INFLUENCE OF CROP MANAGEMENT PRACTICES AND ORGANIC AMENDMENTS ON NEMATODES POPULATION AND DIVERSITY IN BANANA ORCHARDS IN EMBU COUNTY

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A thesis report submitted in partial fulfillment of the requirement for the award of degree of Masters of Science in Crop protection

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OCTOBER, 2015

DECLARATION	
This thesis is my original work and has not bee	en presented for a degree in any other
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DEDICATION

I dedicate this work to my parents for bringing me up and sponsoring my education to university level. This work is also dedicated to my children; Glorious, Cyrus and Fidel and all who will find value and use the findings in this research report for the improvement of human life and future development.

ACKNOWLEDGEMENT

First and foremost I would like to thank the Almighty God for giving me good health and strength to accomplish this work. I would like to acknowledge my university supervisors Dr. W. M. Muiru, Prof. J. W. Kimenju and Dr. P. M. Wachira of the University of Nairobi for their constructive criticism, suggestions, advice and encouragement that led to accomplishment of this work. My special thanks also go to Dr. J. J. Muturi of Embu University whose informed guidance made this research work a success.

Special thanks to my employer, KEPHIS, for employment and giving me the opportunity to further my studies. The KEPHIS staff at JKIA Plant Inspection Unit, especially Dorcas Mugambi, Deborah Shituvi and Peterson Munene, are also appreciated for assisting me during official working days to attend class work.

To Jackline Gataka, MoA-LF Kyeni Division, Embu, who assisted me in reaching out to farmers, may the almighty God richly bless her. Also appreciated are all the households in Embu who allowed me to carry out research in their farms and responding to my questionnaire and the young men who assisted in transport of samples during data collection.

Special thanks goes to my wife Mrs. Alice Paul, children; Glorious Musau, Cyrus Mumo and Fidel Kimanzi for their patience, support and encouragement during my study.

To all my friends, classmates, lecturers and entire staff in PS & CP department, thank you for your support and encouragement special thanks going to Philip Kibet for his guidance on extraction and identification of nematodes.

Finally to all who stood with me and contributed in one way or another to see to the success of my studies may the almighty God bless you.

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LIST OF ABBREVIATIONS

AEZ: Agro-Ecological Zones

CABI: Centre for Agriculture and Biosciences International

DAP: Di-Ammonium Phosphate

DNA: Deoxyribonucleic acid

EPA: United States Environmental Protection Agency

FAO: Food and Agricultural Organization of United Nations

G.O.K: Government of Kenya

GDP: Gross Domestic Product

HCDA: Horticultural Crops Development Authority

IPM: Integrated Pest Management

KEPHIS: Kenya Plant Health Inspectorate Service

KIPPRA: Kenya Institute for Public Policy Research and Analysis

LH: Lower Highland

LM: Lower Midland

LU: Lower Upper land

ME: Milli-equivalent

MOA-LF: Ministry of Agriculture-Livestock and Fisheries

NEMA: National Environment Management Authority

PPM: Parts per Million

PS & CP: Plant Science and Crop Protection

RCBD: Randomized Complete Block Design

RNA: Ribonucleic acid

TA: Tropical Alpine

TSP: Triple Superphosphate

UH: Upper Highland

UM: Upper Midland

UNESCO: United Nations, Educational, Scientific and Cultural Organization

ABSTRACT

Bananas (Musa spp) play an important role as a source of food and income for many households in Kenya. However production of this fruit crop has not been easy due to pests and disease among other challenges. The objective of this study was to determine the diversity of parasitic nematodes in banana production under varying crop management practices, identify the main soil factors that influence nematode communities in banana production systems and assess the effect of organic amendments on nematode communities. A research was conducted in Runyenjes, Embu County in three agro-ecological zones namely upper midland zone 2, upper midland zone 1 and lower highland zone whereby a questionnaire was administered to farmers to get information on importance and challenges of banana production. Soil and root samples were taken from 30 farm fields from three agroecological zones for determination of nematodes occurrence and diversity. Soil samples were also analyzed for organic carbon, Potassium, soil pH, Nitrogen and Phosphorous as the main soil fertility elements. Field trials were then conducted in farmers' fields in the 3 agroecological zones in three replicates. The experiment was laid using Randomized Complete Block Design (RCBD) with 3 replicates for two seasons. Treatments used were cow manure, goat manure, chicken manure, fertilizer and a control. Soil and roots samples were collected before treatment application, after six weeks and twelve weeks after treatment application. The same procedure was repeated for season two. Nematodes were extracted and identified to genus level from the soil and roots samples. The genera found were eight and they were Pratylenchus, Meloidogyne Radopholus, Tylenchus, Helicotylenchus, Filenchus and, Scuttelonema. There was no significant different in total number of nematodes. The results showed that the nematodes population is diverse across the three agro-ecological zones. The results also showed that agronomic practices affect the level of nematodes population. In addition, there was a strong negative correlation between parasitic nematodes and soil fertility elements. Moreover, soil amendment reduced the population of parasitic nematodes in the soil and roots but had no effect on free living nematodes. From the study, it was concluded that different agronomic practices affect the population of banana parasitic nematodes and this can be incorporated with other management practices to manage the nematodes. Increase of soil elements such as Nitrogen, pH, organic carbon, Phosphorous and Potassium reduces the population of banana parasitic nematodes. Finally it was also confirmed that addition of organic amendments in the soil reduces the level of nematodes' population. The general conclusion from this research work is that variations in climate, agronomic activities and soil type, as a result of altitude variation influence the population and diversity of plant parasitic nematodes affecting bananas.

CHAPTER ONE: INTRODUCTION

1.1 Background information

Kenya's economy majorly relies on agriculture which contributes to 25 per cent of GDP and accounting for 80 per cent of the total employment (KIPPRA, 2013). One of the major sectors in agriculture is horticulture which involves production of flowers, vegetables and fruits. In Kenya, production of fruits is undertaken in various areas with banana being one among the major fruits produced. The fruit is a major diet for many people and is ranked fourth agricultural crop in the world after rice, wheat and maize (FAO, 2005). Banana is leading among fruit crops in Kenya and it is grown by both small and large scale farmers (Brooks, 2004). In the year 2012, bananas contributed to 30 per cent of the total fruits produced in Kenya. This was followed by mangoes at 22 per cent, pineapples 16 per cent and the rest of the fruits contributing the remaining percentage (HCDA, 2012).

In the year 2012, area under banana production in Kenya was estimated to be 58,000 hectares producing about 2,781,000 metric tons valued at Kshs. 23,443 million. This was followed by mangoes and pineapples with quantities produced estimated to be 1,394,000 metric tons and 466,000 metric tons valued at Kshs. 13,463 million and 10,125 million, respectively (HCDA, 2012). Banana is grown not only for subsistence use but also for commercial purposes and as livestock feed including being an important food crop in many parts of Kenya (Kimenju *et al.*, 2013).

The demand for bananas has continued to increase due to increasing Kenya's population both in urban and rural areas. This increase in demand has necessitated Kenyan farmers to invest highly in banana production. In its effort, Government of Kenya continuously provides support to farmers by undertaking important reforms (KIPPRA, 2013). These reforms include legal and institutional, increased allocation of resources in irrigation and access to inputs

especially fertilizer and seeds. However, farmers encounter various challenges during production of the fruit crop. These challenges include pests and disease damages. Among these pests, are the plant parasitic nematodes which are the most economically important pests of banana (Crow and Sekora, 2012). The annual banana production in Kenya in the year 2002 was 580,100 tons. The total yield per hectare was estimated to be 4 tons per hectare. This was below the potential yields of 30 tons per hectare (Wachira *et al*, 2013). This was attributed to poor crop production and management practices, insect pests and diseases. Other factors that caused this decline were declining soil fertility and socio-economic factors such as inadequate markets and labor.

Many farmers in Kenya have not been adequately informed on how best and economically banana nematodes can be managed (Michel *et al.*, 2010). Variations in climate, agronomic activities and soil type, as a result of altitude variation, have been reported to directly or indirectly influence the biological activities of plant parasitic nematodes affecting bananas (Lambert, 2009). In addition, factors such as soil texture and organic matter content have been found to influence occurrence of plant parasitic nematodes (Wachira *et al.*, 2013).

Traditionally, management of nematodes has been done through the use of nematicides in most parts of Kenya. Nematicides are pesticides of chemical nature which are expensive and highly toxic especially to farmers who lack skills and knowledge on how they can be used effectively. Other nematicides like methyl bromide have also been banned from use because they pollute and damage the environment (Inga *et al.*, 2010). Moreover, nematodes infestation of soils is increasing rapidly with the increasing occurrence of some genera in areas and on plant varieties including bananas formerly free of these pests (Ciancio and Mukerji, 2009). This calls for search and development of sustainable alternative means for management of banana nematodes which is affordable and manageable by farmers. Use of soil amendments such as cow manure, chicken manure and goat manure could become the

long term solution for the management of nematodes affecting banana. These amendments are affordable, easily available and safe to use. They not only reduce the nematodes population, but also improve soil fertility, plant health and are environmentally sound.

1.2 Problem statement and justification

Banana pests (which include a complex of nematodes), diseases, declining soil fertility, poor crop management, lack of clean planting materials, inefficient marketing infrastructure, postharvest losses, crop competition and lack of credit facilities have been documented as the constraint to banana production in Kenya (Martin, 2006). The main pests affecting banana are the parasitic nematodes which include, burrowing nematode (*Radopholus similis*) lesion nematode (*Pratylenchus loosi*), spiral nematode (*Helicotylenchus multicinctus* and

Helicotylenchus dihystera), root-knot nematode (Meloidogyne incognita and Meloidogyne javanica) and reniform nematode (Rotylenchulus reinformis), (Michel and John, 2005).

These nematodes cause yield losses of up to 30-60% in many countries (Brooks, 2004). They destroy the roots making them unable to supply the crop with enough water and nutrients. This slows down the rate of growth, lengthens time for fruiting and reduces bunch size as well as decreasing productivity. The top heavy plant parts often fall over due to the loss of power by the anchoring roots (Brooks, 2004). The occurrence and severity of these nematodes is influenced by different environmental and soil factors and thus differ from region to region.

Breeding for banana hybrids that have high resistance to banana parasitic nematodes and tissue culture propagation have been reported to yield good results in nematodes control (Michel *et al.*, 2010). However, these techniques are time consuming and expensive. It is also expensive to replant banana hence the use of clean planting material may not be practical for smallholder banana producers (Gaidfashovia *et al.*, 2008). Hence it is important to research

on other affordable methods of reducing the population of banana parasitic nematode below damage threshold such as cultural control and application of organic manure (Koon and Cerruti, 2009).

1.3 Objective of the study

1.3.1 Broad objective

The overall objective of this project was to improve management of plant parasitic nematodes in banana orchards through establishment of the best agronomic practices, organic amendments and determination of nematodes diversity.

1.3.2 Specific objectives

- To determine the diversity of parasitic nematodes in banana production under varying crop management practices on nematodes population.
- 2. To assess the effect of nitrogen, organic carbon, phosphorous, potassium and soil pH on nematode population in banana production.
- 3. To assess the effect of organic amendments on nematodes population in banana production.

1.4 Hypothesis

- a) Variation of crop management practices has no effect on the diversity of nematode communities in banana production.
- b) Nematode communities in banana production are not influenced by soil factors.
- c) Organic amendments have no effect on nematodes population.

CHAPTER TWO: LITERATURE REVIEW

2.1 Overview of banana crop

The economic review of agriculture in 2007 indicates that 51% of the Kenyan population lack access to adequate food which is linked to poverty which stands at 46% (G.O.K, 2008). Over the years, the Kenyan Government has strived to achieve national, household and individual food security throughout the country (M.O.A-RD, 2009). Banana, which is ranked as the fourth most important crop in the world has been cited as an important contributor to improved nutrition. However, the production of this fruit is limited by various factors among them the plant parasitic nematodes. Attempts to control these nematodes have been met with limited success because of limited knowledge of nematode diversity and symptoms (Martha *et al.*, 2011).

