INTEGRATION OF THYMOL, SOIL IMPROVERS AND BIOSTIMULANTS FOR MANAGEMENT OF TOMATO PESTS, DISEASES, NEMATODES AND RHIZOSPHERE MICROBIAL DYNAMICS KIRINYAGA COUNTY, KENYA

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A99/30848/2015

A THESIS SUBMITTED IN FULFILMENT OF THE REQUIREMENT FOR THE AWARD OF DOCTOR OF PHILOSOPHY IN PLANT PATHOLOGY IN THE SCHOOL OF AGRICULTURE AND ENTERPRISE DEVELOPMENT, KENYATTA UNIVERSITY

JUNE, 2022

DECLARATION

This thesis is my original work and has not been presented for an award of a degree in any other university.

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To all my loved ones for their endless support and encouragement throughout the entire period of this study

ACKNOWLEDGEMENTS

I am grateful to my supervisors Prof. Waceke Wanjohi, Prof. Joseph Onyango Gweyi and Dr Ruth Kahuthia-Gathu for their support and overall guidance during the entire period of this study and writing of thesis. I would like to thank the entire School of Agriculture and Enterprise Development and the Department of Agricultural Science and Technology for their dedication and support in ensuring I complete my studies.

I wish to also acknowledge the National Research Fund (NRF) for funding this research project.

I also thank all the farmers Mr. Lawrence Gichoki in Kagio and Ms. Millicent Kivaa in Kimbimbi that graciously agreed to offer their farms in Kirinyaga County to use them as experimental sites for this study.

Lastly, I would like to thank the Kenyatta University farm staff the University farm manager Mr Chepkwony and farm staff Mr Barnaba for the support they accorded me during the greenhouse trials at Kenyatta University.

Above all, I thank God for the protection and providence that enabled me to further my studies.

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ACRONYMS AND ABBREVIATIONS

ANOVA	Analysis of Variance
ASL	Above Sea Level
CFU	Colony Forming Unit
DMRT	Duncan's Multiple Range Test
EC	Electric Conductivity
GC	Gas Chromatography
GDP	Gross Domestic Product
HPLC	High Performance Liquid Chromatography
IPM	Integrated Pest Management
KALRO	Kenya Agricultural and Livestock ResearchOrganization
LM	Lower Midland
LSD	Least Significant Difference
MPR	Minjingu Phosphate Rock
MBC	Microbial Biomass Carbon
MS	Mass Spectrometry
NRF	National Research Fund

PCR–DGGE	Polymerase Chain Reaction and Denaturing Gradient Gel Electrophoresis
PDI	Percent Disease Index
PLFA	Phospho-Lipid Fatty Acids
RCBD	Randomized Complete Block Design
RKN	Root Knot Nematode

ABSTRACT

Soil-borne pathogens, root knot nematodes and insect pests remain a major constraint to tomato *Solanum lycopersicum* L. (Solanales; Solanaceae) production in Kenya. The study was carried out with an aim of evaluating the effect of integrating selected plant essential oils, soil improvers and bio-stimulants on rhizosphere microbial, soil-borne pathogens, nematodes and insect pests of tomato. The plant essential oil used (treatments) in this study were thyme oil, bio-stimulants (*Trichoderma harzianum*), sea weed (*Ascophyllum nodosum* and *Fucus vesiculosus*), a sea weed with humic acids, a sea weed with amino acids, peptides and an amino acid; while the soil improver was based on pellets of plant-based raw materials and seaweeds containing 7.5% N in pure vegetable form, soft ground potassium sulphate with magnesium (4% K_2O) and soft ground rock phosphate (2% P_2O_5). The study was conducted in farmer fields in Kimbimbi and Kagio in Kirinyaga County from September 2016 to July 2017 and in the greenhouse at Kenyatta University from February to October 2018. Data collection was carried out on soil-borne diseases, insect pest population and diversity, plant growth and yield parameters. It was subjected to ANOVA using Genstat statistical packages and means were separated using Fisher's Least Significant Difference (LSD) at $P \leq 0.05$. The results indicated that thyme oil was effective in suppressing the fusarium wilt and early blight by between 49.2% to 65.9% and 58.9% to 84.8%, respectively. Plant essential oils were effective in the management of thrips (mainly *Frankliniella occidentalis*) and whitefly (Mainly *Bemisia tabaci* and *Trialeurodes vaporariorum*) infestations causing a reduction in infestation of between 47% to 68.6 % and 41.3% to 66.2%, respectively. The application of the soil improver had a positive effect on the microbial population of fungi resulting in an increase of active fungi at the soil rhizosphere of tomato plants of up to 292%. Application of *T. harzianum* was effective in the management of Fusarium wilt (39.6% to 48.6% reduction) and root knot nematodes (48.8% to 60.9% reduction). The integration of thyme oil, soil improver, *T. harzianum* and bio-stimulants was effective in the management of a reduction of up to 66.9% for Fusarium Wilt, 83.4% for early blight, 60% for root knot nematodes, 66% for thrips and 47.3% for whitefly as well as increasing yield of tomato by up to 125.6%. Based on these results, the integration of thyme oils, *Trichoderma harzianum* and bio-stimulants can be recommended for management of, fusarium wilt, early blight, root knot nematodes, thrips and whitefly in tomato production.

CHAPTER ONE: GENERAL INTRODUCTION

1.1 Background information

Tomato *Solanum lycopersicum* L., is an economically important crop which belongs to the family *Solanaeace*, contains more than 3,000 species, including plants of economic importance such as potatoes *Solanum tuberosum*, eggplants *Solanum melongena*, tobacco *Nicotiana tabacum* and peppers *Capsicum annuum* (Bai and Lindhout, 2007). In Kenya with an annual production of 599,458 tons on 28,263 Hectares in 2018 (FAOSTAT, 2020). Tomato, contributes significantly to Kenyan economy through meeting nutritional food requirements, generation of income and creation of employment (Sigei *et al*., 2014; Mwangi *et al.,* 2020).

Tomato is a major source of vitamin A and C and supplies a sufficient amount of the antioxidant lycopene pigment that helps to protect consumers against other diseases (Adhikari *et al.,* 2019). In spite of its economic benefits to the farmers, tomatoes production in Kenya is constrained by soil-borne diseases and arthropod pests (Moreno *et al.,* 2017; HCDA, 2018; Ochilo *et al.,* 2018).

The major arthropod pests infesting tomatoes are the leaf miner moth *Tuta absoluta* (Meyrick) (Lepidoptera: Gelichiidae), Western flower Thrips *Frankiliniella occidentalis* Pergande (Thysanoptera: Thripidae), Whitefly *Bemisia tabaci* Gennadius (Homoptera: Aleyrodidae), Leaf miner fly *Liriomyza* spp. (Diptera: Agromizydae), Red spider mite *Tetranychus evansi* Baker (Acari: Tetranychidae) and African bollworm *Helicoverpa armigera* (Hubner)

(Lepidoptera: Noctuidae) (Gacheri, 2016; Wakil *et al.*, 2018; Infonet biovision, 2019).

On the other hand, the major soil-borne diseases that cause high losses of up to 100% in tomato production are bacterial wilt caused by *Ralstonia solanacearium* Smith (Burkholderiales: Burkholderiaceae) and Fusarium wilt caused by *Fusarium oxysporum* f. sp. *lycopersici* Schlecht (Hypocreales: Nectriaceae) (Sigei *et al.*, 2014). The pests and diseases outbreak are aided by continuous cultivation of arable land that has resulted in ecological imbalance (Siameto *et al*., 2010).

Foliar fungal diseases also greatly affect tomato production especially early blight *Alternaria solani* Sorauer (Pleosporales; Pleosporaceae), which is more prevalent in the humid tropics (Sanoubar and Barbanti, 2017). In greenhouse environment, the disease is common due to the favourable microclimate that exacerbates its development (Sanoubar and Barbanti, 2017).

Other important foliar diseases that affect tomato production include late blight caused by the fungus *Phytophthora infestans* (Mont) de Bary, anthracnose caused by the fungus *Colletotrichum lagenarium* (Pass.) Ell. and Halst., powdery mildew caused by the fungus *Podosphaera xanthii* U. Braun and N. Shishkoff, fruit rots caused by Monilinia or Botrytis and bacterial soft rots caused by several types of bacteria, but most commonly by species of gram-negative bacteria, Erwinia, Pectobacterium, and Pseudomonas among others.

Plant-parasitic nematodes are among the many pests affecting tomato production and the concomitant infection of root knot nematodes (RKN) and

soil-borne pathogens in tomato production areas results in greater damage and yield losses of over 70% (Syed, 2012; Infonet-Biovision, 2020). The main plant parasitic nematodes that cause damage in tomato include *Meloidogyne incognita* Sasser. Generally it is admitted that there are four major species Meloidogyne such as *Meloidogyne incognita, M. javanica, M. arenaria and M. hapla,* as well as a few emerging species such as *M. enterolobii* and *M. chitwoodi*, that cause the vast majority of crop damage (Moens *et al.*, 2009, Syed, 2012; Infonet-Biovision, 2020). Apart from excessive pest and disease damage, other challenges that prevent farmers from achieving potential tomato yields are unavailability of quality seed, the use of unadapted varieties, low soil fertility, post-harvest losses and the lack of appropriate cultural practices (Ogbomo, 2011; Ayandiji and Adeniyi, 2011).

Currently, the management of soil-borne diseases and arthropod pests mainly rely on chemical pesticides, which are widely used (Moreno *et al.*, 2017; Nguetti *et al.*, 2018). However, the use of chemical pesticides poses significant risks to human health, environment and non-target organisms ranging from beneficial soil microorganisms, to pollinators, predators, parasitoids, plants, fish and birds (Onkendi *et al*., 2014). In view of this, the most common available alternative pest control methods include biological, natural chemical and genetic control.

Plant essential oils are complex natural mixtures of volatile secondary metabolites normally extracted from plants have antimicrobial properties (Kalemba *et al*., 2003; Seow *et al.*, 2013). The use of plant essential oils in

tomato production systems for pest management in Kenya is still below potential (Moreno *et al.,* 2017; Nguetti *et al.*, 2018). Also, soil improvers play a critical role in restoring ecological balance in heavily cultivated soils, which are usually prone to high infestation by soil-borne pathogens (Siameto *et al*., 2010).

Biological control agents such as *Trichoderma harzianum* Rifai (Hypocreales; Hypocreaceae), have been shown to be effective for management of soil-borne fungal diseases and promoting growth (Siameto *et al*., 2010; Hýsek *et al*., 2011; Samia *et al*., 2012; Kipng'eno *et al*., 2015, Kariuki *et al.*, 2020). As well, some bio-stimulants works as growth-promoting agents eliciting induced plant resistance in the areas where they are applied (Murthy *et al.*, 2014). The induced plant resistance plays an important role in pest management and increasing productivity.

An integrated pest management system that has potential to offer a holistic management strategy for soil-borne pathogens and insect pests is the most sustainable alternative. This study was therefore carried out to assess the mechanism and efficacy of integrating plant essential oils, soil improvers, *T. harzianum* and bio-stimulants in the management of soil-borne diseases and the resulting effect of on arthropod pests of tomato was determined.

1.2 Statement of the problem

Tomatoes are grown extensively by small scale farmers in Kenya as high value horticultural crops for domestic market, processing and export. Despite the economic benefit and poverty alleviation, soil-borne pathogens, root knot

nematodes and arthropod pests remain a major constraint to tomato production in Kenya (Masinde *et al*., 2011, Kariuki *et al.,* 2020) resulting in reduced fruit yield and quality.

In order to avoid heavy losses due to early blight infection, tomato farmers have adopted intensive application of fungicides, and other pesticides. The overreliance on fungicides and other chemical pesticides causes several disadvantages due to accumulation of fungicide residues on the tomato fruits above recommended limits (Nguetti *et al.,* 2018). Also, these pesticides causes adverse effects on environment, human health, soil, water resources and the development of resistance among most pathogens, arthropod pests following repeated use have been reported (Sahu *et al.,* 2013).

Information on the effect of integration of thymol, soil improvers and biostimulants for management of tomato pests, diseases, nematodes and rhizosphere microbial dynamics could lead to identifying alternative sustainable and safer management strategies that would be an alternative to frequent use of chemicals.

1.3 Justification of the study

In Kenya, tomato plays a critical role in meeting domestic and nutritional food requirements, generation of income and creation of employment for both the rural and urban populations (Sigei *et al.,* 2014). In spite of its capabilities to alleviate hunger, and it is socio-economic benefits and employment opportunities, arthropod pests remain the major challenge to it is production

resulting in both low yields and poor quality (Sabbour, 2014, Ochilo *et al.*, 2018). On the other hand, synthetic pesticides are being used heavily by farmers as the main strategy for the management of the pests (Bhattacharjee and Dey, 2014).

The effect of plant growth promoting agents *T. harzianum,* bio-stimulants, plant essential oils and soil improvers on soil borne pathogens has been documented (Shen *et al.,* 2015). However, the effect of integrating plant essential oils, soil improvers, *T. harzianum*, plant growth promoting agents and bio-stimulants in the management of soil-borne diseases in tomato production has not been used in managing rhizosphere soil borne pathogens, nematodes, insect pests of tomatoes in Kenya, and there is also need of evaluating their efficacy.

Plant growth promoting agents contribute significantly towards management of soil-borne diseases (Sutanu and Chakrabartty, 2014). Plants respond to insect attack through natural defenses that include induced resistance, which causes reduction in the level of pest attack and damage (War *et al.*, 2012). Biocontrol agents for such as *T. harzianum* and plant growth promoting agents such as rhizobium and *Bacillus* spp., have been reported to elicit induced resistance in plants (Kloepper *et al*., 2004; Harman *et al*., 2008; Murthy *et al*., 2014).

Application of plant essential oils can increase levels of phenolic compounds in plants, which act as elicitors of induced resistance (Mandal *et al*., 2013). Through this study, the efficacy of integrating plant essential oils, soil

improvers, *T. harzianum* and bio-stimulants in the management of soil-borne diseases and the resulting effect on insect pests of tomato was evaluated. However, consumers needs in terms of health and safety and sustainable environmental management can only be achieved by use of alternative mechanisms of pests, diseases and soil fertility enhancement using new technologies that utilizes essential oils, soil improvers and bio-stimulants.

1.4 Research objectives

1.4.1 General objective

To improve productivity in tomato production systems through integration of plant essential oils, soil improvers and bio-stimulants for sustainable suppression of important tomato diseases and pests in Kirinyaga County.

1.4.2 Specific objectives

i. To evaluate the effect of plant essential oils on the control fusarium wilt, early blight and arthropod pests of tomato

ii. To determine the effect of soil improvers on control of fusarium wilt, rhizosphere microorganisms population dynamics and yield of tomatoes.

iii. To determine the effect of bio-stimulants on the control of root knot nematodes and yield of tomatoes.

iv. To determine the effect of integrating plant essential oils, soil improvers and bio-stimulants on the control soil borne pathogens, root knot nematodes, arthropod pests and yield of tomatoes.

1.5 Hypotheses

i. Plant essential oils are effective on fusarium wilt, early blight and arthropod pests of tomato

ii. Soil improvers influence the soil rhizosphere microbial dynamics, incidence of fusarium wilt and yield of tomato.

iii. Bio-stimulants are effective in the management of fusarium wilt, insect pests of tomato in addition to increasing yield

iv. Integrating plant essential oils, soil improvers and bio-stimulants plant essential oils, soil improvers and bio-stimulants is an effective approach for management of soil borne pathogens, root knot nematodes, arthropod pests and yield of tomato.

1.6 Conceptual framework

The concept of sustainable pest and disease management is one of the ways to overcome threats to food security. Attaining food security, will on the other hand lead to improved livelihoods and incomes. The conceptual framework (Figure 1.1) was drawn to assist in implementation of the integrated pest management system that will contribute to sustainable pest and disease management.

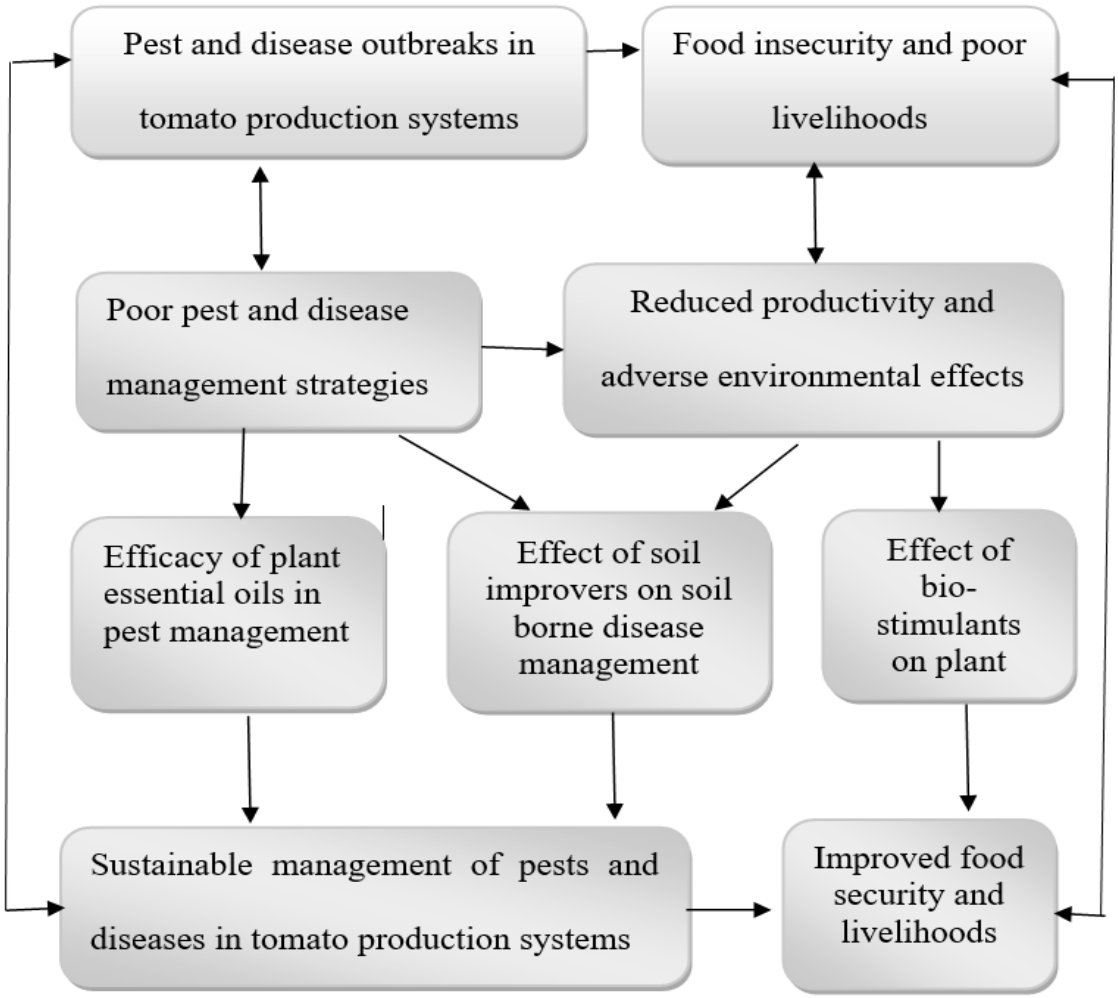

Figure 1.1: Conceptual framework

Sustainable pest and disease management in tomato production depends on an integrated pest and disease management system that will include plant essential oils, soil improvers and bio-stimulants. This integrated system will lead to an increase in tomato productivity, which will contribute to food security and improve the livelihoods of many smallholder famers.

