Diversity of Plant Parasitic Nematodes of Tree Tomato (*Solanum betaceum* Cav.) In Kiambu and Embu Counties, Kenya

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ABSTRACT

Surveys were conducted in five areas representing different agro-ecological zones (AEZs) in Kiambu and Embu counties in Kenya during January to April 2019 cropping season to determine population density, genera diversity and distribution of plant parasitic nematodes associated with tree tomato farming systems. Nematodes were extracted from 200cc of soil or 5g of roots using modified Baermann technique. The nematode diversity and evenness in the study areas were determined using Shannon and Simpson's indices respectively. Soil nutrients were determined using Mehlich double acid digestion. A structured questionnaire was used to get information on farming practices from sampled farmers. *Meloidogyne* spp. was the most dominant genera followed by *Pratylenchus* spp. and *Helicotylenchus* spp. in both soil and root samples. Githunguri recorded significantly high mean number of 15.92 and 4.29 nematodes in soil and root samples respectively. Kiambaa had the highest diversity index of 1.59 and 0.86 in soil and roots respectively while genera richness was highest in Kiambaa with 13 nematode genera in soil. Simpson's index of evenness was highest in Kiambaa with 0.69 and 0.47 in soil and root samples respectively. Githunguri had the high egg mass index of 2.00. Soil texture and elements of C, P, K and N together with farming practices may have contributed to the nematode community structure in the study areas.

Keywords: Agro-ecological zone, Diversity, Plant parasitic nematodes, Baermann technique

Introduction

Plant parasitic nematodes (PPNs) cause enormous crop yield losses worldwide (Karuri *et al.,* 2017). Of all the PPNs, *Meloidogyne* spp. (Root-knot nematodes) is the most serious damaging nematode globally (Wangai *et al.,* 2014; Jones *et al.,* 2013) and it is polyphagous. In Kenya, PPNs are widely distributed in many farming systems (Karuri *et al.,* 2017; Mwangi *et al.,* 2014; Nzensya *et al.,* 2014; Waceke, 2007). In Kenya, tree tomato is an important small holder fruit in most areas with an increasing market demand (Muriithi *et al.,* 2013). Apart from home consumption, the fruits are sold and therefore provide local farmers with rural income generation, food and nutrition security and hence important in poverty alleviation (Muriithi *et al.,* 2013). The occurrence of PPNs in tree tomato cropping systems in Kenya has not been scientifically documented previously and therefore they have not been given keen attention in terms of awareness and management.

Soil physicochemical properties play an important role in influencing the nematode community structure. Soil texture plays a critical role in nematode activities ranging from reproduction to infection. It is reported that sandy soils are favorable to PPNs Bertrand et al, 2001)

The major challenges of nematode management in tree tomato in Kenya are lack of adequate knowledge, low PPN awareness and lack of data on nematode problems. This research study was carried out to determine occurrence, diversity and distribution of PPNs ass0ciated with tree tomato growing areas in Kenya.

Materials and Methods

A nematological survey was conducted in five areas each representing different agro-ecological zones (AEZs) in Kiambu and Embu counties in Kenya to determine population density, diversity and distribution of PPNs on tree tomatoes. The study areas were: Gatundu South (1820-2280 meters above sea level) in LH1, Githunguri (1590-1850 meters a.s.l) in UM1, Embu-Runyenjes (1680-1760 meters

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a.s.l) in UM2, Kiambaa (1520-1580 meters a.s.l) in UM3 and Ruiru (1360-1520 meters a.s.l) in UM4. A total of sixty farms were surveyed where ten samples were randomly taken on each farm in a zigzag fashion. The samples were composited for each farm to make one composite sample, labeled and transported in a cool box to Kenyatta University Nematology laboratory for nematode assay.

A structured questionnaire was used to get information on farming practices where the check list included farmer's name, crop variety, date of orchard establishment, altitude, farm size, crop management, previous crops and intercropping during the survey.

Nematodes were extracted from 200cc of soil or 5g of roots using modified Baermann extraction tray method as described by Hooper et al., (2005). Roots were chopped to 1cm, blended in 100 ml of water and the resultant suspension processed the same way as soil samples. Two (2ml) of the nematode suspension was pipetted in to a gridded counting dish and put under a microscope for counting. This was done thrice and the average recorded. Isolated nematodes were fixed, mounted on slides and identified to genus level under stereo microscope basing the identification on morphological features as described by Mai. (2018); Mekete *et al*. (2012); and Siddiqi (2000). Identification was based on adult female nematodes, but in some occasion second stage juveniles (J2) were also used. Another 5g sub-sample of roots were washed, stained in phloxine-B (15g/L) and scored for egg mass index on a scale of 0-5, where 0=no egg masses, 1=1-2 egg masses, 2=3-10 egg masses, 3=11-30 egg masses, 4=31-100 egg masses and 5=>100 egg masses (Taylor & Sasser, 1978). The egg masses were counted under a dissecting microscope.

