

Full Length Research Paper

Occurrence and distribution of soil nematodes in cotton (*Gossypium hirsutum* L.) production areas of Kenya

Karuri, H. W^{1,2*}, Amata, R.¹, Amugune, N.² and Waturu, C.³

¹National Agricultural Research Laboratories, Kenya Agricultural Research Institute, P.O Box 14733-00800, Nairobi, Kenya.

²School of Biological Sciences, University of Nairobi, P.O. Box 30197-00100, Nairobi, Kenya.

³Horticultural Research Centre, Kenya Agricultural Research Institute, P.O. Box 220, Thika, Kenya.

Accepted 24 June, 2010

A baseline survey was conducted to determine the occurrence and distribution of soil nematodes associated with cotton in major growing areas in Kenya. Such baseline data on soil nematode abundance, diversity and ecosystem function in cotton ecosystems are valuable in providing a basis for comparison with organisms from transgenic cotton fields. Transgenic cotton plants expressing Cry1Ac and Cry2Ab proteins, from the soil bacterium *Bacillus thuringiensis* (Bt), provide effective control of lepidopteran pests. However, the potential effects of these proteins on soil nematofauna are unknown in Kenya. Soil samples were collected from nine locations of western (Odiado, Angorom and Ochundo locations), coast (Baharini, Mpeketoni and Witu locations) and central (Kajiji, Tebere and Nyangati locations) Province. Nematodes were extracted and recovered from soil samples using the Whitehead and Hemming tray method and identified under a light microscope according to their morphological characters. They were classified according to their feeding habits. Twenty seven genera of plant parasites, bacteriovores, fungivores, predators and omnivores were identified. Bacterial, fungal feeding and parasitic nematodes were the most abundant trophic groups across all Provinces. There were significant differences in the numbers of bacteriovores ($P \leq 0.01$) and plant parasites ($P \leq 0.05$) between the Provinces but no difference was observed in the numbers of fungal feeding nematodes. There was a significant difference in genus richness within locations in western and coast Provinces ($P \leq 0.001$). The combined maturity index ($\sum MI$) did not vary significantly within the locations. The Shannon index (H') showed variations within locations in western ($P < 0.001$) and coast Province ($P \leq 0.01$). Soil texture, P and K were correlated with abundance of some nematode genera. The bacteria feeders, *Acrobeles* and *Rhabditis* showed positive correlations to K ($r = 0.592$, $P < 0.05$ and $r = 0.128$, $P \leq 0.05$) and P ($r = 0.406$, $P \leq 0.05$, and $r = 0.252$, $P < 0.05$) while *Aphelenchus* was positively correlated to P ($r = 0.375$, $P \leq 0.05$). The plant parasitic genera *Meloidogyne* and *Pratylenchus* showed significant negative correlation to N ($r = -0.513$, $P \leq 0.05$ and $r = -0.226$, $P \leq 0.05$). It is clear from this baseline data that plant parasitic and free living nematodes are widespread in cotton fields and any potential effects of Bt cotton on these nematodes may affect the nematode community structure and their ecosystem functions.

Key words: Cotton, soil nematodes, survey.

INTRODUCTION

Soil species and their interactions can influence different

ecosystem processes. Nematodes are abundant, diverse in all soils and participate in many functions at different levels of the soil food web (Yeates, 1979). Bacterial and fungal feeding nematodes release a large percent of nitrogen when feeding on their prey groups and are thus

*Corresponding author. E-mail: hkwangari@yahoo.com.

responsible for much of the plant available nitrogen in majority of soils (Bongers and Bongers, 1998). Cotton fields contain high nematode diversity that is influenced by cultural practices including tillage, use of pesticides and fertilizers (Koenning and Barker, 2004). In Kenya, cotton is mainly grown in the semi-arid regions of Eastern, Central, Nyanza, Coast, Western and Rift valley Provinces. It is an important fibre crop that provides a source of income to farmers and fibre to the textile industries. The seeds provide an important source of oil and feed cake for humans and livestock (Lutrell et al., 1994). For decades now, cotton production in Kenya has been characterized by low yields. Pest management in cotton accounts for about 57% of the total production cost (Ikiara and Ndirangu, 2003). Considering the environmental and human health concerns associated with high pesticide usage on cotton, there has been a strong need for more integrated approaches to pest management that minimize pesticide requirement. Transgenic crops expressing insecticidal protein genes from *Bacillus thuringiensis* (Bt) represent one new approach for integrated pest management (IPM) of cotton (Fitt, 2003; Wilson et al., 2004).

