

Full Length Research Paper

Integrated management of *Meloidogyne incognita* in tomato (*Solanum lycopersicum*) through botanical and intercropping

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Received 19 March, 2019; Accepted 25 April, 2019

Root-knot nematode, *Meloidogyne incognita*, is the major limiting factor in tomato production in many regions of the world, including Ethiopia. Hence, there is a need for development of root-knot nematode management methods that are cheap and environmentally friendly. A greenhouse experiment was conducted with an objective to evaluate the integrated effect of botanicals and intercropping against *M. incognita* on tomato. The experiment was laid out in a 2x3x3 factorial completely randomized design (CRD) with four replications. About 30 ml of lantana leaf extract (at 5% v/v concentration) was applied one week before transplanting of convert to days tomato cv Moneymaker seedlings into 20 cm diameter size pot containing 2 kg sterilized soil. Seeds of mustard and garlic were sown directly into the plastic pots on either side of the tomato seedlings. *M. incognita* was inoculated at the rate of 1000 and 2000 second-stage juveniles (J2) per pot one week after transplanting. A control without any nematode treatment was also maintained. The results revealed that combination of lantana leaf extract and tomato-mustard intercropping at both nematode inoculum levels proved to be the most effective treatment that reduced the soil and root population of the nematode. Application of lantana leaf extract alone and in combination at both inoculation levels showed superiority on tomato growth characteristics. Hence, this ecofriendly approach could be incorporated into integrated nematode management in tomato. However, further research is needed to evaluate their efficacies under field conditions.

Key words: Egg-mass, gall, garlic, juvenile, *Lantana camara*, mustard, management.

INTRODUCTION

Tomato (*Solanum lycopersicum*) is one of the most popular vegetables worldwide, owing to its high nutritive

value and diversified use. Tomato is rich in minerals, vitamins, essential amino acids, sugars and dietary fibers.

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Ripped tomato fruit has high nutritive value and it is a good source of vitamins and minerals (USDA, 2005). Red tomato contains lycopene, an anti-oxidant that may contribute to protection against carcinogenic substances and which is known to prevent prostate cancer and improves the skin's ability to guard against harmful ultraviolet radiation (Rao and Rao, 2007). As it is a relatively short duration crop and gives a high yield, it is economically attractive. Area under world tomato cultivation is ever increasing. Over 170 million tons of fresh tomato was produced in the world in 2017 (FAOSTAT, 2018). In Ethiopia, tomato is an important cash crop to farmers and a widely cultivated vegetable crop both under irrigation and rain fed throughout the year (Lemma, 2002). Jiregna et al. (2012) reported as tomato ranking 8th in annual national production.

The quest for increased production is often constrained by biotic and abiotic factors. Plant parasitic nematodes (PPN) constitute one of the biotic factors negatively influencing increased tomato production. The latest statistics showed that the estimated losses induced by PPN worth US\$ 157 billion worldwide (Singh et al., 2015). There are over 4,100 species of PPN described to date (Decraemer and Hunt, 2009). Of these nematode species, the most economically important PPN genus is root-knot nematode (RKN) (*Meloidogyne* species) (Jones et al., 2013). This genus alone consists of more than 101 described species until August 2015 (Seid et al., 2015). The genus consists of extremely polyphagous nematode groups because it has more than 3000 host species, including vegetables, fruit trees, oil, fiber, grains and leguminous crops, next to weeds that are considered secondary hosts to nematodes (Khalil, 2013). These nematode species parasitize almost every species of vascular plant (Jones et al., 2013). The most well-known species of RKN include *Meloidogyne incognita*, *Meloidogyne javanica*, *Meloidogyne arenaria* and *Meloidogyne hapla*, which are responsible for high economic losses to varied crops. Average crop yield losses due to nematode are estimated to be in the neighborhood of 25% with damage in individual fields even reaching up to 100% (Sasser et al., 1982; Seid et al., 2015). RKN species (*M. incognita*, *M. javanica*, *M. arenaria*, *Meloidogyne ethiopica* and *M. hapla*) have also been reported to occur in Ethiopia (Stewart and Dagnachew, 1967; Seid et al., 2017). Among them *M. incognita* is the most widespread species (Wondirad and Kifle, 2000). The occurrence of *M. incognita* had been reported on tomato in the eastern part of the country, particularly in eastern Hararghe where many vegetable crops were attacked by this RKN (Solomon, 1987; Tadele and Mengistu, 2000; Seid et al., 2017). The RKN, particularly *M. incognita*, is the major problem in tomato cultivation in the central and western parts, too (Mandefro and Mekete, 2002; Seid et al., 2017).