CHAPTER TWO: LITERATURE REVIEW

2.1 Tomato production in Kenya

Tomatoes *Solanum lycopersicum* L. are among the most widely cultivated vegetables crops, not only in Kenya but also in other parts of East Africa and the whole world at large. It is ranked third after potatoes and sweet potatoes (FAO, 2020). Tomato is an important vegetable in the Kenyan agricultural sector which contributes over 27% to the Gross Domestic Product (GDP) of the country (FAOSTAT, 2020).

Tomato, contributes significantly to Kenyan economy through meeting nutritional food requirements, generation of income and creation of employment (Sigei *et al.*, 2014; Mwangi *et al.*, 2020). Tomato is a major source of vitamin A and C and supplies a sufficient amount of the antioxidant lycopene pigment that helps to protect consumers against diseases (Adhikari *et al.,* 2019).

Tomato contains 16 calories per 100 g serving, 0.2 g of fat, 1.2 g of protein and 3.2 g of carbohydrate (USDA, 2020). Tomatoes, raw, orange contains 0 g of saturated fat and 0 mg of cholesterol per serving. 100 g of tomatoes, raw, orange contains 1496.00 IU vitamin A, 16.0 mg of vitamin C and 0.00 mcg of vitamin D as well as 0.47 mg of iron, 5.00 mg of calcium and 212 mg of potassium (USDA, 2020). Commercial tomato production has emerged as a viable agribusiness opportunity in Kenya with the area under production increasing (Wachira *et al*., 2014; FAOSTAT, 2020).

The production of tomato plays an important role towards poverty alleviation and economic growth. Kenya recorded an annual production of 599,458 tons in 2018 on 28,263 hectares under production (FAOSTAT, 2020). Tomato crop is reasonably adaptable and grows vigorously in warm conditions with optimum temperatures range of 15°C to 25°C.

Adverse environmental conditions for instance high temperature, reduce fruit set and yields (Bawa, 2016). Extremely low temperatures delay colour formation and ripening while temperatures above 30°C inhibit fruit set, lycopene and flavour development (Bawa, 2016).

2.2 Soil-borne pathogens in tomato production systems

In Kenya, soil-borne pathogens remain a major constraint to tomato production (Mwangi *et al.*, 2020). Tomato growers in the country have reported bacterial wilt with over 64% crop loss reported for open field production and up to 100% in greenhouses thus threatening production of the crop (Kariuki *et al.,* 2020).

The major soil-borne diseases that cause high losses in tomato production are bacterial wilt caused by *Ralstonia solanacearium* Smith (Burkholderiales: Burkholderiaceae), Fusarium wilt caused by *Fusarium oxysporum* f. sp. *lycopersici* Schlecht (Hypocreales: Nectriaceae) (Kago *et al.,* 2016).

Bacterial wilt caused by *Ralstonia solanacearium* Smith (Burkholderiales: Burkholderiaceae) is a soil-borne gram-negative bacterium which affects over 200 families of plants, especially those in the family solanaceae family (Manani *et al.*, 2020). The bacterium normally infects plant roots from the soil through wounds or natural openings, colonizes the intercellular space of the root cortex and vascular parenchyma, and eventually enters the xylem vessel and spreads up into the stem and leaves. Generally, *R. solanacearum* manifests through sudden wilting of foliage.

The bacterium multiplies rapidly in the vascular system. The rapid multiplication causes the xylem elements to fill with bacterial cell and slime (Monther and Kamaruzaman, 2010). The disease develops rapidly in warm weather, especially after heavy rain or flooding. Symptoms start as leaf drooping, followed by the complete plant wilting within a few days. Recently wilted plants are green which is a distinct symptom compared to other vascular diseases like Fusarium wilt, which develop yellowing of leaves (Manani *et al.*, 2020).

Fusarium wilt causes immense losses given the pathogenic variations in the causative pathogen makes it difficult to manage (Chopada *et al*., 2014). The causative pathogen occurs in most tomato growing regions in the world causing vascular wilt that can severely affect the crop (Moretti *et al.*, 2008). The interactions of soil-borne plant pathogens with plant parasitic nematodes result in severe incidences of the disease since the damage caused by the nematodes increase serve as ingress points for the pathogens (Westphal, 2011).

Currently, management of soil-borne diseases mainly rely on chemical pesticides, which are widely used (Moreno *et al.*, 2017; Nguetti *et al.*, 2018). However, control of bacterial wilt remains a challenge since it has a wide host range, long survival in the soil and a wide biological variation. No single management strategy has been found to be significantly effective. The current available management options for Fusarium wilt include the use of fungicides, resistant cultivars, cultural practices and biological control agents (Wagacha *et al*., 2007).

2.3 Soil microbe prevalence and insect pest interactions

Production of tomatoes in Kenya is mainly by small scale farmers who mainly rely on soil as the media for production. Various types of soils are rich in high numbers of phytopathogenic microbes that are capable of devastating crops contain substantial numbers of phytopathogenic microbes capable of devastating (Singh *et al*., 2017). Some of these microbes include groups of fungi and bacteria. During plant growth, plants interact with the microbes in the soil that impact either positively or negatively to their growth (Vishwakarma *et al.*,2020).

The microbial organisms are mainly found at the rhizosphere and depending on type present, could possess disease causing properties, growth promotion properties, disease suppression properties among other attributes (Vishwakarma *et al.*,2020). The interactions between soil microbes and plants result in mediated interactions between below ground micro-organisms and above ground insect pests (Wondafrash *et al*., 2013).

Herbivore-induced responses are usually systemic and therefore can influence the performance and preference of organisms for certain plants (Wondafrash *et al*., 2013). This Herbivore-induced interactions can be useful in the management of insect pests in agricultural production systems.

2.4 Pests of tomatoes

Arthropod pests are the one of the most significant constraints in tomatoes production. The insect pests account for about 34% of the biotic factors impeding tomato production in Kenya (Ochilo *et al.,* 2018). Some of the agricultural pests infecting the plant during their growing season include; whiteflies *Bemisia tabaci*, the tomato leaf miner *Tuta absoluta* , the green peach aphids *Myzus persicae* and the red spider mite *Tetranychus urticae* (Abbas *et al.*, 2020).

The spider mites damage tomatoes by sucking up plant sap using their stylet-like mouth parts. They are usually prevalent in dry areas and are found on the underside of the leaves near the veins (Omukoko *et al.,* 2017). Infested tomato leaves and fruits show a white to yellow speckling. Heavy spider mite infestation causes defoliation while attacked plants yield very small fruits with reduced level of vitamin C. Increased infestation of spider mites causes webbing on plants which can result to death of plants (Infonet-biovision, 2022).

The leaf miner fly *Liriomyza* spp., is equally a very destructive pest of tomatoes (Stuart *et al.,* 2016). This dipteran family Agromyzidae is a distinguished group of small, morphologically similar flies composing of about 1,800 species worldwide, with 75% of them producing mines in leaves (Bader, 2006).

The pest is polyphagous, infesting many host plants including horticultural crops and weeds. They cause damage by puncturing the leaf surface where larvae of flies proceed to feed on the tissue (DEFRA, 2007). The adult females cause damage on the mesophyll cells of host plant due to ovipositor probing (Lopez *et al.,* 2010) to lay eggs. When eggs are hatched, the larvae begin to tunnel within the leaf tissue forming damaging and disfiguring mines usually serpentine in nature.

Leaf mines reduce the quality of crops in addition to reducing the photosynthetic capability of the plant (DEFRA, 2007) and causing tissue death (Lopez et al., 2010). During feeding and egg laying, the leaf miners create small punctures on the side of tomato leaves which may act as entry points for harmful micro-organisms such as *Alternaria alternata* Keissl that causes leaf spot *Cercospora capsici* Heald on tomato (Shailendra, 2018).

Whitefly *Bemisia tabaci*, is a key pest of tomatoes which attacks the crop at all stages of its growth (Sudeepa and Manoj, 2017). Members of the *B. tabaci* group are highly prolific, polyphagous and invasive crop pest found all over the world, and cause significant yield decline (Abate *et al.,* 2000). For tomato, *B. tabaci* has been associated with both direct and indirect damage caused by feeding directly on the tomato plants, sucking sap from the phloem resulting in leaf and fruit spotting, weakening of plants and irregular fruit ripening (Muigai *et al.*, 2002).

In addition, *B. tabaci* has also been associated with transmission of many viral diseases which negatively impact crop yield (Mansoor *et al.,* 2003). The

whitefly nymphs secrete honeydew a clear sugary liquid which lead to development of black sooty mould. The coating of the sooty mould decreases the photosynthetic area of the plants leading to reduced plant growth. They also cause indirect damage to tomatoes as key vectors of viral diseases such as Tomato Yellow Leaf Curl Virus (TYLCV), a major disease in Kenya (Marabi *et al.,* 2017).

The African bollworm *Helicoverpa armigera* is a serious insect pest and has been reported to cause damage in tomato production in Kenya (Wabule, 1997). *Helicoverpa armigera* main damage is caused by old larvae which feed on flower buds, flowers and by burrowing into the young tomato fruits (Syngenta, 2011) causing secondary infection which is followed by rotting of the fruit. In addition, it attacks many other different crops such as beans, pigeon peas, cotton, maize and different weeds (Jallow *et al.*, 2005).

The immature stage (caterpillar) feeds on the young and developed fruits of tomato boring into the fruit leading to severe losses (Konje *et al.*, 2019). The damaged fruits eventually decay and rot due to secondary infections caused by other pathogens for example fungi and bacteria. Through boring the fruits, they can cause up to 70% yield loss (Sumitra *et al.*, 2012; Kumar, 2013).

2.5 Current management practices for soil-borne pathogens

The Solanaceae family of crops which tomato belong to is prone to infestation by soil borne pathogens (Mamphogoro *et al.*, 2020). Farmers have to deploy different pest management strategies for them to be able to produce

tomatoes of good quality and quantity. The current pest management practices for soil-borne diseases is use of pesticides (Nguetti *et al.,* 2018). The indiscriminate use of chemical pesticides poses significant risks to human health, environment and non-target organisms (Bawa, 2016).

The adverse effects caused by some of this chemical pesticides for instance nematicides has led to some being withdrawn from the market (Onkendi *et al*., 2014). Indiscrimate use of the pesticides has contributed to reduction in bio-divesrsity resulting in ecological imbalance. Ecological imbalance has been observed to be a fertile ground for thriving of soil-borne pathogens which affect production of tomato (Siameto *et al*., 2010).

Other pest management options that have been deployed include biological control and cultural practices especially grafting for resistance (El-Shennawy *et al*., 2012), crop rotation, mulching, spacing and use of tolerant varieties (Mandal *et al*., 2013; Bawa, 2016). The pest and disease management practices deployed have some level of success but the soil borne pathogens continue to pose a challenge to production of tomato (Mwangi *et al.*, 2020). Other alternative sustainable approaches have to be for farmers to produce tomatoes in a sustainable way.

2.6 Plant essential oils in pest management

Plant essential oils and phenolic compounds are natural volatile mixtures usually produced by various plants for pigmentation, growth, reproduction, pest resistance among other functions (Lattanzio *et al.*, 2006). Phenolic compounds

are usually as a result of secondary metabolism, which is a process, required mainly for plants survival in the environment (Lattanzio *et al.,* 2006). For many years, plant essential oils have had a wide application in folk medicine, food flavouring and preservation as well as in fragrance industries (Kalemba *et al.,* 2003). They have anti-microbial activity, which is an important property for pest and disease management purposes (Kalemba *et al.,* 2003).

Some plant essential oils for example salicylates act as feeding barriers to insect pests (Lattanzio *et al.,* 2006). Plant flavonoids have also been reported to affect development of some insects and this can be utilized in pest management. Plant essential oils have been reported to have nematicidal effects on the root-knot nematode (Cetintas *et al.,* 2010). Plant essential oils have also been repoterd to inhibit pathogen growth and also control of bacterial speck in tomato (Silva *et al.,* 2014)

2.7 Management of soil-borne pathogens using *Trichoderma* spp.

Trichoderma spp, is an ubiquitous fungal species found in a wide variety of environment worldwide (Akladious *et al.,* 2012). The species colonizes the rhizosphere of plants and are usually beneficial to plants. Apart from protecting the plants from pathogenic microorganisms, they increase productivity (Harman *et al.,* 2008). Application of *Trichoderma* spp. by drenching in the soil or seed inoculation results in the microorganism colonizing the roots of the plants in a symbiotic relationship and providing protection to the plants against diseases (Harman *et al.,* 2008).

The application of *Trichoderma* spp. can therefore be an alternative to chemical pesticides for the management soilborne pathogens that include Fusarium wilt in tomatoes (Mushtaq *et al.,* 2011). The beneficial fungi are reported to be effective in the management of the fusarium wilt fungus and root –knot nematodes (El-Shennawy *et al*., 2012). *Trichoderma* spp. contributes to disease management and plant growth promotion in plant production in various ways. One of the modes of action is through eliciting induced systemic resistance in plants (Kloepper *et al*., 2004; Harman *et al*., 2008; Murthy *et al*., 2014). By developing induced resistance, plants are to tolerate disease attack and thereby reduce the need for application of chemical pesticides.

Competition is another important mode of action. The *Trichoderma* spp. competes aggressively with pathogenic microorganisms thus inhibiting their disease-causing abilities. *Trichoderma* spp. has ability to produce antibiotics and act as a mycoparasite all of which contribute to the management of soil-borne diseases (Harman *et al.,* 2008). Incorporation of *T. harzianum* in cropping systems results in differences in soil microbial populations, which could be a contributing factor to the differences in the levels of plant resistance to diseases (Meijun *et al.,* 2011).

Biological control agents and bio-pesticides integrated with some chemical pesticides provide a sustainable alternative to the current overreliance on chemical pesticides. Plant essential oils are phenolic compounds being utilized in many applications (Kalemba *et al*., 2003; Vagiri *et al.,* 2017), offers an alternative to chemical pesticides.

Soil improvers play an important role in restoring soil balance, which is a key element in management of soil borne diseases (Siameto *et al*., 2010). The integration of soil improvers contributes positively to pest management efforts. The application of *Trichoderma* spp. can therefore be a suitable alternative to chemical pesticides for the management of soil-borne pathogens that include Fusarium wilt in tomatoes (Mushtaq *et al.*, 2011). These potential management strategies can play a role in management of soil-borne pathogens and insect pests in tomato production systems when harnessed synergistically.

CHAPTER THREE: MATERIALS AND METHODS

3.1 Study sites

The field experiments were carried out in a farmer's field in Kimbimbi whose area coordinates are 0.6204° S, 37.3654° E and Kagio at 0.6251° S, 37.2531° E in Kirinyaga County (Figure 3.1). Kagio lies on 1248m above sea level (ASL) with an average annual temperature of 20.6 °C. The area receives a bimodal rainfall with an average annual rainfall of 1,149 mm. The area has soils classified as Rhodic Ferralsols and Niti-Rhodic Ferralsols (Jaetzold, 2005).

The study site in Kimbimbi is situated in the lower midland zone 4 (LM4) at an altitude of 1,050 m asl. It is under a semi-arid area with soils classified as Nitisols (Njinju *et al.,* 2018). The area experiences a bimodal rainfall with an average rainfall of about 850 mm and temperature of 22ºC. These conditions make both areas conducive for tomato production throughout the year. The greenhouse experiments were conducted at Kenyatta University whose coordinates are 1.0500° S, 36.5541° E. The greenhouse had an irrigation system and soil as the planting media where the crop was established.

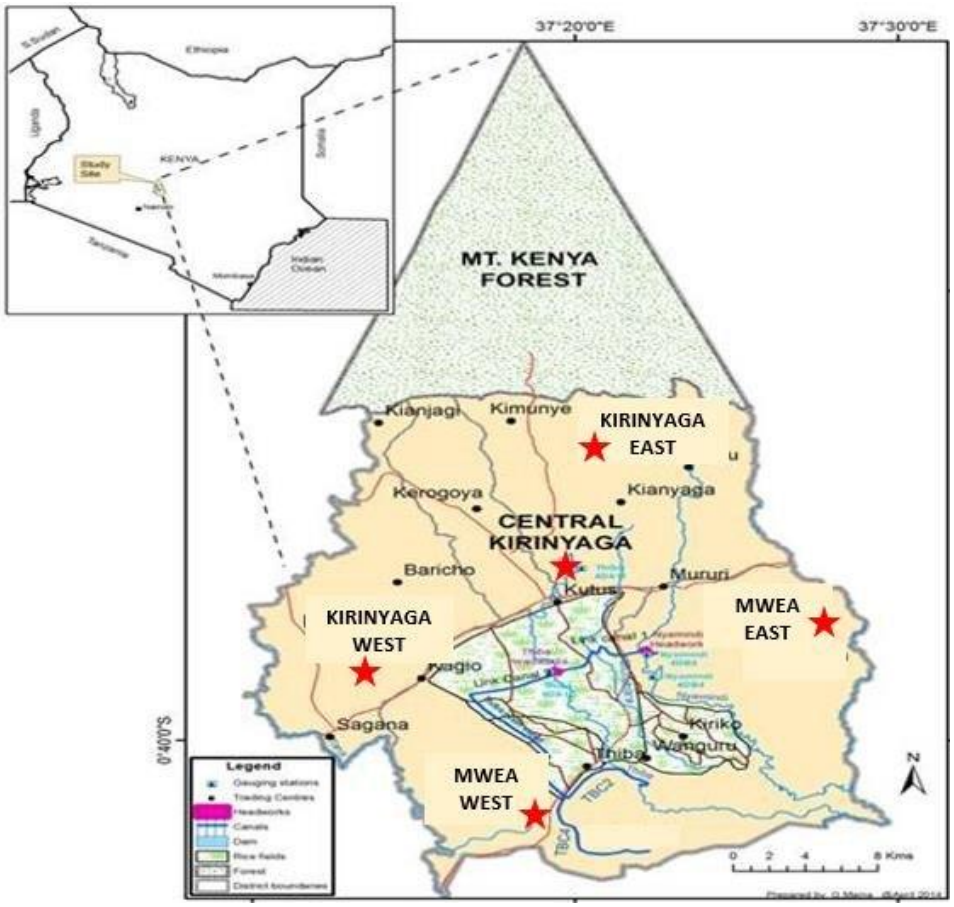

Figure 3.1: Map showing Kirinyaga County- Kenya.
Source: Google.com

3.2 To evaluate the effect of plant essential oils on fusarium wilt, early blight and arthropod pests of tomato

3.2.1 Experimental treatments

The field experiments to determine the effect of plant essential oils on fusarium wilt and insect pests of tomato, were laid out in Kimbimbi and Kagio between September to December 2016. The treatments comprised of varying rates of the plant essential oils, an insecticide, a fungicide and an untreated control as shown in (Table 3.1).