Concerns about the impacts of genetically modified crops on soil biota have been raised, in part because of the chemical and biological properties of soil (McGregor and Turner, 2000). Soil materials have large sorptive capacities for biological molecules, including insecticidal bacterial proteins and DNA. Laboratory studies (Saxena and Stotzky, 2001; Saxena and Stotzky 2002; Tapp and Stotzky, 1998) have shown that insecticidal Cry proteins from Bt are readily adsorbed and bound to clay minerals and humic acids, and they persist in soil. Consequently, the issue of the impact of Bt proteins released to soil from roots and biomass of Bt crops on soil nematodes is an important one. Transgenic plants produce and release relatively large amounts of a variety of novel proteins, including the active toxins. Microbial processes have been shown to be particularly responsive to protein substrates (Wheatley et al., 2001). Changes in the root exudates of a transgenic plant may significantly alter the rhizosphere community associated with it (Bruseti et al., 2004). Bt proteins are present as active toxins in most of the cells of a Bt transformed plant and so are present in all plant residues, and may be released into the soil through various routes depending on the crop and environment (Mendonca et al., 2006). A baseline study to document species diversity and abundance for a particular ecosystem is crucial before cultivation of transgenic plants. Lack of baseline information on soil fauna, found in an agro-ecosystem, to compare with microorganisms in ecosystems containing transgenic crops have been cited as a major problem in evaluating the impact of transgenic crops on soil microbial diversity (Dale et al., 2002). Therefore, documentation of nematodes found in cotton ecosystems in different agro-ecological zones was carried out to provide a basis for comparison with nematodes from cotton fields cultivated with Bt cotton.

MATERIALS AND METHODS

Soils were sampled from cotton growing fields in nine locations of Western (Odiado, Angorom and Ochundo locations), Coast (Baharini, Mpeketoni and Witu locations) and Central (Kajiji, Tebere and Nyangati locations) Provinces of Kenya. Two composite samples (5 cm diameter, 30 cm deep) were independently collected per field. Two linear transects were randomly and diagonally located along rows of crops and composite samples were collected along these transects (Neher, 1999). The samples were thoroughly mixed and 200 ml of each soil samples was used for nematode extraction using Whitehead and Hemming tray technique. Nematode numbers were counted and identification to genus level was done under a compound microscope at a magnification of $\times 400$ - $\times 1000$.

Nematode genera were assigned to trophic groups (bacterial and fungal feeders, plant parasites, omnivores and predators) as described by Yeates et al. (1993). Taxonomic groups were also assigned to colonizer-persistor (cp) values according to Bongers (1990). Physical and chemical properties of soil were measured. Total nitrogen (N) was measured through Kjeldahl method. Total organic carbon (C) was estimated through calometric method. Phosphorous (P) was analyzed by colometry, potassium (K) and sodium (Na) by flame emission spectrophotometry, and calcium (Ca) and magnesium (Mg) by atomic absorption spectro-photometry (Gallaher et al., 1975). Cation exchange capacity (Jackson, 1958), pH, electrical conductivity (Smith and Doran, 1996) and soil texture (Gee and Bauder, 1985) were also analyzed.

The following nematode parameters were computed: Genus richness index ($d=S-1 \log N$), where S = number of genera and N = total number of nematodes, abundance and Shannon Wiener's diversity index ($H'=-\sum p_i \log_2 p_i$). Maturity index for all free living nematodes except taxa with cp value of 1 (MINO), combined maturity index for free living and plant parasitic nematodes ($\sum MI$) and ratio of fungivorous to bacterivorous nematodes was also computed. Nematode data based on relative abundance of genera was subjected to analysis of variance. Relationships between soil chemical properties and nematode genera/indices were derived using correlation analysis. All statistical computations were performed using XLSTAT version 7.5 (Addin software, New York).

RESULTS

Twenty seven genera were identified in nine locations of Western (Odiado, Angorom and Ochundo locations), Coast (Baharini, Mpeketoni and Witu locations) and Central (Kajiji, Tebere and Nyangati locations) Province. The plant parasitic genera identified were *Meloidogyne*, *Pratylenchus*, *Trichodorus*, *Helicotylenchus*, *Rotylenchus*, *Hoplolaimus*, *Xiphinema*, *Tylenchus*, *Filenchus*, *Longidorus*, *Tylenchorhynchus* and *Scutollenema*, the bacteriovores consisted of *Acrobeles*, *Rhabditis*, *Cervidellus*, *Eucephalobus*, *Cephalobus*, *Heterocephalobus*, *Plectus*, *Wilsonema* and *Tylocephalus*. Two fungal genera (*Aphelenchus* and *Aphelenchoides*) were recorded. Nematodes from higher trophic groups in the genera *Labronema*, *Chromadora* and *Prodorylaimus* were also identified (Table 1). Only one genus (*Mononchus*) of predatory nematodes was observed.

Bacterial, fungal feeding and parasitic nematodes were the most abundant trophic groups across all Provinces. There were significant differences in the numbers of bacteriovores ($P \leq 0.01$) and plant parasites ($P \leq 0.05$)

Table 1. Nematode genera, cp values and abundance in Central, Coast and Western Province.

