

## Diversity of migratory nematode endoparasites of banana

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### ABSTRACT

A roving survey was conducted on banana (*Musa* sp.) at Nadia and North 24-Parganas district of West Bengal and Vellayani areas of Thiruvananthapuram, Kerala, India during 2013-2014. Sixty seven soil and root samples (200cc) were collected from the banana fields for the study. Diversity of rhizospheric nematodes associated with banana revealed prevalence of 17 species of phytoparasitic nematodes, 3 species of predatory nematodes (*Mylonchulus*, *Iotonchus* and *Ironus*) and huge number of saprozoic nematodes. At Haringhata block of Nadia district *Pratylenchus crenatus* and *P. coffeae* were recorded as important migratory endoparasitic nematode pests of banana. Occurrence of *P. crenatus* is probably the first record of their existence on banana in West Bengal. Here, *Meloidogyne incognita*, *M. javanica*, *Rotylenchulus reniformis*, *Hoplolaimus indicus*, *Helicotylenchus dihystrera*, *Tylenchorhynchus mashhoodi*, *Criconemoides onoensis*, *Aphilenchus avenae* and *Xiphinema insigne* were also recorded from the crop rhizosphere. *Meloidogyne* spp. and *Rotylenchulus reniformis* were also found as most abundant endoparasites of banana at Habra-II block of North 24-Parganas. *Radopholus similis*, *Helicotylenchus multicinctus*, and *Pratylenchus coffeae* were recorded as abundant migratory nematode endoparasites of banana from Vellayani, Thiruvananthapuram, Kerala. Among those *H. multicinctus* was found most numerous in soil. Observation of different banana cultivars for the diversity of nematode fauna indicated that *Musa* (AAB) cv. *Pisang Bale*, *Ney Poovan* (AB) and *CO-I* (AAB) supported very low population of root knot nematodes. While, *Ney Poovan* (AB), *Karpooravelli* (ABB), *Booditha Bontha Bothisa* (CT), *Gross Michel* (AAA) and *Red banana* (AAA) supported very low population of *P. coffeae*. None of the cultivars of banana at the Mondouri farm under AICRP on Fruits, BCKV showed the existence of *R. similis* and *H. multicinctus*. Detailed morphometric and morphological informations on *P. coffeae*, *P. crenatus*, *H. multicinctus*, and *R. similis*, have been documented.

**Keywords:** Banana cultivars, migratory endoparasites, morphometric variability, nematode biodiversity

Banana, *Musa* spp. is an important fruit crop of the tropics and sub-tropics grown in nearly about 107 countries of the world, primarily for their fruit, and to a lesser extent for fibre, banana wine and banana beer. India, China, Philippines and Indonesia are the major banana producers in Asia. India is contributing 48% as against 17% in China to the total production in Asia from 37% and 15% of total area, respectively. Among various continents, Asia has the lion's share of 60% in global banana production (Mustaffa, 2011).

Banana and plantain, are continuously exhibiting a spectacular growth worldwide. India alone produces 27.01 million tons of banana from an area of 0.765 million ha (FAO STAT, 20011). India is the largest producer of banana in the world and also in Asia, and contributes 22.2% to global production from 7.4 % area in 2009 followed by China and Philippines. Increase in production is more significant in India and China due the technological interventions and adoption, while production in Ecuador, Columbia and Costa Rica has shown a declining trend during this period (FAO STAT, 20011; NHB, 2009). The acreage under banana crop in West Bengal is 7.8 lakh hectares with an annual production of 26.5 million tonnes in 2012-13. In India Tamil Nadu has the largest area

followed by Maharashtra and Karnataka. Tamil Nadu also ranked first in production, followed by Maharashtra. However, the highest productivity was recorded in Maharashtra followed by Gujarat, Tamil Nadu and Madhya Pradesh (Mustaffa, 2011). The yield of banana in West Bengal is approximately 24 t ha<sup>-1</sup> which is far lower than its potential yield (36t ha<sup>-1</sup>) in our country. Thus, there is further scope to improve the yield of banana in West Bengal. Like many other agricultural crops banana production is constrained by several factors viz., erratic weather condition, unavailability of quality planting material, assured source of irrigation water, scarce supply of fertilizers and infestation of pests and diseases. Adoption of improved technologies coupled with high density planting, fertigation and growing tissue culture plants the yield of banana could be enhanced.

The insect pests, mite, plant parasitic nematodes and diseases are important biotic constraints against successful cultivation of banana. More than 151 nematode species of 51 genera have been documented worldwide on *Musa* sp. (Gowen and Queneherve, 1990; Koshy and Gulsar Banu, 2000) and yield losses are estimated to be 19.7% amounting to US \$17.8 million (Sasser and Freckman, 1987). In India, 71 species of plant parasitic nematodes are known to be

