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Survey of Plant-Parasitic Nematodes Associated with Cucumber (*Cucumis sativus* L.) in Major Production Areas of Nigeria

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ABSTRACT
A survey to determine the diversity and relative abundance of phytonematodes associated with cucumber in selected South Western and North Central states of Nigeria was conducted in 2014 and 2015 using systematic sampling techniques. A total of 24 farms from each state and 144 farms from the surveyed areas in Nigeria were sampled. Nematodes were extracted and identified from soil and root samples from each farm at the nematology unit of International Institute of Tropical Agriculture (IITA), Ibadan. For each of the surveyed farms, the cropping practices employed were recorded. Percentage frequency of occurrence and percentage nematode population were calculated using standard formulae. Eleven plant-parasitic nematode genera: *Meloidogyne* spp, *Pratylenchus* spp., *Scutellonema* spp., *Helicotylenchus* spp., *Xiphinema* spp., *Aphelenchoides* spp., *Tylenchus* spp., *Criconemoides* spp., *Longidorus* spp. *Belonolaimus* spp. *Rotylenchus* spp. were encountered. The most widely distributed and abundant was *Meloidogyne* (45.1%) followed by *Helicotylenchus* (16.0%) > *Pratylenchus* (12.3%) > others (26.6%). *Meloidogyne* was more abundant in Lagos State (24.6%) > Kaduna (24.3%) > Plateau (15.6%) > others (35.5%). Most plant-parasitic nematodes identified in this survey can be anticipated to develop into major threats to cucumber and vegetable production and should be considered as serious potential pests of cucumber.

Keywords: Cucumber, Frequency of occurrence, *Meloidogyne* spp , phytonematodes , Relative abundance, Survey.

INTRODUCTION

Cucumber production in Nigeria is fast becoming popular since it is a useful ingredient in the preparation of salad. It has medicinal and therapeutic values, such as in the cure of hypertension and skin diseases. It is also a valuable source of potassium, sodium, magnesium, silicon, phosphorous, chlorine and fluorine (Rai and Yadav, 2005). World average cucumber fruit production in 2013 was 71 million MT (FAOSTAT, 2015). Pests and diseases constitute a major threat to production of cucumber in Nigeria. Plant-parasitic nematodes are common pests of vegetables globally (Sikora and Femandez, 2005; Baimey *et al*., 2009). Plant-parasitic nematodes feeding on vegetables can cause premature plant death, reduced vigor and an increased susceptibility

to other biotic or abiotic stresses such as fungi, bacteria, viruses, and drought (Baimey *et al*., 2009). Losses in the range 20-94% due to plant parasitic nematodes were recorded in Nigeria on cowpea (Olowe, 1978). Root-knot nematodes are a group of plant-parasitic nematodes that feed on roots resulting in the formation of irregular, knotty enlargement of the roots (Scurrah *et al*., 2005). Cucumber is a susceptible host of the root–knot nematodes, *Meloidogyne* spp. (Darekar and Bele, 1990). The first observation of root-knot infection was made on cucumber in 1850's (Mai, 1985). The size and shape of the knots or galls depend on the number of nematodes present in the roots, the species of the nematodes present and the plant species involved (Mitkowski and Abawi, 2003). Root-knot nematodes are amongst the most economically important nematode pests in Nigeria. They have a very wide host range (Adesiyan *et al*., 1990, Netscher and Sikora, 1990, Mitkowski and Abawi, 2003). This makes them very difficult to control because they can survive and reproduce on other host crops including weeds (Mitkowski and Abawi, 2003). Some of the reasons for their high rank as plant pests include their world-wide distribution, extensive host range, the debilitating nature of the diseases they cause and their role in many destructive diseases complexes (Mai, 1985, Mitkowski and Abawi, 2003). Other nematodes, such as sting nematodes (*Belonolaimus* spp.) occasionally cause some losses in cucumber production (Natarajan *et al*., 2006). Though these nematodes alone rarely cause death, they predispose the plants to infection by other pathogens, such as fungi and bacteria, which eventually lead to wilting of crops. Root-knot nematodes are hidden enemies of farmers (Sasser,1990) and are commonly encountered plant-parasitic nematodes in vegetables fields (Anwar *et al*., 2006). Above ground symptoms of the root-knot nematode infection include yellowing of leaves, stunting growth, smaller size of leaves, flowers and fruits. The underground symptoms include abnormal swellings of the roots (knot or root-galls), lesions, and rotting of roots (Agrios, 2005; Pajovic *et al*., 2007). The feeder roots also become fewer and this reduces the rate of absorption of soil water and nutrients (Abad *et al*., 2003; Anwar and Javid, 2010) leading to stunted growth of host plants and yield loss (Agrios, 2005; Ullah *et al*., 2011).

