POTENTIAL OF COMBINING HOST PLANT RESISTANCE AND INTERCROPPING IN THE MANAGEMENT OF ROOT-KNOT NEMATODES AND INSECT PESTS IN THE INDIGENOUS LEAFY VEGETABLES

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DECLARATION

I declare that this is my original work and has not been presented for an award of any degree in any other university.

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DEDICATION

I dedicate this work to my parents and siblings for the precious desire for knowledge they sowed in me at a tender age. Special dedication also goes to my wife Elizabeth Kananu and daughter Emma Kathure for giving me a reason to work hard and excel in life.

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List of Acronyms and abbreviations

ANOVA-Analysis of Variance

AVRDC-Asian Vegetable Research and Development Center

CABI- Centre for Agriculture and Biosciences International

CV-Coefficient of Variation

EM-Egg masses

GI-Galling index

GOK-Government of Kenya

ICIPE-International Centre for Insect Physiology and Ecology

ILV-Indigenous leafy vegetables

IPGRI-International Plant Genetic Resources Institute

IPM-Integrated Pest Management

J2- Second stage Juvenile

JICA-Japan International Cooperation Agency

JKUAT-Jomo Kenyatta University of Agriculture and Technology

KARI-Kenya Agriculture Research Institute

Lsd-Least significance difference

PPN-Plant Parasitic Nematodes

RCBD-Randomized complete block design

RKN-Root knot nematodes

S.e.d-Standard error of difference

STD-Sexually transmitted disease

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General Abstract

Indigenous leafy vegetables (ILVs) play an important role as income and food security crops in many rural and urban households in Kenya, yet their potential in alleviating poverty and ensuring household food and nutrition security has not been exploited. Diversification of diets through increased utilization and consumption of these vegetables would go along way in alleviating the hidden hunger and malnutrition. The objective of this study was to, assess the reaction of ILVs to root knot nematodes, identify the insect pests and plant parasitic nematodes (PPNs) associated with them, and evaluate the effective cultural management strategies for sustainable production of ILVs. A greenhouse experiment was carried out where six indigenous leafy vegetables namely spider plant (*Cleome gynandra*), amaranth (*Amaranth hybridus*), black night shade (*Solanum nigrum*), cowpea (*Vigna unguiculata*), jute mallow (*Corchorus spp*) and sun hemp (*Crotalaria juncea*) were assessed. The seeds were planted in six pots and half of the pots were infested with 2000 second stage juveniles of root knot nematodes. On termination data on plant height, fresh and dry shoot weight, galling index, egg mass index and the second stage juvenile count was recorded and analyzed. The field experiments were established in Kahatia in Murang'a County for two seasons to evaluate the effect of insect pests and plant parasitic nematodes on the indigenous leafy vegetables. Black nightshade, sun hemp and spider plant were selected for intercropping. Experiments were laid out in a randomized complete block design replicated three times. Treatments consisted of same row intercropping, same hill row intercropping, single and two rows intercropping, alternate row intercropping, border cropping and control plots which consisted of black night shade only. Insect pests that infested the indigenous leafy vegetables were identified through visual leaf inspection. Shoot damage, fresh and dry shoot weight of the plants and change in the second stage juvenile numbers in the soil were collected to assess root

knot nematodes and insect pests. These plants were rated using galling index on a scale of 1-10, where 1=resistant and 10=most susceptible. Spider plant, sun hemp and amaranth were rated as resistant while jute mallow, cow pea and black night shade were susceptible. Black aphids (*Aphis fabae*), flea beetles (*Chrysomelidae spp*), leaf miners (*Lyriomyza spp)*, red spider mites (*Tetranchus spp*), cutworms (*Agrotis spp*), diamond back moth (*Plutella xylostella*), African bollworm (*Helicoverpa armigera*), thrips (*Thrips tabaci*), whiteflies (*Bemisa spp*) and root knot nematodes (*Meloidogyne spp*) infested the ILVs. Same hill and same row arrangement pattern for the intercrop were the most effective and significant ($P \leq 0.05$) in reducing the effect of plant parasitic nematodes and insect pests infestation. The identified resistant varieties can be used as intercrops in agricultural production systems as a component of root knot nematode suppression in the soil. Intercropping resistant and susceptible vegetables can be integrated with other control methods for effective management of plant parasitic nematodes and insect pests. It is a practice that can easily be adapted by farmers with low external inputs.

CHAPTER ONE

INTRODUCTION

1.1 Background information

There is rekindled interest and increased demand for indigenous leafy vegetables (ILVs) by rural and urban dwellers of all socio-economic classes in Kenya. This has stimulated production of these vegetables but the supply is lower than the demand, leading to low consumption levels (Gotor and Irungu, 2010). The most widely cultivated indigenous leafy vegetables (ILVs) in Kenya include spider plant (*Cleome gynandra*), amaranth (*Amaranth hybridus*), black night shade (*Solanum nigrum*), cowpeas (*Vigna unguiculata*), jute mallow (*Corchorus spp*) and sun hemp (*Crotalaria juncea*) (AVRDC, 2003). Traditionally, these ILVs are used as food. They are rich in mineral nutrients and have medicinal value (GOK, 2002). For example, amaranth and black night shade can be used to feed those people with human immune deficiency virus (HIV/AIDS) since they are both nutritive and therapeutic (GOK, 2002). According to Adebooye *et al*. (2004) the vegetables can be used to eliminate malnutrition and promote healthy diets in Africa, through increased production and consumption. The leaves and seeds provide vitamins A and C, calcium, iron, protein, carbohydrates and lipids (IPGRI, 2003). Despite the named benefits, indigenous leafy vegetable production faces a myriad of challenges. These include lack of seeds that have limited ILVs production in many areas, damage by pests, diseases and nematodes (Cetintas and Yarba, 2010). Root knot nematodes (RKNs), especially *Meloidogyne species*, are key vegetable production constraints in Kenya (Atkins *et al*., 2004). They reduce plant growth by lowering water and mineral uptake and by enhancing crop damage by other pathogens such as fungal pathogens (Atkins *et al*., 2004).They form synergies with plant pathogenic fungi causing great yield losses of up to 80% if left uncontrolled and in fields that are highly infested (Cetintas and Yarba, 2010). On the other hand, insect pests are also a major constraint in the indigenous vegetable production. Some of the insect pests prevalent in vegetable growing areas include aphids, African bollworm, flower or blister beetles, thrips, pod sucking bugs, legume pod borers and the weevils, which can cause yield losses ranging from 40-60% if appropriate measures are not taken to control them (AVRDC, 2003).

1.2 Problem statement and justification

Indigenous leafy vegetables (ILVs) are an important commodity in the diet of many African communities. Most of the vegetables are grown by low-income small holder farmers and thus, play a crucial role in food security and in improving the nutritional status of poor families (Gotor and Irungu, 2010). However, efforts to unlock the full potential of the industry are hindered by lack of clean seeds, limited knowledge on production practices and pests and diseases. Insect pests, diseases and nematodes can cause upto 80% losses in vegetables yield (Cetintas and Yarba, 2010). Root knot nematodes (RKNs) mainly *Meloidogyne incognita, M.javanica* and *M.arenaria* are a major cause of yield decline in the production fields. They alter the plant physiology by producing specific enzymes that induce giant cell formation within the root at the feeding site (Karssen *et al.*, 2006). The giant cells then act as sinks by “attracting" energy rich plant metabolites, which are consumed by nematodes. The abnormal cells disrupt moisture and nutrient transport within the plant (Anwar and McKenry, 2010). The RKNs attack a wide variety of vegetable crops globally. In particular, they damage vegetables in tropical and subtropical countries and cause losses of upto 80% in heavily infested fields (Anwar and McKenry, 2010).

Insect pests are also a major constraint on the indigenous leafy vegetables production, they damage the plant parts especially the foliage lowering the quality and quantity of the yields and in heavily infested fields they can cause yield losses of up to 60% (AVRDC, 2003). The use of non-host or poor host plant species in cropping systems with susceptible crops is one of the most effective insect pests and nematode management method. Such non-host or poor host crops include sun hemp (*Crotalaria spp*.) and spider plant (*Cleome spp.*) that produce compounds that are allelopathic or repellant to different pests and nematodes (Wang *et al*., 2003). Moreover, *Crotalaria spp*. are known to associate with rhizobium bacteria in nitrogen fixation (Vargas *et al.*, 2000). This is important, as nutrient availability is central to plant tolerance to nematodes. However, there is no definite cropping system that has been developed in relation to insect pests and nematode management (Wang *et al*., 2003). Use of resistant varieties is a good option but little in terms of research has been done to develop them. Even where the resistant varieties have been developed, the cultivars are inaccessible to farmers (Wang *et al.*, 2003). Use of biological and cultural control methods would reduce pesticide use and the risk of pesticides residues while preserving environment quality and maintenance of ecological balance hence the need for research on these management options (Chitra and Anith, 2009).

1.3. General objective

The general study objective was to develop an effective cultural practice of managing arthropod pests and plant parasitic nematodes for sustainable production of indigenous leafy vegetables

1.3.1 Specific objectives

Specific objectives of the study were:

1. To assess the effect of root knot nematodes on the indigenous leafy vegetables

2. To identify arthropod pests that infest selected indigenous leafy vegetables

3. To identify intercropping arrangement patterns of the indigenous vegetables that are suppressive to arthropod pests and plant parasitic nematodes.

1.3.2. Hypothesis

a) Root knot nematodes and arthropod pests are the most limiting factors in the production of the indigenous leafy vegetables in Kenya.

b) Use of resistant indigenous vegetables in intercropping and rotational systems enhances suppression of arthropod pests and plant parasitic nematodes.

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CHAPTER TWO

LITERATURE REVIEW

2.1 Production aspects of the indigenous leafy vegetables

Indigenous leafy vegetables (ILVs) have short growth period, where some of these vegetables are ready for harvest within 3-4 weeks. They have the ability to produce seeds under tropical conditions, respond well to organic fertilizers, tolerant to both biotic and abiotic stress and require minimum production inputs (Abukutsa-Onyango, 2007). They are rich in vitamins and minerals and also have phytochemicals and anti-oxidant properties. The ILVs have potential to generate income for the growers if properly cultivated (Mbugua *et al.*, 2005). The leaves can be cooked or steamed before being eaten or ground into flour for immediate cooking or preservation (Abukutsa-Onyango, 2007). Although, these traditional vegetables resources are diverse with respect to the number of species, only a few of the cultivated taxa are widely used in the country (Grubben and Denton, 2004).

Efforts to conserve the traditional vegetables in the country are short of what is desirable. Genetic resources of the indigenous vegetables are to a large extent left to traditional practices and natural processes. Most of these species are grown for their edible leaves and some species for their edible grain (Mbugua *et al.*, 2005).The Indigenous vegetables grow fast and whole young plants are eaten or young tender leaves are harvested continuously from the established plants (Mbugua *et al.*, 2005). They are resistant to diseases, are drought tolerant, establish well, yield fairly good and are more acceptable to farmers in terms of taste compared to exotic vegetables (Mbugua *et al.*, 2005).

Basically, most of the indigenous vegetables can grow in a wide range of soils. They are commonly grown as monocrops in rows of 30cm to 40 cm apart and 8 to 12 cm between plants. The temperature required for their optimum growth and development ranges from 20^{0}c to 35^{0}c (Grubben and Denton, 2004). Field practices that need to be undertaken to improve the production of ILVs include proper land preparation, manure use, proper seed rates ranging between 2 to 5 kg/ha depending on the ILVs species, weeding, thinning, top dressing, pinching out and crop protection. Manure application of 30-40 tons/ha is recommended and should be mixed well into the soil before sowing. Similarly, Diammonium phosphate (DAP) fertilizer at 200 kg per hectare at planting can be used followed later by top dressing with nitrogenous fertilizer (Mbugua *et al*., 2005).