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associated with banana (Krishnappa and Reddy, 1995; Koshy and Sosamma, 2001). Only seventeen species recorded from West Bengal (Mukherjee and Dasgupta, 1983). The major nematode pests of banana include burrowing nematode, *Radopholus similis*; root lesion nematodes, *Pratylenchus coffeae*, *P. goodeyi*, *P. brachyurus* and *P. reniformi*; spiral nematodes, *Helicotylenchus multicinctus* and *H. dihystra*; reniform nematode, *Rotylenchulus reniformis* and root knot nematodes, *Meloidogyne arenaria*, *M. incognita* and *M. javanica* (Gantait *et al.*, 2010; Wang and Hooks, 2009; Jackson *et al.*, 2003; Bridge, 2000; De Waele and Davide, 1998; McSorley, 1986; McSorley and Parrado, 1986; [http:// en.wikipedia.org](http://en.wikipedia.org)). The most economically important nematode parasite of banana in the world is migratory endoparasitic nematodes; they feed on the root cortex of banana plants. These are burrowing nematode, *R. Similis*; root lesion nematode, *P. coffeae* and *P. goodeyi*; and spiral nematode *Helicotylenchus multicinctus*. Their feeding results in dead root cells, or lesions. Sedentary endoparasitic nematodes that feed on banana include the root-knot (*Meloidogyne* spp.) and reniform (*Rotylenchulus reniformis*) nematodes. These nematodes penetrate banana roots, migrate and settle at a feeding site, and then start injecting growth-regulating substances into the root. This causes the surrounding cells, which now serve as their feeding site, to enlarge (Wang and Hooks, 2009). Infection by burrowing nematode causes toppling disease of banana (Jackson *et al.*, 2003). These diseases are the result of burrowing nematode infection destroying root tissue, leaving plants with little to no support or ability to take up water and translocate nutrients. Because of the damage that it causes to citrus, ornamentals and other crops, burrowing nematode is one of the most regulated nematode pests of plant worldwide (Hockland *et al.*, 2006). *Pratylenchus coffeae* (Zimmermann, 1898) Filipjev & Schuurmans Stekhovem, 1941, *P. goodeyi* (Sher & Allen, 1953) and other *Pratylenchus* species cause symptoms in banana similar to those of *R. similis*. The host range of *Pratylenchus coffeae* includes over 250 plant species. The presence of *P. coffeae* and *P. goodeyi* in banana root lesions is generally highly correlated with infection by fungi such as *Fusarium oxysporum*, *F. redolens*, *F. sambucium*, *Nigrosporangium* and *Rhizoctonia solani*. *P. goodeyi* is also associated with bacterial wilt of *Ensete*. This disease, caused by *Xanthomonas campestris* is highly destructive as it kills plants of all growth stages. *P. goodeyi* may aggravate the disease

expression and play a role in the spread of the disease (Bridge *et al.*, 1997).

The present investigation was therefore undertaken to study the diversity and community of the phytonematodes and in particular the morphometric variability of the migratory endoparasites *viz.*, *Radopholus similis*, *Pratylenchus* spp., and *Helicotylenchus multicinctus* associated with banana plant.

## **MATERIALS AND METHODS**

### ***Survey and collection of sample***

To study the nematode population, sixty seven soil and root samples (200 cm<sup>3</sup>) were collected from the banana fields of Nadia and North 24 Parganas district of West Bengal, and Thiruvananthapuram district of Kerala, India during 2013-2014. Survey was conducted at Mondouri banana orchard under AICRP on fruits, BCKV and Uttar Rajapur areas of Haringhata block of Nadia (West Bengal), Mena areas of Habra-II block of North 24-Parganas (West Bengal) and Vellayani areas of Thiruvananthapuram, Kerala. Soil samples from the mentioned crop field were collected from a depth of 20cm with the help of tubular soil sampler from the rhizosphere of the crop. Each composite sample was representative of ten such sub-samples at each site. The samples were kept in a polythene bag, labelled and brought to the laboratory for storing in refrigerator at 5°C for further extraction and processing of the nematode specimens.

### ***Extraction of nematodes***

The nematodes were extracted from 200 cm<sup>3</sup> of composite soil sample by Cobb's decanting and sieving method (Cobb, 1918) followed by modified Baermann technique (Christie and Perry, 1951). Taking 20g of pencil thickness banana feeder roots wash them gently under running tap water, cut into small pieces of 2-3 cm, split them longitudinally and keep on the double layer facial tissue paper and follow the modified Baermann process (Christie and Perry, 1951). Keep this assembly undisturbed for 48 hours and collect nematode suspension to observe under microscope. The root samples were also examined directly under stereoscopic microscope (Carl Zeiss-Stemi 2000C) after differential staining by NaOCl - acid fuchsin method (Byrd *et al.*, 1983).

### ***Killing and fixing of nematodes***

The nematode specimens were killed by hot-water-bath method. The concentrated nematode

suspension was plunged into a beaker containing hot water at about 60-65°C and shaken the suspension continuously inside the beaker for about 2-3 minutes. Then the nematodes were killed and suspension of nematode was kept out of hot water to bring the suspension at room temperature. Killed nematodes were then fixed in 4:1 formalin glacial acetic acid and kept separately in the labelled vial for further study.

#### **Estimation of nematode population**

Vial containing fixed nematode suspension was taken in a measuring cylinder to measure the total volume of nematode suspension obtained from 200cc of soil. Nematodes remained in the fixed suspension were counted under stereoscopic binocular microscope (Carl Zeiss-Stemi 2000C) using multi-chambered counting dish. The nematode suspension from each location was observed and mean of three aliquot was taken for calculating the population density per 200cc of soil. Root population of sedentary nematode endoparasites was encountered directly under stereoscopic binocular microscope from 2g of stained sample.

#### **Processing of nematodes**

For morphological and morphometric studies the fixed nematode specimens from each population were processed by glycerol-ethanol method (Seinhorst, 1959).

#### **Mounting of nematodes**

The processed specimens were permanently mounted in pure anhydrous glycerol. A small drop of glycerol was placed on the centre of clean glass slide (Borosil make) measuring 76 mm × 26 mm × 1.25 mm. About 8-10 processed nematodes specimens were picked up and placed in the centre of glycerol drop so that their head oriented at same direction, making sure that they were resting on surface of the glass slide and not floating on surface of the drop. Microscopic cover glass of 18 mm was placed over the specimen and sealed by the paraffin wax-ring method (De Maeseneer and D'Herde, 1963).

#### **Observation on morphological and morphometric variation**

Important morphological and morphometric features of taxonomic significance have been studied in detail for each population of *Radopholus similis*, *Pratylenchus* spp., and *Helicotylenchus multicinctus* obtained from different banana growing areas of Nadia and North 24-Parganas district of West Bengal

and Thiruvananthapuram district of Kerala, India. Several populations of mentioned nematode species of banana were examined for morphometric characterization for the generation of information on inter-specific and intra-specific variations. The following set of parameters used to characterize nematode species were developed initially by de Man (1880) and added to, modified and amended by Cobb (1914), Thorne (1949) and others. This is often known as the de Man Formula or the de Man Indices.

n = number of specimens on which measurements are taken

L = total body length

Tail = portion of body from anus or cloaca to posterior terminus

#### **Ratios**

a = body length / greatest body width

b = body length / distance from anterior (head) to basal bulb

c = body length / tail length

c' = tail length / anal body width (abw)

#### **Percentage**

m = length of conus / length of stylet) x 100

o = distance of dorsal esophageal gland opening from sylet knob / stylet length) x 100

P = (Distance of phasmid from anus / tail length) x 100

V = (Distance of vulva from anterior / total body length) x 100

T = (Distance from cloaca to anterior part of testis / total body length) x 100

The measurement of all the specimens was taken under compound microscope (Olympus BX-51) with the help of ocular micrometer as well as with the image analyzing device ProgRes CT5 of Jenoptic version 2.80. All the drawings were made with the help of drawing tube of compound binocular microscope (Olympus BX-51).

