PLANT PARASITIC NEMATODES ASSOCIATED WITH PINEAPPLE (*Ananas comosus* L. *Merr.*) IN KERALA

BY

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THESIS

Submitted in partial fulfilment of the requirement for the degree

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DECLARATION

I hereby declare that this thesis entitled "Plant parasitic nematodes associated with pineapple (*Ananas comosus* L. Merr.) in Kerala" is a bonafide record of research work done by me during the course of research and that the thesis had 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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Vellanikkara,
11 November, 1981.

CERTIFICATE

Certified that this thesis, entitled 'Plant parasitic nematodes associated with pineapple (Ananas comosus L. Merr.) in Kerala" is a record of research work done independently by Shri. Hamza Koya.C.P. under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to him.

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CERTIFICATE

We, the undersigned members of the Advisory Committee of Shri. Hamza Koya. C.P., a candidate for the degree of Master of Science in Agriculture majoring in Agricultural Entomology agree that the thesis entitled "Plant parasitic nematodes associated with pineapple (Ananas comosus L. Merr.) in Kerala" may be submitted by Shri. Hamza Koya. C.P. in partial fulfilment of the requirements for the degree.

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Introduction

INTRODUCTION

Pineapple, Ananas comosus L. Merr., queen of fruits, is one of the most important commercial fruit crops of the world. The crop comes up well in tropical climates. In India this crop is largely cultivated in Assam and Kerala. Cultivation of pineapple as a commercial fruit crop in India during the year 1975-76 was estimated to cover 36,000 hectares with a production of 2,00,000 tonnes. In Kerala the total area under the crop during 1978 was 8971 hectares, the production being 63,000 tonnes (Anon, 1978).

India has good potential to become one of the major exporters of pineapple since optimum conditions for the pineapple cultivation exists in the States of Assam, Kerala, Tripura, West Bengal, Orissa, Goa and Karnataka.

In Kerala, the crop is cultivated mainly in the Districts of Trichur, Quilon, Ernakulam and Cannanore. Planters from most part of Kerala have been reporting progressive decline in fruit yield.

Association of plant parasitic nematodes has been reported by several workers (Guerout, 1975). The first record on association of plant parasitic nematodes on

pineapple in Kerala has been reported by Singh, Rao and Reddy (1979). The nematodes reported were Helicotylenchus spp., Hemicreconemoides spp., Pratylenchus spp. and Rotylenchulus reniformis.

Elsewhere the most destructive nematode associated with pineapple crop has been reported to be Meloidogyne spp. (Linford, 1952; Alvarez-Gracia and Lopez, 1954).

Though the association of nematodes has been suspected in pineapple in Kerala, studies on the nematodes infesting pineapple have not been carried out so far in the State.

The present studies were carried out to gather information on the different nematode genera associated with this crop in Kerala and on the extent of damage inflicted by Meloidogyne spp. which is reportedly the most destructive species.

As part of the present studies, the different pineapple gardens in the State were surveyed for the occurrence of plant parasitic nematodes associated with the crop. The pathogenic effect of the root-knot nematode (M. incognita) on pineapple cv 'kew' was also investigated.

Review Of Literature

REVIEW OF LITERATURE

The first record on association of plant parasitic nematodes, particularly of *Meloidogyne* spp. with pineapple was reported as early as 1911 by Bessy. The other species reported were *Pratylenchus brachyurus* (Godfrey, 1929), *Radopholus similis* (Cassidy *et al.*, 1927), *Rotylenchulus reniformis* (Yap and Linford, 1940) and *Rotylenchus erythrinae* (Parris, 1940). Ayala *et al.* (1970) reported that out of 15 genera of plant parasitic nematodes associated with pineapple, *Helicotylenchus*, *Meloidogyne*, *Rotylenchulus*, *Pratylenchus* and *Creconemoides* were associated with root decay.

Recently Guerout (1975) has reviewed literature on the plant parasitic nematodes associated with pineapple and stated that nearly 23 genera consisting of 36 species were reported by various workers. Out of the 23 genera, *Meloidogyne*, *Pratylenchus*, *Rotylenchulus*, *Helicotylenchus* were of major importance. The commonly occurring nematode species include *Helicotylenchus digonicus*, *H. dihystera*, *Hoplolaimus californicus*, *Meloidogyne* spp., *Pratylenchus brachyurus*, *Rotylenchulus reniformis*.

The details of different species of nematodes reported to occur in pineapple are furnished in Table A.

Table A (Contd.)

Name of the nematode	Country	Author
Helicotylenchus spp.	Puerto Rico	Ayala (1969)
Helicotylenchus spp.	Mexico	Adams and Epinosa (1972)
Helicotylenchus spp.	Jamaica	Hutton (1975)
Helicotylenchus spp.	Carribean Islands	Samuel et al.(1976)
Helicotylenchus spp.	South Africa	Department of Agricultural Service, Praturia (1977).
Helicotylenchus spp.	Venezuela	Suerez (1978)
Helicotylenchus spp.	Panama and Venezuela	Tarjan (1967)
Helicotylenchus digonicus	Thailand	Boduang and Ratnaprapa (1975)
Helicotylenchus dihystra	Hawaii, Puerto Rico, Australia, Ivory Coast & Madagascar	Torrealba (1969)
H. nannus	Indonesia, Cuba and Jamaica	Daiz - Silveira(1967)
H. erythrine	Puerto Rico, Cuba and Jamaica	Daiz - Silveira(1967)

(Contd.)

Table A (Contd.)

Name of the nematode	Country	Author
Helicotylenchus multicinctus	Puerto Rico and Ivary coast	Luc (1964)
Heterodera spp.	Cuba	Diaz - Silveira (1967)
Hemicreconemoides spp.	India	Singh, Rao and Reddy (1979)
Hoplolaimus californicus	Cuba	Diaz - Silveira (1967)
Hoplolaimus indicus	Cuba	Diaz - Silveira (1967)
Hoplolaimus pararobustus	Cuba	Diaz - Silveira (1967)
Longidorus spp.	Mexico	Adams and Epinosa (1972)
Longidorus spp.	Puerto Rico	Torrealba (1969)
Longidorus elongatus	Cuba	Diaz - Silveira (1967)
Longidorus laevieapitatus	Cuba	Diaz - Silveira (1967)
Meloidogyne spp.	India	Singh, Rao and Reddy(1979)
Meloidogyne spp.	Ivory coast	Guerout (1968)
Meloidogyne spp.	Puerto Rico	Ayala (1969)
Meloidogyne spp.	Mexico	Adams and Epinosa (1972)

(Contd.)

Table A (Contd.)