Table 3.1: Experimental treatments: objective one

Treatment	Description
Control (-ve).	Application with water only
Thyme oil	Thyme oil at 0.5% of solution
Thyme oil	Thyme oil at 1% of solution
Thyme oil	Thyme oil at 1.5% of solution
Insecticide	Lambda-cyhalothrin
Fungicide	Carbendazim

Plant essential oils applied at transplanting followed by subsequent applications at 7 days interval. The standard chemical fungicide and insecticide applied at the recommended rates.

3.2.2 Study design and layout

The experiments were set up in a Randomized Complete Block Design (RCBD) with four replications. Treatment plots measured 3m x 4m with 1m spacing between the plots and 1.5m spacing between the blocks. The tomato variety used for the experiments was Zara F1 (which is recommended for both outdoor and greenhouse production) and the treatments (Table 3.1) allocated randomly. The seedlings were raised in small planting pots with media (Plate A) and transplanted when they were 5 weeks old to the in the experimental plots.

Plate A: 5 Week old tomato seedlings ready for transplanting

3.3 To determine the effect of soil improvers on soil microorganisms and yield of tomatoes

3.3.1 Experimental treatments

The field experiments to determine the effect of soil improvers on soil microorganisms and yield of tomato were carried out in Kimbimbi and Kagio between February to July 2017. The treatments comprised of three rates of the soil improvers (200kg/ha, 300kg/ha and 400kg/ha), standard soil amendment (Minjingu Phosphate Rock) at the recommended rate, farmer practice (application of manure) at the recommended rate and an untreated control as shown in (Table 3.2).

Table 3.2: Experimental treatments: objective two

Treatment	Description
T1	Control (-ve).
T2	Soil improver lower rate (200 kg/ Ha).
T3	Soil improver medium rate (300 kg/ Ha).
T4	Soil improver high rate (400 kg/ Ha).
T5	Standard soil amendment (Minjingu Phosphate Rock) at 250kg/ Ha.
T6	Farmer practice (Manure) at the rate of 5 tons per Ha

All the treatments indicated in (Table 3.2) were applied at once after the plots and planting rows were prepared. The treatments were applied in the planting rows and mixed with the top soil. A week later, the tomato seedlings were transplanted into the experimental plots.

3.2.2 Experimental design and layout

The experiments were set up in a Randomized Complete Block Design (RCBD) with four replications. The plots measured 3m x 4m with 1m spacing between the plots, while a 1.5m spacing was maintained between the blocks. The tomato variety used for the experiments was Zara F1 and the treatments were as shown in (Table 3.2). The tomato seedlings were raised in small planting pots with media as shown in (Plate A; sub-section 3.1.2) and they were transplanted after 5 weeks to the experimental plots.

3.4 To determine the effect of bio-stimulants on root knot nematodes and yield of tomatoes

3.4.1 Experimental treatments

The experiment to determine the effect of bio-stimulants on root knot nematodes and yield of tomato was in a greenhouse at Kenyatta University from July to December 2018 for the first season and between January to June 2019. The experimental treatments consisted of different bio-stimulants compared to the untreated control (Table 3.3).

Table 3.3: Experimental treatments: objective three

Treatment	Description
T1	Control (-ve)
T2	*T. harzianum* applied at 2.5kg/ Ha
T3	Seaweed extract for root stimulation applied at 3L/ Ha
T4	Seaweed extracts plus amino acids and peptides applied at 3L/Ha
T5	Seaweed extracts plus humic acids applied at 3L/Ha
T6	Amino acids applied at 2L/Ha

The treatments we applied as follows; *T. harzianum* applied at transplanting followed by two monthly applications of the other four treatments; T3, T4, T5 and T6 as shown in Table (3.3). The treatments were applied at a two weeks interval from transplanting up to week twelve.

3.4.2 Experimental design and layout

The experimental design was Randomized Complete Block Design (RCBD) with four replications. The plots measured 1.5m x 6m with 0.5m spacing between the plots and 1m spacing between the blocks. The tomato variety used for the experiments was Zara F1 and the treatments as indicated in (Table 3.3) were allocated randomly within the blocks. The seedlings were raised in small planting pots with media as shown in (Plate A; section 3.1.2) and transplanted at 5 weeks old to the in the experimental plots.

3.5 To determine the effect of integrating plant essential oils, soil improvers and bio-stimulants on the management of soil borne pathogens, root knot nematodes, arthropod pests and yield of tomato

3.5.1 Experimental treatments

The field experiments to determine the effect of integrating plant essential oils, soil improvers and bio-stimulants on the management of soil borne pathogens, root knot nematodes, arthropod pests and yield of tomato, were carried out in Kimbimbi and Kagio as shown in (Table 3.4). This was conducted between July to November 2019.

The experimental treatments consisted of different combinations of the plant essential oils, soil improver, *T. harzianum* and the bio-stimulants; compared to the untreated control and a standard chemical treatment that involved alternate treatment using insecticides and fungicides.

Table 3.4: Experimental treatments: objective four

Treatment	Description
T1	Control (-ve).
T2	Thyme oil + Soil improver + *T. harzianum* + Seaweeds + Amino acids
T3	Thyme oil + Soil improver + T. harzianum + Seaweeds + Humic acids
T4	Thyme oil + Soil improver + *T. harzianum* + Seaweeds + Peptides.
T5	Thyme oil + Soil improver + *T. harzianum* + Seaweeds + Amino acids + Humic acids.
T6	Thyme oil + Soil improver + *T. harzianum* + Seaweeds + Amino acids + Peptides.
T7	Thyme oil + Soil improver + *T. harzianum* + Seaweeds + Humic acids + Peptides.
T8	Thyme oil + Soil improver + *T. harzianum* + Seaweeds + Amino acids + Peptides + Humic acids
T9	Standard chemical treatment

The treatments were applied subsequently as follows: Thyme oil was applied at planting followed by a biweekly interval. The soil improver was applied once at planting, while the *T. harzianum* was applied at planting followed by two monthly applications, whereas the seaweeds, amino acids, humic acids and peptides were applied at an interval of two weeks from transplanting up to week twelve.

3.5.2 Experimental design and layout

The experiments were set up in a Randomized Complete Block Design (RCBD) with four replications. Treatment plots measured 3m by 4m with 1m spacing between the plots and 1.5m spacing between the blocks. The tomato variety used for the experiments was Zara F1 and the treatments used are as

shown in Table 3.4. The seedlings were raised in small planting pots with media (Plate A; section 3.1.2) and transplanted at 5 weeks old to the experimental plots.

3.6 Data collection and assessments

3.6.1 Assessment of fusarium wilt incidence

The assessment of fusarium wilt was carried out in all the four experiments of the four objectives. During crop development, weekly scouting for fusarium wilt incidences started 4 weeks after transplanting (WAT) of tomatoes. The assessment was done on all the number of plants in each plot and those plants showing fusarium wilt symptoms were recorded. The fusarium wilt score was determined using symptoms showing yellowing on one side of the plant leaf beginning with the older and bottom leaves. This was followed by infected leaves wilting, browning, and defoliation (Infonet-Biovision, 2020). The data as expressed as a percentages and recorded as disease incidence (DI) as shown in the formula (Michel *et al.*, 2006).

$$D = \frac{\text{Number of plants showing wilt symptoms}}{\text{Total number of plants}} \times 100$$

Key: D= % Disease incidence

3.6.2 Assessment of early blight infection

Early blight disease was assessed in experiments under objective one and four. Weekly scouting of early blight started one week after transplanting and was carried out through sampling ten randomly selected plants per plot. The

symptoms used to identify early blight infection included, small dark spots on older foliage near the ground, round, brown leaf spots about half inch in diameter, larger leaf spots with target-like concentric rings and for severe infection, brown leaves falling off, or dead on the stems (Infonet-Biovision, 2020). The percentage leaf area infected by early blight was estimated from five leaves between the 3rd and 7th leaf (from the top) on each of the sampling stems. The disease severity expressed based on the scale in Table 3.5 (Pandey and Pandey, 2002).

Table 3.5: Disease rating scale for early blight infection on tomato

Rate	Percentage of leaf area affected
0	0%
1	1-10%
2	11-25%
3	26-50%
4	51-75%
5	>76%

The individual disease ratings from the sampling stems expressed and recorded as the percent disease index (PDI) per treatment as follows (Pandey and Pandey, 2002):

$$PDI = \frac{\text{Sum of all individual ratings}}{\text{Total number of leaves assessed} \times \text{Maximum disease category}} \times 100$$

Key: PDI= % Disease incidence

3.6.3 Assessment of thrips population

The assessment of thrips population was done in experiments under objective one and four. Weekly scouting between 0800 and 0900hrs for thrips started immediately after transplanting. Ten plants were randomly selected in the net plot and the number of thrips assessed. The thrips population was determined using a mechanical tap method technique where each shoots of the randomly selected plant was shaken in a small box to dislodge any available thrips (Shipp *et al*., 2000). The number of dislodged motile thrips was recorded as per the plant.

3.6.4 Assessment of whitefly population

The whitefly population was assessed in experiments under objective one and four. Weekly scouting between 0700 and 0800hrs of whiteflies, started immediately after transplanting. The assessment was by plucking the third leaf from the top of ten randomly selected plants in the net plot of each treatment. The leaf samples were examined on the underside and the whitefly counted and recorded from each treatment plot according to the procedure described by Hugh and Curtis (2014).

3.6.5 Assessment of the soil rhizosphere microbial population

The assessment of the soil rhizosphere microbial population was carried out in experiment under objective two. The soil rhizosphere microbial composition was determined based on phospholipid fatty acids (PLFA) and microbial biomass Carbon (MBC) analysis (Wang *et al.,* 2018). The microorganisms of focus were bacteria and fungi at the beginning and at the end

of the experiment. The microbial population of fungi and bacteria was expressed in form of total and active fungi and bacteria respectively. The active populations were determined using a plate culture method and the PCR–DGGE analysis (Meijun *et al.*, 2011).

3.6.6 Assessment of root knot nematode infestation

The assessment of the root knot nematode infection was determined in experiments under objective three and four. Before the start of the experiment, a composite soil sample was collected and taken to Kenyatta University's Agriculture Science and Technology laboratory for nematode analysis. During crop development, five plants were destructively randomly sampled in each treatment at eight and sixteen weeks after start of treatments. Five randomly selected tomato plants were uprooted in each treatment at eight and sixteen weeks after tomato planting.

The roots of the uprooted plants were washed and intensity of root galling due to root knot nematode infestation was rated. This was done by using a scale of 0 to 5 based on the percentage of the root system with galls (Hussey *et al.*, 2002). Where 0 = no galling; 1 = trace infection with a few small galls; 2 = ≤ 25% roots galled; 3 = 26 to 50% roots galled; 4 = 51 to 75% roots galled; and 5 = >75% roots galled and visually using Gall Severity Index scale (Gapasin, 1980) as illustrated in Figure 3.2.

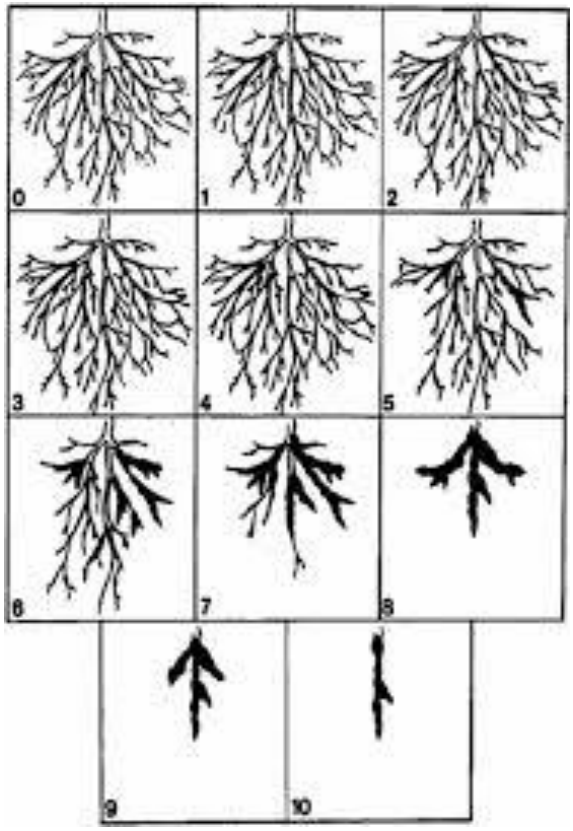

Figure 3.2: Root Knot Nematode Galling Severity Scale.

Source: Google.com, 2021

3.6.7 Pathological analysis of soil samples for soil borne pathogens

The pathological analysis was carried out before the start of every field experiments. A composite soil sample from each field was collected and taken to the Kenyatta University's Agriculture Science and Technology laboratory for pathological analysis. The soil samples were collected by using a soil auger where small soil samples were collected in a zigzag pattern at 20 different spots in the field. Before picking of the sample, the first 3cm of top soil was scrapped off and the sample picked from a depth of 10-15cm. The samples were then thoroughly mixed to make a composite sample from which 500g soil per trial

site was weighed and put in sampling bags and sent to the laboratory for pathological analysis.

3.6.8 Assessment of tomato yield

The yield data was collected in form of the number of tomato fruits harvested. The fruits were harvested weekly from selected five plants starting from eight weeks after transplanting (WAT) in the net plot. The tomatoes were graded as marketable and non-marketable yield. This was in terms of; their size, presence of insect pests, bruising and infection by pathogen. The weight of the tomato fruits was then determined using a digital weighing scale and recorded. The data was extrapolated and expressed as tons per hectare for each treatment.

3.7 Data management and statistical analysis

The data collected on Fusarium wilt, early blight, thrips, whiteflies, soil microbial dynamics, and root nematodes was subjected to Analysis of Variance (ANOVA) using GenStat statistical package. Where treatments had significant differences at $P \leq 0.05$, the treatment means were separated using Fisher's protected least significant difference (LSD) (Maindonald, 1992). For the insect counts, the data was transformed before being subjected to Analysis of Variance using GenStat statistical package. Where treatments had significant differences at $P \leq 0.05$, the treatment means were separated using Fisher's protected LSD (Maindonald, 1992).

CHAPTER FOUR: RESULTS

4.1 Effect of plant essential oils on fusarium wilt, early blight and arthropod pests of tomato

4.4.1 Soil pathological analysis

The pathological analysis on the soil indicated presence of the pathogen *Fusarium oxysporum* in both Kagio and Kimbimbi Figure 4.1. The Kagio site had moderate infection while the Kimbimbi site had light infection. The tomatoes seedlings were able to establish and grow at both locations (Plate B)

		Bacterial Pathogens				Fungal Pathogens						Saprophytic Fungi				
Sample No.	Field Name	*Ralstonia solanocearum*	*Xanthomonas spp*	*Agrobacterium tumefasciens*	*Pseudomonas spp*	*Fusarium spp*	*Fusarium oxysporum*	*Alternaria spp*	*Verticillium spp*	*Rhizoctonia spp*	*Pythium spp*	*Trichoderma spp*	*Aspergillus spp*	*Penicillium spp*	*Rhizopus*	*Mucor*
CK105PT0042	Kagio Sample A	0	0	0	0	0	3	0	0	0	0	0	2	0	0	0
CK105PT0043	Kimbimbi- Sample B	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0

Key:		Colour code
0	Not detected	
1	Starting infection	
2	Light infection	
3	Moderate infection	
4	Infected	
5	Severely infected	

Figure 4.1: Pathological analysis results for Kagio and Kimbimbi trial sites

Plate B: Crop establishment in Kimbimbi

4.4.2 Efficacy of thyme oil on incidences of fusarium wilt in tomato fields

The application of thyme oil in tomato was highly significant P<0.001 in reducing the incidence of fusarium wilt both in Kimbimbi and in Kagio field sites compared to the untreated control (Table 4.1). The application of Carbendazim was highly significant P≤0.05 reducing the Fusarium wilt incidence compared to the untreated control in both trial. All the three rates of thyme oil at 0.5%, 1% and 1.5% tested had highly significant P<0.001 low levels of fusarium incidence compared to the untreated control. The three rates had a reduction of 49.6%, 65.9% and 62.8% respectively in Kimbimbi and 53.8%, 50.8% and 55.4%, respectively in Kagio compared to the untreated control.

There were no significant P≤0.05 differences observed among the three levels of thyme oil in the reduction of fusarium wilt in both locations (Table 4.1). This represents a reduction of 53.5 and 60% respectively in both locations compared to the untreated control. On the other hand, the plots treated with

lambda-cyhalothrin were not significantly P≤0.05 different from the untreated control Table 4.1. Both the Lambda-cyhalothrin treated plots and the untreated control had high fusarium wilt incidences.

Table 4.1: Mean ± SE incidence of fusarium wilt infection in tomato plots sprayed with various concentrations of thyme oil in Kimbimbi and Kagio field sites

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	4.6 ± 0.51^{a}	-	4.6 ± 0.26^{a}	-
Thyme oil 0.5%	2.3 ± 0.23^{b}	49.6	2.1 ± 0.25^{b}	53.8
Thyme oil 1.0%	1.6 ± 0.29^{b}	65.9	2.3 ± 0.10^{b}	50.8
Thyme oil 1.5%	1.7 ± 0.11^{b}	62.8	2.1 ± 0.35^{b}	55.4
Lambda-cyhalothrin.	4.2 ± 0.21^{a}	8.5	4.6 ± 0.58^{a}	1.6
Carbendazim.	2.1 ± 0.41^{b}	53.5	1.9 ± 0.13^{b}	60.0
P-Value	<0.001		<0.001	
LSD	0.901		0.845	
SED	0.423		0.396	

Means followed with the same letter along the columns are not significantly different according to Fisher's Protected LSD test at P≤0.05.