Prevention of the nematode can be done with planting material, seeds, or soil, crop rotation, intercropping,

growing nematode resistant varieties or rootstocks, and through nematicides (Ploeg, 2008). Even though, nematicides are effective in nematode management, however, due to high costs, and the hazards they pose on human as well as on non-target organisms (Nagaraju et al., 2010) made an urge to search for better alternative management strategy.

In spite of wider distribution of the RKN on many crops in Ethiopia, very few work have been done on the management of this nematode species in the country. So far, no efforts have been made to exploit the integrated management of *M. incognita* in tomato using botanicals and intercropping. Hence, the present study was initiated to evaluate the integrated effects of lantana leaf extract and intercropping of tomato with mustard and garlic on *M. incognita* at different inoculation levels under greenhouse conditions.

MATERIALS AND METHODS

Experimental site

A pot culture experiment was conducted at Haramaya University greenhouse, East Hararghe Zone, Oromia Region, Ethiopia at 9°26'N latitude, 42°3' E longitudes. The altitude varies from 1980 to 2000 m above sea level (m.a.s.l.). The mean annual precipitation is 780 mm and the mean annual maximum and minimum temperatures are 23.4 and 8.25°C, respectively. Plants were watered daily and maintained at 23 ± 2°C (12 h day) with 60 to 65% relative humidity for the entire experimental period in a greenhouse at Rarree Research Station of Haramaya University.

Experimental materials

Tomato cv. Moneymaker from (Melkassa Agrioclutural Research Center), garlic (*Allium sativum*) and mustard (*Brassica carinata*) from Debre-Zeit Agricultural Research Center from vegetable research group were obtained, respectively. Leaves of lantana (*Lantana camara*) were collected from the nearby surroundings of Haramaya University. The *M. incognita* population was collected from Jittu farm, Hawassa and identified molecularly by Seid et al. (2017) at Ghent University.

Maintenance of pure culture of *M. incognita*

Pure culture of the *M. incognita* population used in this experiment was raised from a single egg-mass and maintained for ten weeks on Moneymaker at Haramaya University greenhouse. Infected plants were uprooted and roots were gently washed in water to remove the adhering soil particles. Further, the roots were cut into 2 cm pieces and left in a modified Baermann funnel to get fresh second-stage juveniles (J2) (< 24 h old). About 1 ml of *M. incognita* J2 suspension was pipetted into a counting dish and the population was enumerated under a stereomicroscope. Counting was repeated three times to record the mean nematode count.

Preparation of plant extracts

Leaves of lantana (*L. camara*) were collected from the trees grown in the surroundings of Haramaya University campus and used to

prepare the extract. Shade-dried leaves were powdered in an electric blender and 20 g of the botanical powder was soaked in 100 ml distilled sterile water for 24 h in 500 ml flasks. Twenty-four hours after soaking, the materials were filtered through cheese cloth (Wondimeneh et al., 2013) and the filtrates were used for the subsequent experiment.

Treatments and experimental design

There were a total of 18 treatment combinations with three levels of inocula of *M. incognita* (0, 1000 and 2000 J2/pot), including an uninoculated control. Three different cropping patterns viz., tomato with mustard (TM), tomato with garlic (TG) and pure tomato cropping (PT), two levels of the botanicals and untreated control were maintained. Seeds of tomato cv. Moneymaker were sown and raised in a sterilized soil with 1:2:3 proportions of sand, compost and clay, respectively, in a 2 kg capacity pots. About 30 ml of botanical extract (at 5% v/v concentration) was applied one week before transplanting of the seedlings. Seeds of mustard (*B. carinata*) and garlic (*A. sativum*) were planted directly into pots containing sterilized soil and four-week-old-seedlings of tomato cv. Moneymaker were transplanted into pots. The antagonistic plants (mustard and garlic) were planted on either side of the tomato seedlings in line with the treatments. *M. incognita* suspension was inoculated at the rate of 1000 and 2000 J2/pot one week after transplanting. An untreated control was also maintained. Inoculation was done around the root-zone of the tomato seedlings by making three holes and they were covered after inoculation. The experiment was laid out in a factorial completely randomized design (CRD) with four replications. All agronomic practices were maintained as recommended to tomato until termination of the experiment.