Name of the nematode	Country	Author
Meloidogyne spp.	Venezuela	Suerez (1978)
M. incognita	Puerto Rico	Ayala (1968)
M. incognita	Puerto Rico and Cuba	Torrealba (1969)
M. incognita	Venezuela	Suerez (1978)
M. incognita acrita	Puerto Rico, Ivory Coast and Rhodesia	Luc *et al.* (1964)
M. javanica	South Africa	Department of Agricultural Technical Service, Pratoria (1977)
M. javanica	Hawaii, Australia and Rhodesia	Colbran (1962)
Peltamigratus browni	West Indies	Khan, and Zakiauddin (1968)
Pratylenchus spp.	India	Singh, Rao and Reddy (1979)
Pratylenchus spp.	Puerto Rico	Ayala (1969)
Pratylenchus spp.	Mexico	Adams and Epinoza (1972)

(Contd.)

Table A (Contd.)

Name of the nematode	Country	Author
Pratylenchus spp.	Jamaica	Hutton (1975)
Pratylenchus spp.	Venezuela	Suerez (1978)
P. brachyurus	Hawaii, Puerto Rico, Australia, Ivory Coast, Madagascar	Godfrey (1929)
P. brachyrus	Ivory coast	Guerout (1968)
P. brachyrus	Brazil	Monteiro and Lordello (1971)
P. brachyrus	South Africa	Department of Technical Service, Pratoria (1977)
P. brachyrus	Bahia State	Zein and Reinhardt (1975)
P. goodeyi	Cuba	Diaz - Silveira (1967)
P. pratensis	Cuba	Diaz - Silveira (1967)
P. Zeae	Puerto Rico and Australia	Torrealba (1969)
Paratylenchus spp.	Venezuela	Suerez (1978)
Paratylenchus spp.	Puerto Rico, Cuba Panama	Torrealba (1967)

(Contd.)

Table A (Contd.)

Name of the nematode	Country	Author
Paratylenchus minutes	Australia	Colbran (1958)
Psilenchus magnidens	Australia	Colbran (1962)
Psilenchus tumidus	Australia	Colbran (1962)
Radopholus similis	Hawaii and Ivory Coast	Torrealba (1969)
Rotylenchulus spp.	Puerto Rico	Ayala (1969)
Rotylenchulus spp	South Africa	Department of Agricultural Technical Service, Pratoria (1977)
Rotylenchulus spp.	Venezuela	Suerez (1978)
R. reniformis	India (Kerala)	Singh, Rao and Reddy (1979)
R. reniformis	Puerto Rico	Ayala (1968)
R. reniformis	Jamaica	Hutton (1975)
R. reniformis	Carribean	Samuel et al. (1976)
R. reniformis	Hawaii, Puerto Rico, Martinique, Jamaica	Linford and Oleveira (1940)

(Contd.)

Table A (Contd.)

Name of the nematode	Country	Author
Rotylenchoides spp.	Saire (Brazzaville)	Luc *et al.* (1964)
Rotylenchus robustus	Cuba	Diaz - Silveira (1967)
Rotylenchus spp.	Puerto Rico, Kenya, Panama	Heyns (1966)
Scutellonema spp.	South Africa	Department of Agricultural Technical Service, Pratoria (1977)
Scutellonema spp.	Kenya	Hollis (1962)
Scutellonema bradys	Ivory Coast	Luc *et al.* (1964)
S. clathricaudatum	Zaire (Brazzavilla)	Luc *et al.* (1964)
S. brachyurus	Jamaica	Hutton (1975)
Trichodorus spp.	Puerto Rico and Martinique	Merny (1962)
Trichodorus minor	Australia	Colbran (1958)
Trichodorus primativus	Cuba	Diaz - Silveira (1967)
Tylenchornynchus digitatus	Cuba	Diaz - Silveira (1967)

(Contd.)

Table A (Contd.)

Name of the nematode	Country	Author
Tylenchornynchus ebriensis	Ivory Coast	Luc (1960)
Tylenchorynchus spp.	Kenya and Jamaica	Hollis (1962)
Tylenchus spp.	Venezuela	Surez (1978)
Tylenchus spp.	Puerto Rico, Ivory Coast, Panama and Venezuela	Torrealba (1969)
Xiphenema spp.	Puerto Rico, Jamaica, Venezuela	Torrealba (1969)
Xiphenema spp.	Mexico	Adams and Epinosa (1972)
Xiphenema dimorphicaudatum	South Africa	Heyns (1966)

Pathogenic effect caused by different nematodes on pineapple

Godfrey (1929) reported that all the stages of Pratylenchus brachyurus were able to penetrate the roots, even the gravid female which lay their eggs very rapidly after penetration into the cortex parenchyma. In the laboratory, females extracted from the roots laid two to three eggs a day for one week. Most of the time roots still grows, even if the penetration occurs near the root cap. However, if the infestation is serious occurring on the secondary roots, these are quickly destroyed.

Pratylenchus spp. do not cause specific symptoms on the aerial part of the plant. Working on young roots, Godfrey (1931) observed very limited necrosis which was hardly noticeable on plants growing in soil. However, when the parenchyma of the root is destroyed, the cortex separates easily from the central cylinder.

Linford et al. (1949) reported that even continued feeding of Pratylenchus for several days did not produce any pathological changes in the root. As the Pratylenchus feed on plant, without causing their death, these may be considered as well adapted obligatory ectoparasites.

Guerout (1969) recorded that the yield increase in pineapple was closely associated with decrease in populations of Pratylenchus brachyurus in the roots.

The infection of Meloidogyne spp. (Heterodera marioni) on pineapple has been studied in Hawaii (Godfrey, 1936). Root growth was greatly slowed down when this nematode was present. He recorded that root elongation was 8 to 8.1 mm/day in the absence of infestation and this was reduced to 2.4 - 2.88 mm, 1.34 - 1.79 mm and 1 mm when the plants were infested with 10, 250 and 1000 larvae, respectively.

Godfrey and Hagan (1937) compared the plants having only two root knots with those having 72 knots and recorded reduction of plant weight (16.4%), root length (27.6%), and of leaf weight (15%), but the number of leaves were not affected and there was a noticeable stimulation (28.5%) of root proliferation.

Linford (1952) reported that most destructive of the nematode that attacked pineapple roots were the species of Meloidogyne. The characteristic galls were found occurring both on pineapple roots and roots of weed plants growing in pineapple fields. In a field where pineapple was grown as a third crop, the severity of the

root knot was associated with low plant vigour.

Infection by Heterodera marioni (Meloidogyne incognita) on leaves and stems of pineapple was recorded by Linford (1941).

Alvaraz, Gracia and Lopuz (1954) reported the influence of root knot nematode on the decline in vigour of the red spanish variety of pineapple in Pureto Rico. The examination of the roots from the pineapple plants grown in this fields showed heavy infestation with nematodes.

