

**SOIL NEMATODE COMMUNITIES UNDER ORGANIC AND
CONVENTIONAL FARMING SYSTEMS IN CHUKA, THARAKA
NITHI COUNTY, KENYA**

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DECLARATION

DECLARATION

This thesis is my original work and has not been presented for a degree in Kenyatta University /or any other university.

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DEDICATION

This work is dedicated to my parents Andrew Atandi and Rose Teresa Bosibori

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ABBREVIATIONS AND ACRONYMS

ANOVA	Analysis of Variance
AEZ	Agro-Ecological Zone
BF	Bacterial Feeder
BI	Basal Index
Ca	Calcium
CAN	Calcium Ammonium Nitrate
CI	Channel Index
Conv	Conventional Farming System
c-p	Colonizer-persister
d'	Genus Richness
DAP	Di-ammonium Phosphate
DS	Demonstration Site
EI	Enrichment Index
EPN	Entomopathogenic Nematodes
FAO	Food and Agriculture Organization
FF	Fungal Feeder
FFs	Farmer Fields
FiBL	Research Institute of Organic Agriculture
FLN	Free Living Nematodes
FP	Farmers' Practice
GPS	Global Positioning System
H'	Shannon-Wiener Diversity Index

ICIPE	International Center for Insect Physiology and Ecology
K	Potassium
Ln	Natural Log
LSD	Least Significant Difference
MI	Maturity Index
MINO	Maturity Index with no Opportunists
N	Nitrogen
Na	Sodium
N-A	Non-amended Control
OM	Organic Matter
OMN	Omnivorous Nematodes
Org	Organic Farming System
P	Phosphorus
POR	Participatory Organic Research
PPI	Plant Parasite Index
PPN	Plant Parasitic Nematodes
PRC	Principle Response Curves
PRED	Predatory Nematodes
PTD	Participatory Technological Development
RCBD	Randomized Complete Block Design
RKN	Root Knot Nematode
RKNBW	Root Knot Nematode-Bacterial Wilt
SAS	Statistical Analysis Software

SI	Structure Index
TSP	Triple Super Phosphate
UK	United Kingdom
UNEP	United Nation Environmental Program
UM	Upper Midland

ABSTRACT

Plant parasitic nematodes (PPN) are biotic constraints to crop production worldwide resulting in significant yield losses. Management of these nematodes has primarily depended on chemical nematicides; but due to the increased pressure for more economic and environmentally friendly strategies, alternative methods like organic and conventional amendments, have been considered. To test the efficacy of conventional and farmer practice systems against organic farming in the suppression of PPN, on abundance and diversity of free living nematodes (FLN), field trials were conducted in Chuka in January 2015 at two sites (farmers' fields and demonstration site) over three seasons: season 1 (cowpea sole crop), season 2 (maize-bean intercrop) and season 3 (bean sole crop). Organic systems received neem cake + compost + *Tithonia* + ash; conventional received Marshall EC + Calcium ammonium nitrate + Di-ammonium phosphate (DAP) and farmers' practice received DAP + Manure. A non-amended system was included in the study as control. Soil sampling was done every six weeks where ~500 g soil and 50 g cowpea, beans and maize roots were collected per plot from five sampling points. Nematodes were extracted from 100 ml of soil and 5 g of roots then identified to genus level. Data on abundance and diversity were subjected to analysis of variance in R version 3.2.5 with differences at $p \leq 0.05$ considered statistically significant. Twenty nine genera belonging to bacterivores, fungivores, omnivores, predators and PPN were found in the study area. Bacterivores dominated the organic systems while PPN prevailed in the non-amended control system at both sites. High population of bacterivores in the organic system could be attributed to compost which is known to provide soil organisms with a new energy source that increases their diversity and activity. Soils under maize-bean intercrop showed varying population of nematodes as organic system recorded significantly higher population when compared to other systems at farmer fields ($2,182 \pm 89.78$) and demonstration site ($2,014.5 \pm 98.00$). Renyi diversity profile showed no significant differences among the farming systems indicating nematodes were evenly distributed across the systems. Principle response curves were used to establish the effect of farming systems on individual nematode genera over time and only the organic model was significant. *Tylenchus*, *Meloidogyne* and *Helicotylenchus* spp. were effectively reduced up to the second month. Diversity and ecological indices during the bean sole crop differed significantly ($p \leq 0.05$) where organic systems had significantly higher values of plant parasite index (4.16 ± 0.88), enrichment index (85.52 ± 10.61) and structural index (87.42 ± 8.06) at farmers' fields with a similar trend at the demonstration site. This indicated the soils were high in nutrients and very stable. In general, significantly lower numbers of PPN were recorded in the organic system across all seasons at both sites. In conclusion, organic farming appears to suppress populations of PPN whilst promoting that of FLN. Organic farming is therefore recommended for small holder farmers and extension officers to use in awareness creation. Further research should be done to identify nematodes to species level using molecular methods under controlled environment.

CHAPTER ONE: INTRODUCTION

1.1 Background information

Nematodes are microscopic worms that are aquatic in nature. They are common in soils all over the world (Yeates *et al.*, 2009) and live in soil pores that are formed by soil processes. They move in the films of water that cling to soil particles (Blair, 1996). Their number is estimated at over half a million (Ferris *et al.*, 2012), many of which are free-living types found in the oceans, in freshwater habitats, and in soils whereas the parasitic species form the smaller group (Buckley and Schmidt, 2003). Broadly, from a plant health perspective, they can be classified as either free living or plant parasitic nematodes.

The free-living nematodes comprise mostly of beneficial nematodes where some are successfully being used as bio-control agents of other pests and pathogens (Denno *et al.*, 2008). For example, entomopathogenic nematodes *Steinernema* spp. and *Heterorhabditis* spp. are used to control insect pests like grubs within 48 hours. They are often grouped based on their feeding habits, that is, trophic groups (Yeates *et al.*, 2009), whereby bacterivores, fungivores, omnivores-carnivores and predators are present.

On the other hand, plant parasitic nematodes (PPN) are one of the major biological constraints around the world in almost all types of crops as they cause severe losses in production of up to 20% annually (Hamida *et al.*, 2015) with *Meloidogyne* spp. documented to cause yield losses of up to 85% (Coyne *et al.*, 2014). These nematodes can be grouped into relatively restricted specialized groups that either cause direct damage to their host or act as virus vectors (Makete *et al.*, 2008). Directly they affect

crops through feeding on or in plant roots and their feeding also create open wounds that provide entry to a wide variety of plant pathogenic fungi and bacteria (Wurst *et al.*, 2009). Other PPN transmit viruses, for example *Xiphinema* spp. and *Trichodorus* spp. transmit tomato ringspot virus and tobacco rattle virus in tomato and tobacco plants, respectively (MacFarlane and Robinson, 2004). Continuous cropping of susceptible crops in local farming systems in Kenya can lead to the accumulation of these pathogens. Cropping systems affect the types and number of nematodes in a field (Sinha *et al.*, 2004).

Farmers have been seeking alternatives to chemical pesticides for management of plant parasitic nematodes. The continuing environmental problems associated with the use of nematicides have introduced a sense of urgency and a persistent pressure on farmers worldwide to adopt other management strategies that do not contribute to environmental pollution. Techniques such as solarization, flooding, use of resistant cultivars and use of cover crops have been practiced (Gaur and Perry, 1991; Kaskavalci, 2007) but they are not feasible in all locations and arable land is scarce. Therefore, the need to adopt a strategy that is not location limited and is within the capabilities of the farmer arises.

Crop rotation and addition of soil amendments therefore may become necessary and convenient for farmers worldwide. Crop rotation and soil amendments (organic and conventional alike) are known to have beneficial effects on soil nutrition, soil physical conditions, soil biological activities and crop performance (Efthimiadou *et al.*, 2010). Direct applications of organic matter and mineral fertilizers to the soil always affect the abundance and activity of soil organisms (Maina *et al.*, 2012).

Organic soil amendments are fertilizers that are derived from living things such as animals or plants. They are often slow to release nutrients so soil microorganisms have to first break them down so as to make them available for plant use (Efthimiadou *et al.*, 2009). The most commonly used are manures, fishmeal, wild sunflower *Tithonia diversifolia* G. and neem *Azadirachta indica* J. Organic amendments have been investigated as an alternative method of nematode management strategies (Akhtar and Mahmood, 1996). Ferris *et al.* (2012) reported that increasing the organic matter in soil encourages the growth of numerous fungi, bacteria and beneficial nematodes that may provide some level of biological control for root knot nematodes. Hence, various organic amendments have been tested and reported to have nematicidal properties (Sharma, 2001; Devi and Hassan, 2002; Stephan *et al.*, 2002).

Plants grown in soil with high organic matter are often less damaged by nematodes compared to those grown in soil with less organic matter content (Al-Rehiyani, 2001; Efthimiadou *et al.*, 2009). Any kind of organic soil amendment including manure, rock phosphate, *Tithonia* mulch, compost and neem cake can improve tolerance of plants to nematodes and also reduce nematode populations. Neem *Azadirachtin indica* and wild sunflower *Tithonia diversifolia* as botanical materials have been reported to possess nematotoxic activity due to the content of limonoids, azadirachtin and tagitinin C, respectively (Agyarko and Asante, 2005; Adeyemi and Adewale, 2011).

There has been considerable progress in the use of compost as soil amendment for the control of PPN in infested fields (Mc Sorley *et al.*, 1999; Akhtar and Malik, 2000; Zhang and Zhang, 2009). However, they cannot eliminate a severe nematode

infestation rather they are better suited to keep nematode population low (Agyarko and Asante, 2005). Reduction in population densities of plant parasitic nematodes in response to application of organic amendments and their positive effects on host growth have been reported in many studies (Akhtar and Malik, 2000; Sharma, 2001; Devi and Hassan, 2002; Stephan *et al.*, 2002; Summers, 2011; Stirling *et al.*, 2011; Farahat *et al.*, 2012; Renco and Kovacik, 2012; Olabiyi and Oladeji, 2014).

Conventional fertilizers are composed of synthetic artificial ingredients that are manufactured and ready to use on plants. Unlike organic fertilizers, they do not need to decompose over time so as to supply nutrients to plants (Neher, 1999). They mainly contain nitrogen, phosphorus and potassium in different ratios. The use of conventional fertilizers is recommended on soils with very low nutrients and in most cases they are used to complement the use of manure (Akhtar and Mahmood, 1996). However, excessive use of the conventional fertilizer may result in leaching or groundwater contamination in case of buildup to toxic levels and it may also burn or kill plants and their roots (Scow *et al.*, 1994).

1.2 Problem statement and justification

Kenya faces the pressure for more food especially with the high human population levels (MoA, 2013). Consequently, intensification of agriculture coupled with poor crop husbandry has led to an increase in plant parasitic nematodes and other soil pathogens. Optimal pest and disease management is therefore essential. The plant parasitic nematodes have been a major global challenge in ensuring food security and feeding the increasing human population yet so little is known about these pathogens as they are often overlooked. In a review of biotic constraints in Africa, nematodes were

not at all mentioned yet reports show that in the United Kingdom, cyst nematodes alone account for over \$70 million per annum losses or approximately 9% of production (Nicol *et al.*, 2011). As a result, majority of the farmers in developing countries, who are resource poor, are barely aware of the magnitude of nematodes as plant pathogens. They often attribute their damage to other pest or crop problems (Coyne *et al.*, 2014) because of the typical disease symptoms they cause hence nematode control is overlooked. For a few, chemicals (nematicides) can be used for control but still the farmers lack technical skills required for their application (Adediran, 2005). Furthermore, the chemicals are too toxic which has led to their banning (for example, methyl bromide in Kenya, 2015) and restricted use (only liquid formulations are allowed). They are also too costly, kill beneficial soil organisms, lead to resistance development, lead to mammalian toxicity and environmental pollution (Chen and Ferris, 2004). Currently, there is no known systemic nematicide that has been developed to be applied safely to plants killing endoparasitic nematodes (Chen and Ferris, 2004) thus there is a need to look into other alternatives.

Management of PPNs over time has been based on singular strategies which do not consider the beneficial nematodes as well as other soil organisms (Neher, 2012). For instance, integrated pest management (IPM) of soil pathogens has mainly focused on plant parasitic nematodes leaving out the beneficial nematodes. Therefore, a comprehensive study of soil nematode communities was necessary in determining the effect of management strategies on the nematodes communities. A wholesome approach which took into perspective equally vital associated characteristics other than productivity and incorporated soil health and biodiversity was developed, tested and

validated. Therefore, the focus of this research was to compare the effects of organic and conventional amendments on the abundance and diversity of soil nematode assemblages.

1.3 Objectives

1.3.1 General objective

To determine the effect of organic and conventional farming systems on soil nematode populations under maize, beans and cowpeas cropping systems in Chuka, Tharaka Nithi County, Kenya.

1.3.2 Specific objectives

- i. To characterize nematode assemblages present on cowpeas under organic and conventional farming systems based on their trophic groups in Chuka, Tharaka Nithi County
- ii. To determine the effect of organic and conventional farming systems on population densities and genus diversity of soil nematode communities on intercrops (maize and beans) in Chuka, Tharaka Nithi County
- iii. To evaluate the influence of farming systems on soil nematode community structure under sole crop (beans only) in Chuka, Tharaka Nithi County

1.4 Hypotheses

- i. Nematode assemblages present on cowpeas are not different under organic or conventional farming systems.
- ii. Organic and conventional farming systems have no effect on the population densities and diversity of soil nematodes under intercrop of maize and beans.

- iii. Soil nematode community structure under sole bean crop is not influenced by organic and conventional farming systems.

1.5 Significance of the study

There has been insufficient information regarding the importance of free living nematodes and therefore this study will help highlight their significance in soil health and how soil amendments influence their population and diversity. Plant parasitic nematodes on the hand have been shown to cause diseases leading to economic yield losses worldwide yet very little has been done to control and manage their populations. Chemical control of nematodes is based on the use of nematicides, both fumigants and non-fumigants. The use of cultural practices and organic amendments will significantly reduce the cost of production as farmers will not need to purchase expensive inputs such as nematicides to control plant parasitic nematodes. With this knowledge of affordable and convenient control strategies, farmers will be able to adopt such techniques.

1.6. Conceptual framework

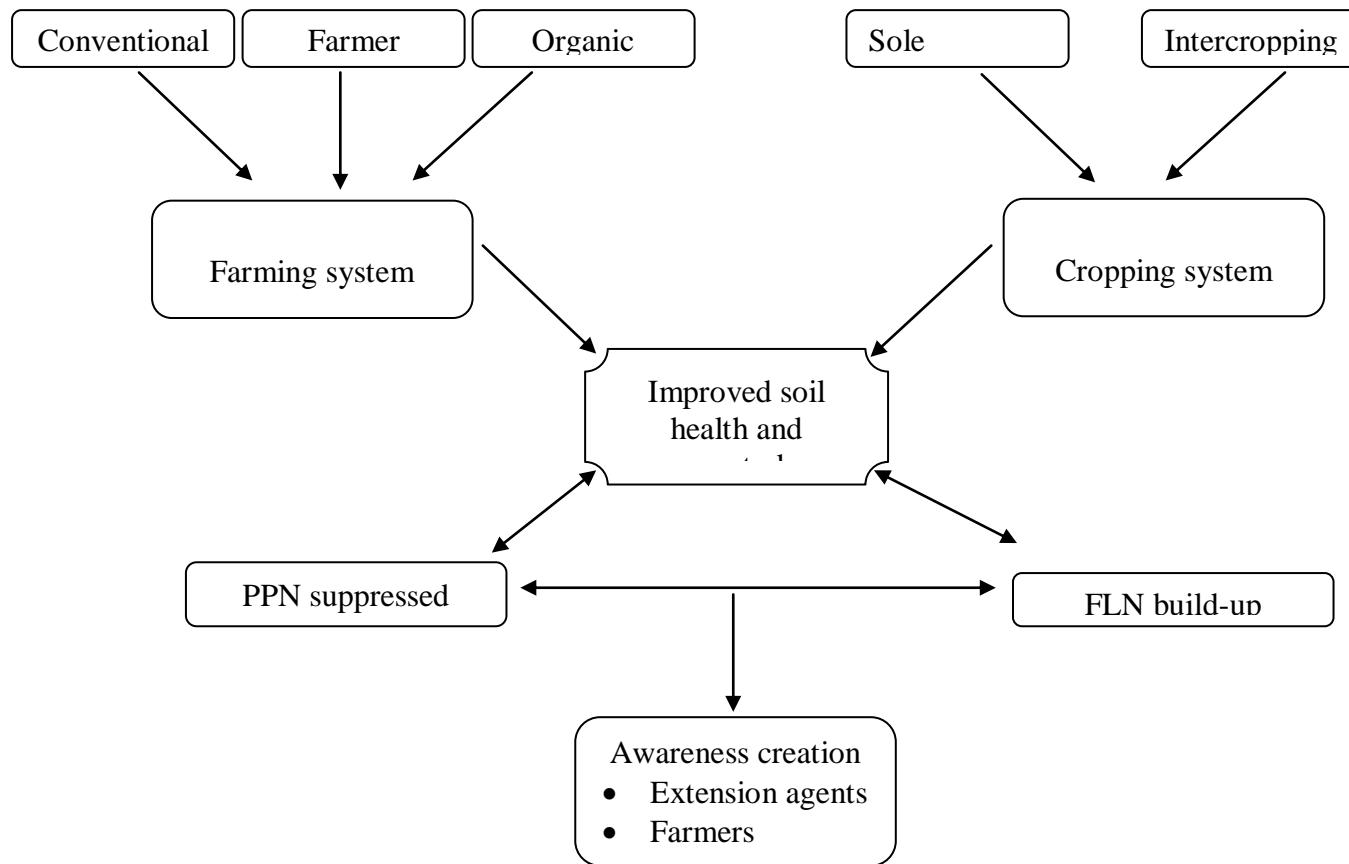


Figure 1.1 Conceptual Framework

CHAPTER TWO: LITERATURE REVIEW

2.1. Cropping systems

A cropping system is the crops and crop pattern/sequence cultivated on a piece of land over a given time as well as their management techniques (Cardinale *et al.*, 2003). The patterns include practices such as mixed cropping, monoculture, double cropping, intercropping, mono cropping, sole cropping and strip cropping.

