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Nematode Infestations

Part II: Industrial Crops

Editors

Mujeebur Rahman Khan

Ph. D, Formerly Chairman Department of Plant Protection Aligarh Muslim University Aligarh 202 002, India

M Shamim Jairajpuri

Ph. D, D. Sc, FNASc, FTWAS, FNA, FASc, FNAAS, FISP, FZS, FLS, FIBiol
Formerly Director, Institute of Agriculture, AMU & Vice Chancellor,

MANUU, Hyderabad Honorary Scientist, Department of Zoology Aligarh Muslim University Aligarh 202 002, India

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Nematode Infestation in Spice Cultivations

SANTHOSH J. EAPEN

Indian Institute of Spices Research, Calicut - 673 012, Kerala, India email: sjeapen@spices.res.in

Abstract

About 66 species of plant parasitic nematodes belonging to 27 genera are reported on ten major spices. The most predominant and widely distributed among these are root knot nematodes (Meloidogyne spp.), burrowing nematodes (Radopholus similis) and root lesion nematodes (Pratylenchus spp.). Nematode infestations cause 'slow decline' disease in black pepper plantations. Gradual decline and foliar yellowing coupled with severe root damage on account of galls, lesions and rotting are the predominant symptoms of nematode infestation in black pepper. Widespread occurrence of root-knot nematodes, M. incognita and M. javanica has been reported in cardamom nurseries and plantations in India. Root system of infested plants shows varying degrees of root galling. In ginger and turmeric too, nematodes such as root knot nematodes are widely distributed in all states where these crops are cultivated. Use of planting materials without symptoms of nematode infestation from known and nematode-free sources is very important aspect of successful management of the nematodes. Exclusion of nematode susceptible intercrops, shade trees, supports or standards can also minimise the nematode infestation. Soil amendments like application of organic cakes, mulching etc. are ideal ecofriendly measures to manage these pests. Efforts to identify nematode resistance have helped to recognise a few resistant lines in some spices. Chemical control of nematodes is a less preferred option now a days and as a result intensive research is on at several laboratories to isolate efficient biocontrol agents against these nematodes. Tropical agro-ecosystems are complex, more resilient and therefore, sustainable ways of nematode management have to be evolved by blending different management options.

Introduction

Spices are aromatic substances of plant origin which are commonly used for flavouring, seasoning and imparting aroma in foodstuffs. International spice organisation (ISO) has recognized 109 plants

belonging to 31 families as spices useful as ingredients in food. They are distributed in different countries of the world. During 2006, spices were cultivated in an area of 5.98 million ha globally with a total production of 7.31 million tons (FAO, 2008). India is considered as the home of majority of these spices from ancient times and sizable area of the country is under spice cultivation. The latest statistics shows that the total cultivated area in India under spices is 2.89 million ha with a total production of 3.33 million tonnes. The perennial spice crops like black pepper and cardamom are being cultivated in mixed cropping systems involving an array of crops, shade trees and live standards.

Among the spice crops, black pepper occupies first position in foreign exchange earnings of India. Black pepper (Piper nigrum L.), a perennial climber belonging to the family Piperaceae, has its origin in the Western Ghats of India. It is a major spice crop of India and is mostly grown in the states of Kerala, Karnataka, Tamil Nadu and Andhra Pradesh. Cultivation of cardamom (Elettaria cardamomum Maton), the 'Queen of spices', is confined to the southern states of India. The other two major Zingiberaceae spices are ginger (Zingiber officinale Rosc.) and turmeric (Curcuma longa L.) which are cultivated extensively in India. Coriander (Coriandrum sativum L.), cumin (Cuminum cyminum L.), fennel (Foeniculum vulgare Mill), fenugreek (Trigonella foenum graecum L.), celery (Apium graveolens L.), dill (Anethum graveolens L.), caraway (Carum carvi L.) and aniseed (Pimpinella anisum L.) are some important seed spices (grain spices). Apart from these, there are several tree spices being cultivated in India and some of the prominent ones are clove (Syzygium aromaticum Merril and Perry), nutmeg (Myristica fragrans Hout.), cinnamon (Cinnamomum zeylanicum Blume) and allspice (Pimenta dioica Merr.). Most of these spices are extensively used for culinary or flavouring purposes. They are also being used for preserving processed foods. Several of them are known traditionally for their medicinal properties.

Plant parasitic nematodes infesting spices

Among the biotic factors, problems caused by nematodes singly or with fungi (disease complexes) are major production constraints of spices. Nematodes are also known to break the resistance to fungal/bacterial pathogens in many spice crops. About 66 species of plant parasitic nematodes belonging to 27 genera are reported on ten major spices (Koshy et al., 2005). Nematological problems in spices and condiments have been reviewed recently (Koshy et al., 2005; Eapen,

2005). In this review an attempt is made to update the information already available on all the important spice crops.

The most predominant and widely distributed among plant nematodes are root knot nematodes (Meloidogyne spp.). The burrowing nematode (Radopholus similis) is another important nematode pathogen, a severe constraint in the production of black pepper. Tremendous diversity is observed in the nematode fauna present in the rhizosphere of tropical spices like black pepper, ginger and turmeric.

Black pepper (Piper nigrum L.)

Production and productivity of black pepper are seriously affected by two major diseases in all pepper growing countries. These are Phytophthora foot rot caused by the fungus Phytophthora capsici and slow decline (slow wilt/pepper yellows), mainly due to plant parasitic nematodes, M. incognita and R. similis. Several plant parasitic nematodes belonging to different groups are reported in association with black pepper (Sundararaju et al., 1979a, Ramana and Eapen, 1998). In India, 17 genera of plant parasitic nematodes were recorded on black pepper in Kerala and Karnataka alone (Sundararaju et al., 1979b).

History

The slow-wilt disease was first reported from Wayanad area in Kerala as early as 1902 and Krishna Menon (1949) reported mortality up to 10% of the vines due to the disease. The first record of root knot nematode infestation on black pepper was by Barber (cited by Ridley) who observed root-knot nematode infestation on black pepper in Wayanad, Kerala, India. Butler (1906) in his further investigations on the disease in Wayanad, also reported the association of root-knot nematodes with the diseased vines. Slow decline disease of black pepper was first reported by Van der Vecht (1950) as 'pepper yellows' on the islands of Bangka, Indonesia and R. similis was reported to be responsible for the disease. Later Christie (1957) held this nematode to be for responsible the death of 20 million pepper vines. Association of R. similis with black pepper in India was first reported by D'Souza et al. (1970) and subsequently its wide spread occurrence in black pepper plantation in South India was reported by several workers. A new species of a semi-endoparasitic nematode, Trophotylenchulus piperis was reported on black pepper exclusively from India. It was initially identified as Trophotylenchulus floridensis (Mohandas and Ramana, 1982) and was subsequently raised to a new species, T. piperis (Mohandas et al., 1985).