Linford (1941) reported the feeding habits of the larvae of Heterodera marioni, Rotylenchulus reniformis Pratylenchus paratensis, Rotylenchus crythrinae and Paratylenchus spp. All of these begin feeding on the surface cells and continue to feed while entering into the roots. Cell walls are punctured by the stylet, saliva injusted into the cell followed by sucking out of the cell contents. Cell destruction follows when walls have been weakened by repeated puncture and the nematode then break into the cells. Paratylenchus spp. feeds chiefly at the root surface and on root hairs, less frequently centering the cortex.

In Puerto Rico, the nematode population composed predominantly R. reniformis which reduced the plant

population by more than 83 per cent.

The work published by Ayala (1962) on the pathogenicity of R. reniformis on pineapple was not very conclusive, although 10,000 larvae were inoculated on each plant. He attributed this to the short duration (4 months) of the experiment.

According to Ayala (1968), Rotylenchulus reniformis, Helicotylenchus spp. and Meloidogyne incognita were considered to cause the greatest crop loss in Puerto Rico.

Guerout (1975) reported that due to the attack of Meloidogyne spp. (Heterodera marioni) at Hawaii or Pratylenchus brachyurus in the Ivory Coast resulted in crop losses to a tune of 40 per cent during the first crop.

Yield losses of pineapple in South Africa due to nematodes exceeded 50 per cent as reported by Keetch (1978).

Materials And Methods

MATERIALS AND METHODS

I. Survey

a) Selection of zones for survey

Five districts where pineapple is cultivated on commercial scale were selected for the survey. The locations for surveying was selected in the pockets where large scale cultivation was practiced.

b) Collection of soil and root samples

From the fields in each location, 5 - 10 plants were randomly selected for the sampling. Soil and root samples were collected from the root zones at a depth of 15 - 20 cm of these plants. A representative sample consisting of 250 g of the soil and 25 g of the roots were sampled out and these were put in polythene bags and tied with rubber bands after labelling for further studies. A proforma was developed (Appendix I) for collecting all information on the crop and the locations included in the survey.

c) Extraction of nematodes from soil samples

Plant parasitic nematodes were extracted from the soil samples (100 ml) by modified Cobb's method of

sieving and sifting technique (Christie and Perry, 1951). The nematode suspension was drawn out after 48 hours. Out of 250 ml of soil collected, only 100 ml was used for nematode extraction.

d) Extraction of nematodes from root samples

10 grams of the roots were weighed out from each sample and were gently cleaned of soil adhering to them by holding in a stream of water under a tap. The cleaned roots were then sliced into small bits of less than 1 cm in length. These were kept on moist tissue paper supported by a wire guaze in a petri-dish containing water to the level of the gauze. The setting was left undisturbed and changing water every 24 hours and was continued for 48 hours. The extracts were pooled, concentrated and used for the determination of the nematode counts.

e) Estimation of nematode population in soil and root extracts

The nematode suspension drawn from the petri-plates were allowed for the settlement of the nematodes for three hours and the volume was reduced to 25 ml by pouring off the supernatent water.

From the 25 ml suspension, an aliquot of 0.5 ml of the nematode suspension was drawn out in a nematode counting dish and the different genera present were counted using a wild stereobinocular microscope at 60x. This was repeated for three times and the average of three observations was then multiplied by the factor to get the actual population of different nematode genera in the whole suspension. The nematode specimens were then killed and preserved in 3 per cent formalin for further studies.

II. Identification of nematodes

Permanent slides of the nematode specimens from the suspension were made as per slow glycerine method (Southey, 1970) and identification of major group to species level.

III. Pot culture experiment for determination of the extent of damage caused by M. incognita.

a) Preparation and sterilization of soil

Pot mixture was prepared by mixing sieved field red soil, sand and well decomposed farm yard manure in the ratio 1:1:1. This pot mixture was denematized by injecting 5 per cent Formaldehyde per every 900 cm^2 area of the bed with 30 cm depth and kept away from contamination.

Whenever this pot mixture was to be used for experimental purposes, it was examined to confirm that no nematodes were present.

b) Collection of pineapple suckers

70 - 80 days old pineapple suckers of cv 'kew' with uniform growth were collected from the Pineapple Research Centre, Vellanikkara for this experiment.

c) Raising and maintenance of pineapple suckers

70 - 80 days old pineapple suckers with 9 - 10 leaves and an average weight of 300 g were selected and planted in 12' x 12" pots filled with 7.5 kg denematized pot mixture. These were maintained by adopting the recommended agronomic practices.

d) Pure culture of Meloidogyne incognita

Pure cultures of M.incognita were raised from single egg masses collected from the stock culture plants of tomato maintained in the Department, after identifying this species by preparing and examining the perennial pattern. Enough numbers of whole plant cultures with pure population were maintained regularly to obtain sufficient larval population for inoculation purposes.

e) Method of inoculation

One day old larvae were used for inoculations. For obtaining one day old larvae of the nematode, a large number of egg masses from the culture plants were hand picked and kept in many cavity blocks containing sterile water. Care was taken to see that the egg masses were in contact with water. After 24 hours, the suspension in each cavity block was collected and pooled. The number of larvae per ml of suspension was determined by taking the mean of three counts. This suspension was made into suitable aliquots to get the larval population of 10, 100, 1000 and 10,000 larvae for inoculation. Each aliquot wasused for inoculating each potted plant. The inoculation was done by boring five holes of about 10 cm deep around the plant at 7.5 cm away from plant base. The holes were dug using a glass rod and closed immediately with denematised soil immediately after inoculation. The inoculation of all the pots were completed on the same day (16.2.1981). Pots were irrigated to keep the soil just moist. Then four inoculum levels with a check (no nematodes) were tested for the pathogenic effect. Each treatment (inoculum level) was replicated six times. Randomised block design was adopted for the experiment.

f) Observations

The following observations on each plant were recorded after 180 days (18.8.1981) of the nematode inoculation.

1) Plant height
2) Fresh top weight
3) Total number of leaves per plant
4) Length of leaves (a) 5 leaves from the first whorl
   (b) 5 leaves from the second whorl
   (c) 5 leaves from the third whorl
5) Length of stem
6) Weight of stem
7) Total fresh weight of the roots
8) Root-knot index
9) Final soil population per pot

Plant height

The height of the plants were measured from the base of the plant to the highest leaf length available, using a meter scale.

Fresh top weight

The plants were cut at the ground level and the entire fresh top weight was recorded.

Total number of leaves per plant

Number of leaves were counted from each plant after separating all the leaves of the plant from the ground level.

Length of leaves

From each plant 15 leaves namely five leaves from the first whorl, five from the second whorl and the five from the third whorl were selected for recording the length using a meter scale.

Length of stem

After recording the stem weight, the stem was vertically cut into two equal halves and the actual length of the stem was measured using a centimeter scale.