2.2 Importance of banana fruit

Banana is the fourth most important world food commodity after rice, wheat and maize (UNCST, 2007). The crop is grown in an area approximated to be 10 million hectares in more than 100 countries worldwide. The global annual production has been estimated to be 88 million tones (UNCST, 2007). Banana contributes significantly to food and income security to many people especially in developing countries (Martha *et al.*, 2011). The crop serves as a source of food and income to many farmers and families in Kenya both in rural and urban households (Kimenju *et al.*, 2013).

2.3 Growth requirement of banana, cultivars and area of production in Kenya

Banana crop is grown in a wide range of agro-ecological zones ranging from coastal low lands to highlands zones (Frison *et al.*, 2007). The crop does best at altitude of below 1800m above sea level. For optimal growth, a warm humid climate and an average temperature of 30°C is required (Amugune *et al.*, 2007). The annual rainfall requirement ranges from 1000

mm to 2500 mm. Banana does well in well drained loam soil although it can also be grown in a wide range of soils with good drainage and adequate fertility. For best performance, the pH should range from 5.5 to 6.5 (Amugune *et al.*, 2007).

Manure and fertilizer should be applied to achieve a healthy and strong crop growth for high production. On average banana plant will require a pH range of 5 to 7, soils of organic carbon ranging from 0.5 to 3.0, 200 gms of Nitrogen, 400 gms of potassium and 30 gms of phsphorous for otimum growth (Noor *et al* 2010). Manure should be applied at the rate of 20kgs to 40 kgs of decomposed Farm Yard manure per stool per year (Kimenju et al., 2013). A survey carried out by Martha *et al.*, (2011) on banana farming in Central and Eastern Kenya showed that banana are suitable for intercropping making it more attractive to small scale farmers who often grow different crops at the same time.

Banana is a popular fruit for most communities in Kenya and production is scattered all over the country. Among the main areas of production include Kisii, Meru, Embu, Murang'a, Kirinyaga, Kakamega, Bungoma, Nyeri, Kerio Valley and coastal areas (HCDA 2013). Local cultivars include Muraru, Kiganda, and Sukari while improved cultivars include apple, kampala, dwarf Cavendish and williams (HCDA, 2013). These cultivars are adapted to various agro-ecological zones (AEZs).

In Murang'a the main variety grown is israel (>60%) followed by Kampala in Meru and Kirinyaga. In other areas, the main varieties grown are the traditional and cooking varieties such as Muraru, Mutahato, Nyahobe, Mbiri, Kibunda, Kiganda/Githumo (Kasyoka *et al*, 2010).

2.4 Banana production constraints

Production of bananas across Africa has been declining in the last two decades (UNCST, 2007). In Kenya this decline has been attributed to pests and diseases, drought, limited land

and poor markets both local and international (Marta *et al.*, 2011). Declining soil fertility, poor crop management, lack of clean planting materials, lack of inputs and post harvest losses have also been reported to affect banana production (Barker, 2013). Other constraints in banana production include poor storage facilities damages during transport and lack of adequate information on production (Hossain 2014).

Among the pests affecting bananas are plant parasitic nematodes (which include *Radopholus similis*, *Pratylenchus* spp, *Helicotylenchus* spp and *Meloidogyne* spp), banana weevil (*Cosmopolites sordidus*), banana silvering thrips (*Hercinothrips bicinctus*), fruit flies (*Bactrocera invadens* and *Ceratitis rosa*) and banana aphids (*Pentalonia nigronervosa*) (Michel and John, 2005). All these pests attack banana plants and reduce the productivity of the crop (CABI, 2005).

2.5 Parasitic nematodes of bananas

Nematodes are small worm-like members of animal kingdom found in many habitats including water and soil (Brooks, 2004). Hundreds of nematodes species attack plants. Some are endo-parasites which live and feed within the root tissues, buds, seeds and tubers hence destroying them. Others feed externally through root walls and are called ecto-parasites. The population of parasitic nematodes is generally higher in warmer areas as compared to cooler ones (Martin, 2006). Nematodes usually cause heavy yield losses in various crops other than bananas. They damage plant roots making them difficult for water and nutrients to be absorbed. In bananas, as well as other plants, infestation usually slows down growth, reduces the reproductive life of orchard, prolong fruiting time and reduce bunch weight (Indra, 2007).

Occurrence of parasitic nematode species can vary depending on temperatures, cropping systems, soil types and husbandry practices (Brooks, 2004). Survival of plant parasitic nematodes can also be influenced by conditions such as soil organic content, soil aeration,

soil structure, predators, pathogens of nematodes, soil moisture content, distribution of host plant roots and organic matter content (Zalpuri, 2013). In cultivated soils, most nematodes occur in upper layers. Free living nematodes play a key role in essential soil processes. They directly contribute to nitrogen mineralization and distribution of biomass within plants by feeding on decomposer microbes, excreting ammonium, and immobilizing nitrogen in live biomass. Predatory nematodes also regulate nitrogen mineralization by feeding on microbial feeding nematodes (Deborah 2010).

Nematodes adapt to unfavorable conditions through production of eggs, in cysts, having a wide host range, production of high number of eggs and resistance to adverse conditions by some juvenile stages. They have stylets that pierce cells and allow them to feed on cellular contents. Most of them spend all or part of their life in the soil. Plant parasitic nematodes can cause yield losses of up to 30-60% in many countries through mechanical or chemical injuries (Zalpuri, 2013). The main plant parasitic nematodes affecting bananas include;

2.5.1 Burrowing nematodes

One of the main parasitic nematodes affecting banana crop worldwide is burrowing nematode (*Radopholus similis*). It is an endo-parasitic migratory nematode which causes decay of the cortex (Sekora & Crow, 2012). Burrowing nematodes causes toppling diseases in bananas (Tholkappian and Rajendran, 2011). Motile juvenile stages and female attack and penetrate the roots at any point and feed from inside (Sekora & Crow, 2012). The female lays eggs inside the root as they migrate through the roots, with an average of four to five eggs per day for 2 weeks. The complete life cycle from egg to adult takes 20 to 25 days at a temperature range of 24 to 32°C. The eggs hatch into second-stage juvenile after 8-10 days and the juvenile stages are completed in 10 to 13 days. The second-stage juvenile completes its life cycle within the root or moves out to look for other healthy root of the host plant. After

entering the roots, the nematodes occupy an intercellular position in the cortical parenchyma where they feed on the roots cells (CABI 2014).

Symptoms of *R. similis* damage are dark and necrotic lesions similar to those caused by other endo-parasitic nematodes (Whitehead, 2001). Nematodes are less affected by soil variables such as soil texture since they are endo-parasites (Michel and John, 2005).

2.5.2 Lesion Nematodes

Root-lesion nematodes (*Pratylenchus loosi*) are ranked third after root-knot and cyst nematodes as nematodes of the greatest economic impact worldwide in crops (Davis and MacGuidwin, 2005). They produce characteristic necrotic lesions on the surface of infected roots. The nematodes move into the soil from the over-parasitised roots. They then penetrate the growing part of the root tip. It takes 15-17 days for eggs to hatch, 15-16 for the juvenile stage and 15 days as adults before egg laying (CABI 2014). At low level of infestation lesion nematodes does not cause significant damage to banana plant. However at high population, the parasites can lead into water and nutrient deficiencies, slow growth and finally dieback disease. In India these nematodes have been reported to cause significance losses (CABI, 2015). Lesion nematodes are confirmed through microscopic examination (Perry and Ploeg, 2010).

2.5.3 Root-knot nematodes

Root-knot nematodes (*Meloidogyne incognita* and *Meloidogyne javanica*) forms galls on injured plant tissues (Guerena, 2006). Under average conditions a female produces 300 to 800 eggs between 30 to 40 days which hatch into juvenile stage. The juveniles penetrate the root tips of the host plant. Invaded nematodes initiate the development of giant cells in the root tissues and galling of roots occurs (CABI 2014). This makes it difficult for water and nutrients to flow from the roots to other sections of the plant causing stunting and impairing

nutrients flow. Leaves turn yellow and eventually the plant wilts. *Meloidogyne* spp have a wide host range (Eric and Mellisa, 2005). Management of these nematodes can be achieved through maintaining field sanitation and choice of pest free planting material. Irrigation during dry season as well as addition of organic manure has been found to reduce root-knot nematodes below economic damage thresholds (Perry, 2014).

2.5.4 Spiral nematodes

Spiral nematodes (*Helicotylenchus* spp) are mainly found in soils characterized by heavy level of clay, silt, low organic matter and low pH. These nematodes penetrate the cell walls of the host plant where they feed and then moves to other feeding sites of the root cells. They are less affected by soil type. Reproduction varies among different species. The pest stays in one location feeding on a single food cell where they lay eggs. The eggs hatch into second stage juvenile which grows into third stage juvenile and then adult (CABI, 2014). The nematodes have a wide host range including fruit crops, vegetables and other plants. Management is achieved through application of nematicides or by use of Integrated Pest Management practices (William, 2012).

2.5.5 Reniform nematodes

Effects of reniform nematode (*Rotylenchulus reinformis*) are influenced by the presence of other root nematodes. The pest is largely distributed in tropical, subtropical and warm temperate zones (Koon-Hui, 2007). These nematodes can survive for more than two years in absence of the host in dry soil through anhydrobiosis (a survival mechanism which enables them enter an ametabolic state and live without water for a long time).

R. reniformis has four juvenile stages. Females lay single-celled eggs, which develop into the first stage juveniles that moult into the second-stage and third stage juvenile. The final moult produces an immature vermiform female or male. The female penetrates the cortex of host

plant root. The females then lay eggs into a gelatinous matrix. The complete life cycle take 3 weeks depending on environmental conditions (CABI 2014).

Crops affected include cotton, pineapple, fruit crops and many vegetable crops. Population level is influenced by soil type, structure and irrigation. Management of these nematodes can be achieved through planting of resistant cultivars, crop rotation, continuous irrigation during dry seasons, weeding and field sanitation (Nematol, 2013).

2.6 Effects of Parasitic Nematodes on Bananas

Generally, plant parasitic nematodes cause damages in banana due to chemical and mechanical injuries, increasing multiplication of other disease causing organisms due to wounds they create and toppling of the plant (Michel and John, 2005). Nematodes affect banana yields by damaging the root system. Burrowing and lesion nematodes are associated with root necrosis, reduced banana biomass and toppling of the plant. Banana plants become unable to take up water and nutrients resulting into stunted growth, delayed maturation time and reduced bunch size. Nematodes also cause breakdown of banana resistance to other disease causing organism as well as reducing the ability of the crop to withstand water stress (Nicholas and William, 2012).

2.7 Management of Banana Parasitic Nematodes

2.7.1 Planting of resistant varieties

Many banana hybrids (such as apple, kampala, williams, grand nain and poyo) from plant breeders are resistant to nematodes and other disease causing micro-organisms. (Mbwika et al, 2009). Farmers can be encouraged to plant these hybrids in order to reduce nematodes population (Brooks, 2004).

2.7.2 Managing soil biology

Routine application of organic matter in the soil is important for maintaining a healthy soil. Addition of organic matter in form of decomposed compost or manure usually leads to reduced nematode population in the soil. This reduces their adverse effects on plants (Guerena, 2006). Addition of organic manure in the soil improves soil fertility and water holding capacity (Kimenju, 2013). It also increases plant resistance, increase population of bacteria and fungi as well as other nematode-antagonistic agents. Nematodes in the soil sometimes feed on the increased bacteria and fungi in the soil thus reducing their adverse effects on host plant (Stock *et al.*, 2011). Some fungi attack and destroy nematodes in the soil. There is evidence that about 50 per cent of nitrogen in crops is made available by the activities of bacteria-feeding nematodes. These nematodes are themselves not necessarily plant parasitic nematodes (Deborah, 2010). Crop rotation, minimum tillage, compost, animal manure and cover crop practices promotes the growth of beneficial organisms while suppressing the plant parasitic nematodes (Vern, 2005).