#### **Statistical analysis**

To study the nematode diversity frequency (absolute and relative), density (absolute and relative), and prominence value (absolute and relative) of major rhizospheric nematodes of banana were determined according to the formulae given by Norton (1978).

## RESULTS AND DISCUSSION

### **Diversity of rhizospheric nematodes associated with banana, *Musa* sp.**

Diversity of rhizospheric nematode associated with banana from the surveyed areas of West Bengal and at Vellayani, Kerala revealed prevalence of 17 species of phytoparasitic nematodes, 3 species of predatory nematodes and quite a good number of saprozoic nematodes (Table 1).

### **Rhizospheric nematode population of banana in West Bengal**

Survey on banana was conducted at two areas viz., Moundouri and Uttar Rajapur of Haringhata block of Nadia district and Mena area of Habra –II block of North 24-Parganas district. At Moundouri root knot nematodes, *Meloidogyne* spp. and reniform nematode, *Rotylenchulus reniformis* were found as most predominant endoparasitic nematode of banana. The migratory endoparasite *Pratylenchus* spp. (lesion nematode) was also recorded from the said area. Here, population of *Meloidogyne* spp. and *Rotylenchulus reniformis* varied from 51-5664 and 0-2118 per 200 cm<sup>3</sup> of soil, respectively (Table 2).

Population of *Pratylenchus* spp. ranged from 0-187 per 200 cm<sup>3</sup> of soil. *Hoplolaimus*, *Helicotylenchus*, *Tylenchorhynchus*, *Criconemoides*, *Aphilenchus* and *Xiphinema* were also recorded as ectoparasitic nematode fauna from the banana rhizosphere on that locality. Studying eight soil and root samples from Uttar Rajapur root knot nematode, reniform nematode and lesion nematode populations were found predominant. Among these, reniform nematode population was found most numerous, ranging from 377-616 per 200 cm<sup>3</sup> of soil (Table 2). At Mena of Habra-II of North 24- Parganas eight samples from banana were studied and found *Meloidogyne* spp. and *Rotylenchulus reniformis* as most abundant endoparasites. All the samples from surveyed areas represented huge numbers of saprozoic nematodes. Predatory nematodes viz., *Mylonchulus* sp., *Iotonchus* sp. and *Ironus* sp. were also encountered from both the districts of West Bengal (Table 2).

### **Rhizospheric nematode population of banana in Kerala**

Altogether eight soil and root samples from Vellayani, Thiruvananthapuram, Kerala were collected and studied for the occurrence and distribution of rhizospheric nematode population of banana. It was found that seven phytonematodes were

associated with the banana crop. Of which, *Radopholus similis*, *Helicotylenchus multicinctus*, and *Pratylenchus coffeae* were recorded as abundant migratory nematode endoparasites. *H. multicinctus* was found most numerous ranging from 764-3840 per 200 cm<sup>3</sup> of soil (Table 3).

### **Community study of root endoparasitic nematodes of banana at Moundouri, Haringhata, Nadia**

Community study of root inhabiting endoparasitic nematodes of banana revealed presence of seven species of nematodes viz., *M. incognita*, *M. javanica*, *R. reniformis*, *Pratylenchus coffeae* and *P. crenatus* (Table 4). Among them *Meloidogyne* spp. were most dominant over others considering frequency, density and prominence value. Two species of *Pratylenchus* viz., *P. coffeae* and *P. crenatus* had been recorded as migratory nematode endoparasite of banana from this locality.

### **Community study of root inhabiting endoparasitic nematodes of banana at Uttar Rajapur, Haringhata, Nadia**

Community study of root inhabiting endoparasitic nematodes of banana revealed presence of four species of nematodes viz., *R. reniformis*, *M. incognita*, *Pratylenchus coffeae* and *P. crenatus* (Table 5). Among them *R. reniformis* was most dominant over others considering frequency, density and prominence value. Two species of *Pratylenchus* viz., *P. coffeae* and *P. crenatus* had been recorded as migratory nematode endoparasite from this locality.

### **Community study of root inhabiting endoparasitic nematodes of banana at 24 Parganas, West Bengal**

Three endoparasitic nematodes species viz., *M. incognita*, *R. reniformis* and *Pratylenchus coffeae* were present in banana at Mena, Habra-II, 24 Parganas (Table 6). Among them *M. incognita* was most predominant over others considering relative frequency, density and prominence value. Here, *P. coffeae* was more frequent over *R. reniformis*.

### **Community study of root inhabiting endoparasitic nematodes of banana in Kerala**

Four endoparasitic nematodes species viz., *Helicotylenchus multicinctus*, *Rotylenchulus reniformis*, *Radopholus similis* and *Pratylenchus coffeae*, were present with banana here (Table 7). Except *Pratylenchus coffeae* all endoparasites had cent per cent frequency of occurrence.