The soil physico-chemical parameters were determined for each farm using 500g of the composite soil. The soil analysis was done in Plant and Crop Laboratory in Agriculture Sciences Department at Kenyatta University. The elements of P, K, Na, Mg, Ca and Mn were analyzed using Mehlich double acid digestion method. Total organic carbon was analyzed using calorimetric method where carbon concentration was read on the spectrophotometer at 600nm. The total nitrogen was estimated using Kjeldahl method in which soil samples were digested using concentrated sulphuric acid containing Potassium sulphate, selenium and hydrated copper sulphate at 350°C. Total nitrogen was then determined calorimetrically. Soil pH was determined using a P^{H} meter in 1:1(w/v) suspension. Extractable phosphorus was estimated using Olsen method (for soils with P^{H}=7.0 or more). The dried soil samples were extracted in a ratio (w/v) with 0.5M solution of sodium bicarbonate at P^{H} 8.5. Phosphorus in the extract was determined spectrophotometrically.

The PPNs were counted and analyzed for population density while PPN diversity indices were calculated for each sample and their means computed for each of the five study areas and compared for genera diversity, richness and evenness. Calculations were based on methods of community analysis (Norton and Norton, 1978) where:

1. Population density = Mean number of a given genus in 200cc dry soil or 5g dry roots
2. Genera diversity using Shannon-Weiner index was used to calculate nematode genera diversity, to determine genera variation in the areas sampled;

   Diversity $H = \sum_{i=1}^{s} \text{pi logepi}$
   $H' = -\sum pi \ln pi$
   Where: Σ refers to "the sum of"
   s= the number of genera in the community
   pi = is the relative abundance (proportion) of the ith genera in the community
   ln = natural log (Shannon and Weaver, 1963).
3. Genera richness (S) = total number of genera in a given soil sample.
4. Genera evenness was calculated using Simpson's index of diversity (Ds) to determine relative abundance with which each genus was represented in areas sampled. This was done using formula below;

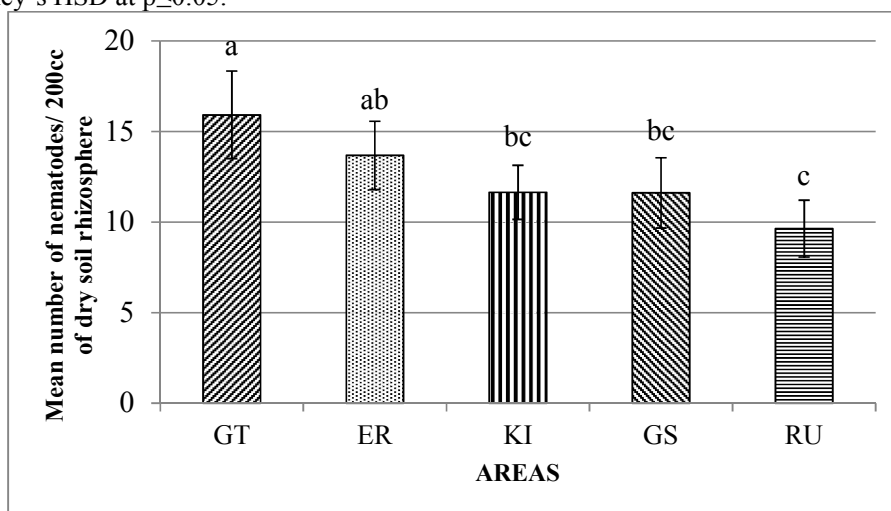$$D_s = 1 - \sum \left(\frac{ni}{N}\right)^2 \text{ (Simpson, 1949).}$$

 Where;
   Ds= Simpson's Diversity Index
    ni= the number of individuals of genera i
   N= the total number of genera in the sample

Data was organized and summarized in MS excel sheets and analysed using Analysis of Variance (ANOVA) using SAS software version 9.1 at p≤0.05. Means significant were separated using Tukey's HSD at p≤0.05.

Fig. 1: Mean number of nematodes in rhizosphere soil of tree tomato in each of the study areas. Bars with the same letter(s) are not significantly different (p≥0.05) with Tukey's HSD test.

The overall mean population density of PPN genera ranged from 0.11 to 84.62 nematodes per 200cc of dry soil (Table 1). *Meloidogyne* had the highest mean population density per 200cc of dry soil followed by *Helicotylenchus* and *Pratylenchus* while *Hoplolaimus* was the least. *Hemicycliophora*, *Longodorus*, *Trichodorus* and *Xiphinema* also recorded high mean population though not significantly different (p≥0.05) from each other (Table 1).

