

Survey and Identification of Plant Parasitic Nematodes on Tomato Crop in Ethiopia

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Abstract: A survey was conducted to identify and quantify the frequency and population density of plant parasitic nematodes genera associated with tomato in Ethiopia, during 2018 cropping season. A total of one hundred nineteen composite soil and root samples were randomly collected from tomato growing areas of Oromia, SNNP, Tigray and Amhara regions. A total of eight plant parasitic genera were detected in the surveyed regions, viz *Meloidogyne*, *Helicotylenchus*, *Pratylenchus*, *Criconema*, *Rotylenchulus*, *Scutellonema*, *Paralongidorus* and *Hoplolaimus*. *Meloidogyne* was the most abundant and frequent genus with 283 nematodes/10g root and 45% frequency occurrence respectively. *Helicotylenchus* was second most frequent genus with (36%) frequency of occurrence. But in terms of abundance *Pratylenchus* was the second most abundant genus with mean population density of 73 nematodes/10g root followed by *Helicotylenchus* (62 nematodes/100ml soil). Of all the genera detected the least frequency of occurrence of (2%) and (3%) were observed with genera *Criconema* and *Paralongidorus* respectively. Additionally, *Criconema* and *Paralongidorus* were observed with lowest equal mean population density of 20 nematodes/100ml. The aim of the study was to know the distribution of plant parasitic nematodes across the four regions. Among the surveyed four regions Oromia was positive for all of the eight detected plant parasitic nematode genera while SNNP for five genera and Amhara and Tigray were positive for four and two nematode genera respectively. *Criconema* and *Rotylenchulus* were only detected in Oromia region. Moreover, the agro-ecological difference present in the sampling sites might be the underlying causes for the variation of nematode prevalence, abundance and diversity across the four regions. The study encourages more research work on establishing economic importance and designing management strategies on reported nematode pests.

Keywords: Distribution, Occurrence, Plant Parasitic Nematodes, Population Density, Tomato

1. Introduction

Tomato (*Solanum lycopersicum*) is among the most valuable agricultural crops worldwide [6]. In Ethiopia it is one of the most important cash and food crops widely grown by smallholder farmers, commercial state and private farms under rain and irrigated condition [18, 14]. According to FAO [10] commercial tomato production in Ethiopia has significantly expanded as the national agriculture strategies gave the highest priority for the production of high value cash crops like tomato. In the year of 2018/19 area under tomato production is about 4,322 hectares with total production of 5.46 t ha⁻¹ [4]. Despite the economic importance of tomato, the production is being

adversely affected by several biotic and abiotic constraints in Ethiopia. Plant-parasitic nematodes (PPNs) are most important biotic constraints which are responsible to cause significant economic losses worldwide on a wide variety of crops including tomato. To date, over 4100 species of plant parasitic nematode have been identified [7]. A number of plant parasitic nematode genera have been reported to be found in association with tomato [12, 3]. Sasser and Freckman [28] reported that the yield losses caused by nematodes to tomato are of about 20.6%, of which the largest proportion is caused by root-knot nematodes. Wondirad and Tesfamariam [32] reported that root knot nematode (*Meloidogyne incognita*) is an important problem and often a limiting factor in tomato cultivation in Ethiopia. Seid *et al.*, [26] have reported tomato yield loss due

to PPN ranged from 25% up to 100% and the main nematode genus attacking tomato was *Meloidogyne*. They feed by piercing plant cell that cause wound on plant which will serve as entryways for secondary pathogens such as fungi and bacteria. Even though, plant parasitic nematodes are causing high damage on crops the damage remains hidden because of multiple biotic and abiotic stresses [2]. This is likely to be a significant underestimate of the actual figure as many growers in developing nations are unaware of the existence of plant-parasitic nematodes [15]. Hence, the study on the distribution, frequency of occurrence and population densities of the nematode genera associated with tomato is very important in giving clear picture of the damage impact of nematodes on the crop. Therefore, the present survey was conducted with the aim to map, identify, quantify the frequency and densities plant parasitic nematodes associated with tomato in four major tomato growing regions of Ethiopia.

