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OCCURRENCE AND ABUNDANCE OF PLANT PARASITIC NEMATODES IN CABBAGE-BASED CROPPING SYSTEMS IN KENYA

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ABSTRACT

A survey was conducted to determine the incidences of plant parasitic nematodes (PPN) and factors enhancing population build-up in cabbage (*Brassica oleracea* L. var *capitata*)-based cropping system in selected agro-ecological zones of Kenya. Samples were collected from sixty cabbage growing farms in Nyandarua and Embu Counties. Nematodes assay was conducted for both root and soil samples. Extraction of PPN from soil and root sample was done using modified extraction tray method and modified maceration extraction technique respectively. Extracted nematodes were enumerated, identified to genus level and their frequencies of occurrence and abundance determined. Nematodes belonging to various genera were found associated with cabbage in six agro-ecological zones. The study revealed that cabbage is a preferable host to lesion nematodes (*Pratylenchus*) with 87% and 58% frequency of occurrence in soil and roots, respectively. The mean population of this nematode in soil and root samples was significantly different from other nematode general. *Meloidogyne* spp. were present at low frequency of 42% and 23% in soil and roots and density 6.36/100 cm^3 of soil and 1.1/10 g roots. *Helicotylenchus* spp. were recovered in significant numbers with 82% frequency of occurrence in rhizosphere and 27% in roots and a density of 26.28/100 cm^3 in soils and less than 1 nematode per 10 g of roots. *Tylenchorhynchus* spp. occurred in 67% of the soil samples while *Paratrichodorus*, *Trichodorus* and *Paratylenchus* spp. were present in 78%, 57% and 58% frequency of occurrence in the cabbage rhizosphere but at a low population density. Other PPN detected in low frequencies of occurrence and density include; *Tylenchus, Scutellonema*, *Xiphinema* spp. among others. The study encourages more research work to establish the economic importance and the management of the reported nematode pests.

Keywords: Agro-Ecological Zones, cabbage (*Brassica oleracea*), root-knot nematode, *Pratylenchus* spp.

1. INTRODUCTION

White cabbage (*Brassica oleracea* L. Var. *capitata*) is a major vegetable crop in Kenya. It is a source of food to high urban and rural population in Kenya besides being an income earner to most Kenyans (Kungu, 2007). Cabbage is rich in vitamin A, B6, C and K in addition to some medicinal attributes (Fahey *et al*., 2001). Cabbage production is however constrained by various biotic and abiotic factors. According to Strange and Scott (2005) approximately 10% of global food production is lost to plant pathogens. Plant parasitic nematodes (PPN) form a group of pathogen that is not highly regarded yet it leads to an approximated loss of $ 125 billion worldwide annually (Chitwood, 2003). Given the nature of symptoms exhibited by PPN infestation, some of the losses caused by these pests are overlooked.

In order to satisfy the market demand for cabbage in Kenya, intensive and continuous cultivation systems favouring the build-up of PPN in the soil have been adopted. This predispose cabbage to PPN infestation and various secondary infection mainly caused by bacteria and fungi (Luc *et al*., 2005). For long, cabbage has been assumed to be resistant to most PPN (Pattison *et al*., 2006) due to glucosinolate compounds contained in their residues (Guerana, 2006). As such, no PPN management strategy in cabbage based cropping system has been designed. On the contrary, cabbage has been used as a rotational crop in the management of these soil pests especially in vegetable based cropping systems (Bello *et al*., 2004) as an alternative to use of nematicides. Most nematicides have been linked to various healthy and environmental problems leading to their withdrawal from market (Lilley *et al*., 2007). Establishing the occurrence and abundance of PPN associated with cabbage is therefore significant in evaluating the effectiveness of cabbage use in nematode management as well as designing a safe and widely acceptable alternative PPN management strategy in cabbage-based cropping systems in Kenya.