2.7.3 Use of synthetic nematicides

In developing countries, control of nematodes has been achieved by the use of nematicides. However, these chemicals are under restricted use and most of them have been banned from use due to their adverse effect on human, animal and environment (Tholkappian and Rejendram, 2011). However, researchers have established that, the use of synthetic pesticides to control pests in crops significantly increases the yield (Atungwi *et al.*, 2009). However these chemicals are likely to be misused causing environmental pollution and negatively affecting balance of nature ((Tholkappian and Rejendram, 2011). They are also costly and some are not available to farmers (Khan and Tarannum, 1999). Green manure significantly reduces cysts, eggs and larval population of *Meloidogyne chitwoodi* and *Heterodera schachtii* for instance (Hafez & Palanisamy, 2006).

2.7.4 Field sanitation (cultural practices)

Field sanitation is an important factor in control and prevention of spread of banana parasitic nematodes as well as other plant pests and diseases. Removing crop remains by hands or mechanically prevents multiplication of nematodes (Michel *et al*, 2010). Control of weeds has been found to control various pests in crop fields (Martin, 2006). This is because weeds can serve as an alternate host of a pest when the crop is harvested and then attack the crop when planted.

2.7.5 Organic amendments

Soil amendment with crop residues, green manure and other organic manure highly suppresses nematode population in any field (Barker, 2013). Combinations of green manure with other organic amendments have potential for management of parasitic nematodes in large scale.

Micro-organisms that are antagonistic to plant parasitic nematodes are stimulated by organic soil amendments. This is made possible due to the fact that the decomposition of organic matter causes specific compounds such as *Oxamyl* in the soil to have nematicidal effect (MacGuidwin *et al*, 2006). Organic amendments such as cow manure, compost manure, and chicken manure could be waste from animals, industrial wastes, agricultural wastes and other bio-product wastes. The control of plant parasitic nematodes is as a result of improved soil structure & fertility, alteration of the level of plant resistance, release of nemato-toxic compounds and other biocontrol agents (Akhtar and Malik, 2000).

Results by Lawrence *et al.*, (2006), indicated that poultry litter reduced *Rotylenchulus reinformis* by 55 per cent. This was attributed to reduction of number of eggs in roots. Results also showed that bacterial population was also increased, evidence that poultry litter has a potential to reduce *Rotylenchulus reinformis* in cotton (Lawrence *et al*, 2006)

2.7.6 Use of biological control

Use of bio-controls have been reported to be environmentally safe and sustainable (Ciancio and Mukarji, 2009). Moreover, application of micro-organisms was observed to improve health and productivity of tomatoes (Perry and Ploeg, 2010). Application of *Bacillus subtilis*, *Bacillus thuringiensis* and *Pseudomonas stutzeri* against *Meloidogyne incognita* was observed to increase plant growth and yield increase in tomato. These micro-organisms reduced root galling and inhibited the reproduction of the nematodes in tomato fields (Lawrence *et al*, 2006) Myrmicine ants were found to control banana weevil in Cuba. Microbial control, using entomopathogenic fungi and nematodes have also been found to be effective (Gold 2001).

2.7.7 Use of Integrated Pest Management (IPM) in banana production

Effective, economical and ecological-based integrated management of nematodes is a key component of sustainable food production and for enhancing quality of life in an increasing world population (Sterling and Pattison, 2008). This may involve biological control, cultural methods, crop rotation, application of organic amendments and induction of crop defense reaction. Biological control can be done by application of natural substances produced or extracted from micro-organisms (Davis and MacGuidwin, 2005). IPM approach in nematodes management enhances protection of environment, food safety, good working conditions, human health and economic development (Alabouvette *et al*, 2006). This method could serve as the long lasting solution not only to management of banana parasitic nematodes but also in management of other pests (Ritzinger *et al*, 2006).

2.7.8 Phytosanitary regulations

Phytosanitary regulations including restriction of movement of plant materials has been used as a method of preventing introduction, establishment and spread of nematodes from one country to another (Nicholas and William, 2012). In this method, only plant materials that

have been certified are allowed to be exported (Barker, 2013). In Kenya, Kenya Plant Health Inspectorate Service (KEPHIS) is the phytosanitary regulatory institution. The institution is mandated to undertake regulatory activities which ensure that pest risk analysis is done to any plant material intended to be imported into Kenya.

2.8 Effects of soil factors on nematodes population

Most pest control procedures used by farmers can be considered as soil fertility management. Resistance of plants to pests and diseases is related to optimal physical, chemical and biological characteristics of soils. More balanced mineral nutrition makes crops more resistant to pests and disease. Different animal manure applications can result in sustainable soil fertility through enhancement of soil pH, electrical conductivity, moisture content (MC), nitrogen (N), phosphorous (P) and potassium (K), (Akhtar, 2013).

2.9 Diversity of nematodes in soil

Nematode population in the soil is linked to the soil chemical and physical properties (Moreno, 2008). These chemical properties define the microenvironments in which nematodes interact with each other and with other organisms (Moreno, 2008). Nematodes are key players in various function which enables species interaction. Nematode species and their interactions in the soil result in associations in which they affect and reflect soil properties (Campos-Herrera, 2008). Relationships between nematodes and soil properties depend on soil management (Koon and McSorley, 2005). Soil texture, organic carbon, soil nitrogen, bulk density, microbial diversity, and the presence of food sources and natural enemies play roles in determining nematode assemblage composition (Moreno, 2008). At landscape level, longitude, elevation, temperature, pH, vegetation and land use determines the composition of nematode assemblages.

CHAPTER THREE: MATERIALS AND METHODS

3.1 Description of the study area

3.1.1 Location and characteristics of study area

The research was conducted in Runyenjes in Embu County located approximately 120 km North-east of Nairobi on the Eastern slopes of Mt. Kenya. The County occupies among the most prime fertile land in the Kenyan highlands, with its weather being favorable for various agricultural activities. The county has six major Agro-ecological zones, namely tropical alpine (TA), Upper Highlands (UH), Lower Highlands (LH), Upper Midland (UM), Lower Midland (LM), and Inland Lowland (IL). The soils are deep fertile volcanic soils suitable for tea, coffee, banana and other crops. The county lies approximately between latitudes 0°8' and 0° 35' south and longitudes 37°19' and 37°42' east (NEMA, 2009).

3.1.2 Climatic characteristics of the trial site

The trial site was characterized by highlands, midlands hills and valleys with altitude ranging from 1500 m to 4500 m above sea level (Jaetzold and Schmidt, 2006). The research was conducted at the altitude range of 1750 m to 1900 m above sea level. The temperature ranges from 12°C in July to a maximum of 27°C in March (NEMA, 2009). The annual rainfall ranges between 800 mm and 2800 mm. The area experiences two seasons of rainfall with long rains occurring from mid-March to May and short rains from October to December (FAO-UNESCO 1974).

3.1.3 Determination of zones

The research was carried out in three Agro-Ecological Zones (AEZ). The zones were upper midland zone 2 /Coffee zone (UM2), upper midland zone 1 /Coffee tea zone (UM1) and lower highland zone/tea zone (LH). These zones were selected since most of banana

production takes place within the zones. Assistance in zone allocation was given by the agricultural officer from the MoA-LF in Kyeni Division, Ruyenjes, Embu.

3.2 Administration of the questionnaire

A simple questionnaire (appendix 1) was administered to selected farmers, with farms ranging from one to two hectares, in each zone to obtain information on importance of bananas in their livelihood, constraints of production, farmers' knowledge on banana parasitic nematodes and losses incurred by farmers during production due to nematodes infestation. The questionnaire was administered randomly to 10 farmers in each of the three zones where samples were collected, making a total of 30 farmers. The response was gathered and percentages on importance of bananas and challenges of its production in Runyenjes was calculated. Methods applied by farmers to overcome the challenges were also gathered.

3.3 Determination of nematodes diversity and agronomic practices affecting their population

In each Agro-Ecological Zone, 10 farms in, 3 replicates, of banana plantations were selected at random making a total of 90 farms where the samples were collected. The selection of 90 farms was made possible by visiting various farms where enquiry was made on agronomic practices undertaken in banana farms. This enabled different farms with different agronomic practices to be selected as indicated in table 1. Soil and root samples were collected from each selected farm before setting of field trials. The procedure adopted for collection of samples was as described by Kleynhans, (1999) (appendix 2). Some of the farms selected had combinations of various agronomic practices. The agronomic practices were intercropping, manure application, irrigation and application of nematicides. A control farm was also included in which nothing had been planted (Table 1).

Table 1: Agronomic practices in each selected farm in lower highland zone, upper midland zone one and upper midland zone two

Block/Farm	Agronomic practice
1	Intercropping, manure and irrigated
2	Manure, irrigated and nematicides
3	Intercropping and manure
4	Manure and irrigated.
5	Manure and nematicides
6	Manure only
7	Irrigated and nematicides
8	Irrigated only
9	Nematicides only
10	Nothing done

In each of the ninety randomly selected farms, thirty root samples and sixty soil samples were collected from each of the three zones. The root samples where cut randomly from each selected banana stool. The total samples collected from each agro-ecological zone were 90. Soil was gently lifted using a trowel and a knife was used to cut the roots. Thirty pieces of root each 10 cm long were randomly cut from each banana stool from an average depth of 25 cm to make a sub-sample. Each thirty sub-samples were mixed so as to obtain a composite sample of 2 Kg of soil and 100g of roots from each farm.

The soil samples were divided into two samples. One part was used for extraction and identification of nematodes while the other part was used for analysis of soil fertility. The samples were placed in a sturdy plastic bag and closed firmly. The bags were labeled, placed

in a cool box and transported to the nematology laboratory for nematode extraction. The samples for nematodes extraction were stored at 10° C before extraction of nematodes.

3.4 Calculation of nematodes diversity

The diversity of plant parasitic nematodes was calculated to quantify the biodiversity of nematodes in the three agro-ecological zones of study. This was done through taking into account the number of genera present and their abundance. The factor that was taken into account was the richness which was the measure of genera in the three agro-ecological zones. Diversity was calculated using Simpsons Diversity Index.

3.5 Analysis of soil for Organic Carbon, pH, Nitrogen, Phosphorous and Potassium

The other soil samples of 1 kg each was analyzed for Organic Carbon, Nitrogen, Phosphorous, Potassium and soil pH. A comparison was made in terms of the level of each element and the population of nematodes found. Analysis was done to determine if the quantity of each element in the soil influences nematode population. Analysis was also done to find out if the soil factors influence the interaction by modulating nematodes population. Analysis of Phosphorous and Potassium was done using the procedure described by Chech, (1999).

3.5.1 Analysis of soil samples for Phosphorous

Soil was mixed with water to make 5ml solution. The solution was put into a test tube and 1ml of ammonium vanadate-molybdate mixture was added. The solution was mixed and the optical density was read on the colorimeter after one hour at 430 nm. The calibration graph of the reading of working standard series was plotted against its concentration in ppm. The concentration of samples was read on the graph.

3.5.2 Analysis of soil samples for Potassium

Analysis of soil for Potassium was done by pipetting 2ml of working standard series, soil extract and blank into 25ml vials. Then 5ml of anion exchange resin and 15ml DW were added, shaken and allowed to stand overnight. The working standard series, soil extract and blank solution was aspirated into flame photometer and transmission was recorded.