Distribution

Plant parasitic nematodes belonging to 14 genera were reported infesting black pepper in the detailed surveys conducted during 1980s in two districts of Kerala viz., Calicut and Kannur (Table 1) (Ramana and Mohandas, 1987). In Karnataka, maximum incidences of M. incognita and R. similis were seen in Dakshina Kannada District (Ramana and Mohandas, 1989). Among the nematodes reported in India, the predominant species are Meloidogyne spp., R. similis, Trophotylenchulus piperis, Helicotylenchulus sp. and Rotylenchulus reniformis (Jacob and Kuriyan, 1980; Ramana and Mohandas, 1987; 1989). Among the four major species of Meloidogyne, M. incognita is of major economic importance in black pepper. A new species of root knot nematode, M. piperis, is also reported on black pepper (Sahoo et al., 2000). In Kerala and Karnataka, about 69.8% and 53.8% of the vines respectively were found infested with the M. incognita (Ramana and Mohandas 1987, 1989). Interestingly both apparently healthy and slow decline affected vines harboured high populations of the nematode (Ramana et al., 1987). T. piperis, a semiendoparasite is wide spread in all major black pepper growing areas in Kerala (Ramana and Mohandas, 1987) and Karnataka (Ramana and Mohandas, 1989).

Table 1- Plant parasitic nematodes associated with black pepper in Kerala.

^{*} Incidence represented as absolute frequencies Source: Ramana et al., 1994.

Symptoms

Nematode infestations often assume alarming proportion leading to 'slow decline', a debilitating disease in black pepper plantations primarily occurs due to burrowing nematode, *Radopholus similis*. Mild to moderate foliar yellowing at different regions of the affected vine is the initial aerial symptom of slow decline disease (Fig. 1). With the advancement of the disease, the infected vines show defoliation and die back leading to loss of vigour, yield and finally death of the vine. Vines in the early stages of the disease look apparently normal with the onset of monsoon, but foliar yellowing becomes more pronounced with the depletion of soil moisture in summer season (Mohandas and Ramana, 1987; 1991; Ramana *et al.*, 1994). Root penetration by *R. similis* causes necrotic lesions on white feeder roots of black pepper. Subsequently the lesions merge and encircle the root cortex leading to disintegration of distal portion of the roots (Fig. 1).



Fig. 1 – A black pepper vine showing symptoms of 'slow decline' disease, and root rot caused by *Radopholus similis*.

Gradual decline and foliar yellowing are also the predominant symptoms of root knot nematode infestation in black pepper. Plants exhibit dense, yellowish discolouration in inter veinal areas of leaves (interveinal chlorosis) (Ramana, 1992; Ramana et al., 1994). Roots of infested plants show varying degrees of galling. Typical galls or knots are seen on secondary and fibrous roots and as elongated swelling on thick primary roots (Fig. 2). Adult females with egg masses are generally embedded within the root tissue.

Occasionally the nematode infects the underground portion of the stem, particularly the rooted cuttings in the nursery (Ramana et al.,

1994). Damage caused to the roots directly affects vigour and productivity of the vines. Foliar yellowing and defoliation is the main above ground symptom expressed by the vines infected with *R. similis*. Venkitesan and Setty (1977) reported that black pepper plants inoculated with *R. similis* @ 1000 and 10,000 showed wilt symptoms after 90 days and all succumbed within 118 days of inoculation. The foliar yellowing and defoliation became severe in the vines inoculated with *R. similis* alone or in combination with *M. incognita* and these symptoms were more pronounced during summer months (Mohandas and Ramana 1991). Necrosis and general drying of the root tissues are observed at regions where *T. piperis* females are found attached to pepper roots.

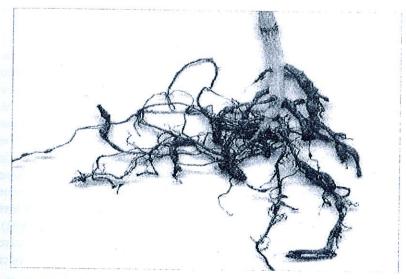


Fig. 2 - A black pepper rooted cutting with root-knot nematode induced galls.

Yield loss

Experiments conducted under artificial inoculation with *M. incognita* showed that there was a reduction of 16% and 50% in the growth of black pepper rooted cuttings at an initial inoculum of 10 and 100,000 second stage juveniles of *M. incognita*, respectively (Koshy *et al.*, 1979). In Indonesia, 'yellows' symptoms appeared in plants having a population level 47J₂/100 g soil and 350 individuals of *M. incognita* /10 g roots (Mustika, 1978). Apart from reducing various growth parameters in adult vines grown under simulated field condition, the yield was reduced from 13 to 47% with the initial inoculum levels ranging from 100 to 100,000 nematodes/plant (Mohandas and Ramana, 1991). Further, foliage yellowing and defoliation were maximum in vines inoculated

with higher inoculum levels (>10,000 nematodes/plant). Similar reduction in the growth of pepper vines due to root knot nematodes was reported by others also (Winoto, 1972; Jacob and Kuriyan, 1980).

Pathogenicity tests with *R. similis* on black pepper vines grown in microplots showed that the nematode caused significant reduction in the growth and yield of the vines at the initial population of 100 to 10,000 nematodes/plant (Mohandas and Ramana, 1991). The onset of yellows diseases in Sumatra (Indonesia) is correlated with *R. similis* population of 2 nematodes/100 g soil and 25 nematodes/10 g root (Mustika, 1978). In India a minimum population of 250 nematodes/g roots was consistently recorded with slow decline affected pepper vines (Ramana *et al.*, 1987a). Black pepper vines at all stages of growth are susceptible to the nematode infestation (Ramana, 1992).

Economic importance

Sitepu and Kasim (1991) reported that this disease may cause up to 32% loss in Indonesia. In Para (Brazil), about 90% of pepper vines were infected with root knot nematodes, mostly *M. incognita* (Ichinohe, 1975). In Guyana, about 30% vines were destroyed by the similar disease (Biessar, 1969). Experiments have shown that black pepper yields could be increased by 50% by controlling nematode infestation through nematicide application (Kueh and Teo, 1978; Davide, 1985). Though, the disease involving *M. incognita* and *R. similis* infestations is prevalent in all major black pepper growing areas in India and the nematodes exact crop loss estimates are not available (Ramana *et al.*, 1987a, 1994; Ramana, 1992). Yield losses may rang from 39 to 65 % when pepper vines were inoculated with *R. similis*, *M. incognita* alone or in combinations under simulated field conditions (Mohandas and Ramana, 1991).