Table 1: Variations in population densities and overall mean of PPN genera from rhizosphere soils in the study areas

| GENUS	ER	GS	GT	KI	RU	Overall mean population
			AREAS			
Belonolaimus	1.20±0.47de	0.00±0.00d	0.00±0.00e	0.00±0.00e	0.00±0.00d	0.24±0.11fg
Cricinemoide	0.00±0.00e	0.00±0.00d	0.00±0.00e	2.21±0.74e	0.92±0.43d	0.63±0.20 efg
Helicotylenchus	20.50±2.77bc	7.04±0.92cd	9.88±2.27d	17.67±2.45bc	20.00±3.30b	15.02±1.29c
Hemicycliophora	8.54±1.72de	3.75±1.32d	0.42±0.31e	15.83±2.07bcd	1.75±0.69d	6.06±0.94de
Hoplolaimus	0.00±0.00e	0.00±0.00d	0.00±0.00e	0.54±0.32e	0.00±0.00d	0.11±0.07g
Longidorus	8.42±1.95de	4.29±0.83d	6.42±1.26dc	7.29±1.40cde	3.79±0.93cd	6.04±0.62de
Meloidogyne	98.88±5.75a	95.92±10.36a	74.42±10.06b	76.71±6.94a	77.17±9.18a	84.62±3.98a
Paralongidorus	1.75±0.77de	0.54±0.32d	0.29±0.29e	0.67±0.32e	0.00±0.00d	0.65±0.20efg
Paratrichodorus	2.91±0.98de	1.25±0.52d	0.00±0.00e	0.25±0.17e	0.92±0.32d	1.07±0.26efg
Pratylenchus	11.13±1.62cd	18.50±1.46c	21.25±2.80c	10.46±1.76cde	8.17±1.04bcd	13.90±1.03c
Radopholus	0.54±0.42de	0.00±0.00d	0.67±0.43ee	0.13±0.13e	0.71±0.40d	0.41±0.15egf
Trichodorus	10.46±2.43cde	3.17±0.90d	3.58±0.91de	6.25±1.06de	5.71±1.35cd	5.83±0.71def
Tylencus	2.75±0.95de	2.04±1.12d	9.27±3.98d	1.54±1.54e	0.00±0.00d	3.30±0.95defg
Xiphinema	10.29±2.87cde	1.50±0.71d	4.75±0.86de	13.75±1.21bcd	8.88±2.73bcd	7.83±1.00d
FLNs	27.88±2.03b	36.17±1.81b	106.92±5.40a	21.25±1.46b	16.63±0.97bc	41.77±4.49b
P-Value	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001

KEY: ER= Embu Runyenjes, **GS**= Gatundu South, **GT**= Githunguri, **KI**= Kiambaa and **RU**= Ruiru. Means are data ±standard error (S.E) of nematodes per 200cc of dry soil in sixty soil samples collected in five study areas. Means with the same letter(s) in a column are not significantly different using Tukey's Honestly Significant Difference (HSD) at p≤0.05.

Distribution of PPN genera in the study areas in soil rhizosphere

There were higher significant (p≤0.05) differences in the population densities of various nematode genera in each of the study areas (Table 1). *Meloidogyne* spp. recorded very high population density in each area and this was significantly different from the other genera. The genera *Meloidogyne*, *Helicotylenchus* and *Pratylenchus* were dominant in each area apart from KI, in KI area Hemicycliophara and xiphinema genera dominated pratylenchus, while in RU area xiphinema dominated praty lenchus. ER recorded the highest mean of *Meloidogyne* followed by GS. Belonolaimus was present in ER only while *criconemoide* occurred at KI and RU while *Hoplolaimus* was present at KI only. The genera *Paralongidorus* and *Tylenchus* were found in all the areas apart from RU while *Paratrichodorus* was found in all areas apart from GT (Table 1).

**Plant parasitic nematodes and free living nematodes in tree tomato soil rhizosphere**

The number of PPNs in tree tomato rhizosphere soil samples was comparatively higher in all the study areas than free-living nematodes (FLNs). However there was significant difference (p ≤ 0.05) between the PPNs and FLNs in the study areas (Figure 2).
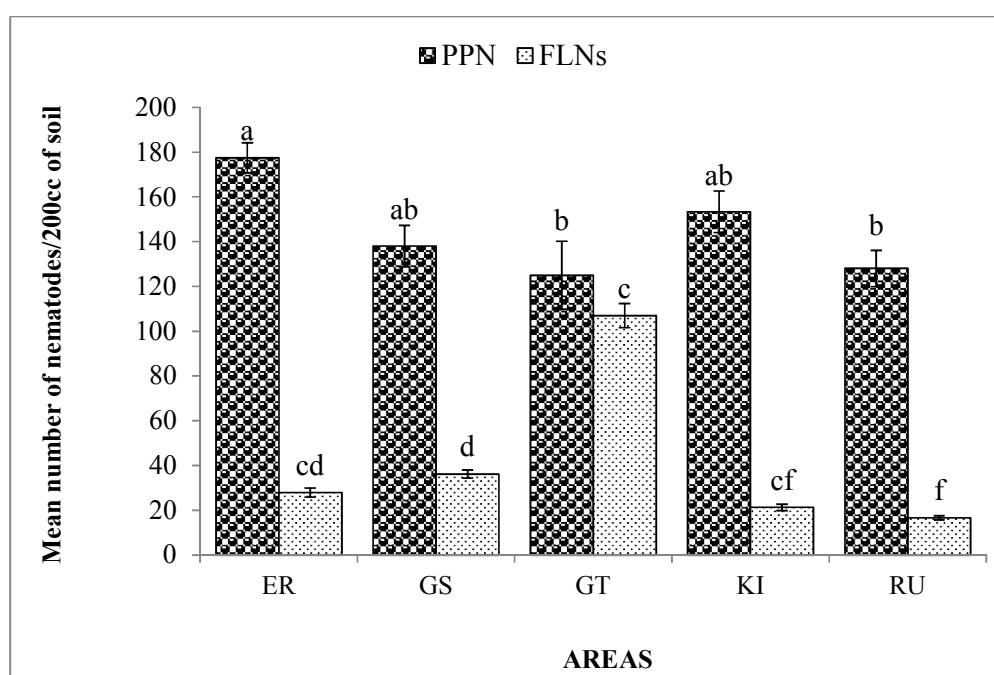
Fig. 2: Number of plant parasitic nematodes (PPNs) and free-living nematodes (FLNs) in rhizosphere soil of tree tomatoes in five study areas. Bars with the same letter(s) are not significantly different (p≥0.05) with Tukey's HSD test

