without R. similis infestatiion



2. Banana root showing symptoms of R. similis infestation

REVIEW OF LITERATURE -

factors that affect their recovery from plant tissues. The burrowing nematode being a migratory endoparasite, special techniques were followed for extracting them from roots. A method of incubating moist roots by placing them in closed jars containing a small quantity of water was the popular technique followed in earlier years (Young, 1954). Maceration of 10-20 g of root bits of 1-3 cm length, followed by incubation at 27-31°C for 2 days in 1-3 % hydrogen peroxide yielded maximum recovery of the nematodes from infected roots (Gowen and Edmounds, 1973; Whyte and Gowen, 1978; Alvarado-soto and Lopez-chaves, 1981).

Culturing of R. similis

Boncato and Davide (1980) reported successful culturing of R. similis from banana roots on sliced carrot discs in petridish containing 1% water agar and 600 ppm streptomycin sulphate at 24° to 26° for 4-6 weeks. Population developed on carrot discs were inoculated to banana and found that the nematodes caused root necrosis within two weeks. Brown and Vessey (1985) reported that R. similis multiplied on 1 g fruit callus after inoculation with 50 nematodes in aqueous streptomycin sulphate and subsequent incubation for 30 days. It can also be reared aseptically on banana shoot cultures in vitro (Mateille, 1990). Axenic culture of the nematodes following this method is advantageous for developing stock cultures for experimental purposes. Castrol and Ferraz (1990) reported multiplication of R. similis in a medium consisting of yeast extract, 2,4 sucrose and agar at 25-30°C. Mass culturing of R. similis on carrot callus tissue gave the highest population of 15,000-20,000 nematodes from an initial inaculum level of 25 nematodes within 60 days (Gnanapragasam and Prematunga, 1991).

The coconut and aracanut isolate of *R. similis* were cultured axenically on carrot discs placed on 1% water agar (Koshy and Sosamma, 1980). It was also cultured within the mesocarp of growing tender coconuts without affecting the size or quality of the nut (Koshy and Sosamma, 1982).

Control of R. similis

Nematode attack on banana arises when planting materials are collected from diseased plantation. Raising healthy suckers in *R. similis* infested soil also bring about infestation. To avoid inducing nematode pest into a new plantation planting materials may be disinfected either by paring or heat therapy. Paring the plant material by trimming away necrotic lesions and immersing in hot water at 50-55°C for 20 minutes were effective to render the planting material nematode free (Inomoto and Monteiro, 1989).

Paring the corms alone was found insufficient to eliminate R. similis (Venkitesan and Charles, 1983; Jager and Rabie, 1991). Dipping pared planting material in a nematicidal solution or coating with a nematicidal mud is useful to kill nematodes in the corm and protect infestation in early stages by nematodes.

A number of organophosphate, Oxime carbamate and carbamate nematicides are used on banana either as granular or emulsifiable concentrate formulation. Immersion of pared rhizome in DBCP at 1% for 5 minutes (Casamayor *et al.*, 1966). Phenamiphos at 100 ppm for 5 minutes (Decker *et al.*, 1971) aldicarb at 0.1 % for 30 minutes (Vankitesan and Charles, 1983), oxamyl at 0.5 % for 30 minutes (Inomoto and Monteiro, 1991) effectively controlled *R. similis*.

Other methods

Flooding the soil for 3 -9 weeks after the removal of all rhizomes lead to the disappearence of *R. similis* (Rajendran *et al.*, 1979, Sarah *et al.*, 1983, Mateille *et al.*, 1988).

Bare fallow and cultivation of horse bean (Zem and Alves, 1983), sweet potato (Ternisien and Melin, 1989), Canavalia ensiformis and Crotalaria juncea or Brachiaria decombans and sorghum (Ternisien, 1989) reduced R. similis population in a banana plantation. Ternisien and Ganry (1990) found that rotation of banana with sorghum, sweet potato, green manure legumes- Canavalia ensiformis, Desmodium pruriens, Crotalaria juncea, Mucuna pruriens, grasses- Brachiaria decumbans, and sorghum plus Macroptilium atropurpureum could eliminate R. similis. Charles et al. (1995) reported reduction of R. similis in a three year crop rotation involving banana - paddy - cowpea - paddy - elephant foot yam. The yield of banana the next best rotation was banana -tapioca - fallow sequence.

Inter cropping of banana with *Crotalaria juncea* was found to reduce *R. similis* with better growth and yield (Charles *et al.*, 1985; Naganathan *et al.*, 1988; Subramaniyan and Selvaraj, 1990).

Soil covers with black polythene, sugercane leaftrash and banana trash were reported to reduce *R. similis* in soil and root of banana (Battacharyya and Rao, 1984)

Culture extract of 17 species of microorganisms were evaluated for nematicidal activity against *R. similis* under laboratory and green house conditions. Purified extract of *Penicillium oxalicum*, *P. anatolicum*, *Aspergillus niger* and *Penicillium* sp.showed high nematicidal activity at 100 to 200 ppm. Root dip treatment of *P. oxalicum* gave best results and the control ranged from 69.1 to 85.3% (Molina and Davide, 1986). Inoculation of VAM (*Glomus fasiculatum*) seven days prior to *R. similis* inoculation resulted in vigorous root growth and reduced nematode population in soil and root (Umesh et al., 1988).

Plant extracts.

It has been known from early days that plant extracts have adverse effects on plant parasitic nematodes. The antihelminthic properties of leaf, seed and flower extracts of various indigenous medicinal plants have been reported as early as 1955 by Singh *et al.* Abivardi (1971) studied the effect of nine Iranian antihelminthic plant extracts on the root knot nematodes and concluded that *Artemisia dina*, *Portulaca oleracea*, *Thymus serpyllum* and *Coriandrum sativum* were effective at 80 ppm. Gommers (1973) tested a wide range of compositae for their ability to supress population of *Pratylenchus penetrans* in field and glass house experiments. Extracts from various parts of neem tree (leaves and fruits) have nematicidal activity against *Pratylenchus brachyurus* in maize (Egunjobi and Afalami, 1976).

Plant extracts of Ocimum basilicum, Asparagus racemosus, Argemone mexicana, Embelia ribis and Vinca rosea (Desai et al., 1973) Curcuma anadalonga (Pillai et al., 1975) Annona squamosa and Tamarindus indica (Hussain and Masood, 1975), Melia azadirachta (Haseeb et al., 1978), were reported to be nematicidal against plant parasitic nematodes. Mahmood et al. (1982) tested different concentrations of leaf and seed extracts of 12 medicinal plants against Rotylenchulus reniformis and Meloidogyne incognita and found that Anagallis arvensis, Linum usitatissimum and Sida cordifolia were highly toxic.

Leaf extracts of Aleurites cordata, Aleurites fordii and Sapium japonicum were proved effective against pathogenic nematodes (Kawazu et al., 1980). Latex of Euphorbia neriifolia, E. tirucalli, Thevetia peruviana and Pedilanthus tithymaloides were highly toxic to Hoplolaimus indicus and Tylenchus filiformis in vitro. The toxicity increased with the increase in concentration of the latex and exposure period (Siddiqui et al., 1984). A large number of plant extracts have been identified to have nematicidal properties against different nematode species The results are furnished below.

EGG STAGE Plant species Extract tested Results Reference S1. Nematode species No. Tagetes signata Leaf and root No nematicidal Sasanelli and Vitro (1991) 1. Globodera rostochiensis T. erecta effect. R. communis was Nandal and Bhatti (1986) 2. Meloidogyne javanica Calotropis procera Leaf Datura stramonium significantly Ricinus communis better in reducing Xanthium strumarium hatching. Clerodendron enermi Root Completely inhi-Patel et al. (1985) 3. M. incognita bited egg hatching at 24 h onwards. 4. M. incognita Leucaena leucocephala Leaf, podshell Inhibited egg Jain and Hasan (1985) and seed hatch.

Nematicidal properties of plants tested against nematode pests

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5.	M. incognita	Cymbopogon flexuosus	Leaf	Significantly inhibited egg hatching.	Tiyagi <i>et al</i> . (1985)
6.	M. incognita	Aloe barbadensis A. perryi Gloriosa superba	root and shoot	Shoot extract inhibited hatc- hing of larvae.	Pandey and Haseeb (1988)
7.	M. incognita	Cosmus bipinnatus Eclipta alba Sondius oleraceus Zinnia elegans	Flower, leaf stem and root	All extracts inhibited egg hatching <i>E. alba</i> showing greatest inhibition.	Banu <i>et al</i> . (1986)
8.	M. incognita	Artabotrys odoratissimus	Leaf	A significant reduction in larval hatch was observed after 24,48 and 72 h.	Chattopadhyay and Mukhopadhyaya (1989)

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9. M. incognita	Cyperus esculentus	Corm, rhizome and root	Inhibited egg hatching and reduced survival of hatched larva.	Haroon (1989)
10. M. incognita	Cassia occidentalis	Root, leaf and pod	The extract showed nematicidal and hatch inhib- itory effect.	Sarosh <i>et al.</i> (1989)
11. M. incognita	Cassia fistula Acacia arabica A. nilotica Eclipta alba Swertia chirata Datura metal Argemone mexicana	Plant	All extracts were inhibitory.	Goswami and Vijayalakshmi (1990)
12. M. incognita	Ammi majus Artemisia annua A.pallens Lactuca sativa	Plant ·	Inhibited hatching.	Pandey (1990) 11

13. M. incognita	Barringtonia sp Afzelia bijuga	Leaf, bark and fruit	Max. inhibition in larval hatch was found in the fruit extract of <i>B. speciosa</i> followed by fruit extract of <i>B.racemosa</i> .	- Salam and Sinha (1990)
14. M. incognita	Ocimum sanctum Euphorbia hitra Artemiesia absinthium Aegle marmalos	Root	Tagetes were most nematicidal foll- owed by Ocimum, Artemisia,Aegle and Euphorbia.	Sharma and Trivedi (1992) .

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 Sl. No.	Nematode species	Plant species	Extract tested	Results	Reference
1.	Anguina tritici	Ricinus communis Calotropis procera Nerium oleander	Leaf	The nematodes were sensitive to <i>N. oleander</i> at 1:5 dilution.	Verma <i>et al</i> . (1989)
2.	A. tritici	Ocimum basilicum O. sanctum Mentha piperita Callistemon lanceolatus Eugenia caryophyllata Syzygium aromaticum Cymbopogon caesius	Essential oil and the major monoterpeno- idal constit- uents.	Essential oil of S. aromaticum was found to be highly nematicidal The major constit- uents like eugenol, linalool and gera- niol were also nematicidal.	Sangwan <i>et al</i> . (1990
3.	Aphelenchoides composticola	Bougainvilleà spectabilis Calotropis procera Cedrela toona Jacaranda acutifolia Melia azedarach Ricinus communis Tagetes patula Melia azadirachta	leaf, flower and seed	All extract were toxic to nematode.	Grewal (1989) 13

4.	Heterodera cajani	Ocimum basilicum O. sanctum Mentha piperita Callistemon lanceolatus Eugenia caryophyllata Syzygium aromaticum Cymbopogon flexuosus	Essential oils and the major monoterpenoidal constituents	Essential oil of S. aromaticum was found to be highly nematicidal. The major consti- tuents like eugenol linalool and gerani were also found to be nematicidal.	l, ol ·
5.	M. javanica	Calotropis procera Nerium oleander Euphorbia caudicifolia Plumeria oblongifolia Ficus religiosa F. elastica Thevetia neriifolia	Latex	C. procera, N. olender and E. caudicifolia were highly toxic.	Zureen and Khan (1984)
6.	M. javanica	Xanthium strumarium	Root and stem	Acceptable larval mortality.	Malik <i>et al</i> . (1987)
7.	M. javanica	Datura stramonium Ipomea carnea Tagetes patula Lowsonia alba	Leaf, stem and buds	67 - 100 % mortality.	Kumari <i>et al</i> . (1987) 14

8. M. javanica	Cleome viscosa Thespesia populnea	Leaf	100% mortality.	Krishnamurthy <i>et al.</i> (1989)
9. M. javanica	Ricinus communis Calotropis procera Nerium oleander	Leaf	R. communis C. procera and N. oleander were excellent.	Verma <i>et al</i> . (1989)
10. <i>M. javanica</i>	Ocimum basilicum O. sanctum Mentha piperita Callistemon lanceolatus Eugenia caryophyllata Syzygium aromaticum Cymbopogon caesius	Essential oils and the major monoterpenoidal constituents	Essential oil of S. aromaticum was found to be highly nematicidal. The major consti- tuents like eugenol, linalool and geraniol were also found to be nematicidal.	Sangwan <i>et al.</i> (1990)
11. M. incognita	Ipomea carnea	Flower	5% solution of extract was found to be highly nematicidal.	Nikure and Lanjewar (1983)

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12. M. incognita		Leaf, stem, root and flower	Both plants exih- ibited high nema- tode toxicity. G. picta was more toxic to juvenile than I. diversifolia The flower extract in both plants showed higher toxicity than other extracts.	Tiyagi <i>et al.</i> (1985)
13. M. incognita	Xanthium strumarium Parthenium hysterophoru	Leaf	Larvae were killed in 75 and 60 minutes respectively.	Bala <i>et al</i> . (1986)
14. M. incognita	Chromolaena odorata	Plant	Nematode movement was markedly slo- wed within 3 h with total inacti- vation in 6h in 1:1 and 1:5 diluted extract. The same result was observed with 1:10 and 1:20 after 4 h.	Subramaniyan (1986) 16

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15.M. incognita	Cosmos bipinnatus Eclipta alba Sonchus oleraceus Zinnia elegans	Flower, leaf stem and root	All plant extracts were toxic with S. oleraceus giving greatest mortality follwed by extract of leaves, stem and	
16.M. incognita	Euphorbia caudicifolia Calotropis procera Opuntia sp Carica papaya Euphorbia tricalli Plumeria oblongifolia	Latex	<i>E. caudicifolia</i> and <i>C. procera</i> were highly toxic to nematode.	Maqbool <i>et al.</i> (1987)

17.M. incognita

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Calotropis gigantea Leaf Datura stramonium Leucaena leucocephala Tridax procumbans

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Effective at Mani and Chitra (1989) 500-1000 ppm con and caused a high mortality of the IInd stage larva.