2. Materials and Methods

In 2018 growing season, one hundred nineteen soil and root samples were collected from four tomato growing regions of Ethiopia (Oromia, SNNP, Tigray and Amhara) Sampled sites of the four tomato growing regions were mapped using geographical coordinate system (figure 1). Samples were obtained by digging the soil to a distance downward of about 15cm-30 cm from the rhizosphere of the tomato plants in a systematic, zigzag-sampling pattern of each field. Composite soil samples of about 1kg and 200 g of adventitious roots were placed in plastic bags and transported

directly to the laboratory of nematology at Ambo Agricultural Research Center. Samples were kept in the refrigerator at 4°C awaiting extraction of nematodes [30]. Nematodes were extracted from both soil and root samples using extraction tray or modified Baermann funnel method [13]. Tomato roots were carefully washed cut into 0.5 mm-sized pieces and standard of a 10 g subsample was used for nematodes extraction. For soil samples processing, each composite soil sample was thoroughly mixed and sub sample of a 100ml soil was used for extraction of nematodes. Nematode suspensions were decanted and nematodes collected on a 38 µm sieve, rinsed into beakers, reduced to 10 ml. Nematodes were identified and quantified from 1 ml aliquots using nematode counting slide under a compound microscope. Nematodes population densities were calculated for each root and soil samples and expressed as the number of plant-parasitic nematodes on tomato in a 10 g root or 100 ml soil. Frequency of occurrence (FO) was calculated as described by. Prominence value (PV) was calculated for each of the nematode genera [9]. Nematode specimens from each sample fixed in TAF were processed to anhydrous glycerin for permanent slides, following a modified glycerin-ethanol method [8]. Finally for confirmation nematodes were further mounted on a glass slide and identified to genus/species level under compound microscope following keys and references of Siddiqi [27] and [19]. Genera were considered widespread when they occurred in more than 30% of the sites. A genus whose mean density was more than 10 individuals/100 g of root was considered abundant [30].

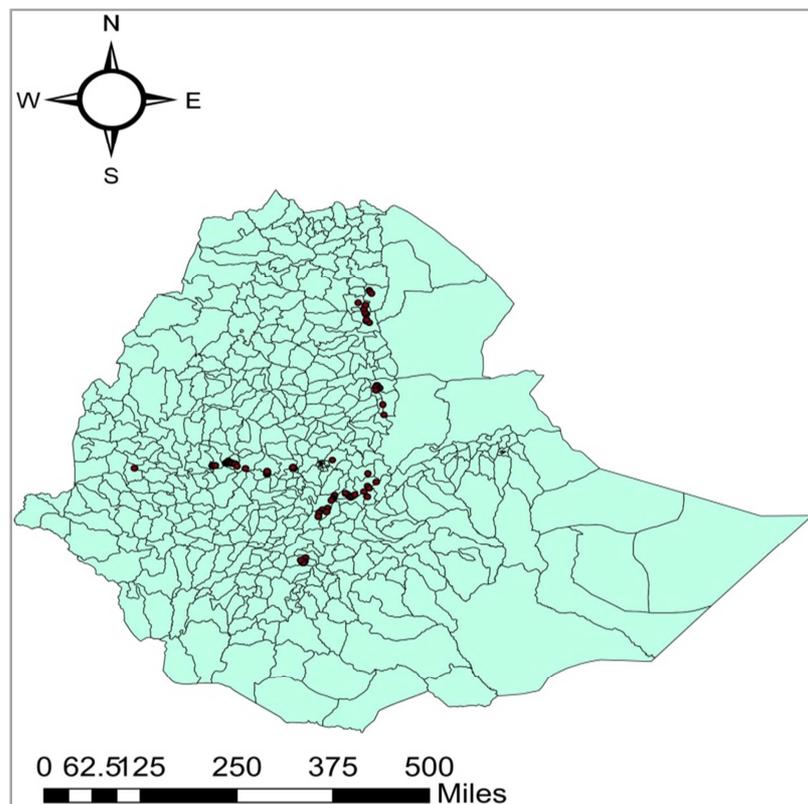


Figure 1. Map of Ethiopia showing sampled fields in the four tomato growing regions of Ethiopia.