Weight of the stem

The stem formation was noticed in most of the plants. After removing the leaves and the fibrous roots the weight of stems were recorded.

Total fresh weight of the root

Fresh weight of the roots removed from the stem was recorded.

Root knot index

The root system of each plant was carefully

examined, for gall formation and the number of galls were recorded.

Final nematode population in the pot soil

A sample of 100 ml of the soil was taken from each pot by thorough mixing of the soil in the pot and after removal of the plants. The nematodes were extracted and estimated as per the procedure already indicated. The total population in the pot soil was determined by computing and multiplying with the factor 75.

The data were analysed as per the method of Cochran and Snedecor (1969).

Results

RESULTS

I. Survey of nematodes associated with pineapple in Kerala

The survey was conducted during the months of January, February, March and April 1981. A total of 120 samples were collected. The districts covered and the locations from samples drawn and nematodes encountered are listed in Table 1.

Quilon District

Out of 20 samples collected three samples were free of plant parasitic nematodes. The remaining 17 samples contained Rotylenchulus spp., Helicotylenchus spp., Melodogyne spp., Creconemoides spp. and Pratylenchus spp. Among the above genera of nematodes, the population of Rotylenchulus was most predominant. The population of Rotylenchulus in soil samples varied from 0 - 316 per 100 ml soil with an average of 54. In the case of Helicotylenchus spp. the population range was 0 - 130 with average of 11. Meloidogyne showed a range between 0 - 15 with an average of four. The population of Creconemoides spp. and Pratylenchus spp. were very low.

Ernakulam District

Out of the 30 samples collected nine were found to be free of plant parasitic nematodes. The other 21 samples collected contained Rotylenchulus spp., Helicotylenchus spp. and Creconemoides spp. Of these Helicotylenchus spp. was the most predominant one. The population of Helicotylenchus spp. in soil sample varied from 0 - 450 per 100 ml soil with an average of 54. In case of R. veniformis, the population range was 0 - 150 with an average of 15. The population of Creconemoides was very low.

Trichur District

It was observed that out of the 40 samples collected and analysed 19 samples were free of any plant parasitic nematodes. The remaining 21 samples contained R. veniformis, Helicotylenchus spp., Creconemoides spp. and Pratylenchus spp. Rotylenchulus spp. was the most widely prevalent nematode in this district. The population of this nematode in soil samples varied from 0 - 516 per 100 ml soil with an average of 82. Helicotylenchus spp. was the next important nematode and their number ranged from 0 - 550 with an average of 18. The population of Creconemoides and Pratylenchus was found in negligible numbers

Calicut District

Among 10 samples collected, one sample was free of plant parasitic nematodes. The other nine samples contained mostly Tylenchus veniformis, Helicotylenchus spp., Meloidogune spp., Hoplolaimus spp. and Hemicyclophora spp. The dominant nematode among them was Rotylenchulus spp. and its population varied from 0 - 293 with an average of 43. Helicotylenchus spp. population ranged from 0 - 100 and the average was 25. The population of Meloidogyne ranged from 0 - 33 and the average was four. The population of Hoplolaimus and Hemicyclophora spp. were negligible.

Cannanore District

Out of the 20 samples collected, four samples were free of plant parasitic nematodes. In the remaining 16 samples nematodes consisted of Rotylenchulus spp., Helicotylenchus spp., Creconemoides and Pratylenchus spp. Among these Rotylenchulus was the most dominant one. The population of this nematode ranged from 0 -516 per 100 ml soil with an average of 161. The Helicotylenchus spp. population varied from 0 - 180 with an average of 28. The population of other nematodes were very negligible.

Table 1. Details of locations surveyed and population densities of different nematodes (per 100 ml soil)

Districts	Rotylenchulus	Helicotylanchus	Meloidogyne	Criconemoides	Hoplolaimus	Pratylenchus	Hemicyclophora
1	2	3	4	5	6	7	8
Quilon							
Malayalapuzha	50	33	-	1	-	-	-
Malayalapuzha	---	33	-	-	-	-	-
Malayalapuzha	150	--	50	16	-	-	-
Konni	-	-	-	-	-	-	-
Konni	316	33	33	-	-	-	-
Piravanthur	50	-	1	-	-	-	-
Vettithitta	16	-	-	-	-	-	-
Vettithitta	33	130	-	-	-	-	-
Vettithitta	116	33	-	-	-	-	-
Piraventhur	66	---	-	-	-	-	-
Piraventhur	83	-	-	-	-	-	-
Piravanthur	16	-	-	-	-	-	-
Pimananthapura	33	-	-	-	-	-	-
Pimananthapura	-	-	-	-	-	-	-
Pimananthapura	21	-	-	-	-	-	-
Pimananthapura	3	2	-	-	-	-	-
Pinanthepuram	-	-	-	-	-	-	-
Pinanthepuram	-	1	-	-	-	-	-
Pravanthur	11	-	-	-	-	-	-
Pravanthur	100	-	-	-	-	-	-

(Contd.)

Table 1 (Contd.)

1	2	3	4	5	6	7	8
Ernakulam							
Avoli	1	-	-	-	-	-	-
Avoli	150	-	-	-	-	-	-
Avoli	33	-	-	-	-	-	-
Vazhakulam	-	50	-	-	-	-	-
Vazhakulam	50	-	-	-	-	-	-
Vazhakulam	-	-	-	-	-	-	-
Vazhakulam	5	-	-	-	-	-	-
Vazhakulam	5	2	-	-	-	-	-
Vazhakulam	-	65	-	-	-	-	-
Vazhakulam	-	-	-	-	-	-	-
Neerumpuzha	-	-	-	-	-	-	-
Neerumpuzha	33	180	-	1	-	-	-
Neerumpuzha	50	-	-	-	-	-	-
Neerumpuzha	-	1	-	-	-	-	-
Neerumpuzha	50	450	-	-	-	-	-
Kallurkad	-	4	-	-	-	-	-
Kallurkad	-	33	-	-	-	-	-
Kallurkad	-	-	-	-	-	-	-
Kallurkad	-	-	-	-	-	-	-
Kallurkad	-	-	-	-	-	-	-
Manjellur	-	-	-	-	-	-	-
Manjellur	5	-	-	-	-	-	-
Manjellur	33	330	-	-	-	-	-
Menjummel	-	1	-	-	-	-	-
Menjummel	-	-	-	-	-	-	-
Menjummel	83	200	-	-	-	-	-
Menjummel	-	280	-	-	-	-	-

(Contd.)

Table 1 (Contd.)