The root-knot nematode infection, with other fungal, bacterial and viral plant diseases, has been one of the major obstacles to successful cultivation of vegetables in many parts of the world (Kathy, 2000; Anwar *et al*., 2006). The average field crop damage by root-knot nematodes is estimated at 15% while damage in individual fields ranges from 60 % - 100% (Sasser and Carter, 1982). However, yield losses of vegetable crops, in particular due to *Meloidogyne* spp., ranged from 17 to 20% for *Solanum melongena* (Kathy, 2000), and over 80% for *tomato* (Sasser, 1979), 74-100% in pepper (Olabiyi and Oyedunmade, 2008). Several studies on the distribution/ occurrence of root-knot nematode species in vegetable crops carried out by several workers worldwide show that distribution/occurrence of *Meloidogyne* spp. vary significantly from location to location, and also from crop to crop (Anamika *et al*., 2011; Chaudhary *et al*., 2011) . So far, few studies have been conducted on the distribution of nematodes associated with cucumber. This survey was therefore carried out to provide information on the occurrence and diversity of plant-parasitic nematodes associated with cucumber.

MATERIALS AND METHODS

The survey was carried out in Lagos, Ogun, Osun, Oyo (South Western Nigeria), Kaduna (North West Nigeria) and Plateau (North Central Nigeria). The cropping patterns, crop

grown and irrigation system employed were recorded for each of the surveyed farm. Farm location coordinates and altitude were documented with GPS device. This selection was based on information of high seeds sales rate by a major local distributor (Agrictropic Nigeria Limited). Six farms per local Government were selected from four cucumber producing Local Government Areas. The locations were visited for the survey from October to March during 2013/14 and 2014/15 growing seasons. This gave a total of 24 farms from each State and a total of 144 farms surveyed. Soil samples of 1 kg consisting of 10 to 15 soil cores were taken along a zig-zag pattern, Samples was taken 0 to 30 cm deep with the aid of a soil auger. The cores were combined per farm to form a single composite sample. About 150 g of lateral roots were sampled from 10 individual plants randomly selected per farm. The soil and root samples from each farm were collected in plastic bags, bulked separately, properly labeled, and were taken to the Nematology Research Laboratory of International Institute of Tropical Agriculture (IITA), Ibadan for processing. The nematodes were extracted from the soil and root samples using the tray method described by Whitehead and Hemming (1961). The set-up consists of two sieves separated by a double-ply facial tissue placed on a collection tray. After thorough mixing, 250 ml of soil was poured on the upper sieve and water was added to the collection tray to field capacity. Also 10 g of roots were cut into 2 cm pieces and placed in the tray set-up. Vermiform nematodes from the soil and root samples were collected in beakers after 48 hours and counted under the dissecting microscope. Nematode identification was carried out at the Nematology Laboratory of International Institute of Tropical Agriculture (IITA), Ibadan with the use of plant-parasitic identification key (Lyon and Mai, 1975; Makete *et al*., 2012).

Statistical analyses

Microsoft Excel was used to calculate the percentage frequency of occurrence (i.e. frequency rating) and percentage nematode population for each State using the formulae below:

Percentage frequency of occurrence=

$$\frac{n}{N} X \frac{100}{1}$$

Where, n is the number of times an individual nematode occurred in all the samples and N is sample size.

Percentage nematode population =

$$\frac{In}{TN} X \frac{100}{1}$$

Where In is the individual nematode population in all the samples, while TN is the total population of all the nematodes extracted in all the samples.