Data collection

The following nematode and plant related data parameters were collected sixty days after nematode inoculation (DAI).

Nematode related parameters

Number of galls per root system: After cutting the top parts of the plants, all the pots were turned upside down with care, to discharge the soil and the roots were uprooted and rinsed with tap water to remove adhering soil particles. Number of galls per root system was counted manually aided with hand lens.

Number of egg-masses per root system: Roots containing egg-masses were soaked in a solution of Phloxine B (15 mg 100 ml⁻¹ in tap water) for 15 to 20 min and then the roots were rinsed in tap water to remove residual stain. The egg-masses were stained pink to red and observed and counted under stereomicroscope (Coyne et al., 2014).

Number of eggs per egg-mass: Ten egg-masses per plant were randomly taken using forceps. The egg-masses were shaken with 5% sodium hypochlorite (NaOCl) in stopper flasks for 2 min (Hussery and Barker, 1973). The numbers of eggs were counted under stereomicroscope.

Final nematode population density per pot (Pf): Soil and root samples from the pots were collected in marked plastic bags and were brought to the laboratory and nematode extraction was made using the modification of Baermann funnel (Southy, 1970). The Pf of *M. incognita* was counted by transferring the suspension to nematode counting dish.

Reproduction factor (RF): It was obtained from the ratio of Pf to Pi (initial population density).

Plant related parameters

Plant height: Plant height was measured in centimeters from the soil line to the tip of the tomato stem in each pot.

Root length: Root length was measured in centimeters from soil line to the tip of fibrous root after the adhering soil was washed using tap water.

Number of leaves per plant: Number of leaves per plant was counted in each pot.

Fresh shoot weight (g): The tomato plant was cut at the crown level in each pot and the fresh shoot weight was measured in gram using electronic balance soon after cutting.

Dry shoot weight (g): The shoots were put in paper bag and brought to laboratory just after taking the fresh weight and kept in an oven at 105°C for 24 h, and allowed to come to room temperature and the dry shoot weight was measured in gram using an electronic balance.

Data analysis

The data were subjected to the standard analysis of variance (ANOVA) procedures using Gens tat 15th edition statistical software package. The differences among treatment means were separated using Fisher's protected LSD test at 5% level of probability.

RESULTS

Nematode-related parameters

Number of galls per root system

The result revealed that the main factor botanical, intercropping and inocula level, as well as all possible two factors and three factors interactions had highly significant ($p \leq 0.01$) influence on number of galls produced per root system of tomato (Table 1). The highest (997.2) mean number of galls was observed in 2000 J2 inoculated, un-treated and tomato sole cropping treatment. The lowest (177.5) mean number of galls was recorded for 1000 J2 inoculated, treated and tomato-mustard intercropping treatment. Larger-sized galls were observed on 2000 J2 inoculated treatment than on 1000 J2 inoculation level to all cropping patterns and for both botanical treated and untreated tomato plants. Both tomato-garlic and tomato-mustard cropping patterns when added together with lantana leaf extracts recorded significant reduction in the number of galls per root compared to the treatment where either botanical or intercropping was used alone. The result of this investigation thus clearly showed the significant performance of the cumulative effect by botanical and intercropping.