Nematode genera	cp value	Central Province	Coast Province	Western Province	ANOVA		
					Province	Genera	Province×Genera
Bacterial feeders							
<i>Acrobeles</i>	2	190.125	97.500	29.083			
<i>Cephalobus</i>	2	360.750	224.250	.000			
<i>Cervidellus</i>	2	124.313	372.750	.000			
<i>Eucephalobus</i>	2	124.313	.000	.000			
<i>Heterocephalobus</i>	2	131.625	338.813	.000			
<i>Plectus</i>	2	.000	4.875	.000			
<i>Rhabditis</i>	1	34.125	24.375	50.000			
<i>Tylocephalus</i>	2	19.500	87.292	.000			
<i>Wilsonema</i>	2	29.250	.000	.000			
Mean number of bacterial feeders		112.660	127.76	8.79	**1	**	ns
Fungal feeders							
<i>Aphelenchoides</i>	2	58.500	282.750	4.167			
<i>Aphelenchus</i>	2	168.188	173.063	31.250			
Mean number of fungal feeders		113.344	227.906	17.708	ns	ns	ns
Plant parasites							
<i>Filenchus</i>	2	7.313	.000	.000			
<i>Helicotylenchus</i>	3	319.000	309.667	87.500			
<i>Hoplolaimus</i>	2	33.000	180.000	104.167			
<i>Longidorous</i>	5	17.063	.000	.000			
<i>Meloidogyne</i>	3	112.917	191.208	68.750			
<i>Pratylenchus</i>	3	97.667	75.042	91.667			
<i>Rotylenchus</i>	3	18.667	346.000	91.667			
<i>Scutellonema</i>	3	225.000	320.000	56.250			
<i>Trichodorous</i>	4	71.667	139.333	39.583			
<i>Tylenchorhynchus</i>	2	12.188	.000	.000			
<i>Tylenchus</i>	2	80.667	67.667	116.667			
<i>Xiphinema</i>	5	50.333	10.000	97.917			
Mean number of plant parasites		87.123	136.576	62.847	*	***	ns
Omnivores							
<i>Chromadora</i>	3	48.750	.000	20.833			
<i>Labronema</i>	4	104.813	143.813	.000			
<i>Prodorylaimus</i>	5	7.312	58.500	.000			
Mean number of omnivores		53.625	67.438	6.944	ns	ns	ns

1*, **, indicate significance at $P < 0.05$ and $P < 0.01$, respectively; ns = not significant.

^aColonizer-persister scale (cp1-5) describing life history strategy of nematodes.

between the Provinces but no difference was observed in the numbers of fungal feeding nematodes. In western Province the common genera were *Acrobeles*, *Rhabditis*, *Aphelenchoides*, *Aphelenchus*, *Meloidogyne*, *Pratylenchus*, *Trichodorous*, *Helicotylenchus*, *Rotylenchus*, *Hoplolaimus*, *Xiphinema*, *Tylenchus*, *Scutellonema* and *Mononchus*. Similar genera were found in coast and central Province except *Mononchus*. In addition, the bacteria feeding genera, *Cervidellus*, *Eucephalobus*, *Cephalobus*, *Heterocephalobus* *Plectus* and *Tylocephalus* were also identified in the two Provinces. The genus *Wilsonema*

was only found in central Province. Omnivorous nematodes from the genera *Chromadora* were identified in western and central Province while *Labronema* and *Prodorylaimus* were present in coast and central Provinces (Table 1).

There was a significant difference in genus richness within locations in western and coast Provinces ($P \leq 0.001$). The Shannon index (H') showed variations within locations in western province ($P \leq 0.001$) and coast province ($P \leq 0.01$). The combined maturity index (ΣMI) did not show significant variation within locations.

Table 2. Nematode richness, abundance and diversity indices in western, central and coast province.

	Western province			Central province			Coast province		
	Odiado	Angorom	Ochundo	Tebere	Nyangati	Kajiji	Witu	Mpeketoni	Baharini
Genus richness	4.925±0.095 ^b	5.38±0.067 ^a	5.2±0.051 ^a	5.228±0.142	5.637±0.142	4.976±0.201	5.05±0.177 ^a	4.128±0.177 ^b	4.119±0.102 ^b
Abundance	172±27.823	262.5±19.674	221.43±14.872	739.875±56.963	769.109±56.963	652.594±80.559	934±54.037 ^a	807±54.037 ^{ab}	680.50±31.198 ^b
MINO	0.395±0.111	0.5975±0.078	0.396±0.059	1.263±0.146	1.343±0.146	1.310±0.207	1.405±0.123	1.125±0.123	0.93±0.071
Combined MI	3.225±0.231	3.238±0.163	2.964±0.124	2.480±0.084	2.508±0.084	2.620±0.119	2.695±0.0128	2.415±0.0128	2.723±0.074
F/B+F	0.415±0.084	0.35±0.060	0.336±0.045	0.273±0.043 ^a	0.165±0.043 ^a	0	0.225±0.124	0.383±0.124	0.242±0.072
Shannon index	2.34±0.018 ^c	2.498±0.012 ^a	2.431±0.009 ^b	2.525±0.037	2.428±0.037	2.385±0.052	2.64±0.065 ^a	2.365±0.065 ^b	2.325±0.038 ^b

NB: Means followed by different letters along the row for each Province are significantly different ($P \leq 0.05$).

