DEVELOPMENT OF MEAN CONCENTRATION STIMULATION POINT FOR FERMENTED *LANTANA CAMARA* PHYTONEMATICIDE ON TOMATO PRODUCTION

BY

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DECLARATION

I, Kgashane Phillip Malatji, declare that the mini-dissertation hereby submitted to the University of Limpopo, for the degree Master of Science in Agriculture (Agronomy) has not previously been submitted by me for degree at this or any other university; that it is my work in design and in execution, and that all material contained herein has been duly acknowledged.

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Malatji K.P. (Mr) Date

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DEDICATION

To my late father, Mr M.J. Malatji, and my late grandmother, Mrs M.R. Monyela.

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ABSTRACT

Root-knot nematodes (*Meloidogyne* species) are the major soil-borne pests of tomato (*Solanum lycorpesicum*) plants. Due to the global withdrawal of effective chemical nematicides from the agrochemical markets, nematodes are difficult to control under the production systems. Currently, botanicals are being researched and developed as alternative to chemical nematicides with promising results, although they have challenge of phytotoxicity. The objective of this study was to determine the Mean Concentration Stimulation Point (MCSP) of Tickberry (*Lantana camara*) extracts for tomato plant-infected with *M. javanica*. Treatments consisted of six levels of *L. camara* extracts, namely, 0, 2, 4, 6, 8 and 10% per pot, which were arranged in a randomised complete block design, with ten replicates. Tomato seedlings were inoculated with 2500 second-stage juveniles (J2S) of *M. javanica* at five days after transplanting, with treatments applied at seven days after inoculation. At 56 days after inoculation, *L. camara* extracts had positive effects on plant height, stem diameter, number of leaves, number of fruits and fruit mass, contributing 65, 74, 61, 25 and 61% in total treatment variation (TTV), respectively, under greenhouse conditions. Under microplot conditions, treatments contributed 55, 85, 61, 36 and 85% in TTV of the respective plant variables. Under greenhouse it contributed 60, 35 and 77% and 29, 79 and 70% under microplot on dry shoot mass, dry root mass and galling index respectively. Treatments did not have any effects on soil pH and electrical conductivity (EC). Under greenhouse conditions, treatments contributed 88, 94 and 92% in TTV of nematode in roots, soil and final population, respectively, whereas under microplot conditions 94, 97 and 95% in

TTV of the respective nematode stages. The derived mean concentration of *L. camara* extracts for tomato was 5.76 and 5.31% under microplot and greenhouse conditions, respectively. The overall sensitivity of tomato plants to *L. camara* extracts under microplot and greenhouse were 3 and 0, respectively. In conclusion *Meloidogyne* species can be managed using *L. camara extracts* 5.31 and 5.76% under glasshouse production and field production system respectively.

CHAPTER 1
GENERAL INTRODUCTION

1.1 Introduction

Root-knot (*Meloidogyne* species) nematodes are the major plant-parasitic nematodes affecting the quantity and quality of crop production in different annual and perennial crops. *Meloidogyne* species are considered the most damaging and cause serious yield losses on a wide range of crops (Javad *et al*., 2006). Generally, nematodes are difficult to control because of their wide host ranges and high rate of reproduction, with females capable of producing up to thousand eggs per hatch (Natarjan *et al*., 2006). The withdrawal of synthetic chemical nematicide such as methyl bromide from agrochemical markets following the adoption of the Montreal protocol (1997) increased the incidence of crop failures in most active agricultural soils (Pelinganga *et al*., 2011). Economic damage in tomato (*Solanum lycorpersicum*) could occur when densities of *Meloidogyne* species were from of 0.1 to 1.0 nematode per cm^3 soil at transplanting (Sikora and Fernandez, 2005). Seinhorst (1965) indicated that crop damage due to nematodes was directly proportional to the initial nematode population densities (Pi).

Meloidogyne species are among the most destructive nematodes in agriculture with estimated yield loss of US $100 billion worldwide (Oka *et al*., 2007). Adesiyan *et al*. (1990) reported that the estimated yield loss of tomato ranged from 28 to 68%, whereas Sasser (1979) reported 85% yield loss. In South Africa, the estimated crop loss annually due to nematode damage in all crops was previously standing at 14% (Swart, 2010). The genus *Meloidogyne* constitutes a number of species, with the major species

considered the most destructive being *M. incognita*, *M. javanica*, *M. arenaria* and *M. hapla* (Esfahani, 2009). Additionally, the listed four *Meloidogyne* species are characterised by having biological races. Within the same nematode species, the races are morphologically similar and are distinguished using differential host plants and molecular approaches (Garcia and Sánchez-Puerta, 2012).

Methods for managing nematodes such as chemical control, organic amendments, resistant varieties, soil solarisation and biological control had been tried with different levels of success in tomato production (Sakhuja and Jain, 2001). Chemical nematicides have been effective in management of plant-parasitic nematodes; however, due to their deleterious effects on the environment and non-target organisms focus had been redirected to the use of alternative methods with potential harmless effects on the environment. Due the problems unsustainable nature of nematicides, sustainable natural products with nematicidal activities such as nematode resistance and endophytic microbes (Vetrivelkalai *et al*., 2010) and plant extracts (Mashela *et al*., 2015; Muniasamy *et al*., 2010; Pavaraj *et al*., 2010).

Plant-based pesticides have found favour as alternatives to chemical pesticides and are being exploited commercially in pest management (Agnihotri *et al*., 1999). Different plant species are being tested to identify the sources of nematicidal substances and many of them have shown promising results in the control of plant parasitic nematodes (Abdi, 1996). However, it had been reported that not all plant organs contain allelochemicals with nematicidal properties (Mashela *et al*., 2015). Secondary

metabolites are used in development of botanical extracts which are low-molecular weight compounds that do not play a role in primary plant metabolism (Van Wyk and Wink, 2004).

Development of phytonematicides depended on allelochemicals which are considered as the active ingredients and are naturally phytotoxic. Allelochemicals have great potential for pesticides because they are free from problems associated with present pesticides. Tickberry (*Lantana camara*) had been reported to inhibit germination, growth, development or metabolism of neighbouring plants due to secretion of allelochemicals (Qasem, 2006). Application of allelochemicals at higher concentrations might well produce phytotoxic symptoms. Faheem *et al.* (2016) suggested that prior to field application for suppression of plant-parasitic nematodes the concentration of *L. camara* extracts which could be toxic to nematodes but not to plants to be protected should be determined.

1.2 Problem statement

Phytotoxins are used as agricultural chemicals such as bio-nematicides to substitute the withdrawal of synthetic chemicals and their hazardous effects on the environment (Awan *et al*., 2012). Toxicity of *L. camara* had been researched and reported to possess nematicidal properties (Ahmad *et al*., 2010; Kalita and Bhola, 2013). Nematicidal and nematostatic activities of *L. camara* against *Meloidogyne* species have also been *in vitro* and in soil experiments (Ahmad *et al*., 2010; Qamar *et al*., 2005). However, there

are no reports on Mean Concentration Stimulation Point (MCSP) of *L. camara* extracts in tomato production.

The MCSP is the concentration of a phytonematicide that would stimulate plant growth, while inhibiting nematode population densities (Mashela *et al.,* 2015). It is quantified as: MCSP = $D_m + (R_h/2)$, where D_m is the threshold stimulation point while R_h is saturation level of the applied dosage of phytonematicides.

1.3 Rationale

Fermented *L. camara* has been used for managing nematode population densities at the ZZ2 in tomato production as an alternative to synthetic chemical nematicides (Taurayi, 2011). The plants are harvested locally in the wild and fermented in airtight containers until the pH of the mixture is at approximately 3.7 (Mashela *et al*., 2015). Currently, without empirically-based information, the product is applied weekly at 3.68% per hectare, with some evidence of phytotoxicity to tomato plants (Daneel *et al*., 2014; Hiten *et al*., 2011). In order to avoid the incidence of phytotoxicity on tomato plants, it is imperative that the MCSP of the product on tomato be empirically developed.

1.4 Purpose of the study

1.4.1 Aim

The aim of the study was to develop the MCSP of fermented *L. camara* crude extracts on tomato plants for managing the root-knot nematodes.

1.4.2 Objective

To determine whether the MCSP of *L. camara* extracts for tomato plants inoculated with *M*. *javanica* could be established under greenhouse and microplot conditions.

1.5 Hypothesis

The MCSP of *L. camara* extracts for tomato plants inoculated with *M*. *javanica* would be established under greenhouse and microplot conditions.

1.6 Structure of mini-dissertation

The mini-dissertation was structured using the Senate-approved format of the University of Limpopo (UL). Findings were summarised in the abstract, followed by detailed background to the research problem (Chapter 1), which was in turn followed by the review of relevant literature on the research problem (Chapter 2) with the objective of the study being addressed as a standalone (Chapter 3). Finally, findings were summarised, with related recommendations being provided regarding the use of fermented *L. camara* as phytonematicide (Chapter 4).

CHAPTER 2
LITERATURE REVIEW

2.1 Work done on the problem statement

2.1.1 Phytotoxicity of phytonematicide

Phytonematicides are depended on allelochemicals which are considered as the active ingredients and are naturally phytotoxic. The use of phytonematicides is often limited by the observed phytotoxicity to the protected plants (Mashela *et al*., 2015). In tomato seedlings the application of crude extracts of garlic bulb at 5% concentration reduced population densities of plant-parasitic nematode but was also highly phytotoxic. In South Africa, Mashela *et al.* (2011) developed phytonematicides from fruits of indigenous *Cucumis* species in granular formation, Nemarioc-AG and Nemafric-BG phytonematicides or in liquid formulation, Nemarioc-AL and Nemafric-BL phytonematicides (Pelinganga *et al*., 2013). Granular formulation of the two phytonematicides was observed to be phytotoxic to eight monocotyledonous and ten dicotyledonous crops when applied at 5 g (Mafeo and Mashela 2009a, b, 2010), while in liquid formulation the material was phytotoxic to tomato seedlings when applied at 10% concentration (Pelinganga and Mashela, 2012). *Lantana camara* had been reported to inhibit germination, growth development or metabolism of neighbouring plants due to secretion of allelochemicals (Qasem, 2006).