Population densities of PPNs from tree tomato roots

There was a significant difference (p≤0.05) in the mean number of nematodes in the study areas. Githunguri (GT) and Embu-Runyenjes (ER) recorded the highest mean number of nematodes in roots of tree tomatoes (Figure 3). This was not significantly different (p≥0.05) from each other but significantly different with other areas. Kiambaa (KI) area recorded the lowest mean number of nematodes in roots followed by Ruiru (RU) and Gatundu South (GS) at 2.65, 3.31 and 3.30 respectively (Figure 3).
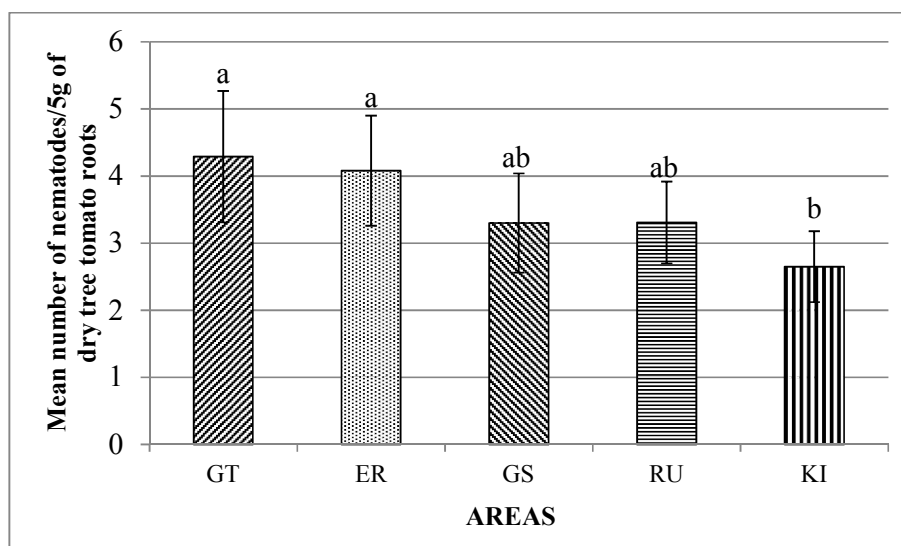
Fig. 3: Mean numbers of nematodes in roots of tree tomato in each of the study areas Bars with the same letter(s) are not significantly different (p≥0.05) with Tukey's HSD test

The overall mean population density of PPN genera in roots of tree tomato ranged from 0.13 nematodes for *Longodorus* spp. to 20.22 nematodes for *Meloidogyne* spp. per 5g of dry roots (Table 2). The root knot nematodes (*Meloidogyne*) recorded significantly ($p<0.05$) the highest population density than other PPN genera (Table 2). On the other hand, *Helicotylenchus*, *Hemicycliophora* and *Pratylenchus* had their population densities not significantly different ($p≥0.05$) from each other but were significantly different ($p≤0.05$) to other PPN genera from roots of tree tomatoes (Table 2). Mean numbers of *Belonolaimus*, *Longidorus*, *Trichodorus* and *Xiphinema* though varied were not significantly different ($p≥0.05$) from each other and occurred in very low numbers (Table 2).

Distribution of PPN genera in roots of tree tomato of the study areas
The PPN genera in roots differed significantly ($p≤0.05$) in in each study area. *Meloidogyne* was found in significantly high population density in roots in all the five study areas than any other genera. The other nematode genera were not significantly different from each other. However, there were variations with *Pratylenchus* and *Helicotylenchus* as second to *Meloidogyne* (Table 2).