Predominance of plant parasitic nematodes associated with banana root in two districts of West

Table 1: Diversity of rhizospheric nematodes associated with banana, *Musa* sp. in selected areas of West Bengal and at Vellayani, Kerala

Trophic Guild	Nematode Species	West Bengal		Kerala
		Nadia	North 24-Parganas	
<b>Plant Parasitic</b>	1. <i>Meloidogyne incognita</i>	+	+	-
	2. <i>Meloidogyne javanica</i>	+	-	-
	3. <i>Rotylenchulus reniformis</i>	+	+	+
	4. <i>Pratylenchus coffeae</i>	+	+	+
	5. <i>Pratylenchus crenatus</i>	+	-	-
	6. <i>Helicotylenchus multicinctus</i>	-	-	+
	7. <i>Helicotylenchus dihystra</i>	+	+	-
	8. <i>Helicotylenchus</i> sp.	+	+	-
	9. <i>Radopholus similis</i>	-	-	+
	10. <i>Hoplolaimus indicus</i>	+	+	-
	11. <i>Scutellonema</i> sp.	-	-	+
	12. <i>Tylenchorhynchus mashhoodi</i>	+	+	-
	13. <i>Tylenchorhynchus</i> sp.	-	-	+
	14. <i>Criconemoides onoensis</i>	+	-	+
	15. <i>Aphelenchus aveane</i>	+	-	-
	16. <i>Xiphinema insigne</i>	+	-	-
	17. <i>Longidorus</i> sp.	+	-	-
<b>Predatory</b>	1. <i>Mylonchulus</i> sp.	+	+	+
	2. <i>Iotonchus</i> sp.	+	-	-
	3. <i>Ironus</i> sp.	+	-	-
<b>Saprophagous</b>	1. <i>Acrobeles</i> sp.	+	-	-
	2. <i>Mesorhabditis</i> sp.	+	+	-
	3. <i>Amphidellus</i> sp.	-	+	-

**Table 2: Rhizospheric nematode population of banana in West Bengal**

Block	Locality/ No. of sample	Population per 200 cm <sup>3</sup> of soil +20g of root										
		Mel	Hoplo	Heli	Tyl	Roty	Crico	Praty	Aph	Xiph	Pred	Sapro
Haringhata (Nadia)	Mondouri / 43	578 (51-5664)	46 (0-696)	57 (0-172)	23 (0-234)	239 (0-2118)	4 (0-28)	35 (0-187)	14 (0-84)	4 (0-47)	13 (0-65)	437 (46-924)
	Uttar Rajapur / 8	396 (88-704)	197 (87-306)	—	1670 (319-3020)	497 (377-616)	—	81 (30-132)	—	—	15 (0-29)	695 (320-1069)
Habra -II (North 24-Parganas)	Mena/8	229 (164-294)	21 (0-42)	26 (21-30)	11 (0-21)	32 (0-63)	—	—	—	—	17 (0-35)	558 (94-1022)

Note: Mel- *Meloidogyne incognita* & *M. javanica*, Hoplo- *Hoplolaimus indicus*, Heli- *Helicotylenchus dihystra* & *Helicotylenchus* sp. Tyl- *Tylenchorhynchus mashhoodi*, Roty- *Rotylenchulus reniformis*, Crico- *Criconemoides onoenis*, Praty- *Pratylenchus coffeae*, P. *crenatus*, Aph- *Aphelenchus avenae*, Xiph- *Xiphinema insigne*, Pred- Predatory nematodes *Mylonchulus* sp., *Iotonchus* sp. & *Ironus* sp., Sapro- Saprozoic nematode viz., free living dorylaimids, rhabditids etc.

**Table 3: Rhizospheric nematode population of banana at Vellayani, Thiruvananthapuram, Kerala**

Locality	No. of sample	Population per 200 cm <sup>3</sup> of soil +20g of root									
		Rado	Heli	Tyl	Roty	Crico	Praty	Scut	Pred	Sapro	
Vellayani	8	529 (193-1049)	2240 (764-3840)	78 (0-225)	1754 (214-3790)	18 (0-50)	217 (0-510)	1.5 (0-6)	11 (0-42)	262 (64-550)	

Note: Rado- *Radopholus similis*, Heli- *Helicotylenchus multincinctus*, Tyl- *Tylenchorhynchus* sp., Roty- *Rotylenchulus reniformis*, Crico- *Criconemoides* sp., Praty- *Pratylenchus coffeae*, Scut- *Scutellonema* sp., Pred- Predatory nematode *Mylonchulus* sp, Sapro- Saprozoic nematode viz., free living dorylaimids, rhabditids etc.

Bengal and one area of Kerala is presented in descending order of their relative prominence value (Table 4-7). With reference to the density (absolute and relative) and prominence value *Meloidogyne incognita* and *M. javanica* ranked first as root inhabiting nematode endoparasites of banana crop followed by *Rotylenchulus reniformis*, *Pratylenchus coffeae* and *P. crenatus* at Mondouri of Haringhata block of Nadia district (Table 4). Similar trend was noticed at Mena of Habra-II block of North 24-Parganas (Table 6). While at Uttar Rajapur area of the Haringhata block of Nadia district *Rotylenchulus reniformis* ranked first as root inhabiting nematode endoparasites of banana crop followed by *Meloidogyne incognita*, *Pratylenchus coffeae* and *P. crenatus* (Table 5). It was pertinent to workout prominence value for determining the parasitic importance of nematodes in a diverse community (Beals, 1960). The prominence value calculated did not seem to clearly reflect the pathogenic significance of the members of the community. It was reported that *Rotylenchulus reniformis* is a most frequently occurring nematode parasite of crops in West Bengal at present (Mukhopadhyay and Roy, 2006; Roy et al., 2007; Roy and Mukhopadhyay, 2011). Along with root-knot and reniform nematodes, root lesion nematode, *P. coffeae* and *P. crenatus* were recorded as a parasitic migratory nematode pest of banana in West Bengal.

Four endoparasitic nematodes species viz., *Helicotylenchus multicinctus*, *Rotylenchulus reniformis*, *Radopholus similis* and *Pratylenchus coffeae*, were present with banana at Vellaynai, Kerala (Table 7). Except *Pratylenchus coffeae* all endoparasites had cent per cent frequency of occurrence here. This seems to be at par with the observation of Tiwari et al. (2000).