2.1.2 Allelopathic effect of *Lantana camara*

Allelopathy has been defined in several literatures as the act of one plant to inhibit or stimulate the growth of other neighbouring plants through the release of allelochemicals

to the surrounding environment (Ghafarbi *et al*., 2012). *Lantana camara* secretes allelochemicals to the rhizosphere of the nearby crops and inhibit germination, growth and metabolism development (Qasem, 2006). These allelochemicals affect many cellular processes in target crops including disruption of membrane permeability. Different concentration of leaf extracts on *L. camara* causes reduction on germination of agricultural crops. Maiti *et al.* (2010) showed that allelopathic effects of different concentration of aqueous leaf extracts and leaf leachates were inhibitory to mung bean seed germination.

2.1.3 Nematicidal properties of *Lantana camara*

Aerial parts of *L. camara* are known to possess nematicidal properties. The nematicidal and nematostatic activities of *L. camara* against root-knot nematodes have been evaluated *in vitro* and *in vivo* experiments (Ahmad *et al*., 2010 and Qamar *et al*., 2005). Several compounds from aerial parts such as monoterpenoids and triterpenoids have been tested for their nematicidal activity against *M. incognita* (Begun *et al*., 2000). Qamar *et al*. (2005) isolated lantanilic acid, camaric acid and oleanolic acid from the methanolic extract of the aerial parts of *L. camara* through bio-assay guided fractionation. These compounds exhibited significant mortality against *M. incognita* at 0.5% concentration (Qamar *et al*., 2005).

Studies has shown that the use of Lantana extracts inhibit plant parasitic nematodes. Aqueous extracts of *L. camara* caused high mortality of *M. javanica in vitro*. It has also been reported that incorporation of *L. camara* materials in the soil has significance

reduction in population density of *M. javanica* and subsequent root-knot infection of Mung bean growing in the same plots (Ali *et al*., 2001).

2.1.4 Development of fermented lantana extracts

Fermented *Lantana* extracts has been developed and used by ZZ2 since 2002 and it is currently the main product used for root-knot nematode management on tomato. The product is developed through anaerobic fermentation of *L. camara* with the addition of effective micro-organisms (EM).

2.1.5 Effective micro-organisms

Effective microorganisms (EM) are a commercially available concoction of microorganisms used to enhance plant growth and yield (Zimmermann and Kamukuenjandje, 2008). The concoction comprises predominant populations of lactic acid bacteria, yeast, photosynthetic bacteria, actinomycetes and other types of organisms. In development of phytonematicides, EM is used to mine the chemicals out of the plant materials prior to application through irrigation system.

2.1.6 Curve-fitting Allelochemical Response Dosage

The Curve-fitting Allelochemical Response Dosage (CARD) model (Liu *et al*., 2003) was adapted to develop the mean concentration stimulation point (MCSP) concept (Mashela *et al*., 2015). Studies were conducted using CARD model to determine concentrations where phytonematicides developed from fruits of *Cucumis* species had no effect and inhibitory effect on germination of various crops in order to establish the pre-emergent

quantities of crude extracts of *Cucumis* fruit (Mafeo and Mashela, 2010; Mafeo *et al*., 2010). The development of phytonematicides is depended upon allelochemicals that serve the active ingredients, which are naturally phytotoxic to plants. The use of phytonematicides is often limited by its phytotoxicity to the crop being protected against nematode damage. Organisms respond to different concentrations of phytonematicides through density-dependent growth (DDG) patterns (Liu *et al*., 2003). Mashela *et al*. (2015) reported that the CARD model could provide the biological indices that could be used in computing the mean concentration that, when properly used, would consistently reduce nematode numbers without causing phytotoxicity. Also, the CARD model provides the sensitivity of organisms to the test phytonematicide (Liu *et al*., 2003).

In short, the CARD model generates biological indices which are used to calculate the MCSP, which was defined as the concentration of a phytonematicide that would stimulate plant growth while inhibiting nematode population densities (Mashela *et al*., 2015). Among the biological indices, the sensitivity index (k) provides information on the susceptibility of the organism to the applied extracts (Liu *et al*., 2003). Generally, the k value is inversely proportional to the degree of sensitivity of the test organism to the phytonematicide (Liu *et al*., 2003). Mashela *et al.* (2015) reported that the sensitivity of plants to the phytonematicide is an aggregate of all k values generated from separate plant variables, which could be expressed as an overall sensitivity (Σk) value (Liu *et al*., 2003).

2.2 Work not done on problem statement

The MCSP of *L. camara* extracts under tomato product with respect to managing plant-parasitic nematodes, along with the overall sensitivities of tomato to this product, had not been empirically-developed. The development of the MCSP for *L. camara* would allow for the development of the application interval, and then the dosage model (Mashela *et al*., 2015).

CHAPTER 3
RESPONSES OF TOMATO GROWTH AND NEMATODE NUMBERS TO FERMENTED *LANTANA CAMARA*

3.1 Introduction

In South Africa, tomato is the second most important vegetable crop grown and its production is limited by various fungal, bacterial, viral and nematode pests (Lin *et al*., 2009). *Meloidogyne* species are the main soil borne pests of tomato plants across the world (Jacquet *et al*., 2005), due to their wide host range. *Meloidogyne* species have a wide host range and are considered to be the greatest threat in plant production (Ameer-Zareen *et al*., 2003). Due to their short life cycle of four to six weeks, *Meloidogyne* species are difficult to manage in the presence of a suitable host (Pakeerathan *et al*., 2009).

Plant-parasitic nematodes are recognized as the causes of serious yield losses on a wide range of crops (Javad *et al.,* 2006). The most destructive species are *M. incognita* and *M. javanica* which cause serious problem in various agricultural crops. It had been estimated that parasitic nematodes causes yield reduction of about 5 to 15% (Stirling, 2014). Currently there has been an increase in the intensity of search for alternative sources of effective, ecologically sound and safe control methods against nematodes.

Due to the problems concerning environmental pollution by chemical nematicides, the use of botanical extracts of controlling *Meloidogyne* species is becoming appealing. Deregistration of some hazardous nematicides such as methyl bromide, there is been

pressure on farmers to use non-chemicals pest control methods that do not pollute the environment (Agbenin, 2005). A wide variety of plant species representing 57 families have been shown to possess nematicidal compound (Sukul, 1992), which includes isothiocunates, thiophenics glycoside, alkaloids, phenolics and fatty acids (Gommers, 1973). Chitwood (2002) reported that nematicidal phytochemicals are generally safe for environment and humans.

During the past two decades, nematode control was largely depended on synthetic nematicides, which besides being costly they also have negative impact on the environment and non-target organisms. Currently there is intensity in search for alternatives to chemicals whereby possibilities are being investigated of exploiting nematode antagonistic plants for the management of plant parasitic nematodes (Akhtar, 2004; Chitwood, 2002). Certain weeds extracts have been demonstrated to cause substantial mortality of plant-parasitic nematodes with improved plant growth and yield (Shaukat *et al*., 2004).

Lantana camara is an invasive shrub in some tropical countries, like in the lowveld of South Africa (Urban *et al*., 2011). The plant species possesses nematicidal properties that suppress the development and reproduction of *Meloidogyne* species (Ahmad *et al*., 2010). The active ingredients had been reported to be present in all parts of the shrub, and released to the surroundings to interfere with growth of unrelated plant species (Choyal and Sharma, 2011). Allelochemicals from *L. camara* had been investigated for nematicidal, termiticidal, insecticidal and repellent bioactivities (Kalita and Bhola, 2011).

Phytonematicides are depended on allelochemicals that are the active ingredients, which are naturally phytotoxic to plants. Phytotoxicity limits the use of phytonematicides as they induce high yield losses unintentionally (Mahmood *et al*., 1979). Phytotoxicity of phytonematicides may even lead to total plant losses of the protected plants (Satia *et al.*, 2007). *Lantana camara* had been reported to inhibit germination, growth, development or metabolism of neighbouring plants due to the secretion of allelochemicals (Qasem, 2006). There are less scientific studies on the use of fermented *L. camara* extracts for the suppression of root-knot nematodes and its phytotoxicity to tomato plants. The objective of this study was to determine whether the mean concentration stimulation point of *L. camara* extracts will be similar under greenhouse and micro-plot conditions on tomato seedlings.

3.2 Materials and methods

3.2.1 Experimental site and growth conditions

The study was conducted under greenhouse and microplot conditions at the Green Technologies Research Centre, University of Limpopo (23˚53’10’’S, 29˚44’15’’E) in the Summer/Autumn season of 2016. Under greenhouse, ambient day and night temperatures were averaged to 28˚C and 21°C, respectively, with maximum temperatures controlled using thermostatically activated fans. Other greenhouse variables, such as relative humidity, photosynthetically active radiation and solar radiation, were not measured.