Table 1. Interactions effect of botanical and intercropping at different inocula levels of second-stage juveniles (J2) of *Meloidogyne incognita* on number of galls per root system, number of egg-masses per root system, number of eggs per egg-mass and final population density per pot at HU, Rarree greenhouse.

| Botanical | Inocula (J2s) | PT | TG | TM |
|--|---------------|---------------------|---------------------|---------------------|
| Intercropping on number of galls | | | | |
| T | 0.00 | 0.0 ^k | 0.0 ^k | 0.0 ^k |
| | 1000 | 282.8 ^h | 277.0 ^h | 177.5 ^j |
| | 2000 | 529.8 ^d | 500.5 ^e | 263.3 ^l |
| UT | 0.00 | 0.0 ^k | 0.0 ^k | 0.0 ^k |
| | 1000 | 459.0 ^f | 452.5 ^f | 299.0 ^g |
| | 2000 | 997.2 ^a | 976.8 ^b | 658.5 ^c |
| LSD (5%) | | 11.89 | | |
| CV (%) | | 2.6 | | |
| Intercropping on number of egg-masses | | | | |
| T | 0.00 | 0.00 ^l | 0.00 ^l | 0.00 ^l |
| | 1000 | 253.5 ^{fg} | 260.0 ^f | 161.0 ^h |
| | 2000 | 705.2 ^b | 706.2 ^b | 251.8 ^g |
| UT | 0.00 | 0.00 ^l | 0.00 ^l | 0.00 ^l |
| | 1000 | 404.2 ^d | 403.5 ^d | 290.0 ^e |
| | 2000 | 842.2 ^a | 844.2 ^a | 609.2 ^c |
| LSD (5%) | | 6.67 | | |
| CV (%) | | 1.5 | | |
| Intercropping on number of eggs per egg-mass | | | | |
| T | 0.00 | 0.00 ^j | 0.00 ^j | 0.00 ^j |
| | 1000 | 996.8 ^f | 975.5 ^g | 859.2 ⁱ |
| | 2000 | 1108.0 ^c | 1115.3 ^c | 968.0 ^g |
| UT | 0.00 | 0.00 ^j | 0.00 ^j | 0.00 ^j |
| | 1000 | 1035.5 ^e | 1032.0 ^e | 901.7 ^h |
| | 2000 | 1156.8 ^a | 1138.0 ^b | 1043.5 ^d |
| LSD (5%) | | 7.87 | | |
| CV (%) | | 8 | | |
| Intercropping on final population density per pot | | | | |
| T | 0.00 | 0.00 ^m | 0.00 ^m | 0.00 ^m |
| | 1000 | 2003 ^g | 1503 ⁱ | 1003 ^l |
| | 2000 | 3078 ^b | 2068 ^d | 1061 ^k |
| UT | 0.00 | 0.00 ^m | 0.00 ^m | 0.00 ^m |
| | 1000 | 2056 ^e | 1516 ^h | 1088 ^j |
| | 2000 | 4016 ^a | 3034 ^c | 2017 ^f |
| LSD (5%) | | 4.94 | | |
| CV (%) | | 3 | | |
| Intercropping on reproduction factor | | | | |
| T | 0.00 | 0.0000 ^l | 0.0000 ^l | 0.0000 ^l |
| | 1000 | 2.0030 ^c | 1.5030 ^f | 1.0032 ^j |
| | 2000 | 1.5391 ^d | 1.0340 ^h | 0.5306 ^k |
| UT | 0.00 | 0.0000 ^l | 0.0000 ^l | 0.0000 ^l |
| | 1000 | 2.0560 ^a | 1.5162 ^e | 1.0875 ^g |
| | 2000 | 2.0079 ^b | 1.5171 ^e | 1.0086 ⁱ |

| | |
|----------|-------|
| LSD (5%) | 0.003 |
| CV (%) | 2 |

Means followed by the same letter(s) within a row and column in each character are not significantly different at 5% level of significance. LSD (5%) = least significant difference; CV (%) = coefficient of variation, J2 = second stage juveniles of *Meloidogyne incognita*, T = treated with leaves extract of lantana at 5% concentration, UT = untreated treatment with lantana leaves extract, PT = pure tomato plant cropping, TG = tomato plant intercropped with garlic, TM = tomato plant intercropped with mustard.