4.4.3 Efficacy of thyme oil on early blight infestation in tomato fields

The application of the three rates of thyme oil were as affective as the carbendazim treatment in the management of early blight in both Kimbimbi and Kagio field sites. The results indicated a highly significantly P<0.001 lower percent disease index in plots treated with the three rates of thyme oil compared

to the untreated control and the lambda-cyhalothrin treated plots in both locations (Table 4.2).

Plots treated with the fungicide recorded a lower percent disease index (PDI) compared to the three rates of thyme oil in Kagio with a reduction of 77.5% in early blight infestation. However, this was not significantly at $P>0.05$ different from the application of thyme oil at the three rates of 0.5%, 1% and 1.5% which caused a reduction in early blight infection by 77.0%, 73.9% and 58.9% respectively. The application of thyme oil at the rate of 1.0% spray solution caused the highest reduction of early blight infection of 84.8% in Kimbimbi field site. This reduction was not significantly $P>0.05$ different from the application of carbendazim with a reduction of 83.1%, application of thyme oil at 0.5% with a reduction of 79.1% and application of thyme oil at 1.5% which resulted in a reduction of 81.7% (Table 4.2).

Plots treated with insecticide (Lambda-cyhalothrin) were not significantly $P>0.05$ different from the untreated plots with both recording the highest percent disease index in both locations. The early blight infection was higher in Kimbimbi compared to Kagio. This is evident in the untreated control that had a mean percent disease of 33.0 in Kimbimbi and 24.8 in Kagio (Table 4.2).

Table 4.2: Mean ±SE of early blight infection on tomatoes sprayed with various concentrations of thyme oil in Kimbimbi and Kagio field sites

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	33.0 ± 1.65[a]	-	24.8 ± 1.49[a]	-
Thyme oil 0.5%	6.9 ± 1.58[b]	79.1	5.7 ± 0.57[b]	77.0
Thyme oil 1.0%	5.0 ± 0.93[b]	84.8	6.5 ± 0.95[b]	73.9
Thyme oil 1.5%	6.1 ± 0.99[b]	81.7	10.2 ± 3.68[b]	58.9
Lambda-cyhalothrin.	31.8 ± 1.62[a]	3.6	23.5 ± 1.70 [a]	5.0
Carbendazim.	5.6 ± 0.96[b]	83.1	5.6 ± 0.67[b]	77.5
P-Value	<0.001		<0.001	
LSD	3.913		5.935	
SED	1.836		2.785	

Means followed with the same letter along the columns are not significantly different according to Fisher's Protected LSD test at P≤0.05.

4.4.4 Efficacy of thyme oil on Thrips infestation on tomatoes

The treatments had a significant (P<0.001) effect on Thrips population in both Kimbimbi and Kagio field sites (Table 4.3). Application of thyme oil at the rate of 1.0% spray solution had the highest reduction of Thrips population at 54.4 % reduction in Kimbimbi. This was not significantly different from the other thyme oil rates (0.5% and 1.5%) and the insecticide which elicited a reduction of 51.2%, 47.0% and 53.7% thrips, respectively.

In Kagio, the application of insecticide had the highest reduction of thrips population at 68.6 %. This was not significantly different at P≤0.05 from thyme oil rates at 0.5%, 1.0% and 1.5% which had 56.9%, 56.6% and 55.5% reduction respectively. Also, there were no significant different at P≤0.05 between

applications of carbendazim with the untreated plots with both recording the highest levels of thrips infestation in both locations (Table 4.3).

Table 4.3: Mean ±SE of Thrips infestation on tomatoes sprayed with various concentrations of thyme oil in Kimbimbi and Kagio field sites

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	4.3 ± 0.41^{a}	-	4.0 ± 0.24^{a}	-
Thyme oil 0.5%	2.1 ± 0.24^{b}	51.2	1.7 ± 0.05^{b}	56.9
Thyme oil 1.0%	2.0 ± 0.27^{b}	54.4	1.7 ± 0.18^{b}	56.6
Thyme oil 1.5%	2.3 ± 0.19^{b}	47.0	1.8 ± 0.25^{b}	55.5
Lambda-cyhalothrin	2.0 ± 0.10^{b}	53.7	1.3 ± 0.23^{b}	68.6
Carbendazim	4.4 ± 0.35^{a}	-	3.7 ± 0.17^{a}	7.3
P-Value	<0.001		<0.001	
LSD	0.887		0.551	
SED	0.416		0.259	

Means followed with the same letter along the columns are not significantly different according to Fisher's Protected LSD test at $P \leq 0.05$.

4.4.5 Efficacy of thyme oil on whiteflies infestation on tomatoes

The application of thyme oil at the three rates caused a significant ($P<0.05$) reduction in the whiteflies population compared to the untreated plots and the carbendazim treated plots in both Kimbimbi and Kagio field sites (Table 4.4). Application of thyme oil at the rate of 1.5% spray solution resulted in the highest reduction of whiteflies population of 66.2% in Kimbimbi. This reduction was not significantly different from thyme oil at 0.5%, thyme oil at 1% and lambda-cyhalothrin treatment that caused 65.8%, 61.5% and 63.8% reduction respectively).

In Kagio, the application of the insecticide (lambda-cyhalothrin) caused the highest reduction in whiteflies population on tomato at 58.6% reduction (Table 4.4). This reduction was not significantly (P≤0.05) different from application of thyme oil at 1% and 1.5% which had a reduction of 44.1% and 43.1% respectively. However, the application of lambda-cyhalothrin treatment was differed significantly from application of thyme oil at the rate of 0.5% which resulted in a reduction 38.6%.

Table 4.4: Mean ±SE of whitefly infestation on tomatoes sprayed with various concentrations of thyme oil in Kimbimbi and Kagio field sites

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	28.8 ± 3.65^{a}	-	31.4 ± 1.79^{a}	-
Thyme oil 0.5%	9.9 ± 0.87^{c}	65.8	19.3 ± 2.16^{b}	38.6
Thyme oil 1.0%	11.1 ± 0.42^{c}	61.5	17.6 ± 1.56^{bc}	44.1
Thyme oil 1.5%	9.8 ± 1.02^{c}	66.18	18.5 ± 1.76^{bc}	41.3
Lambda-cyhalothrin.	10.4 ± 0.56^{c}	63.8	13.0 ± 1.26^{c}	58.6
Carbendazim.	21.1 ± 0.79^{b}	27.0	26.6 ± 1.95^{a}	15.5
P-Value	<0.001		<0.001	
LSD	4.902		5.542	
SED	2.300		2.600	

Means followed with the same letter along the columns are not significantly different according to Fisher's Protected LSD test at P≤0.05.

In both locations, the lowest reduction in whitefly population was in the carbendazim treated plots that recorded 27.0% and 15.5% reduction in Kimbimbi and Kagio, respectively. This reduction however, was significantly

P<0.05 different from the untreated plots in Kimbimbi but was not in Kagio (Table 4.4).

4.4.6 Effect of application of thyme oil on tomato yield

The three rates of thyme oil significantly at P≤0.05 increased the tomatoes yield as compared to the untreated plots in both Kimbimbi and Kagio field sites (Figure 4.2). Application of thyme oil at the rate 0.5%, 1% and 1.5% increased the yield of tomatoes by 29.8%, 59.6% and 87.2% respectively compared to the untreated plots in Kagio and 22.6%, 58.5% and 71.7%, respectively in Kimbimbi (Table 4.5).

At both locations, the three thyme oil rates differed significantly as well as the lambda-cyhalothrin treated plots and the carbendazim treated plots. The application of thyme oil at 1.5% spray solution was not significantly (P≤0.05) different from the insecticide and fungicide treated plots in increasing yield in Kagio and Kimbimbi in field sites (Figure 4.2).

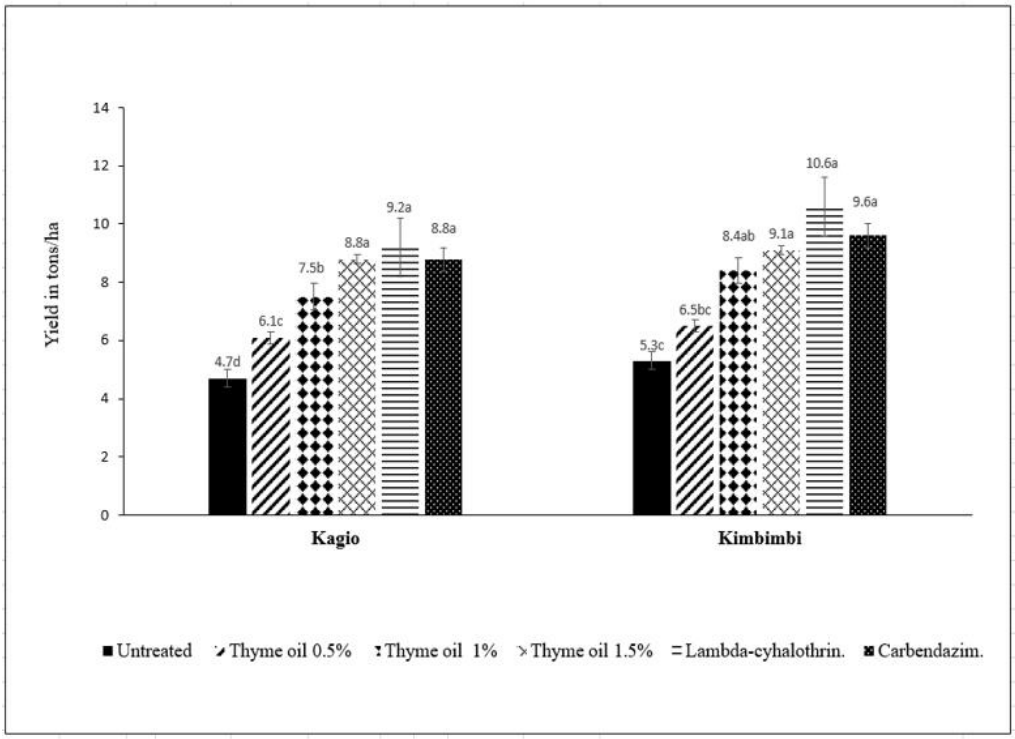

Figure 4.2: Mean marketable yield of tomato per treatment in tons/ha at 16 Weeks after transplanting sprayed with various concentrations of thyme oil on tomato yield in Kimbimbi and Kagio

Means followed by the same letter are not significantly different according to Fisher's Protected LSD test at $p \leq 0.05$

The lambda-cyhalothrin treated plots had the highest yield increase that represented a 95.7% in Kagio and a 100.0% increase in yield compared to the untreated plots (Table 4.5). Among the three thyme oil rates, plots treated with the 0.5% rate had the lowest increase in yield with an increase of 29.8% in Kagio and 22.6% in Kimbimbi. The application of thyme oil at 1.5% resulted in the highest increase in yield among the three rates of thyme oil treatments with an increase of 87.2% in Kagio and 71.7% in Kimbimbi.

Table 4.5: Percent yield increase in tomatoes at 16 weeks after transplanting sprayed with various concentrations of thyme oil in Kimbimbi and Kagio field sites

Treatments	Kagio	Kimbimbi
Untreated control	-	-
Thyme oil 0.5%	29.8	22.6
Thyme oil 1.0%	59.6	58.5
Thyme oil 1.5%	87.2	71.7
Lambda-cyhalothrin.	95.7	100.0
Carbendazim.	87.2	81.1

4.2 Effect of soil improvers on rhizosphere microbial dynamics and yield of tomato

4.2.1 Effect of soil improver on rhizosphere microbial population

In the all the plots the microbial populations was high in comparison with the baseline study that was done at the start of the experiments in both locations as shown in (Table 4.6) and (Table 4.7). Among the three rates of the soil improver, the application of the soil improver at 400kg/ha had the highest increase the number of active fungi of 575.5% than the other treatments in Kagio (Table 4.6). The farm yard manure treatment that had an increase of 454.7% followed closely. The lowest increase was in the untreated plots that had an increase of 67.9% Table 4.6. In Kimbimbi, application of manure at five tons per hectare resulted in the highest increase of active fungi of 359.2% (Table 4.7).

Table 4.6: Microbial populations of Fungi as influenced by application of different rates of the soil improver at end of the trial period in µg per gram of dry soil in rhizosphere of tomato in Kagio as compared with Baseline population

Treatments	Total Fungi	% increase	Active Fungi	% increase
Control (-ve)	118.0	51.3	89.0	67.9
Soil improver (200kg/ha)	254.0	225.6	187.4	253.6
Soil improver (300kg/ha)	301.0	285.9	275.0	418.9
Soil improver (400kg/ha)	356.0	356.4	358.0	575.5
Mijingu Phosphate Rock (250kg/ha)	312.0	300.0	200.0	277.4
Farm Yard Manure (5tons/ha)	387.0	396.2	294.0	454.7
Baseline (at the beginning)	**78**	**-**	**53**	**-**

The application of the soil improver at the rate of 400kg/ha followed closely with an increase of 292.2%. The application of the soil improver at the rate of 200Kg/Ha had the lowest increase of 65.6% in active fungi population of (Table 4.7). The application of Minjingu Phosphate Rock led to an increase in active fungi of 277.4% in Kagio and 151.6% in Kimbimbi. Among the three rates of the soil improver, the 200Kg/ Ha rate resulted in the lowest increase in the microbial population of active fungi in both locations with an increase 67.9% in Kagio and 65.6% in Kimbimbi.

Table 4.7: Microbial populations of fungi as influenced by application of different rates of the soil improver at the beginning (Baseline) and at end of

the trial period in µg per gram of dry soil in rhizosphere of tomato in Kimbimbi

Treatments	Total Fungi	% increase	Active Fungi	% increase
Control (-ve)	253.0	155.6	176.0	175.0
Soil improver (200kg/ha)	288.0	190.9	106.0	65.6
Soil improver (300kg/ha)	265.0	167.7	114.0	78.1
Soil improver (400kg/ha)	355.0	258.6	251.0	292.2
Mijingu Phosphate Rock (250kg/ha)	212.0	114.1	161.0	151.6
Farm Yard Manure (5tons/ha)	487.0	391.9	294.0	359.1
Baseline (at the beginning)	**99**	**-**	**64**	**-**

4.2.2 Effect of soil improver on fusarium wilt incidence in tomatoes

There was no significant difference at P≤0.05 among treatments in suppressing the fusarium wilt incidence in both Kimbimbi and Kagio as shown in (Table 4.8). The application of the soil improver at the rate 300kg/ha recorded the lowest incidence of Fusarium wilt in Kimbimbi which represented a reduction of 24.8%. In Kagio, the application of the soil improver at the rate of 400kg/ha resulted in the lowest disease index with a Mean ± SE disease index of 5.9 ± 0.86. This represented 55.4% reduction compared to the untreated control.

Table 4.8: Mean ±SE incidence of fusarium wilt infection in tomato plots treated with different rates of the soil improvers in Kimbimbi and Kagio field sites

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	11.1 ± 1.71	0.0	11.4 ± 2.24	-

Soil improver (200 kg/ha)	10.1 ± 1.23	8.9	10.6 ± 0.70	23.3
Soil improver (300 kg/ha)	8.4 ± 0.62	24.8	11.3 ± 0.82	1.0
Soil improver (400 kg/ha)	9.8 ± 1.98	12.2	5.9 ± 0.86	55.4
Minjingu Phosphate Rock (250kg/ha)	9.3 ± 1.77	16.9	8.6 ± 0.23	35.6
Farm Yard Manure (5tons/ha)	11.8 ± 0.78	-	11.1 ± 1.70	8.4
P-Value	NS		NS	

4.2.3 Effect of soil improver on tomato yield

The three rates of the soil improver at 200kg/ha, 300kg/ha and 400kg/ha significantly $P \leq 0.05$ increased the tomatoes yield compared to the untreated control in both Kimbimbi and Kagio (Figure 4.3). This represnted an increase of 29.2%, 37.5% and 20.8% in Kagio respectively and 141.7%, 125.0% and 125.0% increase in Kimbimbi respectively as in (Table 4.9). The application of the soil improver at 400Kg/Ha was not differ significantly from the application of Minjingu Phosphate Rock at 250kg/ha and application of manure at 5tons/Ha in increasing yield of tomato in both Kimbimbi and Kagio (Figure 4.3).

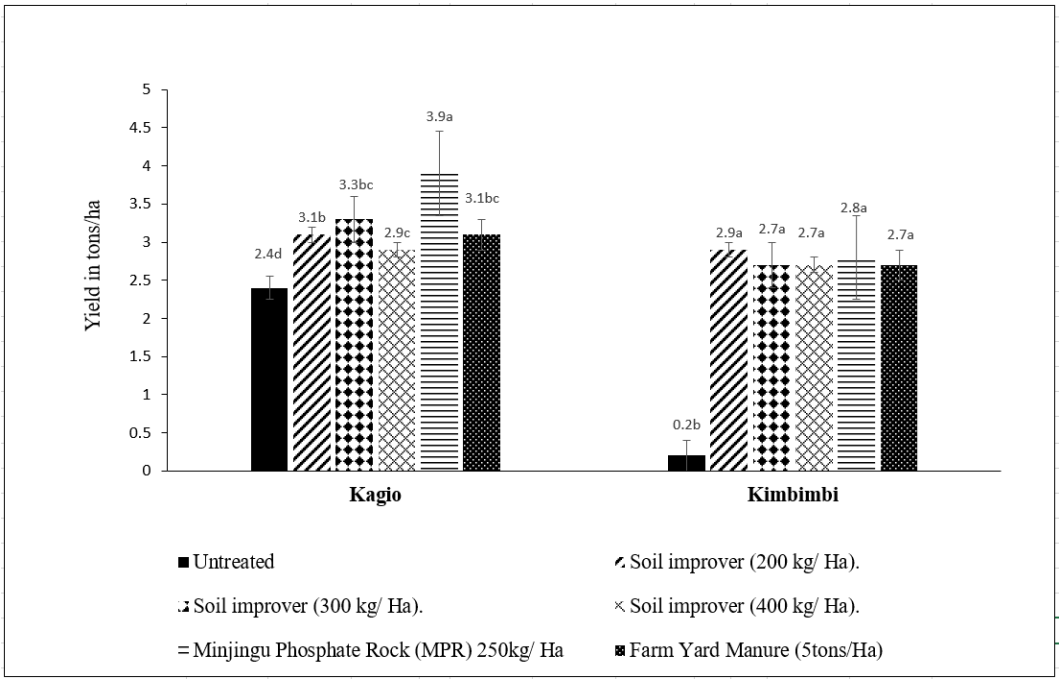

Figure 4.3: Mean marketable yield of tomato per treatment in tons/ha in tomatoes at 16 weeks after transplanting as influenced by application of different rates of the soil improver tomatoes yield in Kimbimbi and Kagio

Means followed by the same letter are not significantly different according to Fisher's Protected LSD test at $p \leq 0.05$

Table 4.9: Percent tomato yield increase at 16 weeks after transplanting as influenced by application of different rates of the soil improver in Kagio and Kimbimbi filed sites

Treatments	Kagio (%)	Kimbimbi (%)
Untreated	-	-
Soil improver (200 kg/ Ha).	29.2	141.7
Soil improver (300 kg/ Ha).	37.5	125.0
Soil improver (400 kg/ Ha).	20.8	125.0
Minjingu Phosphate Rock 250kg/Ha	62.5	133.3
Farmyard Manure (5tons/Ha)	29.2	125.0

4.3 Effect of bio-stimulants on fusarium wilt and root knot nematodes in tomatoes

4.3.1 Effect of bio-stimulants on fusarium wilt incidence in tomatoes

There were significant ($P \leq 0.05$) differences in the effect of *T. harzianum* in reducing the fusarium wilt incidence in tomato compared to the untreated plots in the first and second season (Table 4.10). Plots treated with *T. harzianum* had a reduction of 39.6% fusarium wilt incidence in the first season and 48.6% in the second season. During the first season, there were no significant ($P \leq 0.05$) differences in the effect of *T. harzianum* in suppressing the fusarium wilt incidence in tomatoes compared to the plots treated with seaweed for root stimulation, seaweed plus amino acids and peptides, seaweed plus humic acids and amino acids only.