3.5.3 Analysis of soil samples for Nitrogen

Nitrogen was analyzed using macro KJELDAHL method as described by Hinga *et al*, (1982). This was done by transferring 1 gm of dried soil into digestion tube. Using a scoop, 1 gm of catalyst mixture was added together with 10ml of concentrated sulphuric acid. The digestion tube was placed on the digestion rack and the mixture was heated gently to 350°C. The digestion was continued until the digest turned colorless. The mixture was allowed to cool and 30 ml of distilled water was added. The mixture was transferred into 100 ml volumetric flask and let to stand overnight. This was followed by distillation and titration to determine the quantity of Nitrogen in the sample.

3.5.4 Analysis of soil samples to determine organic carbon

Analysis of soil for organic carbon was done using the procedure described by Anderson and Ingram, (1993). This was done by weighing 1 gm of soil. This was put into the digestion tubes and 2 ml of deionised water was added. This was followed by adding 10 ml of 5% potassium dichromate solution and 5 ml of concentrated sulphuric acid. The tubes were placed into the digestion block. This was heated gradually at a temperature of 150°C for 30 minutes. The digests were then removed from the block and left to cool then 50 ml of 0.4% barium chloride solution was added to each tube. This was mixed using a vortex mixer and allowed to stand overnight. Then a calibration curve was plotted and the percentage of organic carbon determined.

3.5.5 Determination of soil pH

Analysis for pH in the soil was done using a pH meter. The soil sample was placed in a khaki bag and dried in a oven for 40°C. The dried soil was ground using a pestle and motor. The sample was then sieved using a 0.5 mm sieve. This was followed by adding 20 ml of distilled water using a pipette. The sample was mixed using a vortex mixer and left to stand for one hour. The pH meter was then placed in the mixture and the pH reading was taken.

3.6 Field experiment

Field trials were conducted in the 3 agro-ecological zones in which one farm was used in each zone. The area of each farms used for laying the experiment was one hectare. The trials were conducted between the month of July 2013 and February 2014. The experiments were carried out in two seasons for four months in each season. Season one (dry season) July, 2013 to October, 2013 and season two (wet season) November, 2013 to February, 2014.

The treatments were replicated three times in each zone in the farm selected. The experimental design adopted was the Randomized Complete Block Design (RCBD). Treatments used were cow manure, goat manure, chicken manure, compound fertilizer (NPK 17:17:17) and a control banana stool, where no treatment was applied, (Table 2).

Table 2: Experimental design

Block 2 Goat manure

Farm 1 (Lower highland zone)

Block 1 Control Cow manure Goat manure Fertilizer Chicken manure

Block 2 Goat manure Fertilizer Chicken manure Control Cow manure

Block 3 Fertilizer Chicken Manure Cow manure Goat manure Control

Farm 2 (Upper midland zone 1)

Block 1 Control Cow manure Goat manure Fertilizer Chicken manure

Block 2 Goat manure Fertilizer Chicken manure Control Cow manure

Block 3 Fertilizer Chicken Manure Cow manure Goat manure Control

Farm 3 (Upper midland zone 2)

Block 1 Control Cow manure Goat manure Fertilizer Chicken manure

Block 3 Fertilizer Chicken Manure | Cow manure | Goat manure | Control

The treatments applied were cow manure (40 kg per banana stool), goat manure (40 kg per banana stool), chicken manure (20 kg per banana stool), compound fertilizer (17:17:17 100 gms/stool) and the fifth treatment where nothing was applied. The quantities applied were based on Nitrogen Ratio in each manure type, (Table 3). Cow, goat and chicken manure comprised of well-composted and cured manure which had been dried for a period of six months. One banana stool, with at least four plants, of the same variety (Israel) was selected for each treatment application. This was replicated three times for each treatment. Each stool

Fertilizer Chicken manure Control Cow manure

comprised of 5 banana plants at fruiting stage. The amendments were incorporated in 10 cm of the soil in each banana stool by use of a spade.

Table 3 Carbon Nitrogen Ratio

Type of manure	%Nitrogen	C:N Ratio
Cow manure	1.5 - 4.2	11 – 30
Chicken manure	4 – 10	3 – 10
Goat	1.3 – 3.9	13 – 20

Source: Richard, 1999

3.7 Data collection

Roots and soil from the treated banana stools were sampled before application of the amendments. Ten roots and ten sub-soils were collected from each banana stool where treatments were to be applied. Soil was gently lifted using a trowel and a knife was used to cut the roots to an average depth of 25 cm. Each 10 sub-samples were mixed so as to obtain a composite sample of 1Kg of soil and 100 gms of roots from each banana stool.

Treatments were then applied and more roots and soil samples were collected after six weeks from the treated banana stools. During this period care was taken to ensure that the treated banana plants were not interfered with. This was repeated after twelve weeks for the first season and season two. After samples were collected, they were labeled and placed in a sturdy plastic bag and closed firmly. The bags were placed in a cool box and transported to the Nematology laboratory for nematode extraction. The samples were then stored at 10°C before extraction of nematodes. Nematodes from each sample were extracted and identified to genera level.

3.8 Isolation/characterization and counting of nematodes from the soil and roots samples

Nematodes were extracted from the soil and root samples using the procedure described by Kleynhans, (1999). To extract nematodes from the soil samples, soil was mixed thoroughly and 200 cc was placed in a bucket. The bucket was three quarters full with water. Soil samples were soaked in water for two hours. The mixture was stirred to free nematodes from the soil and allow them to be suspended in the water. The mixture was allowed to settle for one minute and then decanted over a 2 mm aperture sieve into another bucket. Less water was added to the sediment in the first bucket. This step was repeated three times to increase nematode recovery. Any sediment left in the first bucket was discarded and the bucket washed out. The sieve was rinsed over the second bucket. The contents in the second bucket were stirred, allowed to settle for about 10 seconds and then poured through a 710 µm aperture sieve into the clean bucket. The residue nematodes were collected on the sieve by directing a gentle stream of water on the upper surface of the sieve so as to wash out small nematodes and eggs. The process was repeated using 250, 125 and 90µm aperture sieves. The contents of the collecting beakers were allowed to settle for 1 hour and the supernatant liquid was carefully decanted leaving about 20 ml in the bottom. In addition to this, nematodes were also extracted from the roots by chopping them into pieces of 1 cm long and then submerging them (roots) into the bucket containing water to free the nematodes. Samples were concentrated to 3 ml aliquot. Nematodes were then extracted and identified using the same procedure described above.

Nematodes were killed by transferring them into a drop of water on a glass slide which was heated for a few minutes by placing the slide on a hot plate at 65°C. The nematodes were then transferred to a fixative (40% formaldehyde topped with distilled water to 100 ml). After fixation, nematodes were transferred to glycerine via dehydration so as to avoid decaying.

Nematodes were then identified to genera level using nematodes identification key. The population level of nematodes was determined from a counting slide under microscope observed at magnification of 10 and expressed as the number per 200 cc of soil or 10gm of roots.

3.8 Data analysis

Data collected was entered in Microsoft Excel 2007 software and exported to GenStat Windows, Thirteenth Edition (Genstat Release, 2010) for analysis of variance. Separation of means and determination of probabilities for the significance of F values was done at 5% probability levels.

CHAPTER 4: RESULTS

4.1 Banana production and production constraints in Embu

Banana crop is grown by 68% of the households mainly for subsistence consumption. Banana is mainly produced for subsistence consumption with 30% of the total production by farmers being sold within the county and neighboring counties including Nairobi. About 2% of the total banana products and banana plants are used for other purposes such as aesthetic purposes, wind break and as a wrapping material (Figure 1).

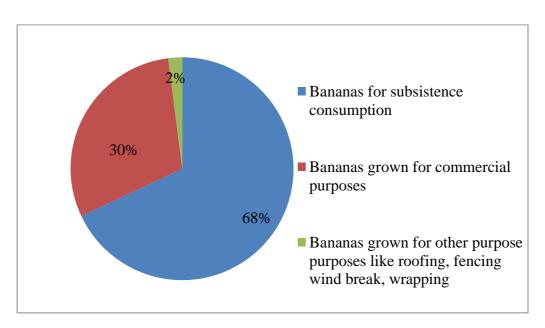


Figure 1: Importance of banana crop in Embu

According to the findings obtained from the survey banana is affected by various pests and diseases reducing its quality and hence the productivity. Pests identified include nematodes, banana silvering thrips, moths, mites, fruit flies and banana aphids. Pests' damage varies across the year with high level of nematodes infestation being experienced during dry season. Other challenges affecting banana farming include lack of planting materials, lack of enough farming land, poor rainfall distribution and fruit storage facilities. Lack of information on

pest and disease management and lack of ready markets were identified also as other challenges (Table 4).

To overcome the challenges of banana production in Embu, farmers mainly use pesticides. The pesticides were sourced from agro-vets within the county. Other methods used include irrigation during dry season, intercropping, change of varieties with higher pest resistance and shifting the place of production to new areas perceived to have low levels of pest infestation.

Table 4: Challenges of banana production in Embu based on 30 respondents

Challenge	Percentage (%) of farmers
Inadequate information on pest and disease management	70
Damages by nematodes	30
Damages by banana diseases	25
Damages by other pests like banana thrips	40
Lack of clean planting materials	50
Lack of ready market	15
Lack of enough land for banana production	40
Poor storage facilities	10

4.2 Effects of farm management practices on nematodes population

Farm management practices affect the population of parasitic nematodes. Results showed that 90% of banana farmers in Embu use various agronomic practices in banana production (Table 5, Figure 2 and Figure 3). Parasitic nematodes from the soil and root samples in which different agronomic practices had been practiced differed significantly. The lowest numbers of nematodes were found in the soil where practices such as application of manure, irrigation and nematicides had been done (Table 5). The highest number of parasitic nematodes was found in the farm which had been neglected. The results also showed that there was no

significant effect of the various agronomic practices on the free living nematodes. Generally, in all the samples obtained from the farms where different agronomic practices had been done, low counts of parasitic nematodes were recovered. There was no significant difference in parasitic nematodes from the soil samples obtained from the farms where intercropping was incorporated with manure, irrigation, application of nematicides or application of manure (Table 5). Sample where nothing had been done differed significantly from the other samples. Nematodes population was low in farms where irrigation, intercropping and application of manure was done (Table 5, 6 and Figure 2).

Table 5: Average number of nematodes in 200cc of soil sample in relation to agronomic practices in Lower Highland Zone

Agronomic practice	Parasitic nematodes in soil samples (densities in 3ml aliquot)	Parasitic nematodes in roots samples (densities in 3ml aliquot)	Free living nematodes in soil samples (densities in 3ml aliquot)
Intercropping, manure and irrigated	27 ^{ab}	6^{a}	27ª
Manure, irrigated and nematicides	47 ^{bc}	33 ^b	53 ^{cd}
Intercropping and manure	5 ^a	68 ^{cd}	50 ^{cd}
Manure and irrigated	90^{d}	74 ^d	57 ^{cd}
Manure and nematicides	53°	77 ^{de}	52 ^{cd}
Manure only	31 ^{bc}	89 ^{ef}	43 ^{ab}
Irrigated and nematicides	28^{ab}	71 ^{cd}	61 ^{cd}
Irrigated only	35 ^{bc}	59c	45 ^{bc}
Nematicides only	32 ^{bc}	60°	61 ^{cd}
Nothing done	89 ^d	96^{f}	65 ^d
P-value	< 0.001	< 0.001	0.007
LSD (5% significance level)	25	13	17
CV (%)	36.6	9.2	18.3

Values with different letters in the same column are significantly different at 5% probability while treatments with the same letters are not significantly different

Table 6: Average number of nematodes in 200cc of soil sample in relation to agronomic practices in Upper Midland Zone one