Number of egg-masses per root system

The main factor botanical, intercropping and inocula levels, as well as all possible two factors and three factors interactions had highly significant ($p \leq 0.01$) influence on the number of egg-masses per root system. The combined effect of botanical and intercropping to all inoculation levels showed significant suppression of egg-masses per root system (Table 1). The maximum (842.2) mean number of egg-masses was observed in 2000 J2 inoculated, un-treated and tomato sole cropping treatment. The lowest (161.0) mean number of egg-masses was recorded for 1000 J2 inoculated treated and tomato mustard intercropping treatment. There was no significant difference in the number of egg-masses in tomato sole cropping and tomato-garlic intercropping in all inoculation levels of J2s of *M. incognita*. Combined effect of botanical and tomato-mustard intercropping was found to be the most effective in reducing the number of egg-masses compared to all other treatments.

Number of eggs per egg mass

The result showed that the main factor botanical, intercropping and inocula, as well as all possible two factors and three factors interactions had highly significant ($p \leq 0.01$) influence on the number of eggs per egg-mass (Table 1). Significant reduction in fecundity was observed when plants were treated using lantana leaf extract and intercropping of tomato with garlic and mustard alone. But combination of lantana leaf extract and tomato-mustard intercropping treatment resulted in a maximum (859.2) decline in eggs of *M. incognita*. Tomato alone without botanical treatment and without intercrops but inoculated 2000 J2 of *M. incognita* caused the highest (1156.8) number of eggs egg-mass⁻¹ as compared to other treatments.

Final population density per pot

The obtained result revealed that the main factor botanical, intercropping and inocula, as well as all possible two factors and three factors interactions had a significant ($p \leq 0.01$) influence on nematode population (Table 1). Maximum (1003) reduction of nematode population occurred in soil treated with botanical, 1000 J2

inoculated and tomato-mustard intercropped treatment and the highest (4016) number of nematode population was recorded due to 2000 J2 inoculated, un-treated and tomato sole cropping treatment.

Reproduction factor (RF)

The main factor botanical, intercropping and inocula as well as all possible two factors and three factors interactions had highly significant ($p \leq 0.01$) influence on reproduction factor (Table 1). Tomato plants inoculated with 1000 J2 *M. incognita* without any treatment showed the highest (2.0560) reproduction rate. Application of botanical and intercropping with garlic and mustard either alone or in combination showed a significant reduction in reproduction of nematode but the reduction was more pronounced in tomato plant intercropped with mustard and treated with leaf extract of lantana (0.5306).

Plant parameters

Plant height

The main factors botanical, intercropping, inocula levels, and interactions effect of botanical and inocula as well as three factors interactions (botanical \times intercropping \times inocula) had highly significant ($p \leq 0.01$) influence on tomato plant height. However, the remaining two factors interactions (botanical \times intercrop and intercropping \times inocula) did not significantly affect plant height (Table 2). The tallest (73.75 cm) plant was observed in un-inoculated, botanical treated and tomato sole cropping treatment. The shortest (39.75 cm) plant height was recorded for 2000 J2 inoculated, un-treated and tomato-mustard intercropping treatment. The reduction of plant height might be due to the root damage of the plant by the *M. incognita* population used.

Root length

The experimental result revealed that all the main factors (botanical, intercropping and inocula) and the interactions effect of botanical and inocula as well as inocula and intercropping had highly significant ($p \leq 0.01$) influence on root length of tomato (Table 3). The longest (18.62

Table 2. Interaction effects of botanical, intercropping and inocula levels of second-stage juveniles (J2s) of *Meloidogyne incognita* on plant height of tomato at HU, Rarre greenhouse.

| Botanical | Inocula (J2s) | Plant height (cm) in intercropping system | | |
|-----------|---------------|---|--------------------|---------------------|
| | | PT | TG | TM |
| T | 0.00 | 73.75 ^a | 65.00 ^c | 51.75 ^e |
| | 1000 | 68.00 ^{bc} | 68 ^b | 48.50 ^e |
| | 2000 | 56.5 ^d | 57.25 ^d | 36.75 ^g |
| UT | 0.00 | 70.75 ^{ab} | 70.5 ^{ab} | 49.00 ^e |
| | 1000 | 60.5 ^d | 51.25 ^e | 42.75 ^f |
| | 2000 | 49.75 ^e | 49.50 ^e | 39.75 ^{fg} |
| LSD (5%) | | 4.07 | | |
| CV (%) | | 5.1 | | |

Means followed by the same letter(s) within row and column in each character are not significantly different at 5% level of significance. LSD (5%) = Least significant difference; CV (%) = Coefficient of variation, J2 = Second stage juveniles of *Meloidogyne incognita*, T = treated with leaf extract of lantana at 5% concentration, UT = untreated treatment with lantana leaf extract, PT = pure tomato plant cropping, TG = tomato plant intercropped with garlic, TM = tomato plant intercropped with mustard.