2.1.1 Crops

2.1.1.1 Maize

Maize *Zea mays* L. belongs to the Poaceae family (Plate 2.1) and is indeed the most important food crop in Kenya as it plays an important role in food security (Short *et al.*, 2012). Having been introduced in Africa in the 1500s (McCann, 2005), maize has become a dominant crop. In Kenya, it is grown on approximately 1.6 million hectares annually (MoA, 2013) majority of which belongs to small scale farmers (Kamidi *et al.*, 1999). Its production is mainly dependent on rain.

Despite the high reliance on maize, it is subject to biotic (pests and diseases) and abiotic stresses which significantly reduce the yield. Its production has stagnated between 30 and 40 million bags (MoA, 2013). Among the diseases are the nematodes and particularly *Meloidogyne* spp. and *Pratylenchus* spp. (Sikora *et al.*, 2005). Other common nematodes include *Helicotylenchus* spp., *Rotylenchus* spp., *Hoplolaimus* spp., *Radopholus* spp., *Criconema* spp., *Tylenchulus* spp., *Xiphinema* spp., *Trichodorus* spp., *Hirschimaniella* spp., *Anguina* spp. and *Ditylenchus* spp. which have been reported to be found on the roots or the soil around the roots (Nicol *et al.*, 2011).



Plate 2.1 Maize plants and maize cob with seeds

Source - Atandi, 2016

Studies have been carried out to analyze the effects of organic amendments on maize yields (McSorley and Gallaher, 1996) and results have shown that the use of compost on agricultural sites may be a useful means for crop production. On the other hand, conventional methods of managing nematodes like use of chemicals has been limited due to environmental, economic as well as political reasons in most places (Sikora *et al.*, 2005).

2.1.1.2. Beans

Beans *Phaseolus vulgaris* L. are edible legumes in the family Fabaceae (Plate 2.2) used mostly for human or animal feed. They are the most important legumes grown in Kenya on more than 500,000 hectares of land (GoK, 2010). Generally, common bean is considered a short-season crop with most varieties maturing in a range of 65 to 110 days from emergence to physiological maturing (Otipa *et al.*, 2006). Regular consumption of common bean and other pulses is now promoted by health

organizations because it reduces the risk of diseases such as cancer, diabetes or coronary heart diseases (Leterme and Munoz, 2002).



Plate 2.2 Bean plant with pods

Source - Atandi, 2015

Bean production is influenced by a number of factors such as edaphic (soil), environmental (weather) as well as biotic factors (pests and diseases). Among the diseases affecting yields of beans are the nematodes. Nematodes that are associated with beans prevent nodulation and consequently affect nitrogen fixation (Kimenju *et al.*, 1999) and include the *Meloidogyne* spp. followed by *Pratylenchus* spp. and *Helicotylenchus* spp. *Meloidogyne* spp. are highly damaging and their estimated yield losses are up to 60% in fields that are heavily infested (Widmer and Abawi, 2000). Karanja *et al.* (2002) showed that chicken manure is the most effective organic amendment for suppressing nematodes in beans when compared to neem, compost and farmyard manure. They found chicken manure was able to reduce the galling index thrice as much as other organic amendments. Performance of beans under intercropping systems has not been intensively researched (Ntukamazina, 2008). Therefore, there is need to formulate an integrated management program that considers

all possible and economical strategies that can be used to effectively control nematodes affecting bean production.

2.1.1.3. Cowpeas

Cowpea *Vigna unguiculata* L. is a dicotyledonous crop in the order *Fabacea* that's rich in protein and starch content; and have high capacity to fix nitrogen (Tarawali *et al.*, 2002). Both grains and leaves (Plate 2.3) are used as food and are a source of income for resource poor farmers. It is mostly grown as a substitute for proteins in areas where production of beans is not suitable as they are more drought tolerant (Brader, 2002). This crop is also important as it helps improve farming systems and soil fertility as it aids in soil erosion reduction and weed suppression (Tarawali *et al.*, 2002).



Plate 2.3 Cowpea plant and pods

Source - Atandi, 2015

Research has been carried out on cowpeas and nematodes that affect cowpeas. Sosanya (2006) found *Meloidogyne* spp. and *Pratylenchus* spp. to be the most common nematodes affecting yield of different cowpea varieties. Since they are susceptible hosts for *Meloidogyne* spp., they cannot be used in rotations to control the nematodes

(Vargas-Ayala and Rodríguez-Kábana, 2001). High densities of nematodes (*M. incognita* C.) have been confirmed to negatively affect nodulation and decrease nitrogen levels in plants (Ferris *et al.*, 2012). Claudius-Cole *et al.* (2010) established that the use of plant extracts like neem, drumstick tree *Adenanthera pavonina* L., basil *Ocimum basilicum* L. and the African shrub *Vernonia amygdalina* D. are effective in the management of *M. incognita* on cowpea as they lower their populations while increasing the crop yields.

Breeders have come up with resistant cowpea lines (Cowpea breeding line IT84S-20149 and Iron Clay) which have been shown to suppress population densities of *M. incognita* (Matthews *et al.*, 1998; Ehlers *et al.*, 2002). Other strategies involve the use of available management methods such as chemical and cultural control methods (Sikora *et al.*, 2005).

2.1.2. Intercropping maize and beans

This is the cultivation of two or more crops in the same field (Plate 2.4) at the same time arranged strategically (Sinha *et al.*, 2004). The main goal is to produce more yields on a piece land by maximizing on the resources that would not have been used by a single crop (Ouma and Jeruto, 2010) as land use is optimized. When compatible crops are intercropped, biodiversity is encouraged as more insects and soil organisms get a better habitat than when a single crop was planted. This is useful as it increases predators that help control pests and diseases (Cardinale *et al.*, 2003; Altieri and Nicholls, 2004; Sinha *et al.*, 2004).



Plate 2.4 A maize-bean intercrop system

Source - Atandi, 2015

Intercropping is widely practiced in Kenya whereby legumes are often included to improve the status of nitrogen in the soil (Clermont-Dauphin, 1995). It is an important strategy that has so far been used in the management of nematodes especially *Meloidogyne* spp. (root knot nematodes) and *Globodera* spp. (potato cyst nematodes) (Akhtar, 1997). Studies on sugarcane by Berry *et al.* (2009) have revealed that intercropping can be used by small holder farmers to manage nematodes as well as improve the overall crop productivity.

However, some studies have shown that intercropping could result in more damaging effects like increasing the populations of plant parasitic nematodes (when susceptible hosts are intercropped) or insect pests (Netscher and Sikora, 1990; Pitan and Odebiyi, 2001). The effect of intercropping has been contradictory in many studies (Sinha *et al.*, 2004) and is very dependent on the choice of principal and companion crops (Berry *et al.*, 2009). The companion crop can either be useful or detrimental by attracting or multiplying the pathogens hence it is important to choose crops considerately.

2.1.3. Sole cropping

This refers to the agricultural practice of growing a single crop at a time in a field. This is useful as it allows for specialization in crop production and equipment but it can result in damage of the soil ecology. This is due to reduction in soil diversity and depletion of nutrients (Cardinale *et al.*, 2011). Monoculture is subject to greater parasitization by plant parasitic nematodes (Wasilewska, 2000). De Deyn *et al.*, (2004) established that plant diversity affects nematode diversity in soil contrast to plant abundance which does not affect the nematode diversity. When it comes to yield, studies in Costa Rica by Henriksen *et al.*, (2002) showed that beans grown as intercrops yielded 15% to 50% more than those grown as sole crops.

However, studies done by Griffin *et al.* (2009) showed that there were no significant differences on nematode genus richness between monoculture and polycultures in soil ecosystems. Wardle (2002) and Hooper *et al.* (2002) mentioned that it is unpredictable to determine the effect of plant diversity on the abundance and diversity of soil organisms. The results found are often inconsistent each time making it difficult to come up with a conclusion.

2.2. Farming systems

A farming system is a decision-making unit comprising the farm household, cropping and livestock system that transform land, capital and labour into useful products that can be consumed or sold (Fresco and Westphal, 1988). It takes into account the main technologies used, which determine the intensity of production and integration of crops and livestock

2.2.1. Organic farming system

The basis of sustainable nematode control is the maintenance of a healthy soil. This begins with routine application of organic matter. There is considerable evidence that addition of organic matter in the form of compost or manure will decrease nematode pest populations and associated damage to crops (Stirling, 1991; Akhtar and Mashkoor, 1993). This is because of improved crop production via improved soil structure and fertility, alteration of the level of plant resistance, release of nematotoxins, or increased populations of fungal and bacterial parasites and other nematode-antagonistic agents (Akhtar and Malik, 2000). Organisms found to be involved in nematode suppression are nematophagous fungi like *Verticillium chlamydosporium*, *Hirsutella rhossiliensis*, *Trichoderma* spp. and bacteria like *Pasteuria penetrans* which parasitize their nematode host (Huanga and Zhang, 2004).

Organic fertilizers influence both yield and plant micronutrient contents and thus help sustain crop productivity (Mottaghian *et al.*, 2008). Akinyemi *et al.*, (2009) confirmed that organic manures were very effective in reduction of plant parasitic nematode populations. They found that *Tithonia* mulch was able to suppress populations of *Pratylenchus* spp. and *Radopholus* spp. on banana and plantain. Application of amendments rich in Nitrogen is responsible for suppressing them hence amendments with low C: N ratio i.e. below 10, are often found to be successful (Bailey and Lazarovits, 2003).

Higher organic matter content increases soil's capacity to hold water as well as support thriving communities of the decomposers and predators that make up the soil's system. Nematodes are important participants in this underground energy-transfer

system as they consume living plant materials, fungi, bacteria, mites, insects, and each other, and are themselves consumed in turn (Ingham *et al.*, 1996). Some fungi, for example, capture nematodes with traps and sticky knobs (Wachira *et al.*, 2009b).

Nematodes and protozoa regulate mineralization processes in the soil. There is evidence that between 30% and 50 % of the nitrogen present in crops was made available by the activity of bacterial feeding nematodes (Ingham *et al.*, 1996). Research done in Denmark indicates that nematodes convert about as much energy as earthworms in certain forest soils (Dropkin, 1980) and the vast majority of nematodes found in the soil are not plant parasites.

The nematode stability is challenged by the yearly turning of the soil, which reduces the numbers of organisms that displace or prey on plant parasitic nematodes, while bringing more nematodes to the surface from deeper soil. If the same host crop is planted year after year, plant parasitic nematodes may increase to damaging levels. Root feeding nematodes are very opportunistic, and are among the first organisms to invade after a disturbance (Dropkin, 1980; Ingham *et al.*, 1996).

It is therefore important to actively manage soil biology using *Tithonia*, neem *Azadirachtin indica*, compost, animal manures, green manures, and crop rotations as these practices help promote the growth of beneficial organisms while suppressing plant parasites nematodes. Certain organisms that are associated with well managed crop soils for example, *Rhizobacteria* and mycorrhizae may induce systemic host resistance to nematodes (Barker and Koenning, 1998). In general, there is need for large amounts of soil amendments to significantly reduce nematode infestation (Barman and Das, 1996)

though their effects on the soil microbial communities are hard to interpret (Sikora *et al.*, 2005).

2.2.3. Conventional farming systems

Inorganic fertilizers are often used to improve soil fertility and crop production in that they provide plants with the necessary nutrients needed for a healthy growth. Furthermore, they help reduce plant stress which enables plants to withstand nematode attack. Fertilizer application and watering of plants less frequently encourages the development of a deep root system that will reduce stress on plants and can help minimize nematode problems (Akhtar and Mahmood, 1996).

Bednarek and Gaugler (1997) reported that addition of inorganic amendments, particularly NPK, suppressed nematode densities. They confirmed that prolonged exposure to high inorganic fertilizer concentrations inhibited their reproduction. However, it affected the beneficial nematodes specifically entomopathogenic nematodes by reducing their infectivity. The research approach of using inorganic fertilizers to diminish nematodes while maximizing the benefits of the fertilizer has been used for a while. Oteifa (1955) made the first report that ammonia decreased the counts of *Meloidogyne incognita* females and egg masses produced on infected lima beans *Phaseolus lunatus* L.

Proper management of diseases and pests that is mostly done through soil macro-nutrient management in cases of nutrient deficiencies can also reduce plant stress and help reduce damage from nematodes (Yeates *et al.*, 2009). Nutrient deficiencies and soil compaction can inhibit root development and increase plant sensitivity to nematode

damage (Li *et al.*, 2007). The combined use of organic and inorganic systems is common due to the limitations that each one has (Pimentel *et al.*, 2005).

2.3. Soil nematodes

2.3.1. Introduction

Many genera and species of nematodes have particular soil and climatic requirements (Efthimiadou *et al.*, 2009) whereby certain species do best in sandy soils, while others favor clay soils. Nematode populations are generally denser and more prevalent in the warmer regions, where longer growing seasons extend feeding periods and increase their reproductive rates (Wang and McSorley, 2005). In colder regions, the life cycle of the nematodes tends to be prolonged by up to two weeks after 21 days.

Light, sandy soils generally harbor larger populations of nematodes than clay soils (Yeates *et al.*, 2009). This is attributed to the more efficient aeration of sandy soil, presence of fewer organisms that compete with and prey on the nematodes, and the ease with which nematodes can move through the root zone. Also, plants growing in readily drained soils are more likely to suffer from intermittent drought, and are thus more vulnerable to damage by parasitic nematodes (Buckley and Schmidt, 2003). Desert valleys and tropical sandy soils are particularly challenged by nematode over population (Efthimiadou *et al.*, 2009).

2.3.2. Plant parasitic nematodes

Plant parasitic nematodes are said to cause worldwide yield losses of more than 20% annually as they are mostly root feeders, completing their lifecycles in the root zone of most plants (Coyne *et al.*, 2014). They possess a stylet that is used in piercing and penetrating the roots of host plants (Figure 2.1). Some are endoparasitic, living and

feeding within the tissue of roots, tubers, buds, and seeds while others are ectoparasitic, feeding externally through plant walls. A single endoparasitic nematode can kill a plant or reduce its productivity while several hundred ectoparasitic nematodes might feed on a plant without seriously affecting production thus making the endoparasitic ones more severe (Summers, 2011). Most affect crops through feeding on or in plant roots, whilst minority are aerial feeders. In addition to direct feeding and migration damage, nematode feeding facilitates subsequent infestation by secondary pathogens, such as fungi and bacteria (Moon *et al.*, 2010).

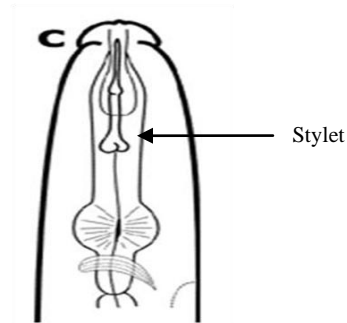


Figure 2.1 Head region of a typical plant parasitic nematode
Source: Ugarte and Zaborski, 2014

On a global scale, the distribution of nematode species varies greatly. Some are cosmopolitan while others are particularly restricted geographically, for example *Nacobbus* spp., or are highly host specific, such as *Heterodera carotae* which attacks only carrots *Daucus carota* L. A few species are highly host-specific, such as *Heterodera glycines* on soybeans *Glycine max* L. and *Globodera rostochiensis* on potatoes *Solanum tuberosum* L. (Sasser, 1990). But majority of the nematodes have a wide host range cutting across families like Solanaceous, Cucurbitaceae and Brassicaceae.

Direct feeding by nematodes can drastically decrease a plant's uptake of nutrients and water resulting in stunted growth and in severe cases, plant death (Sikora *et al.*, 2005; Coyne *et al.*, 2014). They have the greatest impact on crop productivity when they attack the roots of seedlings immediately after seed germination (Ploeg, 2001). Endoparasitic root feeders include economically important pests such as the root-knot nematodes *Meloidogyne* spp., the cyst nematodes *Heterodera* spp., and the root-lesion nematodes *Pratylenchus* spp. (Sasser, 1990).

These microbial infections are often more economically damaging than the direct effects of nematode feeding. In some cases the nematodes form disease complexes with the other pathogens resulting in more devastating effects as compared to the damage of the pathogens individually (Agrios, 2005). A good example is the root knot nematode-bacterial wilt (RKNBW) on Solanaceous and *Fusarium* wilt-lesion nematode complex (Akinsanmi and Adekunle, 2003).

Nematode control is essentially prevention because once a plant is parasitized it is impossible to kill the nematode without also destroying the host. For sustainable approach, there is need to integrate several tools and strategies, such as cover crops, crop rotation, organic soil amendments, least-toxic pesticides and resistant plant varieties (Renco and Kovacik, 2012). These methods work best in the context of a healthy soil environment with sufficient organic matter to support diverse populations of microorganisms (Mottaghian *et al.*, 2008; Renco and Kovacik, 2012). A balanced soil ecosystem will support a wide variety of biological control organisms that will help keep nematode pest populations below the economic damaging levels.

2.3.3. Free living nematodes

The free living nematodes are often grouped based on their feeding habits (Yeates *et al.*, 2009a) whereby some feed on fungi or bacteria; while others are predators or omnivores. Majority of the nematodes in biologically active and productive soils are not necessarily plant feeders but are bacterial-feeding and fungal-feeding species (Ferris, 2010). These free living nematodes form the higher portion of nematodes in the soil yet more attention is paid to the parasitic species (Andrássy, 2009).

They consist of the bacterivores which consume bacteria, fungivores which rapture fungal cell walls, omnivores and predators which form the lesser group (Yeates *et al.*, 2009b). They are essential to soil health in that some of them regulate mineralization processes. The nematodes contribute indirectly to nitrogen mineralization by excreting ammonium and immobilizing nitrogen in live biomass (Ferris *et al.*, 1998). Some nematode species like Entomopathogenic nematodes (EPNs) parasitize insects by infecting them with bacteria hence are essential for biological control (Denno *et al.* 2008).