Agronomic practice	Parasitic	Parasitic	Free living
	nematodes in	nematodes in	nematodes in
	soil samples	roots samples	soil samples
Intercropping, manure and irrigated	28 ^b	22ª	83 ^e
Manure, irrigated and nematicides	7^{a}	24 ^a	66 ^{bc}
Intercropping and manure	63 ^d	21 ^a	56 ^{abc}
Manure and irrigated	18 ^{ab}	39 ^b	73 ^{de}
Manure and nematicides	42 ^c	61 ^c	48 ^{ab}
Manure only	81 ^e	95 ^{ef}	66 ^{cd}
Irrigated and nematicides	96 ^f	83 ^d	45 ^a
Irrigated only	79 ^e	99 ^f	72 ^{de}
Nematicides only	60 ^d	91 ^{def}	45 ^a
Nothing done	83 ^e	87 ^{de}	62 ^{bc}
P-value	< 0.001	0.02	0.001
LSD (5% significance	13	11	15
level)			
CV (%)	10.5	9.9	14.0
level)			

Values with different letters in the same column are significantly different at 5% probability while treatments with the same letters are not significantly different

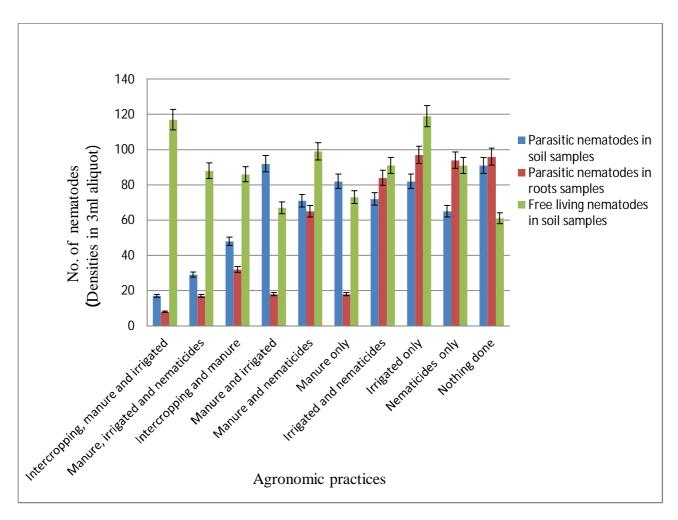


Figure 2: Average number of nematodes in 200cc of soil in relation to agronomic practices in upper midland zone two

4.3 The main parasitic nematodes in lower highland zone, upper midland zone one and upper midland zone two

The main parasitic nematodes genera found in all the three zones were Pratylenchus, Meloidogyne and Radopholus (Figure 3). In all, the nematodes population was low where intercropping, application of manure and intercropping had been done. The population was high where nothing had been done.

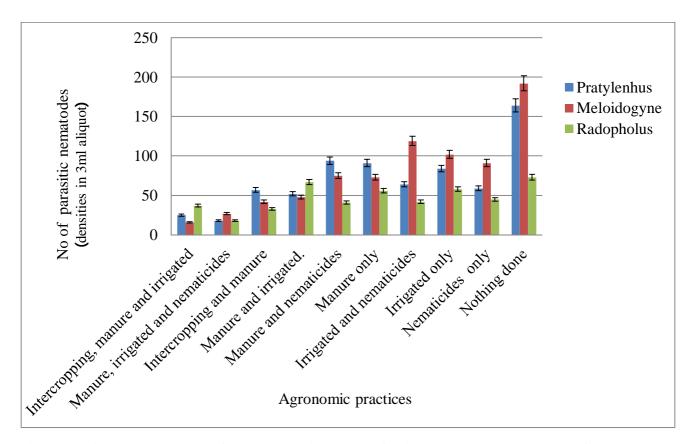


Figure 3: Average number of nematodes in 200cc of soil samples and 100 gms of root samples of the main parasitic nematodes genera in banana farms as per agronomic practices in lower highland zone, upper midland zone one and upper midland zone two

The main parasitic nematodes genera recovered in soil samples were *Pratylenchus*, *Meloidogyne* and *Radopholus* with *Meloidogyne* spp being the most common nematodes in terms of frequency of occurrence. These nematodes were mainly found in farms where nothing had been done to improve production. The farms which had been intercropped with application of manure and irrigation had the lowest population of nematodes.

In roots samples, the main nematodes genera recovered was *Radopholus* although other genera such as *Meloidogyne* and *Pratylenchus* were also recovered (Figure 4). Like in soil samples, samples from the farms which had nothing done to improve production had the largest number of nematodes. Fewer nematodes were found in farms which and been irrigated

with application of manure and nematicides to control nematodes, for example application of manure.

4.4 Occurrence of banana parasitic nematodes in Embu in root and soil samples

4.4.1 Roots samples

Analysis of roots samples collected from the three agro-ecological zones of the selected farms, showed that three main banana parasitic nematodes genera which were Pratylenchus, Meloidogyne and Radopholus occur in all the three agro-ecological zones (Figure 4). The number of Pratylenchus and Meloidogyne differed significantly. The number of the three main parasitic nematodes also differed significantly. There was no significant difference in number of nematodes between these nematode genera. There were no non-plant parasitic nematodes found in root samples.

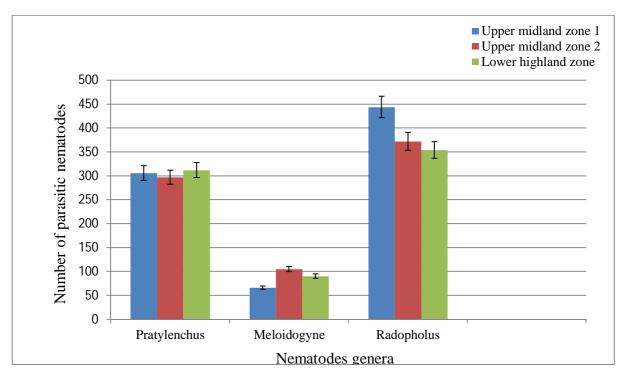


Figure 4: Population of main parasitic nematodes in upper midland 2, upper midland zone one and lower highland zone in root samples

4.4.2 Soil samples

Analysis of soil samples showed that three main banana parasitic nematodes genera which were Pratylenchus, Meloidogyne and Radopholus occur in all the three agro-ecological zones (Figure 5). The number of the three main parasitic nematodes differed significantly. The other nematodes identified from the soil samples were *Tylenchus*, *Helicotylenchus*, *Filenchus* and, *Scuttelonema*. There was no significant different in total number of nematodes between these nematode genera. Free living nematodes were also found in soil samples.

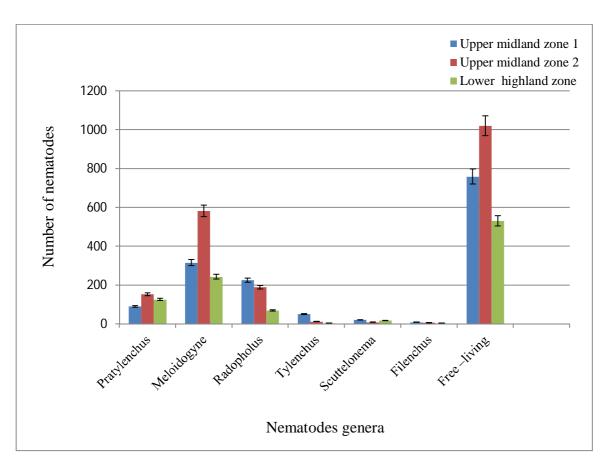


Figure 5: Population of nematodes in upper midland 2, upper midland zone 1 and lower highland zone as recovered from the soil samples

The results showed that different genera of parasitic nematodes occur in banana production zones in all the three ecological zones. These were *Pratylenchus, Meloidogyne, Radopholus, Helicotylenchus, Tylenchus, Hoplolaimus, Scuttelonema, Filenchus* and free living

nematodes. The genus Meloidogyne was mostly found in the soil samples while *Radopholus* and *Pretylenchus* were more in root samples. The main genera from both the root and soil samples were *Radopholus*, *Meloidogyne* and *Pratylenchus*. Free living nematodes were also recovered. The population for other nematodes genera was insignificant.

4.4.2 Diversity of plant parasitic nematodes in lower highland zone, upper midland zone one and upper midland zone two

The results from the study showed that the population of plant parasitic nematodes depends on agro-ecological zone and agronomic practices. These two factors play a key role in influencing the diversity of nematodes. There were eight genera of parasitic nematodes found in addition to free living (Table 7). The population was low in upper highland zone and high in upper midland zone two which was a lower zone. There were more plant parasitic genera found in soil samples than in root samples. In upper midland zone two, there were more varieties of genera which were more equitably distributed (Table 8). In all the zones, more than 34% of the farms were occupied by one species.

Parasitic nematodes were more diverse in lower highland zone than in all other zones. Nematodes in roots samples were less diverse as compared to the ones in soil samples. The results also show that variations in nematodes diversity occurred in different agro-ecological zones.

Table 7: Mean number of plant parasitic nematodes in lower highland zone, upper midland zone one and upper midland zone two as influenced by agro-ecological zones

Nematode genera	Soil samp	oles (densit	ies in 3ml	Root sam	ples (densi	ties in 3ml
	Lower highland zone	Upper midland zone 1	Upper midland zone II	Lower highland zone	Upper midland zone 1	Upper midland zone II
Pratylenchus	47	30	51	104	102	118
Meloidogyne	87	108	194	30	22	35
Radopholus	27	65	63	118	138	124
Tylenchus	1	17	4	0	0	0
Helicotylenchus	8	0	0	1	0	0
Scuttelonema	0	3	2	0	0	0
Filenchus	4	7	13	0	0	1
Hoplolaimus	0	0	0	0	0	1
Total parasitic nematodes	174	230	327	253	262	278
Free –living	231	253	37	0	0	0

Table 8: Diversity indices of nematodes genera in lower highland zone, upper midland zone one and lower midland zone two

	Value						
	Soil sampl	es		Root samp	Root samples		
	Lower highland zone	Upper midland zone one	Upper midland zone two	Lower highland zone	Upper midland zone one	Upper midland zone two	
Number of genera (parasitic)	9	9	9	6	6	6	
Richness	7	7	8	4	3	5	
Diversity	3.4	3.8	3.3	2.7	2.5	2.8	

4.5 Effects of soil factors on nematodes population

4.5.1 Occurrence of parasitic nematodes in relation to pH

The results indicated that increase in soil pH resulted in slight change in nematodes population (Figure 6). This indicates that the population of nematodes is slightly affected by acidic soil and nematodes slightly increase as soil tends to be alkaline. The number of free living nematodes remained fairly constant and increase or decrease in pH did not influence the number of these nematodes.

4.5.2 Occurrence of parasitic nematodes in relation to soil organic carbon

Soil samples with high level of organic carbon were found to have comparatively lower level of nematodes population (Figure 7). From the graph, it is clear that increase of the quantity of organic carbon in the soil results into reduction of parasitic nematodes.

4.5.3 Occurrence of parasitic nematodes in relation to Nitrogen

Soil samples with high levels of nitrogen had lower levels of nematodes. Figure 8 shows that increase in quantity of nitrogen causes the reduction in population of nematodes. The results indicate a negative correlation between quantity of nitrogen in the soil and nematodes population.

4.5.4 Occurrence of parasitic nematodes in relation to Potassium

Soil samples which had comparatively high level of Potassium were found to have low population of nematodes. This indicates that increase in quantity of Potassium in the soil caused reduction in the population of nematodes. This shows a negative correlation between the level potassium and the population of nematodes as indicated in figure 8. The graph for parasitic nematodes in figure 9 is steeper than the other graph indicating that there was an interaction.

4.5.5 Occurrence of parasitic nematodes in relation to Phosphorous

Soil samples with high level of phosphorous were found to have comparatively lower level of nematodes population (Figure 10). From the graph it is clear that increase of the quantity of phosphorous in the soil resulted into reduction of nematodes population. The results indicate a negative correlation between quantity of phosphorous in the soil and the population of nematodes.