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18.M. incognita	Eucalyptus citriodora Cymbopogon martinii Nardosytachys jatamansi Anethum sowa Acorus calamus Millettia ovalifolia Mesua ferrea Chromolaena odorata	Essential oil and petrolium ether extract.	100% mortality except <i>E. citr- idora</i> and <i>Chromolaena</i> .	Saxena <i>et al</i> . (1990)
19.M. incognita	Lantana camara	Leaf extract.	100 % mortality	Chandel and Mehta (1990)
20.M. incognita	Antigonon leptopus	Flower, leaf	Flower extract was more toxic than leaf or stem extracts.	Ahmad and Khan (1991)
21.M. incognita	Thuja orientalis Ocimum sanctum	Root, stem leaf, fruit and inflorescence.	Inflorescence and fruit extract were morenematicio	Fazal and Husain (1991) ial.

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Juvenile mobility Mani et al. (1986) Root and leaf. Tagetes erecta 22. Tylenchulus was decreased. Brassica compestris semipenetrans Neem showed 19.27% Vinca rosea mobility at 5% Azadirachta indica dilution.

23.T. semipenetrans

Ricinus communis Calotropis procera Nerium oleander

Leaf.

Verma et.al. (1989) Leaves of C. procera and N. oleander showed significant activity.

24.T. semipenetrans

Ocimum hasilicum O. sanctum Mentha piperita Callistemon lanceolatus Eugenia caryophyllata Syzygium aromaticum Cymbopogon caesius

Essential oils and the major monoterpenoidal was found to be constituents.

Sangwan et al. (1990) Essential oil of S. aromaticum highly nematicidal. The major constituents like eugenol, linalool and geraniol were also exihibited nematicidal property.

SI. No.			ADULT STAGE		
	Nematode species	Plant species	Extract tested	Results	Reference
1.	Anguina tritici	Cymbopogon martinii C.flexuosus C.winterianus	Essential oils and major con stituents like geraniol,citr- gnellol and citranellal.	All were toxic to nematode.	Sangwan <i>et al</i> . (1985)
2.	Aphelenchoides composticola	Bougainvillea pectabilis Calotropis procera Cedrela toona Jacaranda acutifolia Melia azedarach Ricinus communis Tagetes patula Melia azadirachta	Leaf, Flower and seed.	All extracts were toxic to nematodes.	Grewal (1989)

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3. A.composticola	Ricinus communis Calotropis procera Chrysanthemum indicum Azadirachta indica Cannabis sativa Eucalyptus hybrida	Plant.	R. communis and C. procera caused 100% mortality after 72 h. C. indicum, A. indica, C. sativ E. hybrida caused 70% mortality.	va
4. Helicotylenchus dihystera	Parthenium hysterophorus	Leaf, Stem and root.	Leaf extract killed more nematodes than ro and stem extracts. 100% mortality w observed after 24 48 h of exposure.	as
5. H. dihystera	Punica granatum Thymus vulgaris Artemisia absinthium	Plant	P. granatum caused 95.7% mortality. T. vulgaris and A. absinthium caused 71.4% and 42.9% mortality after 72 h.	Korayem <i>et al.</i> (1993)

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6. H. indicus	Cymbopogon flexuosus	Leaf	20% mortality. only.	Hasan and Jain (1984)
7. H. indicus	Euphorbia neriifolia E. tirucalli Pedilanthus tithymaloide. Calotropis procera Thevetia peruviana Nerium indicum	Latex. s	Highly toxic.	Siddiqui <i>et al.</i> (1984)
8. Heterodera avenae	Ricinus communis Bougainvillea spectabilis	Leaf extract.	R. communis caused 100% mortality and B. spectabilis resulted in 71.45% mortality.	Bhattiand Verma (1991)
9. H. avenae	Cymbopogon martinii C. flexuosus C. winterianus	Essential oil and major con- stituents like gereniol, citral citronellol and citranellal.	All were toxic to nematode.	Sangwan <i>et al.</i> (1985)

10. Heterodera cajani	Ocimum basilicum O. sanctum Mentha piperita Callistemon lanceolatus Eugenia caryophyllata Syzygium aromaticum Cymbopogon flexuosus		Essential oil of S. aromaticum I was found to be highly nematicidal. The major consti- tuents like eugenol, linalool and geraniol were also toxic.	Sangwan <i>et al</i> . (1990)
11. Hoplolaimus indicus	Euphorbia neriifolia E. tirucallli Pedilanthus tithymaloides Calotropis procera Thevetia peruviana Nerium indicum	Latex	Highly toxic.	Siddiqui <i>et al</i> . (1984)
12. H. indicus	Cymbopogon citratus	Leaf	Highly toxic.	Tiyagi <i>et al</i> . (1986)
13. M. javanica	Cymbopogon martinii C. flexuosus C. winterianus	Essential oils and major con- stituents like geraniol, citral citronellol and citronellal.	All were toxic to nematode.	Sangwan <i>et al.</i> (1985)
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14. M. javanica	Achillea santolinea Euphorbia tinctoria Heliotropium europaeum Serratula carinthefolia	Plant	Killed nematodes.	Al.obaedi <i>et al</i> . (1987)
15. M. incognita	Parthenium hysterophorus	Leaf, stem and root	100% mortality in leaf extract after 24,48 h of exposure at 1:50 concentration.	Hasan and Jain (1984)
16. M. incognita	Cymbopogon flexuosus	Leaf	100% mortality was observed after 12 h of exposure.	Tiyagi <i>et al.</i> (1986)
17. M. incognita	Azadirachta indica Melia azedarach	Fruit, leaf flowers and bark.	Fruit extract was more effective.	Siddique and Alam(1987)

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18. M. incognita	Aloe barbadensis A. perryi Gloriosa superba Scilla indica	root and shoot.	100% mortality was found with root extract of <i>A. barbadensis</i> <i>G. superba</i> and <i>S. indica</i> after 12 h.	Pandey and Haseeb (1988)
19. M. incognita	Helianthus annus Vicia sativa	Root, stem leaf and fruit.	Leaf extract was most inhibitory.	Nisar <i>et al</i> .(1989)
20. M. incognita	Azadirachta indica Calotropis procera Ricinus communis	Leaf extract	More than 50% mortality.	Khanna (1991)
21. M. incognita	Melia azedarach Calotropis procera	Leaf	C. procera was most effective.	Akhtar <i>et al</i> . (1992)
22. M. incognita	Azadirachta indica	Leaf extract	Killed nematodes	Wani (1992)
23. Rotylenchulus reniformis	Mentha viridis Cassia fistula Cordia myxa Carissa carandas Clocaria antiguorum Dalbergia sisso	Plant	All extract were active against nematode.	Haseeb <i>et al</i> . (1982)
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24. R. reniformis	Cymbopogon flexuosus	Leaf	Toxic to nematode.	Tiyagi <i>et al</i> . (1986)
25. R. reniformis	Azadirachta indica Melia azedarach	Seeds were tre- ated with fruit leaf,flower and bark extract.	reduced	Siddiqui and Alam (1987)
26. R. similis	Tagetes patula	Leaf	All were killed or inactivated after 4h in 1:1 and 1:5 dilutions.	Subramaniyan and Selvaraj (1988)
27. R. similis	Glyricidia maculata Ricinus communis Crotallaria juncea	Leaf	All were lethal to the nematode at 1:10 concent- ration within 24h.	Jasy and Koshy (1992)

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28.	Tylenchulus filiformis	Euphorbia neriifolia E. tirucalli Pedilanthus tithymaloides Calotropis procera Thevetia peruviana Nerium indicum	Latex	Highly toxic.	Siddiqui <i>et al</i> . (1984)
29.	T.semipenetrans	Cymbopogon martinii C. flexuosus C. winterianus	Essential oils and major cons- tituents like geraniol,citral citronellol and citronellal.		Sangwan et al. (1985)
30.	T. semipenetrans	Cyperus esculentus	Corn, rhizome and root	mortality was higher when treated with corm and rhizome extract.	Haroon (1989)

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31. T. semipenetrans	Ocimum basilicum O. sanctum Mentha piperita Callistemon lanceolatus Eugenia caryophyllata Syzygium aromaticum Cymbopogon caesius	Essential oil and the major monoterpeno- idal constit- uents.	Essential oil of S. aromaticum was was found to be highly nematicidal The major constit- uents like eugenol, inalool and gera- niol were also nematicidal.	Sangwan <i>et al.</i> (199
32. Xiphinema basiri	Tagetes patula	Leaf	Better nemato- static property.	Rajvanshi <i>et al.</i> (198
33. X. index	Capsicum annum	Pod	Higher nematode mortality.	Sasanelli and Catala: 1991).

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MATERIALS & METHODS-

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MATERIALS AND METHODS

Laboratory experiments were conducted at the College of Horticulture, Vellanikkara to screen medicinal and aromatic plants for antihelminthic properties against different life stages of banana burrowing nematode *R. similis*.

A. Collection and extraction of R. similis.

The *R. similis* infested banana roots collected from banana Research Station, Kannara were cleaned free of soil particles adhering to them by holding them in a stream of tap water. The cleaned roots were then split longitudinally into two halves and then cut into small bits of 2.5 cm and were kept in water contained in 15 cm diameter Petri dishes for 72 h at 20-25°C. At every 24 h the nematodes were collected by passing the suspension through 20 (40 u), 60 (250 u) mesh sieves to remove the root bits and finally collected on 400 (38 u) mesh sieve and transferred to 100 ml beaker. The nematode suspension, thus obtained was cleared by pouring it on a layer of tissue paper supported by a wire gauze in a Petri dish containing water to touch the bottom of the wire gauze and was kept for 24 h. The nematode suspension from the Petri dishes was poured in to 100 ml beakers and the volume was reduced to 25 ml after the settling of the nematodes. The active *R. similis* (females and larvae) were hand picked from the nematode suspension under a stereo microscope.

B. Preparation of Carrot Callus tissue for culturing R. similis.

Fresh and healthy Carrot tubers (*Dacus carota* L) were selected and washed thoroughly with a strong jet of water to remove the adhering dust



3. carrot kept for callus initiation on Agar media.

particles. Under the laminar flow individual tubers were dipped in 95% ethyl alcohol, flammed, pared and sliced into disc of 8 to 10 cm thickness using a sterile razor blade. Individual disc was then transferred in to a sterile 100 ml Erlenmayer conical flask containing 10 ml of 1% sterilised solidified agar. These flasks were kept on a laboratory table in the air conditioned room for one week to observe contaminations, if any , and also for initiation of callus growth (Koshy and Sosamma, 1980).

C. Inoculation of R. similis on Carrot Callus tissue.

Pure suspension of *R. similis* collected from banana roots were pipetted out into sterile centrifuge tubes and centrifuged for 1 minute at 3000 rpm. The supernatant was decanted leaving about 0.5 ml suspension at the bottom of the tube. Two ml of mercuric chloride (0.1%) was added to the tubes and the centrifuge was run again for 1 minute followed by the removal of the supernatant and rinsing of the nematode suspension twice with sterile water with 15 seconds centrifugation each time. The mercuric chloride treated population was then washed thoroughly with 0.1% streptomycin sulphate similar to mercuric chloride.