1	2	3	4	5	6	7	8
Menjummel	-	1	-	-	-	-	-
Menjummel	-	-	-	-	-	-	-
Menjummel	-	4	3	-	-	-	-
Trichur							
Pineapple Research Station (Vellanikkara)	-	1	-	-	-	-	-
Pineapple Research Station (Vellanikkara)	2	-	-	-	-	-	-
Pineapple Research Station (Vellanikkara)	2	-	-	-	-	-	-
Pineapple Research Station (Vellanikkara)	-	-	-	-	-	-	-
Pineapple Research Station (Vellanikkara)	166	-	-	-	-	-	-
Amala Cancer Institute Estate	-	-	-	-	-	-	-
Amala Cancer Institute Estate	-	-	-	-	-	-	-
Amala Cancer Institute Estate	-	-	-	-	-	-	-
Amala Cancer Institute Estate	-	-	-	-	-	-	-
Amala Cancer Institute Estate	-	33	-	-	-	1	-
Chittilapalli	-	-	-	-	-	-	-
Chittilapalli	-	-	-	-	-	2	-
Chittilapalli	-	-	-	-	-	-	-
Chittilapalli	-	-	-	-	-	-	-
Allapat Estate	-	-	-	-	-	-	-
Allapat Estate	-	-	-	-	-	3	-
Allapat Estate	-	-	-	-	-	-	-
Churanattukkara	-	-	-	-	-	-	-
Churanattukkara	-	-	-	-	-	-	-
Churanattukkara	-	-	-	-	-	-	-
Kuttanellur	1	-	-	-	-	-	-
Kuttanellur	116	-	-	-	-	-	-
Kuttanellur	116	-	-	-	-	-	-

(Contd.)

Table 1 (Contd.)

1	2	3	4	5	6	7	8
Manathakkara	68	-	-	-	-	-	-
Manathakkara	68	-	-	-	-	-	-
Elanthuruthi	450	-	-	-	-	-	-
Elanthuruthi	-	550	-	-	-	-	-
Kalyan Products (Nadathara)	-	-	-	-	-	-	-
Kalyan Products (Nadathara)	400	-	-	-	-	-	-
Kalyan Products (Nadathara)	200	1	-	-	-	-	-
Banana Research Station, Kannara	116	-	-	-	-	-	-
Banana Research Station, Kannara	416	66	-	-	-	-	-
Panancherry	-	-	-	9	-	-	-
EVU Estate (Alpara)	-	-	-	-	-	-	-
EVU Estate (Alpara)	8	5	-	-	-	-	-
EVU Estate (Alpara)	250	66	-	-	-	-	-
Alpara	516	2	-	-	-	-	-
Alpara	466	-	-	-	-	-	-
Instructional Farm, (Vellanikkara)	-	-	-	-	-	-	-
Instructional Farm (Vellanikkara)	-	-	-	6	-	-	-
Instructional Farm (Vellanikkara)	-	-	-	-	-	-	-
<u>Calicut</u>							
A.R.S.Koothali	-	16	-	-	-	-	-
A.R.S.Koothali	83	-	-	-	-	-	-
Mukkola	33	-	-	-	-	-	-
Pulivayal	-	3	-	-	2	-	-
Kalladanod	-	100	-	-	-	-	-
Karachalil	33	-	-	-	-	-	1
Karachalil	-	-	-	-	-	-	-
Chamal	-	83	33	-	-	-	-

(Contd.)

Table 1 (Contd.)

1	2	3	4	5	6	7	8
Kunchukulam	-	-	3	-	-	-	-
Kunchukulam	-	66	-	-	2	-	-
Kuppayakod	283	2	-	-	-	-	-
Cannanore							
Malur	100	-	-	-	-	-	-
Malur	100	-	-	-	-	1	-
Malur	133	33	-	13	-	-	-
Malur	433	-	-	-	-	-	-
Pazhassi	500	50	-	-	-	-	-
Pazhassi	283	83	-	-	-	-	-
Pazhassi	83	66	-	-	-	-	-
Pazhassi	50	50	-	-	-	-	-
Pazhassi	316	50	-	-	-	-	-
Koothuparambu	200	-	-	-	-	-	-
Kalliyasseri	-	-	-	-	-	-	-
Kalliyasseri	--	-	-	-	-	-	-
Sivapuram	215	-	-	-	-	-	-
Sraniachal	-	-	-	-	-	-	-
Sraniachal	30	-	-	-	-	-	-
Chalode	516	15	-	-	-	-	-
Chalode	-	-	-	-	-	-	-
Koodali	1	-	-	-	-	-	-
Koodali	166	33	-	-	-	-	-
Sreekandapuram	50	180	-	-	-	-	-

The roots collected along with soil samples did not show any infection by plant parasitic nematodes.

II. Frequency of occurrence of nematodes in the samples in the area surveyed

Among the 120 samples collected and examined Rotylenchulus spp. were found in 64 locations followed by Helicotylenchus in 43 locations, Meloidogyne and the Creconemoides in 6 each, Pratylenchus in 5, followed by Hoplolaimus and Hemicyclophora in two and one respectively. Thirty-six samples were found free of any plant parasitic nematode. The data are presented in Table 2. The results indicated that Rotylenchulus and Helicotylenchus spp. were the predominant nematodes associated with pineapple cultivation in Kerala.

III. Nematode population as influenced under two systems of planting and age of the crop

The details of nematode population under the two systems of planting and age of the crop was separately compiled from the detailed gathered in the proforma (vide appendix I). The results are presented in Table 3. It was observed that R. reniformis population increased with the age of the crop from 1st to 4th year under ridge system from 66 per 100 ml soil during first year followed by 160, 169 and 271 during second, third and fourth years

respectively. But in the case of the pineapple planted in the bed method the population density of the nematode was highest during first year (157) and showed an erratic trend in density levels with 80, 107 and 95 during second, third and fourth years respectively.

In the case of Helicotylenchus spp. nematode population density showed an increasing tendency with a population of 33, 63 and 87 from the first year to the third year respectively and this was followed by a sudden decrease of 45 during the 4th year in the ridge method of planting. The same tendency was noticed in the bed system of planting also with a population of 17, 107, 123 and 17 for the 1st, 2nd, 3rd and 4th year respectively.

The population of other genera of nematodes observed were very negligible and population levels was eratic.

IV. Identification of nematodes

The species of Rotylenchulus was identified as Rotylenchulus reniformis, Helicotylenchus was suspected to be a new species and the description and differential diagnosis of the same are given herewith.

Head annules - Either three or four

Body - Open, spiral when relaxed

Head - Conical, not set off from the body

Stylet - 20 - 21 µ with metachium and telachium of almost same length

Stylet knob - Slightly intended anteriorly, Anterior cephaline 2 to 3 annuli's below basal plate and posterior six annuli's below basal plate.