Figure 2. Tomato root attacked by *Meloidogyne* spp (A&B) and Patch and yellow symptom of tomato field infested with Plant parasitic nematodes (C).

3. Results

A total of one hundred nineteen soils and root samples were collected from rhizosphere of tomato plants in 2018 growing season. Out of one hundred nineteen (119) samples collected from tomato fields, fifty-five (55) of the samples were found to be infested with different genera of plant parasitic nematodes. A total of eight (8) genera of plant parasitic nematodes were detected, namely *Meloidogyne*, *Helicotylenchus*, *Pratylenchus*, *Criconema*, *Rotylenchulus*, *Scutellonema*, *Paralongidorus* and *Hoplolaimus* (Table 1). All nematodes belonged to the order Tylenchida with the exception of *Paralongidorus* which belonged to the order Dorylaimida.

The distribution of these plant parasitic nematodes varies across the four regions. The highest number of plant parasitic nematode genera (8) were recorded in Oromia region followed by SNNP (5), Amhara (4) and Tigray (2) (Table 2). *Meloidogyne* and *Helicotylenchus* were detected in all of the four surveyed regions (Table 2). *Pratylenchus* and *Paralongidorus* were recorded only from Oromia and Amhara regions. On the other hand, *Criconema* and *Rotylenchulus* were only observed in the oromia region. The genera *Pratylenchus* and *Meloidogyne* were recovered only from root samples while *Helicotylenchus* *Longidorus*, *Criconema*, *Rotylenchulus*, *Scutellonema*, *Paralongidorus* and *Hoplolaimus* recorded only on the soil samples (Table 4).

Meloidogyne was the most frequently encountered genus, observed in 53 of the samples with frequency of occurrence (45%) followed by *Helicotylenchus* with (36%) frequency of occurrence (Table 3). Comparatively, *Rotylenchulus*, *Scutellonema* and *Hoplolaimus* were less frequent genera with equal frequency of occurrence (4%). However, among all the genera detected the least frequency of occurrence of (2%) was observed with genus *Criconema*. With regard to population density *Meloidogyne* was the most abundant plant-parasitic nematode detected in tomato root, with mean population density of 283 nematodes/10g root followed by *Pratylenchus* (73 nematodes/10g root) and *Helicotylenchus* (62nematodes/100ml soil) (Table 4). Population density as high as 14,975 of individuals per 10g root was also observed for genus *Meloidogyne* in a particular field in oromia region. The genera *Criconema* spp and *Paralongidorus* showed the least abundance of (20 nematodes/100ml soil) each of them. The other genera detected only in the soil samples includes,

Hoplolaimus, *Scutellonema*, *Rotylenchulus*, *Criconema* and *Paralongidorus* with mean population density of (32, 28, 24, 20, and 20 nematodes/100ml soil). The highest prominence value of 206 was recorded with genus *Meloidogyne* (Table 4). *Helicotylenchus* was the next most prominent nematode genus on tomato with prominence value of 41 followed by *Pratylenchus*, *Hoplolaimus* *Scutellonema* *Rotylenchulus*, *Paralongidorus* and *Criconema* with prominence value of 13 7, 6, 5 4 and 3 respectively.

Table 1. Plant parasitic nematodes genera isolated from soils and roots of tomato in four regions of Ethiopia.

Order	Sub-order	Family	Genus
Tylenchida	Tylenchina	Hoplolaimidae	<i>Helicotylenchus</i>
			<i>Haplolaimus</i>
		Criconematidae	<i>Rotylenchulus</i>
			<i>Scutellonema</i>
			<i>Criconema</i>
Dorylaimida	Dorylaimina	Paratylenchidae	<i>Paratylenchus</i>
		Heteroderdae	<i>Meloidogyne</i>
		Paralongidordae	<i>Paralongidorus</i>

Table 2. Distribution of plant parasitic nematode genera on tomato in Oromia, SNNP, Tigray and Amhara regions of Ethiopia during the 2018 growing season.