Table 3. Interaction effects of inocula levels of second-stage juvenile (J2) of *Meloidogyne incognita* with botanical and intercropping on root length of tomato at HU, Rarre greenhouse.

| Botanical | Root length (cm) | | |
|---------------|---------------------|---------------------|----------------------|
| | 0.00 J2 | 1000 J2 | 2000 J2 |
| T | 15.79 ^a | 13.33 ^b | 12.21 ^b |
| UT | 16.62 ^a | 8.83 ^c | 5.96 ^d |
| LSD (5%) | 0.96 | | |
| Intercropping | 0.00 J2 | 1000J2 | 2000 J2 |
| PT | 18.62 ^a | 11.88 ^{bc} | 10.00 ^{bcd} |
| TG | 18.12 ^a | 12.25 ^b | 9.94 ^{bcd} |
| TM | 11.88 ^{bc} | 9.12 ^{cd} | 7.31 ^d |
| LSD (5%) | 1.18 | | |
| CV (%) | 9.7 | | |

Means followed by the same letter(s) within row and column in each character are not significantly different at 5% level of significance. LSD (5%) = least significant difference; CV (%) = coefficient of variation, J2 = second stage juveniles of *Meloidogyne incognita*, T = treated with leaves of lantana at 5% concentration, UT = untreated treatment with lantana leaves extract, PT = pure tomato plant sole cropping, TG = tomato plant intercropped with garlic, TM = tomato plant intercropped with mustard.

cm) root length was recorded on the un-inoculated tomato planted alone treatment, while the shortest (5.96 cm) length was recorded in un-treated, 2000 J2 inoculated treatment. The interaction effects of botanical and inocula and also inocula and intercropping had significant influence on root length. No significant difference was detected between 1000 J2 and 2000 J2 inoculation level treatments on root length but in un-treated pots raising inocula levels of *M. incognita* reduced the tomato root length. Generally, it was observed that the effect of nematode was more pronounced in roots than on shoots on *M. incognita* inoculated un-treated pots. There were no remarkable difference between

tomato sole cropping and tomato-garlic intercropping at 1000 J2 and 2000 J2 inoculation level.

Number of leaves

The highest (444.5) number of leaves per plant was recorded on un-inoculated, tomato sole cropping treatment. The smallest (153.4) number of leaves was recorded on 2000J2 of *M. incognita* inoculated tomato-mustard intercropped treatment. Leaf number was significantly influenced by the interaction effects of intercropping and inocula levels (Table 4). Raising

Table 4. Interaction effects of intercropping and inocula levels of second-stage juvenile (J2) of *Meloidogyne incognita* on leaf number of tomato at HU, Rarre greenhouse.

| Intercropping | Number of leaves | | |
|---------------|--------------------|--------------------|---------------------|
| | Inocula (0.00 J2) | Inocula (1000 J2) | Inocula (2000 J2) |
| PT | 444.5 ^a | 320.7 ^b | 207.4 ^d |
| TG | 411.5 ^a | 257.6 ^c | 183.6 ^{de} |
| TM | 250.1 ^c | 207.7 ^d | 153.4 ^e |
| LSD (5%) | | 28.32 | |
| CV (%) | | 10.4 | |

Means followed by the same letter(s) within a row and column in each character are not significantly different at 5% level of significance. LSD (5%) = least significant difference; CV (%) = coefficient of variation, J2 = second stage juveniles of *Meloidogyne incognita*, PT = pure tomato plant cropping, TG = tomato plant intercropped with garlic, TM = tomato plant intercropped with mustard.