Median bulb - Oval

Oesophagial gland - Overlap intestine ventrally

Excretory pore - Just above the oesophagial intestinal valve

Vulva - A transverse slit without any lateral mumbrane (epiptegma)

Tail - Dorsally convex, consists of about 12 - 15 ventral annules. Tail ends in a digitate process with 1 - 2 annules.

Phasmids - Located slightly below the annus

Diagnosis - Helicotylenchus n. spp. close resemblance to Helicotylenchus digitatus (Siddiqui and Hussain, 1964) but can be distinguished by the presence of anteriorly intended spear knobs, more posterior portion of excretory pore and absence of epyptegma.

Table 2. Frequency of occurrence of the nematodes in the samples in the area surveyed

Nematode	Frequency of occurrence
Rotylenchulus reniformis	64
Helicotylanchus spp.	43
Meloidogyne spp.	6
Creconemoides spp.	6
Pratylenchus spp.	5
Hoplolaimus spp.	2
Hemicyclophora spp.	1
Samples free of nematode	36

Table 3. Nematode population in 100 ml soil as influenced by the two system of plantings and age of the crop

Nematodes	Ridge				Bed			
	I year	II year	III year	IV year	I year	II year	III year	IV year
Rotylenchulus	66	160	169	271	157	80	107	95
Helicotylanchus	33	63	87	45	17	107	123	17
Meloidogyne	---	---	33	---	---	17	26	3
Creconemoides	1	5	-	---	---	9	6	-
Pratylenchus	-	-	3	---	1	2	1	1
Hoplolaimus	-	2	-	2	-	-	---	-

V. Extent of damage caused by Melodogyne incognita on pineapple

Attempts made on the visual observations of the plants under different treatments did not reveal any marked variation in their growth or health.

Observations on the plant characters at different inoculum levels of second stage larvae of M. incognita are presented in Tables 4 and 5. The observations were recorded after 180 days of inoculation of the test organism.

1. Height of the plant

The height of the plants were taken from the base to the highest leaf length. The data presented showed that plants without nematodes were having the maximum height (79.33 cm) whereas the plants inoculated with 10,000 larvae had the minimum (68.33 cm). A difference of 11 cm in height have been recorded between the plants with no nematodes and maximum larval population of 10,000 but this was not statistically significant.

2. Fresh top weight

The highest mean top weight of 505 g was recorded in treatment with T_1 plants and the minimum was recorded in T_4 plants (339.17 g). Eventhough the observations

(Table 5) revealed that there consistent was reduction in top weights at increased inoculum levels, the data were not statistically significant.

3. Total leaf production

The plants in T_4 produced the minimum number of leaves (26.16), whereas maximum number of 30.60 was produced by plants in Treatment T_2.

4. Length of leaves

The highest leaf length (43.63 cm) was observed in plant in T_0 compared to the lowest of 38.8 cm in plant in T_4 in the first whorl.

In case of the 2nd whorl the highest leaf length of 72.5 cm was observed in plants under T_3 whereas T_4 recorded the lowest leaf length (63.27 cm).

In the case of the leaves in the 3rd whorl the maximum leaf length (58.2 cm) was recorded by plants in T_0 whereas the plants in treatment T_4 recorded the lowest (48.17 cm).

Eventhough the leaf lengths in the three whrols recorded a difference of 4.63 cm, 9.23 cm and 9.5 cm respectively, the differences were not statistically significant.

any gall development. A few cases of slight swellings on the roots noticed were teased out to liberate any developing stages of the nematode. But stages were not detected and hence the Root knot index could not be worked out.

9. Soil population

The data on the soil population of nematodes is presented in Table 5. The larval population present in the pot soil at the end of the duration of the experiment showed that over 50 per cent of population were still surviving in the soil. However, there was no increase in their number when compared to the initial inoculum levels. This indicated that they did not readily multiplying on the host.

Table 5. Final nematode (larval) population in the pot soil - after 180 days of nematode inoculation

Treatment	Mean	F value	Inference
T_0	00.00		
T_1	67.00		
T_2	108.00	278*	Significant
T_3	567.00		
T_4	5158.00		

Discussion

DISCUSSION

Pineapple is one of the important fruit crops cultivated in Kerala. Canning industries in Kerala depend mostly on the fruits produced in the State. Recently some cultivators have been reporting that the production of the crop is on the decline particularly in ratoon crop from the second year onwards (Balakrishnan, 1979, Personal communication). Singh, Rao and Reddy (1979) reported the presence of parasitic nematodes in the rhizosphere of pineapple plants in Kerala.

The survey revealed that Rotylenchulus reniformis is the nematode commonly associated with the crop and this was observed in more than 76 per cent of the locations. Helicotylenchus spp. were observed in nearly 50 per cent of the locations. The other parasitic nematodes encountered were Meloidogyne spp. and Creconemoides spp. and Pratylenchus spp. The observation of Hoplolaimus and Hemicyclophora were negligible. All these nematodes have been reported to be associated with pineapple plants in several other countries (Guerout, 1975).

LOCATIONS SURVEYED IN DIFFERENT DISTRICTS

Quilon	Ernakulam	Trichur	Calicut	Cannanore
1. Malayalapuzha	1. Avoli	1. Pineapple Research Station (Vellanikkara)	1. A.R.S.Koothali	1.Malur
2. Konni	2. Vazhakulam	2. Estate Amala Cancer Institute	2. Mukkola	2.Pazhassi
3. Piravanthur	3. Neerumpuzha	3. Chittilapalli	3. Puliyavayal	3.Kuthuparamba
4. Vettithitta	4. Kallurkad	4. Allapat Estate	4. Karachalil	4.Kalliyasseri
5. Pimananthapura	5. Manjillur	5. Chooranattukara	5. Chamal	5.Sivapuram
6. Pravanthur	6. Menjummel	6. Kuttanellur	6. Kalladanod	6. Sraniachal
		7. Manathakara	7. Kunchukulam	7. Chalode
		8. Elanthuruthy		8.Koodali
		9. Kalyan Products (Nadathara)		9.Sreekandapuram
		10. Banana Research Station, Kannara		
		11. Pananchery		
		12. K.V.U. Estate(Alpara)		
		13. Instructional Farm, Vellanikkara.		