2.3.3.1. Bacterial feeders

It is known that between 30 and 50 % of the nitrogen present in crop plants was made available by the activity of bacteria-consuming nematodes (Ingham *et al.*, 1996). Under field conditions, bacterivorous and predatory nematodes are estimated to contribute approximately 8% to 19% of nitrogen mineralization in conventional and integrated farming systems respectively (Beare, 1997). The entomopathogenic nematodes are often categorized as bacterivores though they are in a symbiotic

relationship with Gammaproteobacteria which aid in the parasitization and killing of insect pests (Denno *et al.* 2008).

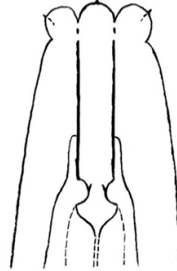


Figure 2.2 Head region of bacterial feeding nematode
Source: Ugarte and Zaborski, 2014

The mouth parts of bacterivores are often open as they have ornate lip structures (Figure 2.2) that help distinguish them from other nematodes (Ingham *et al.*, 1996). This group consists of families like Rhabditidae (*Rhabditis* spp, *Rhabdolaimus* spp. and *Acrobeles* spp.), Cephalobidae (*Cephalobus* spp. and *Cervidellus* spp.), Monhysteridae (*Monhystera* spp.) and Panagrolaimidae (*Panagrolaimus* spp).

2.3.3.2. Fungal feeders

When nematodes eat bacteria or fungi, ammonium (NH_4^+) is released because bacteria and fungi contain much more nitrogen than the nematodes require (Blair, 1996). The fungal-feeding nematodes have small, narrow stylets or spears, in their stoma (mouth) (Figure 2.3) which they use to puncture the cell walls of fungal hyphae and withdraw the cell fluid (Ingham *et al.*, 1996). This interaction releases plant-available nitrogen from fungal biomass. Some have a large square-shaped basal bulb which further aids in their identification.

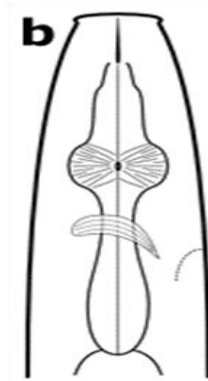


Figure 2.3 Typical head region of fungivorous nematode
Source: Ugarte and Zaborski, 2014

The most common genera here are *Aphelenchoides*, *Aphelenchus*, *Ditylenchus*, *Tylenchus*, *Antarctenchus* and some Dorylaimida. They feed on plant pathogenic fungi like *Fusarium oxysporum* S. and *Pythium ultimum* T., nematophagous fungi like oyster mushroom *Pleurotus ostreatus* K. and saprophytic fungi such as *Rhizoctonia solani* K., *Chaetomium globosum* K, *Coprinus cinereus* S. and *Flammulina velutipes* S. (Okada and Harada, 2007).

2.3.3.3. Predatory nematodes

Predators are nematodes that feed on invertebrates, such as rotifers, enchytraeids, protozoa, and other nematodes. The predators feed indiscriminately on both plant parasitic and free-living nematodes but their potential for use as bio-control agents against plant parasitic nematodes has not been considered effective as they don't intentionally prey on specific nematodes (Bilgrami *et al.*, 2008). These nematodes may regulate populations of bacterial- and fungal-feeding nematodes (Ingham *et al.*, 1996) and are often very large, dark bodied with huge teeth-like structures in the mouth parts as seen in Figure 2.4. Most common genera found here are the *Mononchus*, *Mononchoides*, *Neoactinolaimus*, *Mylonchulus* and *Clarkus* (Yeates *et al.*, 1993).

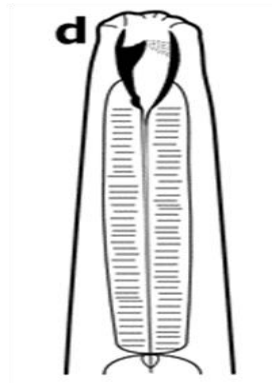


Figure 2.4 Predatory nematode head region

Source: Ugarte and Zaborski, 2014

2.3.3.4. Omnivores

Omnivores consume a range of food including plants, bacteria, fungi, unicellular eukaryotes, and invertebrates (Yeates *et al.*, 1993). Some members of the order Dorylaimida may feed on fungi, algae, and other animals and therefore considered omnivorous (Yeates *et al.*, 1993). The genera found in this group consist of *Dorylaimus*, *Mesodorylaimus*, *Eurodorylaimus*, *Aporcelaimellus* and *Enchodelus* among others (Yeates *et al.*, 1993).

CHAPTER THREE: MATERIALS AND METHODS

3.1. Description of study area

The research was carried out in Chuka, Tharaka Nithi County located in the mid-altitude eastern region of Kenya (Appendix I). The climate is favorable for agricultural activities with products such as livestock, tea, coffee, horticulture, cotton, millet, sorghum, cowpeas, bananas and cassava (MoA, 2009). It lies in the agro ecological zone 2 (AEZ 2).

Chuka receives an annual rainfall of 1500 mm in two seasons a year (long rains from March to June and short rains from October to December) (Table 3.1). The area falls under upper midland 2 and 3 (UM2 - UM3). The soils in the area are mostly humic nitisols (Jaetzold and Schmidt, 1983). They are moderate-high in fertility with high water holding capacity and high organic matter content (Appendix II).

Table 3.1 Ecological conditions of the study area

Parameter	Range
Rainfall (bimodal)	1500 - 2400 mm
Temperature	17.9 - 25.9°C
Altitude	1600 m asl
Latitude	0.33°S
Longitude	37.65°E

Source: MoA, 2006

3.2. Field trials

3.2.1. On-farm and on-station experiment

Two field trials were conducted concurrently; one on-farm and one on-station. The on-farm trials were conducted on farmers' fields whereby four farmers were selected prior to the start of the experiment through survey within the framework of the "Farming systems comparison trials in the tropics" (SysCom; www.systems-comparison.fibl.org). The farmers were selected following group discussions and interactions with extension officers from the Ministry of Agriculture (MoA) in the study area. Type 2 on-farm experimental design was used, that is, designed by the researcher but managed by the individual farmers. They were in close proximity with each other and served as a block (Appendix III). The on-station trials were carried out at the participatory technological development (PTD) trial site herein referred to as demonstration site (Appendix IV).

3.2.2 Crop establishment

The experimental crops were cowpeas (*Vigna unguiculata*, cv. K80) in the first season as a solecrop (October 2014 - February 2015); an intercrop of maize (*Zea mays*, cv. H513) and common beans (*Phaseolus vulgaris*, cv. KATB9) during the second season (April – August 2015); and beans as a solecrop in the third season (October 2015 – February 2016). Plot sizes at both farmer fields and demonstration site measured 5 x 5 m. Tillage was done using a hand hoe up to a depth of 20 cm and planting holes made to a depth of 10 cm with all seeds being hand sown. Row spacing varied within each season and each crop: cowpea solecrop were sown on 45 x 30 cm row spacing; maize bean intercrop spacing was 75 x 60 cm between maize with beans being row

intercropped at a spacing of 30 x 37.5 cm; sole bean crop were sown on 45 x 30 cm row spacing.

3.2.3. Treatments (Farming systems)

The experiment was set up to compare four farming systems: farmer practice system (based on farmer management practices that were determined from the farmers participating in the project); organic farming system (restricted to organic amendments); conventional farming system (based on non-organic amendments administered); and a non-amended control (served as a bare control). The amendments were incorporated into each system during planting and are listed in Table 3.3 below; their physical and chemical characteristics are shown in Table 3.4.

Table 3.2. Details of amendments applied to each system

Farming system	Amendments
Farmers' practice	DAP (0.64g*) Animal manure (6.6g) Compost (Crop residues + Wood ash + Manure) (11.6g)
Organic	<i>Tithonia</i> mulch (10.78g) Neem cake (1.12g) DAP (0.736g)
Conventional	CAN (1.28) Marshal EC (seed coating)
Non-amended	Nil

*Values in parenthesis refer to the quantity of amendment applied in each plot in grams

Table 3.3. Nutrient analysis of amendments applied

Property	N (%)	P (%)	K (%)	Ca (%)	C:N	Dry matter	pH
					Ratio	(%)	
DAP	18	46	0	-	-	-	8
Compost	1.15	0.24	2.03	1.42	12.7	94.8	9
Manure	1.41	0.26	1.52	1.24	9.86	94.8	8.78
Neem cake	2.16	0.87	1.46	2.68	-	90	-
<i>Tithonia</i>	0.17	0.3	1.3	-	-	-	-
Ash	-	5.4	0.4	0.24	-	-	-

3.2.4. Agronomic practices

Routine agronomic practices were conducted during the growing period of the crops. Hand weeding was done twice in each season. The species and populations of weeds were similar across sites and farming systems and the most common were black jack (*Bidens pilosa*) and couch grass (*Elymus repens*). The experiment was strictly rain fed therefore no irrigation was performed. No chemicals/pesticides, besides those listed in each particular system, were added to any of the experimental plots.

3.4. Data collection

Soil and root sampling for determination of characteristics affecting nematode assemblages was done in the plots demarcated for the study. Sampling bags were labeled with numeric codes describing plots prior to sampling.

3.4.1. Sampling pattern

A systematic sampling pattern, cross diagonal pattern (Figure 3.1) was used in collection of root and soil samples. Five subsamples per plot were collected making a composite soil sample of 1 kg. At the same point, approximately 70 g of roots was

taken. This was done using sampling tools such as a trowel, for scooping soil at a depth of 5 - 25 cm, knives and pangas for cutting roots. Both samples were later placed in the same labeled plastic bags, so as to preserve the roots, and stored in cool boxes for transportation to the laboratory for processing.

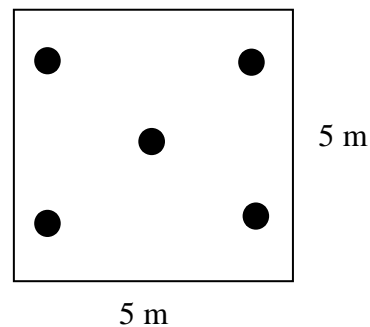


Figure 3.1 Cross diagonal sampling pattern used for sampling nematodes in each plot

3.4.2. Sampling regime

During the first cropping season of cowpea monocrop, soil samples were collected during the flowering (Month 1.5) and harvesting period (4th and 5th month). In the second cropping season (maize and bean intercrop) soil samples were collected five times, that is, at planting (Month 0), at bean flowering (Month 1.5), at flowering (Month 3), at bean harvest (Month 4) and finally during harvesting of maize (Month 5). In the last cropping season where beans were planted as a sole crop, soil samples were collected during planting, flowering and at harvest. All crop root samples were collected at harvest when the experiment was terminated.

3.5. Sample processing and identification of nematodes

3.5.1. Nematode extraction

The nematode extraction technique followed the modified Baermann funnel technique as described by Coyne *et al.* (2014). This involved taking a subsample of 100

ml of soil from the composite sample after thoroughly mixing and placing it on a sieve lined with tissue then inside a plate with 400 ml of water as shown in Figure 3.2 below. For the roots, 5 g out of the 70 g were finely chopped and blended then were used on the baermann plates. After two days the sieve was gently removed and the water in the plates transferred into small beakers and left overnight for the nematodes to settle. The remaining suspension was decanted through a 25 μm sieve and washed off into sample bottles using a wash bottle prior to identification.

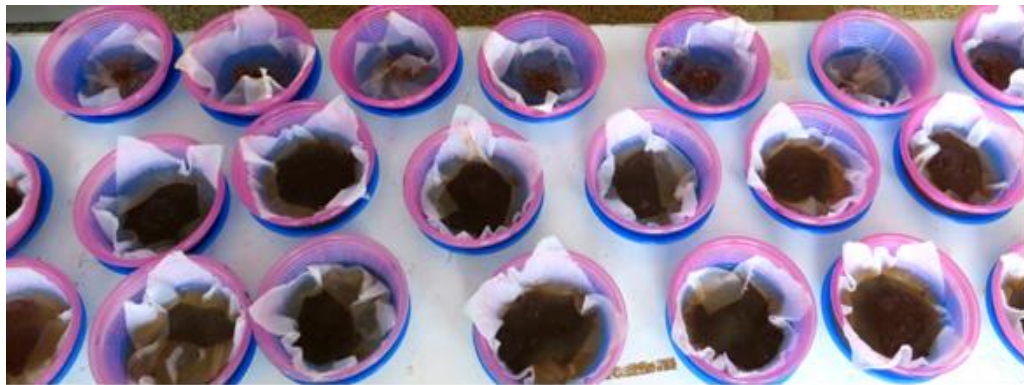


Plate 3.1 Set up for 100 ml soil and 5 g root extraction

3.5.2. Counting of nematodes

Estimation of nematode density was done by counting the total number of nematodes present in each treatment and replicate. This was done by taking a known volume of nematode sample (2 ml) using a pipette and placing it on a counting dish. Nematodes were observed under a Leica MZ12 dissecting microscope at a magnification of x40 and counting done using a tally counter (Figure 3.3). Total nematode counts were taken three times and the mean obtained was used to determine the population using the formula:

$$\text{Population} = \frac{49/7 \times \alpha}{y}$$

Where α = Total volume of nematode suspension (7.5 ml)

y = Quantity of suspension used for counting (2 ml)

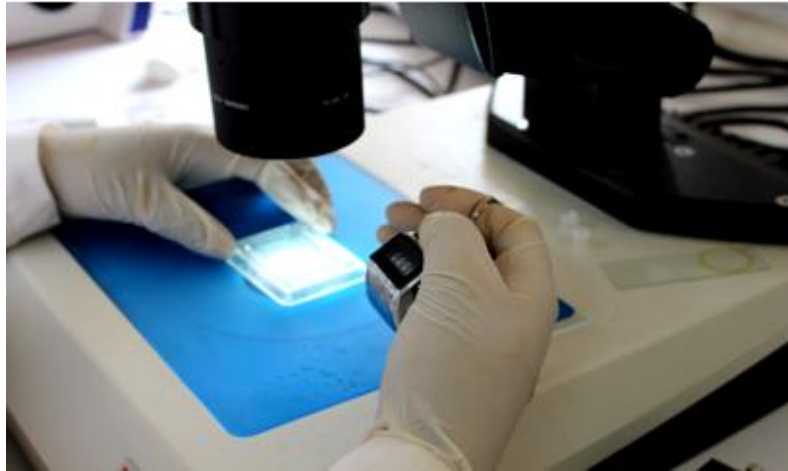


Plate 3.2 Dissecting microscope, counting dish and tally counter for counting nematodes

3.5.3. Fixing of nematodes

The nematodes were first killed by the hot water bath method as described by Coyne *et al.* (2014). This involved immersing 50 ml sample bottles containing nematodes in hot water (approximately 80°C) for 2 minutes. Fixing followed immediately after using 4% formaldehyde (10% formalin). It is the most common fixative used in nematology (Van Bezooijen, 2006). Three drops of formaldehyde were added into the sample bottles. The processed samples were stored at 20°C to allow the fixed nematodes to adequately settle at the bottom of the vial for later identification.

3.5.4. Nematode identification

Nematodes were identified up to the genera level where at least 100 nematodes per plot were identified based on morphological features as described by Siddiqi (2000),

Sikora *et al.*, (2005) and the University of Nebraska Lincoln nematode identification website (KSU, 2015). Key morphological features used in distinguishing the different PPN nematodes included type of stylet, size of stylet, shape of the tail, length of the nematode, shape of cephalic region, esophagus-intestinal overlap, position of vulva in females and the presence/absence of bursa in males, among others.

On the other hand, the morphological features that were used to distinguish the free living nematodes included the type of mouth region, presence/absence of a ‘tooth’, position of the ‘tooth’, number of parts of the esophagus, type of esophagus-intestinal overlap, shape of the tail, length of the tail, bursa presence, size of the nematode, presence of probolae on the cephalic region and position of the vulva, among others.