Table 5. Interaction effects of intercropping and inocula levels of second-stage juvenile of *Meloidogyne incognita* on fresh shoot weight of tomato at HU, Rarre greenhouse.

| Intercropping | Fresh shoot weight (g) | | |
|---------------|------------------------|---------------------|---------------------|
| | Inocula (0.00 J2) | Inocula (1000 J2) | Inocula (2000 J2) |
| PT | 295.8 ^a | 256.6 ^{bc} | 218.3 ^d |
| TG | 261.8 ^b | 247.6 ^{bc} | 231.5 ^{cd} |
| TM | 161.5 ^e | 208.1 ^d | 175.9 ^e |
| LSD (5%) | | 22.24 | |
| CV (%) | | 9.7 | |

Means followed by the same letter(s) within row and column in each character are not significantly different at 5% level of significance. LSD (5%) = least significant difference; CV (%) = coefficient of variation, J2 = second stage juveniles of *Meloidogyne incognita*, PT = pure tomato plant cropping, TG = tomato plant intercropped with garlic, TM = tomato plant intercropped with mustard.

inocula levels of J2 of *M. incognita* in all cropping patterns reduced tomato leaf numbers.

Fresh shoot weight

The main factors intercropping, inocula and interaction effect of inoculums and intercropping had highly significant ($p \leq 0.01$) influence on fresh shoot weight of tomato. However, the remaining possible two factors and three factors interactions did not significantly affect tomato fresh shoot weight (Table 5). Among the treatments, the highest (295.8 g) and the lowest (175.9 g) fresh shoot weight were recorded from un-inoculated tomato sole cropped treatment and 2000 J2 inoculated tomato plant intercropped with mustard, respectively. Interaction effects of inocula levels and intercropping showed significant effect on fresh shoot weight. Increasing inocula levels of second-stage juveniles of *M. incognita* in all cropping patterns reduced fresh shoot weight of tomato. There was no significant difference between tomato sole cropping and tomato-garlic intercropping at 1000 J2 of *M. incognita* and 2000 J2 of *M. incognita* level.

Dry shoot weight

The inocula levels and intercropping had highly significant ($p \leq 0.01$) influence on dry shoot weight of tomato plants. However, dry shoot weight was not significantly influenced by botanical; the interaction effect of all the possible two and three factors interactions (Table 6). Tomato sole cropping resulted in mean dry shoot weight (66.1 g) at par with tomato-garlic intercropping but both exceeded the tomato-mustard intercropping. Dry shoot weights of plants inoculated with 2000 J2 of *M. incognita* showed minimum mean value (58.3 g) than 1000 J2 of *M. incognita* inoculated and un-inoculated control.

DISCUSSION

The results of this study showed that raising the inoculation levels of *M. incognita* recorded significant reduction on growth characteristics of tomato and significant increase on nematode related parameters. This might be due root infection that resulted in stunting

Table 6. Effect of inocula of second-stage juveniles of *Meloidogyne incognita* and intercropping on dry shoot weight of tomato at HU, Rarre greenhouse.

| Inoculum | Dry shoot weight (g) |
|----------------------|----------------------|
| 0.00 J2 | 66.9 ^a |
| 1000 J2 | 63.3 ^b |
| 2000 J2 | 58.3 ^c |
| LSD (5%) | 1.39 |
| Intercropping | |
| PT | 66.1 ^a |
| TG | 66.1 ^a |
| TM | 57.08 ^b |
| LSD (5%) | 1.39 |
| CV (%) | 3.8 |

Means within a row in each treatment and character with similar letter(s) are not significantly different at 5% level of significance, LSD (5%) = least significant difference, CV (%) = coefficient of variation, J2 = second stage juveniles of *Meloidogyne incognita*, PT = pure tomato plant cropping, TG = tomato plant intercropped with garlic, TM = tomato plant intercropped with mustard.

action of *M. incognita* and distortion of the plant roots and nematode colonization of vascular bundles that reduce supply of nutrients to plants. Sikora and Fernandez (2005) reported that the galls on the root system might disturb important root functions like uptake and transport of water and nutrients.