Nematode abundance, richness and H' varied significantly within coast Province (Table 2.). All Provinces had high $\sum MI$ which did not significantly vary across locations. Nyangati had the highest genus richness (5.64) while the lowest was in Baharini (4.12). MINO varied across locations in the 3 Provinces with Witu having the highest value. The lowest value of F/B+F was in Kajiji and the highest H' was found in Witu (Figure 1). There were differences in soil properties across the Provinces (Table 3). Soil texture, Phosphorous (P) and Potassium (K) were correlated with abundance of some nematode genera. *Acrobelles* showed strong positive correlations to K ($r = 0.592$, $P \leq 0.05$) while *Rhabditis* exhibited weak positive correlation ($r = 0.128$, $P \leq 0.05$). The two genera showed weak positive correlation to P (for *Acrobelles* and $r = 0.406$, $P \leq 0.05$ and $r = 0.252$, $P \leq 0.05$ for *Rhabditis*). There was a weak positive correlation between and P ($r = 0.375$, $P \leq 0.05$). The plant parasitic genera *Meloidogyne* and *Pratylenchus* showed significant negative correlation to nitrogen ($r = -0.513$, $P \leq 0.05$ and $r = -0.226$, $P \leq 0.05$). A strong positive correlation was also found between $\sum MI$ and silt content ($r = 0.666$, $P \leq 0.05$).

DISCUSSION

Nematodes in an agro-ecosystem are potential bio-indicators due to their diversity and their relationships to soil processes (Yeates and Bongers, 1999). The nematode community structure can be used to assess changes in soil since these organisms respond rapidly to new resources (Bongers and Bongers, 1998). Cotton fields in different regions had different abundances of nematode functional groups. Common genera of parasitic nematodes associated with cotton were identified across the Provinces, which is in agreement with the findings of Lawrence and Mclean (1995). The root knot nematode genus, (*Meloidogyne*) that causes serious yield losses in cotton, was found in all the regions surveyed indicating the need for nematode management strategies in cotton. Bacteria and fungal feeding nematodes indirectly affect primary production due to their role as key intermediaries in the decomposition process and nutrient cycling (Ruess and Ferris, 2004). Bacteriovores were among the dominant trophic groups present in all the Provinces. The genera were mainly in the cp 2 category which is associated with dry conditions

in the soil environment (Griffiths et al., 1995). Large numbers of fungal feeders were also identified and this could be an indication of decomposition of substrates with high C : N ratios by fungi. The high abundance of fungal and bacterial feeding nematodes could also be attributed to colonization of cotton roots by large numbers of bacteria and fungi (Hallman et al., 1999; Feng et al., 2003; Koenig and Barker, 2004). Predators and omnivores which are at a higher trophic level were not abundant in the surveyed areas. Freckman and Ettema (1993) associated low numbers of these groups of nematodes to disturbances in agro-ecosystems such as use of fertilizers and cultivation.

The number of genera in a soil habitat is a reflection of its biodiversity (Ou et al., 2005). Variation in nematode diversity shown by values of H' is an indication of environmental disturbances (Yeates and Bongers, 1999). Genus richness, MINO and H' values in cotton fields were comparable to values in other agricultural systems in Kenya (Kimenju et al., 2009). Nematode genus richness varies across soil ecosystems and an abundant food source at the rhizosphere accommodates great richness (Macchia et al., 2003). The