3.6. Data analysis

In order to meet assumptions of normality, count data were log transformed to their natural log $\ln(x+1)$ prior to analysis then subjected to analysis of variance (ANOVA) using R version 3.2.3 (R Core Team, 2015). ANOVA was used in determining the effect of the treatments on abundance of PPN and FLN. Differences at $p \leq 0.05$ level were considered statistically significant and means were separated using Fisher’s LSD (least significant difference) test using the package “agricolae” (De Mendiburu, 2015). Nematode abundance was based on trophic groups (Yeates *et al.*, 1993), and assigned to functional guilds then classified along the colonisation-persistence gradient (c-p values) according to Bongers (1990). They were further arranged to functional guilds which are portions of trophic groups that share the same c-p value (Bongers and Bongers, 1998; Ferris *et al.*, 2001). Diversity of trophic groups,

genus richness and maturity indices were computed. These community indices were calculated as follows:

3.6.1. Renyi diversity

The Renyi profile was used in analyzing the diversity of the nematode communities under different farming systems on soils sown to maize and bean intercrops. Alpha diversity was used as it refers to the average genus number found on a single farm or single sample plot (Kindt and Coe, 2005). Renyi Diversity index [HR (α)] was used in the evaluation of nematode diversity functional groups using the package “BiodiversityR” (Kindt and Coe, 2005) with the formulae (Tóthmérész, 1995):

$$HR(\alpha) = \frac{1}{1-\alpha} \log \sum p_i^\alpha$$

where α = scale parameter (with values, 0, 1, 2, 3, 4 and 5)
 p_i = relative abundance of the genus i
 s = number of genera

3.6.2. Genus richness

The genera richness represents the number of taxa in a particular treatment. However, it fails to mention the identity or diversity of the taxa present (Neher *et al.*, 2004). It is calculated using the formula (Neher *et al.*, 2004):

$$d = (S - 1) \log N$$

where d = genus richness
 S = number of genera
 N = total number of nematodes

3.6.3. Maturity indices

Maturity indices (MI) were computed for free living nematodes with colonizer-persister (c-p)1 through c-p 5 whereas maturity index with no opportunists (MINO) was calculated excluding free living nematodes with cp value of 1. Plant parasite index (PPI) was also calculated for the plant parasites using the same formula (Neher *et al.*, 2004):

$$\text{MI/ MINO/ PPI} = \frac{\sum (V_i \times f_i)}{\sum N}$$

where V_i = colonizer-persister value of i
 f_i = frequency of genus i
 N = total number of nematodes

3.6.4. Shannon diversity

The Shannon wiener, sometimes called the Shannon weaver index, is a measurement of diversity that takes into account both the genus richness and the proportion of each genus within the community (Begon *et al.*, 1996). The Shannon entropy quantifies the uncertainty (entropy or degree of surprise) associated with this prediction. It is most often calculated as below:

$$H' = - \sum_{i=1}^s (p_i \ln p_i)$$

where s = number of species

\sum = sum of the calculations

ln = natural log

p = proportion (n/N) of individuals of one particular species found (n) divided by the total number of individuals found (N)

3.6.5 Principle response curves

The results for each of the treatments (farming systems) were evaluated using the multivariate principle response curves (PRC) method on nematode communities that were repeatedly sampled in time using the package “vegan” (Oksanen *et al.*, 2015) in R. It enabled the quantitative interpretation of effects towards the genera level (Van den Brink and Ter Braak, 1999). The PRC analysis was used to show the effects of the treatments on specific nematode genera over time and this was achieved by modeling the abundance of each particular genus as a sum of three terms: mean abundance in the control, a month-specific treatment effect, and an error (Van den Brink and Ter Braak, 1998):

$$Y_{d(j)tk} = y_{0tk} + b_k C_{dt} + \sum_{d(j)tk}$$

where $Y_{d(j)tk}$ = abundance of genus k (=12) in replicate j (= 4) of treatment d (= 4) at time t (= 0-5 months)

y_{0tk} = mean abundance of taxon k in month t in the control

b_k = genus weight

C_{dt} = least-squares estimate of the coefficients

$\sum_{d(j)tk}$ = a random error term

CHAPTER FOUR: RESULTS AND DISCUSSION

4.1. Characterization of nematode communities associated with cowpea on organically and conventionally managed soils

4.1.1. Genera of free living nematodes found at farmers' fields and demonstration site in the farming systems

Free living nematodes from farmers' fields and demonstration site were identified and then classified based on Bongers (1990) and Yeates *et al.* (1993) as depicted in Table 4.1. Highest number of genera recorded belonged to bacterial feeders (10) followed by omnivores with four genera then fungal feeders and predators each consisting of two genera at both sites (Plate 4.1 and 4.2).

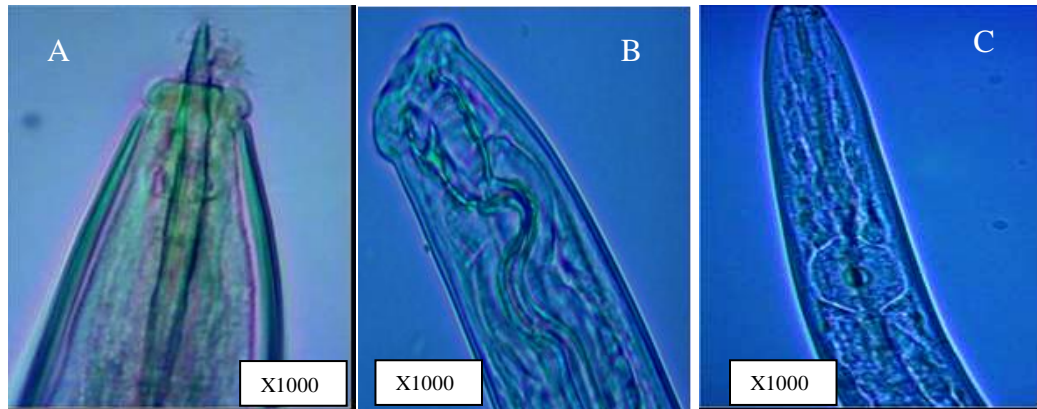


Plate 4.1. Free living nematodes present in the study site

A: Omnivore – *Labronema* spp., B: Predator – *Mylonchulus* spp. and C: Fungivore – *Aphelenchus* spp.

Table 4.1 Occurrence of free living nematode genera at farmers' fields and demonstration site in Chuka, Tharaka Nithi County

Genus	cp value	Functional guild	Trophic group	Site detection	
				On-farm	On-station
<i>Acrobeles</i> spp.	2	Ba2	Bacterial feeder	√	√
<i>Cephalobus</i> spp.	2	Ba2	Bacterial feeder	√	√
<i>Chiloplacus</i> spp.	2	Ba2	Bacterial feeder	√	X
<i>Diplogasterid</i> spp.	1	Ba1	Bacterial feeder	X	√
<i>Eucephalobus</i> spp.	2	Ba2	Bacterial feeder	√	√
<i>Monhystera</i> spp.	1	Ba1	Bacterial feeder	√	√
<i>Oscheius</i> spp.	2	Ba2	Bacterial feeder	√	√
<i>Plectus</i> spp.	2	Ba2	Bacterial feeder	√	√
<i>Rhabditis</i> spp.	1	Ba1	Bacterial feeder	√	√
<i>Wilsonema</i> spp.	2	Ba2	Bacterial feeder	√	X
<i>Aphelenchoides</i> spp.	2	Fu2	Fungal feeder	√	√
<i>Aphelenchus</i> spp.	2	Fu2	Fungal feeder	√	√
<i>Dorylaimus</i> spp.	4	Om4	Omnivore	√	√
<i>Eudorylaimus</i> spp.	4	Om4	Omnivore	√	√
<i>Labronema</i> spp.	4	Om4	Omnivore	√	√
<i>Prodorylaimus</i> spp.	5	Om5	Omnivore	√	X
<i>Discolaimus</i> spp.	5	Ca5	Predator	√	√
<i>Mylonchulus</i> spp.	4	Ca4	Predator	√	√

¹Ba: Bacterivores. Fu: Fungivores. Om: Omnivores. Ca: Carnivores. Numbers following the trophic groups' abbreviations represent the colonizer-persister values belonging to each genus. The c-p values represent generation cycle where 1 is shortest while 5 is longest.

Nematodes with colonizer-persister (c-p) value 1 (Plate 4.2 C) are generally considered to be colonizers and can be found in very disturbed soils or environments/habitats (Ferris and Bongers, 2009). They are primarily bacterivores with high metabolic activity, short generation time and are tolerant to pollutants. The c-p 2 nematodes (Plate 4.1 C, Plate 4.2 A, B and E) also have a short generation time and high reproduction rates although lower than the c-p 1 nematodes. c-p 3 nematodes have longer generation times than c-p 2 and are more sensitive to disturbances. They include some bacterivores, fungivores and a few predators. On the other hand, nematodes in c-p 4 and c-p 5 (Plate 4.1 A and B, respectively) are considered to be persisters, that is, they can stay in soils for very long periods and have long generation times (Bongers and Bongers, 1998). They have low reproduction rates, low metabolism and are very slow in movement. Most omnivores and predators belong to c-p 4 and c-p 5, respectively.

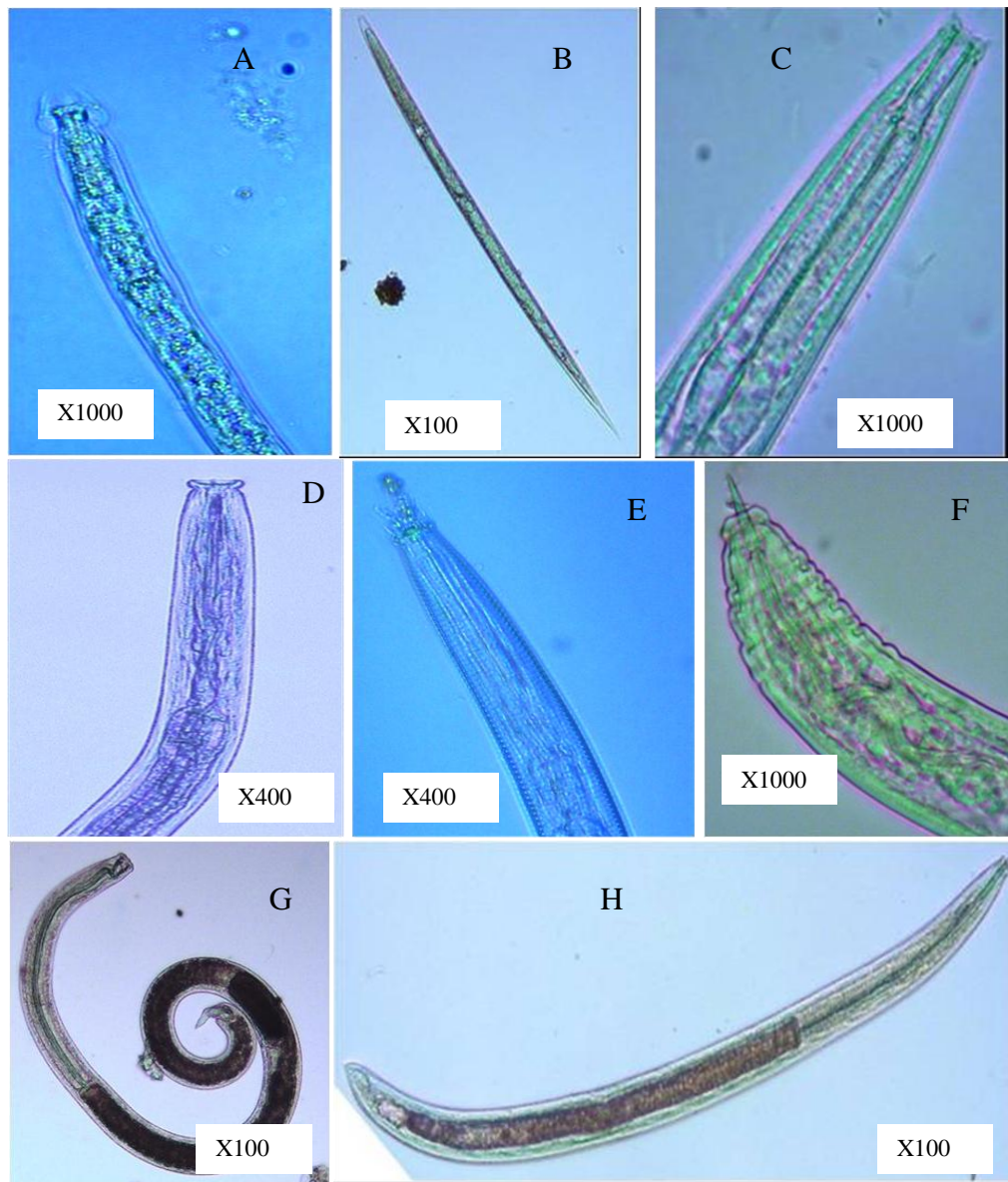


Plate 4.2 Free living nematodes found occurring under different farming systems.

A - *Wilsonema* spp., B - *Oscheius* spp., C - *Rhabditis* spp., D - *Discolaimus* spp., E - *Acrobeles* spp., F - *Dorylaimus* spp., G – *Mylonchulus* spp. and H - *Labronema* spp.

4.1.1.1. Genus composition of free living nematodes under farming systems

In farmer fields, the number of genera of free living nematodes that were recovered under the non-amended control, conventional, farmers practice and organic

system were 9, 13, 14 and 17, respectively. Bacterivores were the dominant trophic group in the organic treatments. Similar trends were observed at the demonstration site. Significant differences ($P \leq 0.05$) were observed for the proportion of free living nematode genera across the farming systems in both sites (Table 4.2).

The abundance of *Aphelenchus* spp. (15.42%) was significantly ($P \leq 0.05$) higher in the conventional system; abundance of *Cephalobus* spp. and was significantly higher in the conventional (30.65%) and organic (23.33%) farming systems; *Dorylaimus* spp. population was significantly higher in the farmer practice system; abundance of *Labronema* spp. was significantly higher in the non-amended control system; while abundance of *Monhystera* spp. and *Rhabditis* spp. was significantly higher in the organic system when compared to other farming systems as shown in Table 4.2 below.

The abundance of other genera comprising *Acrobeles* spp., *Aphelenchoides* spp., *Chiloplacus* spp., *Discolaimus* spp., *Eucephalobus* spp., *Eudorylaimus* spp., *Mylonchulus* spp., *Oscheius* spp., *Plectus* spp., *Prodorylaimus* spp. and *Wilsonema* spp. were comparable across the farming systems at farmer fields. Similar trends were observed at the demonstration site where 7, 10, 10 and 13 genera were identified from the non-amended, conventional, farmers practice and organic system, respectively (Table 4.3). Bacterial feeders were the most dominant group of free living nematodes followed by omnivores, fungivores and predators.

Table 4.2 Percentage mean of free living nematode genera in 100 ml of soil under different farming systems at the farmers' fields in Chuka, Tharaka Nithi County

Genera	Farming system			
	Farmers practice	Organic	Conventional	Non-amended
<i>Acrobeles</i> spp.	0.13 a	4.27 a	0.00 a	0.00 a
<i>Cephalobus</i> spp.	4.63 c	23.33 b	37.65 a	2.57 c
<i>Chiloplacus</i> spp.	1.68 a	0.93 a	0.00 a	0.00 a
<i>Eucephalobus</i> spp.	1.32 a	1.86 a	2.75 a	5.66 a
<i>Monhystera</i> spp.	0.00 b	16.98 a	0.00 b	0.00 b
<i>Oscheius</i> spp.	0.00 a	0.30 a	0.07 a	2.00 a
<i>Plectus</i> spp.	0.67 a	1.91 a	2.74 a	2.31 a
<i>Rhabditis</i> spp.	9.83 b	19.31 a	5.42 b	6.66 b
<i>Wilsonema</i> spp.	3.41 a	8.72 a	0.00 a	0.00 a
<i>Aphelenchoides</i> spp.	0.36 a	3.56 a	0.31 a	0.00 a
<i>Aphelenchus</i> spp.	4.98 b	10.66 a	15.42 a	0.72 b
<i>Dorylaimus</i> spp.	29.66 a	2.17 b	0.13 b	4.13 b
<i>Eudorylaimus</i> spp.	0.46 a	0.00 a	0.89 a	1.00 a
<i>Labronema</i> spp.	1.71 c	1.42 c	32.13 b	57.83 a
<i>Prodorylaimus</i> spp.	2.66 a	0.67 a	0.71 a	1.71 a
<i>Discolaimus</i> spp.	0.00 a	1.25 a	0.52 a	1.00 a
<i>Mylonchulus</i> spp.	1.50 a	2.66 a	1.26 a	4.13 a

Means followed by same letter(s) within rows are not significantly different at $p \leq 0.05$ (Least significant different test)

Table 4.3 Percentage mean of free living nematode genera in 100 ml of soil under different farming systems at the demonstration site in Chuka, Tharaka Nithi County

Genera	Farming system			
	Farmers practice	Organic	Conventional	Non amended
<i>Acrobeles</i> spp.	0.00 a	2.85 a	1.45 a	0.00 a
<i>Cephalobus</i> spp.	19.00 a	18.96 a	21.00 a	25.62 a
<i>Diplogasterid</i> spp.	0.00 a	1.25 a	0.00 a	0.00 a
<i>Eucephalobus</i> spp.	5.89 a	3.87 a	0.00 a	3.28 a
<i>Monhystera</i> spp.	0.00 a	3.11 a	0.00 a	0.00 a
<i>Oscheius</i> spp.	0.17 b	12.54 a	0.00 b	0.00 b
<i>Plectus</i> spp.	2.69 a	5.66 a	1.98 a	0.00 a
<i>Rhabditis</i> spp.	13.11 b	35.78 a	7.56 b	11.43 b
<i>Aphelenchoides</i> spp.	0.78 a	0.00 a	2.71 a	0.00 a
<i>Aphelenchus</i> spp.	16.45 a	0.00 b	11.31 a	16.63 a
<i>Dorylaimus</i> spp.	29.22 a	8.67 b	36.71 a	35.64 a
<i>Eudorylaimus</i> spp.	0.00 a	0.17 a	2.71 a	0.00 a
<i>Labronema</i> spp.	9.55 a	5.21 a	7.24 a	5.29 a
<i>Discolaimus</i> spp.	0.00 a	1.27 a	0.33 a	0.00 a
<i>Mylonchulus</i> spp.	3.14 a	0.66 a	0.00 a	2.11 a

Means followed by same letter(s) within rows are not significantly different at $p \leq 0.05$ (LSD test)

The differences in numbers of free living nematodes suggest that different genera dominate under different farming systems. The high bacterivore population in the organic system could be attributed to organic amendments like manure which are generally known to provide soil organisms with a new energy source that result in increased diversity and activity of soil microbes (Widmer and Abawi, 2000).

Rhabditis spp. and *Monhystera* spp. were highest in the organic system probably because nematodes in c-p 1 are usually the most responsive to organic enrichment (Bongers, 1990) and are thus termed as enrichment opportunists. Studies by Bongers and Ferris (1999) and Porazinska *et al.* (1999) showed that organic amendments increase the population of bacterial feeders in c-p 1 and maintain them till bacteria is exhausted. The c-p2 Cephalobids (*Cephalobus* spp., *Eucephalobus* spp. and *Acrobeles* spp.) were the most numerous bacterial feeders across all the treatments in total and this confirms results by Gomes *et al.* (2003) but members of the genus *Cephalobus* spp. were predominant among the bacterivores. Populations of *Cephalobus* spp. were consistently more abundant in conventional than organic system at both farmer fields and demonstration site and these results are similar to Neher (1999).

Fungal feeding nematodes consisted of only 2 genera *Aphelenchus* spp. and *Aphelenchoides* spp. The key distinguishing feature between the two closely related families was the absence of bursa in males of *Aphelenchoides* spp. and presence of mucron in females of *Aphelenchus* spp. *Aphelenchus* spp. was more abundant in the conventional than in the organic system. These results concur with Neher (1999) who found nematodes belonging to the families Aphelenchidae, Panagrolaimidae and Anguinidae to be more common in conventionally managed soils than organically managed soils. This could presumably be due to the increased food source as conventional system contained synthetic fertilizers that have been shown to increase population of fungi thus boosting their numbers (Nakhro and Dkhar, 2010). Similarly, *Aphelenchoides* spp. was higher in the conventional than the other treatments.