Sun hemp (*Crotalaria spp*) for example is a popular indigenous vegetable in some regions of the country and some farmers harvest it from the wild. Its seeds and leaves can be used as feed/fodder for livestock (Abukutsa-Onyango, 2007). When intercropped with other crops, they benefit from its nitrogen fixing abilities and suppression of nematodes (Anwar and McKenry, 2010). On the other hand, spider plant leaves and tender stems are highly nutritious with more protein and vitamins than kale and cabbage (Abukutsa-Onyango, 2007). It is an important crop in the rural areas of Africa but in recent years, production has decreased dramatically and there is a danger of genetic erosion as traditional lines are being lost (Abukutsa-Onyango, 2007).

2.2 Harvesting, utilisation and storage

Harvesting is done weekly or fortnightly for 2 to 3 months. The first harvest is made of thinnings uprooted within the rows at three weeks. Thinning is continued until 30cm spacing between plants is attained. The next harvest is by cutting the main shoot at 10 cm above the ground to stimulate the growth of the side shoots which are harvested later. Spider plant fruit or pods attain a yellow colour when mature. Amaranth inflorescence also turns yellow at maturity. They must be harvested at this time to prevent damage by birds (Keding *et al.*, 2007).

The cumulative yields from the ILVs range between 20 and 40 tons/ha (Mbugua *et al.*, 2005). Sorting and cleaning of vegetables is done. More so, removal of cooking water, frying in fat, addition of milk or cream and mixing with different types of other foods to mask any bitter taste associated with these vegetables is done (Mbugua *et al.*, 2005). These vegetables can be served with any starch staple foods like rice, irish potatoes, chapatis and ugali. Table 2.1 below shows the yield levels and net value production for the commonly cultivated ILVs.

Table 2.1 Mean, Median and Maximum yield levels and net value of production for selected indigenous vegetables in Tengeru location, Uganda in the year 2003.

CROP SPECIES		Yield(Kg/ha)	Average Per ha	Net Return(Us$) per labour hour	Per kg	N
Cowpeas	Mean	512	178.04	0.24	1.47	78
	Median	198	151.99	0.20	0.16	
	Maximum	7413	757.74	0.72	8.20	
	SD	1238	209.55	0.21	2.80	
Amaranths	Mean	3757	968.28	0.46	0.42	47
	Medium	1305	417.49	0.30	0.24	
	Maximum	37065	1680.84	2.30	1.76	
	SD	6247	3126.23	0.52	0.43	
Night shades	Mean	3184	3184	0.78	0.30	24
	Medium	1661	1661	0.63	1.12	
	Maximum	12335	12335	4.16	1.10	
	SD	3572	3572	0.86	0.35	

Source: Survey conducted by AVRDC in Co-operation with Hort-Tengeru, 2003.N=205 Plots.

The pods are dried, threshed, winnowed and cleaned to extract the seed. The dried seed is stored in clean, air-tight containers. The seeds may fail to germinate the first 2 to 3 months due to immature embryos hence the farmers are advised not to plant fresh seeds. However, the Ilvs seeds can remain viable for up to three years (Keding *et al*., 2007). The seeds are sold to seed companies, suppliers and farmers (IPGRI, 2003).

2.3 Plant parasitic nematodes that infest the indigenous leafy vegetables

Root knot nematodes (RKN) affect many crops and are reported to be one of the leading problems for vegetable growers (Atkins *et al*., 2004). They are favoured by warm temperatures that are prevalent in the tropical and subtropical regions (Coyne *et al.,* 2007). However, some species are able to adapt to local climatic conditions and may be found in temperate climate. The most widely distributed root knot nematode species are *M. incognita, M. javanica, M. arenaria* and *M. hapla.* In the tropics with warm climates, *M. incognita, M. javanica and M. arenaria* are the most important while in the temperate regions *M. hapla, M. chitwoodi* and *M. fallax* are prevalent (Coyne *et al,* 2005). Nematodes affect ILVs during the seedling, flowering, podding and vegetative growth of the vegetables (Mbugua *et al.*, 2005). The leaves, roots and whole plants are attacked by the nematodes causing stunted growth on the plants, discoularation on the leaves and galls on the infected roots (Wesemael and Moens, 2008). In severe cases, there is leaf chlorosis followed by wilting and reduction of yield in quality and quantity (Wesemael and Moens, 2008).

2.4 Symptoms and life cycle of root knot nematodes

The basic life cycle of RKN is not much different from that of the other nematodes. The eggs are retained within a gelatinous matrix in which they are embedded outside the root or inside the galls where the infective second stage juveniles (J2) hatch (Anwar and McKenry, 2010). When the J2 enter the plant roots they establish a feeding site of specialized giant cells where they develop and molt into third stage juveniles(J3) and later into fourth stage juveniles(J4) which moult either to adult males or females (Karssen *et al.,* 2006). Many RKNs including those that are of major economic importance are parthenogenetic and males are not necessary in order for the females to produce viable eggs. Males may migrate from the roots while females remain sedentary within the root tissues (Karssen *et al.,* 2006). The female lays eggs outside the gall in a gelatinous matrix, on the root surface (Karssen *et al.,* 2006). The female can lay between 30 and 80 eggs per day. Only the eggs, J2 and males can be found in the soil around the rhizosphere while the females and other juvenile stages remain inside the roots. The life cycle may be completed in about 25 days, depending on the host, climatic conditions and nematode species (Karssen *et al.,* 2006).

According to Karssen *et al*. (2006), at the temperature of 27^{o}C, the probability of having more generations is high as the life cycle is rapid. This is important in management of nematode problems. The roots are the feeding sites for RKN where they form giant cells and become stationary in the roots causing galls and the galls may be confused with nodules on leguminous vegetables (Wesemael and Moens, 2008). Roots, corms and tubers form galls leading to their abnormal formation and functioning of the root system and blockage of the vascular cylinders. Broad-leaved plants wilt, become yellow and show stunted growth and die (Wesemael and Moens, 2008).

2.5 Major arthropod pests of the indigenous vegetables

Indigenous vegetables are susceptible to damage by foliar insects such as leaf miners (*Lyriomyza spp)*, cutworms (*Agrotis spp*), black aphids (*Aphis fabae*) and cotton aphids (*Aphis gossypi*), red spider mites (*Tetranchus spp*), African bollworm (*Helicoverpa armigera*), flea beetles (*Chrysomelidae spp*), diamond back moth (*Plutella xylostella*) and systates weevils (*Systates nosus*) (AVRDC, 2003). The black aphids (*Aphis fabae*) are widespread. They suck sap on the stems, terminal shoots and petioles of seedlings, pods and flowers. Heavy infestation can cause death of young seedlings, stunting and delay in flowering and these pests are also vectors of viral diseases that affect ILVs (Schippers, 2002). The African bollworm caterpillars' cause extensive damage to seedlings and are considered the most important pests of indigenous leafy vegetables (Varela *et al.*, 2003; KARI, 2004).

Beetles and thrips are the most widespread causing over 80% losses on the indigenous vegetables (Varela *et al.*, 2003; KARI, 2004). Attacked flower buds become brown and eventually fall off leaving behind dark red scars (Varela *et al.,* 2003; KARI, 2004). Damaged flowers are distorted, malformed and show discoloration and may fall off. Bugs are difficult to control since they usually feed on a wide range of crops and are highly mobile. Adult weevils that are 2.0 to 3.5 mm long are the principal storage pests of indigenous vegetables seeds (Varela *et al.*, 2003; KARI, 2004). The bruchid may cause up to 100% loss to the stored seeds within 3 to 6 months under ordinary storage conditions (Varela *et al.*, 2003; KARI, 2004).

Controlling pests infesting pods in the field significantly reduces bruchid carry over in the storage (Palada and Chang, 2003). Weevils have been found to associate with fungi (mainly *fusarium spp*) that cause tissue decay and a cannier disease (Palada and Chang, 2003). Cutworms attack young seedlings and the damage causes plants to wilt and die. Cutworm damage is usually minor and does not normally warrant control. However, in severe outbreaks a young crop maybe destroyed (Palada and Chang, 2003). Leaf miners are small flies, 1.3 to 1.6mm in length (ICIPE, 2004). The maggot makes long, slender, white mines (tunnels) in the leaves. Severely mined leaves may turn yellow and drop. Severely attacked seedlings are stunted and may eventually die (ICIPE, 2004). Spider mites feed on the plants causing reduction in plant growth, flowering and the number of seeds produced (Varela *et al.,* 2003; KARI, 2004). The damage is more severe when mites attack young plants and during the dry season (ICIPE, 2004).

2.6 The potential of intercropping and plant tolerance in pests and nematode management

The presence of few galls and egg masses in some of the indigenous vegetables like the amaranth, spider plant and sun hemp makes them poor hosts of nematodes. They can be used in intercropping and crop rotational programmes to reduce nematode build up in a cropping system (Abdul-baki *et al.,* 2001). Suppression of plant parasitic nematodes by sun hemp has been known for decades. According to Marshall (2002), sun hemp is a poor host to RKN with only a few root galls from RKN infection compared to other ILVs. Most of the plant parasitic nematodes suppressed by sun hemp are sedentary endoparasites nematodes, which remain and feed in one place within the root system (Abdul-baki *et al.,* 2001). Sun hemp can enhance natural enemies of plant parasitic nematodes such as fungi that trap nematodes or feed on their eggs (Wang *et al.,* 2003).

Besides suppressing plant parasitic nematodes directly, sun hemp can also manage nematode damage on crops indirectly by increasing plant tolerance against these pests. Sun hemp amendments enhance free living nematodes in the soil that are involved in nutrient cycling

(Wang *et al.,* 2003) thus increasing nutrients for plant intake. A healthier plant has a higher resistance to damage by plant parasitic nematodes. Although, sun hemp has good potential as an intercrop and rotational crop for managing several important plant parasitic nematodes, the residual effect is short term (few months) (Wang *et al*., 2003). Nematode numbers can resurge to damaging levels on subsequent host crops (Mcsorley *et al.*, 2004). This scenario strongly suggests that integrating sun hemp in intercropping and rotational systems with other management strategies is necessary. Among the possibilities for integration are crop resistance, enhanced crop tolerance, selection for fast growing crop varieties, soil solarisation and biological control (Mcsorley *et al*., 2004).

Spider plant is a branched, hairy herb, growing to three feet or so with purplish stem that have longitudinal parallel lines. The crop is drought tolerant but it grows best in moist well drained soils and in full sunlight (Palada and Chang, 2003). The leaves and the tender stems of spider plant (*Cleome gynandra*) are highly nutritious with more proteins and vitamins than kales and cabbages (Abukutsa-Onyango *et al*., 2003). It is an important crop in rural areas of Africa but production has decreased dramatically and there is a danger of genetic erosion as traditional lines disappear (Palada and Chang, 2003). Spider plant is a useful intercrop and companion crop in reducing diamond back moth in cruciferous crops (Schippers *et al.*, 2002). It is pollinated by ants or bees and it produces oil which has been reported to be repellant to insect pests such as aphids, diamond back moth and weevils (AVRDC, 2003). Spider plant repellant oil and hairy surface which deters insect pests makes it a potential intercrop to benefit other crops. Insect pests which cause huge losses to indigenous vegetables such as leaf miners, leaf roller caterpillars, cutworms, aphids, flea beetles and mites require an integrated pest management strategy (Palada and Chang, 2003).

2.7 Methods of insect pests and nematodes management in the indigenous vegetables

2.7.1 Resistant host cultivars

Plant resistance or tolerance as a pest and nematode management tactic can be used. Some plants such as garlic, castor, spider plant, marigold, amaranth, sun hemp, cowpea, sweet potatoes and tomatoes have been reported to be tolerant to certain species of insect pests and nematodes (Marshall, 2002). Most of the indigenous leafy vegetables are susceptible to root knot nematodes. Resistance of plants to nematode infection is highly desirable. It is often conferred by genes for resistance, but not always particularly, where hypersensitivity takes place (Marshall, 2002). Spider plants, on the other hand, have the potential to repel insect pests. These resistant cultivars help in reducing effects of pest damage. The cultivars may repel insects away from the target host while others impair the insect metabolic process through consumption of toxic plant metabolites. Others exhibit tolerance where the plant is capable of withstanding pest injury and give satisfactory yield (Palada and Chang, 2003).