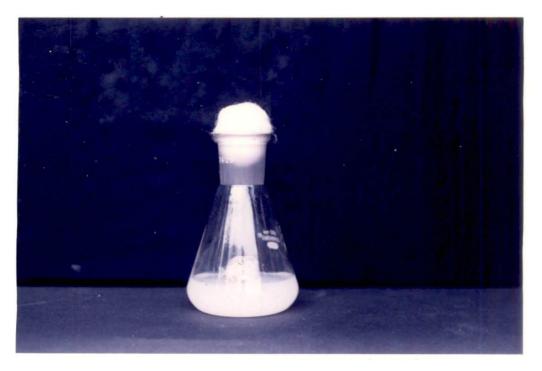
About 0.5 ml of this treated nematode population was then drawn out with a sterile syringe and inoculated directly on callus tissue developed on the carrot disc under the laminar flow. The inoculated flasks were labelled and kept in a B.O.D incubator at $25\pm1^{\circ}$ C.

D. Subculturing of R. similis in carrot disc.

Discolouration of the carrot disc was taken as indication of multiplication of nematodes. After 45 days of incubation browning of the



4. Carrot callus for R. similis multiplication



5. Callus showing multiplication of R. similis

carrot disc occurred and subculturing was done at this time. Five ml sterile water was syringed out on to the infested carrot disc and the flask was shaken gently for 2 minutes. The suspension thus formed was drawn out and a few drops were syringed out on to each new carrot disc.

Nematode suspension obtained by washing the infested carrot disc was cleared by passing it through tissue paper kept over a wire gauze. The active larvae and adults were used for test in the laboratory.

E. Plants screened for antihelminthic property.

The following 20 species of plants were selected from the herbal garden maintained by AICRP on Medicinal and Aromatic plants at The College of Horticulture, Vellanikkara to screen them for antihelminthic property against larval and adult stages of banana burrowing nematode.

SI. No.	Selected plant spp.	Common Name* Famil	tested.
1.	Azadirachta indica	Ncem	
	A.Juss Syn. Melia azadirachta	(Veppu) Meliaceae	Lcaf
2.	Piper betle L	Betel vine Piperaceae (Vettila)	Leaf
3.	Moringa oleifera Lam Syn. M. pterygosperma Gaertn	Drumstick Moringaccae (Muringa)	Leaf
4.	Mentha piperita L	Mint Labiatae (Simapothina)	Leaf
5.	Cassia angustifolia	Vahl Senna Caesalpiniacea (Chinnamukki)	ae Lcaf
6.	Piper longum. L	Thippalli Piperaceae	Lcaf
7.	Annona squamosa L	Custard apple Annonaceae (Sitaphal)	Lcaf

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* Name given in parenthesis are Malayalam names.

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51. No.	Selected plant spp.	Common Name [•]		Plant part tested.
8.	Kalanchoe pinnata Pers Syn. Bryophyllum Galy calycinum Salish	Bryophyllum (Murikootti)	, Crassolaceae	
9.	Lawsonia inermis L	Henna (Mylanchi)	Lythraceac	Leaf
10.	Glycosmis pentaphylla (Retz) correa Syn. G. cochin Chinensis	Bannimbu (Panal) L	Rutaceae	Lcaf
11.	Carica papaya L	Papaya	Caricaceae	Unripc fruit
12.	Psidium guajava L	Guava (Pera)	Myrtaceae	Unripe fruit
13.	Melia azedarach L	Persian lilac (Malaveppu)	Meliaceae	Unripc fruit 4
14.	Cleome viscosa	Sticky cleome (Kattukaduku)	Cleomaceae	Seed

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* Name given in parenthesis are Malayalam names.

SI. No.	Selected plant spp.	Common Name	Family	Plant part tested.
15.	Entada scandens Benth	Nicker bean	Mimosaceae	Seed
	Syn. <i>E. phaseoloides</i>	(Paranda) Merril		
16.	Cyperus rotundus L	Nut grass (Muthanga)	Cyperaceac	Rhizome
17.	Acorus calamus L	Sweet flag (Vayambu)	Araceae	Rhizome
18.	Indigofera tinctoria L	Indigo (Neela amari)	Papilionaceae	Root
19.	Solanum indicum L	Indian night shade (Puthari chunda)	Solanaceae	Root
20.	Euphorbia hirta	Wal dudhi (Nila pala)		hole ant
Na	ame given in parenthesis are	Malayalam names.	·	

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F. Preparation of stock solution and their dilutions

Aqueous extract of fresh plant parts specified in this study was prepared by following the technique of Kumari *et al.* (1986).

Six grams each of leaf, fruit and root was separately ground in 15 ml of sterile distilled water using a pestle and mortar. To remove the plant debris the macerated material was passed through fine muslin cloth, centrifuged for 5 minutes at 1000 rpm and then filtered through Whatman filter paper No:1. The resultant solution was treated as stock solution. Different dilutions , ie, 1:5, 1:10, 1:20 and 1:40 were prepared by adding required quantity of sterile distilled water to 1 ml of the stock solution . These dilutions were used for testing their antihelminthic property.

G. Evaluation of Plant extract for the control of R. similis

Ten active nematodes of both larval and adult stages were picked into a drop of water kept on a glass slide. Then they were transferred to sterile cavity blocks containing different dilutions of the plant extracts using a pipette. The nematode stages kept in sterile distilled water served as control. There were three replications for each treatment . The cavity blocks containing nematodes were kept undisturbed in a BOD incubator at $25\pm1^{\circ}$ C. The number of dead and surviving nematodes were counted after 24, 48 and 72 h under the stereo microscope. Death of the nematode was ascertained by touching the inactive nematode with a feather pick. The dead nematodes were transferred to sterile cavity blocks containing distilled water for 24 h to find out its reversibility.

H. Statistical analysis

Statistical analysis was done by MSTAT package available at the Computer Centre, College of Horticulture, Vellanikkara. In each table first dilution *is* considered as C1 and first time factor as T1. The interaction between dilution and time is expressed as C1T1, C2T2 etc.

RESULTS-

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RESULTS

Nematicidal and nematostatic effects of twenty different plant spp at 1:5, 1:10, 1:20 and 1:40 dilutions on R. *similis* were tried. The effects of these dilutions were observed at 24,48 and 72 h intervals and were replicated three times. The results were statistically analysed and presented as follows.

4.1 Effect of A. Indica, M. azedarach and G. pentaphylla.

4.1.1 Larva

Of the three plant extracts tried G. pentaphylla was found to be highly effective at 1:10 dilution, the assertive results be that all the larvae were dead. So only further dilutions of the same namely 1:20 and 1:40 were included in the analysis. But it was found that there was no significant cumulative mortality when observations were recorded at the three different time intervals. On an average 81% larvae were found dead at 1:20 dilution whereas the same was around 42% at 1:40 dilution. Besides having a lethal effect G. pentaphylla also left on an average 27.7% larvae static at 1:40 dilution where as the same was only around 11.1% at 1:20 dilution. Further influential static effect over time was not noticed (Table 1 and Fig. 1).

A. indica and M. azedarach were also effective in causing significant mortality at all the tested doses. These were more effective at the lowest dilution of 1:5 where complete mortality was observed. On an average 90% larvae were found dead at 1:10 dilution of A. indica and M. azedarach. When dilutions of the same at 1:20 and 1:40 were tried, the mortality percentage were 55.5 and 33.3 for A. indica whereas the same was 56.6 and 18.8 for M. azedarach (Table 2 and Fig. 2). Table 1. Nematicidal and nematostatic effects of *G. pentaphylla* and nematostatic effects of *K. pinnata*

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	Larva dead	Larva st	Larva static	
	G. penta . phylla	G.penta phylla	K. pinnata	G.penta phylla
1:20	81	11.1	48.8	13.3
	(1.133)	(0.338)	(0.774)	(0.364)
1:40	42	27.7	16.6	40
	(0.704)	(0.540)	(0.441)	(0.683)
T1	56	20	35	30
	(0.863)	(0.442)	(0.620)	(0.567)
T2	61.6	18.3	33.3	25
	(0.916)	(0.425)	(0.603)	(0.509)
Т3	66.6	20	30	25
	(0.977)	(0.449)	(0.554)	(0.502)
C1T1	7.66	10	50	16.6
	(1.081)	(0.322)	(0.785)	(0.416)
C1T2	80	13.3	46.6	13.3
	(1.116)	(0.369)	(0.752)	(0.369)
C1T3	86.6	30	50	1.
	(1.202)	(0.562)	(0.785)	(0.322)
C2T1.	36.6	26.6	20	4.332
	(0.645)	(0.529)	(0.455)	(0.718)
Č2T2	43.3	26.6	20	36.6
	(0.717)	(0.529)	(0.455)	(0.648)
C2T3	46.6	26.6	10	40
	(0.752)	(0.524)	(0.322)	(0.682)
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CD1	0.13	0.151	0.09	0.1045
CD2	NS	NS	NS	NS

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.

NS

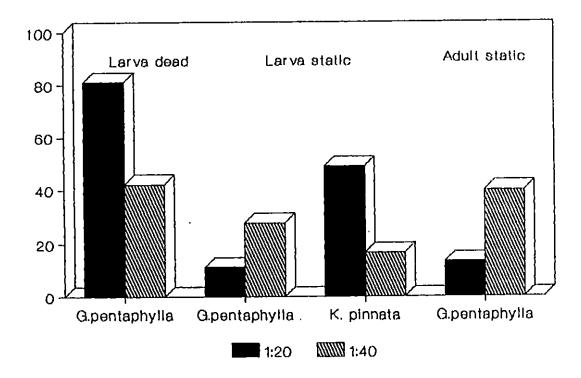
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Nematostatic and Nematicidal effect of G.pentaphylla and K.pinnata

Flg. 1

In contrast to the low retention effect of G. pentaphylla, A. indica and M. azedarach showed slight retention effect, as could be evidenced from the fact that 67.7% larvae were found dead after 72 h for A. indica and 61.1% for M. azedarach. But no interactive effect of the dilution over time was observed.

4.1.2. Adult

All the three plant extracts had lethal effects on adult also. In close analogy with the results discussed in the larval stage, *G. pentaphylla* was most effective resulting in 94.4% mortality where as 86.6% and 82.2% adults were respectively killed by *A. indica* and *M. azedarach* at 1:10 dilution. At 1:20 dilution around 83.3% adults were found dead due to the effect of *G. pentaphylla*, where as the same was around 61.1% and 56.6% for *A. indica* and *M. azedarach* respectively. For all the three plant extracts the mortality rate dropped at 1:40 dilution considerably (Table 2 and Fig. 2).

In contrast to its low retension effect on larval stage G. pentaphylla did have slight but significant retention effect at all the three time lapse intervals. A significant interactive effect of the dilution over time was observed only for A. indica. The mortality percentage being 100 at 1:10 dilution when observed after 72 h.

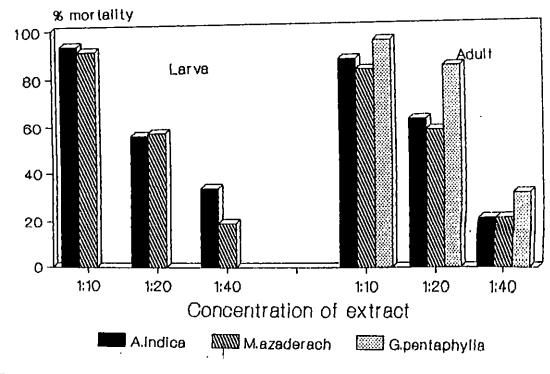
Besides having lethal effects, G. pentaphylla exhibited significant nematostatic effect on adult at 1:20 and 1:40 dilutions (Table 1 and Fig. 1).

Table 2.