Lantana camara was collected at maturity from ZZ2 fields, Mooketsi (23˚56'51''S, 30˚15'83''E). The above ground parts of *L. camara*, *viz*., shoots, leaves, seeds and flowers were washed and chopped into pieces while still fresh and fermented for 14 days in 20 L sealed containers having 15 L tapwater, 300 ml molasses, 100 g brown sugar and 200 ml effective micro-organism (EM). Prior to use, fermented *L. camara* product was stored at room temperature in sealed containers. Nematode inocula were prepared by extracting eggs and second-stage juveniles (J2) of *M. javanica* from roots of greenhouse grown nematode susceptible tomato plants cv. 'Money maker' by blending and maceration method for 30 seconds in 1% NaOCI (Hussey and Barker, 1973).

3.2.2 Experimental design and cultural practices

Twenty-cm diameter plastic pots were arranged at 0.3 m inter-row spacing and 0.25 m intra-row spacing, each filled with 1 800 ml steam-pasteurised sand mixed with Hygromix-T (Hygrotech, Pretoria North, South Africa) at 3:1 (v/v) ratio. Six-week-old tomato seedlings cv. 'Romanita' were transplanted and inoculated with 2 500 eggs and J2 *M. javanica* into 5-cm-deep holes around the base of each stem at five days after transplanting, whereas appropriate concentration of fermented *L. camara* were applied once weekly. Six treatments, *viz*. 0, 2, 4, 6, 8 and 10% fermented *L. camara* extracts were arranged in a randomised complete block design with ten replications.

Three days after transplanting, each plant was fertilised with 20 g multi-feed applied for two weeks followed by 3 g N-P-K 2:3:2 (22) to provide the total 186 mg N, 126 mg P

and 156 mg K per ml water. Plants were irrigated with 250 ml tapwater every other day. Plants were monitored for pest and disease incidence and when necessary, control measures were applied. Biomectin was applied at the rate of 0.6 l ha $^{-1}$ for the control of leafminer and whitefly, while Benomyl was applied at the rate of 0.8 l ha $^{-1}$ for the control of powdery mildew.

3.2.3 Data collection

At 56 days after inoculation, plant height, stem diameter, number of leaves and number of fruits per plant were measured. Plant height was measured from the soil surface to the tip of the flag leaf. Stems were cut off at the soil surface and the stem diameter was measured at 5 cm above the root area using a Vernier caliper. Roots were removed from pots, immersed in water to remove soil particles. Fruit yield, fresh shoots and roots were weighed. Shoots were oven dried at 70˚C for 72 hours and weighed. Soil sample per pot were collected, shade dried and 15 g soil mixed with 75 ml distilled water and shaken for 1 hour at 175 cycles per minute (cpm). The solution was filtered using Whatmann no. 42 into 100 ml beakers and electrical conductivity (EC) of the filtrates was measured with EC meter (model WTW CF 318) using Longenecker and Lyerlys (1964) method. Five grams soil sample was mixed with 25 ml of distilled water plus 75 ml of KCl for 50 minutes while being stirred and pH meter (model 420 A) was placed in a solution for 10 minutes prior to measuring the pH.

Roots were weighed to facilitate the calculation of nematode density/total roots/plant. Root galling was based on the scale of 0 to 5, in which 0 = no galls, 1 = 1 to 2 galls, 2 =

3 to 10 galls, 3 = 11 to 30 galls, 4 = 31 to 100 galls and 5 = >100 galls/root system (Taylor and Sasser, 1978). Nematodes were extracted from total root system/plant by maceration and blending for 30 seconds in 1% NaOCl (Hussey and Barker, 1973). The material was passed through nested 75- and 38 µm mesh sieves. The contents of the 38-µm mesh sieve were collected for further separation of nematodes from debris using the sugar-floatation and centrifugation method (Jenkins, 1964). Juveniles from root and soil samples were counted using a stereomicroscope.

3.2.4 Data analysis

Data was subjected to analysis of variance (ANOVA) through Statistix software. Mean of squares were partitioned, while treatment mean separation was achieved using Fisher LSD test then subjected to CARD computer based model to generate appropriate biological indices (Liu *et al*., 2003). Then, after adjusting R_h for D_m in plant variables, MCSP was computed by halving the sum of D_m and adjusted R_h. Also the CARD model was used to generate a regression curve estimations using the quadratic equation: $Y = b_2x + b_1x + a$, where Y = plant variable value and x computed from $x = -b_1/2b_2$, where x = the optimum concentration level, which is a concentration value where saturation sets in (Salisbury and Ross, 1992), along with the biological indices, *viz*. D_m, R_h, D_0, D_{50}, D_{100}, k and R^2 (Liu *et al*., 2003). Significant variables were further assessed using relative impact, which was expressed as: Relative impact (RI) = $(\frac{extract}{control} - 1) \times 100$, where the increase was depicted through positive (+) values and the decrease through negative (-) values.

3.3 Results

Treatments effects were significant for plant height, stem diameter, number of leaves and dry shoot mass from greenhouse experiment, while in microplot, conditions treatments effects were only significant for stem diameter, number of leaves, number of fruits, and fruits mass. Different levels of *L. camara* extracts contributed 65, 74, 61, 25 and 61% of plant height, stem diameter, number of leaves number of fruits and fruit mass in greenhouse, while in the microplot contributed 55, 80, 61, 65 and 85% plant height, stem diameter, number of leaves, number of fruits and fruits mass to the total treatment variation (TTV) respectively (Table 3.1 and 3.2).

Both greenhouse and microplot trials, treatments were significant in nematode numbers from roots, soil and final populations. *Lantana camara* extracts contributed 88, 94 and 92% in the greenhouse condition whereas in microplot condition contributed 94, 97 and 95% of nematodes in roots, soil and final population respectively to total treatment variations (Table 3.5 and 3.6). Different concentrations of *L. camara* in greenhouse condition reduced nematode number by 42-71, 40-73 and 46-72%, whereas under microplot condition by 8-85, 18-74 and 10-83% for root, soil and final nematode populations, respectively (Table 3.7).

Biological indices for stem diameter, number of leaves, fresh root mass, number of fruits and fruit weight produced by CARD model were strongly explained by the different levels *L. camara* extracts as shown by the coefficient of determination (R^2) at 70, 89, 76, 79 and 70%, respectively, under microplot conditions, while under green house plant

height, stem diameter and number of leaves were strongly explained by the different levels of *L. camara* extracts as shown by the R^2 at 81, 80 and 87% as the concentrations increased. Under microplot condition the overall sensitivity of tomato plants was 3 units, while under greenhouse was 0 units. The overall calculated MCSP for *L. camara* extracts for tomato were 5.76 and 5.31% in microplot and greenhouse trials, respectively (Table 3.9). Treatment effects were not significant for soil pH and EC in greenhouse and microplot trials. Total treatment variations in soil pH were 42 and 39% under greenhouse and microplot conditions, respectively, while EC were 41 and 31% from both growing condition, respectively (Appendices 3.23 and 3.24).

Graphic presentations of plant variables and concentrations of *L. camara* demonstrated that at low concentrations the material stimulated growth, while at high concentrations inhibition was observed. However, the difference was observed in stem diameter, where the opposite was observed.

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Table 3.1 Partitioning of mean of sum of squares for plant height (PH), stem diameter (SD), number of leaves (NL), number of fruits (NF) and fruit weight (FM) of tomato plant treated with different concentrations of *Lantana camara* extracts grown under greenhouse conditions.

		PH (cm)		SD (cm)		NL		NF		FM (g)	
Source	DF	MS	TTV (%)	MS	TTV (%)	MS	TTV (%)	MS	TTV (%)	MS	TTV (%)
Rep	9	90.53	19	1.05	13	1.22	13	1.62	25	35.32	12
Treatment	5	307.70	65**	5.72	74***	5.67	61*	1.64	25^{ns}	174.66	61^{ns}
Error	45	73.54	16	1.05	13	2.34	26	3.32	50	79.10	28.
Total	59	471.77	100	7.82	100	9.23	100	6.58	100	289.08	100

^{ns} Not significant at P≤ 0.05, *Significant at P ≤ 0.05, ***Highly significant at P ≤ 0.001.

Table 3.2 Partitioning of mean of sum of squares for plant height (PH), stem diameter (SD), number of leaves (NL), number of fruits (NF) and fruit mass (FM) of tomato plant treated with different concentrations of *Lantana camara* extracts grown under microplot conditions.

		PH (cm)		SD (cm)		NL		NF		FM (g)	
Source	DF	MS	TTV (%)	MS	TTV (%)	MS	TTV (%)	MS	TTV (%)	MS	TTV (%)
Rep	9	15.50	16	0.74	13	1.56	18	2.07	16	15.00	3
Treatment	5	53.12	55^{ns}	9.80	80^{***}	5.42	61^{*}	8.36	65^{*}	437.47	85^{***}
Error	45	28.69	29	0.84	7	1.89	21	2.50	19	61.38	12
Total	59	97.31	100	11.38	100	8.87	100	12.93	100	513.85	100

^{ns} Not significant at P≤ 0.05, * Significant at P ≤ 0.05, ***Highly significant at P ≤ 0.001.

Table 3.3 Partitioning of mean of sum of squares for dry shoot mass (DSW), Fresh root mass (FRW) and galling index (GI) of tomato treated with different concentrations of *Lantana camara* extracts grown under greenhouse condition.

		DSM (g)		FRM (g)		GI	
Source	DF	MS	TTV (%)	MS	TTV (%)	MS	TTV (%)
Block	9	2.57	18	39.35	39	0.60	12
Treatment	5	8.87	60*	35.68	35ns	3.72	77*
Error	45	328	22	25.33	25	0.54	11
Total	59	14.72	100	100.96	100	4.86	100

ns Not significant at P≤ 0.05, * Significant at P ≤ 0.05, *** Highly significant at P ≤ 0.001.