2.7.2 Cultural practices

Cultural control measures used in management of nematodes and insect pests include crop rotation, soil disinfestations, soil amendments and green manures, use of pest and nematode-free planting materials, nursery management, sanitation and physical soil treatments (dry heat, steam, solar heat and flooding) (Keller, 2004). Other cultural practices include; cultivation practice, trap cropping, antagonistic plants, destroying of alternate hosts, tillage, irrigation, water management, cover cropping and adoption of appropriate planting date (Schippers, 2002). According to Schippers (2002), the named practices are important in preventing pests and nematode problems before they occur.

2.7.3 Chemical methods

Plant-parasitic nematodes are at their most vulnerable during their active phase in soil when searching for the roots of host plants so nematicides are most effective at this stage (Karssen *et al.*, 2006). Use of chemicals is not sustainable because the indigenous vegetables are harvested frequently pausing a danger of pesticide residue presence in the edible leaves. Therefore, little attention has been paid in the use of chemicals. Fumigants are commonly applied as pre-plant treatments to control nematode numbers, but they must thoroughly penetrate large volumes of soils to be effective (AVRDC, 2003). In addition to broad spectrum fumigants, nervous system toxins such as Oxamyl and Fenamiphos are extremely effective in controlling root knot nematodes (AVRDC, 2003). Insect pests can be controlled using many different chemicals available in the market which are sold in different formulations though majority of indigenous vegetable farmers rarely use them (Karssen *et al*., 2006).

2.7.4 Biological control

Biological control is defined as the reduction of pest populations by natural enemies and typically involves an active human role. Biological control of pests is a method of controlling pests (including insects, nematodes, mites, weeds and plant diseases) that relies on predation, parasitism, herbivory or other natural mechanisms (Cory and Myers,2000). It can be an important component of intergrated pest management (IPM) programmes (Cory and Myers, 2000). Natural enemies of insect pests, also known as biological control agents include predators, parasitoids, and pathogens. There are three basic types of biological control strategies; conservation, classical biological control and augmentation (Cory and Myers, 2000). Plant parasitic nematodes (PPN) can be controlled using biological organisms such as predators, nematophagous fungi, endophytic fungi and bacteria (Cory and Myers, 2000).

2.7.5 Integrated pest management

The most successful approaches to nematode and insect pests control relies on integrated pest management (IPM). Integrated pest management utilises options to keep pest populations below economic threshold levels. A combination of management tactics or tools, including cultural practices (rotations with non host crops and cover crops that favour the build up of pest antagonistics), resistant cultivars and judicious chemical treatments, generally provide acceptable control of pests (Nampala *et al.*, 2002). The extent of success, however, is dependent upon having accurate damage threshold densities and readily acceptable resistant cultivars (Nampala *et al*., 2002).

CHAPTER THREE

EFFECT OF ROOT KNOT NEMATODES ON INDIGENOUS LEAFY VEGETABLES

3.1 Abstract

Indigenous leafy vegetables play an important role as income and food security crops in many rural and urban households in Kenya. Plant parasitic nematodes are a major hindrance to production with yield losses of 80 to 100 percent being recorded on some of the vegetables depending on susceptibility and inocula levels in the soil. The objective of this study was to investigate the effect of root knot nematodes on the growth of popular indigenous leafy vegetables. A greenhouse experiment was conducted twice, where six indigenous leafy vegetables namely spider plant (*Cleome gynandra*), amaranth (*Amaranth hybridus*), black night shade (*Solanum nigrum*), cowpea (*Vigna unguiculata*), jute mallow (*Corchorus spp*) and sun hemp (*Crotalaria juncea*) were tested. The seeds were planted in six pots per ILVs and half of the pots were infested with 2000 second stage juveniles of root knot nematodes. The experiment was terminated at 60 days after planting and data on plant height, fresh and dry shoot weight, galling index, egg mass index and the second stage juvenile count was recorded and analyzed. Fresh shoot weight was significantly ($P \leq 0.05$) different among the different indigenous vegetables which were affected by the root knot nematode. For instance, fresh shoot weight of black night shade was 21 percent lower in inoculated compared to the non- inoculated plants. On a scale of 1-10, where 1 = resistant and 10 = most susceptible, galling index was 1.7 in amaranth and 7.0 in the black night shade. Spider plant, sun hemp and amaranth were rated as resistant while jute mallow, cow pea and black night shade were rated as susceptible. The identified resistant varieties can be used as intercrops/rotation crops in agricultural production systems as a component of root knot nematode suppression in the soil.

3.2 Introduction

Indigenous leafy vegetables (ILVs) play an important role as food and nutritional security to many rural and urban households in Kenya. The crops also provide a source of income to resource-poor rural populations with small land units since they are cheaper to produce compared to exotic crops such as maize (IPGRI, 2003). The ILVs are generally richer in minerals such as calcium, iron and vitamins compared to exotic vegetables (Abukutsa-Onyango, 2003).The indigenous leafy vegetables are associated with several health benefits such as antioxidant, anticarcinogenic, analgesic and immunomodulatory properties (Kalpesh *et al*., 2008). Consumers and growers prefer indigenous leafy vegetables to other vegetables because they are adapted to low-input agriculture, readily available, have a short maturity period and have a high potential for yield per unit area.

Although ILVs have great potential in food, nutrition and income security, their productivity is hampered by root knot nematodes (*Meloidogyne spp*). Plant parasitic nematodes cause yield reduction ranging between 80 – 100 percent through galling depending on the crop variety and levels of inocula in the soil (Cetintas and Yarba, 2010). Galls disrupt water and nutrient uptake abilities of roots thus interfering with growth and photosynthesis. *Meloidogyne incognita* is the most widespread and most injurious nematode to a wide range of crops in tropical and subtropics (Atkins *et al.*, 2004). Knowledge on the effects of root knot nematodes on the growth of indigenous leafy vegetables is scanty. Research on ILVs, in Kenya, has concentrated on nutrient chemical composition neglecting biotic constraints in production especially nematodes of economic importance. Identification of resistant indigenous leafy varieties would contribute greatly to the management of the pests in the cropping systems. Therefore, the objective of this study was to assess the effect of root knot nematodes on growth of the ILVs.

3.3 Materials and methods

3.3.1 Preparation of root knot nematodes inocula

Extraction and preparation of nematode inoculum was carried out at Kabete plant pathology laboratories in the department of Plant science and crop protection. The eggs and second stage juveniles (J2) were obtained from the nematode infested spinach plants and soil at Kabete field station. Infected roots and soils were collected and used to prepare the inoculums (Plate 3.1). The root maceration method described by Coyne *et al*. (2007) was used to extract nematode eggs and the juveniles. Briefly, roots were gently washed with tap water and cut into 1cm long pieces. About 20g of roots were weighed to which, a ratio of 1g of root to 20ml water and 0.5% sodium hypochlorite (NaOCl) was added to the root water mix. The mixture was loaded into a domestic blender and blended for 15 seconds at high speed (Hooper *et al*., 2005) and the process repeated to obtain the required inoculum. The mixture was sieved and using a dissecting microscope the eggs and the second stage juveniles (J2) were counted to estimate the concentration per milliliter of the fluid from the sieving. The extracted juveniles were used to inoculate half of experimental plants grown in the greenhouse.

⇨Galled portion of the root

Plate 3.1 Spinach roots with galls for nematode inoculum extraction

3.3.2 Experimental material

Screening of the selected species of indigenous vegetables (ILVs) was done in the greenhouse where artificial inoculation of nematodes was done in pots with growing plants (Plate 3.2).

Plate 3.2 Greenhouse experiment set up

Six different indigenous leafy vegetables were planted in pots containing sterilized soil which had a mixture of sand (Volcanic ash) and top forest soil mixed in the ratio of 1:3. The selected (ILVs) were sun hemp, jute mallow, amaranth, cow peas, spider plant and black nightshade each replicated thrice to determine their susceptibility or resistance to plant parasitic nematodes. Below are some of the crops that were grown in the greenhouse (Plates 3.3-3.5).

Plate 3.3 Sun hemp plant

Plate 3.4 Spider plant

Plate 3.5 Black nightshade

Seeds of the selected plant species were planted four per pot and watered on daily basis. After sprouting they were thinned to two seedlings per pot and CAN top dress fertilizer was added at a rate of 20 grams per pot. The nematode inoculum which was previously prepared was used to artificially inoculate the potted ILVs. The first inoculation with 2000 J2 juveniles was done ten days after planting on three pots per ILV species and the same was repeated two weeks later after the first inoculation(Coyne *et al.* 2007). The plants were monitored for symptoms such as changes in leaf colour, height, stem size and growth vigour. Soil and root samples were taken from the rhizosphere of the plants in each pot by gently removing the soil. Both roots and soil samples were placed in labeled polythene sample bags and transported to the laboratory in a cool box where the samples were stored at 10^{o}C before nematode bioassays were conducted. The roots were carefully and gently washed with tap water and they were blotted dry.

3.3.3 Parameters measured

The data on plant height on all ILVs in the pots was taken after every two weeks interval. The fresh and dry biomass weights on all ILVS in pots were also recorded 60 days after planting during flowering. The galling index rating was assessed using a chart illustrated by Coyne *et al.* (2007) with a scale ranging between 1 -10 where 1 indicated no galling and 10 indicated severe galling. After assessing the galling index the root knot nematodes juveniles (J2) were extracted by use of the modified Baermann technique (Hooper *et al*., 2005) to identify and count the nematodes. The egg mass index at a scale of 1-5 was also assessed. Galling index, egg mass index and the juveniles in the soil samples from each plant were used to rate nematode infestation and levels of infestation for the selected six indigenous leafy vegetables to root knot nematodes and determine those which were susceptible or resistant to root knot nematodes.

3.3.4 Data analysis

Data on the counts was log transformed before being subjected to analysis of variance (ANOVA) using Genstat computer software package (Lawes Agricultural Trust Rothamsted Experimental Station 2006, version 9). Analysis of variance was conducted to compare the six plant species and determine the most susceptible ones to RKNs. Significance of the differences between treatments was measured by *T*-test, while the treatment means were compared using Fisher's protected least significant difference (lsd) at p=0.05.

3.4 Results

Plants infected with the root knot nematodes had significant ($P \leq 0.05$) reduced heights compared to the untreated controls. The greatest height reduction was recorded in black night shade (18.5 %) followed by the cow pea (15.8 %), jute mallow (12.5 %), spider plant (4 %), amaranth (3.5 %) and sun hemp (2.9 %), respectively (Table 3.1a). The heights of cowpea and jute mallow were not significantly different. Similar trend was observed when the experiment was repeated (Table 3.2a). Black night shade was greatly affected with 31% height reduction followed by cow pea (25 %) (Table 3.2a). Fresh shoot weight differed significantly ($P \leq 0.05$) among the different plant species. Non inoculated plant species had higher fresh weight compared to the inoculated plants. Amaranth had the highest fresh shoot weight of 119.9 gm and had no significant ($P \leq 0.05$) difference between inoculated and non inoculated crop plants while cowpea had the least weight (50 gm) and there were significant ($P \leq 0.05$) difference between inoculated and non inoculated crop plants. Highest fresh shoot weight reduction was observed in cow pea (26.2%), black night shade (21.9 %), jute mallow (19.3 %), amaranth (6.7 %), spider plant (5.3 %) and sun hemp (5.2%) in decreasing order (Table 3.1b). A similar trend was observed when the experiments were repeated (Table 3.2b)

Table 3.1a: Effect of nematodes on the mean plant height and % reductions in height of the indigenous leafy vegetables grown under greenhouse conditions in season one.