All omnivorous nematodes found at both study sites belonged to the order Dorylaimida. *Labronema* spp. and *Dorylaimus* spp. were more abundant in non-amended control and conventional systems respectively but very few in organic systems. On the other hand, populations of predatory nematodes were consistently low in all the farming systems. Several studies done have found the abundance of omnivorous and predatory nematodes to be relatively fewer in soil samples hence often categorize them together as Omnivorous-predator (Neher, 1999; Pokharel *et al.*, 2012). This could be explained by their long generation time as they are c-p5 nematodes (Bongers, 1990).

4.1.2 Occurrence of plant parasitic nematodes in soils

At the farmers' fields, a total of 11 genera of PPN (Plate 4.3) were found in the soil samples. The non-amended had the highest number of genera (10) followed by the conventional and farmers practice (eight) while organic had six. At the demonstration site, the non-amended again showed highest number of genera (11) followed by farmer practice (nine) and the conventional and organic systems both having seven genera (Table 4.4). Significantly ($p \leq 0.05$) higher populations of PPN genera were recorded in the non-amended control system as compared to other systems at both sites. Conventional and farmer practice had significantly ($p \leq 0.05$) less numbers of PPN as compared to the non-amended system. The lowest populations of PPN were observed in the organic system. The proportion of genera of PPN varied significantly at $p \leq 0.05$ among the treatments at the two sites. *Helicotylenchus* spp. and *Tylenchus* spp. were the most abundant ectoparasitic genera at both sites whereas *Longidorus* spp., *Trichodorus* spp. and *Xiphinema* spp. were rare in all treatments (Table 4.4).

Table 4.4 Percentage mean of plant parasitic nematodes in 100 ml of soil under different farming systems in Chuka, Tharaka Nithi County

Site	Genera	Farming system			
		Farmers practice	Organic	Conventional	Non-amended
Farmers' fields	<i>Filenchus</i> spp.	8.63 a	12.00 a	6.43 a	2.48 a
	<i>Helicotylenchus</i> spp.	26.67 a	28.89 a	33.52 a	31.67 a
	<i>Hoplolaimus</i> spp.	0.00 a	0.00 a	0.00 a	6.00 a
	<i>Meloidogyne</i> spp.	8.63 a	2.80 b	9.37 a	10.93 a
	<i>Pratylenchus</i> spp.	16.15 a	6.39 b	21.39 a	15.64 a
	<i>Rotylenchus</i> spp.	12.27 a	16.16 a	5.00 b	1.67 b
	<i>Scutellonema</i> spp.	0.00 a	0.00 a	5.00 a	1.67 a
	<i>Trichodorus</i> spp.	9.36 a	0.00 b	0.63 b	1.67 b
	<i>Tylenchus</i> spp.	16.15 b	33.76 a	18.66 b	20.51 b
	<i>Xiphinema</i> spp.	1.67 a	0.00 a	0.00 a	7.76 a
Demonstration site	<i>Filenchus</i> spp.	3.33 a	6.60 a	0.00 a	2.45 a
	<i>Helicotylenchus</i> spp.	32.76 b	49.36 a	39.11 a	26.45 b
	<i>Hoplolaimus</i> spp.	3.33 a	6.60 a	5.63 a	2.45 a
	<i>Longidorus</i> spp.	0.00 a	0.00 a	0.00 a	1.92 a
	<i>Meloidogyne</i> spp.	6.13 b	1.55 b	12.63 a	18.63 a
	<i>Pratylenchus</i> spp.	21.44 a	6.87 b	20.86 a	8.24 b
	<i>Rotylenchus</i> spp.	5.34 a	1.55 a	5.63 a	2.29 a
	<i>Scutellonema</i> spp.	4.00 a	0.00 a	0.00 a	0.92 a
	<i>Trichodorus</i> spp.	4.00 a	0.02 a	0.00 a	0.92 a
	<i>Tylenchus</i> spp.	19.67 a	27.47 a	12.63 b	18.63 a
<i>Xiphinema</i> spp.	0.00 a	0.00 a	3.51 a	0.92 a	

Means followed by same letter(s) within rows are not significantly different at $p \leq 0.05$ (LSD test)

Lack of amendments that are responsible for suppressing plant parasitic nematode populations appears to be the contributing factor to the high numbers and

genera of plant parasitic nematodes in the non-amended control system. *Helicotylenchus* spp. are common ectoparasitic nematodes that are distributed worldwide and are a serious pest of many crops including beans (Karanja *et al.*, 2002; Kimenju *et al.*, 2004), sugarcane (Berry *et al.*, 2009; Stirling *et al.*, 2011), maize (Waceke *et al.*, 2013) and cotton (Zwahlen *et al.*, 2007; Karuri *et al.*, 2013) among others leading to losses worth millions of dollars annually. *Hoplolaimus* spp., *Scutellonema* spp. and *Rotylenchus* spp. also belong in the same family (Hoplolaimidae) and found in the rhizosphere. They are all commonly referred to as spiral nematodes due to their coiled body structure (Coyne *et al.*, 2014) and often have very long stylets and offset heads as seen in plate 4.3. Their damage is significant to plants if they occur in large numbers (Summers, 2011). Their population was highest in the non-amended system and lowest in organic system.

Tylenchus spp. and *Filenchus* spp. (Tylenchidae) have often been classified as fungivores (Wang *et al.*, 2004; Okada and Harada, 2007; Zhang *et al.*, 2011) and sometimes as PPN (Yeates *et al.*, 1990; Olabiyi and Oladeji, 2014) because their feeding behavior is considered unclear (Wang *et al.*, 2003). Here they were both classified under PPN due to the morphology of their mouth parts, that is, possession of stylet (though not as large and distinct as for other PPN). They do not cause significant damage in most cases and are placed under feeding group 1f (Yeates *et al.*, 1993) which is the lowest level of PPN based on damage. They did not seem to respond to the treatments and their population was quite high across all farming systems and in both sites.

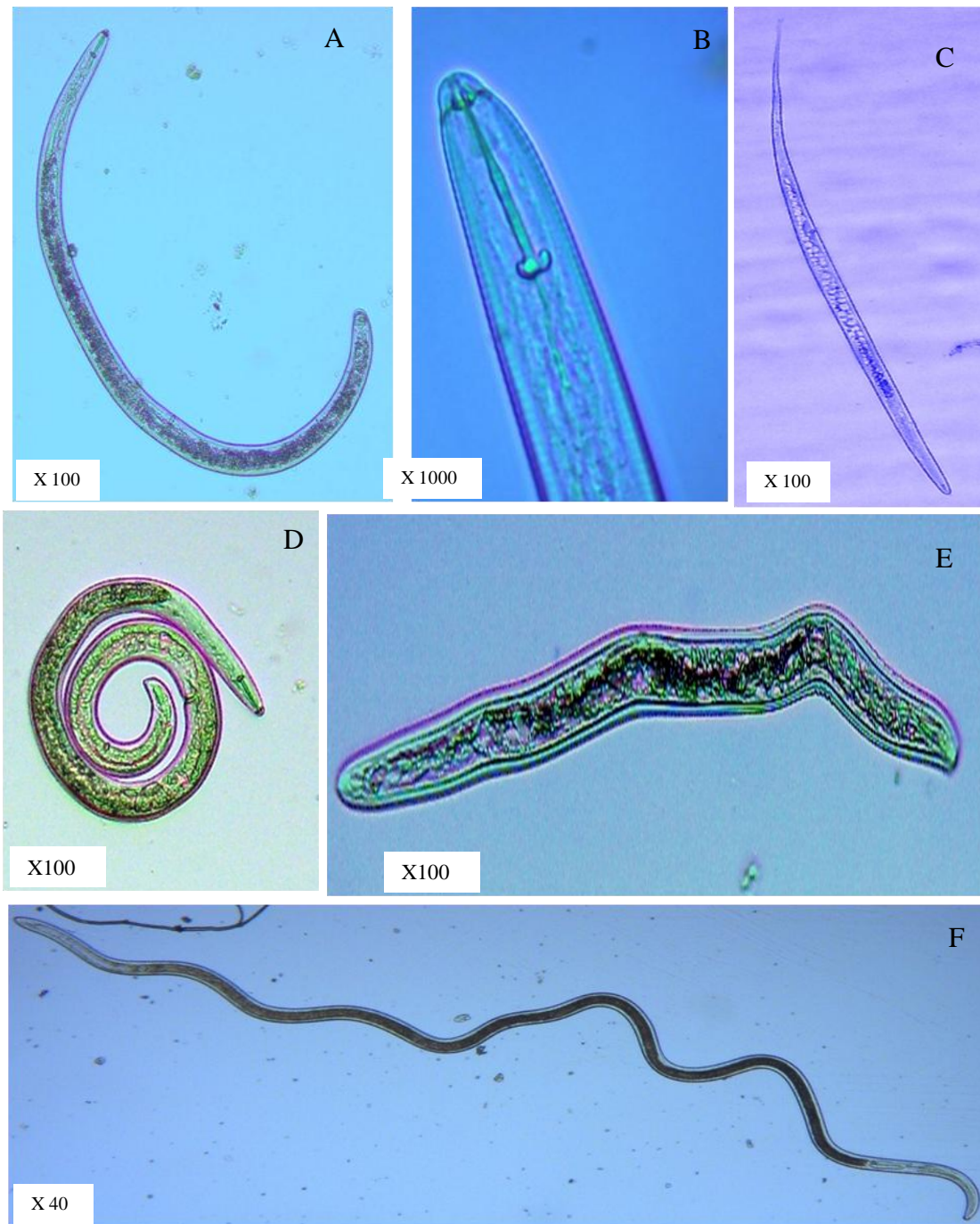


Plate 4.3 Plant parasitic nematodes occurring under the various farming systems.
 Genera A- *Scutellonema* spp., B- *Helicotylenchus* spp., C- *Filenchus* spp., D- *Hoplolaimus* spp.
 (head region), E- *Trichodorus* spp., F- *Longidorus* spp.

Longidorus spp. (needle nematode) and *Xiphinema* spp. (dagger nematode) (order Dorylaimida) and *Trichodorus* spp. (stubby root nematode) were the least abundant nematodes across all farming systems. They are classified under feeding group 1d (Yeates *et al.*, 1993). These nematodes possess a different kind of stylet named odontostylet and onchiostylet, respectively which enable them to act as vectors of plant nepoviruses (MacFarlane and Robinson, 2004). *Xiphinema* spp. is known to transmit tobacco and tomato ringspot virus (TRS and ToRSV) as well as grapevine fanleaf Virus (GFLV); *Longidorus* spp. transmits the tomato black ring virus (TBRV) and cherry leafroll virus (CLRV); and *Trichodorus* spp. transmits raspberry ringspot virus (RpRSV) (MacFarlane, 2003; Makete *et al.*, 2008). They are normally found deep in the soil horizon, below 30 cm deep, and this may explain why the low numbers observed in all the treatments (Mojtahedi *et al.*, 2002).

In the organic system, *Longidorus* spp., *Xiphinema* spp. and *Trichodorus* spp. were all completely absent and these results agree with Zoon *et al.* (2002) who established that green manure and organic amendments were effective in reduction of virus transmitting nematodes. Other research done suggests that intercropping, application of organic amendments and natural products are integrated management options that may be applied to control virus transmitting nematodes in fields that are already infested (Bilevai *et al.*, 2009). McSorley and Gallaher (1996) suggested that doubling of soil organic matter content may have negatively affected the populations of *Trichodorus* spp. and *Paratrichodorus* spp.

4.1.3. Plant parasitic nematodes associated with roots of cowpeas

Nematodes found in the root samples represented four families and six genera namely Hoplolaimidae (*Helicotylenchus* spp., *Hoplolaimus* spp. and *Rotylenchus* spp.), Pratylenchidae (*Pratylenchus* spp.), Meloidogynidae (*Meloidogyne* spp.) and Tylenchidae (*Tylenchus* spp.) across the farming systems. The most abundant genera at both sites were *Pratylenchus* spp. and *Meloidogyne* spp. (Plate 4.4) and were common in all treatments but in numbers varying significantly. The spiral nematodes were the least in abundance whereby *Rotylenchus* spp. and *Hoplolaimus* spp. were absent at the demonstration site and farmers' fields, respectively. *Helicotylenchus* spp. was also in low numbers though present in both sites.

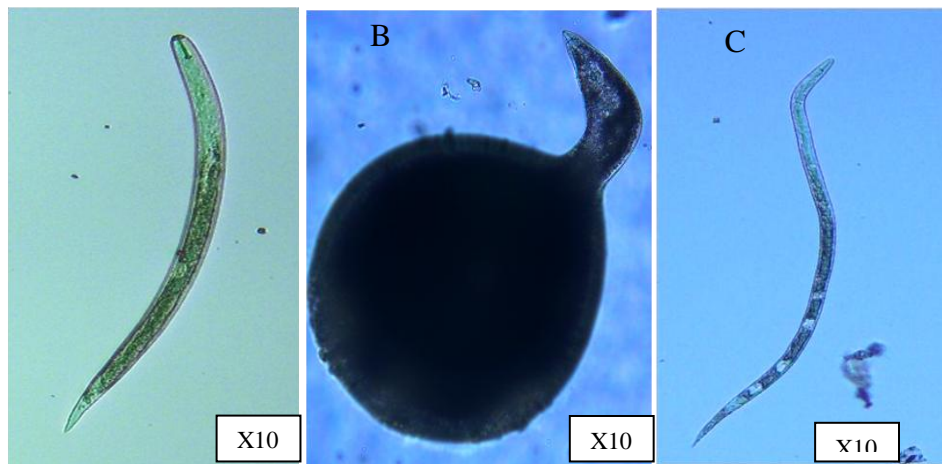


Plate 4.4 Endoparasitic nematodes in root samples.

A - *Pratylenchus* spp., B - *Meloidogyne* spp. juvenile and C- *Meloidogyne* spp. adult female.

Significant treatment effects on the population of PPN genera in root samples were observed. At farmer fields, the population densities of *Helicotylenchus* spp. (28.89%), *Meloidogyne* spp. (2.8%) and *Pratylenchus* spp. (6.39%) were significantly ($p \leq 0.05$) lower in the organic farming system. This same pattern was observed at the

demonstration site for population of *Pratylenchus* spp. and *Meloidogyne* spp. The population of *Rotylenchus* spp. was significantly ($p \leq 0.05$) higher in the organic (12.27%) and farmer practice system (16.16%); whereas the population of *Tylenchus* spp. was significantly ($p \leq 0.05$) higher in the organic farming system at both sites (45.67 and 37.47% at farmers' fields and demonstration site, respectively) as depicted in Table 4.5.

Root feeding nematodes are usually abundant and cause severe damage to crop plants in agroecosystems (Wurst *et al.*, 2009). The populations of *Pratylenchus* spp. and *Meloidogyne* spp. were significantly ($p < 0.05$) different across the treatments. *Pratylenchus* spp. (lesion nematode) are among the most common migratory endoparasitic nematodes and are responsible for causing the root lesion disease (Agyarko and Asante, 2005). They have a worldwide distribution and a wide host range (Agrios, 2005). They had the highest population in the non-amended systems, and lowest in the organic system. This low numbers in the organic system may be attributed to the presence of neem cake and *Tithonia* mulch which are said to have nematicidal compounds that negatively affect them (Agyarko and Asante, 2005; Odeyemi and Adewale, 2011).

Table 4.5 Percentage mean of plant parasitic nematodes in 5 g of roots under different farming systems in Chuka, Tharaka Nithi County

Site	Genera	Farming system			
		Farmers practice	Organic	Conventional	Non-amended
Farmer fields	<i>Helicotylenchus</i> spp.	46.81 a	28.89 b	45.58 a	51.25 a
	<i>Meloidogyne</i> spp.	8.63 a	2.80 b	9.37 a	10.93 a
	<i>Pratylenchus</i> spp.	16.15 a	6.39 b	21.39 a	15.64 a
	<i>Rotylenchus</i> spp.	12.27 a	16.16 a	5.00 b	1.67 b
	<i>Tylenchus</i> spp.	16.15 b	45.67 a	18.66 b	20.51 b
Demonstration site	<i>Helicotylenchus</i> spp.	49.81 a	59.36 a	48.88 a	6.45 b
	<i>Hoplolaimus</i> spp.	2.95 a	0.00 a	5.00 a	1.55 a
	<i>Meloidogyne</i> spp.	6.13 c	1.55 c	12.63 b	36.63 a
	<i>Pratylenchus</i> spp.	21.44 b	6.87 c	20.86 b	46.74 a
	<i>Tylenchus</i> spp.	19.67 b	37.47 a	12.63 b	3.63 c

Means followed by same letter(s) within rows are not significantly different at $p \leq 0.05$ (Least significant difference test)

Meloidogyne spp. (root knot nematode) is a sedentary endoparasite found inside roots of host plants with a wide host range. It is among the most damaging nematode pest in a number of crops and ranked under feeding habit 1a by Yeates *et al.*, (1993). High densities of root knot nematodes (*M. incognita*) lead to poor nodulation and decreased nitrogen levels in plants (Kaskavalci, 2007). In this study, their population was highest in the non-amended control system and very limited in the organic systems. Organic amendments in the organic system may be responsible for reducing growth and development of the RKN. Neher *et al.*, (2014) found similar results stating that compost aided in diminishing nematode populations. However, other studies have stated that the beneficial effects of compost may not necessarily be responsible for this reduction in nematode populations. *Tithonia diversifolia* and neem (*Azadirachta indica*) are botanicals that have been shown to be effective in management of RKN as they contain active ingredients which exhibit nematicidal properties (Akpheokhai *et al.*, 2012). Organic amendments appeared to negatively affect the population of most plant parasitic genera.