Table 3.4 Partitioning of mean of sum of squares for dry shoot mass (DSM), Fresh root mass (FRM) and galling index (GI) of tomato treated with different concentrations of *Lantana camara* extracts grown under microplot condition.

		DSM (g)		FRM (g)		GI	
Source	DF	MS	TTV (%)	MS	TTV (%)	MS	TTV (%)
Rep	9	3.10	35	5.97	5	0.23	8
Treatment	5	2.57	29ns	86.05	79*	1.98	70*
Error	45	3.20	36	17.63	16	0.64	22
Total	59	8.87	100	100.96	100	2.85	100

ns Not significant at P≤ 0.05, * Significant at P ≤ 0.05, *** Highly significant at P ≤ 0.001.

Table 3.5 Partitioning of mean of sum of squares for final nematode population density (Pf) as affected by different concentrations of fermented *Lantana camara* under greenhouse conditions.

		Pf/total root		Pf/ total soil		Pf	
Source	DF	MS	TTV (%)	MS	TTV (%)	MS	TTV (%)
Rep	9	11831	8	472.2	3	12461	5
Treatment	5	131334	88***	12879.5	94***	225883	92***
Error	45	6615	4	385.2	3	7744	3
Total	59	149780	100	13736	100	246088	100

ns Not significant at P≤ 0.05, * Significant at P ≤ 0.05, *** Highly significant at P ≤ 0.001.

Table 3.6 Partitioning of mean of sum of squares for final nematode population density (Pf) as affected by different concentrations of fermented *Lantana camara* under microplot conditions.

		Pf/total root		Pf/ total soil		Pf	
Source	DF	MS	%	MS	%	MS	%
Rep	9	1729	3	141.96	2	16591	3
Treatment	5	403715	94***	7960.55	97***	516882	95***
Error	45	11714	3	74.24	1	11982	2
Total	59	432728	100	8176.75	100	545455	100

ns Not significant at P≤ 0.05, * Significant at P ≤ 0.05, *** Highly significant at P ≤ 0.001.

Table 3.7 Relative impact (%) effects of different concentrations fermented *Lantana camara* on final nematode population density under greenhouse conditions.

Extract level	Pf/total root nematode	RI (%)	Pf/total soil nematode	RI (%)	Pf nematodes	RI (%)
0	432.50[a]	–	143.40[a]	–	575.10[a]	–
2	228.50[b]	–42	86.20[a]	–40	314.70[b]	–46
4	193.70[b]	–56	76.30[bc]	–47	270.00[bc]	–54
6	121.30[b]	–72	60.70[bcd]	–58	229.50[bc]	–61
8	182.10[b]	–58	59.50[cd]	–59	182.00[c]	–69
10	126.30[b]	–71	39.90[d]	–73	166.20[c]	–75

[y]Column with means with the same letter were not different (P≤ 0.05) according to Fishers Least Significant Test.

[z]Relative impact (%) = [(extract/control) – 1) × 100].

Table 3.8 Relative impact (%) effects of different concentrations of fermented *Lantana camara* on final nematode population density under microplot conditions.

Extract level	Pf/total root nematode	RI (%)	Pf/total soil nematode	RI (%)	Pf nematode	RI (%)
0	531.80[a]	–	143.40[a]	–	675.20[a]	–
2	523.80[a]	–8	86.20[a]	–40	613.30[a]	–10
4	266.30[b]	–53	76.30[bc]	–47	346.00[b]	–49
6	175.90[bc]	–69	60.70[bcd]	–58	236.20[bc]	–65
8	161.60[c]	–72	59.50[cd]	–59	217.70[bc]	–68
10	87.60[c]	–85	39.90[d]	–73	116.1[c]	–85

[y]Column with means with the same letter were not different (P≤ 0.05) according to Fishers Least Significant Test.

[z]Relative impact (%) = [(extract/control) – 1) × 100].

Table 3.9 Biological indices and mean dosage stimulation range of , stem diameter (SD), Number of leaves (NL), fresh root mass (FRM), number of fruit (NF), fruit mass (FM) and Plant height of tomato seedlings exposed to different concentrations of fermented *Lantana camara* at 56 days after transplanting.

	Microplot						Greenhouse			
Biological Index	**SD**	**NL**	**FRM**	**NF**	**FM**	**Mean**	**PH**	**SD**	**NL**	**Mean**
Threshold stimulation (D_m)	6.314	1.051	4.936	4.806	2.372	3.895	5.018	1.709	4.12	3.905
Saturation point (R_h)	1.675	1.344	4.927	1.701	9.035	3.736	5.668	0.073	-0.455	5.286
0% Inhibition (D_0)	0	6.043	9.873	9.612	10.37	7.180	10.036	3.417	8.24	7.231
50% Inhibition (D_{50})	0	100.629	12.971	13.071	23.381	30.010	18.366	14.501	19.72	17.529
100% Inhibition (D_{100})	0	0	15.2	15.5	40.9	14.320	23.2	19.070	25.8	22.69
R^2	0.697	0.887	0.758	0.789	0.695	0.765	0.806	0.798	0.865	1.235
Sensitive index (k)	0	2	0	0	1		0	0	0	
Total plant sensitivity		Σk= 3						Σk=0		
Mean concentration stimulation point (MCSP)										
Threshold stimulation (D_m)	6.314	1.051	4.936	4.806	2.372	3.896	5.943	1.709	4.062	3.905
Adjusted saturation point(R_h)^y	7.989	2.395	9.863	6.507	11.407	7.632	14.69 1	1.782	3.668	6.714
						MCSP = 5.76				MCSP = 5.31

Adjusted $R_h = D_m + R_h$, while MCSP = $D_m + (R_h/2)$

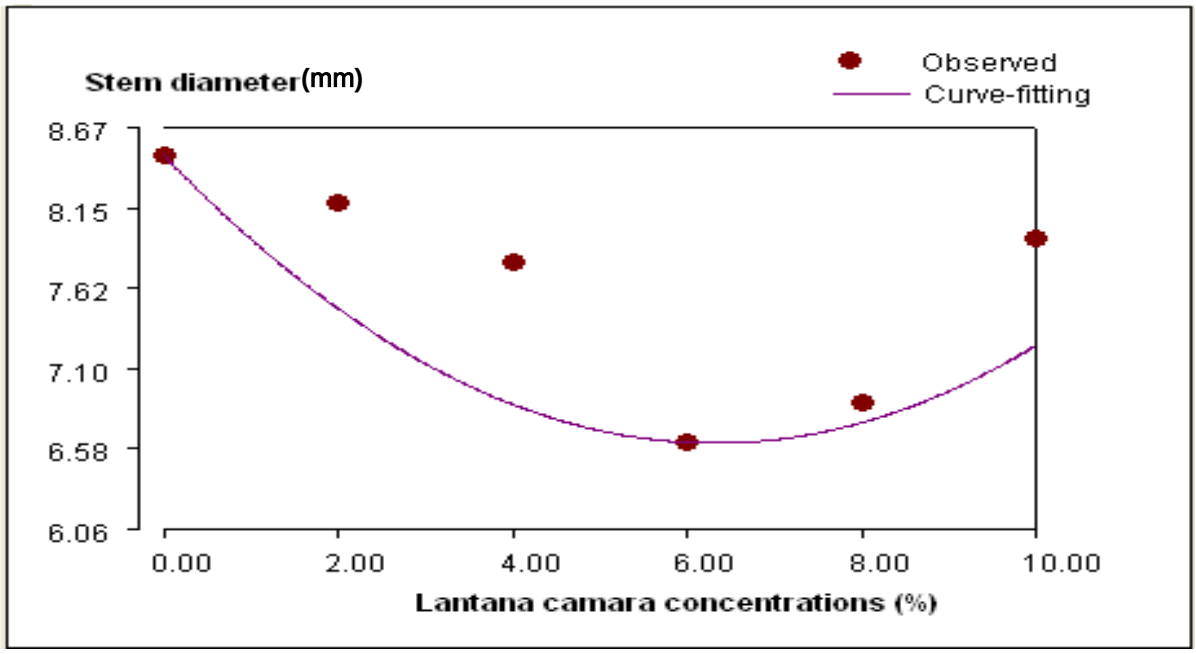

Figure 3.1 Response of stem diameter to concentrations of fermented *Lantana camara* extracts of at 56 days after initiating treatments under microplot condition.