Effect of A. indica, M. azedarach on larval and A. indica, M. azedarach and G. pentaphylla on adult stages of R. similis

M. azeda		l stage	Adult stages of R. similis Adult stage		
	A. indica	M.azedar ach	A.Indica	M.azedar ach	G.pentaph ylla
1:10	93.3	91.1	86.6	82.2	94.4
	(1.308)	(1.272)	(1.220)	(1.160)	(1.329)
1:20	55.5	56.6	61.1	56.6	83.3
	(0.844)	(0.853)	(0.901)	(0.853)	(1.157)
1:40	33.3	18.8	20	20	30
	(0.609)	(0.442)	(0.458)	(0.454)	(0.592)
Tl	52.2	48.8	43.3	44.4	62.2
	(0.811)	(0.770)	(0.706)	(0.721)	(0.925)
Т2 `	62.2	56.6	58.8	54.4	70.0
	(0.942)	(0.870)	(0.887)	(0.834)	(1.029)
ТЗ	67.7	61.1	65.5	60.0	75.5
	(1.008)	(0.927)	(0.987)	(0.907)	(1.104)
CITI	83.3	83.3	73.3	73.3	86.6
	(1.154)	(1.154)	(1.030)	(1.042)	(1.127)
C1T2	96.6	93.3	86.6	83.3	96.6
	(1.358)	(1.303)'	(1.202)	(1.176)	(1.358)
С1Т3	100	96.6	100	90.0	100
	(1.412)	(1.358)	(1.424)	(1.262)	(1.412)
C2T1	46.6	50.0	43.3	46.6	76.6
	(0.750)	(0.758)	(0.718)	(0.750)	(1.068)
С2Т2	53.3	56.0	66.6	60	83.3
	(0.819)	(0.853)	(0.456)	(0.888)	(1.154)
C2T3	66.6	63.3	73.3	63.3	90
	(0.961)	(0.921)	(1.030)	(0.921)	(1.244)
C3T1	26.6	13.3	13.3	13.3	23
	(0.529)	(0.369)	(0.364)	(0.369)	(0.490)
СЗТ2	36.6	20	23.3	20	30
	(0.650)	(0.455)	(0.502)	(0.455)	(0.576)
СЗТЗ	36.6	23.3	23.3	26.6	36.6
	(0.650)	(0.502)	(0.502)	(0.537)	(0.650)
CD1	0.1199	0.08	0.06	0.149	0.115
CD2	0.1199	0.08	0.06	0.149	0.115
CD3	NS	NS	0.116	NS	NS

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.



Effect of M.azedarach, A.Indica & G.pentaphylla on the mortality % of R.similis

Fig. 2

Effect of M.azedarach, A.indica & G.pentaphylia on the mortality % of R.similis

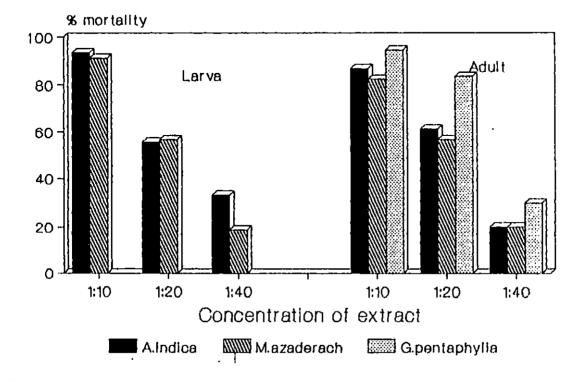


Fig. 2

4.2 Effect of K. pinnata and E. hirta

4.2.1 Larva

The toxic effect of K. pinnata and E. hirta reduced considerably when the strength of the solution was diluted from 1:5 to 1:10 and 1:20. The effect of 1:40 dilution was on par with control. The mortality percentage reduced from 92.2% to 25.5% for K. pinnata and 92.2% to 20% for E. hirta, when the stock solution was diluted by 15% from 1:5. Cumulative effect over time was evidenced only for K. pinnata whereas no such effect was observed for E. hirta. No interactive effect was observed for both the extracts (Table 3 and Fig. 3).

Besides lethal effect K. pinnata showed significant nematostatic effect at 1:20 and 1:40 dilutions. At 1:20 dilution around 48.8% larvae were found to be static where as it was 16.6% at 1:40. At the same dilution there was no nematicidal property (Table 1 and Fig. 1).

4.2.2 Adult

The toxicity of *E. hirta* on the adult was observed only at lower dilutions of 1:5 and 1:10 (Table 8 and Fig. 7). But significant cumulative effect was noticed over time. Whatever be the dilution, on an average *E. hirta* left 20% - 30% adult as nematostatic (Table 7 and Fig. 6). Significant interactive as well as cumulative effect over time could not be noticed. In contrast to the toxic effect of *E. hirta* (Table 8 and Fig. 7). *K. pinnata* was effective at the three doses tested. But no interactive effect of dilution over time was noticed. The percentage mortality dropped from 80 % to 18.8 % as the concentration was diluted from 1:5 to 1:20. At 1:40 there was no mortality (Table 3 and Fig. 3).

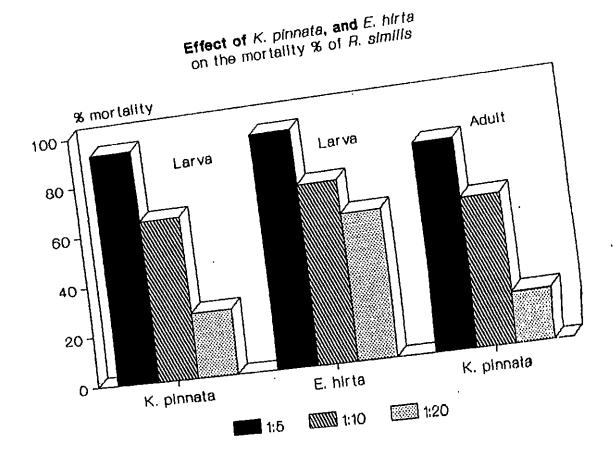
Table 3. Effect of K. pinnata and E. hirta on the mortality of R. similis larva and K. pinnata on the adult.

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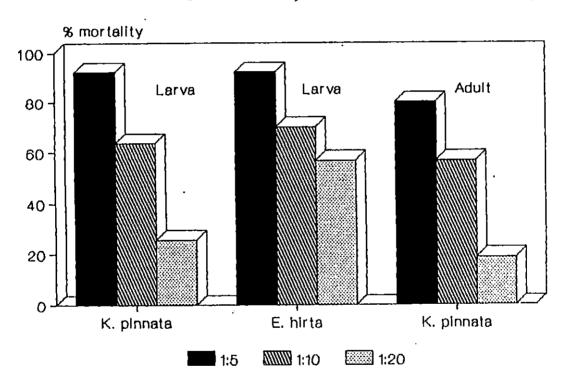
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	Larva	Larva	Adult
	K. pinnata	E. hitra	K. pinnata
1:5	92.2	92.2	80
	(1.295)	(1.295)	(1.122)
1:10	63.3	70	56.6
	(0.925)	(0.996)	(0.855)
1:20	25.5	20	18.8
	(0.523)	(0.447)	(0.438)
T1	53.3	55.5	42.2
	(0.824)	(0.850)	(0.689)
T2	61.1	61.1	51.1
	(0.925)	(0.916)	(0.795)
Т3	66.6	65.5	62.2
	(0.994)	(0.972)	(0.931)
CITI	83.3	86.6	70
	(1.163)	(1.217)	(0.995)
C1T2	93.3	93.3	80
	(1.310)	(1.310)	(1.116)
C1T3	10	96.6	90
、	(1.412)	(1.358)	(1.256)
C2T1	56.6	63.3	46.6
	(0.854)	(0.426)	(0.750)
C2T2	63.3	70	56.6
	(0.926)	(0.995)	(0.854)
C2T3	70	76.6	66.6
	(0.995)	(0.068)	(0.961)
C3T1	20	16.6	10
	(0.455)	(0.408)	(0.322)
C3T2	26.6	20	16.6
	(0.537)	(0.443)	(0.416)
СЗТЗ	30	23.3	30
	(0.576)	(0.490)	(0.576)
CD1	0.13	0.16	0.13
CD2	0.13	NS	0.13
СD3	NS	NS	NS

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.







Effect of K. pinnata, and E. hirta on the mortality % of R. similis

Flg. 3

4.3 Effect of P. longum

4.3.1 Larva

The nematicidal activity of leaf extract of *P. longum* is described in Table 4. A persual of the data indicate that the extract was very effective in causing significant larval mortality at lower dilutions. The nematicidal effect reduced sharply from 82.2% to 11.1% when the ecxtract was diluted from 1:5 to 1:40. Cumulative effect over time was evidenced from the fact that 50% larvae were dead after 72 h of exposure. A significant interactive effect of the dilution over time was observed. It is clear from the Table 4 and Fig. 4 that the mortality rate was 100% at 1:5 dilution when observed after 72h.

4.3.2 Adult

In contrast to the effect on larval stage the nematicidal activity was observed only at three dilutions of the plant extract. At 1:40 the effect was on par with control. Here also the effect dropped from 65.5% to 30% on dilution from 1:5 by 15%. A significant retention effect was noticed at the three time lapse intervals (Table 4 and Fig. 4).

4.4 Effect of C. rotundus

4.4.1 Larva

The data pertaining to the nematicidal effect of *C. rotundus* is presented in the Table 5 and Fig. 5. At lower dilution of 1:5 *C. rotundus* killed all larvae within a short duration of 24 h. Hence only the other higher dilutions were satistically analysed. On further dilution of the extract the .

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and adult. • Larva Adult				
Larva				
Dilution	P. longum	Dilution	P. longum	
1:5	82.2 (1.180)	1:5	65.5 (0.991)	
1:10	42.2 (0.704)	1:10 .	57.7 (0.867)	
1:20	27.7 (0.549)	1:20	30 (0.567)	
1:40	11.1 (0.333)			
Т1	26.6 (0.516)	Tl	28.8 (0.553)	
Т2	44.1 (0.735)	T2	54.6 (0.845)	
ТЗ	51.6 (0.824)	ТЗ	70 (1.027)	
C1T1	53.3 (0.819)	CITI	26.6 (0.537)	
C1T2	93.3 (1.310)	C1T2	73.3 (1.067)	
C1T3	100 (1.412)	C1T3	96.6 (1.369)	
C2T1	30 (0.576)	C2T1	43.3 (0.713)	
C2T2	43.3 (0.718)	C2T2	60 (0.893)	
C2T3	53.3 (0.819)	C2T3	70 (0.995)	
C3T1	20 (0.455)	С3Т1	16.6 (0.408)	
C3T2	26.6 (0.541)	СЗТ2	30 (0.576)	
СЗТЗ	36.6 (0.650)	СЗТЗ	43.3 (0.718)	
C4T1	3.3 (0.213)			
C4T2	13.3 (0.369)			
C4T3	16.6 (0.416)			
CD1	0.0986		0.18	
CD2	0.085		0.18	
CD3	0.170		NS	

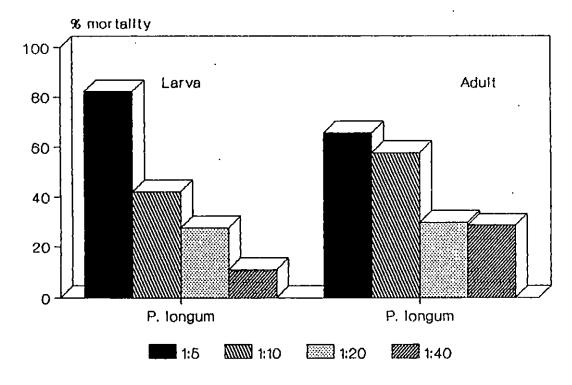
Table 4. Effect of P. longum on the mortality of R. similis larva and adult.

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CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.

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Effect of P. longum on the mortality % of R. similis

Flg. 4

mortality rate started diminishing from 83% to 15.5% when extract was diluted from 1:10 to 1:20. A cumulative effect over time was noticed as 63.3% mortality of the larvae were observed after 72 h exposure. Significant interactive effect of the dilution over time was noticed, the mortality rate being 100% at 1:10 dilution after 48 h interval. Besides nematicidal effect *C. rotundus* has shown significant nematostatic effect. It left 16.6%, 65.5% and 14.4% larvae static at 1:10, 1:20 and 1:40 dilutions (Table 6 and Fig. 6).

4.4.2. Adult.

In close analogy to the results observed in case of the larva, C. rotundus was effective in killing all the adults at 1:5 dilution. The nematicidal effect dropped considerably on further dilution of the extract. On an average 73.3% nematodes were found dead at 1:10 dilution, whereas the same was only about 12.2% at 1:20 dilution. Further, a significant cumulative effect over time was noticed. As in the case of larvae, a significant interactive effect of dilution over time was noticed, the mortality rate being 100% at 1:10 dilution after 72 h. In addition to the nematicidal effect, C. rotundus exhibited significant nematostatic effect (Table 5 and Fig. 5).

4.5 Effect of P. betle and I. tinctoria

4.5.1 Larva

The data showing the effect of *P. betle* and *I. tinctoria* is presented in the Table 8 and Fig. 7.