Results

The plant-parasitic nematodes and frequency of occurrence associated with cucumber in Nigeria was found to vary from State to State (Table 1). Eleven genera of plant-parasitic nematodes were encountered in varying proportions in the States surveyed. *Meloidogyne* spp. was most encountered in Lagos State with population density of 2849 per 250 ml soil and the least encountered was recorded in Oyo State with population density of 1197/250 ml soil (Table *Helicotylenchus* spp. with percentage nematode population of 16.0 was most encountered in Kaduna State with population density of 1044 per 250 ml soil. *Pratylenchus* spp. was most encountered in the soil extracted from Lagos State with population density of 1234 per 250 ml soil and percentage population of 38.9. *Xiphinemoides* spp was absent in Kaduna and Plateau States but most encountered in Ogun State with population density

Table 1: Population of plant-parasitic nematodes (with percentages) from 250 ml soil associated with cucumber in selected production States

State	*Meloidogyne*	*Helicotylenchus*	*Pratylenchus*	*Tylenchus*	*Xiphinema*	*Aphelenchoides*	*Criconemoides*	*Scutellonema*	*Longidorus*	*Rotylenchus*	*Belonolaimus*
Lagos	2849 (24.6)	894 (21.7)	1234 (38.9)	384 (27.3)	249 (30.0)	61 (10.0)	515 (45.0	781 (35.9)	138 (46.5)	64 (100.0)	187 (65.8)
Ogun	1589 (13.7)	517 (12.5)	492 (15.5)	130 (9.2)	271 (32.6)	132 (21.5)	142 (12.4)	388 (17.8)	16 (5.4)		0
Osun	1332 (11.5)	576 (14.0)	227 (7.1)	35 (2.5)	113 (13.6)	110 (17.9)	14 (1.2)	392 (18.0)	0	0	0
Oyo	1197 (10.3)	679 (16.5)	473 (14.9)	44 (3.1)	198 (23.8)	180 (29.4)	36 (3.1)	615 (28.3)	16 (5.4)	0	0
Kaduna	2819 (24.3)	1044 (25.3)	619 (19.5)	617 (43.8)	0	61 (10.0)	412 (36.0)	0	89 (30.0)	0	0
Plateau	1814 (15.6)	413 (10.0)	136 (4.1)	199 (14.1)	0	69 (11.3)	26 (2.3)	0	38 (12.8)	0	97 (34.2)

Values in parenthesis are Percentage Nematode Population across the State

of 271/250 ml soil (Table 1). *Aphelenchoides* spp. was most encountered in Oyo State with population density of 180/250 ml soil and the least was recorded in soils from Lagos and Kaduna States, with population densities of 61/250 ml soil each (Table 1).

Populations of *Scutellonema* spp. was highest in Lagos State (781/250 ml soil) and it was least encountered in Ogun State (388/250 ml soils); the nematode was absent in the soils from Kaduna and Plateau States (Table 1). *Rotylenchus* spp was only present in Lagos State with population density of 64/250 ml soils while *Belonolaimus* spp. was also only present in the soils from Lagos and Plateau States with populations densities of 187/250 ml soil and 97/250 ml soils respectively (Table 1). The most encountered plant parasitic nematodes was *Meloidogyne* spp. (38) followed by *Pratylenchus* spp. (17) and the least encountered was *Rotylenchus* spp and *Aphenlenchoides* spp. with each, respectively across the LGAs in Lagos State (Figure 1). However, in Ogun State, the most encountered plant- parasitic nematodes across the LGAs was also *Meloidogyne* spp (33) and the least was *Longidorus* spp (2) (Figure 2). In Osun state, Meloidogyne spp was most often encountered (37) followed by Helicotylenchus (16), while the nematodes least encountered were *Tylenchus* spp. and *Criconemoides* spp (Figure 3). In Oyo State *Meloidogyne* spp. was also the most encountered plant-parasitic nematodes (37) while the least were *Criconemoides* spp and *Xiphinema* spp 5 each (Figure 4). The most encountered plant-parasitic nematodes in the soil across LGAs in Kaduna State was *Meloidogyne* spp (53) and the least encountered were *Aphelenchoides* spp. and *Longidorus* spp. 1 each (Figure 5) The most encountered was also *Meloidogyne* spp. (60) while the least were *Aphelenchiodes* spp and *Longidorus* spp. 2 each across LGAs in Plateau State (Figure 6).