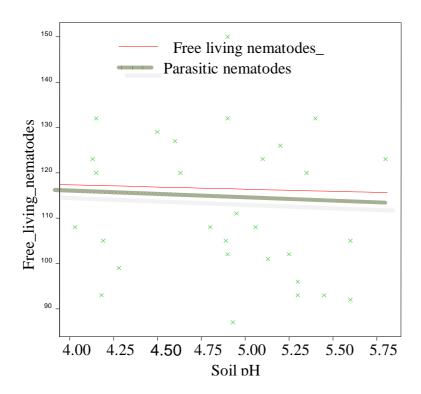


Figure 6: Occurrence of nematodes in soil in relation to pH

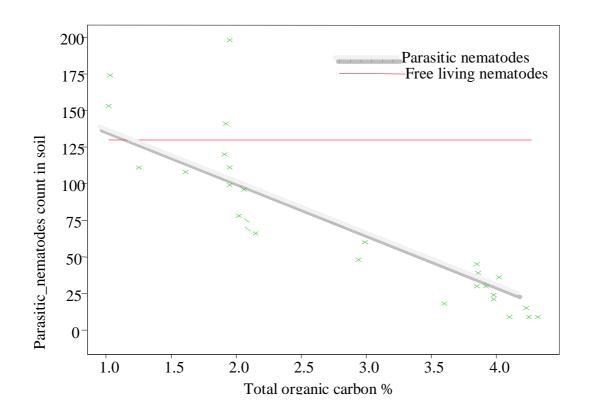


Figure 7: Occurrence of parasitic nematodes in relation to soil organic carbon

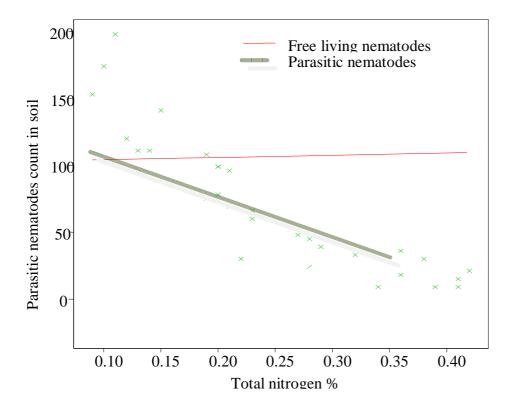


Figure 8: Occurrence of parasitic nematodes in relation to Nitrogen

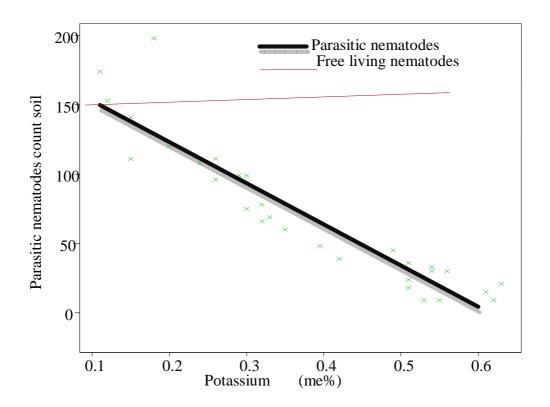


Figure 9: Occurrence of parasitic nematodes in relation to Potassium

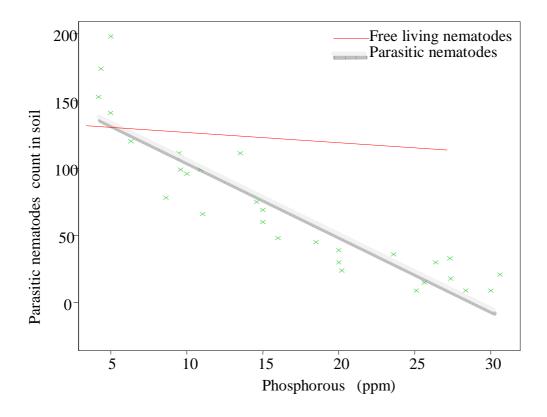


Figure 10: Occurrence of parasitic nematodes in relation to phosphorous

The distribution of plant-parasitic nematodes negatively correlated with the soil organic carbon, potassium, phosphorous and nitrogen. In all parasitic nematodes, the population was high where low levels of these soil parameters were recorded. Low levels of nematodes were recorded in high levels of soil organic carbon, potassium, phosphorous and nitrogen.

4.6 Effects of organic amendments on nematodes population

In lower highland zone, there was a significant difference in the number of plant parasitic nematodes as a result of the effect of treatments used (Table 9). Treatments had no effect on free living nematodes. There was no significant difference on cow manure and fertilizer on plant parasitic nematodes in soil in season one and season two while control treatment was different from all the other treatments (Table 9,10 and 11). The effect of treatments was significantly different in first season for both populations of parasitic nematodes in soil and root samples and free living nematodes in the three agro-ecological zones. In season two, chicken manure was significantly different from all other treatments for it reduced the population of nematodes to lower level than the other amendments.

Table 9: Effects of organic amendments on nematodes population in banana roots and soil from the lower highland zone

	First season			Second season			
Treatment	nematodes (densities in (densities in 3ml		Plant parasitic nematodes (densities in 3ml aliquot)		Free living (densities in 3ml aliquot)		
	Soil	Roots	Soil	Soil	Roots	Soil	
Fertilizer	25 ^{ab}	69 ^{ab}	85a	20 ^{ab}	59 ^{ab}	63 ^a	
Cow manure	32 ^{bc}	51 ^a	81 ^a	21 ^{ab}	41 ^a	68 ^a	
Chicken manure	20 ^a	73 ^{ab}	82ª	12ª	46 ^a	63 ^a	
Goat	19 ^a	106 ^c	85ª	12 ^a	96°	65 ^a	
Control	41 ^c	89 ^{bc}	83ª	28 ^b	82 ^{bc}	64 ^a	
P-value	0.001	0.002	0.08	0.008	<0.001	0.3	
LSD (5% significance level)	10	27	4	10	24	5	
CV (%)	14.2	16.6	11.9	17.1	17.5	8.5	

Values with different letters in the same column are significantly different at 5% probability level while treatments with the same letters are not significantly different

In upper midland zone one, there was larger population of nematodes in control treatment as compared to the other treatments (Table 10). In season one and season two, all the treatment had no effect on the population of free nematodes (Table 10). Cow and chicken manure had

the same effect on nematodes population in soil in season one. In season two, all the treatments had different effects on the population of nematodes in roots. From the table (Table 10), it is also seen that all the treatments had no effect on the population of free living nematodes for both season one and season two.

Table 10: Effects of organic amendment on nematodes population on banana roots and soil from the upper midland zone one

Upper midland zone 1

	First seas	on		Second se	eason	
	Parasitic nematodo (densities aliquot)		Free living (densities in 3ml aliquot)	Parasitic (densities aliquot)	nematodes s in 3ml	Free living (densities in 3ml aliquot)
Treatment Fertilizer	Soil 35 ^b	Roots 109 ^b	Soil 46 ^a	Soil 27 ^b	Roots 99°	Soil 37 ^a
Cow manure	33 ^{ab}	69 ^a	42 ^a	26 ^b	58 ^a	38 ^a
Chicken manure	30^{ab}	93 ^{ab}	61 ^a	19 ^{ab}	68 ^{ab}	51 ^a
Goat	21 ^a	116 ^b	56 ^a	13 ^a	101 ^c	52 ^a
Control	56 ^c	94 ^{ab}	42 ^a	52 ^c	84 ^{bc}	42 ^a
P-value	< 0.001	0.007	0.054	< 0.001	0.002	0.349
LSD (5% significance level)	13	26	20	12	24	19
CV (%)	18.7	16.2	5.3	17.4	19.1	22.3

Values with different letters in the same column are significantly different at 5% probability while treatments with the same letters are not significantly different

In upper midland zone two, the treatments had significant difference, when compared to control, on the population of parasitic nematodes in soil in season one and season two.

Treatments were not significant in the population of parasitic nematodes in roots. Fertilizer, goat manure and cow manure equally reduced the population of parasitic nematodes in soil in both season two and season one. The control treatment did not reduce the population of nematodes and it was significantly different from all other treatments. The treatments did not reduce or increase and had no effect on free living nematodes.

Table 11: Effects of organic amendment on nematodes population on banana roots and soil- Upper midland zone two

	First seaso	n		Second season			
	Plant parasitic nematodes (densities in 3ml aliquot)		Free living nematodes (densities in 3ml aliquot)	Plant parasitic nematodes (densities in 3ml aliquot)		Free living nematodes (densities in 3ml aliquot)	
Treatment	Soil	Roots	Soil	Soil	Roots	Soil	
Fertilizer	48 ^b	106 ^a	103 ^{ab}	38 ^b	89 ^{ab}	66 ^a	
Cow manure	43 ^b	85 ^a	86 ^a	35 ^b	73 ^{ab}	71 ^a	
Chicken	44 ^b	96 ^a	96 ^{ab}	29 ^{ab}	66 ^a	70^{a}	
manure Goat	27 ^a	102 ^a	102 ^{ab}	18 ^a	90 ^{ab}	68 ^a	
Control	90°	113 ^a	106 ^b	89 ^c	106 ^b	66 ^a	
P-value	< 0.001	0.62	0.6	< 0.001	0.168	0.5	
LSD (5% significance level)	15	37	18	15	34	6	
CV (%)	15.9	20.7	13.9	21.1	25.7	15.5	

Values with different letters in the same column are significantly different at 5% probability while treatments with the same letters are not significantly different

CHAPTER 5: DISCUSSION

5.1 Importance of banana crop and challenges facing its production in Embu County

Banana crop is a very important food crop for the livelihood of majority of people in Embu County. The fruit crop is grown by more than 68% of the households. Majority of farmers in Embu generate income through banana production. This agrees with the findings of (Martha *et al.*, (2011) who found that banana is a very important food crop to the people of central and eastern Kenya.

Despite this crop being an important fruit crop for these people, many challenges are encountered during its production. Chief among these challenges are pests and diseases. According to the findings from this research, the main pests attacking bananas in the region are nematodes, banana weevil, banana silvering thrips, fruit flies and banana aphids. The nematodes attacking banana were found to be widespread throughout the 3 agro-ecological zones where the research was conducted. According to the information obtained from the farmers, majority of them were not aware of the nematodes attack on bananas although the symptoms of nematodes attack were evident. This is because most farmers are not able to differentiate symptoms of nematodes from other pests. This concurs with the findings of (Michel *et al*, 2010) who referred nematodes as silent killers as they destroy crops without being identified.

5.2 Diversity of nematode genera in banana production areas under varying environmental and crop management practices

In accordance with the findings from this research, eight genera of plant parasitic nematodes and free living nematodes occur in the three agro-ecological zones. Eight genera of banana parasitic nematodes identified were *Pratylenchus*, *Meloidogyne*, *Radopholus*, *Helicotylenchus*, *Tylenchus*, *Hoplolaimus*, *Scuttelonema* and *Filenchus*. The first three

nematodes were the main parasitic nematodes found with *Radopholus* being the most occurring genera in roots while *Meloidogyne* was the main parasitic nematodes recovered in roots. These other five genera were not frequently found. These findings agrees with the findings of (Michel and John, 2005) who found *Radopholus similis*, *Pratylenchus* spp, *Helicotylenchus* spp, *Meloidogyne* spp and *Rotylenchulis* spp to be the main parasitic nematodes affecting bananas. Sekora and Crow, (2012) also found *Radopholus similis to be* the most economically important parasitic banana nematode in the world.

The genus *Radopholus* complete its life cycle within the root tissues hence it is less affected by agronomic practices unlike other nematodes. In their research on plant parasitic nematodes in subtropical and tropical agriculture, Michel and John, (2005) found *Radopholus similis* to be less affected by soil variables since it is an endoparasite. In addition to parasitic nematodes, free living nematodes were also found.

This study also indicates that high population levels of banana parasitic nematodes were found in upper midland zone 2 which was the lower zone. Low population levels were found in lower highland zone which was an upper zone of the study area. This is in agreement with the findings of Martin, (2006) who found that population of nematodes was denser in warmer areas as compared to cooler ones. This is because high altitude areas experiences low temperatures which are unfavorable for survival of many nematodes. Low altitude areas which are relatively warmer provide a favorable environment for the survival of many parasitic nematodes, Ruess *et al.* (1999).