	Larva	Adult	Mobility adult
	C. rotundus	C. rotundus	C. rotundus
1:10	83 (1.201)	73.3 (1.091)	24 (0.449)
1:20	15.5 (0.385)	12.2 (0.343)	16.6 (0.413)
T1	25 (0.472)	11.6 (0.331)	46.6 (0.742)
T2	60 (0.931)	55 (0.863)	6.6 (0.264)
Т3	63.3 (0.976)	61.6 (0.957)	8.3 (0.288)
CIT1	50 (0.785)	23 (0.502)	73 (1.030)
_C1T2	100 (1.407)	96 (1.358)	0 (0.159)
C1T3	100 (1.412)	100 (1.142)	0 (0.159)
C2T1	0 (0.159)	0 (0.159)	20 (0.455)
C2T2	20 (0.455)	13.3 (0.369)	13.3 (0.369)
C2T3	26.6 (0.541)	23 (0.502)	16.6 (0.416)
CD1	0.074	0.065	0.077
CD2	0.09	0.68	0.077
CD3	0.128	0.113	

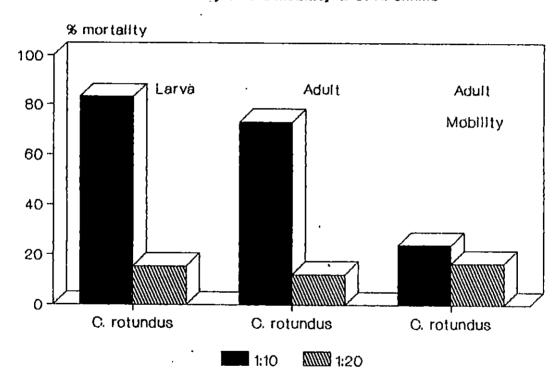
Table 5. Effect of C. rotundus on the mortality (Larva and Adult) and mobility (Adult) of R. similis.

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.

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	C. rotundus	C. rotunaus	C. LOCUMUNS
1:10	83 (1.201)	73.3 (1.091)	24 (0.449)
1:20	15.5 (0.385)	12.2 (0.343)	16.6 (0.413)
T1	25 (0.472)	11.6 (0.331)	46.6 (0.742)
T2	60 (0.931)	55 (0.863)	6.6 (0.264)
ТЗ	63.3 (0.976)	61.6 (0.957)	8.3 (0.288)
C1T1	50 (0.785)	23 (0.502)	73 (1.030)
C1T2	100 (1.407)	96 (1.358)	0 (0.159)
C1T3	100 (1.412)	100 (1.142)	0 (0.159)
C2T1	0 (0.159)	0 (0.159)	20 (0.455)
C2T2	20 (0.455)	13.3 (0.369)	13.3 (0.369)
C2T3	26.6 (0.541)	23 (0.502)	16.6 (0.416)
CD1	0.074	0.065	0.077
CD2	0.09	0.68	
CD3	0.128	0.113	

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.



Effect of *C. rotundus* on the mortality % and mobility % of *R. similis*

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Fig. 5

From the data it is clear that both the plants showed lethal effects at lower dilutions of 1:5 and 1:10. The effect was significant in case of *P. betle* only. On an average 54% and 36% mortality was recorded at 1:5 and 1:10 dilutions respectively for *P. betle* whereas the same was only 34.4% and 22.2% for *I. tinctoria*. A high retention effect was noticed in both the cases the mortality rate being 70% after 72 h of exposure in case of *P. betle* and 41.6% in case of *I. tinctoria*. Significant interactive effect of dilution over time was also noticed for *P. betle*.

In addition to the nematicidal effect, both the plants exhibited significant nematostatic effect. *I. tinctoria* was more effective in causing immobility of the nematode. About 64% larvae were left static where as it was 45.5% at the same dilution of 1:5 in *I. tinctoria* and *P. betle* respectively. Static effect started diminishing in both cases as the strength of dilution was increased. There was no cumulative immobility when observations were recorded at 24, 48 and 72 h interval for *I. tinctoria*. On the contrary *P. betle* has shown a cumulative immobility and interactive effect of concentration over time (Table 9 and Fig. 8).

4.5.2. Adult

Among these two plant species only *I. tinctoria* has shown significant lethel effect on the adult. Mortality percentage of 28.8% and 21.1% was observed at 1:5 and 1:10 dilutions respectively. Cumulative effect over time was also noticed. But no significant interactive effect of dilution over time was noticed (Table 8 and Fig. 7).

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	C. rotundus
1:10	16.6 (1.103)
1:20	65.5 (1.050)
1:40	14.4 (0.736)
T1	41.1 (0.985)
T2	28.8 (0.948)
Т3	26.6 (0.956)
C1T1	50 (1.157)
.CIT2	0 (1.102)
C1T3	0 (1.050)
C2T1	73.3 (1.078)
C2T2	60 (1.021)
C2T3	63.3 (1.050)
C3T1	0 (1.721)
C3T2	26.6 (0.721)
СЗТЗ	16.6 (0)
CD 1	0.113
CD 2	NS
CD 3	NS

Table 6. Effect of C. rotundus on larval mobility of R. similis

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CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values. Both the plants exhibited significant nematostatic effect. The effect decreased gradually as the dilution of extract increased. At 1:5 dilution, 63.3% and 51.1% nematodes were left static for *I. tinctoria* and *P. betle* respectively. The same was around 21% and 16.6% at the higher dilution of 1:20 for *I. tinctoria* and *P. betle*. An interactive effect was also observed. The immobility rate being 86.6 at 1:5 dilution after 24 h for *P. betle*. Whereas no such effect was noticed with *I. tinctoria* (Table 7 and Fig. 6).

4.6 Effect of M. oleifera, S. indicum, M. piperta

4.6.1 Larva

The data on the effect of these plant extracts is presented in the Table 9. From the table it can be seen that among these three plant extracts M. *oleifera* was found to be highly nematostatic at 1:5 dilution followed by S. indicum and M. piperita. At the lower dilution M. oleifera left 63.3% larvae static, but at the higher dilution it was around 28.8%. For S. indicum the static effect reduced from 56.6% to 18.8% and for M. piperita from 46.6% to 16.6%. Eventhough all the effects were statistically significant, there was neither cumulative immobility nor interactive effect (Table 9 and Fig. 8).

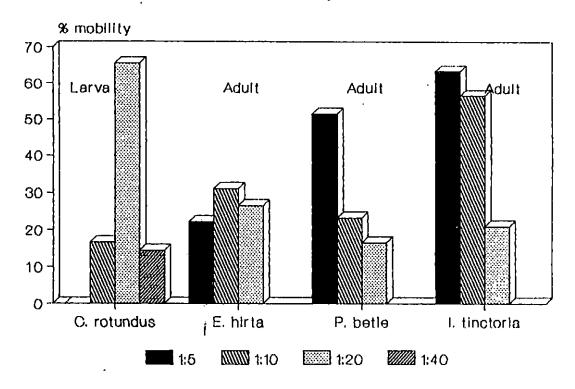
4.6.2 Adult

In close analogy to the results observed in case of larvae, here also

E. hirta	P. betle	I. tinctoria
22.2 (0.478)	51.1 (0.805)	63.3 (0.923)
31.1 (0.585)	23.3 (0.494)	56.6 (0.855)
26.6 (0.527)	16.6 (0.413)	21 (0.471)
34.4 (0.618)	44.4 (0.734)	50 (0.781)
24.4 (0.507)	32.2 (0.597)	45.5 (0.732)
21.1 (0.465)	14.4 (0.382)	45.5 (0.736)
33.3 (0.610)	86.6 (1.209)	70 (0.995)
20 (0.455)	46.6 (0.750)	63.3 (0.921)
13.3 (0.369)	20 (0.455)	56.6 (0.853)
36.6 (0.648)	30 (0.576)	60 (0.893)
30 (0.571)	26.6 (0.537)	53.3 (0.819)
26.6 (0.537)	13.3 (0.369)	56.6 (0.854)
33.3 (0.597)	16.6 (0.416)	20 (0.455)
23.3 (0.494)	23.3 (0.502)	20 (0.445)
23.3 (0.490)	10 (0.322)	23.3 (0.502)
NS	0.117	0.1418
NS	0.117	NS
NS	0.204	NS
	22.2 (0.478) 31.1 (0.585) 26.6 (0.527) 34.4 (0.618) 24.4 (0.507) 21.1 (0.465) 33.3 (0.610) 20 (0.455) 13.3 (0.369) 36.6 (0.648) 30 (0.571) 26.6 (0.537) 33.3 (0.597) 23.3 (0.494) 23.3 (0.494) 23.3 (0.490) NS NS	22.2 (0.478) 51.1 (0.805) 31.1 (0.585) 23.3 (0.494) 26.6 (0.527) 16.6 (0.413) 34.4 (0.618) 44.4 (0.734) 24.4 (0.507) 32.2 (0.597) 21.1 (0.465) 14.4 (0.382) 33.3 (0.610) 86.6 (1.209) 20 (0.455) 46.6 (0.750) 13.3 (0.369) 20 (0.455) 36.6 (0.648) 30 (0.576) 30 (0.571) 26.6 (0.537) 13.3 (0.597) 16.6 (0.416) 23.3 (0.494) 23.3 (0.502) 23.3 (0.494) 23.3 (0.502) 23.3 (0.490) 10 (0.322) NS 0.117 NS 0.117

Table 7. Effect of *E. hirta, P. betle and I. tinctoria* on the mobility of *R. similis* adult

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.



Effect of C.rotundus, E.hirta, P.betle & I.tinctoria on mobility % of R.similis

Fig. 6

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M. oleifera exhibited higher static effect compared to *M. piperita* and *S. indicum.* Static effect was seen only at the two lower dilutions of 1:5 and 1:10 (Table 10 and Fig. 9). On further dilution the effect was on par with control.

4.7 Effect of A. calamus and C. viscosa

4.7.1 Larva

Nematostatic effect was observed only at the two dilutions of 1:5 and 1:10. Around 51.1% larvae were left static at 1:5 dilution and 33.3% at 1:10 dilution. Cumulative static effect was observed. For *C. viscosa* 38.8% and 25% static effect was recorded at the two dilutions, but it was not statistically significant (Table 11 and Fig. 10).

4.7.2 Adult

The static effect dropped from 48.8% to 23.3% for *A. calamus* when the extract was diluted from 1:5 to 1:10. There was no cumulative effect. In case of *C. viscosa* the effect reduced from 36% to 18.8% when diluted from 1:5 to 1:10 (Table 11 and Fig. 10).

4.8. Effect of C. angustifolia, A. squamosa, C. papaya,P. guajava, E. scandens and L. inermis.

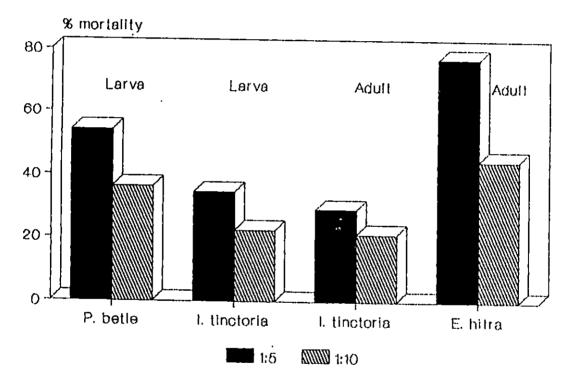
At all the dilutions these extracts failed to cause mortality of the tested nematodes. Even nematostatic property could not be seen after 72 h of exposure. All the treatments were on par with control



Table 8. Effect of *P*. betle and *I*. tinctoria on the mortality of *R*. similis larva and *I*. tinctoria and *E*. hirta on *R*. similis adult.

	LARVA			ADULT	
	P. betle	I. tincto	nia J	I. tinctomia	E. hirta
1:5	54 (0.832)	34.4 (0.617)		28.8 (0.554)	77 (1.093)
1:10	36 (0.640)	22.2 (0.478)		21.1 (0.467)	44.4 (0.727)
Т1	18.3 (0.430)	16.6 (0.412)	_	13.3 (0.369)	50 (0.786)
T2	48 (0.769)	26.6 (0.532)		25 (0.522)	63 (0.934)
ТЗ	70 (1.010)	41.6 (0.697)		36.6 (0.647)	70 (1.010)
C1T1	13.3 (0.369)	20 (0.455)		16.6 (0.416)	66.6 (0.961)
C1T2	63.3 (0.929)	33.3 (0.610)		26.6 (0.541)	80 (1.116)
C1T3	86.6 (1.202)	50 (0.785)		43.3 (0.71 <u>8)</u>	86.6 (1.202)
C2T1	23 (0.490)	13.3 (0.369)		10 (0.322)	33.3 (0.611)
C2T2	33.3 (0.611)	20 (0.455)		23.3 (0.502)	46.6 (0.752)
C2T3	53.3 (0.819)	33.3 (0.610)		3 (0.576)	53.3 (0.819)
CD1	0.128			0.07	0.12 .
CD2	0.15	0.18		0.09	0.14
CD3	0.22	_			

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.



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Effect of P.betle, I.tinctoria & E.hirta on the mortality % of R. similis

Flg. 7

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Table 9. Effect of M. oliefera, I.tinctoria, S. indicum, M. piperta and P. betle on the mobility of R. similis - larval stage.