Table 4.10: Mean ±SE levels of Fusarium wilt incidence index in tomatoes as influenced by application of various bio-stimulants in season one and season two

Treatments	Season 1	% Reduction	Season 2	% Reduction
Untreated	10.8 ± 0.30 [a]	-	12.3 ± 0.97 [a]	-
T. harzianum	6.5 ± 1.20 [b]	39.6	6.3 ± 0.70 [b]	48.6
Seaweed	8.9 ± 0.79 [ab]	18.1	10.2 ± 0.29 [a]	17.2
Seaweed + amino acids + peptides	8.7 ± 0.57 [ab]	19.3	9.8 ± 0.56 [a]	20.1
Seaweed + Humic acids (AA)	6.6 ± 1.00 [b]	38.9	11.2 ± 1.44 [a]	8.8
Amino acids (AA)	8.4 ± 0.66 [ab]	22.8	11.9 ± 1.36 [a]	2.9
P-Value	0.024		0.008	
LSD	2.545		2.994	
SED	1.194		1.405	

Means followed by the same letter along the columns are not significantly different according to Fisher's Protected LSD test at P≤0.05.

During the second season, there were significant differences in the effect of treatments on fusarium wilt. *T. harzianum* highly supressed fusarium wilt in tomatoes compared to the other treatments (Table 4.10). In season two, the treatments with seaweed for root stimulation, seaweed plus amino acids and peptides, seaweed plus humic acids and amino acids only did not differ significantly from one another as well as the untreated control. As well, the

untreated control recorded the highest fusarium wilt incidence throughout the trial period both in season one and two.

4.3.2 Effect of bio-stimulants on Nematode infection on tomatoes

The nematode statistical analysis results revealed a low infection of the root knot nematode (Figure 4.4). There were significant $P<0.05$ differences in the effect of *Trichoderma harzianum* in reducing the galling index on tomato compared to the untreated control in the first and second season (Table 4.11) with a 60.9% and 48.8% reduction respectively. This represented the highest reduction in the galling index on tomato in all treatments.

The treatment with Seaweed plus amino acids plus peptides and the plots treated with amino acids only were also significantly ($P\leq0.05$) different in reducing (18.4% and 43.8% reduction respectively) the galling index compared to the untreated plots in the first season. The galling index on tomato in treatments with seaweed for root stimulation and Seaweed plus Humic acids (AA) were not significant at $P\leq0.05$ different from the untreated plots in the first season (Table 4.11).

During the second season, apart from the *Trichoderma harzianum* treatment, the rest of the treatments did not differ significantly at $P\leq0.05$ from each other as well as the untreated plots. Although, the nematode infestation was generally higher in second season compared to first season (Table 4.11).

Field: **KU Greenhouse** Top Soil

To maintain the correct history ensure that the next sample sent from this Field is labelled: Kagio-KU Greenhouse

History (Last 3 analysis)

Parameter	Unit	Result	Guide Low	Guide High	Low	Optimum	High	Symbol	Current				Method
Saprofagic	100 ml	2040		> 1000				Sap	2040				
Meloidogyne	100 ml	10.0		< 30.0				Mel	10.0				
Pratylenchus	100 ml	20.0		< 50.0				Pra	20.0				
Radopholus	100 ml	0.00		< 50.0				Rad	0.00				
Tylenchus	100 ml	40.0		< 100				Tyl	40.0				
Tylenchorhynchus	100 ml	40.0		< 50.0				Tyle	40.0				
Helicotylenchus	100 ml	0.00		< 100				Hel	0.00				
Scutellonema	100 ml	0.00		< 100				Scu	0.00				
Aphelenchus	100 ml	0.00		< 1000				Aps	0.00				
Xiphinema	100 ml	0.00		< 200				Xip	0.00				
Longidorous	100 ml	0.00		< 40.0				Lo	0.00				
Hemicyclophora	100 ml	0.00		< 100				Hem	0.00				
Criconema	100 ml	0.00		< 100				Cri	0.00				

Figure 4.4: Nematode analysis reports for greenhouse at Kenyatta University

Table 4.11: Mean ±SE levels of Nematode infestation in tomatoes as influenced by application of various bio-stimulants in season one and season two

Treatments	Season 1	% Reduction	Season 2	% Reduction
Untreated	2.8 ± 0.37^{a}	-	3.6 ± 0.47^{a}	-
T. harzianum	1.1 ± 0.13^{c}	60.9	1.9 ± 0.16^{b}	48.8
Seaweed	2.4 ± 0.16^{ab}	15.1	3.3 ± 0.22^{a}	9.4
Seaweed + AA + peptides	2.3 ± 0.13^{b}	18.4	3.3 ± 0.34^{a}	9.4
Seaweed + (HA)	2.3 ± 0.25^{ab}	17.8	3.2 ± 0.40^{a}	12.1
Amino acids	1.6 ± 0.11^{c}	43.8	3.2 ± 0.45^{a}	11.1
P-Value	<.001		0.024	
LSD	0.5036		0.977	
SED	0.2363		0.458	

Means followed by the same letter along the columns are not significantly different according to Fisher's Protected LSD test at P≤0.05. AA-Amino Acids, HA- Humic Acids

4.3.3 Effect of bio-stimulants on tomatoes yield

The treatment with *T. harzianum* as well as treatments with seaweed for root stimulation, seaweed plus amino acids and peptides, seaweed plus humic acids and amino acids only increased yield of tomatoes significantly at P≤0.05 compared to the untreated plots in the first and second season (Figure 4.5). Furthermore, the treatment with a combination seaweed and humic acids as well as the treatment with seaweeds, amino acids and peptides, recorded the highest yields in both the first and second season. In (Table 4.12) *T. harzianum* treatment recorded the lowest percentage yield of 11.6% and 32.4% in season one and two, respectively.

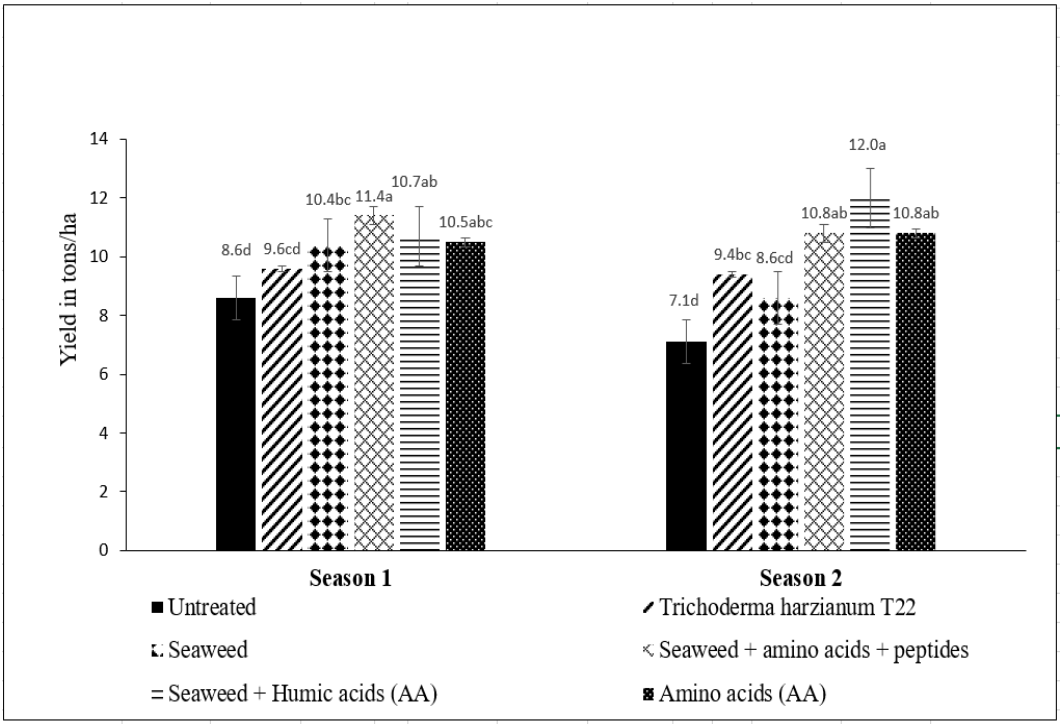

Figure 4.5: Mean marketable yield of tomato per treatment in tons/ha at16 weeks after transplanting as influenced by application of various bio-stimulants in season one and two

Means followed by the same letter are not significantly different according to Fisher's Protected LSD test at p≤0.05

Table 4.12: Percent yield increase in tomatoes at 16 weeks after transplanting as influenced by application of various bio-stimulants in season one and season two

Treatments	Season 1	Season 2
Untreated	-	-
T. harzianum	11.6	32.4
Seaweed for root stimulation	20.9	21.1
Seaweed + amino acids + peptides	32.6	52.1
Seaweed + Humic acids (AA)	24.4	69.0
Amino acids (AA)	22.1	52.1

4.4 Effect of integrating plant essential oils, soil improvers and bio-stimulants on management of soil borne diseases, arthropod pests and tomatoes yield

4.4.1 Effect of integrating thyme oil, soil improvers and bio-stimulants on root knot nematode infection

The nematode analysis results showed a low infection of the root knot nematode in both Kagio and Kimbimbi field sites (Figure 4.6 and Figure 4.7). The integration of thyme oil, the soil improver, *T. harzianum* and bio-stimulants had a significant $P<0.05$ effect on the reduction of root knot nematode infection in both Kagio (35.4 to 60.1% reduction) and Kimbimbi (27.6 to 40.8% reduction) compared to the untreated plots (Table 4.13).

The integrated options had a mean ±SE Galling Index in tomato of between 1.71 ± 0.11 and 2.20 ± 0.25 in Kimbimbi and a mean ±SE Galling Index in tomato of between 1.11 ± 0.09 and1.80 ± 0.05 in Kagio as shown in (Table 4.13). The integrated treatments recorded low galling index on tomatoes, which were comparable to the chemical treated plots that had a mean ±SE Galling Index in tomato of 2.00 ± 0.32 in Kimbimbi and 1.14 ± 0.05 in Kagio.

The highest reduction in Kimbimbi in the galling index was in the integrated treatment with thyme oil, soil improver, *T. harzianum* and humic acids with a reduction of 43.6% (Table 4.13). However, there were no significant differences between this integrated option and the other integrated options as well as the chemically treated plots in Kimbimbi. On the other hand, in Kagio the highest reduction was in the integrated treatment with thyme oil, soil improver, *T. harzanium* and amino acids that had 60.1% reduction. This reduction was not significantly ($P \leq 0.05$) different from the chemically treated plots that had 59.2% reduction.

The chemically treated plots had a significantly lower galling index compared to the untreated plots (Table 4.13). The untreated plots had the highest galling index in both Kimbimbi and Kagio with a mean ±SE Galling Index in tomato of 3.04 ± 0.20 and 2.79 ± 0.19 respectively (Table 4.13). There were generally no significant $P>0.05$ differences among the various integrated options in their effect on the galling index in tomato.

Field: Kimbimbi- Sample A — Top Soil

To maintain the correct history ensure that the next sample sent from this Field is labelled: Kimbimbi-Sample A

History (Last 3 analysis)

Parameter	Unit	Result	Guide Low	Guide High	Low	Optimum	High	Symbol	Current				Method
Saprofagic	100 ml	880		> 1000				Sap	880				
Meloidogyne	100 ml	25.0		< 30.0				Mel	25.0				
Pratylenchus	100 ml	0.00		< 50.0				Pra	0.00				
Radopholus	100 ml	0.00		< 50.0				Rad	0.00				
Tylenchus	100 ml	0.00		< 100				Tyl	0.00				
Tylenchorhynchus	100 ml	0.00		< 50.0				Tyle	0.00				
Helicotylenchus	100 ml	0.00		< 100				Hel	0.00				
Scutellonema	100 ml	0.00		< 100				Scu	0.00				
Aphelenchus	100 ml	0.00		< 1000				Aps	0.00				
Xiphinema	100 ml	0.00		< 200				Xip	0.00				
Longidorous	100 ml	0.00		< 40.0				Lo	0.00				
Hemicyclophora	100 ml	0.00		< 100				Hem	0.00				
Criconema	100 ml	0.00		< 100				Cri	0.00				

Figure 4.6: Nematode analysis report Kimbimbi

Customer:	Geoffrey Ongoya Wafula	Crop:	Tomatoes	Date Received:	15-Jul-19
Address:		Crop Stage:		Analysis Date:	18-Jul-19
Farm Name:	KAGIO	Comments:		Report Date:	22-July-19
Contact Person:	Geoffrey Ongoya	Condition:	Moist	Sample ID:	CK107NEM0019

Field: Kagio -Sample B — Top Soil

To maintain the correct history ensure that the next sample sent from this Field is labelled: Kagio Sample B

History (Last 3 analysis)

Parameter	Unit	Result	Guide Low	Guide High	Low	Optimum	High	Symbol	Current				Method
Saprofagic	100 ml	1040		> 1000				Sap	1040				
Meloidogyne	100 ml	20.0		< 30.0				Mel	20.0				
Pratylenchus	100 ml	20.0		< 50.0				Pra	20.0				
Radopholus	100 ml	0.00		< 50.0				Rad	0.00				
Tylenchus	100 ml	40.0		< 100				Tyl	40.0				
Tylenchorhynchus	100 ml	40.0		< 50.0				Tyle	40.0				
Helicotylenchus	100 ml	0.00		< 100				Hel	0.00				
Scutellonema	100 ml	0.00		< 100				Scu	0.00				
Aphelenchus	100 ml	0.00		< 1000				Aps	0.00				
Xiphinema	100 ml	0.00		< 200				Xip	0.00				
Longidorous	100 ml	0.00		< 40.0				Lo	0.00				
Hemicyclophora	100 ml	0.00		< 100				Hem	0.00				
Criconema	100 ml	0.00		< 100				Cri	0.00				

Figure 4.7: Nematode analysis report Kagio site

Table 4.13: Mean ±SE levels of Nematode infestation in tomatoes as influenced by application of various integrated options in Kimbimbi and Kagio

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	3.04 ± 0.20^{a}	-	2.79 ± 0.19^{a}	-
TO+SI+TH +SW+AA	2.20 ± 0.25^{b}	27.6	1.11 ± 0.09^{c}	60.1
TO+SI+TH +SW+HA	1.71 ± 0.11^{b}	43.6	1.26 ± 0.12^{c}	54.7
TO+SI+TH +SW+PEPTIDES	2.03 ± 0.22^{b}	33.1	1.34 ± 0.26^{bc}	52.1
TO+SI+TH +SW+HA+AA	1.88 ± 0.21^{b}	38.3	1.36 ± 0.26^{bc}	51.1
TO+SI+TH+SW +AA+PEPTIDES	1.85 ± 0.21^{b}	39.2	1.13 ± 0.15^{c}	59.6
TO+SI+TH+SW +HA+PEPTIDES	2.18 ± 0.18^{b}	28.1	1.31 ± 0.21^{c}	52.9
TO+SI+TH+SW +AA+HA+PEPTIDES	1.80 ± 0.16^{b}	40.8	1.80 ± 0.05^{b}	35.4
Chemical treatment	2.00 ± 0.32^{b}	34.2	1.14 ± 0.05^{c}	59.2
P-Value	0.014		<.001	
LSD	0.6525		0.4731	
SED	0.3162		0.2292	

Means followed by the same letter along the columns are not significantly different according to Fisher's Protected LSD test at $p \leq 0.05$. TO- Thyme oil; SI- Soil improver; TH- *Trichoderma harzianum*, SW- Seaweeds; AA-Amino acids; HA-Humic acids

4.4.2 Effect of integrating plant essential oils, soil improvers and bio-stimulants on incidence of fusarium wilt

The integration of thyme oil, the soil improver, *T. harzianum* and bio-stimulants had a significant (P≤0.05) effect on the incidence of fusarium wilt in tomato compared to the untreated plots in both Kimbimbi and Kagio field sites (Table 4.14). The highest reduction in the fusarium wilt incidence was in the integrated treatment with thyme oil, soil improver, *T. harzianum*, seaweeds, humic acids and amino acids that had 56.6% reduction in Kimbimbi and 66.9% reduction in Kagio. This reduction was significantly P<001 different from the chemically treated plots that had a reduction of 44.4% in Kimbimbi and 43.3% in Kagio.

The untreated plots had the highest incidence of fusarium wilt throughout the trial period with a mean ± SE fusarium wilt incidence of 3.21 ± 0.38 in Kimbimbi and 3.46 ± 0.38 in Kagio. In addition, the chemical treatment had significantly P<0.001 lower fusarium wilt incidence compared to the untreated plots in Kimbimbi and Kagio field sites (Table 4.14).

There were significant (P≤0.05) differences between the various integrated options in their effect on reduction of fusarium wilt incidence in both locations Table (4.14). The lowest reduction among the integrated options was in the integration option with thyme oil, soil improver, *T. harzianum*, seaweeds and humic acids at 31.4% reduction in Kimbimbi and the integrated option with thyme oil, soil improver, *T. harzianum*, seaweeds and peptides that had a reduction of 27.8% in Kagio.