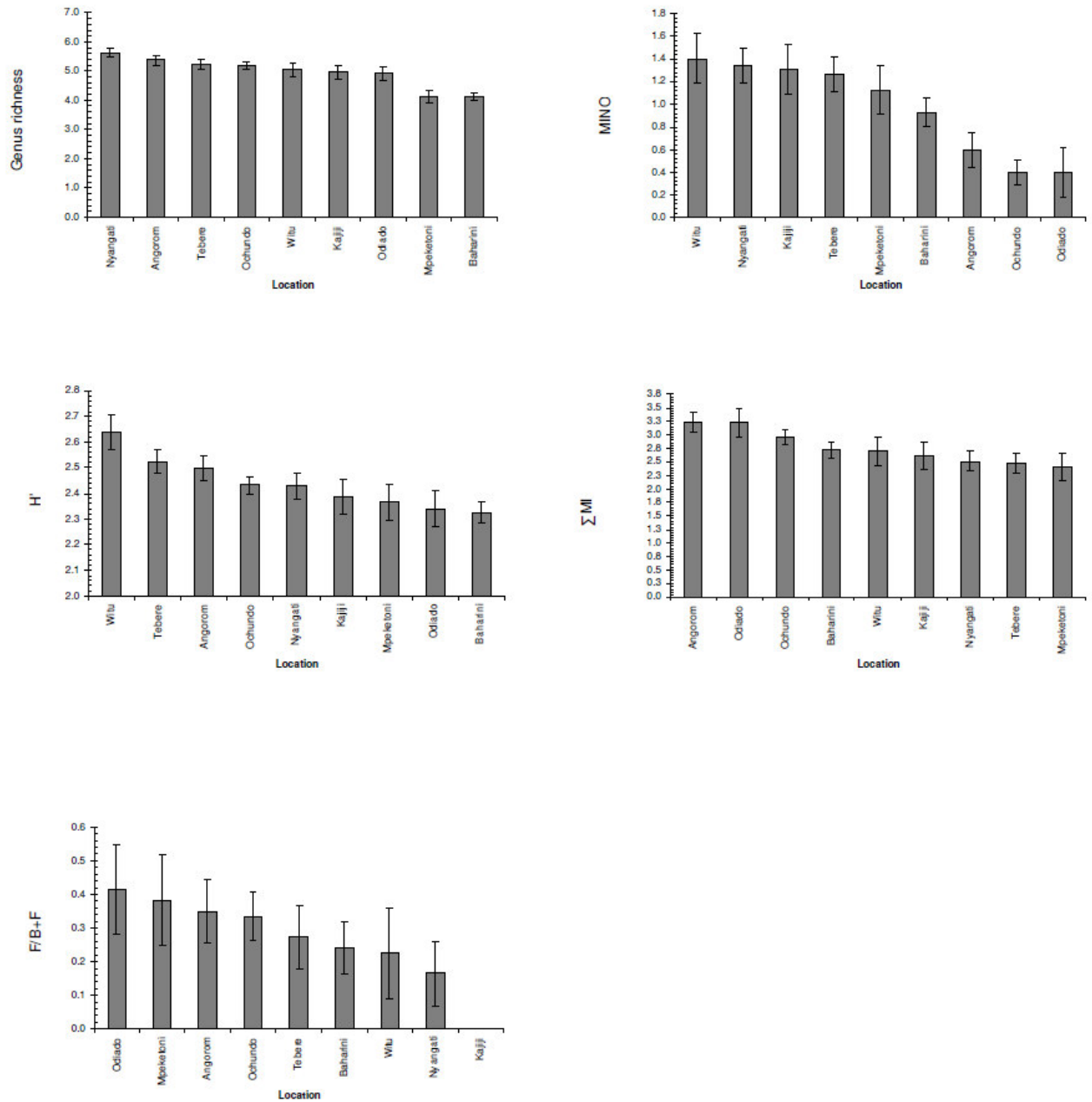


Figure 1. Nematode richness and indices in cotton fields from different locations of western, central and coast Province. **NB:** MINO-Maturity index with no ones; H'- Shannon Wiener's diversity index; ΣMI-Combined maturity index; F/B+F-Fungivores:Bacteriovores ratio.

ecological measure MI is important in assessing the status of soil food webs in terrestrial ecosystems (Yeates et al., 1999; Neher et al., 2005). The soil food web

condition can also be assessed using ΣMI. High values of ΣMI in this study were realized due to the inclusion of plant feeding nematodes in the calculation (Zuckerman

Table 3. Physical and chemical properties of soil samples collected from different locations in western, central and coast province.

Soil properties	Western Province			Central Province			Coast Province		
	Odiado	Angorom	Ochundo	Tebere	Nyangati	Kajiji	Baharini	Witu	Mpeketoni
pH	6.7	5	5	5.83	5.54	5.09	6.18	6.88	7.05
Total nitrogen (%)	0.16	0.12	0.09	0.09	0.11	0.11	0.05	0.19	0.09
Phosphorous (ppm)	130	30	24	179	19	24	31	204	32.5
Electrical conductivity (*mS/cm)	0.2	0.1	0.1	0.08	0.08	0.06	0.06	0.13	0.12
Carbon (%)	1.3	0.8	0.7	0.9	0.7	0.7	0.3	1.1	0.4
Sand (%)	20	20	56	8	2	4	90	70	86
Silt (%)	28	58	24	20	24	16	0	6	2
Clay (%)	52	22	20	72	74	80	10	30	12
Texture class	Clay	Clay	Sandy clay	Clay	Clay	Clay	Sandy	Sandy clay loam	Loam sandy
Calcium (**me%)	7.8	4.1	1.5	7.5	5.2	2.4	3.1	14.2	5.6
***CEC (me %)	20	23.7	19.3	19.2	14	6.8	3.5	12.4	3.1
Magnesium (me%)	2.5	1.3	0.5	2.3	1.4	0.8	0.1	1.1	0.5
Potassium (me%)	1.8	0.2	0.3	2.7	1.3	0.9	0.4	1.2	0.6
Sodium (me%)	0.8	0.6	0.6	0.2	0.1	0.2	0.4	0.5	0.5

*mS/cm-millisiemen/centimeter; **me-milliequivalents. ***CEC- Cation exchange capacity.