Nematode genera	Regions/ location			
	Oromia,	SNNP	Tigray	Amhara
<i>Meloidogyne</i>	+	+	+	+
<i>Helicotylenchus</i>	+	+	+	+
<i>Pratylenchus</i>	+	+	-	-
<i>Criconema</i>	+	-	-	-
<i>Rotylenchulus</i>	+	-	-	-
<i>Scutellonema</i>	+	-	-	+
<i>Paralongidorus</i>	+	+	-	-
<i>Hoplolaimus</i>	+	+	-	+

+ =Present - =Absent PPN

Table 3. Frequency of occurrence of plant parasitic nematode genera on tomato crop.

No	Nematode Genera	Frequency of occurrence of nematode genera on tomato plant	
		No.	F.O%
1	<i>Meloidogyne</i>	53	45
2	<i>Helicotylenchus</i>	43	36
3	<i>Pratylenchus</i>	3	3
4	<i>Criconema</i>	2	2
5	<i>Rotylenchulus</i>	5	4
6	<i>Scutellonema</i>	5	4
7	<i>Paralongidorus</i>	3	3
8	<i>Haplolaimus</i>	5	4
Total		119	

Table 4. Abundance and prominence value (PV) of plant-parasitic nematodes recovered from soils and roots of tomato (*Solanum lycopersicum L.*) in the four surveyed regions of Ethiopia.

Nematode genera	Soil (100cm ³)		Root (10 g)	
	Abundance	PV	Abundance	PV
<i>Meloidogyne</i>	-	-	283	206
<i>Helicotylenchus</i>	62	41	-	-
<i>Pratylenchus</i>	-	-	73	13
<i>Criconea</i>	20	3	-	-
<i>Rotylenchulus</i>	24	5	-	-
<i>Scutellonema</i>	28	6	-	-
<i>Paralongidorus</i>	20	4	-	-
<i>Hoplolaimus</i>	32	7	-	-
Total				

No=Number of samples containing a genus; F.O=Frequency of Occurrence; *Total number of sample=119

A=Abundance is mean number of individuals of a genus over the sampling sites where the genus was detected.

Frequency of occurrence (FO%) = number of sites where a genus detected/total number of sites sampled*100.

Prominence value (PV) = Mean population density *(Frequency of occurrence)^{1/2} *10⁻¹.

4. Discussion

The present study shows the distribution and occurrence plant parasitic nematode genera associated with tomato across the four tomato growing regions of Ethiopia including Oromia, Amhara, SNNP and Tigray. Eight plant parasitic nematodes genera were detected in association with tomato in Ethiopia namely *Criconea*, *Helicotylenchus*, *Hoplolaimus*, *Meloidogyne*, *Paralongidorus*, *Pratylenchus*, *Rotylenchulus* and *Scutellonema*. In Ethiopia, only *Helicotylenchus* spp., *Heterodera* spp., *M. incognita*, *M. ethiopia*, *Pratylenchus* spp were mentioned to be found in association with tomato [21]. However, all the eight plant-parasitic nematodes recorded in this survey were reported to be associated with tomato cultivation in many parts of the world [1, 29]. Of all detected Plant parasitic nematode *Meloidogyne* was the most predominant genus observed at higher frequency of occurrence (45%) and abundance (283 nematodes/10g root) as well as prominence value (206). This study is in line with the previous study of Wondirad Mandefro and Tesfamariam Mekete [33] who have reported *Meloidogyne* spp were the most widely spread and predominant plant parasitic nematode affecting tomato producing areas of Ethiopia. So far Wondirad Mandefro and Tesfamariam Mekete [33] have reported three *Meloidogyne* species including *M. incognita*, *M. javanica* and *M. ethiopia* on tomato from Ethiopia. MoARD [20] also reported that, the most common diseases in tomato production fields are the root knot nematode, *M. incognita* which is the dominant disease in Rift Valley of Ethiopia. Lately based on molecular and biochemical analysis [25] has been reported the presence of *M. arenaria*, *M. hapla* as well as *M. incognita* and *M. javanica* on tomato in Ethiopia. Similar results were obtained in other surveys conducted in Tanzania [16]. Additionally, in study conducted in Ghana [12, 22] have reported that *Meloidogyne* was the most frequent and abundant plant