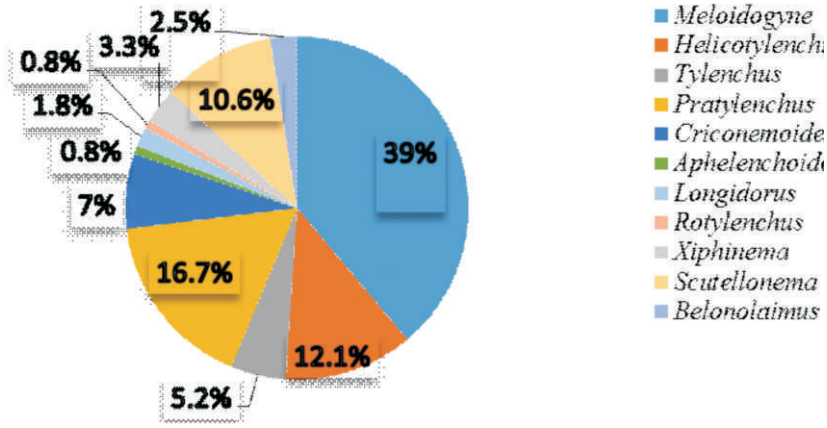

Figure 1: Percentage representation of plant-parasitic nematodes associated with Cucumber soil per 250ml across the LGAs in Lagos State

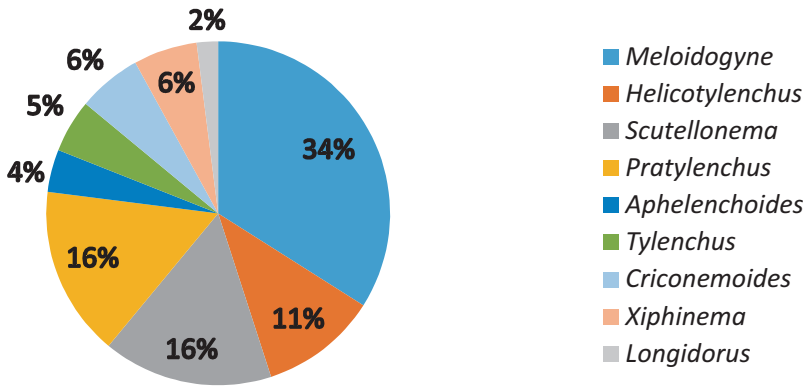

Figure 2: The percentage representation of plant-parasitic nematodes associated with Cucumber soil per 250 ml across the LGAs in Ogun State.

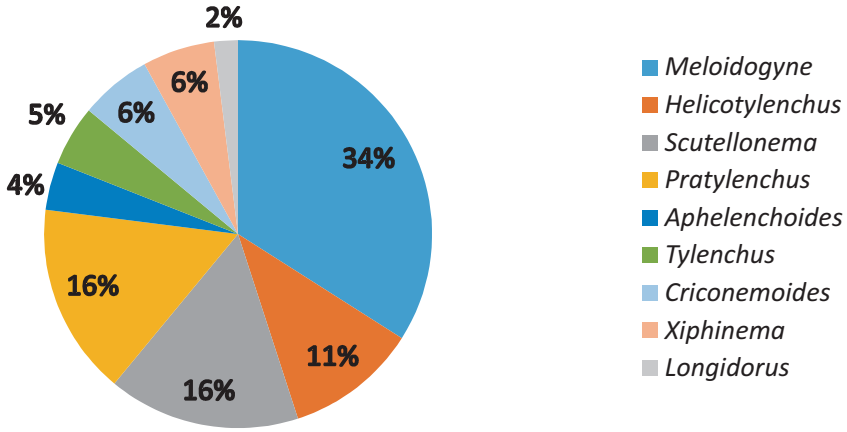

Figure 3: The percentage representation of plant-parasitic nematodes associated with Cucumber soil per 250ml across the LGAs in Osun State.

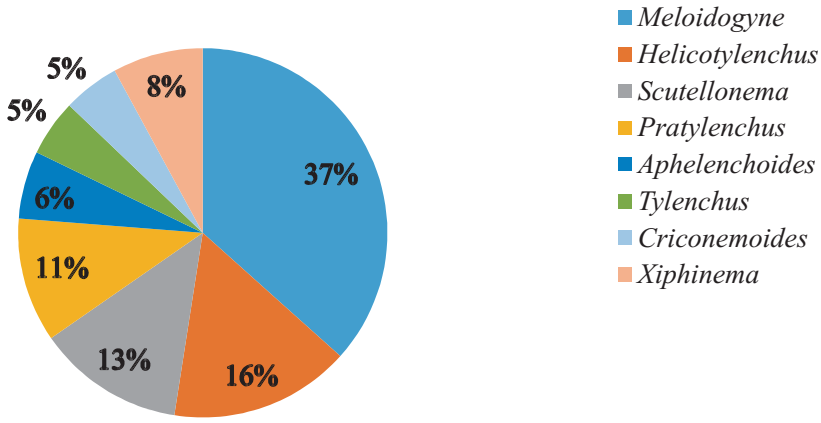

Figure 4: The percentage representation of plant-parasitic nematodes associated with Cucumber soil per 250 ml across the LGAs in Oyo State.