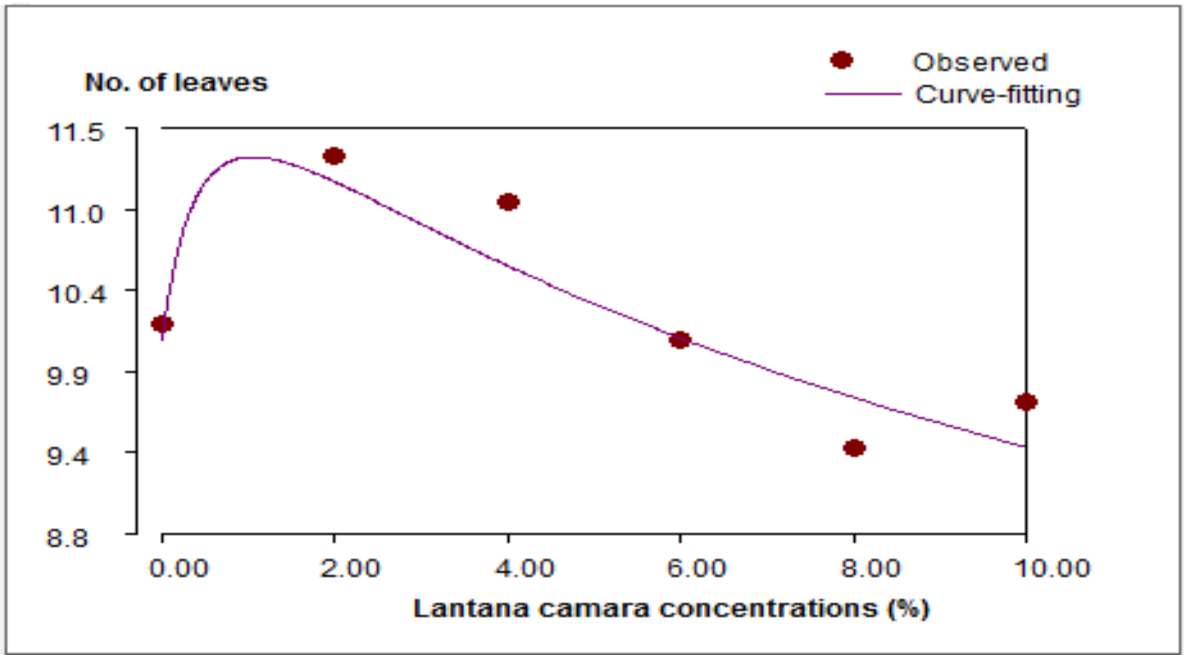

Figure 3.2 Response of number of leaves to concentrations of fermented *Lantana camara* extracts of at 56 days after initiating treatments under microplot condition.

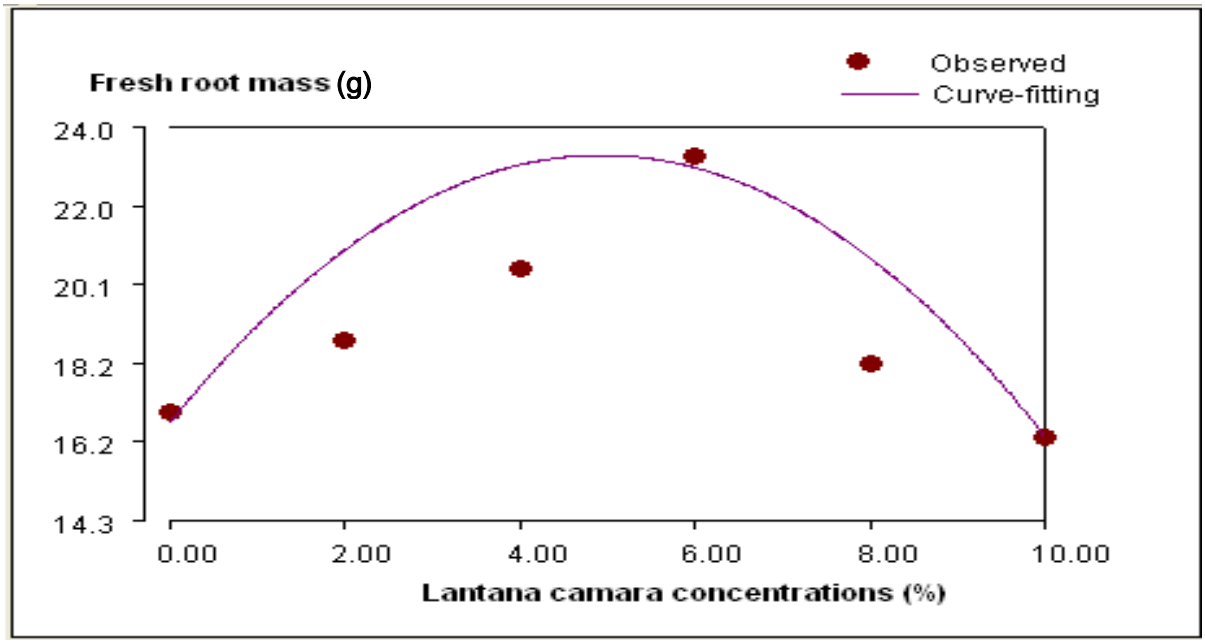

Figure 3.3 Response of fresh root mass to concentrations of fermented *Lantana camara* extracts of at 56 days after initiating treatments under microplot condition.

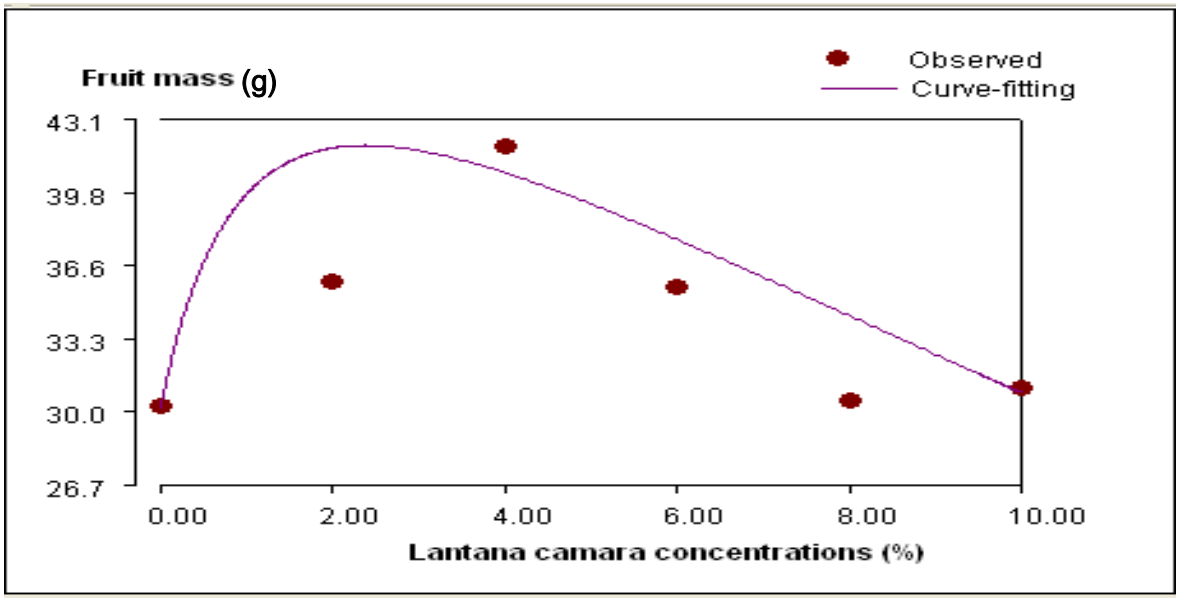

Figure 3.4 Response of fruit mass to concentrations of fermented *Lantana camara* extracts of at 56 days after initiating treatments under microplot condition.

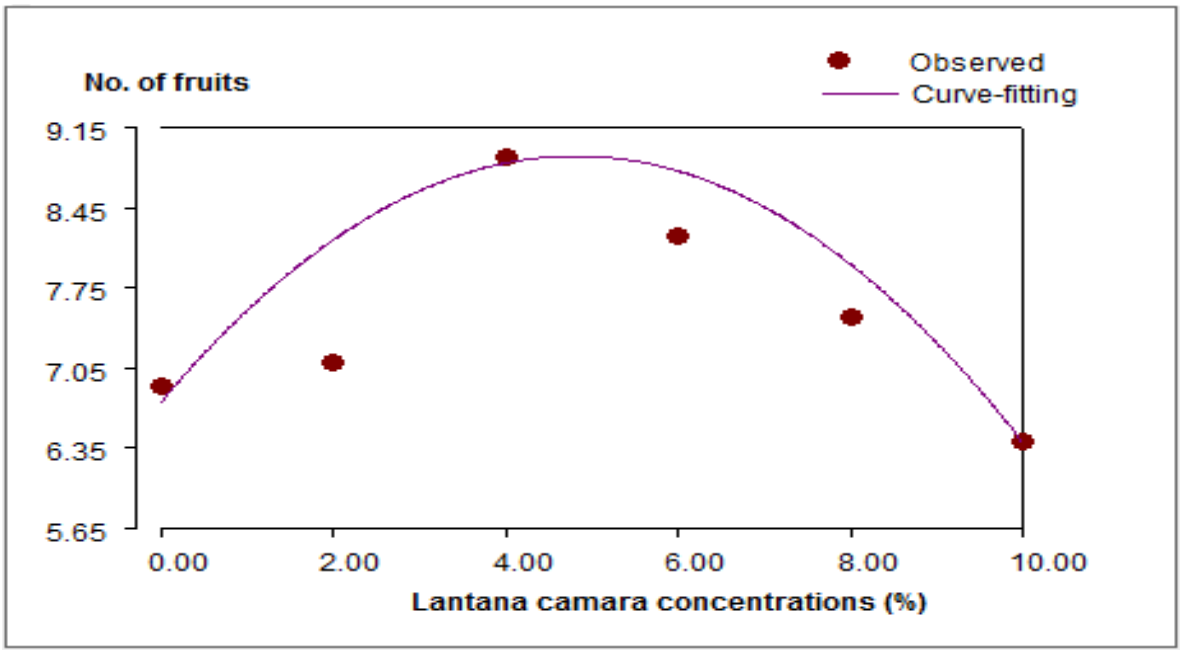

Figure 3.5 Response of number of fruits to concentrations of fermented *Lantana camara* extracts of at 56 days after initiating treatments under microplot condition.

Table 3.10 Quadratic relationships, coefficient of determination and computed optimum response concentration for variables of tomato from the Curve-fitting Allelochemical Response Dosage against different concentrations of fermented *Lantana camara* extracts at 56 days after treatments under microplot conditions.