Khan (1999) after survey in West Bengal reported widespread occurrence of *Radopholus similis* in banana with a frequency of 42.8%. In this study, none of the surveyed banana growing areas of West Bengal showed the presence of *R. similis* and *H. multicinctus*. Those areas are free from the mentioned nematode pests of banana. Besides phytonematodes, predatory nematodes viz., *Mylonchulus* sp., *Iotonchus* sp. and *Ironus* sp. with a frequency ranging from 25-27% were also encountered from both the districts of West Bengal under survey. However, saprozoic nematodes comprising of free living dorylaimids and rhabditids were also observed as very prominent member among soil nematode communities of banana.

### **Diversity of nematode fauna on different banana cultivars**

Observation of different banana cultivars (22) for the diversity of nematode fauna indicated that *Musa* (AAB) cv. Rajapuri, cooking type cv. Booditha Bontha Bothisa, Patcha Bontha Bothisa and Cuba-03 (ABB) supported huge population of root knot nematodes, *Meloidogyne* spp. ranging from 594-5664 J2 per 200 cm<sup>3</sup> of soil (Table 8). However, varieties like, Pisang Bale, Ney Poovan (AB) and CO-I (AAB) supported very low population of root knot nematodes, *Meloidogyne* spp. ranging from 51-76 J2 per 200 cm<sup>3</sup> of soil (Table 8). The banana cultivars viz., Red banana (AAA), Raskadali (AB) and Sannachen Kadali (AA) recorded *R. reniformis* population ranging from 1118-2118 per 200cc of soil. These variations in nematode population in the rhizosphere of banana cultivars are attributed to the varietal effects on nematode survival and multiplication. Varieties or cultivars usually respond differently to nematode parasite presumably due to their constituent chemicals or toxic principles. Positive correlations occur between the concentration of phenolics, chlorogenic acid, total sugars of tomato and banana cultivars and resistance response to root-knot and burrowing lesion nematodes, respectively (Devrajan and Rajendran, 2002). Root exudates of plants also play an important role for attracting and repelling nematodes (Huang, 1985). Therefore, abundance of nematode fauna in a crop is greatly influenced by the chemical nature of crop rhizosphere.

*P. coffeae* was also found infecting Sannachen, Kadali (AA), Srimanti (AAA), Patcha Bontha Bothisa (CT), Budu Bale, Matti (AA) and Pisang Bale cultivars of banana. *P. coffeae* produced lesions on feeder roots leading to rotting and final decay of the root system. The low nematode population in Ney Poovan (AB), Karpooravelli (ABB), Booditha Bontha Bothisa (CT), Gross Michel (AAA), Red banana (AAA) could be presumably due to a suppressive effect or resistance response on *P. coffeae*. The ratio between plant feeding nematode and non plant feeding nematode revealed less than 1 in Raskadali (AB), Matta Poovan (AAB), Sannachen Kadali (AA), Rajapuri (AAB), Cuba-03 (ABB/CT), Amritpani (AAB), Booditha Bontha Bothisa (CT), Patcha Bontha Bothisa (CT), Nendran (AAB), Mahalaxmi (AAA), Budu Bale, Red banana (AAA) and Pisang Bale. This indicated that these cultivars are highly susceptible to phytophagous nematode, because here phytophagous nematodes are dominating over

saprozoic and predatory nematodes and able to build up population in banana plants (Table 8).

None of the cultivars of banana examined here showed the existence of *R. similis* and *H. multicinctus*. As almost all the selected cultivars were procured from southern part of India, there may be possibility of their existence in the banana plantation at Mondouri under AICRP on Fruits, BCKV. Present study revealed non existence of those two notorious migratory endoparasitic nematode pests of banana here. Studies on absolute biomass and importance values of major phytonematodes revealed that *Meloidogyne incognita*

was more important in banana. In West Bengal, *Rotylenchulus reniformis* and *Hoplolaimus indicus* were observed as second and third important phytonematodes of banana, respectively (Table 9).

#### Identification of *Pratylenchus* populations associated with banana in West Bengal

All the populations of *Pratylenchus* Filipjev, 1936 studied were identified either as *P. crenatus* Loof, 1960 or *P. coffeae* (Zimmermann, 1898) Filipjev & Stekhoven, 1941. *P. crenatus* had been recorded from banana plantations at Mondouri and Uttar Rajapur areas of Haringhata block of Nadia district.

**Table 4: Community study of root inhabiting endoparasitic nematodes of banana at Mondouri, Haringhata, Nadia (Based on 21 samples)**

Nematode species	Absolute frequency (%)	Relative frequency (%)	Absolute density	Relative density (%)	PV	RPV (%)
<i>Meloidogyne incognita</i> <i>M. javanica</i>	90.5	70.4	447.5	93.0	425.6	96.2
<i>Rotylenchulus reniformis</i>	28.6	22.2	29.1	6.1	15.6	3.5
<i>Pratylenchus coffeae</i> <i>P. crenatus</i>	9.5	7.4	4.3	0.9	1.3	0.3

Note: PV=Prominence Value; RPV=Relative Prominence value, PV=Absolute density,

**Table 5: Community study of root inhabiting endoparasitic nematodes of banana at Uttar Rajapur, Haringhata, Nadia (Based on 4 samples)**

Nematode species	Absolute frequency (%)	Relative frequency (%)	Absolute density	Relative density (%)	PV	RPV (%)
<i>Rotylenchulus reniformis</i>	100.0	44.4	418.8	53.9	418.8	58.3
<i>Meloidogyne incognita</i>	75.0	33.3	297.0	38.3	257.2	35.8
<i>Pratylenchus coffeae</i> <i>P. crenatus</i>	50.0	22.2	60.5	7.8	42.8	6.0

**Table 6: Community study of root inhabiting endoparasitic nematodes of banana at Mena, Habra-II, 24 Parganas (N) (Based on 4 samples)**

Nematode species	Absolute frequency (%)	Relative frequency (%)	Absolute density	Relative density (%)	PV	RPV (%)
<i>Meloidogyne incognita</i>	75.0	50.0	212.0	97.2	183.6	98.1
<i>Rotylenchulus reniformis</i>	25.0	16.7	3.3	1.5	1.6	0.9
<i>Pratylenchus coffeae</i>	50.0	33.3	2.8	1.3	1.9	1.0