Interaction with other organisms

Several other microorganisms particularly fungi have been reported to be associated with the slow decline disease. Hubert (1957) and Bridge (1978) opined that though *R. similis* is primarily responsible for the disease, an association with fungus like *Fusarium* sp. is, however, necessary to cause 'yellows' disease. An Indonesian isolate of *R. similis* predisposed black pepper seedlings to attack by a weak pathogenic isolate of *Fusarium solani* resulting in severe root damage (Freire, 1982). A synergistic effect of *Meloidogyne* sp., *R. similis* and *Fusarium* sp. on reduction in growth of pepper vines and increase and severity of foliar yellowing was observed (Lopes and Lordello, 1979; Hamada *et al.*, 1985; Sheela and Venkitesan, 1990; Mustika, 1990; 1992). The fungus

also showed antagonistic effect on the multiplication and production of root galls by *M. incognita* (Sheela and Venkitesan, 1990; Mustika, 1992). Varughese and Anuar (1992) reported the association of *Pythium* sp. and *Fusarium* sp. with the roots of slow decline affected vines in Malaysia. In India, presence of *Fusarium* sp., *Rhizoctonia bataticola* and *Pythium* sp. in the roots of slow decline affected vines was reported by Nambiar and Sarma (1977). However, the possible role of *Fusarium* sp. in the slow decline disease complex could not be established in India (Ramana *et al.*, 1992).

Winoto (1972) observed that plants infested with root knot nematodes were more susceptible to *Phytophthora* infections. However, Holliday and Mowat (1963) could not find any relation between *M. javanica* infestation and foot rot caused by *P. palmivora* in pepper plantations in Sarawak, Malaysia. Experiments conducted in microplots under simulated field conditions to assess the role of *M. incognita*, *R. similis* and *P. capsici* in slow decline disease showed that *R. similis* and *P. capsici* alone or in association and *M. incognita* in association with either *R. similis* or *P. capsici* or both resulted in root rotting leading to typical slow decline disease (Anandaraj *et al.*, 1996 a, b).

Ferraz and Sharma (1979) found *M. incognita* alone and in combination with *Rotylenchulus reniformis* had a highly pathogenic effect on the growth of rooted cuttings of black pepper, and *R. reniformis* had an inhibitory effect on the multiplication of *M. incognita*. Sheela and Venkitesan (1981) reported that simultaneous inoculation of *R. similis* and *M. incognita* suppressed the plant growth to a great extent. Root knot nematode population buildup was adversely affected under combined inoculation.

Cardamom (Elettaria cardamomum Maton)

Plant parasitic nematodes belonging to 19 genera have been found being associated with cardamom (Table 2). The most important nematode is root-knot nematodes, *Meloidogyne* spp., although lesion nematodes *Pratylenchus* spp. and burrowing nematodes, *R. similis* are known to cause root rotting to this spice (D'Souza *et al.*, 1970; Sundararaju *et al.*, 1979b). Reniform nematode, *Rotylenchulus reniformis*, has also been found infesting cardamom (Eapen, 1995a).

Distribution

Widespread occurrence of root-knot nematodes, *M. incognita* and *M. javanica* has been reported in cardamom nurseries and plantations in

India (Kumar *et al.*, 1971; Koshy *et al.*, 1976; Ali and Koshy, 1982a; Ali, 1984, 1986a; Raut and Pande, 1986).

Symptoms

The infested cardamom seedlings at the two-leaf stage show marginal yellowing and drying of leaves and severe galling on roots. On transplantation to a secondary nursery, the seedlings exhibit curling of the unopened leaves. These leaves mostly emerge after the breaking open of the pseudostem. In secondary nurseries, the infested plants are stunted and yellowed with poor tillering, drying of leaf-tips and margins, and heavy galling on roots (Ali and Koshy, 1982a; Eapen, 1995b). The leaves show varying degrees of chlorosis, narrowing and drying at leaf tips and margins. Root system of infested plants shows varying degrees of root galling (Fig. 3).



Fig. 3 - Root system of a root-knot nematode infested cardamom plant

It is observed that young seedlings are more susceptible to root knot nematode attack than mature plants (Eapen, 1992). Heavy infestation with root-knot nematode in mature plants in a plantation causes stunting, reduced tillering, yellowing, premature drying of leaf-tips and margins, narrowing of leaf blades, a delay in flowering, immature fruit-drop and reduction in yield (Fig. 4). Though galling of roots is not prominent, the infested roots exhibit a "witches broom" type of excessive branching. The heavily shaded, hot, humid atmosphere and continuous availability of soil moisture usually prevalent in cardamom plantations are congenial conditions for the multiplication of root-knot nematodes. A yield loss of 32-47% has been reported due to root knot nematode infestation (Ali, 1985, 1986b). Microplot studies under simulated field conditions showed

46.6% yield loss at an initial inoculum level of 4 nematodes/100 cc soil (Eapen, 1987; Eapen, 1994).

Interaction with other organisms

Root knot nematode, *M. incognita* is the predisposing factor to *Rhizoctonia solani* infection, causing damping off and rhizome rot, which is quite prevalent in cardamom nurseries (Eapen, 1987; Ali and Venugopal, 1992; 1993). *Meloidogyne* population was 5-10 times more in 'katte' (a virus disease) affected cardamom plants (Ali, 1989).

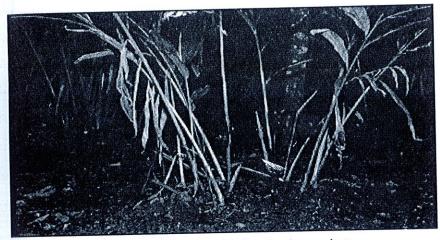


Fig. 4 - Root-knot nematode infested cardamom plants.

Ginger (Zingiber officinale Rosc.)

Plant parasitic nematodes belonging 17 genera were reported on ginger (Sundararaju et al., 1979b; Kaur, 1987; Ramana and Eapen, 1998; Rama and Dasgupta, 1998) (Table 2). However, the most important nematode pests are *Meloidogyne* spp., R. similis and P. coffeae.

Table 2 – Plant parasitic nematodes associated with Zingiberaceous spices in India.