and Coleman, 2007). However, there was no variation in Σ MI across the Provinces. Neher (1999) also reported a lack of variation in the Σ MI across conventionally and organically managed soils. The highest F/B+F ratio was in Odiado (Western Province) and lowest in Kajiji. Fungivore: bacteriovore ratios provided information on detrital pathways in soils (Zuckerman and Coleman, 2007) and can also be used as an index reflecting the microbial community structure (Yeates et al., 1993). The F/B+F ratios in this study were within the range reported for agricultural ecosystems. Agro-ecosystems have an inter-mediate fungivore to bacteriovore ratios (0.38) compared to grasslands (0.19) and forests (0.74) (Wasilewska, 1979). The low F/B+F ratio in Kajiji may be an indication of a bacteria-based food web which is characterized by higher decomposition rates than fungi-based food webs (Porazinska and Coleman, 1995). Crop species, use of fertilizers and herbicides may also influence the F/B+F ratio (Neher, 1999).

Soil physical characteristics, different nutrients and elements influence the occurrence, distribution and population dynamics of nematodes (Norton et al., 1971; Mcsorley, 1998; Wang et al., 2004). Significant positive correlations were found between K and the bacteriovores, *Acrobeles* and *Rhabditis* while the fungal feeder *Aphelenchus* was positively correlated to P. Similar results were obtained by Ingham et al. (1985) and Wang et al. (2004) and they attributed the correlations to high levels of nutrient mineralization that occur where large numbers of free living nematodes are present (Ferris et

al., 1998). Negative correlations between plant parasitic genera and soil nutrients may be due to the fact that plant roots may have taken up nutrients from the soil (Wang et al., 2004). Correlations between nematode genera and soil physical characteristics in Kenya have been documented (Kandji et al., 2001; Chirchir et al., 2008). Soils with sand, silt or loam texture have different nematode composition and maturity indices (Yeates et al., 1997). Although some soil physico-chemical properties are correlated with nematode community structure and indices, there are other factors such as season and type of crop that may influence soil quality and ultimately the nematode composition (Norton et al., 1971; Mendoza et al., 2008).

Parasitic nematodes that cause damage to cotton (Gazaway and Mclean, 2003) and microbial grazers that regulate rates of decomposition (Yeates and Coleman, 1982) and nutrient mineralization were identified in the cotton fields surveyed. Crops affect the quality of resources that enter the soil through supply of organic substrates from sloughing off plant cells, root exudates and aging root epidermis (Nguyen, 2003). These substrates consequently influence the community composition (Wardle et al., 2003). Genetically modified plants offer various benefits over conventional crops; however their products may have potential adverse effects on soil organisms (Rui et al., 2005). Bt toxins engineered into plants could persist, accumulate and retain their insecticidal activity due to their binding to humic acids and clay particles (Tapp and Stotzky, 1998). This could be

hazardous to non target organisms such as nematodes. Transgenic plants have been reported to have no toxic effects to nematodes (Saxena and Stotzky, 2001) but there are Bt crystal proteins that have been shown to contain nematicidal properties (Wei et al., 2003). Manachini and Lozzia (2002) reported a decrease in bacteriophagous nematodes and an increase in mycophagous nematodes in Bt maize field trials in Italy. There are no reports on the effects of Bt cotton on soil nematodes in Kenya and it is therefore important to evaluate the potential risks of its release to the environment. Post release monitoring should also be done once Bt cotton is commercially cultivated in order to detect and prevent adverse effects on soil nematodes. Despite these concerns, Bt crops such as cotton have the potential to significantly increase yield and reduce pesticide use if the technology is applied in an environmentally acceptable and sustainable manner.

ACKNOWLEDGEMENTS

This is publication No. 18 of the BiosafeTrain project funded by the Danish International Development Agency (DANIDA). The authors would like to acknowledge the Kenya Agricultural Research Institute, National Agricultural Research Laboratories (KARI-NARL) for providing laboratory space and equipments. We also thank Maurice Okomo and Mary Ndunguli for technical support and Elias Thurairira for assistance in data analysis.