Plant variables	Quadratic relationship	R^2	$(X)^z$	P ≤
Stem diameter	$-0.420x^2+0.531x+8.764$	0.697	0.632	0.01
No of leaves	$-4.586x^2+4.965x+10.200$	0.887	0.541	0.05
Fresh root mass	$-0.204x^2+2.015x+16.661$	0.758	4.939	0.01
No of fruits	$-0.074x^2+0.708x+0.074$	0.865	4.784	0.01
Fruit mass	$-0.612x^2+14.867-29.799$	0.695	12.156	0.01

[z]Calculated optimum response concentration (x) = $-b_1/2b_2$

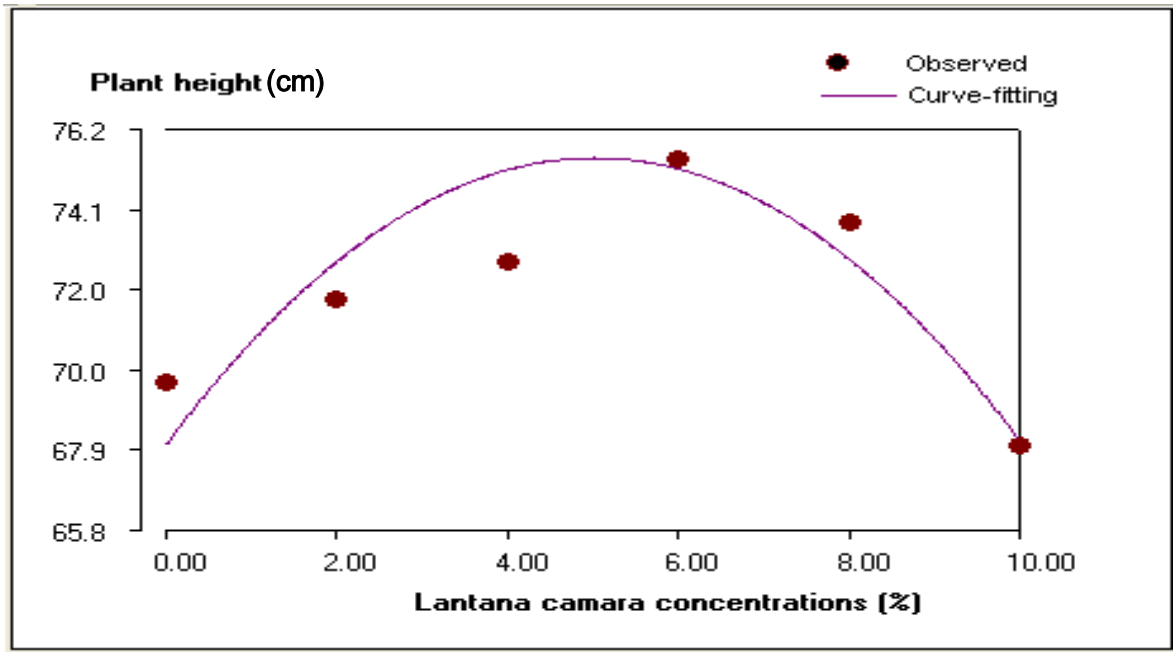

Figure 3.6 Response of plant height to concentrations of fermented *Lantana camara* extracts of at 56 days after initiating treatments under greenhouse condition.

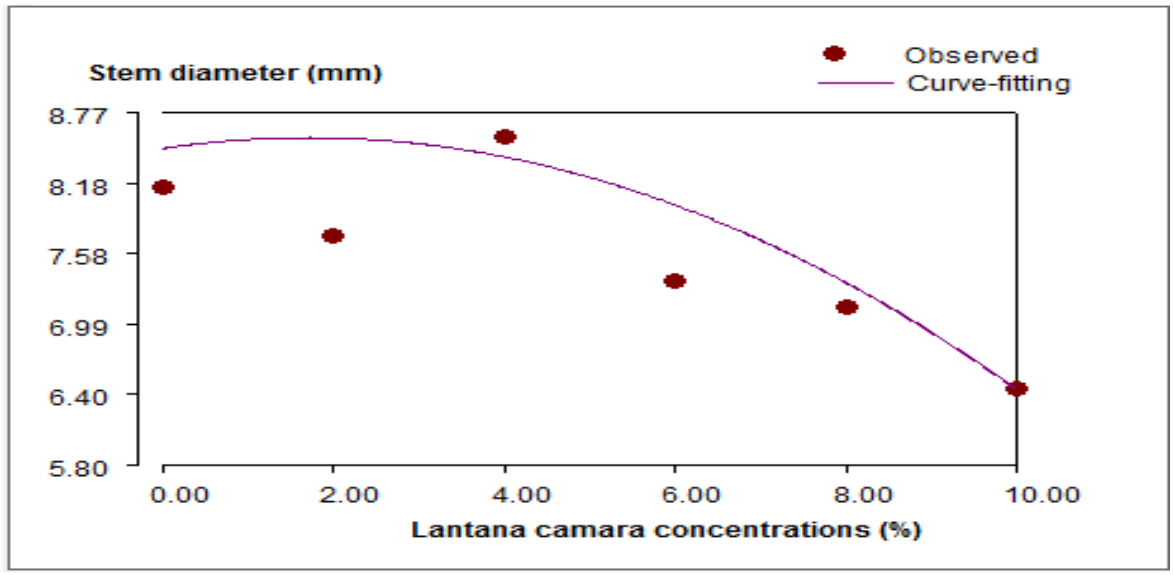

Figure 3.7 Response of stem diameter to concentrations of fermented *Lantana camara* extracts of at 56 days after initiating treatments under greenhouse condition.

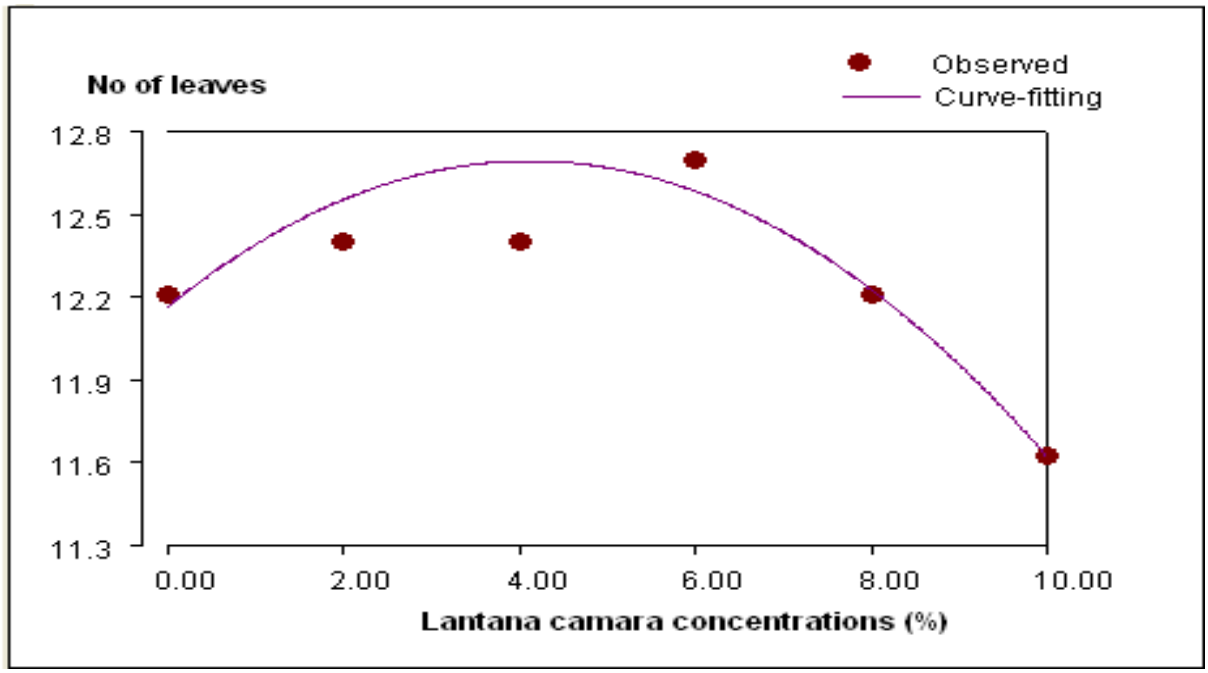

Figure 3.8 Response of number of leaves to concentrations of fermented *Lantana camara* extracts of at 56 days after initiating treatments under greenhouse condition.

Table 3.11 Quadratic relationships, coefficient of determination and computed optimum response concentration for variable of tomato from the Curve-fitting Allelochemical Response Dosage against different concentrations of fermented *Lantana camara* extracts at 56 days after treatments under greenhouse conditions.

Plant variables	Quadratic relationship	R^2	$(X)^z$	P ≤
Plant height	$-0.224x^2+2.253x$ 68.825	0.806	5.029	0.01
Stem diameter	$-0.025x^2+0.086x + 8.050$	0.798	1.72	0.01
No of leaves	$-0.027x^2+ 0.227x + 12.129$	0.895	4.093	0.05

[z]Calculated optimum response concentration (x) = $-b_1/2b_2$

3.4 DISCUSSION

3.4.1 Effects on tomato plant growth

The fact that some of assessed plant organs in this study were not affected by the increasing concentrations of the phytonematicides, suggested that the organs were concentration saturated at harvest (Mashela *et al*., 2015). Similar observation were made using *Cucumis* species in tomato plants (Pelinganga *et al*., 2013), geranium (Sithole *et al*., 2016) and *M. incognita* J2 hatch trials (Dube and Mashela, 2016). From the two conducted studies, treatment effect were not significant on number of leaves and fruit mass under greenhouse conditions and plant height under microplot conditions. Mashela *et al*. (2015), indicated that when concentrations were within the stimulation range, growth is invariably stimulated, with the neutral range having no effect whereas within the inhibition range, growth is inhibited.