parasitic nematode genus on tomato. The results of the current study have revealed that *Helicotylenchus* was second most frequent genus with (36%) frequency of occurrence. In agreement with the current finding [12] have also reported *Helicotylenchus* as the second most frequent genus next to *Meloidogyne* on tomato. The genera *Criconea*, *Hoplolaimus*, *Paralongidorus*, *Pratylenchus*, *Rotylenchulus* and *Scutellonema* were observed at very low frequency, abundance and prominence value. Of the detected plant parasitic nematodes genera *Meloidogyne*, *Rotylenchulus*, *Pratylenchus*, *Paralongidorus* and *Helicotylenchus* are among the most economical plant parasitic nematodes in tomato. Root knot nematode species including *M. arenaria*, *M. hapla*, *M. incognita*, and *M. javanica* were considered to have great economic impact in tomato [11]. Sikora & Fernandez, [29] have reported (2-200) juveniles/100ml soil as a threshold level for *M. arenaria* and *M. incognita* on tomato.

However, the impact of *Rotylenchulus* often get masked when it concomitantly presents with root knot nematodes [29]. The genus *Pratylenchus* is migratory endoparasitic nematode causes severe damage to economically important crops including vegetables [5]. *Helicotylenchus* is also one of the common a parasite of various economically important crops worldwide [17, 24, 23, 31]. The rest genera including *Criconea* spp, *Hoplolaimus* spp and *Scutellonema* spp were known to parasitize wide range of crops including tomato but their economic impact on tomato is not known. Moreover, the distribution of plant parasitic nematodes detected in this study varies across the four regions. The difference on the distribution of plant parasitic nematode across the regions might be associated with various factors present in sampling location. For instance, soil texture, altitude, pH, temperature, cultural practices, cropping sequence, existence bio-control agent and management practices have been reported to affect the distribution plant parasitic nematodes. The present study stated that lighter sandy soils are better than heavier clay soils in supporting nematode populations. The highest frequency and density of plant parasitic nematode were recorded in Oromia region; this might be due to the reason that most of tomato cultivation areas of this region have conducive climatic and edaphic condition which favors the survival and reproduction of plant parasitic nematodes. Additionally, tomato producing areas of Oromia region are positioned at lower altitude where there is higher temperature that enhances the survival of plant parasitic nematodes. Furthermore, most of the farmer in this region does not practice widely spaced crop rotation and multiple cropping systems. Therefore, these might create additional conducive condition for survival and consistent population buildup of plant parasitic nematodes in this region. However, further research will be needed on various factors that affect the occurrence and distribution of plant parasitic nematodes on tomato in Ethiopia. Moreover, further research also needed on species identity, economic importance as well as on their damage potential on tomato under Ethiopia condition.

5. Conclusion

The present study reported the presence of eight plant parasitic nematodes genera belonging to 5 families, found associated with the rhizosphere of tomato plantations at four regions, Oromia, SNNP, Tigray and Amhara. Among the eight phytonematode genera, *Meloidogyne* and *Helicotylenchus* genera seemed to be the prevailing nematode pests as they occurred at the rates of 53 and 43 times with percent occurrence of 45% and 36%, respectively. Moreover, the nematode genera, *Rotylenchulus*, *Scutellonema* and *Hoplolaimus* showed modest distributions as they occurred at equal rates of 5 times with percent occurrence of 4%, whereas, the genus, *Criconea* shows less frequent as it occurred at the rate of 2 times with percent occurrence of 2%. In general, the essence of this survey would facilitate in choosing planting systems in the selected locations and evade planting the susceptible hosts, as well as should use in design of nematode management programs. Higher occurrence and density of major nematode pests such as *meloidogyne* and *Helicotylenchus* may constrain tomato production in the country.

6. Recommendations

However, the economic importance of these nematodes in relation to yield loss and their impact on national production of these crops still remains unknown. There is need to establish the economic importance of the reported nematodes in Ethiopia as well as continued research for an effective plant parasitic nematode management strategy in tomato based cropping systems.

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