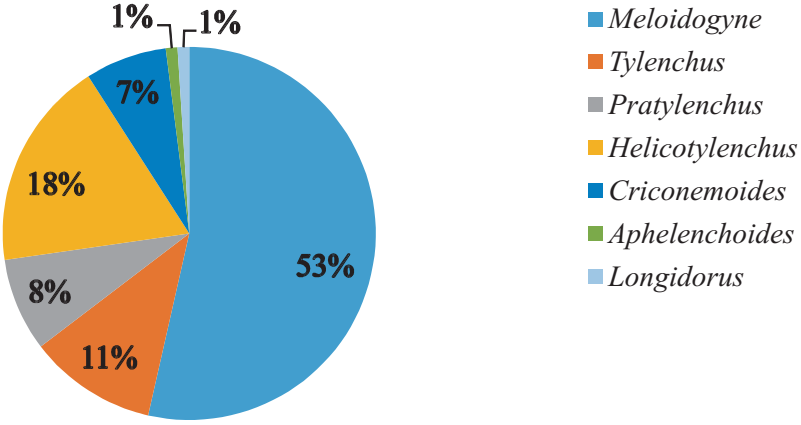

Figure 5: The percentage representation of plant-parasitic nematodes associated with Cucumber soil per 250 ml across the LGAs in Kaduna State

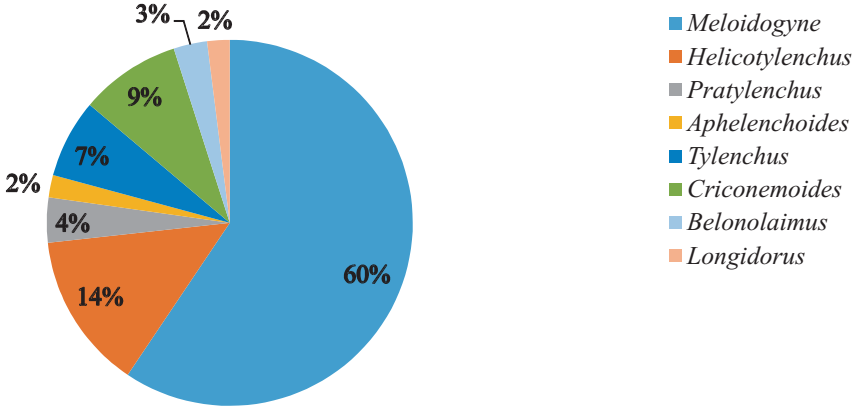

Figure 6: The percentage representation of plant-parasitic nematodes associated with Cucumber soil per 250 ml across the LGAs in Plateau State

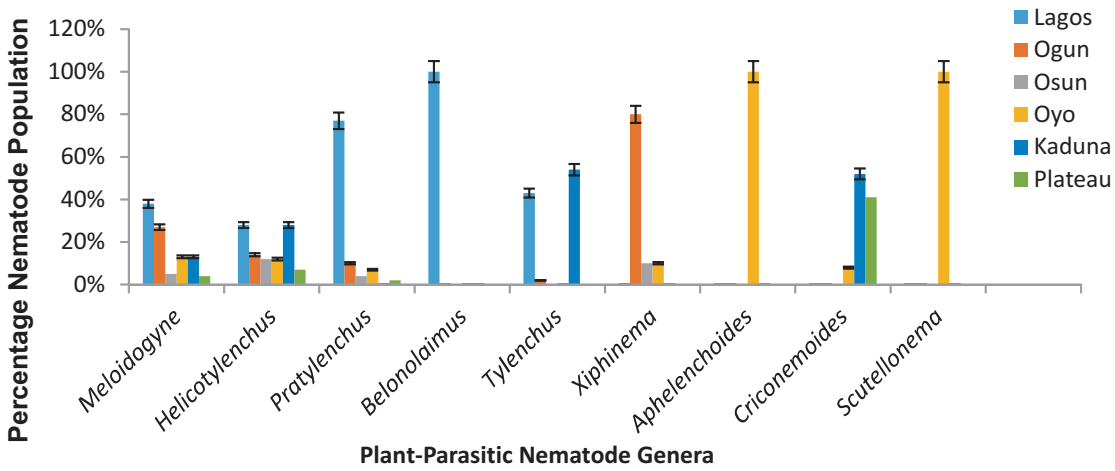

Figure 7: The percentage plant-parasitic nematodes population in 10g of the roots of cucumber in major production States