Table 2: Variatons in population densities and overall mean of PPN genera from tree tomato roots in the study areas

| GENUS | AREAS | | | | | |
	ER	GS	GT	KI	RU	Overall mean population
Belonolaimus	1.50±0.59b	0.00±0.00b	0.42±0.42b	0.00±0.00b	0.00±0.00c	0.38±±0.16c
Helicotylenchus	1.63±0.64b	0.67±0.32b	2.17±0.72a	1.38±0.42b	1.54±0.61c	1.48±0.25[bc]
Hemicycliophora	2.21±0.45b	1.08±0.38b	0.83±0.49b	2.75±0.41b	1.42±0.47c	1.66± 1.66[bc]
Longidorus	0.13±0.13b	0.13±0.13b	0.29±0.29b	0.13±0.13b	0.00±0.00c	0.13±0.07[c]
Meloidogyne	19.85±1.65a	21.58±1.24a	27.96±2.46a	14.63±1.82a	17.08±0.81a	20.22±0.94a
Pratylenchus	3.29±1.46b	1.96± 0.96b	2.25±0.88b	1.50±0.49b	6.42±1.36b	3.08±0.52[b]
Trichodorus	3.75±3.75b	0.42±0.31b	0.29±0.29b	0.00±0.00b	0.00±0.00c	0.89± 0.75[c]
Xiphinema	0.25±0.17b	1.29±0.49b	0.13±0.13b	0.83±0.40b	0.00±0.00c	0.50± 0.14[c]
P-Value	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001

KEY: ER= Embu Runyenjes, **GS**= Gatundu South, **GT**= Githunguri, **KI**= Kiambaa and **RU**= Ruiru. Means are data ±standard error (S.E) of nematodes per 200cc of dry soil in sixty soil samples collected in five study areas. Means with the same letter(s) in a column are not significantly different. Means were separated using Tukey's Honestly Significant Difference (HSD) at p≤0.05

Plant parasitic nematode genera diversity indices in soil and roots of tree tomato

The Shannon-Wiener diversity indices were significantly different (P≤0.05) across the five study areas in both soil and root samples. The PPNs were more diverse in the soils in Kiambaa that had a higher diversity index > 1.5 while the PPNs less diverse in ER, GT, RU and GS in descending order that had lower diversity indices < 1.5 (Table 3).

Kiambaa had higher nematode genera richness of 13 nematode genera in soil samples followed by Embu-Runyenjes (12), Gatundu South, Githunguri and Ruiru (10 each)). In root samples, Embu-Runyenjes and Githunguri had the highest number of nematode genera (8) followed by Gatundu South, Kiambaa and Ruiru respectively (Table 3).

The Simpson's evenness index in soil was significantly (P≤0.05) varied across the five areas. The Simpson's index was high in Kiambaa followed by GT, ER, RU and GS in descending order respectively for soil samples (Table 3). In root samples, Kiambaa had the highest Simpson's index of genera evenness while GS had the least (Table 3).

The egg mass indices were significantly different (P≤0.05) among the study areas. GT had the highest index followed by ER.

Table 3: PPN diversity and egg mass indices in soil and roots of tree tomato in five study areas.

| AREAS/AEZ	Genera Diversity (H)		Genera Richness		Genera Evenness		EMI
	Soil	Roots	Soil	Roots	Soil	Roots	
ER (UM2)	1.47±0.07ab	0.82±0.07a	12	8	0.64±0.03ab	0.44±0.04a	1.17±0.03ab
GS (LH1)	1.09±0.08c	0.56±0.09a	10	7	0.49±0.04b	0.30±0.05a	0.83±0.17b
GT(UM1)	1.37±0.08abc	0.55±0.08a	10	8	0.65±0.03a	0.31±0.04a	2.00±0.35a
KI (UM3)	1.59±0.06a	0.86±0.09a	13	6	0.69±0.02a	0.47±0.04a	0.58±0.23b
RU(UM4)	1.20±0.11bc	0.73±0.01a	10	4	0.56±0.05ab	0.43±0.06a	0.67±0.22b
P-Value	<0.0006(0.0019)	<0.048(0.0253)			<0.001(0.0036)	<0.03(0.0188)	<0.0019(0.0088)

KEY: ER= Embu Runyenjes, **GS**= Gatundu South, **GT**= Githunguri, **KI**= Kiambaa and **RU**= Ruiru, EMI = Egg-mass index. Means are data ± S.E. Means with the same letter(s) in a column are not significantly different with Tukey's Honestly Significant Difference (HSD) at (p≥0.05). P-values in parenthesis are from transformed data

Farming practices in tree tomato production in Kiambu and Embu counties

The proportions of farms under tree tomato were small in all the areas. There were 75% of the farmers who grew tree tomatoes on less than 0.5 acres and only 25% used between 0.5-1 acres of lands. None of all the sixty farmers interviewed had practiced crop rotation. However, intensive intercropping was practiced in all the study areas with 66.7% of the farmers using multiple crops 5% with cereal crops, 1.7% with vegetable crops, 20% with cash crops and 5% with fruit crops.

There were different varieties of crops previously grown where tree tomato were planted. 36.7% had grown cash crops, 33.3% multiple crops and 13.3% vegetable crops (Figure 4). The purpose of growing tree tomatoes were different with13.3% of the farmers producing for home consumption, 38% for sale and 48.3% for both.