Table 4.14: Levels of Fusarium wilt incidence index in tomatoes as influenced by application of various integrated options in Kimbimbi and Kagio

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	3.21 ± 0.38 [a]	-	3.46 ± 0.38 [a]	-
TO+SI+TH +SW+AA	1.61 ± 0.12 [bc]	50.0	1.61 ± 0.28 [cd]	53.6
TO+SI+TH +SW+HA	2.20 ± 0.09 [b]	31.4	1.99 ± 0.15 [bc]	42.6
TO+SI+TH +SW+PEPTIDES	1.93 ± 0.18 [bc]	40.0	2.50 ± 0.09 [b]	27.8
TO+SI+TH +SW+HA+AA	1.40 ± 0.18 [c]	56.6	1.15 ± 0.15 [d]	66.9
TO+SI+TH+SW +AA+PEPTIDES	1.64 ± 0.28 [bc]	48.9	1.64 ± 0.24 [cd]	52.6
TO+SI+TH+SW +HA+PEPTIDES	1.82 ± 0.23 [bc]	43.3	2.04 ± 0.42 [bc]	41.2
TO+SI+TH+SW +AA+HA+PEPTIDES	1.57 ± 0.34 [bc]	51.1	1.57 ± 0.24 [cd]	54.6
Chemical treatment	1.79 ± 0.15 [bc]	44.4	1.96 ± 0.34 [bc]	43.3
P-Value	<.001		<.001	
LSD	0.6683		0.7558	
SED	0.3238		0.3662	

Means followed by the same letter along the columns are not significantly different according to Fisher's Protected LSD test at $p \leq 0.05$. TO- Thyme oil; SI- Soil improver; TH- *Trichoderma harzianum*, SW- Seaweeds; AA-Amino acids; HA-Humic acids

4.4.3 Effect of integrating thyme oil, soil improvers and bio-stimulants on early blight infection

The integration of thyme oil, the soil improver, *T. harzianum* and bio-stimulants significantly $P<0.001$ suppressed early blight infection in tomato (Table 4.15) compared to the untreated plots in Kimbimbi and Kagio field sites. The integration of thyme oil, soil improver, *T. harzianum*, seaweeds and humic acids has the highest reduction in early blight infection of 83.4% in Kimbimbi. This was not however, significantly $P>0.05$ different from the other integrated options with a mean ±SE of early blight disease index of between 5.03 ± 0.93 to 7.90 ± 1.25 and the chemically treated plots with a mean ± SE of early blight disease index of 5.94 ± 0.15.

In Kagio, the chemically treated plots had the highest reduction in early blight infection with a reduction of 79.8%. This reduction was not significantly different from the integrated options treatments with a mean ±SE of early blight disease index of between 6.28 ± 0.50 to 7.62 ± 1.86 but was significantly $P<0.001$ different from the untreated plots Table 4.15. The untreated control had the highest early blight disease index throughout the trial period in both Kimbimbi and Kagio with a mean ± SE of early blight disease index of 30.26 ± 2.78 in Kimbimbi and 26.53 ± 3.81 in Kagio (Table 4.15).

Table 4.15: Mean ±SE levels of early blight infection in tomatoes as influenced by application of various integrated options in Kimbimbi and Kagio

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	30.26 ± 2.78 [a]	-	26.53 ± 3.81 [a]	-
TO+SI+TH +SW+AA	6.91 ± 1.58 [b]	77.2	6.58 ± 1.12 [b]	75.2
TO+SI+TH +SW+HA	5.03 ± 0.93 [b]	83.4	7.62 ± 1.86 [b]	71.3
TO+SI+TH +SW+PEPTIDES	6.06 ± 0.99 [b]	80.0	6.50 ± 0.72 [b]	75.5
TO+SI+TH +SW+HA+AA	7.90 ± 1.25 [b]	73.9	7.28 ± 1.10 [b]	72.6
TO+SI+TH+SW +AA+PEPTIDES	5.59 ± 0.96 [b]	81.5	7.02 ± 0.52 [b]	73.5
TO+SI+TH+SW +HA+PEPTIDES	5.66 ± 0.60 [b]	81.3	6.81 ± 1.29 [b]	74.3
TO+SI+TH+SW +AA+HA+PEPTIDES	6.38 ± 0.69 [b]	78.9	6.28 ± 0.50 [b]	76.3
Chemical treatment	5.94 ± 0.15 [b]	80.4	5.37 ± 1.13 [b]	79.8
P-Value	<.001		<.001	
LSD	3.231		4.122	
SED	1.565		1.997	

Means followed by the same letter along the columns are not significantly different according to Fisher's Protected LSD test at $p \leq 0.05$. TO- Thyme oil; SI- Soil improver; TH- *Trichoderma harzianum*, SW- Seaweeds; AA-Amino acids; HA-Humic acids

4.4.4 Effect of integrating thyme oil, soil improvers and bio-stimulants on thrips infestation

From the study the integration of thyme oil, soil improver, *T. harzianum* and bio-stimulants effectively reduced the thrips population compared to the untreated plots in Kimbimbi and Kagio field sites (Table 4.16). The integrated application of thyme oil, soil improver, *T. harzianum*, seaweeds and amino acids had the highest reduction in thrips population of 53.5% in Kimbimbi.

On the other hand, the integrated application of thyme oil, soil improver, *T. harzianum*, seaweeds and humic acids had the highest reduction in thrips population of 66.0% in Kagio (Table 4.16). The chemically treated plots had significantly $P<0.001$ lower thrips population compared to the untreated plots with a mean ± SE of motile thrips per tomato plant of 2.44 ± 0.43 in Kimbimbi and 1.67 ± 0.37 in Kagio representing a reduction of 41.9% and 40.2% respectively.

There were no significant ($P\leq0.05$) differences among the various integrated in Kimbimbi with a mean ± SE of motile thrips per tomato plant among the treatments ranging from 1.95 ± 0.27 to 2.61 ± 0.26 (Table 4.16). Similarly, the various integrated options were not significantly ($P\leq0.05$) different from the chemically treated plots that had a mean ± SE of motile thrips per tomato plant of 2.44 ± 0.43. On the other hand, in Kagio, there were significant $P<0.001$ differences among the various integrated options in reducing thrips populations as well as the chemically treated. The untreated plots recorded

the highest number of thrips per plant in both locations with a mean ± SE of motile thrips per tomato of 4.20 ± 0.34 in Kimbimbi and 2.80 ± 0.38 in Kagio.

Table 4.16: Mean ±SE levels of thrips infestation on tomatoes treated with various integrated options in Kimbimbi and Kagio field sites

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	4.20 ± 0.34^{a}	-	2.80 ± 0.38^{a}	-
TO+SI+TH +SW+AA	2.09 ± 0.24^{b}	50.3	0.95 ± 0.20^{e}	66.0
TO+SI+TH +SW+HA	1.95 ± 0.27^{b}	53.5	1.59 ± 0.20^{cde}	43.3
TO+SI+TH +SW+PEPTIDES	2.27 ± 0.19^{b}	45.9	2.04 ± 0.38^{bc}	27.0
TO+SI+TH +SW+HA+AA	1.98 ± 0.10^{b}	52.8	1.74 ± 0.07^{bc}	37.8
TO+SI+TH+SW +AA+PEPTIDES	2.61 ± 0.26^{b}	37.9	1.59 ± 0.22^{cde}	43.2
TO+SI+TH+SW +HA+PEPTIDES	2.05 ± 0.23^{b}	51.2	0.96 ± 0.26^{de}	65.5
TO+SI+TH+SW +AA+HA+PEPTIDES	2.08 ± 0.14^{b}	50.4	2.34 ± 0.15^{ab}	16.5
Chemical treatment	2.44 ± 0.43^{b}	41.9	1.67 ± 0.37^{bcd}	40.2
P-Value	<.001		<.001	
LSD	0.7312		0.7163	
SED	0.3543		0.3471	

Means followed by the same letter along the columns are not significantly different according to Fisher's Protected LSD test at $p \leq 0.05$. TO- Thyme oil; SI- Soil improver; TH- *Trichoderma harzianum*, SW- Seaweeds; AA-Amino acids; HA-Humic acids

4.4.5 Effect of integrating plant essential oils, soil improvers and bio-stimulants on whitefly infestation

The integration of thyme oil, soil improver, *T. harzianum* and bio-stimulants had a highly significant (P≤0.05) reduction on the whitefly infestation compared to the untreated plots in Kimbimbi field site (Table 4.17). In Kagio, there were no significant (P≤0.05) differences among the various integrated options in their effect on reducing the whitefly population compared to the untreated plots. The chemically treated plots recorded the highest reduction of whitefly in both locations with a reduction of 47.6% in Kimbimbi and 47.1% in Kagio.

This reduction was highly significant from the untreated plots in Kimbimbi but was not significantly P>0.05 different from the untreated plots in Kagio (Table 4.17). Similarly, there were no significant (P≤0.05) differences between the various integrated options and the chemically treated plots in their effect of reducing whitefly infestation on tomato in Kimbimbi and Kagio. The untreated plots recorded the highest number of whitefly infestation in both locations with mean ± SE of whitefly per tomato leaf of 37.72 ± 2.49 in Kimbimbi and 38.00 ± 6.49. Generally, integrated options had a reduction of a mean ± SE of whitefly per tomato leaf of 19.89 ± 2.59 to 23.06 ± 2.64 in Kimbimbi and 21.3 ± 3.83 to 31.7 ± 1.51 in Kagio.

Table 4.17: Mean ±SE levels of whitefly infestation on tomatoes treated with integrated options in Kimbimbi and Kagio field sites

Treatments	Kimbimbi	% Reduction	Kagio	% Reduction
Untreated	37.72 ± 2.49 a	-	38.00 ± 6.49	-
TO+SI+TH +SW+AA	23.06 ± 2.64 b	38.9	31.7 ± 1.51	16.6
TO+SI+TH +SW+HA	21.34 ± 1.15 b	43.4	31.3 ± 5.71	17.6
TO+SI+TH +SW+PEPTIDES	20.97 ± 2.24 b	44.4	27.3 ± 4.87	28.2
TO+SI+TH +SW+HA+AA	19.89 ± 2.59 b	47.3	29.3 ± 12.00	22.9
TO+SI+TH+SW +AA+PEPTIDES	21.37 ± 2.49 b	43.3	23.9 ± 6.01	37.1
TO+SI+TH+SW +HA+PEPTIDES	21.21 ± 2.72 b	43.8	22.9 ± 7.83	39.7
TO+SI+TH+SW +AA+HA+PEPTIDES	20.88 ± 2.30 b	44.6	21.3 ± 3.83	43.9
Chemical treatment	19.76 ± 2.71 b	47.6	20.1 ± 3.69	47.1
P-Value	<.001		NS	
LSD	5.180			
SED	2.510			

Means followed by the same letter along the columns are not significantly different according to Fisher's Protected LSD test at p≤0.05. TO- Thyme oil; SI- Soil improver; TH- *Trichoderma harzianum*, SW- Seaweeds; AA-Amino acids; HA-Humic acids

4.4.6 Effect of integrating plant essential oils, soil improvers and bio-stimulants on marketable yield of tomatoes

The integration of thyme oil, soil improver, *T. harzianum* and bio-stimulants significantly ($P \leq 0.05$) increased the yield of tomato compared to the untreated plots in Kimbimbi and Kagio field sites (Figure 4.8). The various integrated options were not significantly ($P \leq 0.05$) different in their effect of increasing yield in tomato in Kagio (Figure 4.8). The highest yield increase among the integrated options treatments in Kagio was in the treatment with the integration of Thyme oil, Soil Improver, *T. harzianum,* Seaweeds and Peptides that had a 126.5% increase compared to the untreated plots (Table 4.18). The lowest increase in yield among the integrated options treatments was in the treatment with integration of thyme oil, Soil Improver, *T. harzianum*, Seaweeds and Amino Acids with 55.1% increase in Kimbimbi.

In Kimbimbi, the integration of thyme oil, Soil Improver, *Trichoderma harzianum*, Seaweeds, Humic Acids and Peptides had the highest increase in yield of 94.2 % compared to the untreated control (Table 4.18) that was significantly $P<0.05$ different from the other integrated options as well as the chemically treated plots. The lowest increase in yield among the integrated options treatments was in the treatment with integration of Thyme oil, Soil Improver, *Trichoderma harzianum,* Seaweeds and Amino Acids with 76.0% increase in Kagio (Table 4.18). The various integrated options did not differ significantly at $P \leq 0.05$ from the chemically treated plots in their effect of increasing yield in Kagio (Figure 4.8).

The chemically treated plots had the lowest yield increase compared to the integrated option treatments in both Kimbimbi and Kagio with a yield increase of 42.7% and 74% respectively (Table 4.18). The untreated plots had the lowest yield in both locations with a yield of 5 tons per hectare in Kagio and 5.6 tons per hectare in Kimbimbi (Figure 4.8).

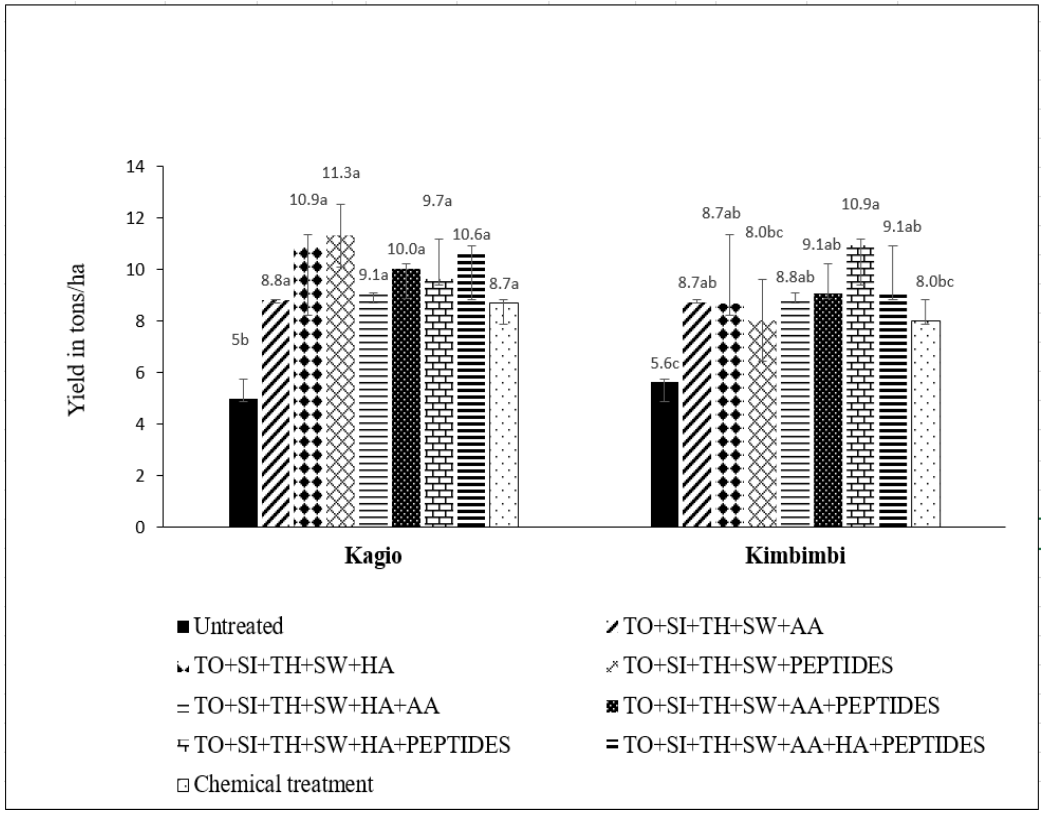

Figure 4.8: Mean marketable yield of tomato per treatment in tons/ha at 16 Weeks after transplanting treated with various integrated options in Kimbimbi and Kagio

Means followed by the same letter are not significantly different according to Fisher's Protected LSD test at $p \leq 0.05$. TO- Thyme oil; SI-Soil improver; TH-*Trichoderma harzianum*, SW- Seaweeds; AA-Amino acids; HA-Humic acids

Table 4.18: Percent yield increase in tomatoes treated with integrated options in Kimbimbi and Kagio field sites

Treatments	Kimbimbi	Kagio
Untreated	-	-
Thyme oil + Soil Improver + *T. harzianum* + Seaweeds + Amino Acids	55.1	76.0
Thyme oil + Soil Improver + *T. harzianum* + Seaweeds + Humic Acids	54.2	118.0
Thyme oil + Soil Improver + *T. harzianum* + Seaweeds + Peptides	42.7	126.5
Thyme oil + Soil Improver + *T. harzianum* + Seaweeds + Humic Acids + Amino Acids	55.6	81.0
Thyme oil + Soil Improver + *T. harzianum* + Seaweeds + Amino Acids + Peptides	61.3	100.5
Thyme oil + Soil Improver + *T. harzianum* + Seaweeds + Humic Acids +Peptides	94.2	93.0
Thyme oil + Soil Improver + *T. harzianum* + Seaweeds + Amino Acids + Humic Acids + Peptides	62.2	112.0
Chemical treatment	42.7	74.0

4.6.7 Association between variables; Correlation analysis between variables

The combined Pearson's correlation analysis for the various parameters assessed showed that early blight infection, fusarium wilt incidence, thrips infestation and whitefly infestation had a negative significant $P \leq 0.05$ relationship with yield in Kimbimbi (Table 4.19). On the other hand, the nematode infection had a negative but non-significant at $P \leq 0.05$ relationship with yield in Kimbimbi.

In Kagio, the combined Pearson's correlation analysis for the various parameters assessed showed that early blight infection and nematode infection had a negative significant P≤0.05 relationship with yield (Table 4.20). On the other hand, fusarium wilt incidence, thrips infestation and whitefly infestation had a negative but non-significant at P≤0.05 relationship with yield. The relationship between early blight infestation, fusarium wilt incidence, nematode infection, thrips infestation and whitefly infestation was positive in both locations (Table 4.19 and Table 4.20).