**Table 7: Community study of root inhabiting endoparasitic nematodes of banana at Vellayni, Thiruvananthapuram, Kerala (Based on 4 samples)**

Nematode species	Absolute frequency (%)	Relative frequency (%)	Absolute density	Relative density (%)	PV	RPV (%)
<i>Helicotylenchus multicinctus</i>	100.0	26.7	1262.5	48.4	1262.5	48.7
<i>Rotylenchulus reniformis</i>	100.0	26.7	905.5	34.7	905.5	34.9
<i>Radoopholus similis</i>	100.0	26.7	312.0	12.0	312.0	12.0
<i>Pratylenchus coffeae</i>	75.0	20.0	128.3	4.9	111.1	4.3



Table 8: Diversity of nematode fauna on different cultivars of banana

Sl. No.	Variety	Population per 200 cm <sup>3</sup> of soil + 20g of root												PFN:NPF		
		Mel	Hop	Heli	Tyl	Roty	Crico	Praty	Aph	Xiph	Pred	Sapro	PFN	NPF	PFN	NPF
1.	Matti (AA)	414	18	144	36	—	—	54	—	—	—	—	773	666	773	1.2
2.	Ney Poovan (AB)	65	65	86	—	85	—	22	—	22	65	—	924	343	988	2.9
3.	Tatilla Kunnan (AB)	322	69	115	—	184	—	—	46	—	—	—	851	736	851	1.2
4.	Raskadali (AB)	98	—	172	26	1204	—	—	—	—	26	—	470	1500	496	0.3
5.	Matta Poovan (AAB)	445	—	118	71	—	27	—	—	—	47	—	347	659	394	0.6
6.	Poovan (AAB)	135	—	46	—	69	23	—	—	—	—	—	46	273	46	0.2
7.	Sannachen Kadali (AA)	126	—	130	234	1118	—	156	52	—	—	—	857	1816	857	0.5
8.	Rajapuri (AAB)	5664	696	169	—	165	—	—	—	—	—	—	639	6693	639	0.1
9.	Srimanti (AAA)	218	—	—	50	—	—	187	—	—	—	—	349	455	349	0.8
10.	Cuba-03 (ABB/CT)	594	—	—	—	—	—	—	—	—	—	—	90	594	90	0.2
11.	Karpooravelli (ABB)	94	118	—	7	—	—	24	—	—	47	—	494	242	541	2.2
12.	Amritpani (AAB)	1034	6	—	2	—	—	—	—	—	—	—	85	1042	85	0.1
13.	Booditha Bontha Bothisa (CT)	1040	—	—	25	—	28	28	48	—	25	—	273	1168	298	0.3
14.	Patcha Bontha Bothisa (CT)	616	—	—	—	210	—	112	—	—	—	—	735	938	735	0.8
15.	Nendran (AAB)	143	—	—	29	—	—	—	—	—	—	—	86	171	86	0.5
16.	CO-I (AAB)	76	—	25	—	26	—	—	25	—	—	—	527	151	527	3.5
17.	Mahalaxmi (AAA)	407	14	27	—	14	—	—	—	—	—	—	382	461	382	0.8
18.	Karpurachakkarkeli (AAB)	373	23	23	—	65	—	—	—	23	—	—	548	507	548	1.1
19.	Budu Bale	361	—	49	21	—	—	—	84	—	—	—	571	515	571	1.1
20.	Gross Michel (AAA)	398	—	99	—	—	—	24	—	47	—	—	296	567	296	0.5
21.	Red banana (AAA)	58	—	55	—	2118	—	28	—	—	55	—	193	2258	248	0.1
22.	Pisang Bale	51	—	—	—	—	—	128	51	—	26	—	77	230	102	0.4

Note: Mel- *Meloidogyne incognita* & *M. javanica*, Hop- *Hoplolaimus indicus*, Heli- *Helicotylenchus dihystra* & *Helicotylenchus* sp., Tyl- *Tylenchorhynchus mashhoodi*, Roy- *Roylenchulus reniformis*, Crico- *Criconemoides onoensis*, Praty- *Pratylenchus coffeae*, Aph- *Aphelenchus avenae*, Xiph- *Xiphinema insigne*, Pred- Predatory nematodes *Myionchulus* sp., *Iotonchus* sp. & *Ironus* sp., Sapro- Saprozoic nematode viz., free living dorylaimids, rhabditids etc; # PFN- plant feeding nematode, NPF- non plant feeding nematode inclusive of free living and predatory.

**Table 9: Absolute biomass and importance values of selected phytonematodes of banana**

Nematode species	Absolute biomass (n)	Biomass nematode species <sup>-1</sup> (µg)	Total biomass species <sup>-1</sup> (µg)	Relative biomass (%)	Relative Frequency (%)	Relative Density (%)	Importance value (IV)
<i>Meloidogyne incognita</i>	579	0.052	30.11	38.8	25.3	57.9	122.0
<i>Rotylenchulus reniformis</i>	239	0.049	11.80	15.2	12.6	23.9	51.7
<i>Hoplolaimus indicus</i>	46	0.365	16.77	21.6	9.2	4.6	35.4
<i>Helicotylenchus dihystra</i>	57	0.118	6.71	8.7	16.1	5.7	30.5
<i>Pratylenchus coffeae</i>	35	0.119	4.17	5.4	11.5	3.5	20.4
<i>Tylenchorhynchus mashhoodi</i>	23	0.168	3.85	5.0	11.5	2.3	18.8
<i>Aphelenchus avenae</i>	14	0.079	1.09	1.4	6.9	1.4	9.7
<i>Xiphinema insigne</i>	4	0.416	1.73	2.2	3.4	0.4	6.0
<i>Criconemoides onoensis</i>	4	0.333	1.33	1.7	3.4	0.4	5.5

Note: Biomass (G) =  $\{(a^2b)/16 \times 100,000\}$  where, a- greatest body width (µm), b- body length (µm), 16- empirical value (Andrassy, 1956). Importance value = relative frequency + relative density + relative biomass (Norton, 1978).