Intercropping is an important agronomic practice for the management of banana parasitic nematodes. According to the findings from this research, banana farms that had been intercropped with other food crops had low population level of nematodes. Nematodes were further reduced when this practice was combined with irrigation and application of manure.

These findings concurs with the findings of Brook, (2004) who found that occurrence of parasitic nematodes species can vary depending on temperatures, cropping systems, soil types and husbandry practices. Moreover Barker (2013) found that seasonal fluctuation of nematodes population is influenced by nematodes biology, environmental factors and management practices.

These finding are attributed to the fact that some crops like legumes contain beneficial bacteria. These bacterial are able to carry out nitrification in the soil. The process helps to make nitrogen available for plant absorption (EPA, 2004). Banana planted together with these crops easily absorbs the availed nitrogen hence are able to grow vigorous and healthy and develop some resistance to nematodes. Some crops also release chemicals which may be toxic to parasitic nematodes.

Addition of water to banana stool in form of irrigation during dry season reduces stress to the plant. Plants which are not stressed are able to assume a vigorous growth. This overcomes the effect of parasitic nematodes and can reduce the population of nematodes.

Application of nematicides to control banana parasitic nematodes was also observed to reduce parasitic nematodes to a significant level. This agrees with the findings of Atungwi *et al*, (2009) who established that, the use of synthetic pesticides to control pests in crops significantly increases the yield. These nematicides act by direct killing of nematodes through poisoning. However, most of these chemicals have been banned from use due to their adverse effects on human beings, animals and environment; hence they cannot be used to control nematodes. Indra (2007) found that in developing countries, control of nematodes had been achieved by the use of nematicides. The chemicals had a restricted use and most of them had been banned due to associated problems of residual, toxicity, environment pollution and public health.

5.3 Influence of soil factors on nematode communities in banana production systems

The factors assessed were soil pH, soil organic carbon, nitrogen, phosphorous and potassium. Increase in soil pH slightly lowered population of parasitic nematodes. For other soil parameters, the results indicated a negative correlation relationship. The free living nematodes were not much affected by the soil pH.

The soil factors here were used to explain the fertility of the soil. Reduction in nitrogen, phosphorous, potassium and organic carbon in the soil leads to reduction in soil fertility. Soil fertility influences nematode communities in the soil. Guerena, (2006) found that about 50 per cent of nitrogen in crops is made available by the activities of bacteria-consuming nematodes. These nematodes are themselves not necessarily plant parasitic nematodes.

Soil factors, which included nitrogen, potassium, phosphorous, organic carbon and soil pH, influence the interaction by modulating the behavior of nematodes. This agrees with the findings of Mathias and Jing (2013) who found that root-feeding herbivores and natural enemies are likely to decrease in abundance in soils with high quantities of abiotic factors.

5.4 Effect of organic soil amendments on nematode communities

This research further indicates that application of organic amendments in banana stools reduces the population of parasitic nematodes. This is in line with the findings of Kleynhans, (1999) who discovered that survival of plant parasitic nematodes could be influenced by organic content, soil aeration and soil structure. Ciancio and Mukarji, (2009) reported that use of biocontrol and soil amendments are environmentally safe and could be effective in management of parasitic nematodes.

Cow manure was the most effective in reduction of plant parasitic nematodes in all amended treatments. In all the amended soils, lower nematodes population was recovered compared to the control. Soils amended with chicken manure and cow manure had significantly lower

nematodes population as compared to the one amended with goat manure and fertilizer.

Although some of the organic amendments performed better than others, it was evidenced that all the organic amendments reduced the population of parasitic nematodes to significant levels.

The results showed that incorporation of these amendments effectively suppresses the population of nematodes. One of the mechanisms involved is that the application of amendment results into increase of fungi and bacteria which feed on nematodes resulting into reduction of their population. Results by the Lawrence *et al*, (2006) reported that poultry litter reduced *Rotylenchulus reinformis* by 55 per cent. This was attributed to reduction of number of eggs in roots. Results further indicated that bacterial population was also increased. This is evidence that poultry litter has a potential to reduce *Rotylenchulus reinformis* in cotton.

The organic amendments, which are cow manure, goat manure and chicken manure, supplies needed food to the micro-organisms thereby increasing their effectiveness in destroying nematodes. This observation concurs with the findings by Wachira *et al* (2009) who found the amendments to be effective in stimulating microorganisms. The other possible mechanism is that the reduction of the population of nematodes was attributed to release of substances with nematicidal effect and organic acid during amendments decomposition in the soil.

The reduction of banana parasitic nematodes observed in both roots and soil indicated that all the amendment were effective in reducing the population of nematodes in both banana roots and in soil. Most parasitic nematodes genera such as *Meloidogyne* have their life cycle both in soil and roots. This is one of the reasons why their reduction in the soil also resulted into their reduction in the roots.

In the banana stools where farms had been neglected, nematodes did not reduce in number. In some of the banana stools, the population increased. This is also a proof that soil amendments reduce the population of parasitic nematodes.

Addition of organic amendments in banana farms increases soil fertility and water holding capacity. The organic matter added releases nemato-toxins, increase fungi and bacteria in the soil which reduces the population of parasitic nematodes. It also increases the population of other nematodes-antagonistic agents (Wachira *et al*, 2009).

Micro-organisms that are antagonistic to plant parasitic nematodes are stimulated by organic matter (Wachira *et al*, 2009). These micro-organisms, which mainly include fungi attack and destroy nematodes hence reducing their population in soil. Addition of organic amendments promotes growth of beneficial organisms while suppressing the plant parasitic nematodes. All these agrees with the findings of Akhtar and Malik, (2000) who reported that control of plant parasitic nematodes can be achieved through improved soil structure and fertility, alteration of the level of plant resistance, release of nemato-toxic compounds and other biocontrol agents. In addition to this, MacGuidwin *et al.*, (2006) discovered that decomposition of organic matter causes nematicidal effect and organic matter increases fungal activities in soil. The findings of Khan and Tarannum, (1999) indicated also that application of microorganisms against *Meloidogyne incognita* increase plant growth and yield in tomato. The micro-organisms reduced root galling and inhibited the reproduction of the nematodes in tomato fields.

Soil nematodes respond to environment disturbance (Zalpuri et al, 2013). Parasitic nematodes are abundant in soil and experience a variation in distribution patterns. Their abundance is influenced by agronomic practices such as irrigation, mixed cropping, application of nematicides and organic amendments.

CHAPTER 6: CONCLUSION AND RECOMMENDATIONS

6.1 Conclusions

Banana is a very important food crop not only to the people of Embu but also to Kenyans as a whole. Special attention must be paid to this crop in pest management. The research established that eight genera of plant parasitic nematodes in banana occur in Embu. This was in addition to free living nematodes. The parasitic nematodes were *Pratylenchus*, *Meloidogyne, Radopholus, Helicotylenchus, Tylenchus, Hoplolaimusa Scuttelonema* and *Filenchus*. It was found that agronomic practices such as irrigation, application of manure, mixed cropping and application of nematicides largely reduce population of plant parasitic nematodes in bananas. Combination of irrigation, application of manure and mixed cropping was found to be the best practice in reduction of nematodes. The research also established that, large population of nematodes was found in upper midland zone two which was the lower zone, as compared to the other two zones of study.

Soil factors such as pH, Nitrogen, Phosphorous, Potassium and soil Organic Carbon highly influenced the occurrence and population of banana parasitic nematodes. This research established that, there is a strong negative correlation between Nitrogen, Phosphorous, Potassium and Organic Carbon and parasitic nematodes in bananas. Change of soil pH was found to have no effect on the population of nematodes. Further research will be required to establish how increase of these factors reduces the population of parasitic nematodes. These factors were also found to affect the diversity of plant parasitic nematodes in bananas. In this research, the potential of organic soil amendment to suppress banana parasitic nematodes was demonstrated. Application of cow manure, goat manure and chicken manure was found to suppress the population of plant parasitic nematodes in bananas with cow manure emerging as the best amendment. Further research will be required to find out the level at which these organic amendments should be added in banana stools to effect a maximum benefit.

This research indicates that environment and crop management, soil factors and mineral & inorganic amendment affect the population level and diversity of plant parasitic nematodes. There is a need for intervention in banana farming to ensure farmers are acquainted with knowledge on existence of nematodes and their management. Since banana parasitic nematodes were found to be widespread, there is a need to implement efforts aimed at their control. Educating farmers on various methods of controlling nematodes such as use of soil amendments will not only boost banana production but also improve the quality of the bananas.

6.2 Recommendations

It is recommended that for proper management of parasitic nematodes in banana orchards, farmers should use organic manure such as chicken manure, cow manure and goat manure. However, further research will be required to establish the period in which the amendments remain effective in the soil especially in banana farms.

Diversity and population of nematodes is affected by environmental factors such as temperatures and soil factors and agronomic practices. It is hence recommended that farmers should be encouraged to apply manure especially cow, goat and chicken manure in their banana crops. This will be in addition to doing irrigation during dry season for these practices have been proved to largely reduce the population of plant parasitic nematodes in bananas. Further research will be needed to establish how diversity and population of nematodes is impacted by different pest management strategies and environmental factors.

For complete management of plant parasitic nematodes in bananas, IPM cannot be ruled out. Integration of compatible pest management strategies remains a key aspect in IPM use as a tool for management of banana nematodes. Moreover, knowledge of nematodes diversity in an area can also be useful in deciding management strategies for parasitic nematodes.

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APPENDICES

Appendix 1: Questionnaire
UNIVERSITY OF NAIROBI
DEPARTMENT OF PLANT SCIENCE AND CROP PROTECTION
Masters of Science in Crop Protection student
A56/79521/2012
BASELINE SURVEY QUESTIONNAIRE
IMPORTANCE OF BANANA AND CONSTRAINTS OF PRODUCTION IN EMBU
Questionnaire number
DETAILS OF THE FARMER
1.1 NameZone
1.2 Age in years: $[1] = 20-30$ $[2] = 31-40$ $[3] = 41-50$
$[4] = 51 - 60 \qquad [5] = \ge 61$
1.3 Highest level of education
[1] = None [2] = Primary [3] = Secondary [4] = Tertiary [5] = University
2.0 SOURCES OF INCOME
[1]= Sale farm produce [2]= Formal employment [3]= Sale of livestock

[4]= Small Business	[5]= Casual labor	[6]=Pensions	[8] =
Dividends	[9] = House rentals	[10] = Interest savings	[11] =
Land lease	[12] =support from family m	embers	
[13]= Others (specify)			
3.0 LAND USE PRACTICE	S		
3.1 What is the size of your f	armAcı	res	
3.2 What are the main farm e	enterprises? Tick the appropri	ately:	
[1]= Crops	[2]= Livestock – (cattle, por	ultry etc)	
[3]= Woodlot//agro-forestry	[4]= Other (specify)		
3.3 If growing crops, what cr	ropping patterns do you praction	ce?	
[1]= Monocropping	[2]= Inter-cropping	[3]= Relay cropping	
[4]= Others (specify)			

3.4 What are the major crops grown on your farm?

Crop	Acreage	Usage
1		
2		
3		
4		
5		

3.5 Are there pests that attack banana in the field? [1]= Yes [2]= No

If **YES**, name the pest and rank them based on the losses that they cause. State also the control method

Pest	Ranking	Control measures	
	1.	1.	
	2.	2.	
	3.	3	
	1.	1.	
	2.	2.	
	3.	3	
	1.	1.	
	2.	2.	
	3.	3	
	1.	1.	
	2.	2.	
	3.	3	

Pest ranking-[1]-Heavy loss [2]-Moderate loss [3]-Slight loss

3.6 Are there diseases that attack bananas?

Pest ranking-[1]-Heavy loss [2]-Moderate loss [3]-Slight loss

3.7 Have you observed a change in damage by pest and diseases over last 5 years?

3.8 If **YES**, state the changes fill the table below

Pest/Disease	Trend 1= Decrease	Remarks
	2= Increase	
	3= remained the same	

3.9 What do you attribute the changes	to:
---------------------------------------	-----

[1]= Increase in temperature [2] = Decrease in temperature

[3]= Emergence of new crop diseases [4] = Change in rainfall patterns

[5]= others specify

3.10 What strategies/measures have you taken in response to these changes?
Planting of different varieties
Planting of tissue culture
Planting treated/disinfected planting materials
Application of conservation agriculture like use of organic manure, mulching, pruning and
removal of old stems from the farm
Use of irrigation
Migrate to other areas
Use of pesticides
Intercropping
Others specify
3.11 Have you noticed attack by nematodes in your farm
[1] = Yes $[2] = No$
3.12 If yes, what proportion of bananas are affected
[1]= less than 10% [2]= up to 25% [3]= up to 50% [4]= up to 75% [5]= Over 75%
3.113 Does damage by nematodes vary across the year? $[1] = Yes$ $[2] = No$
3.14 If YES , in which seasons is the damage most severe?
[1] = Dry and hot [2] = Cool and dry [3]= Cool and wet [4] = Warm and wet
3.15 How would you rate the severity of nematodes attack in your farm?