Nematode species	Cardamom	Ginger	Turmeric
Aphelenchoides spp.	*	*	×
Aphelenchus sp.		*	*
Basirolaimus spp.		*	*
Bitylenchus spp.		*	*
Caloosia sp.		*	*
Criconema spp.	*		
Criconemella spp.			_

Cont'd...

Discocriconemella elettaria	*		*
Helicotylenchus spp.	*		
Hemicriconemoides spp.	*	*	*
Hemicycliophora spp.	*	*	*
Hoplolaimus sp.	*	*	*
Longidorus sp.	*		
Macroposthonia spp.			*
Meloidogyne spp.		*	*
Ogma taylata		*	*
Paratrichodorus sp.	*	*	*
Pratylenchus spp.	*		
Radopholus similis	*		*
Rotylenchus sp.	*	*	*
Rotylenchulus reniformis	*	*	*
Scutellonema sp.	*	*	*
Trichodorus minor	*	*	*
Tylenchorhynchus sp.	*		
Tylenchus sp.	*	*	* *
Xiphinema sp.	*		*

Source: Ramana and Eapen, 1998 * indicates reports of nematode infestation.

History

Root knot nematode infestation in ginger was first reported by Nagakura (1930) while Hart (1956) reported parasitism by *R. similis* in Florida, USA. Later, Butler and Vilsoni (1975) reported heavy infestation of ginger by *R. similis* in Fiji and its further spread through infested seed rhizomes. Occurrence of *R. similis* along with *M. incognita*, *Pratylenchus* sp. and *Helicotylenchus* sp. has also been reported from India (Charles, 1978; Charles and Kuriyan, 1980). Recently a new root knot nematode species, *M. thalandica*, is reported from Thailand (Handoo *et al.*, 2005).

Distribution

Several species of root knot nematodes viz. *M. arenaria*, *M. hapla*, *M. incognita* and *M. javanica* have been reported as parasites of ginger in various countries including India. In Kerala, *M. incognita*, *R. similis* and *Pratylenchus* spp. were the major nematodes species found in the rhizosphere of ginger (Mammen, 1973; Charles, 1978; Charles and Kurian, 1980; Anonymous, 1993; Sheela *et al.*, 1995). *R. reniformis* and

M. incognita were the dominant nematodes associated with ginger in Orissa (Routaray et al., 1987b). The predominant nematode species in ginger rhizosphere are M. incognita and P. coffeae in Himachal Pradesh (Srivastava et al., 1998; Kaur et al., 1989; Khan and Makhnotra, 1998), M. incognita in Madhya Pradesh (Vadhera et al., 1998a), M. arenaria in Himachal Pradesh (Kaur and Sharma, 1988), R. reniformis, Hoplolaimus indicus and P. coffeae in West Bengal (Rama and Dasgupta, 1998, 2000) and Pratylenchus spp. in Sikkim (Thapa et al., 2008a, b).

Symptoms

The root-knot nematodes cause typical galling of roots. The fibrous roots were very much reduced. In addition, nematodes parasitizing the rhizomes produce galls in the outer tissues. Infested rhizomes have brown, water soaked areas in the outer tissues (Huang, 1966; Cheng and Tu, 1979). *Radopholus similis* produce small, shallow, sunken, water soaked lesions on roots (Vilsoni *et al.*, 1976; Sundararaju *et al.*, 1979c).

Heavily infested plants show external symptoms like stunting, poor tillering and chlorotic leaves with marginal necrosis. Several physiological and chemical changes take place in the plant in the early phase of nematode infection (Yin et al., 2005). The affected ginger plants mature, dry faster and die prematurely than healthy ones, leaving a poor crop stand at harvest. Infested rhizomes serve as a source of infection and means of dissemination. P. coffeae is reported to cause 'ginger yellows' disease, prevalent in Himachal Pradesh and some North-Eastern States (Kaur and Sharma, 1990). The nematode infestation caused yellowing of leaves and dry rot like symptoms with dark, brown necrotic lesions on rhizomes. The dry rot symptoms caused by Pratylenchus spp. are indistinguishable from those caused by some fungi.

Economic importance and yield loss

M. incognita is widely distributed in ginger fields in Kerala and causes a loss of upto 46.4% (Charles, 1978). A reduction of 74% rhizome weight has been recorded at 10,000 Pi root-knot nematodes per plant over a period of six months under potted conditions (Sudha and Sundararaju, 1986). In another study on ginger, an avoidable yield loss of 43% was observed at an initial population level of 166 M. incognita larvae per 250 g soil (Sheela et al., 1995). M. incognita significantly reduced the height, root biomass and yield of ginger at an initial inoculum level of 02.2 to 200 J₂/100 cc soil (Ramana and Eapen, 2001). Significant damage may be noticed at 0.5 and 1.25 Meloidogyne J₂/g of soil in sterilized soil under potted conditions. The fibrous roots may be

greatly reduced at $2J_2/g$ soil (Parihar, 1985; Routaray *et al.*, 1987*a*). Carbofuran (@ 1 kg a.i./ha) treated ginger plots showed of 20 % increase in the yield (Makhnotra and Khan, 1997).

Severe infestation of rhizomes with root knot nematodes reduced yield by 57% as determined by fumigation in Queensland, Australia (Pegg et al., 1974). On overall basis in Fiji, R. similis incidence has been reported more than 50% of the total area with 10-50% infection that results about 40% to yield reductions (Vilsoni et al., 1976). A Pi of to nematodes/plant reduced shoot weight, root weight and rhizome weight by 43, 56 and 40%, respectively, in a pot experiment (Sundararaju et al., 1979c). The nematode is highly pathogenic to 15 day old ginger seedlings even at a lower inoculum level such as 10 nematodes in sterilized soil (Kaur, 1987). Rhizomes of ginger harbour very large populations of P. coffeae in Sikkim (Thapa et al., 2008a).

Interaction with other organisms

Incidence of rhizome rot of ginger caused by *Pythium aphanidermatum* is reported to be severe when rhizomes are infested with *M. incognita* and/or *P. coffeae* (Dohroo *et al.*, 1987). Recent studies have shown that, ginger plants inoculated with root knot nematodes developed disease symptoms early on inoculating with *P. aphanidermatum* (Ramana *et al.*, 1998). However, Doshi and Mathur (1987) could not observe any interaction with these two organisms. Similarly, there was no interaction between *M. incognita* and *Pythium myriotylum* also (Lanjewar and Shukla, 1985). Studies have proved that bacterial wilt of ginger caused by *Ralstonia solanacearum* is not influenced by nematode infection (Ramana *et al.*, 1998) even though there are some contradictory reports (Samuel and Mathew, 1983).