There were 68% of farmers who did not know about nematodes while 30% reported nematode attack and 2% reported no nematode attack (Fig.5).
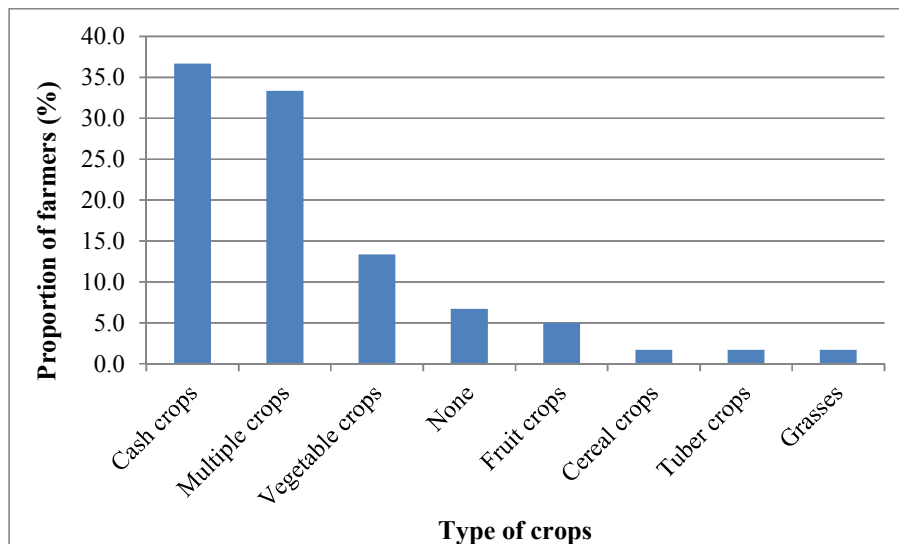
Fig. 4: previous types of crops grown before tree tomatoes by farmers in Kiambu and Embu counties
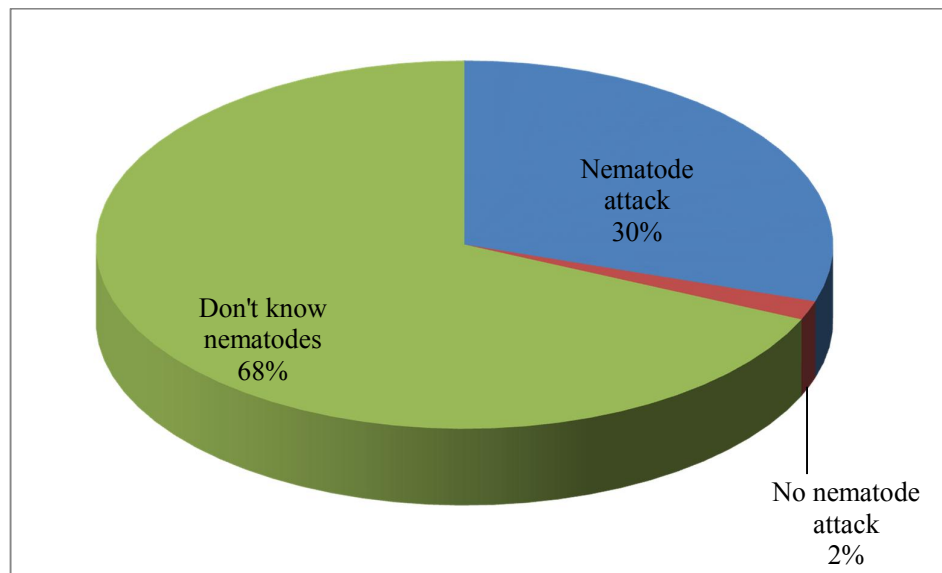
Fig. 5: Percentage of tree tomato farmers aware of nematode attack in Kiambu and Embu counties

Soil physicochemical characteristics on PPN populations in the soil in Kiambu and Embu counties

The soil physicochemical properties varied across the study areas (Table 4). The pH slightly varied in the study areas with ER, GS and GT recording higher pH values than KI and RU. The amount of Phosphorus were high in ER, GS and KI while in GT and RU were lower while, no significant differences were found in amount of potassium, N and C at all studied areasThe levels of C and N were lower in ER, GS and GT while in KI and RU higher. However, the soil texture did not vary much across the study areas (Table 4). The mean numbers of J2correlated with some of the soil elements. There was a significant positive correlation between J2 populations in the rhizosphere soils and phosphorus (y=0.2827x+149.95; R^2=0.0222). In addition, J2s significantly correlated positively with potassium (y=7.1734 x+150.93; R2=0.0039).