#### Morphometric variability of *P. crenatus*

Comparable morphometric data of *P. crenatus* has been presented in the table-10. The body length of *P. crenatus* varied from 520-560µm in females and 440-520µm in males. Frederick and Tarjan (1989) presented in a compendium that, the body length of female ranged from 430-570µm. The lip region was continuous, stylet knob was round and set off from shaft, tail sub-hemispherical to clavate with annulated terminus. Besides that, a (23-27), c (16-21), v% (79-83), stylet (17µm), lip annules (3), and number of tail annules (20-26) are found similar to the findings of Frederick and Tarjan, 1989; Loof, 1960; Ryss, 1988; Karssen and Brinkman, 2003. Present morphometric observations confirmed the studied population of *Pratylenchus* as *Pratylenchus crenatus* Loof, 1960. There was no report on the occurrence of the *Pratylenchus crenatus* in West Bengal. Probably this is the first record of their existence on banana in West Bengal.

#### Morphometric variability of *P. coffeae*

The observation on morphometric variability of *P. coffeae* has been presented in the table-11. The body length of *P. coffeae* varied from 455-675µm in females and 420-520µm in males. According to Frederick and Tarjan (1989) the body length of female ranged from 460-630µm. The lip region was continuous, stylet knob was round and set off from shaft, tail sub-hemispherical with smooth terminus. While, a (23-29), c (19-21), v% (80-81), stylet (17-18µm), lip annules (2), and number of tail annuli (16-24) are found similar to the findings of Bajaj and Bhatti, 1984; Frederick and Tarjan, 1989. Present morphometric

observations confirmed the studied population of *Pratylenchus* as *Pratylenchus coffeae* (Zimmermann, 1898) Filipjev & Stekhoven, 1941. The occurrence of the *Pratylenchus coffeae* on banana in West Bengal has been reported by several earlier workers (Khan, 1999).

#### Identification of *Helicotylenchus* populations associated with banana in Kerala

All the endoparasitic root inhabiting populations of *Helicotylenchus* Steiner, 1945 from Vellayani, Thiruvananthapuram, Kerala were identified as *H. multicinctus* (Cobb, 1893) Golden, 1956. The comparative morphometric diagnostic data of the females and males of *H. multicinctus* is presented in table- 12. All the comparable morphometric observations are found similar to the findings of Siddiqi, 1973 and Ganguly *et al.*, 2013.

#### Morphometric variability of *H. multicinctus*

The observation on morphometric variability of *Helicotylenchus multicinctus* has been presented in the table-12. **Adult female-** Heat relaxed specimens were open-C shaped, annules distinct, lateral field with four incisures, not areolated. Lip region hemispherical, slightly offset with 4-5 annuli, cephalic framework heavily sclerotized. Stylet 22-25mm long with 4.5-5mm wide stylet knob appearing anteriorly concave. Oesophagus with rounded meatcorpus and glandular basal bulb overlap intestine ventrally. Dorsal oesophageal gland orifice at 34% of total spear length. Excretory pore at 93% from anterior end i.e. at the level of median bulb. Hemizonid 0-3 body annules anterior to excretory pore. Female gonad composed of

Table 10: Comparable morphological characteristics of *Pratylenchus crenatus* in  $\mu\text{m}$ 

Morphological features	Present findings			After Karssen and Brinkman, 2003
	Female (n=3)	Male (n=3)	Female	
L	520-560	440-520	320-600 /430-560	430
a	23-27	25-29	19.7-29.9/18-27	28.4
b	4.3-5.5		4.9-7.9/5.1-6.9	
b'	3.7-4.5		—/4.9-5.5	
c	16-21	22.0-24.5	16.4-26.8/16-27	21.4
Stylet length	17	15.0	14-18/16-17.5	16
Vulva % (v)	79-83		78-86/79-84	
Spicule	—	15-16	—	17
Gubernaculum	—	5.0	—	4.5
Lip annulations	3		3	
Lateral lines	6, outer ones strongly crenate and inner ones are interrupted		6, outer ones strongly crenate and inner ones are interrupted	
Tail shape and terminus	sub-hemispherical to clavate, annulated		sub-hemispherical, annulated coarsely and distinctly	
No. of tail annuli	20-26		20-24	

Note: n- number of specimen observed; value in parenthesis indicates mean value

Table 11: Comparable morphological characteristics of *Pratylenchus coffeae* in  $\mu\text{m}$

Morphological features	Present findings		Female (after Frederick and Tarjan, 1989)	Female (after Bajaj and Bhatti, 1984)
	Female (n=4)	Male (n=6)		
L	455-675 (559)	420-520 (469)	460-630 (580)	550-700 (620)
a	23-29 (26)	21-29 (26)	21-30 (25)	23-38 (28)
b	5-5.6 (5.2)	4.1-6.1 (5.2)		6.1-7.8 (7.3)
b'	4.4-5.0 (4.6)	3.6-4.1 (3.9)		4.0-5.5 (4.8)
c	19-21 (20)	21-26 (22)	17-27 (21)	17-26 (20)
Stylet length	17-18 (17.5)	15-16 (15)	15-17 (16)	14-17 (15)
Vulva % (v)	80-81 (81)		76-82 (79)	79-84 (81)
Spicule		15-18		
Gubernaculum		3-5		
Lip annulations	2	2	2	
Tail shape and terminus	sub- hemispherical, terminus smooth		hemispherical to bluntly pointed, terminus smooth	
No. of tail annuli	16-24		17-24	

Note: n- number of specimen observed; value in parenthesis indicates mean value

Table 12: Comparable morphological characteristics of *Helicotylenchus multinctus* in  $\mu\text{m}$ 