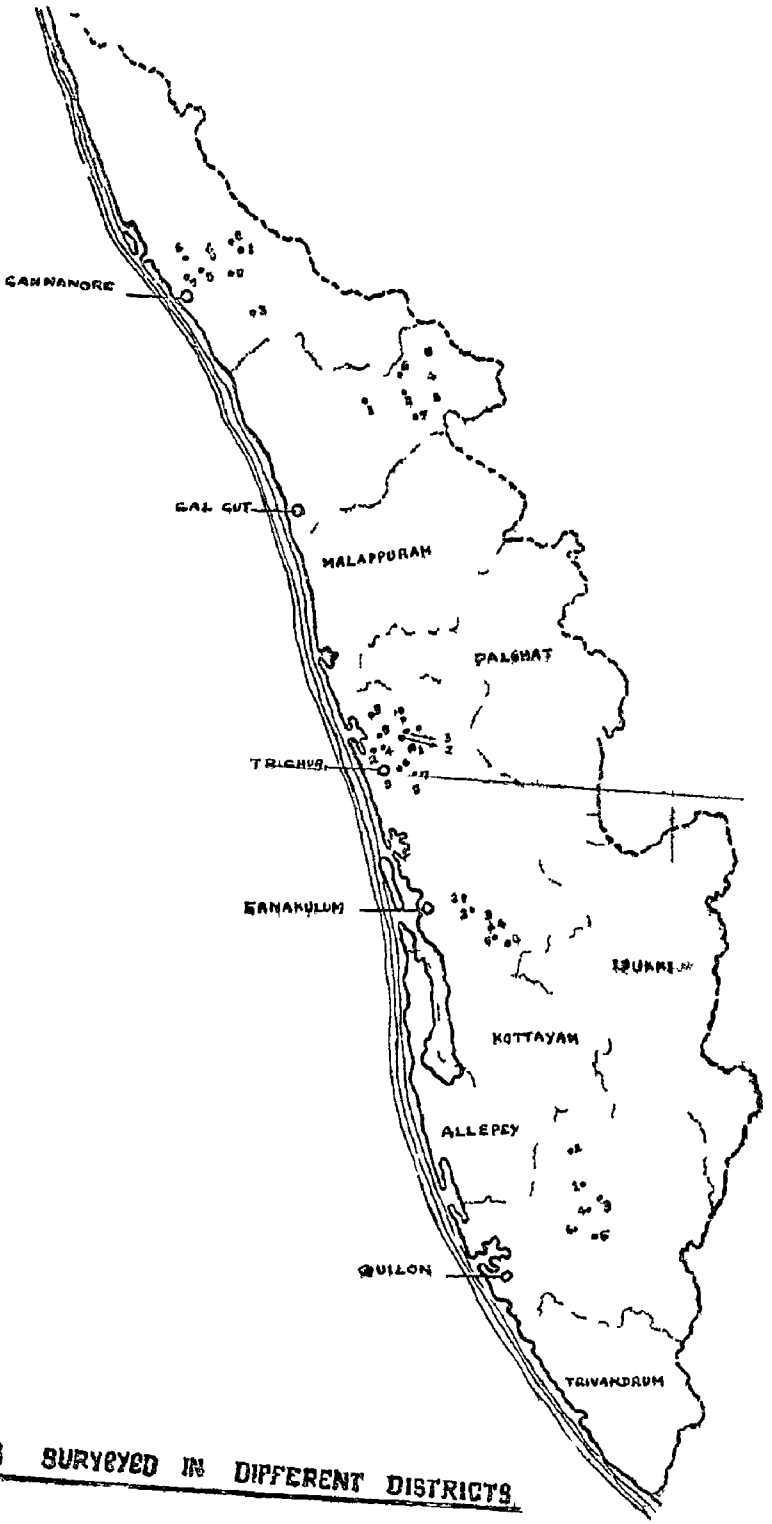

Fig 1 LOCATIONS SURVEYED IN DIFFERENT DISTRICTS

The association of R. reniformis was most predominant in the samples in Quilon, Trichur, Calicut and Cannanore (Fig. 2) whereas in Ernakulam District Helicotylenchus spp. was most dominant one. Pratylenchus spp. was associated with soil samples drawn from the Quilon, Trichur and Cannanore Districts only.

The nematode population in soil as influenced by the system of planting and the age of the crop revealed that the population build up of R. reniformis increased from first to fourth year under the ridge system and the population trends were erratic under the bed system of cultivation in the same crop duration. The nematode build up under ridge system might be due to the availability and concentration of root system for the proper feeding and multiplication of nematode and lesser weed growth in the ridge. In the case of bed system, the presence of erratic population levels might be due to the weed growth prevalent in between the pineapple plants which probably served as alternate host for the nematode population build up.

The species of Rotylenchulus was identified as R. reniformis and this species has been reported to be associated with pineapple in Puerto Rico

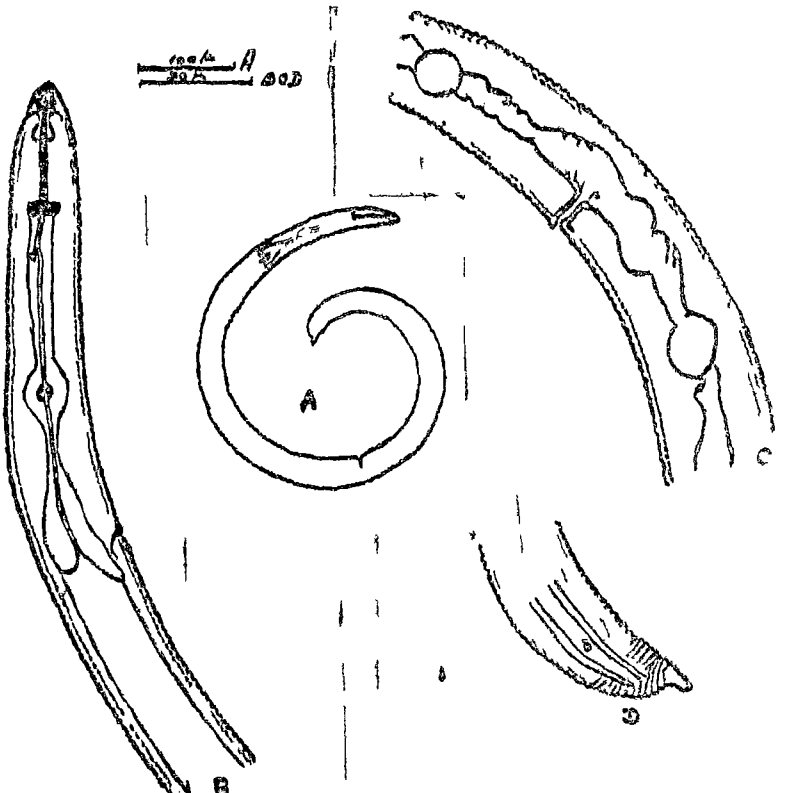

Fig.2. Helicotylenchus np. N. (?) Female
A. Female full size B. Anterior region showing stylet, oesophagus and position of excretory pore C.Vulval opening D.Tail characters; position of phasmid.

(Ayala, 1968), in Hawaii (Linford and Olivera, 1940), in the Carribean islands (Samuel et al. 1976), in Jamaica (Hutten, 1975) and this nematode is considered to be the most common nematode associated with pineapple. The specimens of Helicotylenchus examined from the location (Kalladanod and Neerumpuzha) revealed that it is a new species closely resembling to Helicotylenchus digitatus (Siddique and Hussain, 1964) but can be distinguished by the presence of anteriorly intended spear knobs, more posteriorly located excretory pore and absence of epiptegma.