Parameter		Amaranth (*Amaranth hybridus*)	Cowpea (*Vigna unguiculata*)	Sun hemp (*Crotalaria juncea*)	Jute Mallow (*Corchorus spp*)	Spider plant (*Cleome gynandra*)	Black night shade (*Solanum nigrum*)
		Plant species					
Plant height(cm)	Inoculated	43.1a	27.1a	46.7a	35.6 a	61.7a	41.4 a
	Non inoculated	44.7a	32.2b	48.1 a	40.7b	64.3a	50.8b
	% reduction	3.5	15.8	2.9	12.5	4	18.5
LSD(p≤0.05) value		1.8	4.64	1.58	3.61	2.83	6.42
Significance level		Ns	*	Ns	*	Ns	*

Ns: No significant difference (p≤0.05) (same letters in the columns) *Significant difference (p≤0.05) (different letters in columns) on means in damage incidence between the inoculated and non inoculated plant species.

Table 3.1b: Effect of nematodes on the mean fresh shoot weights and % reductions in weight of the indigenous leafy vegetables grown under greenhouse conditions in season one.

Parameter		Amaranth (*Amaranth hybridus*)	Cowpea (*Vigna unguiculata*)	Sun hemp (*Crotalaria juncea*)	Jute Mallow (*Corchorus spp*)	Spider plant (*Cleome gynandra*)	Black night shade (*Solanum nigrum*)
		Plant species					
Fresh shoot weight(gm)	Inoculated	111.9 a	50 a	70.1a	65.4a	78.6a	83.4a
	Non inoculated	119.9a	67.8b	73.8a	81b	82.7a	105.3b
	% reduction	6.7	26.2	5.2	19.3	5.3	21.9
LSD(p≤0.05) value		8.42	17.1	3.92	13.8	4.46	15.9
Significance level		Ns	*	Ns	*	Ns	*

Ns: No significant difference (p≤0.05) (same letters in the columns) *Significant difference (p≤0.05) (different letters in columns) on means in damage incidence between the inoculated and non inoculated plant species.

Table 3.2a: Effect of nematodes on the mean plant height and % reductions in height of the indigenous leafy vegetables grown under greenhouse conditions in season two.

Parameter		Amaranth (*Amaranth hybridus*)	Cowpea (*Vigna unguiculata*)	Sun hemp (*Crotalaria juncea*)	Jute Mallow (*Corchorus spp*)	Spider plant (*Cleome gynandra*)	Black night shade (*Solanum nigrum*)
		Plant species					
Plant height(cm)	Inoculated	38.8a	27.1a	44.8a	32.9 a	55.6a	41.5a
	Non inoculated	41.7a	36.3b	49.4a	42.6b	61.3a	60b
	% reduction	7.1	25.3	9.3	22.8	9.3	30.8
LSD(p≤0.05) value		3.2	7.41	4.72	6.12	5.76	8.41
Significance level		Ns	*	Ns	*	Ns	*

Ns: No significant difference (p≤0.05) (same letters in the columns). *Significant difference (p≤0.05) (different letters in columns) on means in damage incidence between the inoculated and non inoculated plant species.

Table 3.2b: Effect of nematodes on the mean fresh shoot weights and % reductions in weight of the indigenous leafy vegetables grown under greenhouse conditions in season two.

Parameter		Amaranth (*Amaranth hybridus*)	Cowpea (*Vigna unguiculata*)	Sun hemp (*Crotalaria juncea*)	Jute Mallow (*Corchorus spp*)	Spider plant (*Cleome gynandra*)	Black night shade (*Solanum nigrum*)
		Plant species					
Plant shoot weight(gm)	Inoculated	125.4 a	43.3 a	78.7a	62.7a	85.1a	83.9a
	Non inoculated	129.6a	56b	82.7a	74.8b	88.3a	107.3b
	% reduction	3.3	22.7	5	16.9	3.7	21.8
LSD(p≤0.05) value		4.32	11.35	4.2	9.1	3.56	16.7
Significance level		Ns	*	Ns	*	Ns	*

Ns: No significant difference (p≤0.05) (same letters in the columns).*Significant difference (p≤0.05) (different letters in columns) on means in damage incidence between the inoculated and non inoculated plant species.

Dry shoot weight differed significantly (P≤0.05) between the inoculated and non inoculated plants. Black night shade suffered the greatest dry shoot weight reduction of 47.9 % followed by jute mallow (42.3 %), cow pea (38.2 %), sun hemp (28.1 %), spider plant (13.3 %) and amaranth (10.8 %) in that order (Figure 3.1). A similar trend in the dry shoot weight reduction was observed in season two (Figure 3.2).

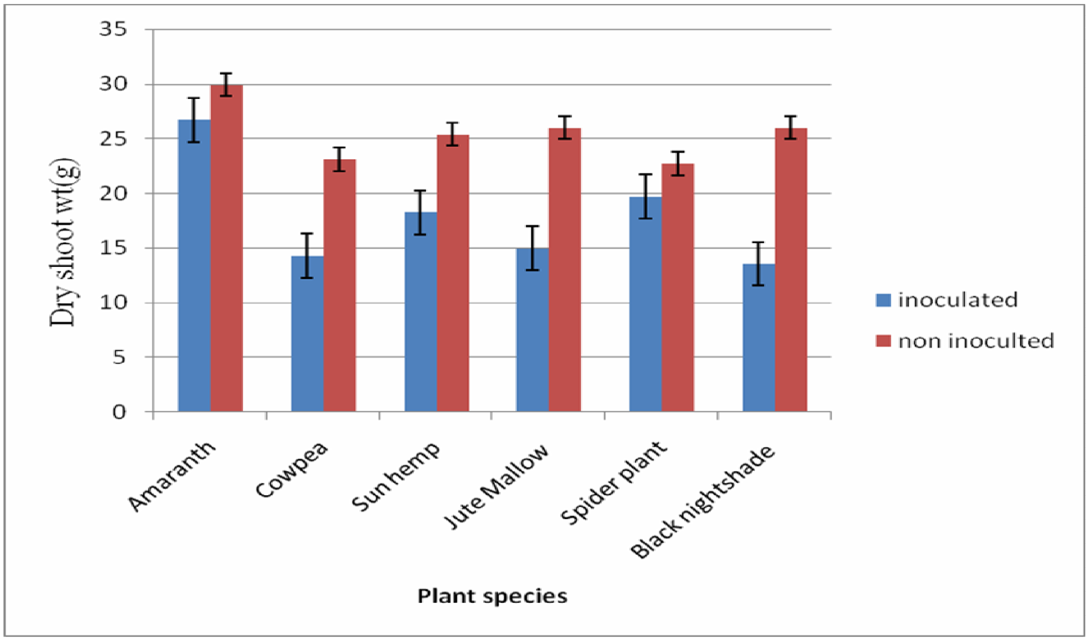

Figure 3.1: Effect of root knot nematodes infestation on the selected indigenous vegetables for season 1

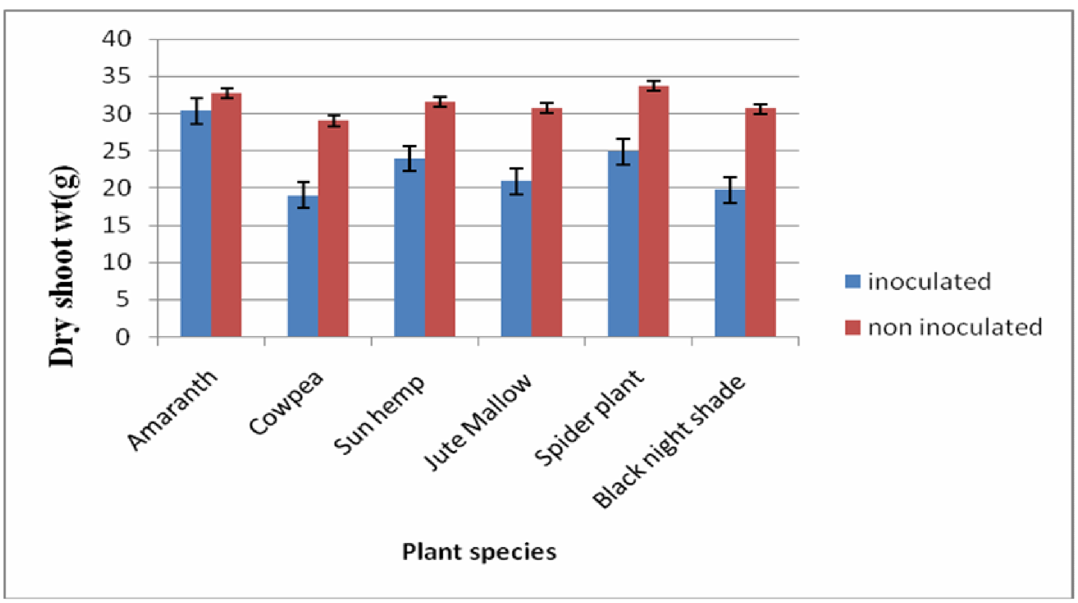

Figure 3.2:Effect of root knot nematodes infestation on the selected indigenous vegetables for season 2.

The indigenous leafy vegetable species inoculated with root knot nematodes formed galls of variable sizes. The galling index, egg mass index and juvenile stage two populations differed significantly ($P \leq 0.05$) among treatments (Table 3.3). The highest galling index rating was observed in the black nightshade at an average of 7.0 followed by jute mallow (6.7), cowpea (6.3), spider plant (3.0), sunhemp (2.0) and amaranth (1.7). Black night shade,cowpeas and jutemallow had the highest egg mass indices with mean of 3.7 each. The least egg mass index was recorded in amaranth (1) (Table 3.3).

The three crops, black night shade, cowpea and jute mallow also had high nematode log transformed counts with mean averages of 8.85, 8.42 and 8.42, respectively (Table 3.3). The least root knot nematode population was recorded in sun hemp at 6.03 and did not differ significantly from 6.45 that was recorded in amaranth (Table 3.3). The spider plant and sun hemp recorded relatively low rating for galling, egg mass indices and root knot nematode population which were recovered from the soils around the root area (Table 3.3). The most resistant plant species to the root knot nematodes under greenhouse condition was amaranth with mean galling index of 1.7, egg mass index of 1.0 and second stage juvenile population log transformed mean of 6.45 in both seasons. (Table 3.3 and 3.4). Egg and galling indices and second-stage juvenile numbers were comparable in Amaranth and sun hemp while the black night shade had the highest egg mass, galling indices and root knot nematode populations. Plate 3.6 and 3.7 shows the amaranth crop plant with least galls on its roots mass and black night shade root mass with the most galls.

Table 3.3: Galling index, egg masses and number of Juveniles observed in the soil and the plant roots for nematode inoculated crop species under greenhouse conditions in season one

	Crop Variety	Egg mass index (1-5)	Galling index (1-10)	J2 /200cm3 (log_2x)	Reaction
1	Amaranth (*Amaranth hybridus*)	1.0	1.7	6.45	Resistant
2	Cowpeas (*Vigna unguiculata*)	3.7	6.3	8.42	Susceptible
3	Sun hemp (*Crotalaria juncea*)	1.7	2	6.03	Resistant
4	Jute mallow (*Corchorus spp.)*	3.67	6.7	8.42	Susceptible
5	Spider plant (*Cleome gynandra*)	1.7	3	6.78	Resistant
6	Black nightshade (*Solanum nigrum)*	3.7	7	8.85	Susceptible
	[1]LSD value ($P \leq 0.05$)	0.92	0.79	0.27	
	Significance level	*	*	*	
	[2]Cv%	19.8	9.8	2	

[1]least significance difference [2]coefficient of variation, *significance difference ($p \leq 0.05$)

Galling index score (1-10) where 1-3 = resistant and > 3= susceptible, Egg masses count in the plant roots score of 1-5 where 1 = resistant and 5= susceptible, J2-Second stage juveniles populations recovered from the soil in $200cm^3$, Log-Logarithm.