Table 4.19: Combined Pearson's correlation between variables at Kimbimbi field site in Kirinyaga

Early blight	-					
Fusarium	0.862*	-				
Nematodes	0.912*	0.794*	-			
Thrips	0.936*	0.848*	0.867*	-		
Whiteflies	0.978*	0.898*	0.923*	0.930*	-	
Yield	-0.796*	-0.734*	**-0.643NS**	-0.804*	-0.757*	-
	Early blight	Fusarium Wilt	Nematodes	Thrips	Whiteflies	Yield

*-Significant, NS-Not Significant

Table 4.20: Combined Pearson's correlation between variables at Kagio field site in Kirinyaga

Early blight	-					
Fusarium	0.807*	-				
Nematodes	0.916*	0.725*	-			
Thrips	0.648*	**0.558 NS**	0.806*	-		
Whiteflies	0.731*	**0.514 NS**	**0.550 NS**	**0.290 NS**	-	
Yield	-0.848*	**-0.564 NS**	-0.714*	**-0.375 NS**	**-0.569 NS**	-
	Early blight	Fusarium Wilt	Nematodes	Thrips	Whiteflies	Yield

*-Significant, NS-Not Significant

CHAPTER FIVE: DISCUSSION

5.1 Effect of Thyme oil on fusarium wilt, early blight and arthropod pests of tomato

5.1.1 Effect of thyme oil on Fusarium wilt incidence and early blight infection.

Thyme oil was effective in suppression of fusarium wilt and early blight in Kimbimbi and Kagio. The high antifungal activity is because of several modes of action through which thyme oil acts on the pathogens. Jabeur *et al.* (2017) reported ability of thyme oil to target genes involved in fungal development and virulence thereby giving it the antifungal properties. The suppression of fusarium could be attributed to the property of plant essential oils that involve disease suppression (Vishwakarma *et al.*, 2020). The effect of thyme oil on fusarium wilt could also be as a direct effect of the thyme oil on the pathogen but also as a result induced acquired resistance by the tomato plants (Maissa *et al*., 2015).

The above results are consistent with findings by Kalemba *et al*. (2003) that reported anti-microbial properties of plant essential oils. The results also agrees with the findings by Park *et al*. (2017) which showed inhibition of growth of fusarium species by plant essential oils. The results also concides with findings by Torre *et al*. (2016) who also reported the use of plant essential oils for management of fusarium wilt in tomatoes. In their findings, Torres *et al.* (2016) observed that application of plant essential oils on tomato plants dipped

in a conidial suspension of *Fusarium oxysporum* f. sp. Lycopersici inhibited mycelial growth and conidial germination.

The effect of thyme on early blight is consistent with findings≤ by Mona *et al.* (2016) who reported inhibition of growth of *Alternaria solani* that casuses early blight in tomatoes by plant essential oils. Similarly El-Mohamedy *et al*. (2015), reported effective management of early blight through application of thyme oil. The carbendazim treatment had significantly P0.05 lower disease pressure compared to the untreated control. This can be attributed to the fungicidal effect of carbendazim on fusarium wilt and early blight (Deo, 2013).

5.1.2 Efficacy of thyme oil on thrips and whitefly

Thyme oil had a significant effect at P≤0.05 on the population of thrips and whitefly. This results are comparable with of Lattanzio *et al.* (2006), that showed some plant essential oils, for example, salicylates to be feeding barriers to insect pests. Plant flavonoids have also been reported to affect development of some insects (Lattanzio *et al.*, 2006). The effect on thrips agrees with findings by Koschier (2008) that showed plant essential oils effect as a feeding and oviposition deterrent for thrips. The effect of thyme oil on whitefly was significant at P≤0.05 and this are similar with the findings by Aroiee *et al.* (2005) and Hanaa Saleh Hussein (2017) which reported repellent and toxicity effect of plant essential oils on whitefly.

The carbendazim treatment had no significant effect on the population of thrips and whitefly compared to the untreated control which had high thrips and whitefly population. This can be attributed to the lack of insecticidal properties

in the fungicide that allowed the whiteflies and thrips populations to build up. The lambda-cyhalothrin had the lowest population of thrips and whitefly compared to all other treatments. This can be attributed to the insecticidal properties of lambda-cyhalothrin that suppressed the population of thrips and whitefly (Badii *et al.*, 2013).

5.1.3 Effect of thyme on tomato yield

Thyme oil had a positive impact tomatoes yield causing an increase in the quantity of marketable yield. This response can be attributed to the properties of plant essential oils that promotes plant growth, which eventually results in increased yields (El-Mohamedy *et al.*, 2015; Vishwakarma *et al*., 2020). The untreated control had the lowest yield compared to other treatments. This can be attributed to the high disease and insect pest pressure, which negatively affected the yields.

Also, the application of plant essential oils resulted in a significant effect at $P \leq 0.05$ in the increase in tomatoes yield. This could be attributed to the effective suppression of fungal diseases and insect pests, which allowed the tomato crop to grow well and hence, have higher yields. The yield in Kagio were slightly higher than the yields in Kimbimbi. This can be attributed to the differences in the soil types and climate in the two areas.

5.2 Effect of soil improvers on soil rhizosphere microbial population and tomatoes yield

Application of the soil improver had a positive effect on the microbial population of fungi resulting in an increase of active fungi at the soil rhizosphere of tomato plants. The application of the soil improver resulted in lower incidences of fusarium wilt in both Kimbimbi and Kagio. The lack of significant effect at $P \leq 0.05$ in the suppression of fusarium wilt can be attributed to the short duration of the experimental trial. For strong effect on the pathogenic populations, continuous application of the soil improver over a longer period is required.

These results are consistent with findings by Mokhtar *et al*. (2014) that reported lower pathogenic fungi in response to an increase in the population of beneficial fungi. The increase in yield in the treatments with the soil improver can be attributed to the improved soil environment due to increased microbial activity and the nutrients made available through application of the soil improver. The lower yield response at the lower rates of the soil improver shows that the crop yield responds positively to an increase in the quantity of the soil improver applied. The application of manure and minjungu phosphate rock also resulted in an increase in the yield of tomato.

However, the fusarium wilt incidences were higher compared to the soil improver treatments but lower than the untreated control. These results agree with findings by Panth *et al.* (2020) that reported positive effect of applying soil amendments in increasing yield and suppressing soil borne diseases. The low

yields in the untreated control can be attributed to the poor soil conditions and high disease pressure that affected normal growth of the tomato plants.

5.3 Effect of bio-stimulants on management of fusarium wilt and root knot nematodes in tomato

The application of bio-stimulants had a positive effect on the growth of tomatoes compared to the untreated control in both season one and two. *T. harzianum* was effective in the management of Fusarium wilt and root knot nematodes. These results are similar to those of Harman *et al.* (2008) who revealed that application of *Trichoderma spp.* by drenching in the soil or seed inoculation gave results in the microorganism colonizing the roots of the plants in a symbiotic relationship and providing protection to the plants against diseases. El-Shennawy *et al*. (2012) and Mushtaq *et al.* (2011) also reported *Trichoderma spp.* as an effective control measure against fusarium wilt and root knot nematodes.

The application of *T. harzianum* resulted in a significant $P \leq 0.05$ increase in yield compared to the untreated plots. These results agree with findings of Harman *et al*. (2008) who reported that application of *Trichoderma spp.* increases productivity in plants. The increase in production can be attributed to the less disease pressure and low root knot nematode infestation that allowed the crops to grow with less competition for nutrients. The low fusarium and root knot nematode infestation can also be attributed to the plants enhanced defence mechanisms that were elicited due to application of *T. harzianum* (Kloepper *et al*., 2004; Harman *et al*., 2008; Murthy *et al*., 2014). *Trichoderma* spp. also has

ability to produce antibiotics and act as a mycoparasite all of which contribute to the management of soil-borne diseases (Harman *et al.,* 2008).

The application of seaweed extracts for root stimulation was not effective in the management of Fusarium wilt and root knot nematodes. However, the levels of Fusarium wilt and root knot nematode infestation were lower than the levels in the untreated control. However, the effect on yield was significant in both season one and two. These results are consistent with findings by Atzmon *et al.* (1994) and Arthur *et al.* (2003) that reported a positive effect on growth and yield of plants due to application of seaweed products.

The application of amino acids did not result in effective management of fusarium wilt and root knot nematodes. However, the application of amino acids resulted in a significant ($P \leq 0.05$) increase in yield compared to the untreated plots. The increase in yield can be attributed to the fact that amino acids do not directly provide nutrients to the plant but can as signals in different metabolic processes, thus inducing greater assimilation of nitrogen by plants (Santi *et al.,* 2017) which influences positively the growth of the plant. These results are also consistent with findings by Teixeira *et al.* (2017) also showed that application the amino acids influences the plant growth parameters of plants.

The application of humic acids did not differ significantly at $P \leq 0.05$ in reduction of fusarium wilt incidence and the root knot nematode infestation in both season one and two. On the other hand, the application of humic acids positively affected the yield of tomato which was significant at $P \leq 0.05$. The increase in yield can be attributed to the increase in assimilation of both the

micro and microelements by plants and the improved rhizosphere conditions because of application of humic acids (Canellas *et al.,* 2014; Puglisi *et al.,* 2013).

The application of peptides did not differ significantly at P≤0.05 in reducing fusarium wilt incidences and root knot nematodes. The impact on yield was however was significant. These results are consistent with findings by Haeffner *et al.* (2014) and Huffaker *et al.* (2011) who reported a positive effect on crops as a result of application of peptides. The combination of seaweeds, humic acids and amino acids as well as the combination of seaweed, amino acids, humic acids and peptides resulted in a higher increase in yield compared to the single treatments of each in both in seasons. This can be attributed to the synergistic effect that comes along with the combination.

The low yields in the untreated control can be attributed to the high fusarium wilt and root knot nematode infestation which had a negative effcet on the growth of tomatoes. In general the fusarium wilt and root knot nematode infestation was lower in season one compared to season two.

5.4 Effect integrating plant essential oils, soil improvers and bio-stimulants on management of soil borne diseases, arthropod pests and yield of tomato

The integration of plant essential oils, soil improver, *T. harzianum* and bio-stimulants was effective in the managing fusarium wilt, early blight, root knot nematodes, thrips, whitefly as well as increasing tomatoes yield. The need to integrated was necessitated by the fact that the individual treatments that is; plant essential oils, the soil improver, *T. harzianum* and the bio-stimulants only

play a small part in the management of soil-borne diseases, insect pests and increasing yield.

Through integration, the various components have a greater effect because of the synergistic effect. Plant essential oil have been shown to be effective in management of Fusarium wilt, early blight, thrips, whitefly and are reported to have nematicidal effects (Lattanzio *et al.*, 2006; Cetintas *et al.*, 2010). Soil improvers can contribute positively to management of soil borne diseases through alteration of the soil microbial dynamics. An increase in beneficial fungi in the soil results in lower population of the pathogenic species that cause diseases in plants (Mokhtar *et al.*, 2014).

Trichoderma harzianum plays an important role in management of Fusarium wilt and root knot nematodes. Harman *et al.*, (2008) that reported that application of *Trichoderma spp.* by drenching in the soil or seed inoculation results in the microorganism colonizing the roots of the plants in a symbiotic relationship and providing protection to the plants against diseases. El-Shennawy *et al.* (2012) and Mushtaq *et al.* (2011) reported *Trichoderma spp.* as an effective control measure against fusarium wilt and root knot nematodes.

Bio-stimulants play an important role in eliciting metabolic activities of plants and stimulating uptake of nutrients by plants. The positive effect of bio-stimulants that included seaweeds, humic acids, peptides and amino acids among others on tomatoes have been widely reported by other scientists including Santi *et al.* (2017), Teixeira *et al.* (2017), Canellas *et al.* (2014) and Puglisi *et al.* (2013). The integration therefore of plant essential oils, the soil improver, *T.*

harzianum and the bio-stimulants resulted in a synergistic effect that caused effective management of Fusarium wilt, root knot nematodes, early blight, thrips and whitefly. The resultant yield increase as compared to the untreated plots was due to the low disease and insect pest pressure.

The correlation analysis between variables revealed a negative relationship for the various parameters between yield and the other variables assessed in the study clearly illustrates the need for effective pest management options for early blight, fusarium wilt, root knot nematodes, thrips and whitefly in tomato production. The positive relationship between early blight, fusarium wilt, root knot nematodes, thrips and whiteflies clearly illustrates that variables are not antagonistic to each other but rather synergistic. These results agree with findings by Hoysted *et al.* (2017) who reported a positive relationship between below ground pathogens affecting plants and above ground sucking pests.

In this case, effective integrated management of blight, fusarium wilt, root knot nematodes, thrips and whiteflies will result in increased yield in tomato production. From the combined Pearson's correlation analysis, the occurrence of soil borne challenges in tomato production in this case fusarium wilt and root knot nematodes makes that plant more susceptible to infestation by thrips and whitefly and therefore pest management strategies should aim to manage all these challenges.

CHAPTER SIX: CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

i. Plant essential oils (thyme oil) was effective in suppression of fusarium wilt and early blight in Kimbimbi and Kagio by between 49.2% to 65.9% and 58.9% to 84.8% respectively. Also, thyme oil was effective in the management of thrips and whitefly infestations causing a reduction in infestation of between 47% to 68.6 % and 41.3% to 66.2% respectively.

ii. The application of the soil improver had a positive effect on the microbial population of fungi resulting in an increase of active fungi in the rhizosphere of tomato plants of up to 292%. Also the application of the three rates of the soil improver at 200kg/Ha, 300Kg/Ha and 400Kg/Ha increased the tomatoes yield compared to the untreated control in both Kimbimbi and Kagio sites. This represnted an increase of 141.7%, 125.0% and 125.0% increase in Kimbimbi and 29.2%, 37.5% and 20.8% in Kagio respectively.

iii. The application of bio-stimulants a positive effect in reduction of Fusarium wilt and root nematodes. For instance, *T. harzianum* reduced Fusarium wilt by 39.6% and 48.6% in first and second season respectively. Also, the treatment with a combination seaweed and humic acids as well as the treatment with seaweeds, amino acids and peptides, recorded the highest tomatoes yields in both the first and second season.

iv. The integration of thyme oil, the soil improver, *T. harzianum* and bio-stimulants reduced the root knot nematode infection in both Kagio (between

35.4 to 60.1%) and Kimbimbi (between 27.6 to 40.8%) compared to the untreated plots.

6.2 Recommendations

1. The adoption of plant essential oils (thyme oil) for the management of fusarium wilt, early blight, thrips and whitefly in tomato production systems by farmers.

2. The adoption of *Trichoderma harzianum* for management of fusarium wilt and root knot nematodes in tomato production systems by farmers.

3. More studies to be carried out on the effective rates of the soil improver that is sufficient to significantly reduce incidences of fusarium wilt in tomato production systems

4. The adoption of the integration of plant essential oils (thyme oils), the soil improver, *Trichoderma harzianum* and bio-stimulants for the management of fusarium wilt, early blight, root knot nematodes, thrips, whitefly as well as increasing yield tomato

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ANNEXES

Annex I: Anova tables

Analysis of variance: Levels of Fusarium wilt incidence index in tomatoes as influenced by application of various integrated options in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	0.9314	0.3105	1.48	
Block.*Units* Stratum Treatment	8	9.4097	1.1762	5.61	<.001
Residual	24	5.0334	0.2097		
Total	35	15.3745			

Analysis of variance: Levels of Fusarium wilt incidence index in tomatoes as influenced by application of various integrated options in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	1.7848	0.5949	2.22	
Block.*Units* Stratum Treatment	8	14.3610	1.7951	6.69	<.001
Residual	24	6.4372	0.2682		
Total	35	22.5830			

Analysis of variance: Levels of early blight infection in tomatoes as influenced by application of various integrated options in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	82.967	27.656	5.64	
Block.*Units* Stratum Treatment	8	2084.150	260.519	53.16	<.001
Residual	24	117.610	4.900		
Total	35	2284.727			

Analysis of variance: Levels of early blight infection in tomatoes as influenced by application of various integrated options in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	88.596	29.532	3.70	
Block.*Units* Stratum Treatment	8	1413.799	176.725	22.15	<.001
Residual	24	191.482	7.978		
Total	35	1693.877			

Analysis of variance: Levels of Nematode infestation in tomatoes as influenced by application of various integrated options in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	0.1416	0.0472	0.24	
Block.*Units* Stratum Treatment	8	5.0462	0.6308	3.16	0.014
Residual	24	4.7978	0.1999		
Total	35	9.9856			

Analysis of variance: Levels of Nematode infestation in tomatoes as influenced by application of various integrated options in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	0.6219	0.2073	1.97	
Block.*Units* Stratum Treatment	8	9.1977	1.1497	10.94	<.001
Residual	24	2.5225	0.1051		
Total	35	12.3421			

Analysis of variance: Levels of thrips infestation on tomatoes treated with various integrated options in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	1.4224	0.4741	1.89	
Block.*Units* Stratum Treatment	8	15.9577	1.9947	7.95	<.001
Residual	24	6.0240	0.2510		
Total	35	23.4040			

Analysis of variance: Levels of thrips infestation on tomatoes treated with various integrated options in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	2.0709	0.6903	2.87	
Block.*Units* Stratum Treatment	8	11.3609	1.4201	5.89	<.001
Residual	24	5.7818	0.2409		
Total	35	19.2135			

Analysis of variance: Levels of whitefly infestation on tomatoes treated with integrated options in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	326.79	108.93	8.64	
Block.*Units* Stratum Treatment	8	1015.97	127.00	10.08	<.001
Residual	24	302.41	12.60		
Total	35	1645.18			

Analysis of variance: Levels of whitefly infestation on tomatoes treated with integrated options in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	1064.2	354.7	2.52	
Block.*Units* Stratum Treatment	8	1086.9	135.9	0.96	0.486
Residual	24	3380.0	140.8		
Total	35	5531.1			

Analysis of variance: Mean yield of tomato per treatment in tons/ha per treatment in Kimbimbi

Variate: Mean

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Block stratum	3	4.517	1.506	0.51	
Block.*Units* Stratum Treatment	8	61.760	7.720	2.61	0.033
Residual	24	71.073	2.961		
Total	35	137.350			

Analysis of variance: Mean yield of tomato per treatment in tons/ha per treatment in Kagio

Variate: Mean

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Block stratum	3	10.172	3.391	0.68	
Block.*Units* Stratum Treatment	8	112.591	14.074	2.82	0.023
Residual	24	119.823	4.993		
Total	35	242.586			

Analysis of variance: Levels of Fusarium wilt incidence index in tomatoes as influenced by application of various bio-stimulants in season one

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	4.336	1.445	0.51	
Block.*Units* Stratum Treatment	5	51.338	10.268	3.60	0.024
Residual	15	42.755	2.850		
Total	23	98.428			

Analysis of variance: Levels of Fusarium wilt incidence index in tomatoes as influenced by application of various bio-stimulants in season two

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	9.805	3.268	0.83	
Block.*Units* Stratum Treatment	5	94.014	18.803	4.77	0.008
Residual	15	59.179	3.945		
Total	23	162.999			