Turmeric (Curcuma longa L.)

Among the 35 nematode species reported infesting turmeric, *Meloidogyne* spp., *R. similis* and *P. coffeae* are of considerable economic importance (Table 2).

History

Root-knot nematode on turmeric was first reported by Ayyar (1926) and is the only nematode that received maximum attention on this spice in the past.

Distribution

A number of species of plant parasitic nematodes have been reported associated with turmeric in India (Nirula and Kumar, 1963:

Sundararaju et al., 1979b; Dasgupta and Rama, 1987; Gunasekharan et al., 1987; Haider et al., 1998a; Rama, 1987; Mani and Prakash, 1992; Routaray et al., 1987b; Bai et al., 1995) of which Meloidogyne spp., R. similis and P. coffeae are economically important. Two species of root-knot nematodes, M. incognita and M. javanica have been reported to be prevalent in India. R. reniformis and M. incognita were the most dominant and frequently recorded nematode species in the Chittoor and Cuddapah districts of Andhra Pradesh (Mani and Prakash, 1992; Sudheer et al., 2008) and in Bihar (Haider et al., 1995) in India. R. reniformis was reported to be more harmful to turmeric than M. incognita, and produced a significantly higher growth reduction at the lower population level (Haider et al., 1998a).

Symptoms

Turmeric plants infested with *M. incognita* have stunted growth, yellowing, marginal and tip drying of leaves and reduced tillering with galling and rotting of roots. In the field, higher populations of *M. incognita* cause yellowing and severe stunting and withering in large patches. Plants die prematurely leaving a poor crop stand at harvest. Infested rhizomes tend to lose their bright yellow colour (Mani *et al.*, 1987). The population density of *M. incognita* increases with crop age and decreased with crop senescence (Poornima and Sivagami, 1999).

Roots of turmeric damaged by *R. similis* become rotted and most of these decayed roots retain only the epidermis devoid of cortex and stelar portions. The infested plants show a tendency to age and dry faster than healthy plants. Infested rhizomes are of a yolk yellow colour compared with the golden yellow colour of healthy rhizomes and have shallow water-soaked brownish areas on the surface. The scale leaves also harbour *R. similis* (Sosamma *et al.*, 1979).

Economic importance and yield loss

Significant reduction in growth and yield of turmeric were noticed in plants inoculated with >1000 root knot nematode juveniles per plant (Sudha *et al.*, 1989). An avoidable yield loss of 33.6% was reported in turmeric due to root-knot nematode infestation (Ray *et al.*, 1995). When four varieties of turmeric were tested against 2 juveniles of *M. incognita* per gram of soil, maximum reduction (18%) of fresh rhizome weight was observed in the cv. Suvarna (Ramana and Eapen, 2001). Poornima and Sivagami (1998a) reported that an initial inoculum level of > 5000 *M. incognita* larvae per plant was highly pathogenic to turmeric. Avoidable

yield loss under field conditions was 45.3% due to *M. incognita* but was only 33.3% in a mixed infestation of *M. incognita* and *R. reniformis* (Bai *et al.*, 1995). Pathogenicity studies using *R. similis* showed that an inoculum level of 10 nematodes per plant caused 35% reduction in the rhizome weight after four months and 46% reduction at the end of the season (8 months). With 100,000 nematodes, the extent of reduction in rhizome weight was 65 and 76%, after 4 and 8 months, respectively (Sosamma *et al.*, 1979).

Other spices

Although a number of spice crops including clove, nutmeg, cinnamon, cassia, tamarind, kokum, vanilla and an array of seed spices are cultivated over large areas in the tropics and subtropics, there is very little information available on the damage and yield loss caused by plant parasitic nematodes on many of them. Random surveys have revealed infestation with a few nematodes on clove (Ghesquiere, 1921; Goodey et al., 1965; Sharma and Loof, 1974; Bridge, 1978; Sundararaju et al., 1979b), nutmeg (Goffart, 1953; Goodey et al., 1965; Kumar et al., 1971; Sundararaju et al., 1979b; Chawla and Samathanam, 1980), cinnamon (Goffart, 1953; Goodey et al., 1965; Sundararaju et al., 1979b; Chawla and Samathanam, 1980; Dasgupta and Rama, 1987; Rama, 1987) and vanilla (Orton Williams, 1980; Stier, 1984 in Bridge, 1988). All these spices are hosts of Meloidogyne spp. The important nematodes are root knot nematodes (Meloidogyne spp.) on cinnamon, nutmeg and clove, Pratylenchus sp. on clove and cinnamon, R. similis on nutmeg and Rotylenchulus reniformis on clove, nutmeg and cinnamon. There are reports on severe galling of roots of cumin by M. incognita and M. javanica (Patel et al., 1986) and reduced growth of vanilla vines by Pratylenchus brachyurus in the Pacific island of Tonga (Stier, 1984 in Bridge, 1988). Celery plants infested by M. incognita showed severe yellowing and stunting, with heavily deformed and damaged root systems (Voylas et al., 2008). M. incognita enhanced the wilting of cumin when inoculated in combination with Fusarium oxysporum f.sp. cumini (Sharma and Trivedi, 2003).

Nematode management in spices

Nematode control is essentially prevention, because once a plant is parasitized it is becomes very difficult to kill the nematode without destroying the host (Guerena, 2006). There is seldom a single method to alleviate the nematode problems. This warrants a sustainable approach to

nematode control integrating several tools and strategies, including cover crops, crop rotation, soil solarization, least toxic pesticides, biological control and plant varieties resistant to nematode damage (Barker and Koenning, 1998). A brief review on sustainable management of plant parasitic nematodes has been published recently by Eapen (2008). The aim should be to bring down the nematode populations below economic injury levels. The management of potential nematodes infesting spices is discussed crop wise.

Black pepper

Preventive: Planting rooted cuttings infested with nematodes in the field leads to slow decline disease in due course of time. Use of certified planting material, use of soil less growing media in greenhouses/nurseries, good agricultural practices and phytosanitation can reduce nematode spread to a greater extent. Uprooting of diseased black pepper vines, destruction of the vines along with root mass and replanting after a period of 9-12 months have to be undertaken in all nematode damaged plantations. This will reduce the source of inoculum and the spread of the pathogens.