Table 3.4: Galling index, egg masses and number of Juveniles observed in the soil and the plant roots for nematode inoculated crop species under greenhouse conditions season two

	Crop species	Egg mass index	Galling index	J2($p/200cm^3$) (log_2x)	Reaction
1	Amaranth (*Amaranth hybridus*)	1.0	1.3	6.54	Resistant
2	Cowpeas (*Vigna unguiculata*)	3.7	7	8.49	Susceptible
3	Sun hemp (*Crotalaria juncea*)	1.0	1.7	6.02	Resistant
4	Jute mallow (*Corchorus spp.)*	3.7	6.3	8.38	Susceptible
5	Spider plant (*Cleome gynandra*)	1.7	2.7	6.92	Resistant
6	Black nightshade (*Solanum nigrum)*	3.7	7.7	8.78	Susceptible
	[1]LSD value($p \leq 0.05$)	0.79	0.92	0.26	
	Significance level	*	*	*	
	[2]Cv%	17.8	11.4	1.9	

[1] least significance difference, [2]coefficient of variation, *significance difference ($p \leq 0.05$)

Galling index score (1-10) where 1-3 = resistant and > 3= susceptible,Egg masses count in the plant roots score of 1-5 where 1 = resistant and 5= susceptible,J2-Second stage juveniles populations recovered from the soil in $200cm^3$, Log-Logarithm.

Plate 3.6 Root mass for Amaranth with
with the least galls (Most tolerant).

Plate 3.7 Root mass for black nightshade
with most galls (Most susceptible).

The arrow points to the galled part of the root

3.5 Discussion

This study has demonstrated that the indigenous vegetables tested had varied reactions to the root knot nematodes. Amaranth was the most resistant vegetable to the root knot nematodes and could be cultivated in areas where the pathogenic nematodes are endemic. This would ensure sustainable food, income and nutrition security among rural and urban households. The black night shade can be used as a susceptible control in experiements evaluating for resistance to the root knot nematodes. Stunted growth, reduced plant height and vigour in the inoculated vegetables were associated with the root knot nematode infestation. These results compare to those of Mcsorley *et al.* (2004), who reported suppressed plant growth on crops that host root knot nematode. The presence of galls on the roots of susceptible varieties such as the cow pea and black night shade was responsible for stunted growth and wilting of the plants. Galls on the plant roots interfere with nutrients and water absorption leading to discoloration of the leaves displaying symptoms that resemble those of nutrient deficiencies (Atkins *et al.,* 2004). The height of amaranth, sun hemp and spider plant did not differ significantly with non-inoculated controls indicating some levels of resistance to the root knot nematode infestation. These findings compares to a greater extent with those by Nchore *et al*. (2011), who reported less damage on amaranth and more damage on black night shade by root knot nematodes while working on these ILVs in Kisii and Transmara districts of Kenya. Cowpea, jute mallow and black night shade were highly susceptible to the root knot nematode infestation which resulted to reduced height and suppressed growth. Similar findings on infestation by root knot nematodes on cowpeas crop species have been reported (Mcsorley *et al.,* 2004).

The fresh and dry shoot weights of amaranth, spider plant and sun hemp were high despite the root knot nematode infection implying resistance to the pest. Fresh and dry shoot weights of cowpea, black night shade and jute mallow were relatively low implying susceptibility to root knot nematode infestation. The reduced fresh and dry shoot weight on the susceptible vegetables could be attributed to the fact that root knot nematode infestation interferes with water, minerals and nutrients absorption and translocation thus interfering with photosynthesis. The infected plants become stunted, leaves turn yellow, wilt and eventually die (Wesemael and Moens, 2008).

The high egg and galling mass indices observed in the black nightshade, cowpea and jute mallow implied that these crops were more susceptible to the root knot nematodes compared to amaranth, spider plant and sun hemp. Black night shade (*Solanum spp)* was the most susceptible and frequently attacked by root knot nematodes compared to the spider plant (*Cleome spp*) and amaranths (*Amaranthus spp).*Similar findings on black night shade and amaranth have been reported by Nchore *et al*. (2011). Susceptible plants to the root knot nematodes, warm temperatures in the greenhouse and sandy soils which were used during experimentation may have contributed to high numbers of second stage juveniles in the soils inoculated with the root knot nematodes. Coyne *et al.* (2007) observed an increased number of root knot nematodes on sandy soils, where susceptible plants were grown under warm greenhouse conditions.

Screening of the six ILVs has shown that they are infested by and react differently to RKNs inoculation. Amaranth was the most resistant vegetable whereas sun hemp and spider plant were mildly resistant. Black night shade, cowpea and jute mallow were susceptible to RKNs infection. The infestation stunts crop growth through galling, shortening and deforming the roots and lowers the biomass yield required for consumption. This knowledge will enable the development of effective strategies for RKNs management through crop rotation or intercropping and/or selection of appropriate crop cultivars/species for nematode infested soils.

CHAPTER FOUR

EFFECT OF INTERCROPPING AND PLANT RESISTANCE ON INSECT PESTS AND NEMATODES OF ILVS

4.1 Abstract

Intercropping is a practice of growing two or more crops in proximity in the same field using different patterns. Intercropping increases the distance between plants of the same species by planting other crops in between them hence interfering with pests host recognition. The objectives of this study were to identify insect pests and plant parasitic nematodes (PPNs) that infest indigenous leafy vegetables (ILVs) and to evaluate intercropping of susceptible and resistant plants for the management purposes. Three plants namely black nightshade (susceptible to RKN and insect pests), sun hemp and spider plant (resistant to insect pests and RKN) were selected for intercropping as a means of managing RKNs and insect pests.

Field experiments were established in Kahatia location of Murang'a County for two seasons from June to October, 2012 and experiments were laid out in a randomized complete block design with 3 replicates. The treatments consisted of different arrangement patterns of intercropping namely same row intercropping, same hill intercropping, one and two row intercropping, alternate row intercropping, border cropping and control plots with black nightshade alone. Insect pests that infest indigenous leafy vegetables were identified through leaf inspection. Shoot damage, plant fresh shoot and dry shoot weight, percentage change in population of the second stage juvenile (J2) in the soil were recorded to estimate RKN damage. Several insect pests which include; Black aphids (*Aphis fabae*), flea beetles (*Chrysomelidae spp*),

leaf miners (*Lyriomyza spp)*, red spider mites (*Tetranchus spp*), cutworms (*Agrotis spp*), diamond back moth (*Plutella xylostella*), thrips (*Thrips tabaci*), whiteflies (*Bemisa spp*) and root knot nematodes (*Meloidogyne spp*) infested indigenous leafy vegetables. Same hill and same row intercrop were the most effective ($p \leq 0.05$) in reducing the effect of PPNs and insect pests infestation compared to the control plots where black night shade was planted singly with no intercropping. Intercropping resistant and susceptible ILVs can be integrated with other methods to provide an easy adaptable technology to apply for effective management of PPNs and insect pests with low external inputs.

4.2 Introduction

Many traditional African vegetables such as *Amaranths hybridus* which has high growth rates, especially in soils rich in organic matter may be classified as edible weeds. Some farmers use them as soil fertility indicators when broadcasted in the field at close spacing. Indigenous vegetables like sun hemp (*Crotalaria juncea*) and spider plant (*Cleome gyandra)* all have a strong smell and farmers have reported little incidence of pests and nematodes compared to exotic ones (Wang and Hooks, 2007). This suggests that these vegetables have some repellant/ suppressive effect on pests and (PPNs) nematodes. In this study, black night shade (BNS), sun hemp and spider plants were used as intercrops for PPNs and insect pest management. Sun hemp (*Crotalaria juncea*) is known to have an effect on egg oviposition of nematodes and as a legume has an added advantage of increasing yields through its nitrogen fixing capacities (IPGRI, 2003) hence reduced nematode numbers in soils where it is grown. Spider plant on the other hand is repellant to insect pests hence can be used for intercropping (Nampala *et al.*, 2002).

Sun hemp planted as a cover crop or intercrop suppresses populations of root knot nematodes by producing allelochemicals that could be toxic or inhibits and encourages major groups of nematode-antagonistic fungi (Wang *et al*., 2003). Hence maintaining low population densities of *Meloidogyne species*. Intercropping has a disruptive effect on the insect pests feeding by physical or chemical confusion or due to frequent encounters with non host plants (Schippers, 2002).This study was conducted to assess the effect of these ILVs on PPNs and insect pest suppression under different intercrop arrangement designs with susceptible black night shade (*Solanum nigrum*).

4.3 Materials and methods

4.3.1 Experimental site

The experiments were established at Kahatia location of Murang'a County from June to October 2012. Kahatia is located at Latitude 0^0 43'30'' South and Longitude 37^0 9' 28'' East at an altitude of 1700m above sea level. The site receives mean annual rainfall of 1800mm while the soils are volcanic in origin, extremely deep and dark reddish brown to dusky red clay (Nitosols). The temperature ranges between a minimum of 14^oC and a maximum of 28^0C with a mean average of 24 ^0C (GOK, 2012).

4.3.2 Experimental material

Black night shade which was selected as the most susceptible crop to root knot nematodes after greenhouse screening was intercropped with sun hemp and spider plant which are both resistant to nematodes. The selected ILVs were first raised in the nursery except the spider plant before transplanting. During the nursery preparation, the soil was loosened and enriched with decomposed manure at a rate of 20 tons/ha. Seeds were mixed with dry sand for uniform sowing on drills. The seedbed was mulched and the mulch was later removed when the seeds started germinating. The nursery was watered daily and seedlings were transplanted when they had developed six true leaves. Before transplanting, the land was ploughed and prepared to a fine tilth and the recommended farm yard manure applied at 20 kgs per plot (9 m^2) (Plate 4.1).

Plate 4.1 Land prepared for field experiment

4.3.3 Experimental design

The seedbed was subdivided into thirty six plots each measuring 3m by 3m to be used for the experiment (Plate 4.1). The treatments were, same row intercropping of resistance and susceptible ILVs, same hill intercropping of resistance and susceptible ILVs, border intercropping of resistance and susceptible ILVs, one row intercropping of resistance and two of susceptible ILVs, alternate row intercropping of resistance and susceptible ILVs and control plots with monocrops (plates 4.2 to 4.6). Black night shade was intercropped with spider plant in one part of the field under six treatments replicated thrice while BNS was intercropped with sun hemp under six treatments replicated thrice in the second part of the field. Both experiments were laid out in a complete randomized block design. The spacing was 40cm between rows by 20cm between plants with one plant per hole. The crops were watered daily and uniformly to maintain the moisture content. First weeding was done two weeks after planting and repeated after another two weeks. The experiment was repeated for the second season following similar procedure.

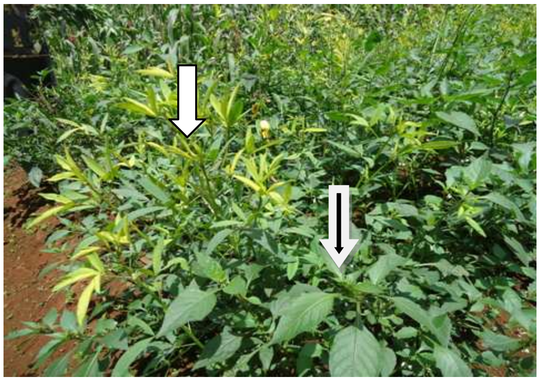

Plate 4.2 Same hill intercrop the best option (BNS and Sun hemp)

Plate 4.3 Same hill intercrop the best option (BNS and Spider plant)

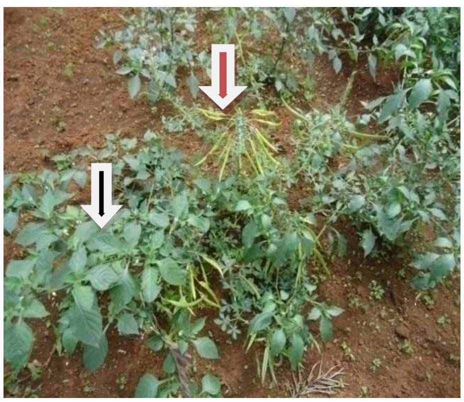

Plate 4.4 Same row intercrop (BNS and spider plant)

Plate 4.5 Border intercrop (BNS and sun hemp)

Plate 4.6 Alternate rows intercrop (BNS and Sun hemp).