Analysis of variance: Levels of Nematode infestation in tomatoes as influenced by application of various bio-stimulants in season one

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	1.5468	0.5156	4.62	
Block.*Units* Stratum Treatment	5	7.7700	1.5540	13.92	<.001
Residual	15	1.6746	0.1116		
Total	23	10.9914			

Analysis of variance: Levels of Nematode infestation in tomatoes as influenced by application of various bio-stimulants in season two

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	2.8858	0.9619	2.29	
Block.*Units* Stratum Treatment	5	7.5921	1.5184	3.61	0.024
Residual	15	6.3054	0.4204		
Total	23	16.7833			

Analysis of variance: Incidence of fusarium wilt infection in tomato plots sprayed with various concentrations of plant essential oils in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	2.1565	0.7188	2.01	
Block.*Units* Stratum Treatment	5	34.4252	6.8850	19.25	<.001
Residual	15	5.3639	0.3576		
Total	23	41.9456			

Analysis of variance: Incidence of fusarium wilt infection in tomato plots sprayed with various concentrations of plant essential oils in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	2.7551	0.9184	2.92	
Block.*Units* Stratum Treatment	5	34.2041	6.8408	21.77	<.001
Residual	15	4.7143	0.3143		
Total	23	41.6735			

Analysis of variance: Levels of early blight infection on tomatoes sprayed with various concentrations of plant essential oils in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	26.180	8.727	1.29	
Block.*Units* Stratum Treatment	5	3764.897	752.979	111.70	<.001
Residual	15	101.116	6.741		
Total	23	3892.193			

Analysis of variance: Levels of early blight infection on tomatoes sprayed with various concentrations of plant essential oils in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	11.65	3.88	0.25	
Block.*Units* Stratum Treatment	5	1631.88	326.38	21.04	<.001
Residual	15	232.63	15.51		
Total	23	1876.16			

Analysis of variance: Levels of whitefly infestation on tomatoes sprayed with various concentrations of plant essential oils in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	36.86	12.29	1.16	
Block.*Units* Stratum Treatment	5	1269.83	253.97	24.00	<.001
Residual	15	158.70	10.58		
Total	23	1465.39			

Analysis of variance: Levels of whitefly infestation on tomatoes sprayed with various concentrations of plant essential oils in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	22.62	7.54	0.56	
Block.*Units* Stratum Treatment	5	897.84	179.57	13.28	<.001
Residual	15	202.78	13.52		
Total	23	1123.24			

Analysis of variance: Levels of thrips infestation on tomatoes sprayed with various concentrations of plant essential oils in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	0.3222	0.1074	0.31	
Block.*Units* Stratum Treatment	5	28.2509	5.6502	16.31	<.001
Residual	15	5.1962	0.3464		
Total	23	33.7693			

Analysis of variance: Levels of thrips infestation on tomatoes sprayed with various concentrations of plant essential oils in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	0.7941	0.2647	1.98	
Block.*Units* Stratum Treatment	5	27.1408	5.4282	40.53	<.001
Residual	15	2.0090	0.1339		
Total	23	29.9439			

Analysis of variance: Incidence of fusarium wilt infection in tomato plots treated with different rates of the soil improvers in Kimbimbi

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	10.674	3.558	1.35	
Block.*Units* Stratum Treatment	5	9.190	1.838	0.70	0.634
Residual	15	39.529	2.635		
Total	23	59.393			

Analysis of variance: Incidence of fusarium wilt infection in tomato plots treated with different rates of the soil improvers in Kagio

Variate: Mean

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	1.459	0.486	0.20	
Block.*Units* Stratum Treatment	5	30.388	6.078	2.45	0.081
Residual	15	37.143	2.476		
Total	23	68.990			

Annex II: Pathological analysis for Kagio and Kimbimbi sites

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Client	Geoffrey Ongoya
Farm	Sample A & B
Analysis	Pathology Test
Receiving date	15th September, 2016
Analysis date	22nd September, 2016
Sample Type	Soil Pathology
Crop	Tomatoes

Key:		Colour code
0	Not detected	
1	Starting infection	
2	Light infection	
3	Moderate infection	
4	Infected	
5	Severely infected	

		Bacterial Pathogens				Fungal Pathogens						Saprophytic Fungi				
Sample No.	**Field Name**	*Ralstonia solanacearum*	*Xanthomonas spp*	*Agrobacterium tumefasciens*	*Pseudomonas spp*	*Fusarium spp*	*Fusarium oxysporum*	*Alternaria spp*	*Verticillium spp*	*Rhizoctonia spp*	*Pythium spp*	*Trichoderma spp*	*Aspergillus spp*	*Penicillium spp*	*Rhizopus*	*Mucor*
CK105PT0042	Kagio Sample A	0	0	0	0	0	3	0	0	0	0	0	2	0	0	0
CK305PT0043	Kimbimbi- Sample B	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0

Remarks/Descriptions

Ralstonia solanacearum and ***Fusarium oxysporum*** were detected in some of the samples.Saprophytic fungi ***Aspergillus* spp and Trichoderma spp** were also detected in some of the samples.

Disease information: The ***Ralstonia solanacearum*** causes the bacterial wilt disease in tomatoes ,capsicums and other solanaceae family crops/plants .The disease causes significant yield losses, through the rapid wilting of the plants.

The first visible symptom of the disease in the crops is wilting of the leaves at the ends of the branches during hot days with recovery at night. The wilt progresses rapidly to other branches and the plants die while green.Sometimes the vascular bundles become bricky red in colour and are usually wet due to the bacteria which easily oozes out.

Disease development is favoured by warm temperatures (above 15°C with optimum of 27°C) and high soil moisture levels. The disease is spread mainly through infected soil, contaminated water and infected planting materials.The disease can survive in infected plant debris.Its mechanically transmitted (Hence can be passed on by people through contaminated hands/tools).

Fusarium oxysporum is a soil borne fungi which causes **fusarium wilts** in tomatoes, capsicum and other crops.They have a wide host range including ornamental plants. They invade the xylem tissue, or vascular tissue of plants leading to browning of vascular tissues,wilts and root rots . They are characterized by wilting; yellowing, stem rot, plant die back and leaf fall when the leaves have dried up .

The fungi are easily transfered through soil , contaminated water and use of infected planting materials.

Saprophytic fungal spp (*Trichoderma* and *Aspergillus* spp).These are saprophytic fungi which cause **rot** diseases on certain fruits , vegetables and grains. Proper aeration both in the field and storage limits their growth.

Pictorial guide on detection for *Ralstonia solanacearum*

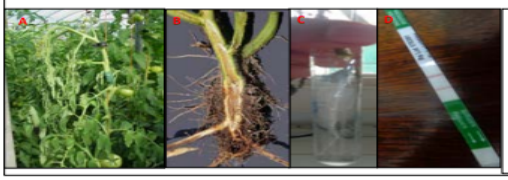

Fig A: The dying plant in the midst of healthy ones.The plant die and remain green.

Fig B: The discolouration (browning) of the vascular bundles.

Fig C: The oozing of the bacteria from the stem.Arrow 1 indicates the bacteria streaming while arrow 2 indicate the accumulation of the bacteria at the bottom of tube.

Fig D: Confirmatory Biorela agristrip test for *Ralstonia solanacearum*.
Two lines indicate positive.

Recommendations

To avoid further spread of the diseases to other areas and to avoid new introductions ;

1) Restrict entry to the field.Only urgent activities should be carried out.

2) If already having a crop; check all symptomatic plants in the field and uproot them plus their immediate neighbors. (The diseased plants should be carried in plastic bags and disposed off by burning. The uprooting should be done end of the day to avoid contamination.

3) Disinfect the areas that have been uprooted with Sodium Hypochlorite (Jik).

4) Practice crop rotation.You can follow this plan;(E.g in **season 1** you can have tomatoes or capsicums or egg plants and any solanacea crop;in season 2 have cabbage or brocolli or brusel sprouts or cauliflowers or any brassicaea family crops and then in **season 3** plant the onions or garlic or leeks or chives or any allium crop) then can go back to the **season 1** crops for **season 4** and repeat.

5) Ensure proper scouting for any symptomatic plants and immediate reporting done for appropriate action.If any plant found; follow the action in 2 with incorporation of the rest of the actions below.

6) Ensure all tools and equipment used per area are disinfected before and after use. If used on the plants, disinfect after each plant.

7) Ensure feet disinfection using sodium hypochlorite is done and hand disinfection with alcohol or Dettol solution.

8) Have dedicated PPEs for each of the areas in the areas.

9) When starting a new crop sterilize the soil and only use certified seeds/seedlings for planting.

In addition to the above; for *Fusarium* ,

10) Incorporate biological control agents during planting e.g. *Trichoderma harzianum* to boost the plant immunity.

11) Drench the plants with **Fosetyl Aluminium** fungicide. This boosts root development in addition to controlling the fungi.

Annex III: Nematode analysis for greenhouse at Kenyatta University

Nematodes Analysis Report

Nematodes in Soil

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Customer:	Geoffrey Ongoya Wafula	Crop:	Tomatoes	Date Received:	21-Aug-18
Address:		Crop Stage:		Analysis Date:	23-Aug-18
Farm Name:	KU Greenhouse	Comments:		Report Date:	29-Aug-18
Contact Person:	Geoffrey Ongoya	Condition:	Moist	Sample ID:	CK105NEM0012

Field: **KU Greenhouse** Top Soil

To maintain the correct history ensure that the next sample sent from this Field is labelled: Kagio-KU Greenhouse

History (Last 3 analysis)

Parameter	Unit	Result	Guide Low	Guide High	Low	Optimum	High	Symbol	Current				Method
Saprofagic	100 ml	2040		> 1000				Sap	2040				
Meloidogyne	100 ml	10.0		< 30.0				Mel	10.0				
Pratylenchus	100 ml	20.0		< 50.0				Pra	20.0				
Radopholus	100 ml	0.00		< 50.0				Rad	0.00				
Tylenchus	100 ml	40.0		< 100				Tyl	40.0				
Tylenchorhynchus	100 ml	40.0		< 50.0				Tyle	40.0				
Helicotylenchus	100 ml	0.00		< 100				Hel	0.00				
Scutellonema	100 ml	0.00		< 100				Scu	0.00				
Aphelenchus	100 ml	0.00		< 1000				Aps	0.00				
Xiphinema	100 ml	0.00		< 200				Xip	0.00				
Longidorous	100 ml	0.00		< 40.0				Lo	0.00				
Hemicyclophora	100 ml	0.00		< 100				Hem	0.00				
Criconema	100 ml	0.00		< 100				Cri	0.00				

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Gakobo Jo Lab Manager	Cordingley Jeremy Managing Director	Approval Date: ***29/08/2018***

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Annex IV: Nematode analysis for the Kagio site

Nematodes Analysis Report

Nematodes in Soil

CROPNUTS Laboratory Services ISO17025 Accredited

Customer:	Geoffrey Ongoya Wafula	Crop:	Tomatoes	Date Received:	15-Jul-19
Address:		Crop Stage:		Analysis Date:	18-Jul-19
Farm Name:	KAGIO	Comments:		Report Date:	22-July-19
Contact Person:	Geoffrey Ongoya	Condition:	Moist	Sample ID:	CK107NEM0019

Field: **Kagio -Sample B** Top Soil

To maintain the correct history ensure that the next sample sent from this Field is labelled: Kagio Sample B

History (Last 3 analysis)

Parameter	Unit	Result	Guide Low	Guide High	Low	Optimum	High	Symbol	Current				Method
Saprofagic	100 ml	1040		> 1000				Sap	1040				
Meloidogyne	100 ml	20.0		< 30.0				Mel	20.0				
Pratylenchus	100 ml	20.0		< 50.0				Pra	20.0				
Radopholus	100 ml	0.00		< 50.0				Rad	0.00				
Tylenchus	100 ml	40.0		< 100				Tyl	40.0				
Tylenchorhynchus	100 ml	40.0		< 50.0				Tyle	40.0				
Helicotylenchus	100 ml	0.00		< 100				Hel	0.00				
Scutellonema	100 ml	0.00		< 100				Scu	0.00				
Aphelenchus	100 ml	0.00		< 1000				Aps	0.00				
Xiphinema	100 ml	0.00		< 200				Xip	0.00				
Longidorous	100 ml	0.00		< 40.0				Lo	0.00				
Hemicyclophora	100 ml	0.00		< 100				Hem	0.00				
Criconema	100 ml	0.00		< 100				Cri	0.00				

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Gakobo Jo
Lab Manager

Cordingley Jeremy
Managing Director

Approval Date: ***22/07/2019***

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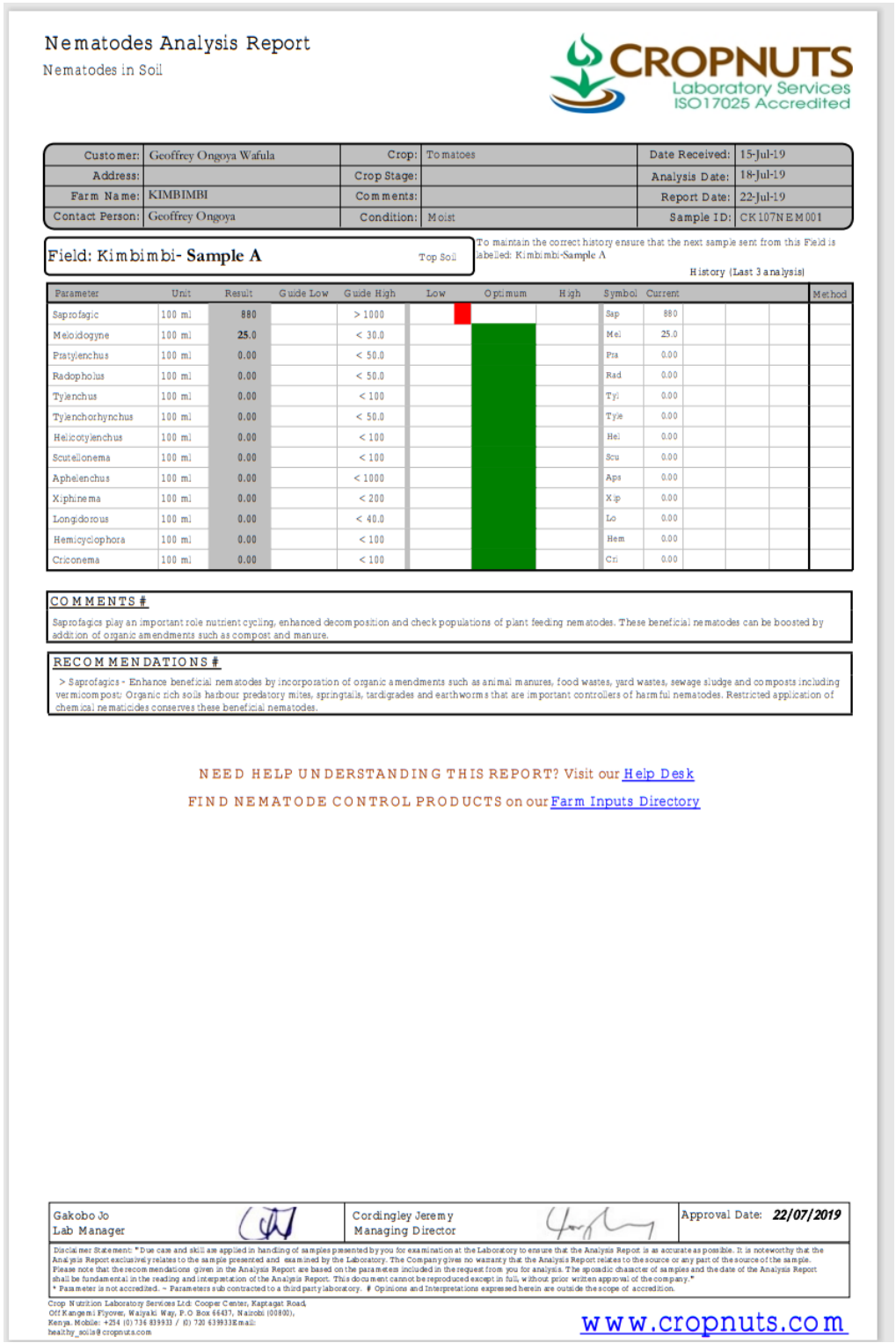
Annex V: Nematode analysis for the Kimbimbi Site



Nematodes Analysis Report

Nematodes in Soil

CROPNUTS Laboratory Services ISO17025 Accredited

Customer:	Geoffrey Ongoya Wafula	Crop:	Tomatoes	Date Received:	15-Jul-19
Address:		Crop Stage:		Analysis Date:	18-Jul-19
Farm Name:	KIMBIMBI	Comments:		Report Date:	22-Jul-19
Contact Person:	Geoffrey Ongoya	Condition:	Moist	Sample ID:	CK107NEM001

Field: Kimbimbi- **Sample A** Top Soil

To maintain the correct history ensure that the next sample sent from this Field is labelled: Kimbimbi-Sample A

History (Last 3 analysis)

Parameter	Unit	Result	Guide Low	Guide High	Low	Optimum	High	Symbol	Current				Method
Saprofagic	100 ml	880		> 1000				Sap	880				
Meloidogyne	100 ml	25.0		< 30.0				Mel	25.0				
Pratylenchus	100 ml	0.00		< 50.0				Pra	0.00				
Radopholus	100 ml	0.00		< 50.0				Rad	0.00				
Tylenchus	100 ml	0.00		< 100				Tyl	0.00				
Tylenchorhynchus	100 ml	0.00		< 50.0				Tyle	0.00				
Helicotylenchus	100 ml	0.00		< 100				Hel	0.00				
Scutellonema	100 ml	0.00		< 100				Scu	0.00				
Aphelenchus	100 ml	0.00		< 1000				Aps	0.00				
Xiphinema	100 ml	0.00		< 200				Xip	0.00				
Longidorous	100 ml	0.00		< 40.0				Lo	0.00				
Hemicyclophora	100 ml	0.00		< 100				Hem	0.00				
Criconema	100 ml	0.00		< 100				Cri	0.00				

COMMENTS #

Saprofagics play an important role nutrient cycling, enhanced decomposition and check populations of plant feeding nematodes. These beneficial nematodes can be boosted by addition of organic amendments such as compost and manure.

RECOMMENDATIONS #

> Saprofagics - Enhance beneficial nematodes by incorporation of organic amendments such as animal manures, food wastes, yard wastes, sewage sludge and composts including vermicompost: Organic rich soils harbour predatory mites, springtails, tardigrades and earthworms that are important controllers of harmful nematodes. Restricted application of chemical nematicides conserves these beneficial nematodes.

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