2. MATERIALS AND METHODS

A survey was conducted in the major cabbage growing areas of Nyandarua and Embu Counties of Kenya. Soil and root samples were collected from 60 cabbage growing farms distributed over five agro-ecological zones (AEZs) (Jaetzold *et al*., 2006; Jaetzold and Schimidt, 1983). These AEZs include upper highland one (UH1), upper highland two (UH2), upper highland three (UH3), upper highland four (UH4) and upper midland one (UM1). Ten soil and roots sub-samples were collected in each of the farms by gently digging out cabbage plant using a garden trowel to a depth of 20 cm. The sub-samples were pooled together and thoroughly but gently mixed and a composite sub-sample of about 1 kg drawn (Young, 1990).

Composite soil samples were put in plastic bags, sealed and then packed in cool box and transported to the laboratory for analysis. The samples were stored at approximately 4^{o} C during nematode extraction period that lasted for two weeks after collection. During nematode

extraction, each sample was thoroughly mixed and a 100 cm^3 sub-sample was collected for nematode extraction using modified Baermann funnel technique as described by Hooper *et al*., (2005). Extracted nematodes were enumerated using a stereomicroscope and identified to genus level based on morphological features as described by Hunt, *et al*., (2005), Siddiqi, (2000) and the University of Nebraska Lincoln nematode identification website (http://nematode.unl.edu/konzlistbutt.htm)

Root samples were washed and examined for galls. A sample of root was stained for egg masses according to Holbrook *et al*., (1983). Nematodes were also extracted from roots using a modified maceration and filtration technique according to Hooper *et al*., (2005) and thereafter enumerated and identified to genus levels at various magnifications as described earlier. Mean nematodes densities were compared using analysis of variance (ANOVA). Means that were significantly different ($P \leq 0.05$) were separated by LSD. Absolute frequency was determined using the following formula;

$$\text{Frequency of occurrence} = \frac{\text{Number of samples containing genera}}{\text{Number of samples collected}} \times 100$$

3. RESULTS

A total of eighteen (18) genera of plant parasitic nematodes were identified from the soil and the root samples in the six agro-ecological zones (Table-1).

Table-1. Plant parasitic nematodes genera isolated from soils and roots of cabbage in Nyandarua and Embu Agro-Ecological Zones.

Order	Sub-order	Family	Genus
Tylenchida	Tylenchina	Pratylenchidae	*Pratylenchus*
		Hoplolaimidae	*Helicotylenchus* *Hoplolaimus* *Rotylenchulus* *Peltamigratus* *Scutellonema*
		Meloidogynidae	*Meloidogyne*
		Criconematidae	*Hemicycliophora*
		Tylenchulidae	*Tylenchus* *Filenchus* *Coslenchus*
		Tylenchorhynchidae	*Tylenchorhynchus* *Quinisulcius*
		Paratylenchidae	*Paratylenchus*
Dorylaimida	Dorylaimina	Longidoridae	*Longidorus* *Xiphinema*
Triplonchida	Diphtherophorina	Trichodoridae	*Paratrichodorus* *Trichodorus*

Plant parasitic nematodes were detected in nearly every sample but at varying frequencies and densities. *Paratrichodorus* and *Filenchus* spp. were present in all AEZs at high frequencies. *Pratylenchus* and *Helicotylenchus* spp. occurred in high frequencies in five out of the six AEZs while *Hemicycliophora* and *Longidorus* occurred in 50% of the sample. Six nematode genera were present in both cabbage roots and soils, namely: *Pratylenchus* (lesion), *Helicotylenchus* (spiral), *Meloidogyne* (root knot), *Xiphinema* (dagger), *Trichodorus* (stubby), *Tylenchorhynchus* (stunt) and *Longidorus* (Needle).

Pratylenchus spp. were present at an absolute frequency 87% in the soil samples. This lesion nematode occurred in high frequency in nearly all sampled areas except in UM1 where it existed in slightly lower frequency. Fifty percent of the nematode genera isolated from soil samples had a high frequency of occurrence (Table-2).

Table-2. Frequency of occurrence and abundance of eight major plant parasitic nematode genera in soil samples from various Agro-Ecological Zones in Nyandarua and Embu County.