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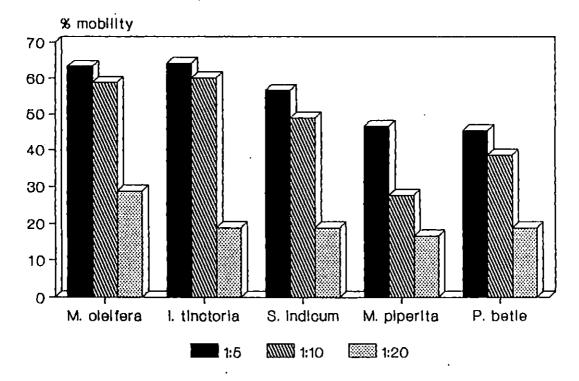
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	M. oliefera	I.tincto ria	S. indicum	M. piperila	P. betle
1:5	63.3	64	56.6	46.6	45.5
	(0.936)	(0.942)	(0.855).	(0.752)	(0.738)
1:10	58.8	60	48.8	27.7	38.8
	(0.879)	(0.891)	(0.774)	(0.549)	(0.669)
1:20	28.8	18.8	18.8	16.6	18.8
	(0.560)	(0.439)	(0.442)	(0.411)	(0.439)
т1	55.5	52.2	35.5	32.2	54.4
	(0.852)	(0.805)	(0.628)	(0.584)	(0.837)
Т2	50.0	48.8	44.4	30	32.2
	(0.784)	(0.766)	(0.718)	(0.566)	(0.596)
ТЗ	45.5	42.2	44.4	28.8	16.6
	(0.739)	(0.766)	(0.726)	(0.561)	(0.413)
C1T1	80	76.6	50	56.6	86.6
	(1.135)	(1.081)	(0.785)	(0.854)	(1.202)
C1.T2	56.6	66.6	66.6	46.6	36.6
	(0.854)	(0.961)	(0.960)	(0.752)	(0.645)
C1T3	53.3	50	53.3	36.6	13.3
	(0.820)	(0.785)	(0.820)	(0.650)	(0.369)
C2T1	60	63.3	40	26.6	56.6
	(0.893)	(0.928)	(0.683)	(0.529)	(0.854)
C2T2	63.3	63.3	50	26.6	36.6
	(0.923)	(0.928)	(0.785)	(0.537)	(0.650)
C2T3	53.3	53.3	56.6	30	23.3
	(0.820)	(0.819)_	(0.854)	(0.580)	(0.502)
C3T1	26.6	16.6	16.6	13.3	20
	(0.529)	(0.403)	(0.416)	(0.364)	(0.455)
C3T2	30.0	16.6	16.6	16.6	20
	(0.576)	(0.408)	(0.408)	(0.408)	(0.494)
С3Т3	30.0	23.3	23.3	20	13.3
	(0.576)	(0.502)	(0.502)	(0.455)	(0.369)
CD1	0.175	0.18	0.118	0.117	0.117
CD2	NS	NS	NS		0.117
CD3	NS	NS	NS	NS	0.2040

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values. 53



Effect of medicinal plant extracts on the mobility % of *R. similis* Larva

Fig. 8

Comparison of toxicity of different plant extracts.

Nematicidal property of different plant extracts was compared by calculating the toxicity index. Higher toxicity index after a short exposure of 24 h was taken as the criteria for selecting the plants as highly nematicidal (Tables 12, 13, 14, 15, 16 and 17 and Fig.11, 12; 13, 14).

Nematicidal property

Larva

G. pentaphylla > A. indica > M. azedarach > E. hirta >

K pinnata > C. rotundus > P. longum > I. tinctoria (Table 12)

Adult

- G. pentaphylla > A. indica > M. azedarach > K. pinnata >
- C. rotundus > E. hirta > P. longum > I. tinctoria. (Table 13)

Nematostatic property

Larva.

M. oleifera > P. betle > I. tinctoria > C. rotundus > A. calamus > M. piperita > C. viscosa > S. indicum > K. pinnata > E. hirta > G. pentaphylla (Table 14).

Adult

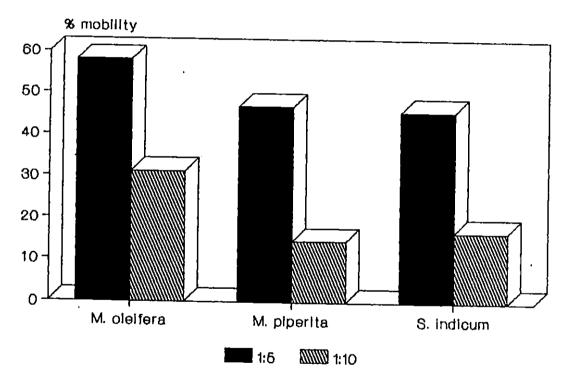
I. tinctoria > P. betle > E. hirta > M. oleifera > C. rotundus > A. calamus > K. pinnata > M. piperita > G. pentaphylla > S. indicum > C. viscosa (Table 15).

Table 10. Effect of M. oleifera, M. piperta and S. indicum on the mobility of R. similis - Adult.

	M. oleifera	M. piperta	S. indicum
1:5	58 (0.878)	46.6 (0.750)	45.5 (0.740)
1:10	31.1 (0.573)	14.4 (0.385)	16.6 (0.413)
Tl	46.6 (0.742)	33.3 (0.594)	28.3 (0.544)
T2	46.6 (0.742)	31.6 (0.584)	35 (0.618)
Т3	41.6 (0.692)	26.6 (0.524)	30 (0.569)
C1T1	63.3 (0.926)	53.3 (0.819)	43 (0.718)
C1T2	60 (0.888)	46.6 (0.752)	53 (0.819)
C1T3	53.3 (0.820)	40 (0.678)	4 (0.683)
C2T1	30 (0.557)	13.3 (0.369)	13.3 (0.369)
C2T2	33.3 (0.597)	16.6 (0.416)	16.6 (0.416)
C2T3	30 (0.564)	13.3 (0.369)	20 (0.455)

CD1	0.21	0.112	0.09
CD2	NS	NS	NS
ÇD3	NS	NS	NS

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.



Effect of M.oleifera, M.piperita & S.indicum on mobility % of R.similis Adult

Flg. 9

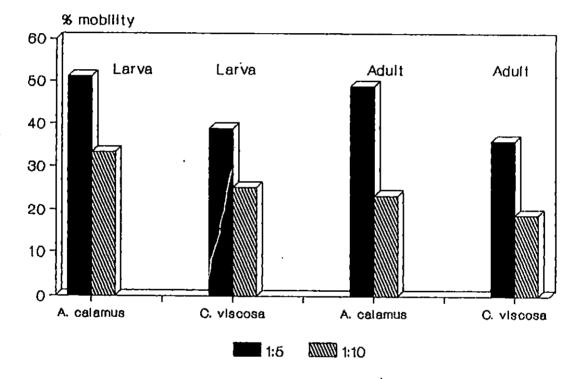
Table 11. Effect of A. calamus and C. viscosa on the mobility of R. similis - Larva and Adult.

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· · ·	I	JARVA	AD	ULT
	A. calamus	C. viscosa	A. calamus	C. viscosa
1:5	51.1	38.8	48.8	36.0
	(0.798)	(0.665)	(0.773)	(0.641)
1:10	33.3	25.0	23.3	18.8
	(0.605)	(0.524)	(0.498)	(0.445)
	53.3	41.6	38.0	25
T1	(0.822)	(0.698)	(0.645)	(0.507)
T2	43.3	28.3	36.0	31.6
	(0.715)	(0.552)	(0.644)	(0.540)
ТЗ	30	26.6	33.3	26.6
	(0.564)	(0.532)	(0.608)	(0.532)
CITI	63.3	53.3	56.6	36.6
	(0.926)	(0.820)	(0.854)	(0.645)
C1T2	50	33.3	50.0	43.3
	(0.785)	(0.611)	(0.785)	(0.717)
C1T3	40	30.0	40.0	30.0
	(0.683)	(0.562)	(0.678)	(0.562)
C2T1	43.3	3.0	20.0	13.3
	(0.717)	(0.576)	(0.455)	(0.369)
C2T2	36.6	23 [.] .3	23.0	20.0
	(0.645)	(0.494)	(0.502)	(0.464)
	20	23.3	.26.6	23.3
C2T3	(0.455)	(0.502)	(0.537)	(0.502)

CD1	0.1615	NS	0.156	0.13
CD2	0.1976	NS	NS	NS
CD3	NS	NS	NS	<u> </u>

CD1- CD dilution, CD2- CD time, CD3- CD interaction. Figures in the parenthesis are transformed values.



Effect of A.calamus & C. viscosa on the mobility % of R. similis

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Flg. 10

In vitro con	nfirma	tory e	valuat	ion o	f plant (: extr Nemat	acts f icidal	for the prope	e toxi erty)	city to) R. S	simili	ls- Lai	cva	
Plant			24	h		Perc	ent mo	ortalit 48 h	cy at	various	dilu	itions	for 72 ł	1	
	1:5		1:20		inde	x				Tox. index			•		Tox. index
G. pentaphylla	100	100								319.9					
A. indica	100	-								286.6					
M. azedarach	100	83.3	50	13.3	246.6	100	93.3	56.6	20	269.9	100	96.6	63.6	23.3	283.6
C. rotundus	96.6	50	0.00	0.00	146.6	100	100	20.0	0.0	220.0	100	100	26.6	0.0	226.6
K. pinnata	83.3	56.6	20	0.0	159.9	93.3	63.3	26.6	0.0	183.3	100	70	30	0.00	200.0
P. longum	53.3	30.0	20.0	3.3	106.6	93.3	43.3	26.6	13.3	176.6	100	53.3	36.6	16.6	206.6
E. hirta	86.6	63.3	16.6	0.0	166.6	93.3	70.0	20.0	0.0	183.3	96.6	76.6	23.3	0.0	196.6
I. tinctoria	20	13.3	0.0	0.0	33.3	33.3	20.0	0.0	0.0	53.3 .	50.0	0 33.3	3 0.0	0.0	33.3

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Toble. 12

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Plant				.	4 6	Perc	ent mo	rtalit	- ·	various	dilu	tions	for	72 ł	
				24	4 h 				48 h				• - • - - - •	74 1	
	1:5	1:10	1:20	1:40	Tox. inde		1:10	1:20	1:40	Tox. index	1:5	1:10	1:20	1:40	Tox. index
3.															
pentaphylla	100	86.6	96.6	23.3	286.4	100	96.6	83.3	30.0	309.9	100	100	90	36.6	326.7
1. azedarach	100	73.3	46.6	13.3	233.3	100	83.3	60	20	263.3	100	90	63.3	26.6	274.9
A. indica	100	73.3	43.3	13.3	229.9	100	86.6	66.6	73.3	276.6	100	100	73.3	23.3	296.6
C. rotundus	96.6	23.3	0.00	0.00	119.9	100	96.6	13.3	0.0	209.9	100	100	23.3	0.0	223.3
K. pinnata	70	46.6	10	0.0	126.6	80	56.6	16.6	6.6	159.9	90	66.6	30	6.66	193.3
P. longum	26.6	43.3	16.6	3.3	89.9	73.3	60	30	3.3	166.6	96.6	70	43.3	13.3	223.3
5. hirta	66.6	33.3	0.0	0.0	99.9	80	46.6	0.0	0.0	126.6	86.6	53.3	0.0	0.0	140.0
I. tinctoria	16.6	10	0.0	0.0	26.6	26.6	5 23.3	0.0	0.0	49.9.	43.3	3 .30	0.0	0.0	73.3

Table.13. In vitro confirmatory evaluation of plant extracts for the toxicity to R. similis- Adult(Nematicidal property)

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Table. 14

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In vitro co	onfirmat	-			- (for the) R. s	imili	s- Lai	rva		
Plant			24			Perc	ent in	nmobil: 48 h	ity at	varic	various dilutions for 72 h					
	1:5	1:10	1:20	1:40	Tox. inde	ex		1:20		Tox. index	1:5	1:10	1:20	1:40	Tox. index	
E. hirta	0.00	26.6	13.3	20.0	59,90	0.00	23.3	26.6	26.6	76.65	0.00	20.0	23.3	20.0	63.30	
P. betle	86.6	56.6	20.0	0.00	163.3	36.6	36.6	23.3	0.00	96.6	13.3	3 23.3	13.3	0.0	49.9	
S. indicum	50.0	40.0	16.6	0.00	106.6	66.6	50	16.6	0.0	133.3	53.3	56.6	23.3	0.0	133.3	
M. piperita	1 56.6	26.6	13.3	0.0	96.6	46.6	26.6	5 16.6	5 0.0	89.9	36.0	5 30	20	0.0	86.6	
M. oleifera	1 80	60	26.6	0.0	166.6	56.6	63.3	30.	0.0	149.9	53.3	53.3	30.0	0.0	136.6	
C. viscosa	53.3	30.	0.0	0.0	53.3	33.3	23.3	0.0	0.0	56.6	30.0	23.3	0.0	0.0	53.3	
I. tinctori	a 76.6	63.3	16.6	0.0	156.6	66.6	63.3	16.6	0.0	146.6	50.0	53.3	23.3	3 0.0	126.6	
A. calamus	63.3	63.3	0.0	0.0	106.0	5 50	36.6	0.0	0.0	86.6	40.0	20.	0 0.0	0.0	60.0	
G. pentaphylla	a 0.00	0.00	10.0	30.0	40.0	0.00	0.0	10.0	26.6	36.60	0.00	0.0	13.3	26.6	39.9	
C. rotundus	s 0.00	50	73.3	0.00	123.3	0.0	0.0	60.0	26.6	86.6	0.00	0.0	0 63.8	3 16.0	5 79.9	
K. pinnata	0.0	0.0	50.0	20.0	70.0	0.00	0.00	46.6	20	66.6	0.0	0.0	50	10	60.0	