The above arrows in plates point to –Sun hemp, – Black night shade and –Spider plant

4.3.4 Sampling procedure and parameters evaluated

Identification and estimation of insect populations were done through monitoring of insect pest infesting ILVs by leaf inspection and tapping them on a white paper. Ten black night shade plants were sampled per every plot to assess the insect damage and those present. Adult oviposition, feeding punctures and mines were used to identify leaf miners on the leaves while aphids were counted on the underside of the leaves and the growing terminals. Thrips were assessed by inspecting plants for larvae and adults by tapping and counting their numbers on a white paper. A hand lens was used to identify the feeding immature and adults of the whiteflies on the leaves. For the red spider mites green/red/yellow specks/ damage symptoms and scores were used for data collection, scales insects appeared as suspicious looking bumps on the plant stems and leaves hence were recorded as present or absent. Flea beetles were identified with characteristic round holes as damage symptoms on the plant leaves and were counted and recorded physically. The insect pest population counts were obtained by tapping the plant leaves and stems on a white paper with insect counting squares for small insect and manual counting for large insects. Insect pests identified were scored as follows; 0= no insect pest present, 1 = ≤50 individual pests present per plant, 2= 50 - 100 individuals pests/ oviposition holes per plant and 3= > 100 individuals pests feeding/oviposition holes. The insect pests were preserved and transported for identification and counting at Kabete entomology laboratories. The total number of plants infested with insect pest and nematodes were counted in each plot and the averages with damage symptoms were recorded for analysis.

At the flowering stage ten plants were sampled in each plot and weighed to get the fresh shoot weight which was used to estimate the total fresh shoot weight per plot. These fresh shoots per plot were oven dried for three days and weighed to estimate the dry shoot weight. Soil samples were taken in all the plots at the beginning and at the end of the experiment, to estimate the second stage juveniles in 200 cm^3 of soil which were recorded. The percentage change in nematode counts were compared for the different treatments and time periods of observation beginning and at the end.

4.3.5 Statistical analysis

All the collected data was subjected to analysis of variance (ANOVA) using Genstat computer software package (Lawes Agricultural Trust Rothamsted Experimental Station, version 9, 2006) to determine the differences in intercrop pattern arrangements for use in managing insect pests and PPNs. Treatment means were compared using the Fisher's protected LSD test at 5% probability level.

4.4 Results

4.4.1 Monitoring of insect pests infesting the indigenous leafy vegetables in Murang'a

Black aphids (*Aphis fabae*), flea beetles (*Chrysomelidae spp*), leaf miners (*Lyriomyza spp)*, red spider mites (*Tetranchus spp*), cutworms (*Agrotis spp*), diamond back moth (*Plutella xylostella*), thrips (*Thrips tabaci*), whiteflies (*Bemisa spp*) and root knot nematodes (*Meloidogyne spp*) were observed at the experimental plots (Plates 4.7 - 4.12).Beneficial insects observed and recorded during this study included ants and lady bird beetles. More than fifty thrips, aphids, whiteflies and the red spider mites were sampled per plant. Leaf miners, flea beetles and scale insects rated low (1-2) since they were less than fifty in total per plant (Table 4.1).

 mgx10

Plate 4.7 Red spider mites on BNS

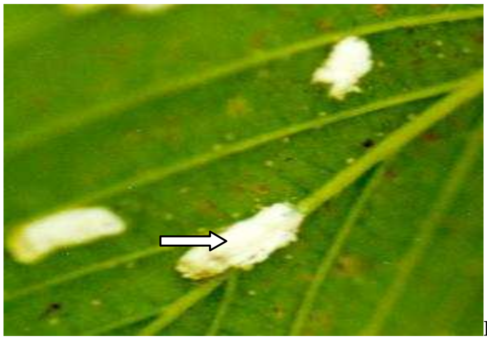 mgx10

Plate 4.8 Scale insects on BNS

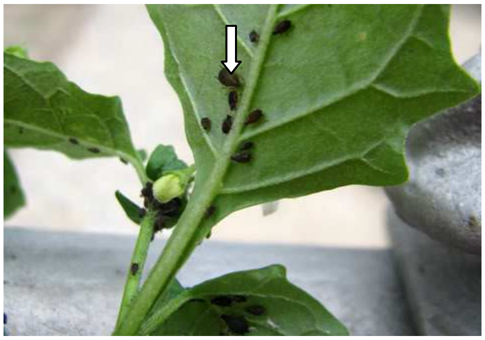

Plate 4.9 Black aphids on BNS

Plate 4.10 curled BNS leaves

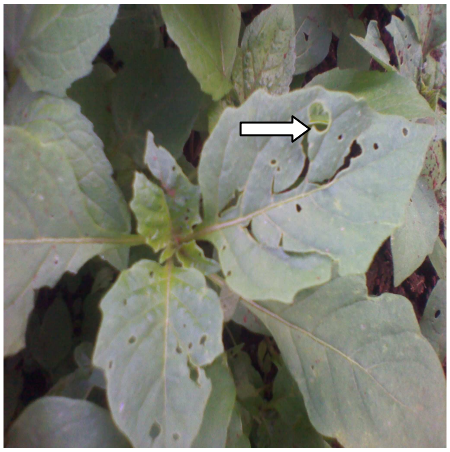

Plate 4.11 Flea beetles damage on BNS

Plate 4.12 Nematode infested plot

Table 4.1 Mean scores of various arthropod pests in black night shade with prevailing temperature for the two seasons

Arthropod pests	Weeks after transplanting	4		5		6		7		8		9	
	Temp ^{0}c	25	14	24	18	26	14	22	20	21	21	24	22
	Season	S1	S2	S1	S2	S1	S2	S1	S2	SI	S2	S1	S2
Leaf miners		0	0	1	1	1	0	1	2	1	2	2	2
Thrips		2	1	2	1	3	1	2	2	3	2	3	2
Aphids		3	1	3	2	3	1	3	2	3	2	3	3
White fly		3	1	3	1	3	1	2	2	2	2	2	2
Flea beetles		1	0	1	1	0	0	1	1	0	1	2	2
Red spider mites		2	0	2	1	3	0	2	1	2	1	2	2

Arthropod pests index score. 0 = no insect visible; 1 = ≤ 50 individual pests per plant; 2 = 50- 100 individuals' pests/oviposition holes per plant; 3 = > 100 individuals' pests feeding/oviposition hole
S1-Season one and S2-Season two

Black aphids, thrips, whiteflies and red spider mites scored 3 (more than 100 individuals per plant) due to high temperatures while at the low temperatures the same insect pests scored 1(less than 50 individuals per plant). Season one which experienced warm temperatures recorded higher insect pests incidence compared to the lower temperatures that were experienced in the following season (Figures 4.1 and 4.2).

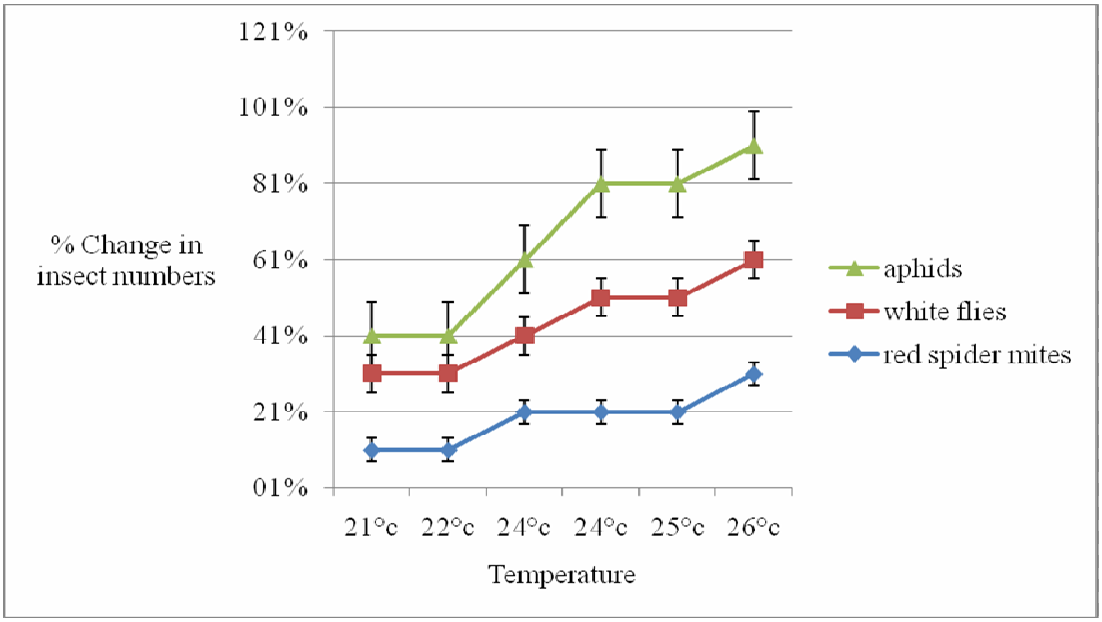

Figure 4.1: Effect of temperature on major athropod pests infesting the indigenous leafy vegetables for season one.

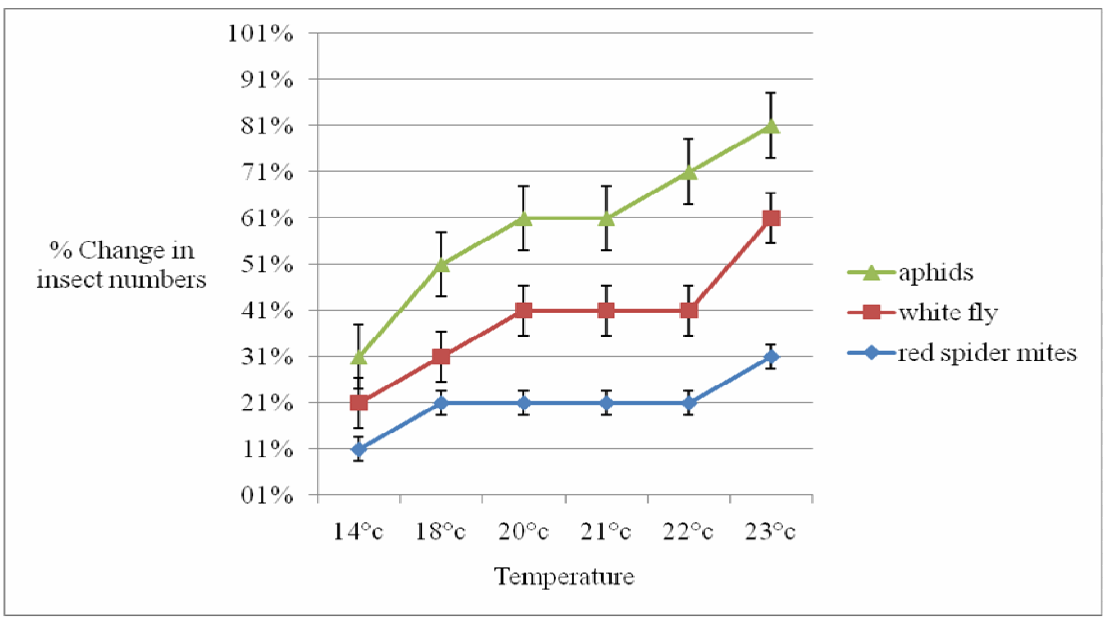

Figure 4.2: Effect of temperature on major athropod pests infesting the indigenous leafy vegetables in season two.