Nematode genera	Abundance†	Frequency of occurrence %
Pratylenchus	41.05±18.01[a]	87
Helicotylenchus	26.28±8.33[ab]	82
Filenchus	16.28±6.11[b]	88
Tylenchorhynchus	11.95±3.44[b]	67
Coslenchus	10.32±4.83[b]	60
Trichodorus	6.72±3.36[b]	57
Meloidogyne	6.38±2.92[b]	42
Paratylenchus	6.20±3.01[b]	58

Data are the mean ± standard error of nematodes per 10g of dry roots in sixty root samples collected in six AEZs.
†Means separated using Fisher's LSD test, means followed by the same letter are not significantly different

When the populations of different PPN genera in soil were evaluated for all AEZs, the population density ranged from 0.47 to 41 nematodes per 100 g of dry soil (Table-2). A significant difference ($P<0.05$) in the mean population densities of various nematode genera isolated from soils was noted. *Pratylenchus* had a high mean density of 41.05 nematodes/100g soil. The predominant species present were *Pratylenchus brachyurus*, *P. zeae*, and *P. neglectus* across all the AEZs.

Mean number of *Pratylenchus* was significantly higher than all other genera but not significantly ($P>0.05$) different from that of *Helicotylenchus*. Similarly, means for *Tylenchorhynchus*, *Paratrichodorus*, *Meloidogyne*, *Paratylenchus* and *Coslenchus* were not significantly ($P>0.05$) different (Table-2).

The sampled cabbage roots also tested positive for endoparasitic nematodes. Fifty eight percent of the total root samples collected had *Pratylenchus* spp. This genus was present in cabbage roots at a high frequency in most of the AEZs except for UH1a and UM1 that had lower frequencies. *Helicotylenchus* and *Meloidogyne* spp. were present in cabbage roots at a moderate frequency, of 27% and 23% respectively. *Tylenchorhynchus*, *Xiphinema*, *Longidorus* and *Trichodorus* spp. occurred at low frequencies of 12%, 8%, 5%, and 3%, respectively (Table-3).

Table-3. Abundance and Frequency of occurrence of plant parasitic nematode genera in root samples from various Agro-Ecological Zones in Nyandarua and Embu Counties.

Genera	Abundance†	Frequency of occurrence %
Pratylenchus	16.38±8.38[a]	58
Meloidogyne	1.10±0.76[b]	23
Helicotylenchus	0.70±0.35[b]	27
Xiphinema	0.42±0.38[b]	8
Tylenchorynchus	0.38±0.20[b]	12
Quinisulcius	0.13±0.08[b]	10
Trichodorus	0.08±0.07[b]	3
Longidorus	0.05±0.03[b]	5

Data are the mean ± standard error of nematodes per 10g of dry roots in sixty root samples collected in six AEZs.
†Means separated using Fisher's LSD test, means followed by the same letter are not significantly different

The general population density of PPN genera in roots of cabbage ranged from 0.1 to 16.38 per 10 g of dry roots with *Pratylenchus* spp. having significantly higher population density of 16.4 individual per 10 g of roots compared to the other genera. The root-knot nematode had a population density of 1.1 nematodes per 10 g of roots (based on J2) that did not differ significantly from other nematodes apart from *Pratylenchus*. Although *Helicotylenchus*, *Longidorus*, *Tylenchorhynchus*, *Quinisulcious*, *Trichodorus* and *Xiphinema* spp. were present in lower numbers with an average density of less

than one nematode per ten grams of roots, their means were not significantly (P>0.05) different (Table-3).

Population densities of various PPN genera from soils differed across all the AEZs. *Pratylenchus* was present in high density in UH1b. This density was significantly (P<0.05) higher than the other AEZs. Upper highland two (UH2) recorded population of *Meloidogyne* that was significantly higher (P<0.05) than other AEZs. The population of *Helicotylenchus* spp. also differed significantly across the five AEZs, with UH1a and UH4 having significantly higher populations. Variations in population densities for other PPN genera with AEZs are as indicated in Table-4.