Ranking of nematodes damage among other pests

Pest	Ranking	Coding for Ranking
		1 = 0% infection
		2 = 10% plants infected
		3 = 25% of the plants infected
		4 = 50% of the plants infected
		5 = more than 50% of the plants infected

4.0 Which of these indicators have you seen on your farm?

Good quality soils	Low quality soils
1.	
2.	
3.	

- 4.1 How would you classify the soils in your farm in terms of quality currently?
- [1] =Very good soils
- [2]= Good quality soils
- [3]=Satisfactory
- [4] = Poor quality soils
- [5] = I don't know
- 4.2 Do you apply any soil fertility inputs to your banana crop?

Input	Type of the input	Amount applied per stool	Remarks/comment
Manure	Chicken	1.	
	Goat	2.	
	3. Cattle Manure	3	
Fertilizer	1. N.P.K.	1.	
	2. D.A.P.	2.	
	3 T.S.P.	3.	
	4. Any other specify		

4.3 What are the main constraints to adoption of the recommended practices on soil fertility?

[1] =They are expensive to undertake [3] =High labor demanding

[2] =Lack of information [4] =Other specify

Appendix 2: Soil sampling procedures as adopted from (Kleynhans, 1999)

Take samples systemically at equal-spaced points according to a grid pattern that covered the entire field. Draw a sketch of how you sampled the soils. Sample at a depth of 30cm.

Bags samples should be labeled. Label should be attached to the outside of the bagged sample. Information in the label included zone, farm no., date of sample collection, agronomic practice, treatment type, replicate etc.

A field book should be there to record the name of the farmer, history of the farm and observed appearance or symptoms of banana infestation, name of collector, locality, the recent cropping history of the sampled field, previous problems and other pest-related problems and control measures taken, size of the area sampled, irrigation and source of water, date the sample was collected.

Appendix 3: Analysis of variance for diversity of nematode communities in banana production areas under varying environmental and crop management practices (parasitic nematodes in soil as per agronomic practice)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Zone stratum	2	11957	5979.	5.39	
Agronomic practice	9	46265	5141.	4.64	0.003
Residual	18	19963.	1109.		
Total	29	78185.			

d.f= degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 4: Analysis of variance for diversity of nematode communities in banana production areas under varying environmental and crop management practices (free living nematodes in soil as per agronomic practice)

Source of variation	d.f.	S.S.	m.s. v.r.	F pr.	
Zone stratum	2	2752.	1376.4 1.86		
Agronomic practice	9	7440.0	826.7 1.12	0.400	
Residual	1 8	13321.	740.1		
Total	29	23514.0			

 $\begin{array}{l} \textbf{d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance\ ratio, F} \\ \textbf{pr=F test} \end{array}$

Appendix 5: Analysis of variance for diversity of nematode communities in banana production areas under varying environmental and crop management practices (nematodes in roots as per agronomic practice)

Source of variation	d.f	S.S.	m.s.	v.r.	F pr.
Zone stratum	2	166.2	83.1	0.10	
agronomic practice	9	72283.5	8031.5	9.35	<.001
Residua	18	15463.8	859.1		
Total	29	87913.5			

d.f= degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 6: Analysis of variance for the effects of organic amendments on parasitic nematodes in soil for lower highland zone for season one

Variate: Parasitic nematodes								
Source of variation	d.f.	s.s.	m.s. v.r.	F pr				
Sampling period stratum	2	464.4	232.2 1.68					
Treatment	4	3073.2	768. 5.56	0.001				
Residual	38	5253.6	138.3					
Total	44	8791.2						

 $\begin{array}{l} \textbf{d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance\ ratio, F} \\ \textbf{pr=F test} \end{array}$

Appendix 7: Analysis of variance for the effects of organic amendments on free living nematodes in soil for lower highland zone for season one

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Sampling period stratum	2	942.4	471.2	1.53	
Treatment	4	7742.8	1935.7	6.29	<.001
Residual	38	11693.4	307.7		
Total	44	20378.8			

d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 8: Analysis of variance for the effects of organic amendments on parasitic nematodes in root for lower highland zone for season one

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	5039.6	2519.8	3.55	
Treatment	4	15766.8	3941.7	5.55	0.001
Residual	38	26990.6	710.3		
Total	44	47797.0			

d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 9: Analysis of variance for the effects of organic amendments on parasitic nematodes in soil for upper midland zone one for season one

Source of variation	d.f	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	1326.4	663.2	3.55	
Treatment	2	6109.2	1527.3	8.19	<.001
D 11 1	4	7000	1066		
Residual	38	7089.6	186.6		
Total	30	14525.2			
	44				

d.f= degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test.

Appendix 10: Analysis of variance for the effects of organic amendments on free living nematodes in soil for upper midland zone one for season one

Source of variation	d.f.	S.S.	m.s.	v.r.
F pr.				
Sampling period stratum	2	1850.8	925.4	2.00
Treatment	4	4665.2	1166.3	0.057
Residual	38	17621.2	463.7	
Total	44	24137.2		

d.f= degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 11: Analysis of variance for the effects of organic amendments on parasitic nematodes in roots for upper midland zone one for season one

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	7344.2	3672.1	5.88	
Treatment	4	11890.9	2972.7	4.76	0.003
Residual	38	23736.0	624.6		
Total	44	42971.1			

d.f= degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 12: Analysis of variance for the effects of organic amendments on parasitic nematodes in soil for upper midland zone two for season one

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	1960.0	980.0	3.46	
Treatment	4	20306.8	5076.7	17.93	<.001
Residual	38	10760.0	283.2		
Total	44	33026.8			

d.f= degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 13: Analysis of variance for the effects of organic amendments on free living nematodes in soil for upper midland zone two for season one

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Sampling period stratum	2	2420.4	1210.2	2.24	
Treatment	4	27174.8	6793.7	12.58	<.001
Residual	38	20527.6	540.2		
Total	44	50122.8			

d.f= degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 14: Analysis of variance for the effects of organic amendments on parasitic nematodes in root for upper midland zone two for season one

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	13051.	6525.	5.60	
Treatment	4	4014.	1004.	0.86	0.496
Residual	38	4307.	1166.		
Total	44	61373.			

 $\label{eq:continuous} \begin{array}{l} \textbf{d.f=} \textbf{degrees of freedom, s.s=} \textbf{sum of squares, m.s=} \textbf{mean square, v.r=} \textbf{variance ratio, F} \\ \textbf{pr=} \textbf{F test} \end{array}$

Appendix 15: Analysis of variance for the effects of organic amendments on parasitic nematodes in soil for lower highland zone for season two

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	313.6	156.8	1.40	
Treatment	4	1643.2	410.8	3.66	0.013
Residual	38	4264.4	112.2		
Total	44	6221.2			

 $\label{eq:continuous} \begin{array}{l} \textbf{d.f=} \textbf{degrees of freedom, s.s=} \textbf{sum of squares, m.s=} \textbf{mean square, v.r=} \textbf{variance ratio, F} \\ \textbf{pr=} \textbf{F test} \end{array}$

Appendix 16: Analysis of variance for the effects of organic amendments on free living nematodes in soil for lower highland zone for season two

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	341.2	170.6	0.89	
Treatment	4	8625.2	2156.3	11.28	<.001
Residual	38	7262.8	191.1		
Total	44	16229.2			

 $\label{eq:continuous} \begin{array}{l} \textbf{d.f=} \textbf{degrees of freedom, s.s=} \textbf{sum of squares, m.s=} \textbf{mean square, v.r=} \textbf{variance ratio, F} \\ \textbf{pr=} \textbf{F test} \end{array}$

Appendix 17: Analysis of variance for the effects of organic amendments on parasitic nematodes in roots for lower highland zone for season two

Source	of d.f.	S.S.	m.s.	v.r.	F pr.
variation					
Sampling	2	3904.2	1952.1	3.23	
period stratur	n				
Treatment		19683.2	4920.	8.15	<.001
	4				
Residual		22950.3	604.0		
	38				
Total	44	46537.6			

 $\begin{tabular}{ll} \textbf{d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance\ ratio, F \\ \textbf{pr=F test} \end{tabular}$

Appendix 18: Analysis of variance for the effects of organic amendments on parasitic nematodes in soil for upper midland zone one for season two

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	1193.2	596.6	4.33	
Treatment	4	7957.2	1989.3	14.44	<.001
Residual	38	5236.8	137.8		
Total	44	14387.2			

d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 19: Analysis of variance for the effects of organic amendments on free living nematodes in soil for upper midland zone one for season two

Source variation	of d.f.	S.S.	m.s.	v.r.	F pr.
Block	2	2062.5	1031.3	2.75	
stratum Treatment	2 4	1724.8	431.2	1.15	0.349
Residual		14273.	375.6		212 12
Total	38	18060.8			
Total	44	10000.8			

d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 20: Analysis of variance for the effects of organic amendments on parasitic nematodes in roots for upper midland zone one for season two

Source of variation	d.f.	S.S.	m.s. v.r.	F pr.
Sampling period stratum	2	7383.2	3691.6 7.09	
Treatment	4	12960.2	3240.1 6.22	<.001
Residual	38	19784.3	520.6	
Total	44	40127.8		

d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test

Appendix 21: Analysis of variance for the effects of organic amendments on parasitic nematodes in soil for upper midland zone two for season two

d.f.	S.S.	m.s.	v.r.	F pr.
2	2348.4	1174.2	4.67	
4	26864.0	6716.0	26.70	<.001
38	9559.6	251.6		
44	38772.0			
	2 4 38	2 2348.4 4 26864.0 38 9559.6	2 2348.4 1174.2 4 26864.0 6716.0 38 9559.6 251.6	2 2348.4 1174.2 4.67 4 26864.0 6716.0 26.70 38 9559.6 251.6

d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr = F test

Appendix 22: Analysis of variance for effects of organic amendments on free living nematodes in soil for upper midland zone two for season two

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	2215.0	1107.5	2.98	
Treatment	4	25223.3	6305.8	16.94	<.001
Residual	38	14142.8	372.2		
Total	44	41581.1			

d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, Fpr=F test

Appendix 23: Analysis of variance for effects of organic amendments on parasitic nematodes in roots for upper midland zone two for season two

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Sampling period stratum	2	14327.0	7163.5	7.80	
Treatment	4	8788.4	2197.1	2.39	0.067
Residual	38	34878.6	917.9		
Total	44	57993.9			

d.f=degrees of freedom, s.s=sum of squares, m.s=mean square, v.r=variance ratio, F pr=F test