High coefficient of determination (R^2) for CARD model in different plant variables suggested strong density-dependent relationship between tomato plant growth and increasing concentrations of *L. camara* extracts. Different k values in all organs demonstrated that in tomato plants, the assessed organs had different sensitivities to concentrations of allelochemicals from fermented *L. camara* extracts. Plant sensitivity is indirectly proportional to k values, with zero suggesting the highest sensitivity to allelochemicals used, while high k values suggested decreased sensitivities (Liu *et al*., 2003). Increasing concentrations of *L. camara* extracts from 2 to 10% were less phytotoxic when tomato plants were exposed to environmental condition as shown by the overall sensitivity ranking (k-value) of three, while under greenhouse conditions the

overall sensitivity was zero. This suggested that the material could be volatile under varying temperature conditions as shown by difference sensitivity of tomato plants exposed to two different growing conditions.

Crude extracts of fermented *L. camara* is currently used as the main product for suppression of plant-parasitic nematode in tomato production. The product is applied on weekly basis at 3.68% (Daneel *et al*., 2014). In this study, the computed MCSP values for products are 5.76 and 5.31% which could be used in field and greenhouse production, respectively. The derived MCSP are higher than those that are currently being used in their production systems. In contrast (Storey, 2012), indicated that the use of the product at 6.5% was too high as seen by phytotoxic effect on roots with no observed phytotoxicity on the aerial parts.

3.4.2 Curve-fitting Allelochemical Response Dosage

The overall calculated MCSP for microplot and greenhouse study were 5.76 and 5.31%, respectively. The findings of this study were different from the observations of Pelinganga (2013) using phytonematicides made from *Cucumis* species, were the derived MCSP of tomato plants was 2.63 and 2.98%. Plants have different respond to different concentrations of phytonematicides. The CARD model demonstrated the importance of choosing appropriate concentrations in phytotoxicity studies as described elsewhere (Mamphiswana *et al*., 2010; Pelinganga, 2013). Mamphiswana *et al*. (2010) reported that when concentrations are already above the saturation point (R_h), the relation between plant growth and increasing concentrations of FPE would be depicted

by a negative linear relationship, while that before R_h would be described by positive linear relationships.

The MCSP value, which is empirically based on a series of phytonematicide concentrations, should be interpreted alongside the overall k-value of the plant to the test phytonematicide (Mashela *et al*., 2015). In this study similar k value was observed in stem diameter, fresh root mass and number of fruits under microplot conditions whereas in greenhouse conditions was only seen on plant height, stem diameter and number of leaves which was equal to zero. The findings of this study are different from the observation of Pelinganga *et al*. (2012), who reported high k value on dry shoot mass, plant height and stem diameter under dilutions from fresh fermented *C. africanus* fruit. Furthermore, in the Ground Leaching Technology (GLT), Mafeo (2012), showed that k values of tomato seedlings ranged from 9 to 20 depending on the investigated organ. According to Pelinganga and Mashela (2012) the k values are affected by various factors, which may include fermentation of dried versus fresh materials, fermented versus unfermented, age of the test plant and/or organ of the test plant.

In the current study, the CARD model indicated the degree of sensitivity of various organs of tomato plants when exposed to different concentration of *L. camara* extracts. Among the two studies conducted, high total plant sensitivity was observed in plant grown under greenhouse condition in comparison to those grown in microplot. This could have been attributed by the different environmental conditions that plants were exposed. Using crude extracts of *C. myriocarpus* as pre-emergent nematicide, Mafeo

(2012) demonstrated that different organs within the same plant species have different sensitivities which may be extended to other extrinsic factors. This was in accordance with the findings of this study that showed that exposing tomato plants to environmental conditions have different sensitivity to increasing concentrations of *L. camara* extracts.

The optimum concentration derived from the quadratic relationships for plant variables were positive, suggesting the positive linear relationship between the variables and increasing concentration of *L. camara* extracts. According to Pelinganga *et al*. (2011); a positive linear relationship between variables and increasing concentrations of a particular phytonematicides, suggested that the concentrations used were within the stimulation range while a negative linear relationship suggest that the concentration of allelochemical were already in inhibition range. Using the CARD model, the variables and concentrations of the extracts are generally characterised by quadratic relationship (Salisbury and Ross, 1992). The DDG patterns between growth variables of tomato plants and increasing concentrations of *L. camara* extracts was depicted through high coefficient.

3.4.3 Effects on nematode population density

The inhibitory effect of *L. camara* extracts on nematode population densities was higher in roots while in the soil was low, suggesting that the material was able to penetrate the plant roots. It has been reported that, the inhibitory effect of plant extracts is due the chemicals properties that are present in the extract that possess nematicidal properties (Agbenin *et al.,* 2005). According to Adegbite and Adesiyan (2005), botanicals with

nematicidal properties affect the embryonic development or kill the eggs. The reduction of root-knot nematodes could be attributed to poor root penetration and later retardation of activities such as feeding and reproduction (Bunt, 1975).

The increase in concentration of fermented *L. camara* extract reduced population densities of *M. javanica* on tomato plants, that's the nematicidal effect of fermented *L. camara* on mortality of *M. javanica* was concentration dependent. Mahmood *et al*. (1979), reported that juvenile mortality decreases with increased extract concentration as the efficacy of the plant extract depends on the concentration and duration of exposure of juveniles to the extract. Plants and microorganisms respond to increasing concentrations of allelochemicals in accordance to the density-dependent growth (DDG) patterns (Liu *et al.,* 2003). The DDG patterns are an advanced modification of the 1933 Nicholson's carrying capacity model, which had been adapted and used in various disciplines for enhancing decision-making (Mashela *et al*., 2015).

Nematodes, particularly J1 and J2 have unique survival strategies (Mashela, 2007), therefore it would not be possible to wipe-out plant parasitic nematodes from plant production system. When J1 and J2 are being exposed to adverse environmental conditions like those of synthetic nematicides and botanicals extracts, nematodes in the two stages enter dauer and cryptobiotic stages, respectively (Mashela, 2007). In these stages the juveniles have reduced metabolic rate and extended survival periods of exposure, but when the unpleasant situations becomes better the juveniles exit the survival stages and assume normal activities (Pelinganga and Mashela, 2012).

Van Gundy and McKenry (1975) described synthetic fumigants as biocidal, meaning that the chemicals kill all forms of life in soil, while non-fumigant was nematostatic. The latter means that synthetic non fumigant nematicides did not kill nematodes, but modified behaviours such as failure to detect chemical cues required in nematode infection of new roots, disruption of development and restriction of reproduction capabilities, as observed for botanicals. Incidentally, all nemastatic properties appear to have the same end result of slowing down increases in final nematode population densities, and therefore initial population densities for subsequent crops, which is all that matters in plant protection (Seinhorst, 1965).

3.4.4 Effects on soil pH and EC

McLean and Lawrence (2000) reported that plant extracts that reduces soil pH are not suitable since they will results in unavailability of alkaline loving nutrients elements. Development of phytonematicide using fever tea (*Lippia javanica)* had to be discontinued due their effect on reduction soil pH (Mashela *et al*., 2010). On contrary, the use of fermented *L. camara* as phytonematicides had no negative effect on the soil pH and EC.

CHAPTER 4
SUMMARY, SIGNIFICANCE OF FINDINGS, RECOMMENDATION AND CONCLUSION

4.1 Summary

In this study, different levels of *L. camara* extracts contributed 65, 74, 61, 43 and 61% of plant height, stem diameter, number of leaves and dry shoot mass in greenhouse, while in the microplot contributed 86, 61, 65, 85 and 79% stem diameter, number of leaves, number of fruits, fruits mass and fresh shoot mass to the total treatment variation (TTV) respectively. Under microplot condition the overall sensitivity of tomato plants was 3 units, while under greenhouse was 0 units. The overall calculated MCSP for *L. camara* extracts for tomato were 5.76 and 5.31% in microplot and greenhouse conditions, respectively. Under greenhouse conditions, nematode numbers were reduced by 42-71, 40-73 and 46-72%, whereas under microplot condition by 8-85, 18-74 and 10-83% for root, soil and final nematode populations, respectively. However, different levels of *L. camara* extracts had no effect on soil pH and EC.

4.2 Significance of findings

Phytonematicides are often toxic to the plants to be protected against nematodes due to high concentrations used in production systems (Mashela *et al*., 2015). Mashela *et al*. (2015) indicated that the CARD model could provide the biological indices that could be used in computing the MSCP. The findings of this study added knowledge on the use of fermented *L. camara* extracts in tomato production in management of *M. javanica* population densities. The derived MCSP of the product would aid in avoiding phytotoxicity of the material to tomato plants while increasing the growth of the plant.

4.3 Recommendations

Currently, at ZZ2 3.68% *L. camara* extracts is being used. The derived MCSP values should be used to establish the application interval of *L. camara* extracts on tomato plants, followed by the dosage model (Mashela *et al*., 2015). The dosage model would eventually allow for tracking the persistence of the active ingredients in the soil. Also, the potential chemical residue of active ingredients responsible for nematode suppression should be assessed in fruit of tomato.

4.4 Conclusions

Meloidogyne species can be managed using fermented *L. camara* extracts at 5.31 and 5.76% when tomato plants are grown under greenhouse and field production systems, respectively. The derived MCSP of fermented *L. camara* extracts would help in managing *Meloidogyne* species with no phytotoxicity of the material to the protected plants. Furthermore, the findings of this study provide the information on efficacy of fermented *L. camara* extracts against population densities of *Meloidogyne* species identified.