REFERENCES

- Bongers T (1990). The maturity index: An ecological measure of environmental disturbance based on nematode species composition. *Oecology*, 83: 14-19.
- Bongers T, Bongers M (1998). Functional diversity of nematodes. *Appl. Soil Ecol.*, 10: 239-251.
- Bruseti L, Fracia P, Bertolini C, Pagliuca AS (2004). Bacterial communities associated with the rhizosphere of transgenic Bt 176 maize and its non-transgenic counterpart. *Plant Soil*, 266: 11-21.
- Chirchir A, Kimenju JW, Olubayo FM and Mutua GK (2008). Abundance and distribution of plant parasitic nematodes associated with sugarcane in Western Kenya. *As. J. Pl. Path.*, 2: 48-53.
- Crecchio C, Stotzky G (1998). Insecticidal activity and biodegradation of the toxin from *Bacillus thuringiensis* subsp *kurstaki* bound to humic acids from soil. *Soil Biochem.*, 30: 463-470.
- Dale PJ, Clarke B, Fontes EM (2002). Potential for the environmental impact of transgenic crops. *Nature Biotech.*, 20: 567-574.
- Feng Y, Motta AC, Reeves DW, Burmester CH, Santen van E, Osborne JA (2003). Soil microbial communities under conventional-till and no-till continuous cotton systems. *Soil Biol. Biochem.*, 35: 1693-1703.
- Ferris HR, Venette C, van der Meulen HR, Lau SS (1998). Nitrogen mineralization by bacterial-feeding nematodes: Verification and measurement. *Plant Soil*, 203: 159-171.
- Fitt GP (2003). Deployment and impact of transgenic Bt cotton in Australia. In: *The Economic and Environmental Impacts of Agbiotech: A Global Perspective* Ed. G Kalaitzandonakes Kluwer NY. pp. 141-164.
- Freckman DW, Ettema CH (1993). Assessing nematode communities in agroecosystems of varying human intervention. *Agric. Ecosys. Environ.*, 45: 239-261.
- Gallaher RN, Weldon C, Futral J (1975). An aluminium block digester for plant and soil analysis. *Soil Sci. Soc. Am. Proc.*, 39: 803-806.
- Gazaway WS, Mclean K (2003). A survey of plant parasitic nematodes associated with cotton in Alabama. *J. Cotton Sci.*, 7: 1-7.
- Gee GW, Bauder J (1985). Particle size analysis, pp 385-411 in *A Kluteed Methods of soil analysis part 1 Agronomy Monograph No 9* 2nd ed. Madison WI: Am. Soc. of Agr.,
- Griffiths BS, Young IM, Caul S (1995). Nematodes and protozoan population dynamics on decomposing barley leaves incubated at different soil matrix potentials. *Pedobiology*, 39: 454-461.
- Hallman J, Kabana-Rodriguez R, Kloepper JW (1999). Chitin-mediated changes in bacterial communities of the soil rhizosphere and within roots of cotton in relation to nematode control. *Soil Biol. Biochem.*, 31: 551-560.
- Ingham RE, Trofymow JA, Ingham ER, Coleman DC (1985). Interactions of bacteria fungi and their nematode grazers: Effects on nutrient cycling and plant growth. *Ecol. Monogr.*, 55: 119-140.
- Ikiara MM, Ndirangu LK (2003). Developing a revival strategy for Kenya's cotton textile industry. A value chain approach KIPPRA Working paper Number 8. Kenya Institute for Public Policy Research and Analysis (KIPPRA) Nairobi Kenya, p. 104.
- Jackson ML (1958). *Soil chemical analysis*. Prentice hall englewood Cliffs NJ, USA.
- Kandji S, Callistus K, Ogol P, Albrecht A (2001). Diversity of plant parasitic nematodes and their relationships with some soil physico-chemical characteristics in improved fallows in western Kenya. *Appl. Soil Ecol.*, 18: 143-157.
- Kimenju JW, Karanja NK, Mutua GK, Rimberia BM, Wachira PM (2009). Nematode community structure as influenced by land use and intensity of cultivation. *Trop. Subtrop. Agroecosys.*, 11: 353-360.
- Koenning SR, Barker KR (2004). Influence of litter applications on nematode communities in cotton agroecosystems. *J. Nematol.*, 36: 524-533.
- Lawrence GW, Mclean KS (1995) Distribution of nematodes. In *Proc. Beltwide Cotton Prod. Res. Conf.*, San Antonio tx 4-7 Jan (1995). Natl. Cotton Council Am Memphis TN. p. 196.
- Luttrell RG, Fitt GP, Ramatho FS, Sugonyaeu ES (1994). Cotton pests management. A worldwide perspective. *Annual Rev. Entomol.*, 39: 517-526.
- Macchia ET, McSorley R, Duncan LW, Syversten JS (2003). Effects of perennial peanut (*arachis glabarata*) ground cover on nematode communities in citrus. *J. Nematol.*, 35: 450-457.
- Manachini B, Lozzia G (2002). First investigations into the effects of Bt. corn crop on Nematofauna. *Boll. Zool. Agr. Bachic. Ser.II*, 34: 85-96.
- Mendonça Hagler LC, Melo IS de, Valadares-Inglis MC, Anyango BM, Siqueira JO, Toan Pham Van, Wheatley RE (2006). Non-target and biodiversity impacts in soil. In Hilbeck, A., Andow, D.A, Fontes, E.M.G. (eds.) *Environmental Risk Assessment of Genetically Modified Organisms Volume 2: Methodologies for Assessing Bt Cotton in Brazil*. CABI Publishing, Wallingford, UK. pp. 225-260.
- Mendoza R, Franti TG, Doran J, Powers T, Zanner C (2008). Tillage effects on soil quality indicators and nematode abundance in loessial soil under long term no till production. *Communications Soil Sci. Plant Anal.*, 39: 2169-2190.
- McGregor AN, Turner MA (2000). Soil effects of transgenic agriculture: biological processes and ecological consequences. *NZ Soil News*, 48: 166-169.
- McSorley R (1998). Population dynamics. In Barker KR, Pederson GA, Windham GL (eds.) *Plant and nematode interactions American society of Agronomy Inc Madison WI USA*. pp. 109-133
- Norton DC, Frederick R, Ponchilla E, Nyhan JW (1971). Correlations of nematodes and soil properties in Soybean fields. *J. Nematol.*, 3: 154-163.
- Neher DA (1999). Nematode communities in organically and conventionally managed agricultural soils. *J. Nematol.*, 31:142-154.
- Neher DA, Wub J, Barbercheck ME, Anas O (2005). Ecosystem type affects interpretation of soil nematode community measures. *Appl. Soil Ecol.*, 30: 47-64.
- Nguyen C (2003). Rhizodeposition of organic C by plants: Mechanisms and controls. *Agronomie*, 23: 375-396.
- Ou W, Liang W, Jiang Y, Li Q, Wen D (2005). Vertical distribution of soil nematodes under different land use types in aquatic brown soil.