4.2. Effect of farming systems on nematode population and genera diversity on maize and beans intercrop

4.2.1. Influence of organic and conventional farming systems on abundance of nematodes in soil and roots under maize and beans intercrops

No significant ($p \leq 0.05$) differences were observed between the farmers' fields and demonstration site for any of the farming systems in both soil and root (bean and maize roots) samples as shown in Table 4.6. For soil samples: in farmers' fields, organic system had significantly ($p \leq 0.05$) higher number of nematodes (2182) when

compared to other systems. On the other hand, farmer practice had significantly ($p \leq 0.05$) lower population of nematodes (969.25) at the demonstration site. Roots of beans were statistically similar among the farming systems at both sites. The maize roots had significantly ($p \leq 0.05$) higher numbers of nematodes in the organic (2157.52) and non-amended control (2319.50) at farmer fields and demonstration site, respectively.

Table 4.6 Mean population of nematodes in soil and root samples under farming systems in farmers' fields and demonstration site in Chuka, Tharaka Nithi County

Sample	Farming system	Site	
		Farmer fields	Demonstration site
Soil	Farmer practice	1,079.25 bA	969.25 bA
	Organic	2,182.00 aA	2,014.50 aA
	Conventional	1,587.00 abA	1,718.75 aA
	Non-amended control	1,314.00 bA	1,565.75 aA
Bean roots	Farmer practice	1,574.92 aA	1,857.75 aA
	Organic	2,160.07 aA	2,281.75 aA
	Conventional	2,106.45 aA	2,311.25 aA
	Non-amended control	1,661.54 aA	1,653.25 aA
Maize roots	Farmer practice	612.58 bA	755.00 bA
	Organic	2,157.52 aA	2,319.50 aA
	Conventional	825.07 bA	1,183.75 bA
	Non-amended control	1,781.42 aA	1,924.00 aA

Means followed by same letter(s) are not significantly different at $p \leq 0.05$ (Least significant difference test); Lowercase letters following the mean represent significance within columns; uppercase letters represent significance within rows

The similarity between the two study sites could be attributed to similar agro ecological zones of the sites. Both farmer fields' and demonstration site were located in the same location (Chuka) and zone (AEZ 2) where the environmental conditions were similar, that is, rainfall, temperature, type of soil (humic nitisols) and sand content in soil were the same (MoA, 2013). These results are in agreement with those of Campos and Villain (2005) who found that factors that influence abundance and distribution of nematodes in different zones are rainfall, soil properties and temperature. This also confirms that the treatments in the farmers' fields were well carried out, comparable to the more controlled demonstration site thus strengthening the results of the treatment effects.

The abundance of nematodes across the farming systems was significantly different particularly between organic and conventional systems at both farmer fields and demonstration site. In both soil and roots of maize and beans, organic systems recorded the highest nematode abundance and this may be as a result of increased population of free living nematodes due to addition of organic amendments (Neher, 1999; Widmer and Abawi, 2000; Olabiyi and Oladeji, 2014). Application of organic substrates serves as a stimulus to processes that lead to a build-up of free living nematodes (Langat *et al.*, 2008).

The non-amended control system had high population of nematodes and this may be attributed to high numbers of plant parasitic nematodes in the system. The conventional and farmer practice systems had significantly fewer nematodes than organic system for both sites and samples. This may be a result of reduced population of PPN with no statistical significant increase in the population of free living nematodes

particularly bacterivores and fungivores (Bulluck *et al.*, 2002). Hamida *et al.* (2015) also found that integrating conventional fertilizers with compost, as was the case in the farmer practice system, could result in reduction or suppression of plant parasitic nematodes.

4.2.2. Effect of farming systems on nematode trophic group composition

The nematodes were classified based on trophic groups and significant differences ($p \leq 0.05$) were observed among the treatments and within trophic groups at both farmers' fields and demonstration site. In soil samples at the farmer fields, bacterial feeders were significantly ($p \leq 0.05$) higher in the organic system (885.94) as compared to other farming systems; PPN were significantly ($p \leq 0.05$) higher in the non-amended control system (947.75). Within the trophic groups, significant ($p \leq 0.05$) differences were observed in the organic farming system (bacterial feeders were significantly higher when compared to other trophic groups) whereas in the non-amended control (PPN were significantly higher). However, the number of fungivores, omnivores and predators were statistically similar between the farming systems as seen in Table 4.7.

In the bean root samples (Table 4.7), among the farming systems, the number of bacterial feeders was significantly ($p \leq 0.05$) higher in the organic system (932.18); fungal feeders significantly ($p \leq 0.05$) higher in the conventional system (681); predators were significantly ($p \leq 0.05$) higher in the organic system (359.07); and PPN were significantly ($p \leq 0.05$) higher in the non-amended control system (987.01). Among the trophic groups, significant ($p \leq 0.05$) differences were observed in each farming system where PPN dominated the farmer practice and non-amended control

system; bacterial feeders were significantly ($p \leq 0.05$) higher in the organic system; and predators were completely absent in the conventional system. Similarly, in the maize root samples (Table 4.7), bacterial feeders and PPN had significantly ($p \leq 0.05$) higher populations in the organic (858.41) and non-amended control (850.07) among the farming systems, respectively. Significant ($p \leq 0.05$) differences were observed within the trophic groups as well in all the farming systems with the exception of conventional system

At the demonstration site, the same trend was observed where population and composition of trophic groups differed significantly ($p \leq 0.05$) among farming systems. For soil samples (Table 4.8), the organic system recorded significantly higher numbers of bacterial feeders; non-amended control system had significantly higher numbers of plant parasitic nematodes among the farming systems. Within the trophic groups, significant differences were observed in the numbers of bacterial feeders in the farmer practice, organic and conventional farming systems which were more. Plant parasitic nematodes were significantly higher in the non-amended control system.

The roots of both beans and maize showed similar patterns in the variations of trophic groups among the farming systems (Table 4.8). The population of bacterial feeders was significantly ($p \leq 0.05$) higher in the farmer practice, organic and conventional system compared to other feeding groups; plant parasitic nematodes were significantly ($p \leq 0.05$) more in the non-amended control system for bean roots. Similarly, bacterial feeders were significantly ($p \leq 0.05$) higher in the organic system and farmer practice while plant parasitic nematodes were significantly ($p \leq 0.05$) higher in the non-amended control system as in Table 4.8.

Table 4.7 Mean number of nematodes under different trophic groups at farmers' fields in soil and root samples in Chuka, Tharaka Nithi County

Feeding groups		Farming system			
		Farmer practice	Organic	Conventional	Non amended
Soil samples	Bacterial feeder	226.25 aC	885.94 aA	226.62 aC	547.75 bB
	Fungal feeder	74.84 bB	124.76 bA	125.08 cA	70.32 eB
	Omnivorous nematodes	48.11 cC	105.12 bB	174.03 bA	205.69 cA
	Predatory nematodes	30.63 cB	123.17 bA	33.26 dB	109.93 dA
	Plant parasitic nematodes	232.75 aB	118.54 bC	266.08 aB	947.75 aA
Bean roots	Bacterial feeder	451.75 bB	932.18 aA	433.53 cA	330.33 bC
	Fungal feeder	197.55 cC	359.64 bB	681.00 aA	203.91 dC
	Omnivorous nematodes	125.40 dC	120.46 cC	421.91 cA	266.80 cB
	Predatory nematodes	95.38 dB	359.07 bA	0.00 dC	73.48 eB
	Plant parasitic nematodes	724.85 aB	388.73 bD	520.00 bC	987.01 aA
Maize roots	Bacterial feeder	305.69 bB	858.41 aA	162.57 aC	289.49 bB
	Fungal feeder	68.89 cB	98.70 bA	115.85 dA	57.85 dB
	Omnivorous nematodes	17.85 dC	36.26 dB	28.78 aB	51.81 dA
	Predatory nematodes	47.11 cC	115.65 bA	87.23 cB	107.51 cA
	Plant parasitic nematodes	457.65 aB	84.73 cC	25.00 dC	850.07 aA

Means followed by same letter(s) are not significantly different at $p \leq 0.05$ (Least significant difference test); Lowercase letters following the mean represent significance within columns; uppercase letters represent significance within rows

Table 4.8 Mean number of nematodes under different trophic groups at the demonstration site in soil and root samples at Chuka, Tharaka Nithi County

	Feeding groups	Farming system			
		Farmers practice	Organic	Conventional	Non amended
Soil samples	Bacterial feeder	205.89 bC	1,184.31 aA	428.38 aB	30.45 bD
	Fungal feeder	123.48 cB	147.44 bB	294.50 bA	30.45 bC
	Omnivorous nematodes	54.89 dA	0.00 cB	12.45 cB	0.00 bB
	Predatory nematodes	13.71 dA	0.00 cA	0.00 cA	0.00 bA
	Plant parasitic nematodes	309.78 aB	129.00 bC	297.69 bB	819.85 aA
Bean roots	Bacterial feeder	444.13 aB	1,243.23 aA	417.89 aB	100.86 cC
	Fungal feeder	153.52 cA	115.58 cB	109.00 cB	155.23 bA
	Omnivorous nematodes	43.84 cC	92.45 cA	99.90 cA	62.44 dB
	Predatory nematodes	21.93 cA	28.89 dA	0.00 dB	26.41 eA
	Plant parasitic nematodes	259.08 bD	306.26 bC	381.47 bB	1,187.31 aA
Maize roots	Bacterial feeder	379.82 aC	1,095.23aA	342.67 aC	487.08 bB
	Fungal feeder	78.45 cA	117.83 bA	389.65 aA	218.20 cA
	Omnivorous nematodes	34.84 dB	50.06 cA	17.71cC	64.51 dA
	Predatory nematodes	12.32 eB	29.90 cA	0.00 cC	9.54 eB
	Plant parasitic nematodes	185.36 bB	094.50 bC	200.50 bB	1,058.36 aA

Means followed by same letter(s) are not significantly different at $p \leq 0.05$ (Least significant difference test). Lowercase letters following mean represent significance within columns; uppercase letters represent significance within rows.

Organic systems have been reported to significantly change the nematode community composition (Neher, 1999; Wachira *et al.*, 2009a). Some studies have shown the rapid increase of free living nematode populations following the application of organic matter (Akhtar and Malik, 2000). Addition of organic amendments has been said to result in increase of bacterial feeding and fungal feeding nematodes whilst reducing that of plant parasitic nematodes in the soil (Wachira *et al.*, 2009a). The bacteria feeding nematodes which were significantly higher in the organic system at both sites could be attributed to organic amendments. Some studies have reported the positive correlation between organic amendments and bacterivorous nematodes (Summers, 2011; Farahat *et al.*, 2012).

The number of plant parasitic nematodes was significantly higher in the non-amended control systems and significantly fewer in the organic system at both sites. This could possibly be explained by the ability of organic amendments to increase antagonists of plant parasitic nematodes (Kimenju *et al.*, 2004; Oka 2010) such as predatory nematodes and nematode trapping fungi (Wachira *et al.*, 2009b). Akhtar and Malik (2000) suggested that organic soil amendments often stimulate the activities of microorganisms which are antagonists of PPN.

Similarly, Summers (2011) suggested that organic amendments such as manure result in multiplication of micro-organisms such as fungi and bacteria that might be pathogenic to plant parasitic nematodes. Agyarko and Asante (2005) proposed the application of organic amendments as an alternative management strategy for control of plant parasitic nematodes especially for small scale farmers who cannot afford pesticides (nematicides). Farahat *et al.* (2012) suggested that organic farming systems

can be used to keep plant parasitic nematode populations under the economic threshold level as well as improve plant performance if used on a regular occasion.

The high population of predatory nematodes in the organic system for both soil and maize and bean root samples at both sites could be attributed to organic amendments. Studies done by Wachira *et al.* (2009a) showed that predacious nematodes respond positively to application of organic amendments. Absence or few populations of the predators in the conventional system suggests that conventional amendments might be negatively impacting the predatory nematodes. They are *k*-strategists that do not perform well in disturbed soils or environments unlike the *r*-strategists such as c-p 1 bacterivores (Ferris and Bongers, 2009). The similarity between the farmer practice and conventional systems could be attributed to the presence of Di-ammonium phosphate (DAP) that might be responsible for the effects observed on trophic group composition (Li *et al.*, 2007).

4.2.3. Effect of farming systems on diversity of nematodes

The results from the Renyi profile analysis at the farmer fields and demonstration site showed that the organic system had the highest diversity with the highest value at scale 0. The non-amended control, conventional and farmer practice systems had same diversity as depicted in Figure 4.1. However, with regards to evenness, organic system was the most uneven system with the most steeply-declining pattern whereas farmer practice was system with most even distribution (Figure 4.2).

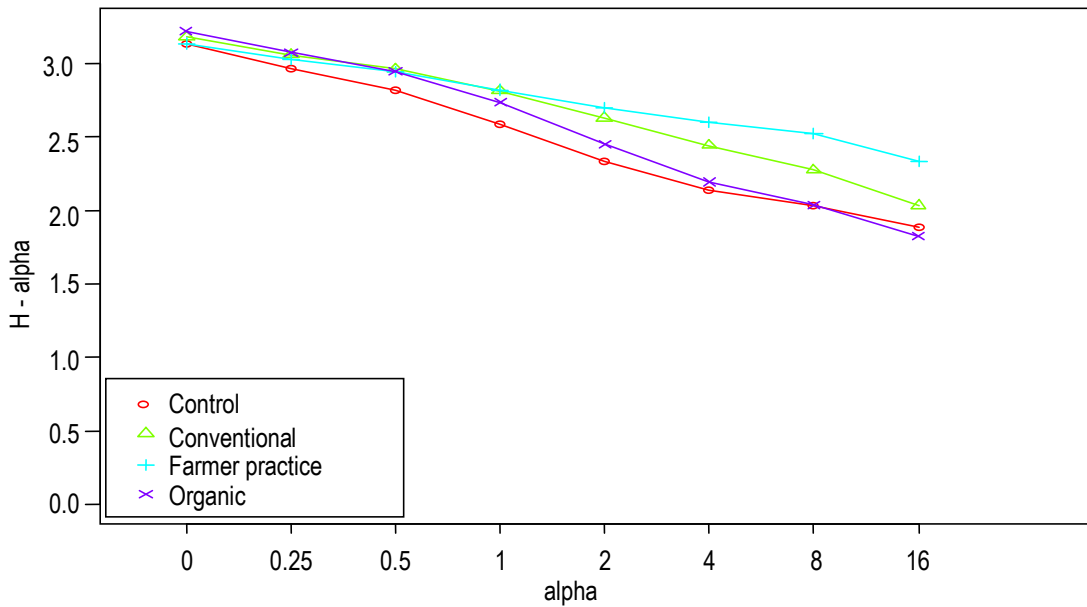


Figure 4.1 Renyi diversity profile calculated for the four farming systems in Chuka, Tharaka Nithi County

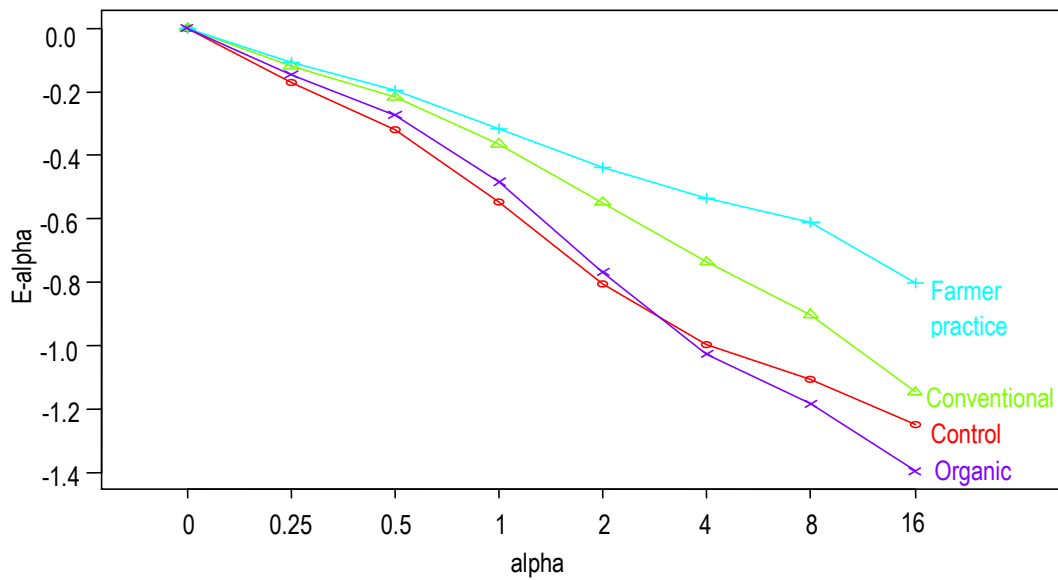


Figure 4.2 Renyi evenness profile calculated for the four farming systems in Chuka, Tharaka Nithi County

Diversity refers to both richness (the number of categories) and evenness (the distribution of items over the categories) of species or genera (Kindt and Coe, 2005).

Although the diversity was almost similar among all the farming systems, the organic system contained more genera of nematodes when compared to the other farming systems at both farmer fields and the demonstration site. These results are in agreement with those by Olabiyi and Oladeji (2014) who found that organic amendments neem and *Tithonia* result in erratic nematode population changes. Another possible explanation for the higher diversity could be due to effect of compost that has been shown to increase populations of free living nematodes (Akhtar and Mahmood, 1996). Similarly, in soybean studies by Lawal and Atungwu (2013), compost resulted in an increase in numbers of free living nematodes in the soil therefore suggesting that organic amendments favor diversity.

4.2.4. Effect of time on nematode communities during maize-bean intercrop

The non-amended control system (at 0) was used as the reference to which other systems were compared as depicted in figures 4.5, 4.6 and 4.7 which represent PRC curves for organic against control, farmer practice against control and conventional against control, respectively. The ordinate axis represents the first principal component of the variance due to treatment effect whereas the abscissa axis represents the sampling time in months. The horizontal line at 0 shows the response of the non-amended control nematode community. The species/genera scores that were associated with the reference system (non-amended control) are shown on the right axis.