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Plant		P 24 h						mobili 48 h	ty at	vario	us di:	lution	s for 72 h		
	1:5	1:10			inde	x				Tox. index	1:5	1:10	1:20	1:40	Tox inde
E. hirta 3	3.3	 36.6	 33.3	0.00				23.3		73.3	13.3	26.6	23.3	0.0	63.2
•	36.6									96.6		13.3			43.3
S. indicum	43.3	13.3	0.00	0.00	56.6	53.3	16.6	0.00	0,0	69.9	40.0	30.0	0.0	0.0	70.
M. piperita :		13.3	0.00	0.00	66.6	46.6	16.6	0.00	0.0	63.3	40.0	13.3	0.0	0.0	53
M. oleifera		30	0.00	0.00	93.3	60.0	33.3	0.00	0.0	93.3	53.3	30.0	0.0	0.0	83
C. viscosa		13.3	0.00	0.00	49.9	43.3	20.0	0.00	0.00	63.3	30.0	23.3	0.0	0.0	53
I. tinctoria		60.0	20.0							136.6					136
A. calamus										0 73.3					
G. pentaphylla	0.00	0.00	16.6	43.3	59.9	0.00	0.0	13.3	36.6	49.90	0.00	0.00	10.0	40.0	50
C. rotundus		73.3	20.0	0.Ò0	93.3	0.0	0.0	13.3	0.00	13.3	0.00	0.00	16.ę́,	0.00	16
K. pinnata		0.0	70.0	0.00	70.0	0.00	0.00	63.3	0.00	63.3	0.0	<u>0</u> .0	53.3	0.00	53

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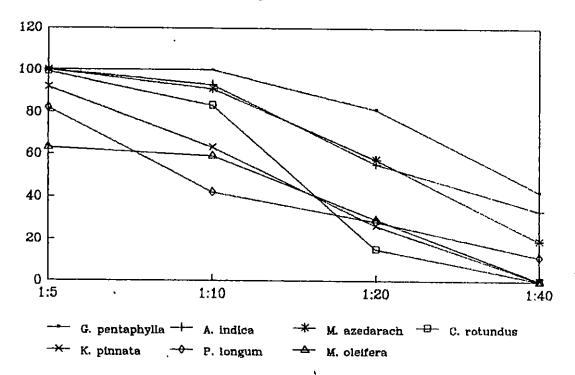
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Table 16. Mortality % of *R*. *similis* (Larva and Adult) with various plant extracts.

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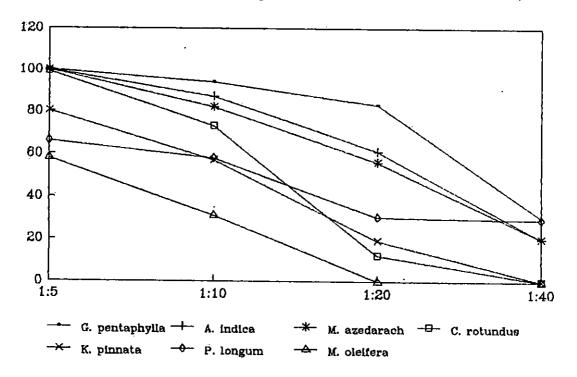
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Plant species	Conc	Larva	Adult
Glycosmis pentaphylla	1:5	100	100
	1:10	100	94
	1:20	81	83
	1:40	42	30
Azadirachta indica	1:5	100	100
	1:10	93	87
	1:20	55	61
	1:40	33	20
Melia azadeach	1:5 1:10 1:20 1:40	100 91 57 19	100 82 56 20
Cyperus rotundus	1:5	99	99
	1:10	83	73
	1:20	15	12
Kalanchoe pinnata	1:5	92	80
	1:10	63	57
	1:20	26	19
Piper longum	1:5	82	66
	1:10	42	58
	1:20	28	30
	1:40	11	29
Moringa delifera	1:5	63	58
	1:10	59	31
	1:20	29	-



Effect of medicinal plant extracts on the mortality % of *R. similis* larva





Effect of medicinal plant extracts on the mortality % of *R. similis* adult



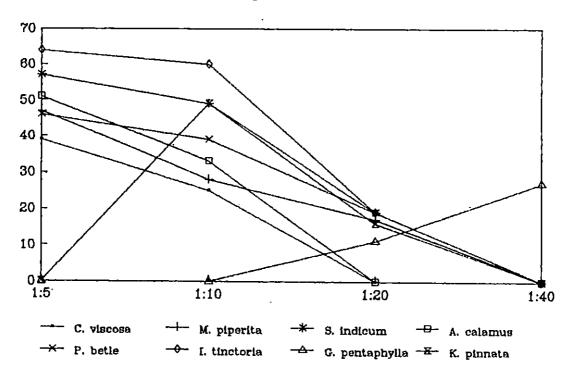
Table 17. Mobility % of *R*. *similis* (Larva and Adult) with various plant extracts.

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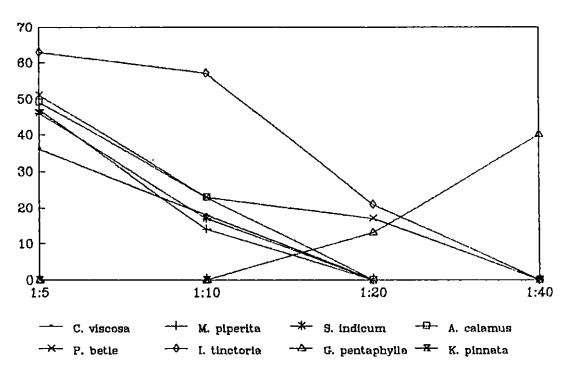
Plant species	Conc	Larva	Adult
Cleome viscosa	1:5	39	36
	1:10	25	18
Mentha piperita	1:5	47	47
	1:10	28	14
	1:20	17	-
Solanum indicum	1:5	57	46
	1:10	49	17
	1:20	19	-
Acorus calamus	1:5	51	49
	1:10	33	23
Piper betle	1:5	46	51
	1:10	39	23
	1:20	19	17
Indigofera tinctoria	1:5 1:10 1:20	64 60 19	63 57 21
Glycosmis pentaphylla	1:20	11	13
	1:40	27	40
Kalanchoe pinnata	1:10 1:20	49 16	-
Cyperus rotundus	1:20	16	24
	1:40	65	17



Effect of medicinal plant extracts on the mobility % of R. *similis* larva

Fig. 13

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Effect of medicinal plant extracts on the mobility % of R. similis adult

Fig. 14

DISCUSSION -

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DISCUSSION

Banana is an important fruit crop cultivated in India and its production ranks high compared with other fruits. This crop is threatened with the attack of the burrowing nematode which is known to be present in all banana growing tracts of the country. Management of this nematode is mainly achieved by the use of chemicals which results in internal residue problems and pollution hazards. If the control of the nematode can be achieved by botanicals having antihelminthic properties, it will be a welcome practice readily accepted by banana farmers. With this objective the present study was initiated. Twenty plant species which are commonly grown in homesteads and having some kind of aromatic and medicinal properties were selected for studying the antihelminthic action against R. similis

The infective stages of the nematodes were used as test organisms for screening the antihelminthic properties of the selected plants. The results obtained are discussed in this chapter.

5.1. Effect of A. indica, M. azedarach and G. pentaphylla

5.1.1. Larva.

The result of the study clearly indicated that *G. pentaphylla* leaf extracts possesS both nematicidal and nematostatic properties. All the tested dilutions exhibited significant mortality ranging from 100 - 42 %. But the time of exposure had no significant effect on the mortality. It is the concentration that is more lethal than the time of exposure. This could be the fact that no interactive effect was noticed between time of exposure

and strength of dilution. Leaf extract of A. *indica* and fruit extract of M. *azedarach* was also effective at all the dilutions ranging from 1:5 to 1:40. In both the cases the time of exposure was significant but the interaction was insignificant.

5.1.2. Adult

Extracts of all the three plant species could bring out significant mortality. Here also *G. pentaphylla* exhibited higher toxicity index. Neither the time interval nor the interaction had significant effect on the mortality. Besides nematicidal property, it exhibited nematostatic property at the higher dilutions of 1:20 and 1:40. *A. indica* left 20% of adult dead even at the highest dilution of 1:40. An interactive effect of dilution over time was noticed. The dilutions 1:10 and 1:20 showed that the duration of their action extended upto 72 h as could be noticed from the significantly higher mortality at 72 h. But in the case of 1:40 dilution longevity of action remained only upto 48 h. Fruit extract of *M. azedarach* was significantly effective at all the dilutions, the percentage mortality being 100 at 1:5 and 20% at 1:40 dilutions. Further it was observed that increase in dilutuion resulted in reduced nematicidal toxicity in all the plant leaf extracts. Also nematodes did not revive when transferred to distilled water indicating their toxic effect as irreversible.

The usefulness of the plant extracts of A. indica and M. azedarach on other nematodes are already reported by Siddiqui and Alam, 1987 and 1989. Grewal (1989) reported that leaf and seed extracts of M. azedarach were highly toxic to A. composticola. Khanna et al. (1988) reported 90% mortality of A. composticola with the plant extract of A. indica. Bare root dip treatment with leaf extract of *M. azedarach* significantly reduced root knot development due to *M. incognita* on tomato and *Capsicum annum* (Akhtar *et al.*, 1992). Wani (1992) reported efficacy of *A. indica* in controlling root knot nematode on okra with seed soaking in neem leaf extract.

Rajvanshi *et al.* (1986) suggested that leaf extract of *Tagetes patula* contains an alkaline water soluble nematotoxic chemical constituent as it kills *Xiphinema basiri*. In the present study a similar action was observed with the extract of *G. pentaphylla*, *A. indica* and *M.azedarach*.

5.2 Effect of K. pinnata and E. hirta

5.2.1 Larva.

The nematicidal effect of both the extracts decreased with an increase in the dilution. The lowest dilution of 1:5 was more effective in causing a mortality of 92% for both the plant extracts. No mortality was observed at the highest dilution of 1:40 for both the extracts. The time factor showed significant effect on the mortality for *K. pinnata* leaf extract only. At the highest dilutions of 1:20 and 1:40 the extract exhibited nematostatic property.

5.2.2. Adult

The toxic effect of K. pinnata was observed at 3 dilutions, where as for E. hirta the effect was seen only at the 2 lower dilutions. K. pinnata left 80% adults as dead at the lowest dilution whereas the same was around 77% for E. hirta. In both cases there was significant difference in the time of exposure but there was no significant difference in their interaction. There were no reports of studies with the extract of K. pinnata. However the results obtained with the plant extract of E. hirta agree with the results obtained by some workers on other nematode pests. (Nandal and Bhatti, 1983; Siddiqui *et al.*, 1984). Zureen and Khan (1984) reported that a related species of *Euphorbia* namely *E. caducifolia* was highly toxic to *Meloidogyne javanica*. Root extract of *E. hirta* inhibited egg hatching of *M. incognita* (Sharma and Trivedi, 1992). Similarly plant extracts of *E. tinctoria* were effective against *M. javanica* on tomato at three dilutions *ie*, 100, 500 and 1000 mg/litre (Al Obaedi *et al.*, 1987).

Here also the treated larvae could not revive when transferred to distilled water. The nematicidal activity of the extract was there for irreversible. As the activity of the extract reduced considerably with dilution, it can be concluded that the effect was dose dependent.

5.3. Effect of P. longum

5.3.1 Larva

Leaf extract of *P. longum* exhibited significant nematotoxic effect at all the four dilutions. A decreasing trend in the mortality was observed from 82% to 11% with an increase in the dilution. Significant effect of the three exposure time was observed on mortality. At all the dilutions, duration of their action extended upto 72 h as could be noticed from the significantly higher mortality at 72 h.