4.4.2 Effect of intercropping the selected indigenous leafy vegetables on insect pests and root knot nematodes

High incidence of insect pests and nematodes were recorded in season one which was warmer compared to the second season. High pest population was associated with severe damage to the crops in season one. Same row and hill intercropping designs varied significantly ($P \leq 0.05$) with other intercropping designs in all the evaluated parameters (Tables 4.2 and 4.3).Spider plant and black night shade intercrops recorded least insect pests damage, for example on the same hill intercrops 19.5% and 21.4% was recorded for insect damage season one and two respectively (Tables 4.2 and 4.3). The control plots where black night shade alone was grown,53.9 % and 38.2% insect damage was recorded for the two seasons respectively (Tables 4.2 and 4.3).The control plots showed the highest damage and lowest mean yields for fresh and dry shoot weight in both seasons (Tables 4.2 and 4.3). The yields of black night shade and spider plant in the same hill intercrop design were significantly ($P \leq 0.05$) higher than other intercropping designs tested with 3.96 kg/plot area and 1.11 kg/plot area mean yields for fresh and dry shoot, respectively. In the plots where sun hemp was intercropped with black night shade in the same hill, a slight increase in the percentage of second stage juveniles was recorded. The increase was 14.6% and 11.9% for season one and two, respectively when compared with juvenile numbers in the soil samples taken at the beginning of experiment (Table 4.4). The highest second stage juvenile increase 31.9% and 17.6% for season one and two, respectively was recorded where black night shade was grown as monocrop (Table 4.4).

Table 4.2: Effect of pests infestation on black night shade under different intercrops under field conditions for season one

	Type of intercrop	Mean number of plants damaged (%)	Mean fresh shoot weight (kg/plot area) 9cm^3	Mean dry shoot weight (kg/plot area) 9cm^3
1	Same row intercrop(black night shade and spider plant)	29.4a	3.62a	1.08a
2	Same Hill intercrop (black night shade and spider plant)	19.5a	3.96a	1.11a
3	One row and two rows (black night shade and spider plant)	48.2b	2.75b	0.95b
4	Border intercrop(black night shade and spider plant)	50.9b	2.61b	0.9b
5	Alternate rows intercrop (black night shade and spider plant)	36.6ab	2.21b	0.81b
6	Black night shade only	53.9c	2.05bc	0.79b
	Grand mean	39.7	3.19	1.05
	[1]LSD ($p \leq 0.05$)	12.2	0.56	0.14
	[2]Cv%	46.3	9.7	7.4

[1]Least significance difference, [2] Co-efficient of variance. Means followed by the same letter(s) within columns are not significantly different ($p \leq 0.05$) while those followed by different letter(s) within columns are significantly different; Means are separated by LSD ($p \leq 0.05$).

Table 4.3 Effect of pests infestation on black night shade under different intercrops under field conditions for season two

	Intercrop pattern/design	Means number of plants damaged (%)	Mean fresh shoot weight (kg/plot area)	Mean dry shoot weight (kg/plot area)
1	Same row intercrop(black night shade and spider plant)	22.8a	4.23a	1.31a
2	Same Hill intercrop(black night shade and spider plant)	21.4a	4.59a	1.37a
3	One row and two rows (black night shade and spider plant)	30.3b	3.24b	1.09bc
4	Border intercrop (black night shade and spider plant)	33.8b	3.24b	1.15b
5	Alternate rows intercrop (black night shade and spider plant)	25.2ab	2.61c	1.01c
6	Black night shade only	38.2c	2.34c	0.89c
	Grand mean	28.6	3.8	1.26
	[1]LSD ($p \leq 0.05$)	3.5	0.24	0.07
	[2]Cv%	36.7	7.8	6.5

[1]Least significance difference, [2] Co-efficient of variance. Means followed by the same letter(s) within columns are not significantly different ($p \leq 0.05$) while those followed by different letter(s) within columns are significantly different; Means are separated by LSD ($p \leq 0.05$).

Table 4.4: Effect of intercropping black night shade with sun hemp on the numbers of second stage (J2) juveniles of root knot nematodes in the soil and % increase in numbers of J2

	Intercrop pattern/design	Mean number of J2/200cm^3 season one			Mean number of J2/200cm^{3}season two		
		J2 initial count (Log_2x)	J2 final count (Log_2x)	% increase in J2	J2 initial count(log_2X)	J2 final Count(log_2x)	% increase in J2
1	Same row intercrop(black night shade and sun hemp)	7.2a	8.25a	14.6	7.33a	8.2a	11.9
2	Same hill intercrop (black night shade and sun hemp)	7.19a	8.23a	14.4	7.35a	8.32a	13.2
3	One row and two rows(black night shade and sun hemp)	6.9b	8.49b	23.04	8.24b	9.52b	15.5
4	Border intercrop(black night shade and sun hemp)	6.46b	8.69b	34.5	8.18b	9.55b	16.8
5	Alternate rows intercrop (black night shade and sun hemp)	6.64b	8.69b	30.8	8.13b	9.46b	16.4
6	Control with black night shade only	6.61b	8.72b	31.9	8.16b	9.6b	17.6
	[1]LSD value($p \leq 0.05$)	0.25	0.23		0.33	0.42	
	Significance level	*	*		*	*	

[1]Least significance difference, Means followed by the same letter(s) within columns are not significantly different ($p \leq 0.05$) while those followed by different letter(s) within columns are significantly different; Means separated by LSD ($p \leq 0.05$).

4.5 Discussion

Insect pests and nematodes were abundant during the experiment period and were a major constraint during field production of the selected indigenous vegetables. A number of insect pests were identified. They included black aphids (*Aphis fabae*), flea beetles (*Chrysomeli- dae spp*), leaf miners (*Lyriomyza spp)*, red spider mites (*Tetranchus spp*), cutworms (*Agrotis spp*), diamond back moth (*Plutella xylostella*), thrips (*Thrips tabaci*), whiteflies (*Bemisa spp*). Plant parasitic nematodes also infested the ILVs while growing. Similar findings were reported by the

African Vegetable Research centre, (AVRDC, 2003) where twelve insect pests species were identified on ILVs.

There was a high incidence of insect pests and nematodes during the first season. This was because of the warm temperatures that prevailed with the maximum temperature recorded at 26^0C. Low incidence of pests was recorded during the second season whose first weeks were extremely cold recording lowest temperature of 14^0C.These findings are in agreement with those by Mohammed *et al*. (2009), who reported that besides host-plant resistance, fluctuations in the populations of pests depends upon variations in weather factors, especially those of temperature and relative humidity.

Sole black night shade plots had the highest shoot damage due to pests indicating high susceptibility of the crop to infestation by a wide range of pests. Same hill and same row intercrops reduced pest and nematode infestations in the plots suggesting that the closer the plants were the better the interaction in reducing nematodes and insect pest infestations. According to Vargas *et al.* (2000), sun hemp is known to inhibit the formation of galls, production of egg masses and release of root exudates toxic to nematodes. This therefore explains the reason for less juvenile numbers where sun hemp was used as an intercrop. In intercropping, the compounds produced from root exudates have a greater allelopathic effect than the individual compounds alone when *Crotalaria spp* is in a monocrop (Vargas *et al.*, 2000). Furthermore, in the same hill and same row intercropping there was closer interaction between the roots of BNS and sun hemp hence benefiting from the sun hemp good attributes. In addition, the same hill and row intercrop black night shade benefited from nitrogen fixed by sun hemp (*Crotalaria spp*) which is leguminous hence the high yields obtained from these two intercrops. This compares with earlier studies by Mbugua *et al.* (2005), which showed that an intercrop with sun hemp led to increased yields presumably by the transfer of biologically fixed nitrogen from the roots of the legumes to the root zone of the companion crop. The result(s) also compare with those reported by Wang *et al.* (2003), that sun hemp (*Crotalaria spp)* is a poor host to many plant-parasitic nematodes including; *Meloidogyne spp, Rotylenchulus reniformis, Radopholus similis, Belonolaimus longicaudatus* and *Heterodera glycines.* It is also a poor or non-host to a large group of other pests and pathogens (Grubben and Denton, 2004). Moreover, *Crotalaria spp.* is known to enhance multiplication of natural enemies of phytopathogenic nematodes, such as fungi that directly feed on nematodes eggs (Wang *et al.,* 2003). They aid in fixing nitrogen and promote the accumulation of decomposers such as free-living nematodes. This increases nutrient availability for plant uptake leads to healthy plants that are resistant to nematode damage (Wang *et al*., 2003)

Same hill and row design of intercropping with spider plant and black night shade had reduced number of plants with shoot damage caused by insects hence high yields. This implies that the closer the interaction between the plants for example in the same hill and same row intercrop the more effective is the intercrop in repelling the insect pest away from the susceptible plants. Cetintas and Yarba (2010), reported that spider plant oil and hairy surface deter insect pests from infesting it and other plants around it. Palada and Chang (2003), also reported that spider plant is effective in reducing insect pests in the cruciferous crop species leading to increased yields. Sun hemp (*Crotalaria juncea*) and spider plant (*Cleome gynandra*) performed well as intercrops due to their inherent genetic characteristics that are not preferred by the pests (Keding *et al*., 2007). Cultural methods such as the intercropping which was evaluated are better than the use of chemicals in pest management because they are friendly to the environment, leave no pesticide residue on the produce for consumption and utilise fewer resources/external inputs. This study has shown that intercropping using resistant crops is effective in suppressing insect pests and plant parasitic nematodes in the cropping system and has also demonstrated that intercropping and plant resistance can be used as a management tool for insect pests and plant parasitic nematodes.

CHAPTER FIVE

GENERAL DISCUSSION, CONCLUSIONS AND RECOMMEDATIONS

5.1 General discussion

There has been renewed interest on indigenous leafy vegetables (ILVs) by the policy makers and the international community on the realization that these vegetables have potential that has yet to be exploited. These indigenous vegetables have many advantages over exotic ones; they have high micronutrients content, medicinal properties, agronomical advantages and thus contribute to food, nutrition security and income generation for farmers (Gruben and Denton, 2004). Studies done by Schippers (2002), revealed that fresh leaves of amaranth, slender leaf, spider plant, cowpeas, amaranth, pumpkin leaves and jute mallow contain more than 100% of the recommended daily allowance for vitamins and 40% proteins for growing children and lactating mothers.

The results obtained in this study on cumulative plant yields, pests and nematodes scores on the plants tested showed significant ($p \leq 0.05$) differences. Sun hemp (resistant to nematodes) was more vigorous in terms of growth and yielded more than black night shade (susceptible to nematodes) both in the green house and in field experiments. Amaranth, sun hemp and spider plants showed resistance to the root knot nematodes with the least reduced yields comparing the inoculated and non inoculated plants. Similar findings were reported by Nchore *et al*. (2011). Black night shade, cowpeas and jute mallow were susceptible, hence high reduction in yields and presence of more galls. Plants infested by nematodes in the field were distributed in patchy or irregular patterns. According to Keding *et al*. (2007), it is difficult to diagnose a nematode infection solely by observing these foliar symptoms. There is, therefore, need to measure other aspects like galling and egg mass indices because these symptoms are similar to those resulting from both biotic and abiotic factors such as insect pests, fungi and bacteria, drought and nutrient deficiencies. Root knot nematodes and insect pests infesting these indigenous leafy vegetables were most active in the warm temperature and their activities greatly suppressed when temperatures were low as was observed during the experimentation period with similar findings reported by Mohammed *et al.* (2009). High infestation on ILVs was observed in the warmer season one compared to season two which was relatively cold. During experimentation several insect pests that infest ILVs were identified and they included black aphids (*Aphis fabae*), flea beetles (*Chrysomelidae spp*), leaf miners (*Lyriomyza spp)*, red spider mites (*Tetranchus spp*), cutworms (*Agrotis spp*), diamond back moth (*Plutella xylostella*), thrips (*Thrips tabaci*), whiteflies (*Bemisa spp*) and root knot nematodes (*Meloidogyne spp*.) with similar findings reported by AVRDC, (2003).