The results for multivariate principle response curves (PRC) on nematode communities showed that only the organic against non-amended control PRC was statistically significant at $p \leq 0.05$ for both farmer fields and demonstration site. The main nematodes that were driving the response were *Tylenchus* spp., *Meloidogyne* spp.

and *Helicotylenchus* spp. The curve shows that these nematodes were initially infrequent (as shown by the negative PRC score at the 0 line), became more frequent after the second month but there was a decrease in activity towards the last month. The control versus farmer practice PRC was being driven by *Rotylenchus* spp., *Rhabditis* spp. and *Monhystera* spp. whereas *Aphelenchus* spp., *Mesorhabditis* spp. and *Rotylenchus* spp. were the main drivers in the conventional PRC.

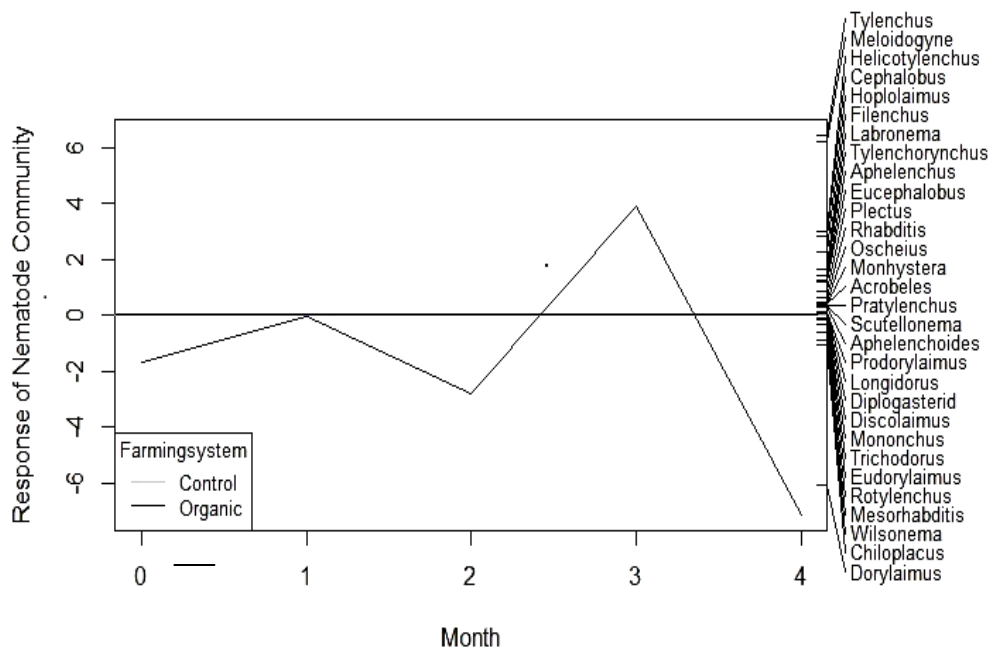


Figure 4.3 Principle response curves of the nematode genera showing the effects of organic system compared to the non-amended control in Chuka, Tharaka Nithi County

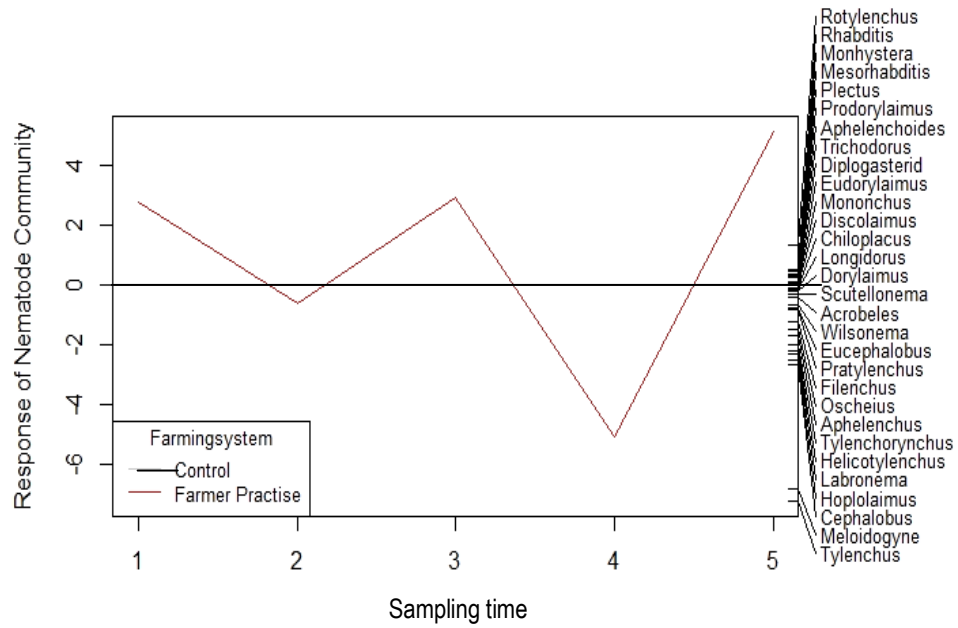


Figure 4.4 Principle response curve for nematode genera showing effects of farmer practice system against non-amended control in Chuka, Tharaka Nithi County

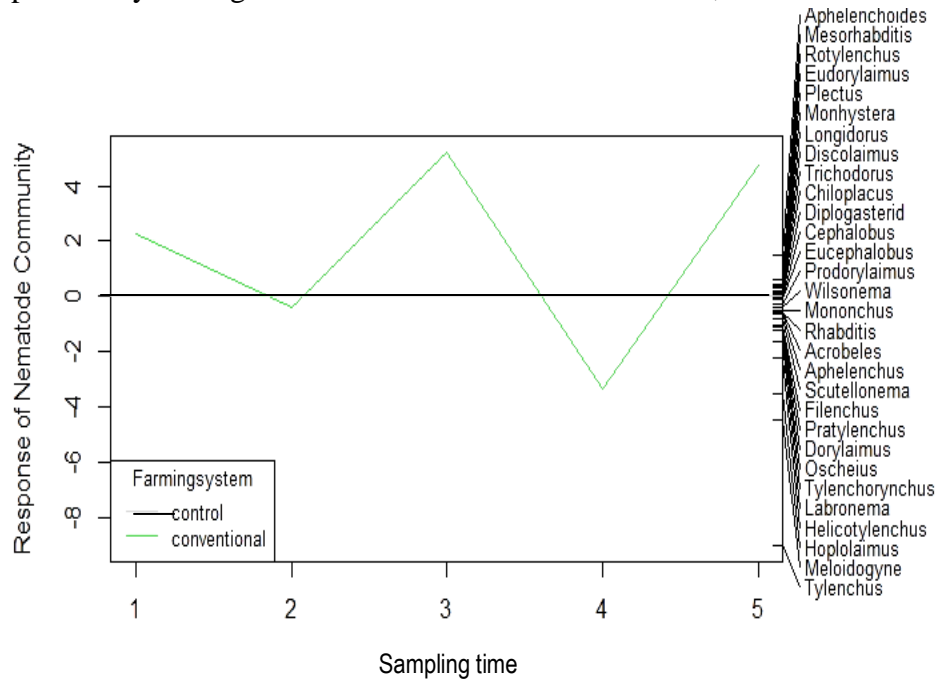


Figure 4.5 Principle response curve for nematode genera showing effects of conventional system against the non-amended control in Chuka, Tharaka Nithi County

The principle response curve model was used in the analysis as it allows for the evaluation of the nematode genera with the most pronounced treatment-related decrease in abundance (Moser *et al.*, 2007). It allows for significance of the treatment to be tested per sampling date therefore enabling one to determine for how long a certain treatment was effective against the pests that are being controlled (Neher and Darby, 2006).

The organic against non-amended control PRC showed the nematode genera that were lost in the non-amended control system were mostly free living nematodes. The main drivers of the model (*Tylenchus* spp., *Meloidogyne* spp. and *Helicotylenchus* spp.) all belonged to plant parasitic taxa. This is an indication that the organic farming system contained amendments that may have been effective in the reduction of plant parasitic taxa up to the second month as they initially had a negative PRC score. In the conventional system, Marshal EC pesticide was present but the PRC model showed no significance at any given time. This may suggest that the quantity of the pesticide was not sufficient enough to reduce the nematode pests or maybe the chemical was not able to control the nematodes beyond the seedling stage (Neher, 1999).

4.3. Influence of farming systems on nematode community structure associated with beans sole crop

4.3.1. Effect of farming systems on nematode genera abundance

Nematodes belonging to 20 genera were recovered from soils sown to sole beans from both farmers' fields and demonstration site as shown in Table 4.9 and 4.10. The majority of the genera were free living nematodes (14) with only 6 genera of plant parasitic nematodes. Significant differences were observed in population densities of nematode genera among the farming systems at $p \leq 0.05$. Bacterial feeding nematodes

particularly *Cephalobus* spp. (51.13) and *Rhabditis* spp. (186.7) were significantly ($p \leq 0.05$) higher in the organic system while conventional system had significantly ($p \leq 0.05$) higher populations of omnivores at both sites (Table 4.9). The farmer practice and non-amended control system had significantly ($p \leq 0.05$) higher numbers of plant parasitic nematodes (*Pratylenchus* spp. (322.98 and 286.12), *Helicotylenchus* spp. (196.9 and 217), *Meloidogyne* spp. (273.38 and 283.25) and *Tylenchus* spp. (322.98 and 257.63)) at farmers' fields and demonstration site, respectively as depicted in Table 4.10.

Rhabditis spp. and *Cephalobus* spp. have been shown to increase in numbers following addition of organic amendments (Neher, 1999). *Acrobeles* spp. and *Monhystera* spp. were quite rare and found only in the organic system. This suggests that organic amendments do boost the presence and populations of bacterial feeding nematodes. *Aphelenchus* spp. and *Mylonchulus* spp. were in high numbers in the conventional farming systems and these results concur with those by Langat *et al.*, (2008) who found that conventional amendments often lead to an increase in fungal feeding nematodes.

Table 4.9 Abundance of free living nematodes across the farming systems at farmers' fields and demonstration site in Chuka, Tharaka Nithi County

Genus	c-p value	Farming system							
		Farmer practice		Organic		Conventional		Non amended	
		On-farm	On-station	On-farm	On-station	On-farm	On-station	On-farm	On-station
<i>Acrobeles</i> spp.	2	0.00 c	0.00 c	9.34 d	33.71 e	0.00 e	9.34 e	0.00 d	16.38 bc
<i>Cephalobus</i> spp.	2	51.13 b	66.00 b	201.04 a	312.88 a	49.58 b	82.45 c	24.60 c	28.82 ab
<i>Eucephalobus</i> spp.	2	6.77 c	3.24 c	60.68 b	106.70 c	36.85 c	28.89 d	25.37 c	21.61 bc
<i>Plectus</i> spp.	2	6.77 c	12.88 c	60.68 b	121.98 c	0.00 e	7.13 de	21.53 c	4.08 c
<i>Rhabditis</i> spp.	1	68.95 b	84.04 b	186.70 a	215.40 b	53.08 b	161.84 a	43.07 b	36.02 b
<i>Aphelenchoides</i> spp.	2	4.60 c	0.00 c	0.34 d	0.00 g	33.78 c	0.00 e	21.53 c	19.61 bc
<i>Aphelenchus</i> spp.	2	4.60 c	125.62 a	56.01 b	82.26 d	62.73 b	127.40 b	48.45 b	45.63 a
<i>Dorylaimus</i> spp.	4	243.62 a	111.34 a	56.01 b	17.89 f	72.38 b	57.58 c	48.50 b	19.21 bc
<i>Eudorylaimus</i> spp.	5	13.79 c	0.00 c	0.00 d	18.00 f	14.48 d	0.00 e	0.00 d	0.00 c
<i>Labronema</i> spp.	5	0.00 c	69.72 b	32.67 c	23.55 f	115.80 a	34.67 d	107.67 a	36.02 a
<i>Prodorylaimus</i> spp.	5	0.00 c	0.00 c	23.34 c	0.00 g	57.90 b	0.00 e	10.77 d	0.00 c
<i>Discolaimus</i> spp.	4	0.00 c	0.00 c	14.00 d	28.32 ef	19.30 d	28.89 d	0.00 d	2.40 c
<i>Mylonchulus</i> spp.	4	9.19 c	0.00 c	18.67 cd	44.67 e	53.08 b	0.00 e	43.07 b	9.61 c

c-p values represent generation cycle where 1 is shortest while 5 is longest. Means followed by same letter(s) within columns are not significantly different at $p \leq 0.05$ (Least significant difference test).

Table 4.10 Abundance of plant parasitic nematodes across the farming systems at farmer fields and demonstration site in Chuka, Tharaka Nithi County

Genus	c-p	Farming system								
		Value	Farmer practice		Organic		Conventional		Non amended	
			On-farm	On-station	On-farm	On-station	On-farm	On-station	On-farm	On-station
<i>Filenchus</i> spp.	2	9.19 d	0.00 c	0.00 c	0.00 c	0.00 c	0.00 c	55.15 c	19.45 c	
<i>Helicotylenchus</i> spp.	3	78.14 c	38.38 b	4.67 c	11.00 c	24.13 c	3.17 c	196.90 b	217.00 b	
<i>Hoplolaimus</i> spp.	3	36.77 d	28.89 b	00.00 c	7.02 c	0.00 c	0.00 c	72.30 c	56.32 c	
<i>Meloidogyne</i> spp.	3	273.38 b	282.25 a	56.00 b	71.45 b	209.65 a	198.83 a	221.53 ab	237.88 b	
<i>Pratylenchus</i> spp.	3	322.98 a	286.12 a	72.09 b	66.22 b	119.30 b	110.06 b	264.60 a	306.76 a	
<i>Tylenchus</i> spp.	2	322.98 a	257.63 a	145.91 a	124.51 a	114.48 b	93.00 b	191.52 b	204.89 b	

c-p values represent generation cycle where 1 is shortest while 5 is longest. Means followed by same letter(s) within columns are not significantly different at $p \leq 0.05$ (Least significant difference test).

Beans are known to be highly susceptible to *Meloidogyne* spp. and trials for resistance have not yielded positive results (Thies *et al.*, 2004). The high population of plant parasitic nematodes particularly *Meloidogyne* spp., *Pratylenchus* spp. and *Tylenchus* spp. in the non-amended system was similar to that of cowpea sole crop as well as maize-bean intercrop season. The soils probably didn't have suppressive effects against the plant parasites as suppressive soils are expected to prevent nematodes from establishing and/or causing disease (Westphal, 2005). Kimenju *et al.* (2004) whilst undertaking studies on coffee nematodes found out that well managed farms have less plant parasitic nematodes as compared to those that are under no management.

4.3.2. Comparison of plant parasitic nematodes and free living nematode abundance in soil and roots of bean sole crop

Generally, there were more nematodes in the root samples than soil samples but the populations of plant parasitic nematodes (PPN) and free living nematodes (FLN) among the farming systems differed significantly in both root and soil samples at farmers' fields. At farmer fields, results from root samples (Figure 4.6) showed the non-amended control system had significantly higher populations of PPN followed by farmers' practice, conventional then organic. Organic systems recorded the highest numbers of free living nematodes followed by conventional, non-amended control and farmer practice. At the demonstration site, lower populations of nematodes were recorded in the farming systems when compared to farmer fields. Significant differences between plant parasitic and free living nematodes were observed in the organic system; plant parasitic nematodes were significantly higher in the non-amended control system when compared to free living nematodes as shown in Figure 4.6.

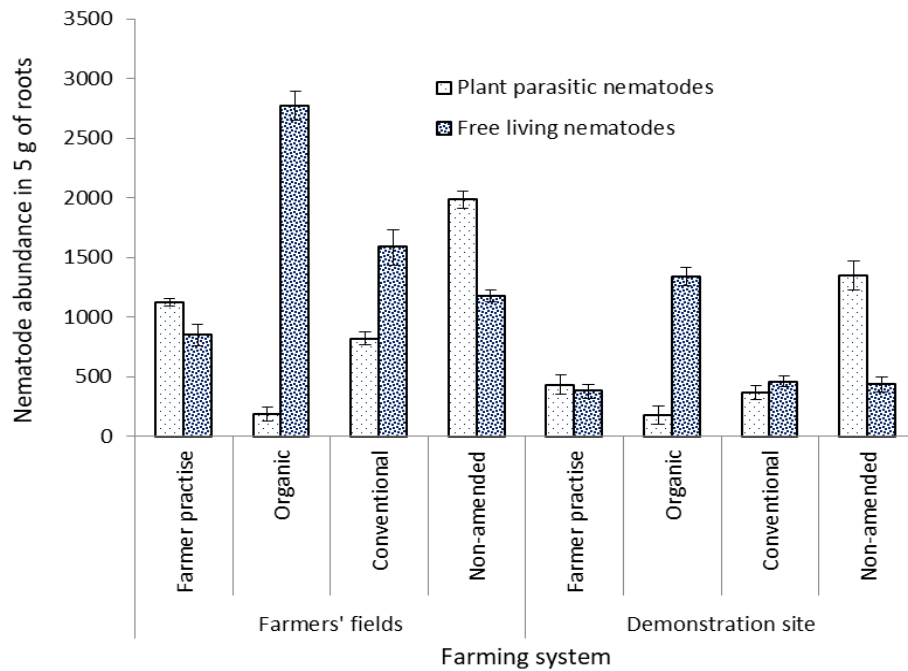


Figure 4.6 Abundance of plant parasitic nematodes and free living nematodes in 5 g of bean roots in Chuka, Tharaka Nithi County

Similar trends between plant parasitic and free living nematode populations were observed in soil samples (Figure 4.7) whereby the differences were most pronounced in the organic and non-amended control system. The numbers of plant parasitic and free living nematodes in the conventional and farmers practice systems were statistically similar. The soil samples at the demonstration site showed significant differences only in the organic system where population of free living nematodes was significantly higher as compared to plant parasitic nematodes. The farmer practice, conventional and non-amended control system showed no significant differences between proportions of plant parasitic and free living nematodes as shown in Figure 4.7.