Morphological features	Present findings		After Siddiqi, 1973/ Ganguly et al., 2003	
	Female (n=22)	Male (n=13)		Female
Habitus	open c shaped	open c shaped	open c shaped	open c shaped
L	475-740 (596)	440-685 (527)	470-680 (540)/400-673	460-680 (540)
a	24-37 (29)	25-34 (31)	24-30	23.8-28.5
b	4.8-7.6 (5.9)	4.4-6.2 (5.4)	4.7-5.4	
b'	4.0-6.2 (4.8)	3.8-5.0 (4.4)	3.4-4.1	
c	35-58 (50)	37-47 (41)	/31-66	
Stylet length	22-25 (23)	21-22 (21)	21-24/20-28	20-24
Shape of stylet knob	anteriorly concave	anteriorly concave	anteriorly concave/flattened	
Width of stylet knob	4.5-5 (5)	4-5 (4.6)	5-6	
DEGO % (o)	34	31	26-35	
Vulva % (v)	62-77 (70)		/61-76	
Lip region	hemispherical	hemispherical	hemispherical	
Lip annulations	4-5 (4.5)	3-5 (4.3)	3-5	
Hemizonid	0-3 body annules anterior to excretory pore	0-3 body annules anterior to excretory pore		
Lateral lines	4, not areolated	4, not areolated	4, not areolated	4, not areolated
Tail shape and terminus	sub-hemispherical, annulated & conoid	short-elongate conoid	sub-hemispherical, annulated & conoid	
No. of tail annules	9-12	9-13	6-13	
Phasmid	Pore like, 1-7 annules anterior to annus		Pore like, 1-6/2-9 annules anterior to annus	

Table 13: Comparable morphological characteristics of *Radopholus similis* in  $\mu\text{m}$

Morphological features	Present findings		After Williams & Siddiqi, 1973; Esser <i>et al.</i> , 1984; Elbadri <i>et al.</i> , 1999	
	Female (n=12)	Male (n=11)	Female	Male
Body length	500-660 (550.4)	440-685 (546.4)	510-820 (655)	450-720 (590)
Stylet length	14-18 (15.8)		16-23 (18.5)	10-16 (13.3)
Excretory pore from anterior end	63-84 (74.3)	61-96 (81.5)	59-115 (88)	65-104 (86)
Anal body width	14-23 (18.4)	10-16 (11.9)	13-24 (17.7)	11-20 (13.9)
Spicule length		16-21 (19.2)		13-24 (19.3)
Gubernaculum length		7-11 (9.0)		7-14 (10.6)
Vulva %	56-59 (58)		50-67 (57)	
a	27-35 (32)	30-38 (34)	20-34 (27)	24-43 (33)
b	4-7 (5.2)	4-6 (5)	6-10 (7.9)	5-10 (7.6)
c	8-10 (9.4)	7-10 (8.8)	7-13 (9.4)	6-10 (8.3)
Lip region	offset	offset	offset	
Lip annulations	3-5	5-7	3-6	
Tail terminus	narrow	narrow	narrow	
Tail annulation	fine to moderate	fine to moderate	fine to smooth	

Note: n- number of specimen observed; value in parenthesis indicates mean value

two symmetrical ovaries, sometimes posterior one may be reduced. Spermatheca round, full of viable sperms. Vulva is depressed transverse slit on ventral aspect of the body. Tail short, sub-hemispherical, with terminus conoid in shape and annulated. Dorsal margin of tail was more curved than ventral, 9-12 annuli on ventral side. Phasmid pore like, 1-7 annuli anterior to anus. **Adult male-** Males are abundant, similar to females except for genital characters. Testis single, outstretched. Caudal alae crenate, enveloping tail. Spicule cephalated, gubernaculum simple.

#### **Identification of *Radopholus* populations associated with banana in Kerala**

All the root inhabiting endoparasitic nematode populations of *Radopholus* Thorne, 1949 from Vellayani, Thiruvananthapuram, Kerala were identified as *Radopholus similis* (Cobb, 1893) Thorne, 1949. The comparative morphometric diagnostic data of the females and males of *R. similis* has been presented in the table- 13.

#### **Morphometric variability of *R. similis***

*Radopholus similis* are small nematodes (less than 1 mm long) with elongate tail and marked sexual dimorphism in the anterior region (Fig. 10). The observation on morphometric variability of *R. similis* has been presented in table-13. **Male-**The knob or button-shaped head is set off by a constriction. Head annules varied from 5-7. Cephalic sclerotization, stylet, median bulb and pharyngeal gland lobe were weakly developed. Four lateral incisures were present. Inner incisures faint. Caudal alae pronounced, arising about one body width anterior to the spicule head and extending to about one to two body widths anterior to the tail tip. Gubernaculum with small titillae. Spicules dorsally arched. **Female-** Head hemispherical, rarely slightly set off. Three to six lip annules. Stylet well defined with rounded knobs. Four to six lateral incisures. Vulva post-equatorial (58%), sometimes with slight protuberant lips. Two functional and equally developed ovaries, each bearing a spheroid spermatheca often filled with sperm. Metacorpus well developed, elongate to ellipsoidal with distinct valve plate. Tail elongate-conoid with narrowly rounded terminus. All the comparable morphometric observations are found similar to the findings of Williams and Siddiqi, 1973; Esser *et al.*, 1984; Elbadri *et al.*, 1999.

In the present experiment community of the rhizospheric nematodes associated with banana crop, their identification and detailed morphometric

variability of major endoparasitic nematode pest of banana viz., burrowing nematode (*Radopholus similis*), root lesion nematode (*Pratylenchus coffeae*, *P. crenatus*), and spiral nematode (*Helicotylenchus multicinctus*), and root knot nematode (*Meloidogyne incognita*, *M. javanica*) had been studied. All the banana growing areas of the state need to be surveyed to get a detail idea about the extent of nematode infestation on the crop. Many plant parasitic nematode species like *Rotylenchulus reniformis*, *Tylenchorhynchus mashhoodi*, *Criconemoides onoensis*, *Hoplolaimus indicus*, *Helicotylenchus dihystra*, *Helicotylenchus* sp., *Scutellonema* sp. and *Xiphinema insigne* were found associated with the crop. Species level identification and morphometric variability (if any) of those parasitic nematode species need to be carried out in future. Importance value, index of similarity, richness of the species and diversity index among nematode species of different locality are need to be worked out in future to have an idea about the interaction among nematode community of banana crop. Studies on the pathogenicity of the parasitic nematodes species associated with banana crop and their eco-friendly management strategy need to be developed in future.

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