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APPENDICES

Appendix 3.1 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on plant height of tomato plant grown under greenhouse condition.

Source	DF	SS	MS	F value	P value
Rep	9	814.77	90.530		
Treatment	5	1535.52	307.703	4.18	0.0033
Error	45	3309.28	73.540		
Total	59	5662.57	471.773		

Appendix 3.2 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on plant height of tomato plant grown under microplot condition.

Source	DF	SS	MS	F value	P value
Rep	9	139.50	15.4997		
Treatment	5	265.61	53.1220	1.85	0.1220
Error	45	1291.09	28.6910		
Total	59	1696.20	97.3127		

Appendix 3.3 Analysis of variance (ANOVA) for different concentration of *Lantana camara* extracts on stem diameter of tomato plant grown under greenhouse conditions.

Source	DF	SS	MS	F value	P value
Rep	9	9.4236	1.04707		
Treatment	5	28.5862	5.71725	5.45	0.0005
Error	45	47.1911	1.04869		
Total	59	85.2010	7.81301		

Appendix 3.4 Analysis of variance (ANOVA) for different concentration of *Lantana camara* extracts on stem diameter of tomato plant grown under microplot conditions.

Source	DF	SS	MS	F value	P value
Rep	9	6.6489	0.73877		
Treatment	5	48.9965	9.79929	11.72	0.0000
Error	45	37.6134	0.83585		
Total	59	93.2588	11.37391		

Appendix 3.5 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on number of leaves of tomato plant grown under greenhouse conditions.

Source	DF	SS	MS	F value	P value
Rep	9	11.017	1.22407		
Treatment	5	28.350	5.67000	5.42	0.0503
Error	45	105.483	2.34407		
Total	59	114.850	9.23814		

Appendix 3.6 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on number of leaves of tomato plant grown under microplot conditions.

Source	DF	SS	MS	F value	P value
Rep	9	14.017	1.55741		
Treatment	5	27.083	5.41667	2.86	0.0250
Error	45	85.083	1.89074		
Total	59	126.183	8.86482		

Appendix 3.7 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on number of fruits for tomato plant grown under greenhouse conditions.

Source	DF	SS	MS	F value	P value
Rep	9.	14.600	1.62222		
Treatment	5	8.200	1.64000	0.49	0.7800
Error	45	149.800	3.32889		
Total	59	172.600	6.59111		

Appendix 3.8 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on number of fruits for tomato plant grown under microplot conditions.

Source	DF	SS	MS	F value	P value
Rep	9	18.667	2.07407		
Treatment	5	41.800	8.3600	3.34	0.0119
Error	45	112.533	2.50074		
Total	59	173.000	12.93481		

Appendix 3.9 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on fruit mass of tomato plant grown under greenhouse conditions.

Source	DF	SS	MS	F value	P value
Rep	9	317.84	35.316		
Treatment	5	873.29	174.659	2.21	0.0700
Error	45	3559.38	79.097		
Total	59	4750.51	289.072		

Appendix 3.10 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on fruit mass of tomato plant grown under microplot conditions.

Source	DF	SS	MS	F value	P value
Rep	9	134.97	14.996		
Treatment	5	2187.34	437.468	7.13	0.0001
Error	45	2761.86	61.375		
Total	59	5084.17	513.839		

Appendix 3.11 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* on dry shoot mass of tomato plant grown under greenhouse conditions.

Source	DF	SS	MS	F value	P value
Rep	9	23.133	2.57028		
Treatment	5	44.348	8.86956	2.70	0.0321
Error	45	147.559	3.27909		
Total	59	215.039	14.71893		

Appendix 3.12 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* on dry shoot mass of tomato plant grown under microplot conditions.

Source	DF	SS	MS	F value	P value
Rep	9	27.871	3.09677		
Treatment	5	12.849	2.56985	0.80	0.5543
Error	45	144.213	3.20472		
Total	59	184.933	8.87134		

Appendix 3.13 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on fresh root mass of tomato plant grown under greenhouse conditions.

Source	DF	SS	MS	F value	P value
Rep	9	359.56	39.9506		
Treatment	5	178.41	35.6813	1.41	0.2395
Error	45	1140.07	25.3348		
Total	59	1678.03	100.9667		

Appendix 3.14 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on fresh root mass of tomato plant grown under microplot conditions.

Source	DF	SS	MS	F value	P value
Rep	9	53.73	5.9702		
Treatment	5	430.24	86.0480	4.88	0.0012
Error	45	793.51	17.6336		
Total	59	1277.48	109.6518		

Appendix 3.15 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on number of root galls of tomato plant grown under greenhouse condition.

Source	DF	SS	MS	F value	P value
Rep	9	5.4000	0.6000		
Treatment	5	18.6000	3.72000	6.86	0.0001
Error	45	24.4000	0.54222		
Total	59	48.4000	4.86222		

Appendix 3.16 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on number of root galls of tomato plant grown under microplot condition.

Source	DF	SS	MS	F value	P value
Rep	9	2.1500	0.23889		
Treatment	5	9.8833	1.97667	3.07	0.0181
Error	45	28.9500	0.64333		
Total	59	40.9833	2.85889		

Appendix 3.17 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on nematodes numbers in roots of tomato plant grown under greenhouse conditions.

Source	DF	SS	MS	F value	P value
Rep	9	106475	11831		
Treatment	5	656668	131334	19.85	0.0000
Error	45	297661	6615		
Total	59	1060804	149780		

Appendix 3.18 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on nematodes numbers in roots of tomato plant grown under microplot conditions.

Source	DF	SS	MS	F value	P value
Rep	9	155693	17299		
Treatment	5	2018577	403715	34.47	0.0000
Error	45	527113	11714		
Total	59	2701383	432728		

Appendix 3.19 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on nematodes numbers in soil of tomato plants grown under greenhouse conditions.

Source	DF	SS	MS	F value	P value
Rep	9	4249.7	472.2		
Treatment	5	64397.7	12879.5	33.44	0.0000
Error	45	17333.9	385.2		
Total	59	85981	13736.9		

Appendix 3.20 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on nematodes numbers in soil of tomato plants grown under microplot conditions.

Source	DF	SS	MS	F value	P value
Rep	9	1277.6	141.96		
Treatment	5	39802.7	7960.55	107.23	0.0000
Error	45	3340.6	74.24		
Total	59	44420.9	8176.75		

Appendix 3.21 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on nematode numbers in roots + soil of tomato plant grown under greenhouse conditions.

Source	DF	SS	MS	F value	P value
Rep	9	112153	12461		
Treatment	5	1129417	225883	29.17	0.0000
Error	45	348492	7744		
Total	59	1590063	246088		

Appendix 3.22 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on nematode numbers in roots + soil of tomato plant grown under microplot conditions.

Source	DF	SS	MS	F value	P value
Rep	9	149323	16591		
Treatment	5	2584408	516882	43.14	0.0000
Error	45	539191	11982		
Total	59	3272923	545455		

Appendix 3.23 Partitioning of mean of sum of squares of tomato plant treated with different concentrations of *Lantana camara* on soil pH and soil electrical conductivity (EC) under greenhouse condition.

		pH		EC	
Source	DF	MS	TTV (%)	MS	TTV (%)
Rep	9	0.06	32	8.96	34
Treatment	5	0.08	42^{ns}	10.68	41^{ns}
Error	45	0.05	26	6.62	25
Total	59	0.19	100	26.26	100

^{ns} Not significant at P≤ 0.05, * Significant at P ≤ 0.05, *** Highly significant at P ≤ 0.001

Appendix 3.24 Partitioning of mean of sum of squares of tomato plant treated with different concentrations of *Lantana camara* on soil pH and soil electrical conductivity (EC) under microplot condition.

		pH		EC	
Source	DF	MS	TTV (%)	MS	TTV (%)
Rep	9	0.33	33	57.74	36
Treatment	5	0.38	39^{ns}	49.64	31^{ns}
Error	45	0.28	28	52.25	33
Total	59	0.99	100	159.63	100

^{ns} Not significant at P≤ 0.05, * Significant at P ≤ 0.05, *** Highly significant at P ≤ 0.001

Appendix 3.25 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on soil pH of tomato plant grown under greenhouse condition.

Source	DF	SS	MS	F value	P value
Rep	9	0.53945	0.05994		
Treatment	5	0.42197	0.08439	1.80	0.1329
Error	45	2.11394	0.04698		
Total	59	3.07536	0.19131		

Appendix 3.26 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on soil pH of tomato plant grown under microplot condition.

Source	DF	SS	MS	F value	P value
Rep	9	3.0051	0.33390		
Treatment	5	1.8742	0.37484	1.35	0.2624
Error	45	12.5273	0.27839		
Total	59	17.4067	0.98713		

Appendix 3.27 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on soil electrical conductivity of tomato plant grown under greenhouse condition.

Source	DF	SS	MS	F value	P value
Rep	9	80.594	8.9549		
Treatment	5	53.416	10.68.32	1.61	0.1761
Error	45	298.074	6.6239		
Total	59	432.084	26.262		

Appendix 3.28 Analysis of variance (ANOVA) for different concentrations of *Lantana camara* extracts on soil electrical conductivity of tomato plant grown under microplot condition.

Source	DF	SS	MS	F value	P value
Rep	9	519.61	57.7345		
Treatment	5	248.21	49.6419	0.95	0.4583
Error	45	2351.35	52.2522		
Total	59	3119.17	159.6286		