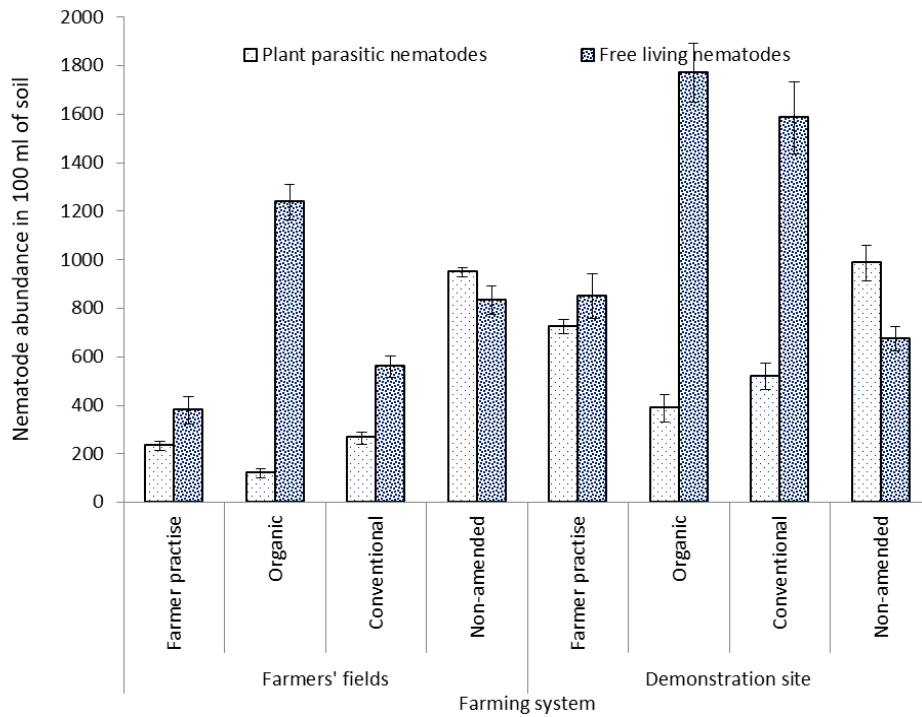


Figure 4.7 Abundance of plant parasitic nematodes and free living nematodes in 100 ml soil in Chuka, Tharaka Nithi County

The organic system appeared to be the most effective in lowering the population densities of plant parasitic nematodes while enhancing that of free living nematodes. These results are similar to those by Kimenju *et al.* (2008) who while carrying out studies on common beans and green manure plants, found that *Tithonia diversifolia* was effective in suppression of plant parasitic nematodes particularly root knot nematode (RKN). In another study by Widmer and Abawi (2000), it was suggested that green manures and other sources of organic matter altered the population of plant parasitic and free living nematodes. They are known for lowering the populations of PPN and increasing FLN. Carnation studies by Langat *et al.* (2008) revealed that organic substrates such as composts lead to a build-up of free living nematodes and were thus recommended for such production systems. Inorganic fertilizers can be useful in

decreasing populations of PPN when integrated with organic manures (Hamida *et al.*, 2015).

4.3.3. Effect of farming systems on nematode diversity indices on bean sole crop

Both genus richness and Shannon diversity did not significantly ($p \leq 0.05$) vary among the treatments at either farmers' fields or demonstration site. However, maturity index (MI) and maturity index with no opportunists (MINO) were significantly ($p \leq 0.05$) different among the farming systems in both sites. The highest MI was recorded in the non-amended control system (3.13 and 3.96) while lowest in the conventional system (1.91 and 2.80) at farmers' fields and demonstration site, respectively. A similar trend was observed with the MINO values. The plant parasite index (PPI) was also significantly ($p \leq 0.05$) varying among the farming systems. The highest values were recorded in the organic system (4.16 and 3.92 at farmers' fields and demonstration site, respectively) while lowest in farmer practice and non-amended control in farmer fields and non-amended control in demonstration site as depicted in table 4.11.

Even though the organic system had the highest genus richness in both farmers' fields and demonstration site, there were no significant differences in the genus richness. This is an indication that there were more genera found in the organic systems. According to Neher *et al.* (2004) values of genera richness merely represent the number of taxa in a sample without mentioning the identity or ecological diversity of the genera. Therefore, Shannon index was also calculated to show diversity of the nematodes. It was, however, not significantly different among the treatments in either site indicating an even diversity of nematodes across the farming systems.

Table 4.11 Diversity indices in Chuka, Tharaka Nithi County

Site	Farming system	Genus richness	Shannon diversity index	Maturity index	Maturity index with no opportunists	Plant parasite index
Farmers' fields	Farmer practice	7.25 a	2.07 a	1.94 b	2.23 c	2.06 c
	Organic	8.32 a	2.12 a	3.04 a	4.16 a	4.16 a
	Conventional	7.31 a	2.56 a	1.91 b	2.10 c	3.67 b
	Non amended	7.53 a	2.22 a	3.13 a	3.39 b	2.06 c
Demonstration site	Farmer practice	3.49 a	2.35 a	2.89 b	2.59 b	2.06 c
	Organic	4.81 a	1.88 a	3.09 b	3.91 a	3.92 a
	Conventional	3.42 a	1.74 a	2.80 b	2.37 b	2.85 b
	Non amended	4.55 a	2.16 a	3.96 a	4.15 a	1.77 c

Means followed by same letter(s) within columns are not significantly different at $p \leq 0.05$ (Least significant difference test).

The maturity index (MI) is a measure of environmental disturbance calculated for non-plant feeding taxa (Neher and Darby, 2006) where low MI indicates a disturbed environment while high MI indicates a more stable environment (Bongers, 1990). MINO is similar to MI but normally excludes the c-p 1 nematodes as they are known to be highly opportunistic nematodes. Ferris and Bongers (2009) stated that when resources become available to soil organisms through soil amendments, environmental changes or disturbance, there is an enrichment pulse of opportunistic guilds. These two indices were both significantly different between the farming systems in both sites whereby highest values were found in the non-amended systems. This may be due to lack of amendments that are known to disturb the soils thus affecting nematode communities' presence.

Plant parasite index (PPI) is computed for plant parasitic nematodes only and is the inverse of MI whereby low PPI is found in nutrient poor conditions while high PPI is found under enriched soils (Neher and Darby, 2009). The organic system had higher PPI than the other systems in both sites. This is an indication that this system had the most enriched soils compared to the others. These results collaborates a study by Neher (1999) who found significantly greater value of PPI in soils managed organically than conventionally. The main reason there were differences in the farming systems could be attributed to higher abundances of *Helicotylenchus* spp. and *Hoplolaimus* spp. in the organic systems. In studies on nematode community structure by Kimenju *et al.*, (2009), the results showed that the PPI was highest in intensively cultivated lands under maize and bean intercrops.

4.3.4. Effect of farming systems on nematode ecological indices on bean sole crop

The results showed that channel index (CI) was not significant ($p \leq 0.05$) at farmers' fields but inverse was true for demonstration site where conventional system had significantly higher value (83.21%) compared to the other farming systems. The enrichment index (EI) differed significantly among the treatments at both sites as organic was highest (85.52% and 82.04% at farmers' fields and demonstration site, respectively) followed by conventional system, farmer practice and non-amended control (Table 4.12). At the farmer fields, significant ($p \leq 0.05$) differences in the structural index (SI) were recorded as organic system had a significantly higher value when compared to farmer practice, conventional and non-amended control system. The pattern was similar at the demonstration site with organic system having a significantly ($p \leq 0.05$) higher value than the other farming systems. However, the basal index (BI) showed no significant ($p \leq 0.05$) differences between the farming systems at both farmers' fields and demonstration site.

The indicators that show the functioning of the ecosystem (enrichment, structure, basal and channel indices) are important as they can be used in environmental monitoring (Neher, 2009). The indices are computed through c-p values and functional guilds as shown by Ferris *et al.* (2001). c-p 1 and 2 fungivores are used to indicate enrichment while cp 3-5 are used to indicate structure. c-p 2 nematodes are basal to both structure and enrichment trajectories while the channel index corresponds to fungal/bacterial decomposition channels. This makes nematodes ideal indicators of the health of the soil (Neher *et al.*, 2012).

Table 4.12 Ecological indices of nematodes at farmers' fields and demonstration site in Chuka, Tharaka Nithi County

Site	Farming system	Channel index	Basal index	Enrichment index	Structural index
Farmers' fields	Farmer practice	55.45 a	22.87 a	58.03 b	40.71 c
	Organic	56.85 a	20.14 a	85.52 a	87.42 a
	Conventional	64.21 a	20.85 a	62.32 b	64.97 b
	Non amended	57.65 a	17.10 a	37.71 c	38.00 c
Demonstration site	Farmer practice	43.66 c	19.38 a	54.18 c	43.36 c
	Organic	64.90 b	13.27 a	82.04 a	89.90 a
	Conventional	83.21 a	15.66 a	67.22 b	72.61b
	Non amended	59.19 b	15.12 a	20.00 d	37.90 c

Means followed by same letter(s) within columns are not significantly different at $p \leq 0.05$ (Least significant difference test)

The high populations of fungivores in the conventional system could be the main cause for the higher channel index (CI) value at the demonstration site. Conventional amendments have been shown to rapidly increase populations of fungivores and omnivores (Neher, 1999). The similarity of basal index (BI) among the treatments could be attributed to the lack of disturbance-tolerant nematode taxa in the farming systems (Ferris and Bongers, 2009). Neher (2001) stated that predators and omnivores are higher in natural habitats as compared to agro-ecosystems as they are highly sensitive to soil disturbance.

High enrichment index (EI) describes a nutrient enriched soil ecosystem whereas a low EI shows that soils are nutrient depleted. The high EI values in organic systems could be attributed to addition of organic amendments such as compost which are known to favour bacterivores. Structural index (SI) varied significantly among the treatments in both sites. It was highest in the organic system then farmers practice, conventional system and non-amended control system. Plots managed organically often result in high abundance of bacterivores and fungivores as they have abundant resources (Bongers, 1990). This would lead to higher SI values in the organic systems which were incorporated with compost and *Tithonia*.

CHAPTER FIVE: CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

The study has demonstrated that the nematodes found on cowpeas in Chuka belonged to five trophic groups (bacterivores, fungivores, omnivores, predators and plant parasitic. There was high nematode diversity in the farming systems of 29 genera. The population of nematodes varied significantly across the farming systems where it was highest under organic system at both farmer fields and demonstration site.

Plots under maize and bean intercrops showed significant differences in population densities and diversity of nematodes with organic farming systems recording the highest abundance followed by control, conventional then farmer practice systems. Different farming systems showed different effects on nematode diversity where it was highest in the organic system while the other systems had similar diversity. Populations of plant parasitic nematodes in the organic systems were significantly reduced.

When beans were grown as a sole crop, there were significant differences in the nematode community structure. Both soil and root samples showed varying proportions of plant parasitic and free living nematodes among the farming systems with organic system having highest population of free living nematodes and least population of plant parasitic nematodes. The diversity and ecological indices varied significantly among the farming systems with plant parasitic index being significantly higher in the organic system.

Overall, this study demonstrated that different farming systems have different effects on population and diversity of nematodes in different cropping systems. Organic systems showed the highest potential in management of both plant parasitic and free

living nematodes. Conventional systems were slightly effective in reduction of plant parasites but they negatively affected some free living nematodes (predators).

5.2 Recommendations

- i. Based on the findings from this study, small holder farmers should practice organic farming as an alternative means of controlling plant parasitic nematodes. However, the type and amount of organic amendments need to be taken into consideration.
- ii. Policy development and extension officers to use this information to educate farmers in sub-Saharan Africa on economic importance of nematodes and management of the parasitic species.
- iii. Effects of organic and conventional farming systems to be performed under controlled environment such as green house or screen house conditions. Field conditions may vary hugely despite blocking and the weather conditions such as too high temperatures may affect the generations of nematodes. Controlled conditions will also enable analysis to be performed on percentage reduction of PPN by the amendments where known numbers of PPN will be introduced to pot experiments and data collected frequently so as to establish the effects that the amendments have on individual nematodes as well.
- iv. Nematode characterization of both plant parasitic nematodes and free living nematodes be done up to species level using molecular methods. Different species of PPN within a genera react differently to management techniques thus it would be ideal if the specific species are identified. On the other hand,

identification of FLN is quite challenging because of the similarities and versatility therefore molecular identification would be more certain and simpler.

- v. Further studies should be done in other agro ecological zones so as to establish if these findings will be consistent in other areas of Kenya where it is much hotter and drier or more humid. Studies to compare lower midland and upper midland areas would deem interesting since altitude has been shown to affect nematode abundance by several studies.
- vi. More studies to be done on other economically important crops such as potatoes and tomatoes. They are known to be very suitable hosts for key plant parasitic nematodes such as PCN (potato cyst nematode) and RKN (root knot nematode) respectively. Their management using these combination of organic and conventional amendments has not been documented much especially in the mid altitude areas of Kenya.

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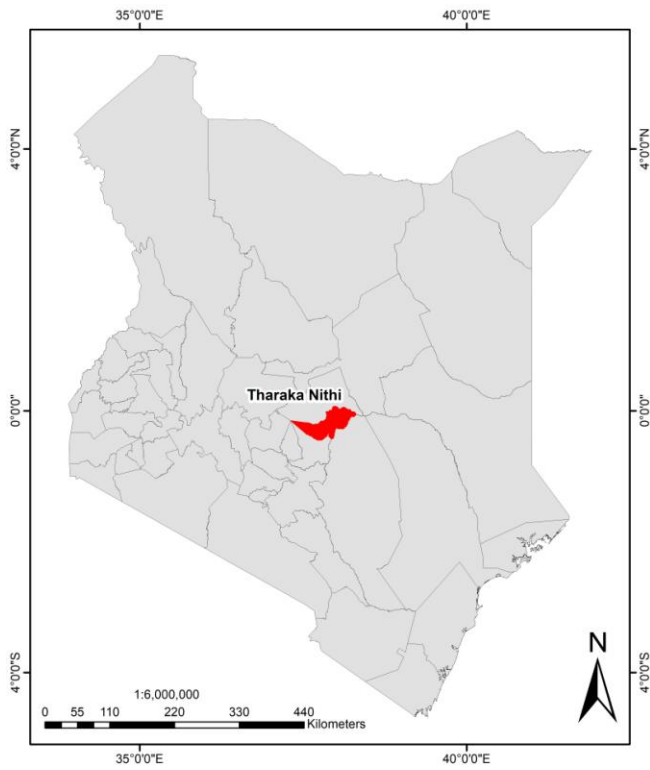
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APPENDICES

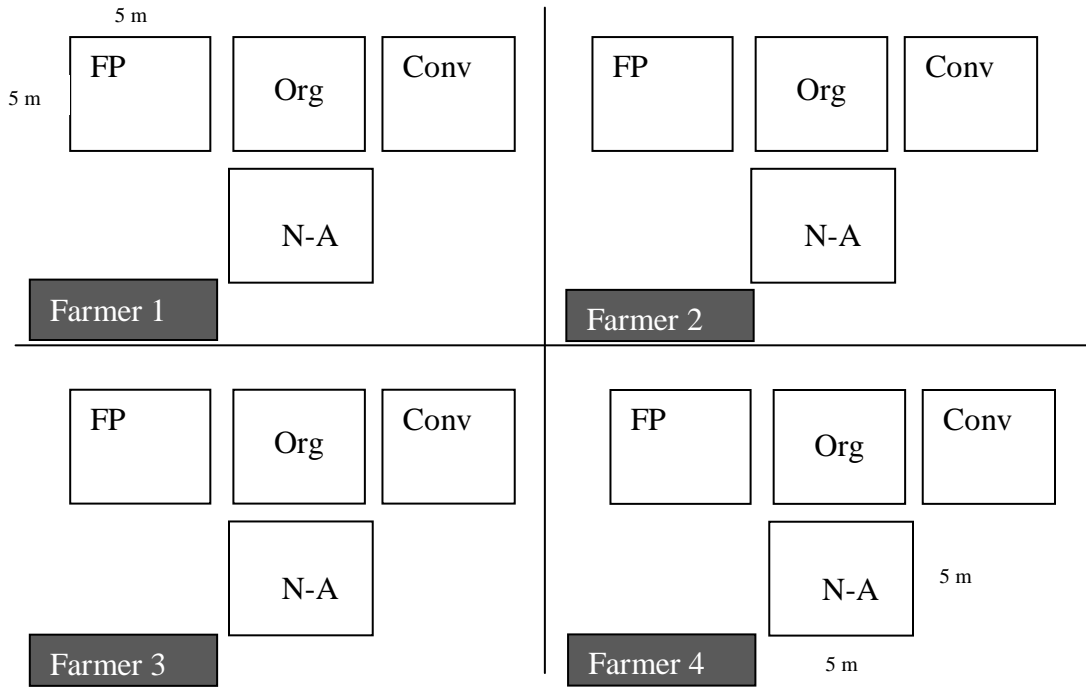
Appendix I: Map of study area (Chuka in Tharaka Nithi County, Kenya)



Appendix II: Physio-chemical characteristics of soils at Chuka in 2015/2016

Soil property	Quantity
pH	5.56
Sand (%)	38.5
Silt (%)	28
Clay (%)	33.5
EC (Salts) uS/cm	170.29
Phosphorus (Olsen) (ppm)	15.97
Potassium (ppm)	328.46
Organic carbon (%)	18
Calcium (ppm)	1640.50
Magnesium (ppm)	339.46
Sodium (ppm)	33.45
C.E.C (meq/100g)	18.32
Total N (g kg ⁻¹)	2.1

Appendix III: Layout of treatments at the farmer fields



Key: FP (farmers practice), Org (organic), Conv (conventional) and N-A (non-amended control)

Appendix IV: Layout of treatments at the demonstration site

Block 1	Block 2	Block 3	Block 4
FP	Org	N-A	Conv
Org	N-A	Conv	FP
Conv	FP	Org	N-A
N-A	Conv	FP	Org

Key: FP (farmers practice), Org (organic), Conv (conventional) and N-A (non amended control)

Appendix V: Data sheet for numbers of nematodes in 100 ml of soil and 5 g roots

Soil nematode		Zone	Treatment			
			Farmers practice	Organic	Conventional	Non amended control
Total		Soil				
		Root				
Plant parasitic		Soil				
		Root				
Free living	Bacterivores	Soil				
		Root				
	Fungivores	Soil				
		Root				
	Omnivores	Soil				
		Soil				
	Predators	Root				
		Soil				