Table-4. Occurrence of plant parasitic nematodes in soils from various Agro-Ecological Zones in Nyandarua and Embu Counties

	Mean nematodes density in 100 g of dry soil							
Nematode	**UH1a**	**UH1b**	**UH2**	**UH3**	**UH4**	**UM1**	**Overall Mean**	**F-probability**
Pratylenchus	14.1^{b}	125.7^{a}	43.2^{b}	31.9^{b}	31.0^{b}	0.4^{c}	41.1	**
Meloidogyne	6.9^{b}	4.5^{b}	20.2^{a}	1.2^{b}	0.8^{b}	4.7^{b}	6.4	**
Helicotylenchus	53.1^{a}	7.6^{bc}	17.1^{bc}	31.1^{ab}	45.6^{a}	3.2^{c}	26.3	*
Filenchus	43.8^{a}	13.9^{bc}	14.6^{bc}	3.6^{c}	19.1^{b}	2.7^{c}	16.2	*
Tylenchorhynchus	24.1^{a}	7.5^{bc}	7.7^{bc}	12.7^{ab}	18.8^{ab}	0.9^{c}	11.9	*
Coslenchus	29.3^{a}	1.8^{bc}	3.8^{bc}	6.5bc	20.5^{ab}	0.0^{d}	10.3	*
Trichodorus	0.7^{c}	14.5^{ab}	3.3^{bc}	0.4^{c}	19.6^{a}	1.8^{bc}	6.7	*
Paratylenchus	20.7^{a}	4.3^{b}	5.7^{b}	4.6^{b}	1.2^{b}	0.7^{b}	5.7	*

*Significant at p<0.01, ** Significant at p<0.05, Means followed by the same letter(s) along rows are not significantly different

Densities of PPN genera in roots differed significantly (P<0.05) across the AEZs with *Pratylenchus* spp. recording significantly (P<0.05) higher populations in UH1b compared with UH2, UH3, UH4 and UM1 that had lower densities (Table-5). Similarly, population density of *Meloidogyne* spp. differed significantly (P<0.05) across the AEZs, with UH3 recording the highest population which was significantly different from all other AEZs. The UH3 also recorded significantly higher densities of *Helicotylenchus* than any other AEZ. The UH1b recorded higher populations of *Xiphinema* than other AEZs. The population densities of other PPN genera isolated in cabbage roots did not differ significantly with AEZs (Table-5).

Table-5. Occurrence of plant parasitic nematodes in cabbage roots from various Agro-Ecological Zones in Nyandarua and Embu Counties

	Mean nematodes density in 10 g of dry roots							
Nematode	**UH1a**	**UH1b**	**UH2**	**UH3**	**UH4**	**UM1**	**Overall Mean**	**F- Probability**
Pratylenchus	4.8^{c}	56.0^{a}	7.6^{bc}	20.2^{b}	9.5^{b}	0.2^{c}	16.4	**
Meloidogyne	0.2b	0.3^{b}	0.5^{b}	4.9^{a}	0.1^{b}	0.6^{b}	1.1	*
Helicotylenchus	0.3^{b}	0.8^{b}	0.1^{b}	2.3^{a}	0.7^{b}	0.0^{b}	0.7	*
Xiphinema	0.0^{b}	2.3^{a}	0.0^{b}	0.2^{b}	0.0^{b}	0.0^{b}	0.4	*S
Longidorus	0.0^{a}	0.0^{a}	0.0^{a}	0.2^{a}	0.0^{a}	0.1^{a}	0.1	NS
Quinisulcius	0.1^{a}	0.5^{a}	0.0^{a}	0.0^{a}	0.1^{a}	0.1^{a}	0.1	NS
Tylenchorhynchus	0.0^{a}	0.2^{a}	0.0^{a}	0.0^{a}	0.08^{a}	0.10^{a}	0.4	NS
Trichodorus	0.0^{a}	0.4^{a}	0.1^{a}	0.0^{a}	0.0^{a}	0.0^{a}	0.1	NS

*Significant at p<0.01, ** Significant at p<0.05, NS: Not Significant Means followed by the same letter (s) along rows are not significantly different

DISCUSSIONS

This research has demonstrated that different genera of PPNs are widespread in cabbage growing fields in various AEZs of Nyandarua and Embu Counties. Extraction of diverse genera from the soil and roots sample in all the five AEZs indicates that cabbage is a potential host to PPN pests. The findings correlate with various other studies with the crop (Mennan and Handoo, 2006; Waceke, 2007). According to Bridge (1996) PPN are a major pests of vegetable crops which highly constrain the crop production.