- Pedobiology, 49: 139-148.
- Porazinska D, Coleman D (1995). Ecology of nematodes under influence of *Cucurbita* spp. And different fertilizer types. Supp. J. Nematol., 27: 617-623.
- Ruess L, Ferris H (2004). Decomposition pathways and successional changes. Nematol. Monographs Perspect., 2: 547-556.
- Rui YK, Yi GX, Zhao J, Wang BM, Li ZH, Zhai ZX, He ZP, Li QX (2005). Changes of Bt toxin in the rhizosphere of transgenic Bt cotton and its influence on soil functional bacteria. World J. Microbiol. Biotechnol., 21: 1279-1284.
- Saxena D, Stotzky G (2001) *Bacillus thuringiensis* (Bt) toxin released from root exudates and biomass of Bt corn has no apparent effect on earthworms nematodes protozoa bacteria and fungi in soil. Soil Biol. Chem., 33:1225-1230.
- Saxena D, Stotzky G (2002). Bt toxin is not taken up from soil or hydroponic culture by corn carrot radish or turnip. Plant Soil., 239: 165-172.
- Smith J, Doran J (1996). Measurement and use of electrical conductivity for soil quality analysis pp. 169-185 in Doran JW, Jones AJ eds. Methods for assessing soil quality. Soil science society of America special publication Madison WI: Soil science society of America.
- Tapp H, Stotzky G (1998). Persistence of the insecticidal toxins from *Bacillus thuringiensis* subsp kurstaki in soil. Soil Biol. Biochem., 30: 471-476.
- Wang KH, McSorley R, Gallaher R (2004). Relationship of soil management history and nutrient status to nematode community structure. Nematropica, 34: 84-93.
- Wardle D, Yeates G, Williamson W, Bonner K (2003). The response of three trophic level soil food web to the identity and diversity of plant species and functional groups. Oikos, 102: 45-56.
- Wasilewska L (1979). The structure and function of soil nematode communities in natural ecosystems and agrocenoses. Pol. Ecol. Stud., 5: 97-145.
- Wei JZ, Hale K, Carta L, Platzer E, Wong C, Fang SC, Aroian RV (2003). *Bacillus thuringiensis* crystal proteins that target nematodes. Proc. Natl. Acad. Sci. USA. 100: 2760-2765.
- Wheatley RE, Caul S, Ritz K, Crabb D, Caul S (2001). Temporal variations in potential nitrification dynamics related to differences in rates and types of carbon inputs. Soil Biol. Biochem., 33: 2135-2144.
- Wilson LJ, Mensah RK, Fitt GP (2004). Implementing IPM in Australian cotton. In Novel approaches to insect pest management in field and protected crops. Arami Horowitz and Ilshaaya (eds.), pp. 97-118.
- Yeates GW (1979). Soil nematodes in terrestrial ecosystems. J. Nematol., 11: 213-229.
- Yeates GW, Bongers T, de Goede RG, Freckman DW, Georgieva S (1993). Feeding habits in soil nematode families and genera—an outline for soil ecologists. J. Nematol., 25: 315-331.
- Yeates GW, Bardgett RD, Cook R, Hobbs PJ, Bowling PJ, Potter JF (1997). Faunal and microbial diversity in three Welsh grassland soils under conventional and organic management regimes. J. Appl. Ecol., 34: 453-470.
- Yeates GW, Bongers T (1999). Nematode diversity in agroecosystems. Agric. Ecosyst. Environ., 74: 113-135.
- Yeates GW, Wardle DA, Watson RN (1999). Responses of soil nematode populations community structure diversity and temporal variability to agricultural intensification over a seven-year period. Soil Biol. Biochem., 31: 1721-1733.
- Yeates GW, Coleman DC (1982). Nematodes in decomposition. In Freckman DW ed. Nematodes in soil ecosystems. Austin TX: University of Texas. pp. 55-80.
- Zuckerman M, Coleman DC (2007). Urbanization alters the functional composition but not taxonomic diversity of the soil nematode community. Appl. soil Ecol., 35: 329-339.