Table 4: Mean physicochemical properties of soils from five study areas

Properties	AREAS					
	ER	**GS**	**GT**	**KI**	**RU**	**P-value**
pH	6.20±0.4ab	6.50±0.5a	5.40±0.2abc	5.10±0.2bc	4.40±0.3c	0.0006
P (ppm)	20.80±6.3ab	20.00±1.3ab	12.50±2.5b	39.50±9.7a	12.50±1.7b	0.0111
Mg (me%)	1.87±0.2b	1.60±0.2b	2.00±0.2b	3.70±0.4a	2.00±0.1b	0.0001
K (me%)	0.81±0.2a	0.80±0.1a	0.60±0.1a	0.70±0.1a	0.60±0.1a	0.3453
Na (me%)	0.40±0.0a	0.41±0.0a	0.33±0.0a	0.40±0.0a	0.40±0.0a	0.3595
Ca (me %)	2.00±0.1b	4.80±1.0a	1.71±0.1b	1.90±0.1b	1.5±00.1b	0.0001
Mn (me%)	1.60±0.1a	1.50±0.1a	1.60±0.0a	1.50±0.1a	1.30±0.0a	0.1145
Fe (ppm)	50.50±2.7a	53.30±2.5a	47.10±1.4a	51.90±1.6a	49.61±0.4a	0.2317
Zn (ppm)	18.10±0.9a	21.80±1.7a	19.52±0.7a	18.71±0.5a	18.10±0.5a	0.0641
Cu (ppm)	1.41±0.2ab	1.52±0.2ab	1.30±0.1b	2.11±0.2a	1.90±0.2ab	0.0195
C (%)	2.50±0.3a	2.61±0.2a	2.42±0.4a	2.80±0.1a	3.67±0.6a	0.2225
N %)	0.24±0.0a	0.28±0.0a	0.25±0.0a	0.36±0.0a	0.43±0.1a	0.2503
Texture	SL	LS	LS	SL	SL	

Key: ER=Embu-Runyenjes; **GS**=Gatundu South **GT**=Githunguri, **KI**=Kiambaa and **RU**=Ruiri. **SL**=sandy loam and **LS**=loamy. Means are data ± S.E. Means followed by the same letter (s) in each row are not significantly different according to Tukey's HSD test at $P \geq 0.05$.

Discussion

Githunguri and Embu-Runyenjes recorded higher PPN populations. The high PPN population density in these two areas could be linked to intercropping tree tomatoes with susceptible crops like beans, kales, spinach, maize and cabbages observed during the survey. This may explain the high numbers of nematodes in the soil. Also the previous crops grown on the farms where tree tomatoes were established may explain the occurrence of high numbers of nematodes in roots. The previous susceptible crops provided the initial inoculum for tree tomato infection after planting. Prolonged mono-cropping of susceptible host plants may lead to plant parasitic nematodes build ups which causes serious crop damage (Bhan, *et al*., 2010). These areas also receives substantial amount of rainfall that ensures prolonged moisture in the soil for nematode movement. Although GS receives good amount of rainfall (Jaetzold and Schmidt, 1983), it had low PPN population density compared to GT and ER. This could be due to farmers applying inorganic fertilizers and fresh un-decomposed cattle manure noticed during the survey. Upon decomposition, organic manure releases toxic chemicals against PPNs and enhances proliferation of antagonistic micro-organisms against PPNS. KI and RU receive depressed rainfall and therefore relatively dry (Jaetzold and Schmidt, 1983). It is reported that areas with hot and dry seasons have reduced PPNs (Hussey, 1996). This may have been the cause of low numbers of PPNs from these areas.

This study revealed that the PPN genera were spread across the tree tomato growing areas with *Meloidogyne* spp. being the most dominant genera isolated followed by *Pratylenchus* and *Helicotylechus* in all the study areas. In Ecuador and Colombia, *Meloidogyne* spp., *Pratylenchus* spp. and *Helicotylechus* spp. were also recorded as dominant genera in the same crop (Vaquez and Soria 2017; Ramirez et al., 2015). These PPNs are polyphagous and their high numbers is linked to intercropping of tree tomatoes with susceptible host crops. It was also noted during the survey that many farmers had previously grown susceptible crops on the farms where tree tomato were established. This could be the reason for high numbers of PPNs isolated from soil and roots. *Hoplolaimus, Belonolaimus*, *Croconemoides*, *Paralongidorus*, *Radopholus*, *Longidorus*, *Xiphinema* and *Trichodorus* recorded very low population densities. This could be due to the presence of other aggressive dominant genera and interspecific competition from other PPNs (Blake 1969). *Belonolaimus* spp., *Criconemoide* spp., *Hoplolaimus* spp., *Paratichodorus* spp., and *Tylenchus* spp. are parasites of many vegetable crops like cabbage, capsicum, tomatoes and cow peas (Anwar *et al*., 2007). Farmers intercropping many vegetable crops with tree tomatoes explain their presence in some of the areas. Also the low numbers of these PPNs may indicate their ability to co-exist as external or internal parasites. However, the presence of *Radopholus* could be attributed to intercropping with

banana plantations in some of the study areas together with warm temperature in the study area that supports thriving of the nematode.

The number of free-living nematodes (FLNs) was low compared to PPNs. The low numbers of FLNs in cultivated farms is an indicator of loss of soil fertility and also due to soil degradations. The FLNs are highly sensitive to soil disturbances. This could have been the case in most of the study areas since many farmers were growing tree tomatoes on less than 0.5 acres of land. These small parcels of land are prone to soil disturbances due to intercropping and continuous cropping making the numbers of FLNs to go down. In this case, PPN numbers go up and became a big menace (Hillocks and Waller, 1997). In Githunguri, the number of FLNs was almost equal with the PPNs. Some of the tree tomato farmers were using cattle manure in the soil which could have increased the population of FLNs in the soil. Use of OAs has been reported to improve soil health, increase yields and enhances biodiversity of soil flora and fauna (Kimpinski *et al.*, 2003).