5.3.2 Adult

When *R. similis* adults were treated with the leaf extract, the nematicidal effect was seen only at three dilutions 1:5, 1:10 and 1:20 and the percentage mortality dropped from 65.5 to 30%. The three exposure timings had significant effect on the mortality. But no significant interactive effect could be noticed.

There were no reports on the nematicidal action of *P. longum* on nematodes.

5.4 Effect of C. rotundus

5.4.1 Larva

The result of the study indicated that rhizome extract of C. rotundus possess both nematicidal and nematostatic properties. The lowest dilution caused maximum mortality of 100%, while there was no mortality at the highest dilution of 1:40. Here both extracts and exposure timings had significant lethal effect on the mortality. The dilutions 1:10 and 1:20showed that the duration of their action prolonged upto 72 h as could be noticed from the significant higher mortality at 72 h. The extract also exhibited nematostatic property at 1:10, 1:20 and 1:40 dilutions. The maximum of 65.5% at 1:20 dilution.

5.4.2 Adult

The treatments with the three lower dilutions of extract also resulted in significant mortality. The time interval had significant effect on the mortality. The interaction of time and strength of dilution was also significant. Besides nematicidal effect, the extract showed significant nematostatic effect at 1:10 and 1:20 dilutions.

A comparable study was undertaken by Hussain and Masood (1975) with the extract of C. rotundus on plant parasitic nematodes. C. rotundus and C. esculentus were evaluated for the control of T. penetrans and M. incognita. C. rotundus was a poor host, while C. esculentus was a nonhost. Water extract from corm, rhizome and root of C. esculentus inhibited egg hatching and reduced survival of hatched larvae of M. incognita (Haroon, 1989).

Nandal and Bhatti (1983) reported that leaf extract of C. rotundus caused 100 and 89% mortality of M. javanica at 1:5 and 1:10 dilutions respectively.

5.5 Effect of P. betle and I. tinctoria

5.5.1 Larva

Present findings revealed that the root extract of *I. tinctoria* was ineffective in causing significant mortality. But significant mortality was observed with the leaf extract of *P. betle* at 1:5 and 1:10 dilutions. An insignificant interactive effect of dilution over time was also noticed. In addition to nematicidal effect, both the plants exhibited significant nematostatic effect. A higher percentage of toxic effect was expressed by *I. tinctoria*. But an interactive effect was observed in case of *P. betle* only.

5.5.2 Adult

Significant lethal effect was shown by *I. tinctoria* extract only. On the other hand both the plants exhibited significant nematostatic effect. The toxic effect of the extract was inversly proportional to the strength of the dilution. No literature is avilable on the nematotoxic effect of both the plants.

5.6. Effect of *M. olefera*, *S. indicum* and *M. piper* .

5.6.1 Larva

The study revealed that all these extracts were nematostatic at the three dilutions of 1:5, 1:10 and 1:20. Among these three, M. oleifera showed the highest nematostatic effect of 63% at 1:5 dilution. From the results it can be concluded that the effect was dependent on the strength of dilution and not on the time of exposure.

5.6.2. Adult

The toxic effects of the extracts were seen only at the two lower dilutions of 1:5 and 1:10. Neither the timings nor the interactions were significant.

There were no reports explaining the nematicidal action of M. *oleifera* extract.

But reports on the nematicidal properties of *M. piperita* and related species of *Solanum* agrees with the findings of this study. A related species of *Mentha*, *M. viridis*, is reported to be significanatly effective against *M. incongnita* larva and R reniformis (Haseeb et al., 1978). Sangwan et al. (1990) reported the nematicidal activity of essential oils of M. piperita and their major monoterpenoidal constituents menthol against juveniles of A. tritici, T. semipenetrans, M. javanica and Heterodera. There were several studies with related species of Solanum like S. tuburosum (Allen and Feildmesser, 1970), S. esculuntum (Onda et al., 1972) and S. bispidium (Haseeb et al., 1978)

Nandal and Bhatti (1983) screened leaf and fruit extracts of S. xanthocarpum against M. javanica and reported that the mortality percentage ranged from 93% to 3% with leaf extract (1:5 to 1:80) and 91% to 76% (1:5 to 1:10) with fruit extract.

An alkaline water soluble nematostatic chemical constituent was detected in the leaf extract of T. patula (Rajvanshi et al., 1986). Presence of similar components in the tested extracts can be attributed to their nematostatic property.

5.7. Effect of A. calamus and C. viscosa

5.7.1 Larva

In this experiment, only rhizome extract of A. calamus expressed significant nematostatic effect. The effect was seen only at the two dilutions of 1:5 and 1:10.

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5.7.2. Adult

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The same result was obtained with adult nematode also. Seed extract of C. viscosa also produced a significant nematostatic effect at 1:5 and 1:10 dilutions. But here, only the strength of dilution was lethal to nematodes.

Comparable results are available with the extract of *A. calamus*. Saxena *et al.* (1990) tested petroleum ether extract of *A. calamus* against *M. incongnita*. Their study revealed 100% mortality of the nematode with the higher concentrations. Similarly for *C. viscosa* 100% mortality was observed with the leaf extract against *M. javanica* (Krishnamurthy *et al.*, 1989).

5.8. Effect of C. angustifolia, A. squamosa, C. papaya, P. guajava, E. scandens and L. inermis.

Present findings revealed that all these plant extracts were ineffective to produce lethal effects on the nematode. It can be seen that neither the plant extracts nor its dilutions have a marked effect on the mortality. A scan through the literature indicate some interesting results contradictory to the findings of this study.

Maqbool et al. (1987) found that latex extract from C. papaya were not toxic against M. incongnita. But incorporation of 100 g chopped shoots of latex bearing C. papaya significantly supressed the population build up of R. reniformis and T. brassicae and reduced root knot development by M. incognita (Siddiqui et al., 1987).

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Several reports are available with the nematicidal action of *L. inermis.* Nandal and Bhatti (1983) reported that leaf extract of *L. inermis* was effective against *M. javanica.* Methanolic extract of *Lawsonia* resulted in 50-100% mortality of *M. javanica, T. semipenetrans* and *A. tritici.* Oil extracted from seeds also exhibited nematicidal property (Kumari *et al.*, 1986, 1987)

No reports are available on the toxic effect of C. angustifolia, E. scandens, A. squamosa and P. guajava.

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- SUMMARY -

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SUMMARY

Laboratory experiments were conducted at the College of Horticulture, Vellanikkara to screen medicinal plants for antihelminthic properties against adult and larval stages of banana burrowing nematode. With this objective, experiments were carried out to test aqueous extract of different parts of 20 medicinal plants at different dilutions to find out its effects on the mortality and mobility of the nematode.

The study resulted in the following findings

Leaf extract of A. indica and G. pentaphylla were found to be highly nematicidal to both larvae and adult at all the tested doses. K. pinnata showed significant mortality at all the tested doses except 1:40. P. longum was effective in causing a significant mortality at all tested dilutions for larval stage, whereas against adult stage. Significant mortality was observed only at the three dilutions of 1:5, 1:10 and 1:20.

Besides nematicidal effects, higher dilutions of 1:20 and 1:40 of G. pentaphylla and K. pinnata expressed nematostatic effect.

A significant nematostatic effect was observed when larval stages were subjected to treatment with the leaf extracts of

M. oleifera, M. piperita and *P. betle* at 1:5, 1:10 and 1:20 dilutions. But these extracts were significantly effective only at the two lower dilutions of 1:5 and 1:10 against the adult stages except in the case of *P. betle*. Leaf extracts of *C. angustifolia*, and *A. squamosa* were not effective at all the tested dilutions. Neither mortality nor mobility was observed with all the plant extracts.

Fruit extract of *M. azedarach* showed significant mortality at all the tested dilutions, but treatments with fruit extract of *C. papaya* and *P. gujuava* were ineffective in expressing lethal effect on adults and larval stages of the nematode.

Significant effect on mobility was observed at 1:5 and 1:10 dilutions when adult stages of the nematode were treated with the seed extract of E. *viscosa*. On the contrary, no significant effect was observed at the same concentration against larval stages. Treatment with seed extract of E. *scandens* was ineffective in causing the mortality of the nematode.

The treatment effects were significant in causing the mortality with the rhizome extract of C. rotundus at the three dilutions ,ie, 1:5, 1:10 and 1:20 against both the larvae and adults. In addition to nematicidal effect, the extract exhibited significant nematostatic effect against both the larvae and adults. Rhizome extracts of A. calamus also exhibited significant nematostatic effect at 1:5 and 1:10 dilutions.

Root extracts of *S. indicum* and *I. tinctoria* were effective in causing significant nematostatic effect. *I. tinctoria* caused immobility at 1:5, 1:10 and 1:20 dilutions against both the adults and larval stages. Beside nematostatic effect it exhibited significant nematicidal effect against adult stages at 1:5 and 1:10 dilutions, whereas the same was insignificant with the larval stages.

Significant nematostatic effect was seen with root extract of S. *indicum* at 1:5, 1:10 and 1:20 dilutions against larval stages, but the effect was seen only at the two dilutions, ie, 1:5 and 1:10 against adult stage.

The plant extract of E. *hirta* was effective in resulting significant mortality at the three tested doses against larval stages. But the effect was seen only at the two dilutions of 1:5 and 1:10 against the adult stages. In addition to nematicidal effect, it also exhibited nematostatic effect but it was not statistically significant.

CONCLUSION

Out of the 20 species of plants tested for anti helminthic properties it is reported that the leaf extract of *Azadirachta indica* (Neem), *Glycosmis pentaphylla* (Panal) and *Kalanchoe pinnata* (Murikootti) have got a high degree of nematicidal effect on the larvae and adult stage of the nematode. The leaf extract of *Piper longum* (Thippali) has the same effect on the larval stages. These informations are new and useful in undertaking further detailed lab tests and field oriented trials on the management of the nematode pest.

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* Orginals not seen

SCREENING MEDICINAL PLANTS FOR ANTIHELMINTHIC PROPERTIES AGAINST DIFFERENT LIFE STAGES OF BANANA BURROWING NEMATODE, Radopholus similis [Cobb, 1893] Thorne 1949

BY

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ABSTRACT OF A THESIS submitted in partial fulfilment of the requirement for the degree MASTER OF SCIENCE IN AGRICULTURE Faculty of Agriculture Kerala Agricultural University

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ABSTRACT

Experiments were conducted at the department of Entomology, College of Horticulture, Vellanikkara to screen medicinal plants for antihelminthic properties against the infective stages of banana burrowing nematode *R. similis*.

The effect of aqueous extract of different parts of 20 medicinal plants were treated at four dilutions and three exposure times. Nematicidal and nematostatic properties of these extracts were studied using the nematode culture developed on carrot callus.

The study resulted in the following findings.

1. Extracts of *A. indica* and *G. pentaphylla* were highly nematicidal to infective stages of the nematode at all the tested doses. *K. pinnata* was significantly effective at all the tested doses except 1:40. *P. longum* resulted in significant mortality at all the tested doses against larval stages whereas only 1:5, 1:10 and 1:20 dilutions were effective against adult stages.

Besides nematicidal effect 1:20 and 1:40 dilutions of G. pentaphylla and K. pinnata exhibited nematostatic effects.

Leaf extract of *M. oleifera* and *M. piperita* at 1:5 and 1:10 dilutions
showed significant nematostatic effects against both larvae and adults. *P. betle* extract was equally effective at the three dilutions.

3. Leaf extracts of C. angustifolia, A. squamosa and L. inermis were not effective at all tested doses.

4. Fruit extract of *M. azedarach* was nematicidal at all the tested dilutions of 1:5, 1:10, 1:20 and 1:40. But extract of *C. papaya* was ineffective.

5. Seed extract of *C. viscosa* expressed nematostatic property at 1:5 and 1:10 dilutions against adult stages, but it was ineffective against larval stages. Treatment with seed extract of *E. scandens* was ineffective.

6. Rhizome extract of C. rotundus was equally effective against infective stages in causing mortality at 1:5, 1:10 and 1:20 dilutions. Besides nematicidal effects, the extract exhibited significant nematostatic effect. A. calamus extract was nematostatic at 1:5 and 1:10 dilutions.

7. Root extract of *I. tinctoria* was nematostatic at 1:5, 1:10 and 1:20 dilutions against both larvae and adults. In addition to immobility, it resulted in the death of adult nematodes at 1:5 and 1:10 dilutions. Extract of *S. indicum* showed significant nematostatic effect at 1:5, 1:10 and 1:20 dilutions against larval stages. But it was effective only at 1:5 and 1:10 dilutions against adult stages.

8. Plant extract of E. *hirta* expressed nematicidal property at 1:5, 1:10 and 1:20 dilutions against larval stages, but the same was effective only at the two lower dilutions against adult stages.