In the roots, nine genera of plant-parasitic nematodes were encountered (Figure 7). *Meloidogyne* spp was encountered in the roots of cucumber in all the States Surveyed, the highest percentage population density was encountered in Lagos State (38) while the least was encountered in Plateau State (4) (Figure7). This was followed by *Helicotylenchus* spp. with the highest population density in Lagos State (28) while the least was encountered in Plateau State (7). *Scutellonema* spp was only encountered in root samples from Oyo State (Figure 7). *Tylenchus* spp was encountered only in Lagos and Kaduna States. *Xiphinema* spp. was encountered in Ogun, Osun and Oyo States with the highest percentage population density in Ogun State (80) while Osun and Oyo States had 10 as population densities each (Figure 7).

DISCUSSION

Eleven genera of plant-parasitic nematodes occurring at varying frequencies and populations were identified in this survey. These include *Meloidogyne* spp, *Pratylenchus* spp., *Scutellonema* spp., *Helicotylenchus* spp., *Xiphinema* spp., *Aphelenchoides* spp., *Tylenchus* spp., *Criconemoides* spp., *Longidorus* spp. *Belonolaimus* spp., and *Rotylenchus* spp. Most plant-parasitic nematodes identified in this survey can be anticipated to be major threats to vegetable production and should be considered as serious pests (Anwar *et al*., 2007; Anwar and McKenry, 2012). *Xiphinema* spp., *Tylenchus* spp, and *Longidorous* spp were among the plant-parasitic nematodes identified, they are ectoparasites of epidermal root tissues and they have not been documented as dangerous pests of vegetables (Anwar *et al*., 2013). Their populations therefore need to monitored with possible consideration of effective management options should large or damaging populations develop.

Meloidogyne spp. a sedentary endoparasite of vascular tissues, was identified from the soil and roots of cucumber in all the states surveyed with frequencies and densities that were variable. *Meloidogyne* spp. was the predominant genus in all surveyed States. *Meloidogyne* spp. are common in vegetable soils worldwide where they parasitize vascular root tissues and induce root galls. The root-knot nematode, *M. incognita,* is among the most common plant-parasitic nematodes (Sasser, 1979; Abawi and Widmer, 2000; Davis *et al*., 2003; Anwar and McKenry, 2010). In addition to extensive root galling leading to malfunctioning of the root systems, root-knot nematodes are often associated with

increased incidence and severity of *Fusarium* wilts (Anwar and Khan, 1973, Martin *et al*., 1994).

Second on the list was *Pratylenchus* spp., migratory endoparasite and cortical feeders, their infection can result in necrotic brown lesions within rootlets. As a result of infection, interference with water and nutrient movement within the plant tissues can occur (Dorhout *et al*., 1991) causing stress-induced yield losses. *Pratylenchus* spp is also known to enhance the severity of *Verticillium* wilt of vegetables (Vrain, 1987). *Aphelenchoides* spp, *Helicotylenchus* spp, and *Tylenchus* spp, were also recorded from each of the states but not at each of the Local Government Area.

The reduction in yield of vegetables crops due to the feeding of plant-parasitic nematodes can be more than 40% (Anwar and McKenry, 2012). The presence of these serious plant-parasitic nematodes in abundance on cucumber production should be taken seriously by the farmers. From the discussions held with selected farmers in the locations from where samples were collected, it was clear that they were not aware of the presence of nematodes, symptoms of damage and yield reduction potentials of nematodes. More so, majority of the farmers practice mixed farming and all the plants involved are known hosts to root-knot nematode. The presence of successive susceptible crops leads to rapid multiplication of root-knot nematode populations on their fields and ultimately a reduction in yield.

The results of this survey indicate that plant-parasitic nematodes are widely distributed on cucumber farms. This information on their occurrence on cucumber will be helpful for growers for planning and administering appropriate management strategies to reduce their populations below economic threshold levels. There is therefore an urgent need for extension officers in the State Ministries of Agriculture and Agricultural Development Projects (ADPs) to enlighten farmers on the presence of plant-parasitic nematodes in their farms and their implications for crop growth and yield. Also, they may need to conduct farm trials in form of demonstration plots for nematode control.

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