Nematode genera diversity was generally low in both roots and soil samples. This could be attributed to the small parcels of land used by farmers and intensive intercropping.. This could have been the case in most of the study areas. Agricultural intensification is linked to soil disturbances through tillage, organic amendments applications, use of pesticides and inorganic fertilizers and planting narrow range of plant genotypes or complete monoculture. This leads to a decline in nematode abundance, richness and diversity due to emergence of a single species and change genera composition (Kimenju *et al.*, 2009; Yeates and Bongers, 1999; Yeates, 1999). Low Shannon diversity numbers are linked to intercropping of tea and coffee monocultures. High application of agrochemicals like pesticides in coffee and fertilizers used in tea could be the factors (Kimenju *et al.*, 2009). Agro-ecosystems with reduced tillage have high nematode diversity while agrochemical use and practice of monoculture causes loss of nematode biodiversity (Kimenju *et al.*, 2009). The Genera richness in the roots was however low as compared to soil and this could be due to host specificity and preference of PPNs. On the other hand, the intensive intercropping in the study areas could be the reason why nematode genera richness was high in the soil samples. Tree tomatoes were highly intercropped with multiple crops like many vegetable varieties, cash crops like tea and coffee, cereal crops like maize and fruit crops all of which are also favorable hosts to various nematode genera. The high EMI in GT and ER may be due to high numbers of *Meloidogyne* spp. in both soil and roots.

It was evident from the study that tree tomato is a suitable host to various PPNs in tree tomato growing areas. Plant parasitic nematodes have been reported in many parts of the world (Ramírez-Gil *et al.*, 2017; Ramirez *et al.*, 2015) and in Kenya (Mwangi *et al.*, 2014; Waceke 2007; Waiganjo *et al.*, 2006). The distribution of PPNs in different areas may have also been influenced by soil physicochemical parameters and farming practices. Kandji *et al.* (2001) and Kimenju *et al.* (2009) showed that soil physic-chemical properties influence the occurrence, abundance, distribution and community structure of nematode. The low numbers of PPNs in Ruiru may be linked to high levels of carbon and nitrogen in that area in comparison, Githunguri and Embu –Runyenjes which recorded high numbers of nematodes in roots had low levels of carbon and nitrogen. Organic carbon is known to increase the number of antagonistic micro-organisms in the soil which reduces the numbers of PPNs. It has been reported that animal manure has been used in the management of RKNs and other PPNs (Nchore *et al.*, 2012). Soil organic amendments (OAs) enhances the build-up of nematode trapping fungi than inorganic fertilizers (Agbenin 2004). A study conducted by Nchore (2012) on indigenous leafy vegetables indicate that mono-cropping with susceptible hosts leads to increased *Meloidogyne* infection. High levels of potassium are known to increase PPNs in the soil. Badra & Yousif (1979) notes that Potassium enhances rapid development of nematodes by reducing their life cycle. High Phosphorus and Potassium levels increase nematode populations because of its essential role in nematode multiplication, nutrition and production of eggs. Potassium also improves root growth which increases the root surface area for nematode attack and feeding (Badra and Yousif, 1979). This explains the high numbers of PPNs in Embu-Runyenjes and Kiambaa which had high potassium. However, Ruiru had low levels of Potassium revealing the occurrence of low numbers of PPNs. This observation was in agreement with studies by Kandji *et al.* (2001) and Badra and Yousif (1979).

Most soils in the study areas were sandy loam (SL) and loam sand (LS). These types of soil structure supports high nematode population densities (Talwana *et al.*, 2008). This might have led to the high numbers of PPNs in the study areas. These types of soil is highly porous with good aeration

and this favors movement of nematodes (Talwana *et al*.,2008; McLean and Lawrence 2000; Norton and Norton, 1978). Most of the study areas receive substantial amount of rainfall around the year with good agricultural potential (Jaetzold *et al*., 2006) and this creates perfect conditions for nematodes to flourish. Moisture increases nematode populations (Wallace 1983). The pH was within range tolerated by most PPNs. Most ago-ecosystems fall in the range of pH 4-8 (Nyasani *et al*., 2008) and therefore tree tomato growing areas are suitable habitats for nematode survival. However, other findings report that extremes of pH affect nematode survival (Nyasani *et al*., 2008).

Conclusion

This study revealed that tree tomato is attacked by diverse PPNs in Kenya. The most dominant genera limiting tree tomato production were *Meloidogyne*, *Pratylenchus* and *Helicotylenchus*. Soil physicochemical properties and farming practices are the major factors contributing to the diversity of PPNs in tree tomato growing areas. Thus, there is need for further research on responses of different tree tomato varieties to root-knot nematodes (RKN) and create farmers awareness to PPN problems.

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