Sun hemp and spider plant were found to be effective candidates for intercropping to reduce the severity of insect pests and plant parasitic nematodes. The intercrop plots showed fewer incidences of insect pests and PPNs compared to those where the sole crops were planted with similar findings reported by Centitas and Yarba (2010), who studied the effect of repellent plants on pests and nematodes. The closer the distance between the resistant and the susceptible plants the lesser the damage on the crops caused by insect pests and nematode thus the same hill intercrop and same row designs were the best option in the management of insect pests and plant parasitic nematodes.

5.2 Conclusions

Indigenous leafy vegetables are considered as minor crops and have been given low priority in most agronomic and development research programmes.Screening of the six ILVs has shown that they are infested by and react differently to RKNs inoculation. Amaranth was the most resistant vegetable whereas sun hemp and spider plant were mildly resistant. Black night shade, cowpea and jute mallow were susceptible to RKNs infection. Infestation by insects and PPNs reduces the biomass which is necessary for consumption. The susceptible crops can be rotated with resistant plants like spider plant, amaranth and sun hemp or intercropped with the same species.

It has also been demonstrated that intercropping can be an efficient tool in the management of insect pests and plant parasitic nematodes. The distance between the intercrops has a role to play in the infestation of the target plant being protected from insect pests and PPNs. Sun hemp (*Crotalaria juncea*) and spider plant (*Cleome gynandra*) in this study emerged as effective intercrops that repelled insects and reduced RKNs infestation on BNS compared to plots where BNS sole crops were planted.

5.3 Recommendations

There is need to screen more indigenous vegetables to obtain more information on their susceptibility or resistance to insect pests or PPNs. With some ILVs being resistant to insect pests and /or root knot nematodes it is possible that they can be evaluated for intercropping and rotational programmes. In addition, it is important to rotate the susceptible vegetables with the resistant ones to reduce nematode build up in the soil. More so, further studies need to be undertaken to understand the mechanisms of resistance/tolerance or poor host status of some indigenous vegetables like sun hemp, spider plant and amaranth with the intention to use their resistant traits for breeding and crop protection. The choice of intercrop and design/distance should be based on the knowledge that has been tested considering the economics, genetics, proximity and spartial orientation of the intercrops to avoid plant-plant competition for sunlight, space and nutrients, hence reducing yields. According to Otipa *et al*. (2009), the challenge to research is therefore, to identify suppressive crops that satisfy the economic considerations in cropping systems.

Intercropping should be employed in the management of insect pests and PPNs. Compared to exotic vegetables ILVs are resistant to diseases, drought tolerant, they establish well, yield fairly well and are more acceptable to many farmers. Therefore, concerted efforts are needed to increase their production and consumption. Further research should be undertaken on various cultural practices such as companion cropping, solarization to improve ILVs production and reduce the biotic constraints by exploiting their genetic potential.

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Appendices

Appendix 1: Mean composition per 100 gram edible portion of selected indigenous leafy vegetables compared to cabbage.

Crop spp	Amaranth	Spider plant	Black nightshade	Jute mallow	Cowpeas	Cabbage
Moisture content(g)	84	86.6	87.2	80.4	89.8	91.4
Iron(mg)	8.9	6.0	1.0	7.2	39	0.7
Protein(g)	4.6	4.8	43	4.5	4.6	1.7
Carbohydrates(g)	8.2	5.2	5.7	12.4	4.8	6.0
Fibre (g)	1.8	1.4	1.3	2.0	1.1	1.2
Ascorbic acid-vit c (mg)	64	13	20	80	87	54
Calcium	410	288	442	360	152	47
Phosphorus	103	111	75	122	120	40
B-carotene (microgram)	5716	10452	3660	6410	5700	100
Thiamine(Vit.B1) mg	0.05			0.15	0.35	0.04
Riboflavin(vit.B2) mg	0.42		0.59	0.53	0.2	0.1

Source: Grubben *et al.* (2004); (KENRIC), National Museums of Kenya; Maundu *et al.* (1999); Onyango, (2001)

Appendix 2: Mean fresh shoot weight, percentage germination and moisture for selected indigenous leafy vegetables.

ILVs	Weight(g)	Germination %		Moisture content %
		Lab	Field	
Night shades	14.4	50	85	12.3
Slender leaf (Sun hemp)	30.6	100	100	11.3
Spider plant	40.6	75	100	11.3
Cowpeas	22.7	100	90	11.9

Source: Onyango, (2003) and Schippers, (2000); Agricultural and rural co-operation page. 214 Chathan.Uk.Technical Centre for international co-operation.

Appendix 3: Weather data for Kahatia in Murang'a during the experimental period.

Month	Mean Max. Temp (^{0}c)	Mean Min Temp (^{0}c)	Total rainfall (mm)	Mean R.H 0600 Z	1200 Z	Number of rainy days
May	24.2	14.2	120.5	66	42	16
June	22.0	14.1	190.0	86	62	20
July	20.4	12.2	80.2	87	62	14
August	22.8	16.1	15.7	57	45	10
September	26.0	16.0	16.8	58	41	5
October	26.0	14.1	160.2	86	56	16
November	24.4	14.2	206.3	84	56	18

Appendix 4: Analysis of variance for plant height on crop species inoculated with root knot nematodes under greenhouse conditions for season one

Variate: Plant Height(cm)					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	71.7	35.8	0.10	
Treatments	5	12109.2	2421.8	6.58	<.001
Residual	100	36791.2	367.9		
Total	107	48972.1			

Appendix 5: Analysis of variance for Fresh shoot weight on crop species inoculated with root knot nematodes under greenhouse conditions for season one.

Variate: Fresh Shoot wt(g)					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	2737.6	1368.8	3.64	
Treatments	5	13286.6	2657.3	7.08	<.001
Residual	28	10515.6	375.6		
Total	35	26539.7			

Appendix 6: Analysis of variance for dry shoot weight on crop species inoculated with root knot nematodes under greenhouse conditions for season one.

Variate: Dry shoot wt(g)					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	134.84	67.42	3.92	
Treatments	5	729.56	145.91	8.49	<.001
Residual	28	481.21	17.19		
Total	35	1345.61			

Appendix 7: Analysis of variance for galling index on crop species inoculated with root knot nematodes under greenhouse conditions for season one.

Variate: Galling index					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	0.1111	0.0556	0.29	
Treatments	5	92.4444	18.4889	97.88	<.001
Residual	10	1.8889	0.1889		
Total	17	94.4444			

Appendix 8: Analysis of variance for egg masses index on crop species inoculated with root knot nematodes under greenhouse conditions for season one.

Variate: Egg masses index.					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	0.7778	0.3889	1.52	
Treatments	5	23.1111	4.6222	18.09	<.001
Residual	10	2.5556	0.2556		
Total	17	26.4444			

Appendix 9: Analysis of variance for second stage juvenile count in the soil on crop species inoculated with root knot nematodes under greenhouse conditions for season one.

Variate: Second stage Juvenile count in the soil					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	8888.	4444.	1.98	
Treatments	5	419929.	83986.	37.39	<.001
Residual	10	22462.	2246.		
Total	17	451279			

Appendix 10: Analysis of variance for plant height on crop species inoculated with root knot nematodes under greenhouse conditions for season two.

Variate: Plant height.					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	19.1	9.6	0.04	
Treatments	5	8759.1	1751.8	6.42	<.001
Residual	100	27272.0	272.7		
Total	107	36050.2			

Appendix 11: Analysis of variance for fresh shoot weight on crop species inoculated with root knot nematodes under greenhouse conditions for season two.

Variate: Fresh shoot wt					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	2452.7	1226.3	3.63	
Treatments	5	24554.4	4910.9	14.56	<.001
Residual	28	9446.4	337.4		
Total	35	36453.6			

Appendix 12: Analysis of variance for dry shoot weight on crop species inoculated with root knot nematodes under greenhouse conditions for season two.

Variate: dry shoot wt

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	105.25	52.62	2.69	
Treatments	5	536.91	107.38	5.49	0.001
Residual	28	547.77	19.56		
Total	35	1189.93			

Appendix 13: Analysis of variance for galling index on crop species inoculated with root knot nematodes under greenhouse conditions for season two.

Variate: Galling index

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	0.7778	0.3889	1.52	
Treatments	5	123.1111	24.6222	96.35	<.001
Residual	10	2.5556	0.2556		
Total	17	126.4444			

Appendix 14: Analysis of variance for egg masses index on crop species inoculated with root knot nematodes under greenhouse conditions for the season two.

Variate: Egg masses index

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	0.7778	0.3889	2.06	
Treatments	5	27.7778	5.5556	29.41	<.001
Residual	10	1.8889	0.1889		
Total	17	30.4444			

Appendix 15: Analysis of variance for second stage Juveniles recovered from the soil on crop species inoculated with root knot nematodes under greenhouse conditions for the season two

Variate: Second stage juvenile count in the soil

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	343.00	171.50	1.79	
Treatments	5	270837.33	54167.47	565.62	<.001
Residual	10	957.67	95.77		
Total	17	272138.00			

Appendix 16: Analysis of variance on the number of plants with shoot damage due pest and PPN infestation under different intercrop designs in the field season one.

Variate: No. of plants with shoot damage					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	210.1	105.1	0.31	
Treatment	5	16632.3	3326.5	9.84	<.001
Residual	100	33818.3	338.2		
Total	107	50660.7			

Appendix 17: Analysis of variance for the fresh shoot weight under different intercrop designs in the field season one.

Variate: Fresh Shoot weight					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	0.13134	0.06567	0.69	
Treatment	5	10.95918	2.19184	23.02	<.001
Residual	10	0.95199	0.09520		
Total	17	12.04251			

Appendix 18: Analysis of variance for the dry shoot weight under different intercrop designs in the field season one.

Variate: Dry shoot weight					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	0.009078	0.004539	0.76	
Treatment	5	0.342361	0.068472	11.43	<.001
Residual	10	0.059922	0.005992		
Total	17	0.411361			

Appendix 19: Analysis of variance on the number of plants with shoot damage due pest and ppn infestation under different intercrop designs in the field season two.

Variate: No. of plants with shoot damage					
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	301.2	150.6	1.36	
Treatment	5	3961.6	792.3	7.18	<.001
Residual	100	11040.9	110.4		
Total	107	15303.7			

Appendix 20: Analysis of variance for the fresh shoot weight under different intercrop designs in the field season two.

Variate: fresh shoot weight

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	0.12054	0.06027	0.70	
Treatment	5	14.40323	2.88065	33.65	<.001
Residual	10	0.85599	0.08560		
Total	17	15.37976			

Appendix 21: Analysis of variance for the dry shoot weight under different intercrop designs in the field season two.

Variate: Dry shoot weight

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Replicates	2	0.011744	0.005872	0.88	
Treatment	5	0.617828	0.123566	18.52	<.001
Residual	10	0.066722	0.006672		
Total	17	0.696294			

Appendix 22: Analysis of variance for the nematode count in the soil at two sampling intervals under different intercrop designs in the field season one.

Variate: Nematode count.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Sampling time	1	227976.	227976.	59.41	
Treatment	5	10802.	2160.	0.56	<.0.032
Residual	5	19187.	3837.		
Total	11	257965.			

Appendix 23: Analysis of variance for the nematode count in the soil at two sampling intervals under different intercrop designs in the field season two.

Variate: Nematode count.

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Sampling time	1	88236.8	88236.8	384.06	
Treatment	5	7457.4	1491.5	6.49	<.0.030
Residual	5	1148.8	229.8		
Total	11	96842.9			