Black pepper is propagated through vegetative stem cuttings. As the conditions provided in the nurseries are highly congenial for nematode multiplication, nurseries are the main source for the spread of nematodes. Using planting materials from known, nematode free sources and selecting healthy propagating materials without symptoms of nematode infestation are important. Manual removal of nematode infested roots in black pepper rooted cuttings can also help to bring down the nematode inoculums. Denematization of nursery mixture either through solarization or fumigation with chemicals is highly effective in reducing the initial nematode load and production of healthy rooted cuttings. For large scale production of nematode free planting materials, soil mixture can be sterilized with solar heat, steam or soil fumigants in black pepper and cardamom nurseries (Ramana et al., 1994; Thankamani et al., 2008). By fortifying potting mixture with biological control agents, nematodefree cuttings can be raised. A number of organisms have been tested and found effective in suppressing root knot nematodes.

Cropping practices: Exclusion of nematode suceptible intercrops, supports or standards can also minimise the nematode infestation. Among the large number of live standards used in black pepper gardens, coconut and arecanut are good hosts of *R. similis* while several of these are highly susceptible to root knot nematodes too. However, *Garuga*

pinnata Roxb. and Macaranga indica Wight are not susceptible to root knot nematodes and the popular live standards, Erythrina indica Lank. and Gliricidia sepium (Jacq.) Walp. are less susceptible. Several inter crops like banana, ginger and turmeric and a large number of weeds are also susceptible to these nematodes (Koshy et al., 1977; Ramana, 1986). Large numbers of weeds that are found in pepper gardens have been recorded as hosts of the root-knot nematodes.

Soil amendments: There is substantial evidence that the addition of organic matter in the form of compost or manure will decrease nematode pest populations and associated damage to crops. Some sources of organic matter known to be nematode-suppressive include oilcakes, sawdust, bone meal, manures, compost, and certain green manures. Application of neem cake @ 2 kg/vine was highly effective against M. incognita than R. similis (Ramana et al., 1992). Mulching and biofumigation ameliorate symptoms of slow-decline/pepper yellows. Addition of chopped leaves of Gliricidia maculata (10g/kg soil) as green manure reduced populations of R. similis and increased the plant growth (Jasy and Koshy, 1992). Mulching the basins with Gliricidia leaves reduced root knot nematodes in Sri Lanka (Ratnasoma et al., 1991). Aqueous leaf extracts of Chromolaena odoratum, Piper colubrinum, all spice, etc. were also reported to exhibit nematicidal properties against M. incognita population of black pepper (Anon, 1993; 1994).

Resistant lines: Developing genetically resistant or tolerant varieties is the best alternative for effective nematode management. A number of black pepper germplasm accessions, including wild types, were screened against M. incognita and R. similis by several workers (Venkitesan and Setty, 1978; Koshy and Sundararaju, 1979; Jacob and Kuriyan, 1979a; Leong, 1986; Paulus et al., 1993). Piper hymenophyllum, P. colubrinum, P. attenuatum and some other accessions like HP 39 and C.820 were found to be resistant to R. similis (Eapen, 2006). One variety PW14 was reported as totally immune to R. similis in Sri Lanka (Gnanapragasam, 1989). P. colubrinum is now widely used as a root-stock to graft cultivated pepper plants (Ramana et al., 1987b; Ramana, 1992). One cultivar, CLT-P-812 was found resistant to M. incognita and was later released as 'Pournami' for cultivation in root knot infested areas (Ravindran et al., 1992) (Fig.5). Some of the wild related species of Piper are also resistant to root knot nematodes (Ramana and Mohandas, 1986; Ramana, 1992). However, a few black pepper lines resistant to R. similis have been reported (Eapen, 2006).

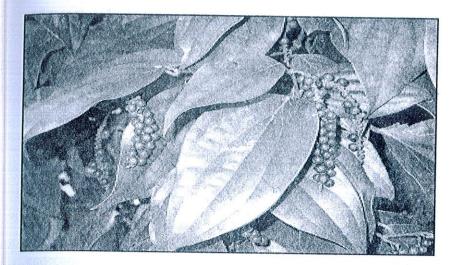


Fig. 5 - 'IISR Pournami', a root-knot nematode resistant black pepper variety.

Chemical control: As a preventive measure, black pepper planting materials can be treated with granular nematicides like phorate or carbofuran @ 0.1g a.i./plant. Application has to be made once in two months (Mohandas and Ramana, 1987). This will ensure eradication of nematodes and production of healthy roots. Carbofuran/phorate at 3 g a.i./vine applied in May/June and again in September/October resulted in the remission of foliar yellowing and reduction in nematode populations in plantations. Various nematicides like phenamiphos @ 20 g/vine (Nambiar and Sarma, 1977), aldicarb sulphone @ 8 kg/ha (Venkitesan and Setty, 1979). Phorate, carbofuran and aldicarb @ 3 g a.i./vine (Venkitesan and Charles, 1979; Venkitesan and Jacob, 1985; Mohandas and Ramana, 1987; Ramana, 1992) are effective in controlling nematodes infecting black pepper in India. In Malaysia, carbofuran @ 114 g/vine (Kueh and Teo, 1978) and phenamiphos and oxamyl (Kueh, 1979) were effective for the control of root-knot nematodes. Leong (1986) reported that fenamiphos @ 1% a.i. was most effective in controlling root-knot nematodes followed by carbofuran and ethoprophos in Sarawak, Malaysia. Similarly, in Indonesia several nematicides were all effective to control nematodes of black pepper (Mustika and Zainuddin, 1978). In Brazil, Ichinohe (1980) found application of aldicarb or carbofuran @ 1.25 g/plant twice a year reduced nematode population and improved the growth. Mustika and others (1984) stated that the severity of pepper yellows was reduced by application of fertilizer (NPK 15:15:15) at 250 g/plant /year and either with aldicarb (50g/plant) of mancozeb (12 g/plant) or both since the

disease can be caused by a multiple attack by nematodes (*M. incognita* and *R. similis*) and *Fusarium oxysporum*.

The chances of rehabilitating the severely affected vines by application of nematicides are slim because of the heavy damage already caused to the root system and the inability of such plants to put out fresh roots for quick rejuvenation. The nematicide may be applied after removing the top soil without causing damage to the roots, followed by replacement of the soil.

Biological control: Considering the export nature of these crops and the high organic status of the soil in which they are cultivated, biological control is an ideal option for managing plant parasitic nematodes infesting spices. Biological control goes hand in hand with organic agriculture being practiced by several farmers. The efforts in this direction have been compiled by several workers (Eapen and Ramana, 1996; Ramana *et al.*, 2002; Anandaraj and Eapen, 2006; Khan *et al.*, 2009).