The Root Knot nematode was selected as the test organism (M. incognita) because this nematode is reported to be present in all agricultural soil in the State infecting several other crops (Venkitesan, 1972; Arthur, 1977, Nadakal, 1963, 1964, 1965; Mammen, 1973a, 1973b; Raveendran and Nadakal, 1975). The pathogenic effects of this nematode have been reported by Godfrey (1936). Linford (1952) attributed this nematode as the most destructive one attacking pineapple. Hence studies were carried out to evaluate the pathogenic effect of M. incognita on this crop. Eventhough the data on the growth characters (Table 4) at different inoculum levels revealed difference

over the control, these were not statistically significant. The plant height, the weight of tops and leaf length did not reveal reduction between the plants not inoculated with nematode (check) and the plants inoculated with maximum number of larvae (10,000). No visual differences in these parameters could be observed. Godfrey (1931) observed that no specific symptoms on the aerial part of the plant were caused due to infection by Pratylenchus spp. Godfrey (1936) studied the infection of Meloidogyne spp. on pineapple and observed that root growth was greatly slowed in the presence of nematode. But observations taken on fresh root weight indicated that the root weight was considerably reduced (12.66 g) in plants inoculated 10,000 larvae compared to the root weight (19.0 g) in plants with no nematodes. Attempts made by Ayala (1962) to establish the pathogenicity of R. reniformis on pineapple did not give conclusive evidence even though a maximum of 10,000 larvae were inoculated to each plant. He attributed this to the short period (4 months) of the experiment. In this case also, the experimental duration was only six months. The final population of larvae in the pot soil (Table 5) revealed that 50 per cent of the test organism were still alive and

there is every possibility that they may be able to do damage to the growing plant subsequently. One of the striking observations was that gall formation did not occur on the root. The variety used for the test was 'kew' which is the only popular variety grown in Kerala and it is not known whether this particular variety has got any resistance to infection by the nematode. In the screening tests for resistance of pineapple against Heterodera marioni (M. incognita) carried out by Hagan and Collins (1932) they had not used 'kew' variety, to establish its resistance. A few swellings on the root of the inoculated plants were teased out to ascertain whether any developing stages of the nematodes were present and these stages were not be detected. Linford (1952) recorded formation of characteristic galls on pineapple roots due to infestation of species of Meloidogyne. However, in this study, the observations are not in confirmity with that of Linford's report. Alvarez et al. (1954) could observe heavy infestation of root knot nematode in the roots of red spanish variety of pineapple.

Since the survey work has indicated that reniform nematode R. reniformis and the spiral nematode

Helicotylencus spp. are of major importance in this State, it will be worthwhile to take up further studies on their role in reducing the crop yield in pineapple.

Summary

SUMMARY

A survey on the plant parasitic nematodes associated with the crop in Quilon, Trichur, Ernakulam, Cannanore and Calicut districts of Kerala State was carried out during the period January-April 1981. Rotylenchulus reniformis was found to occur in more than 76 per cent of the locations and Helicotylenchus spp. were recorded in about 50 per cent of the locations. The other plant parasitic nematodes detected in association with the crop included Meloidogyne spp., Creconemoides spp., Pratylenchus spp. Hoplolaimus and Hemicyclophora occurred in very low population densities.

Studies on the influence of systems of planting on the population build up indicated that the population of R. reniformis increased from the first to the fourth year under the ridge system. The population trends were erratic under the bed system of planting for the same crop duration. The variation in the population trends under the two systems of planting have been discussed.

The specimens of Helicotylenchus collected from Kalladanod and Neerumpuzha appeared to be new and

near to H. digitatus. The new species differed from H. digitatus by the presence of anteriorly indented spear knobs, more posteriorly located excretory pore and absence of epiptegma.

The pathogenic effect of the root knot nematode M. incognita on pineapple was investigated by inoculating sterilized soil around the base of the plant at larval loads of 10, 100, 1000 and 10,000. The plant height, weight of tops and leaf length were not affected by the nematodes even at the highest load of 10,000 larvae per plant.

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APPENDIX - I

KERALA AGRICULTURAL UNIVERSITY

Department of Entomology, College of Horticulture, Vellanikkara.

Survey of Pineapple in Kerala for plant parasitic nematodes.

(Data sheet of samples collected)

Village

NES block

Name & address of the cultivator

Taluk

IPD Unit

Total area under pineapple	I year	II year	III year	IV year and above
	New	Ratoon	Ratoon	Ratoon

Variety grown

Soil type Sandy/sandy loam/loam/clay loam/clay/laterite

Age of the crop (in days)

Nature of planting Ridges/beds/inflower/in fruits

Manuring Straight ferts/FYM/paddy Mix/oil cakes/green leave

Crop Irrigated/rainfed

Condition of the crop

General stand Good/average/bad/very poor/patchy growth

Foliage Health/chlorotic/severe yellowing

Pests noticed.

Disease observed:

Rotation followed

Weeds present (give local name)

(Contd.)

Appendix I (Contd.)

Pesticides used

Chemical sprayed

Granules applied

Name of location Date of collection

Date of processing: Soil Qty. Root

POPULATION PRESENT Soil Root

1. Meloidogyne
2. Heterodera
3. Hoplolaimus
4. Rotylenchulus
5. Helicotylenchus
6. Radopholus/Pratylenchus
7. Tylenchorhynchus
8. Xiphinema
9. Longidorus
10. Others

PLANT PARASITIC NEMATODES ASSOCIATED WITH PINEAPPLE (*Ananas comosus* L. *Merr.*) IN KERALA

BY

HAMZA KOYA, C P

ABSTRACT OF A THESIS

Submitted in partial fulfilment of the requirement
for the degree

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ABSTRACT

As a part of the studies on the occurrence of plant parasitic nematodes associated with pineapple in Kerala, a survey was carried out in different agro-climatic regions of the Kerala State and it was found that R. reniformis occurred in more than 76 per cent of the locations sampled. Helicotylenchus spp. were recorded in about 50 per cent of the locations. The other plant parasitic nematodes detected in association with the crop included Meloidogyne spp., Creconemoides spp., Pratylenchus spp., Hoplolaimus and Hemicyclophora occurred in very low population densities.

A new species of Helicotylenchus closely resembling H. digitatus (Hussain and Siddique, 1964) but differing with respect to spear knob characters and location of excretory pore has been reported to be associated with the crop.

The pot culture trial with M. incognita indicated that this nematode could not induce pathogenic effect on the cv Kew even at the highest inoculum levels of 10,000 larvae per 7.5 kg soil during the period of 180 days.