Upper highland one agro-ecological zone (UH1a and b) recorded a higher density of PPN compared to the other AEZs studied. This can be attributed to the extensive farming system practised including intercropping cabbage with other crops such as maize, potatoes among other as noted during the survey. In addition, the study areas are potential agricultural zones that receive good amount of precipitation which favour rain fed agriculture most parts of the year (Jaetzold *et al*., 2006). This coupled with poor or lack of sound nematode management strategies may have created a favourable environment for nematodes to thrive. Upper midland one (UM1) on the other hand is slightly dry and not so suitable for agriculture. This may explain the low frequency and abundance of plant parasitic nematodes recorded in this zone (Jaetzold and Schmidt, 1983).

Pratylenchus was the most prevalent and abundant nematode in all the AEZs. This lesion nematode has a wide host range including cereals, legumes, fruits and vegetables (Hunt *et al*., 2005). The extensive nature of agriculture in the study area would therefore favour higher population of this nematode. *Pratylenchus* spp. is a common pest in agronomic settings and is responsible for significant yield losses worldwide. In Kenya for example, it is the most important nematode parasite of maize (Kimenju *et al*., 1998) and it has been identified in cabbage in Kenya (Waceke, 2007). In Turkey, Mennan and Handoo (2006) recorded presence of lesion nematode in cabbage while *Pratylenchus penetrans* has been shown to cause wilting and death of cabbage (Acedo and Rohde, 1968).

Root-knot nematode (RKN), *Meloidogyne*, was reported in low number and frequency in both root and soil samples. Root-knot nematode is a potential pest of cabbage having been reported in Uganda (Bafokuzara, 1996) Turkey (Mennan and Handoo, 2006) and Kenya (Waceke, 2007) among other places. Various species of *Meloidogyne* have been associated with crucifer family in various parts of the world (Potter and Oltholf, 1993; Mcsorley and Frederick, 1995; Liébanas and Castillo, 2004). However in this study RKN occurred at low frequency in soil and root samples while the screened cabbage roots had very small egg masses and tiny, inconspicuous galls. This may indicate the suppressive nature of cabbage to RKN as suggested by Bafokuzara (1983). Carneiro *et al*., (2000) found cabbage to be moderately susceptible to *M. javanica, M. incognita* and *M. hapla* but highly susceptible to *M. arenaria*

The *Helicotylenchus* spp. was second in frequency of occurrence in five of the study sites. Since this nematode has a wide host range, its higher presence was probably influenced by cropping history such as mixed cropping thus predisposing cabbage to higher levels of attack. *Helicotylenchus* spp. has been associated with cabbage in various parts of the world (Bafokuzara, 1996; Waceke, 2007).

Other nematodes were encountered at relatively lower frequency and density in various AEZs. Their low occurrence may indicate their poor performances in presence of other root nematodes (Blake, 1969). This could also be attributed to the ability of the cabbage to resist or tolerate nematode attack (Cook and Evans, 1997). Various crop varieties are said to possess varying degrees of resistance and tolerance to disease and pests (Trudgell, 1991). Cabbage for instance is known to produce several thiocyanates and isothiocyanates which have toxicity to certain nematode species (Zasada and Ferris, 2003). These glucosinolates in cabbage residues have suppressive effect (Guerana, 2006; Gardiner *et al*., 1999; Petersen *et al*., 2001) giving brassicas their natural fumigant potential against some nematodes, bacteria, fungi, insect and weeds (Clark, 2007). Different pathogens respond differently to these phytochemicals (Bais *et al*., 2006) leading to varied population in the crop rhizosphere as reported herein.

5. CONCLUSION AND RECOMMENDATIONS

Plant parasitic nematodes occur in cabbage based faming systems in Kenya. Higher occurrence and density of major nematode pests such as *Pratylenchus* and *Helicotylenchus* may constrain cabbage production in the country. There is need to establish the economic importance of the reported nematodes in Kenya as well as continued search for an effective PPN management strategy in cabbage based cropping systems.

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