Manjunath and Bagyaraj (1982) reported the occurrence of vesicular arbuscular mycorrhizae (VAM) on black pepper roots. The beneficial effects of VAM on black pepper were also reported (Shivashanker and Rohini, 1988; Bopaiah and Khader, 1989). Promising isolates of biocontrol agents like vesicular arbuscular mycorrhizae viz., *Glomus mossae*, *G. fasciculatum*, *Acaulospora laevis*, *Gigaspora margarita*, which are suppressive of nematodes on black pepper (Anonymous, 1991; Anandaraj *et al.*, 1991, 1996c) can be incorporated with the nursery soil mixture for better establishment and production of nematode free planting materials. Similarly inoculation of pepper rooted cuttings with *G. fasciculatum* or *G. etunicatum* reduced root knot nematode (*M. incognita*) population in the root and rhizosphere soils (Sivaprasad *et al.*, 1990; 1992).

A number of antagonistic fungi, which parasitize or prey on nematodes, have been identified from spice based cropping systems (Eapen et al., 2005) and further evaluated in greenhouse and field trials for managing nematode pests of spices. Ramana (1994) and Sosamma and Koshy (1997) found that Paecilomyces lilacinus suppressed nematode infestation in black pepper and increased the production of root mass. Freire and Bridge (1985) found high infestation of eggs of M. incognita by P.lilacinus and Verticillium chlamydosporium under artificial inoculation but to a lesser extent in the egg mass from the roots of black pepper inoculated with the fungi. Pochonia chlamydosporia (=Verticillium chlamydosporium) was isolated from black pepper gardens (Sreeja et al., 1996). Several Pseudomonas isolates were

screened against root knot nematodes infesting black pepper (Eapen et al., 1997). Bacillus pumilis, B. macerans and B. circulans significantly reduced M. incognita population and improved the growth of black pepper plants (Sheela et al., 1993). Pasteuria penetrans was also reported to inhibit root knot nematodes infesting black pepper (Sosamma and Koshy, 1997). Field experiments at Kanyakumari showed that treatment with P. fluorescens (Pfbv22) and B. subtilis (Bbv57) @ 10g/vine lead to significant increase in the yield over control (Jonathan et al., 2010). Devapriyanga et al. (2010) tested the efficacy of native strains of Pseudomonas and Bacillus against M. incognita and found that isolate PfIc and BsPC decreased root-knot nematode infection and increased the plant growth.

There are only very few successful attempts to control *R. similis* by using any of the fungal bioagents, probably due to the migratory endoparasitic nature of this nematode (Geetha, 1991; Ramana, 1994; Eapen *et al.*, 2009). The mycorrhizal fungus, *Glomus fasciculatum* suppressed burrowing nematode infestation (Anandaraj *et al.*, 1996c). Recently, rhizobacteria that suppressed both *R. similis* and *M. incognita* infesting black pepper were identified in greenhouse studies (Beena, 2008; Beena *et al.*, 2001, 2003; Aravind *et al.*, 2009). Two promising endophytic bacteria viz. *Bacillus megaterium* and *Curtobacterium luteum* suppressing *R. similis* have been identified recently (Eapen, 2007; Aravind *et al.*, 2010).

Cardamom

Preventive: Pruning roots of cardamom seedlings is advisable to prevent the entry of nematodes to the main plantations. It is also advisable to change nursery sites every year. Disinfestation of nursery beds with methyl bromide at 500 g/10 m² is effective in controlling root-knot infestation in cardamom nurseries (Ali and Koshy, 1982b).

Soil solarization, a method of pasteurization, can effectively suppress most species of nematode wherever summers are predictably sunny and warm. It has been successfully employed in many crops, as a pre-sowing treatment to bring down the initial nematode population in a field. Solarization is more detrimental to plant parasitic than saprophytic nematodes (Ostrec and Grubisic, 2003). In cardamom there was 10% increase in germination, increased growth and vigour of the seedlings resulting in a considerable increase in the number of transplantable seedlings due to soil solarization (Eapen, 1995b).

Resistant lines: Several accessions of cardamom have been screened against root knot nematode using standardized techniques (Eapen, 1990).

None of them were found resistant or tolerant to root knot nematodes (Eapen, 1995b; Hegde *et al.*, 1993).

Chemical control: Disinfestation of nursery beds with methyl bromide at 500 g/10m² is effective in controlling root-knot infestation in both primary and secondary nurseries (Ali and Koshy, 1982b). It has been demonstrated that application of aldicarb at 5 kg a.i./ha three times, every three months, results in increased growth and vigour of seedlings both in primary and secondary nurseries (Koshy *et al.*, 1979*a*; Jacob and Chandrasekharan, 1984; Ali, 1986a, 1987a, b). Drenching of nursery beds with fenamiphos also significantly reduced root knot nematodes (Ali, 1986b). However, in another study, application of phorate @ 2.5-5.0 g a.i./plant reduced the nematode population and increased the yield by more than 40% (Eapen, 1995b).

Biological control: In cardamom there are reports that *Gigaspora margarita* and *Glomus fasciculatum* suppressed *M. incognita* infestation and enhanced growth and vigour of seedlings (Thomas *et al.*, 1989). *Paecelomyces lilacinus* (Eapen, 1995b; Eapen and Venugopal, 1995) and some native isolates of *Trichoderma* spp. (Eapen *et al.*, 2000) are potential antagonists of root knot nematodes.

Ginger and turmeric

Preventive: Excellent reduction in populations of plant parasitic nematodes can be achieved through use of various heat treatments like steam, hot water and sunlight. In ginger and turmeric, using the seed materials collected from healthy fields and also treating the rhizomes in hot water at 50-55°C for 10 minutes were found to reduce the nematode incidence in ginger (Colbran and Davis, 1969; Anonymous, 1971). Disinfestation of ginger (Vadhera et al., 1998 a, b) and turmeric (Chen et al., 1986) rhizomes was achieved by hot water treatment at 45°C for 3 h too. But all these techniques are employed only under special conditions and are not reliable for economic control of nematodes when used in isolation. Soil solarization is another technique that was found effective for the control of soil borne diseases in ginger and turmeric.

Cropping practices: In ginger, the control schedule for *M. javanica* involving the use of clean seed and a ginger-taro-fallow rotation has been recommended in Fiji (Haynes *et al.*, 1973).

Soil amendments: Reduced nematode damage from increased organic matter in soil is likely a combination of improved soil structure and fertility, alteration of the level of plant resistance, release of nematotoxins, or increased populations of fungal and bacterial parasites and

other nematode-antagonistic agents. Mulching or applying well decomposed cattle manure or poultry manure or compost or neem oil cake reduced nematode build up in ginger (Colbran, 1974; Kaur, 1987; Stirling, 1989; Mohanty *et al.*, 1992, 1995; Dohroo *et al.*, 1994; Vadhera *et al.*, 1998b).

Resistant lines: In a preliminary evaluation, a few lines of ginger (Acc. Nos. 36, 59 and 221) were found resistant to *M. incognita* (Eapen *et al.*, 1999). Cultivar UP was identified as tolerant to M. incognita in another study (Nehra and Trivedi, 2005). One of these has been recommended for release as 'IISR Mahima' (Sasikumar *et al.*, 2003). Similarly several workers screened a number of turmeric lines against root knot nematodes and reported a few lines as resistant (Chen *et al.*, 1986; Mani *et al.*, 1987; Gunasekharan *et al.*, 1987; Rao *et al.*, 1994; Eapen *et al.*, 1999).

Chemical control: Soil fumigation or application of granular pesticides like fenamiphos/carbofuran or dip treatment with fenamiphos is all recommended for control of nematodes of ginger (Colbran, 1972; Willers, 1985; Kaur, 1987; Mohanty et al., 1995). In turmeric too, significant yield increase had been noticed on applying granular nematicides like aldicarb, carbofuran, phenamiphos or phorate (Patel et al., 1982; Gunasekharan et al., 1987; Mani et al., 1987; Haidar et al., 1998b). Integrated management of the plant parasitic nematodes infesting these crops has been successful with combined application of carbofuran and neem oil cake (Makhnotra et al., 1997; Mohanty et al., 1992; Mohanty et al., 1995; Ray et al., 1995; Sheela et al., 1995).

Biological control: Recently several fungal bioagents viz. *Aspergillus nidulans*, *Fusarium oxysporum*, *P. lilacinus*, *T. viride*, *Verticillium lecanii* and *Pochonia chlamydosporia* have been successfully used in ginger and turmeric fields to suppress root knot nematodes (Eapen *et al.*, 2008). Application of antagonistic fungi increased the yield by 17.5-45.6 % in ginger and 12.5-40.9% in turmeric. Among the ten fungi evaluated, *P. chlamydosporia* was superior in suppressing root-knot nematodes and increasing ginger yield. Sampat *et al.* (2003) has reported application of mycorrhizal fungi for controlling root knot nematodes infesting ginger.

Conclusion and future prospects

An overview of the nematode problems of various spices and condiments has revealed that plant parasitic nematodes cause serious damage to many of these plants. Most of the nematological investigations in the earlier years were mere survey reports or host range studies. This was followed with experiments to control these pests using nematicides. Considering the export oriented nature of spices, more

emphasis should be given for developing ecofriendly nematode management practices with minimum use of pesticides. Of late, research on biological control is gaining momentum. There is a very wide scope for isolating more efficient biocontrol agents useful against nematodes, in a tropical country like India. Developing resistant lines is another thrust area in spices as India possesses rich genetic resources of most of these crops. Modern biotechnology tools will be of immense use for achieving rapid successes in this field. The changing climate, cropping pattern etc. warrant a fresh look into the distribution pattern of plant parasitic nematodes associated with spice crops. Therefore, nematological investigations on this economically important group of crops have to be intensified and strengthened in the coming years.

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Nematode Infestation in Ginger

B.C. BORA and B.N. CHOUDHURY

Department of Nematology, Assam Agricultural University, Jorhat-785013

email: bcbora05@yahoo.co.in

Abstract

Ginger, Zingiber officinale is presumed to have originated from the Southern Asian and Indian regions. The crop is grown commercially in several countries of the world mostly used as a spice and to some extent for medicinal purposes. India is the largest producer and exporter of dry ginger in the world market. The country also produces and exports value added products of ginger like ginger oil and ginger oleoresin. However, the crop is suffered from many pests and diseases including plant parasitic nematodes, which causes significant losses in yield and acreage. All together there are 32 species belonging to 14 different genera have so far been recorded around the ginger growing countries in the world. Among them, root-knot nematode, Meloidogyne spp. is the prime nematode pest causing serious yield loss to the crops followed by burrowing nematode, Radopholus similis and root lesion nematode, Pratylenchus coffeae. Nematode pests and diseases account for crop losses that are variously estimated to be between 10-50%. The nematodes not only causing direct damage to the crops but also encourage the entry of some of the fungal and bacterial pathogens namely Pythium spp., Fusarium spp. causing rhizome rot and Ralstonia solanacearum causing wilt of ginger. Soil application of carbofuran @ 3 kg a.i./ha as post plant treatment along with adoption of certain alternative options such as cultural control like use of clean planting materials and rotation with non-hosts and nematode suppressive crops proved effective against nematode pest of ginger. Soil solarization, sanitation and use of hot water treatment are some of the effective physical means of management ag. 1st both nematodes and pathogens prior to planting which could help growers to reduce damage to the crop.

Introduction

Ginger (Zingiber officinale Rose) under the family Zingiberaceae is one the important and popular cash crops grown widely all over the

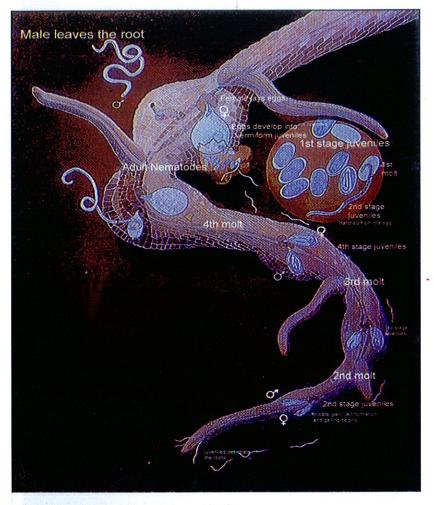


Fig. 3

Nematode Infestation in Spice Cultivations

Santhosh J. Eapen



Fig. 1.



Fig. 2



Fig. 3



Fig. 4



Fig. 5.

Nematode Infestation in Groundnut

P. Kalaiarasan, M. John Sudheer and P. Vetrivelkalai



Fig. 1

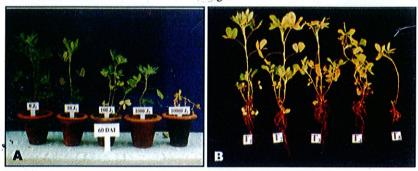


Fig. 2



Fig. 3