Application of botanical (at 5% v/v concentration) had significant improvement on tomato growth and recorded suppressive effect on galling intensity and nematode population when used alone and in combination with other treatments. This might be due to the nematicidal activity of aqueous leaf extract of *L. camara* against *M. incognita* as indicated by the reduced galling and egg-masses production. Udo et al. (2014) also reported that nematostatic properties of *L. camara* leaf extract were attributed to poor coordination and orientation of infective juveniles towards the plant roots. Among the different treatments, combination of leaf extract of lantana and tomato-mustard intercropping was the most effective in reducing galling intensity and nematode population followed by combination of leaf extract of lantana and tomato-garlic intercropping. Untreated control recorded the highest galling intensity and nematode population, thereby showing poor tomato growth. Addition of botanical to soil leads to a better environment for the growth of the roots. This enhances the utilization of soil nutrients, as a consequence, nematode damage might have been markedly reduced (Abubakar et al., 2004).

Plants are important sources of naturally occurring compounds, many of which have nematicidal properties, like alkaloids, diterpenes, fatty acids, glucosinolates, isothiocyanates, phenols, polyacetylenes, sesquiterpenes and thienyls (Chitwood, 2002). Mustard, African marigold,

asparagus, castor, and sesame have been found to reduce population of RKN and may be grown as commercial crops, cover crops, or established in mixed planting with other crops for the management of root-knot nematodes (Bridge, 1996). Use of such antagonistic plants as green manures improves the fertility of the soil too. Certain nematode-toxic chemicals, which kill nematodes, are released during decomposition of the plant materials. Green manures increase the saprophagous bacteria, fungi, and other microorganisms in the soil, which aid in reducing RKN population (Barker et al., 1985). Halbrendt (1996) suggested that plant compounds elicit nematode behavior, such as attraction or repulsion from roots. Naturally occurring plant chemicals for nematode management have advantages being eco-friendly and posing no hazardous effect on produce over the current use of synthetic chemicals.

The efficient management of PPN requires the carefully integrated combination of several methods. Although each individual method of management has a limited use, together, they help in reducing the nematode populations more efficiently. With the ongoing progress in research, a public desire for methods of managing/reducing plant pests in ways that are cheap, easily available, eco-friendly and do not pollute or otherwise degrade the environment, has increased concomitantly. The integrated pest management provides a working methodology for pest management in sustainable agricultural systems. One such method employed for maintaining the populations of PPN below the economic threshold level, is the mixed cropping practice, sometimes also referred to as intercropping, which is a form of multiple-cropping system using host and non-host

crops at the same place and time (Rodriguez-Kabana and Canuilla, 1992).

It has been reported by several workers that different cropping sequences reduced the populations of some harmful PPN to the levels that do not cause economic losses (Idowua and Fawole, 1989; Haider et al., 2001). Haider et al. (2004) reported that the intercropping two rows of yellow mustard sarson (*Brassica campestris* var. *toria* / *B. campestris* var. *sarson*) with sugarcane, recorded the highest reduction (23.7%) in nematode populations followed by sugarcane + one row of yellow mustard at the time of harvest of intercrops. This sequence showed prolonged effect of toxicity as evidenced by 21% reduction in nematode population from initial density level at the time of harvest of sugarcane. Sugarcane and yellow mustard intercropping system exhibited the highest cane yield. Similar results of inclusion of mustard, a poor host for several nematodes, in different cropping sequences for reducing nematode populations have been reported by several other workers (Singh and Sitaramaiah, 1993; Kumar and Khanna (2006). Intercropping mustard could attribute the decrease in nematode populations by intercropping mustard to the presence of 2-propenyl isothiocyanate in mustard having nematicidal activity as reported by Kowalska and Sonalinska (2001). However, no report was found in this type of three factors study to explain the result in relation to others' finding. Since this experiment was carried out under controlled (greenhouse) conditions, further study is required under farmer's field conditions to prove the consistency of the results obtained and improve their application technologies. Moreover, intercrops might have competitive effects on crops for essential growth factors, which may, in turn, affect the yield of crops; thus further trials geared towards using mustard and garlic as a short term fallow plants would give a more conclusive result in their use for management of *M. incognita*.

Conclusion

The present study was conducted to evaluate the integrated effect of botanical and intercropping against different inoculation levels of root-knot nematode, *M. incognita* on tomato cv. Moneymaker. The result of this study showed that raising the inoculation levels of second-stage juveniles of *M. incognita* in all treatments showed a significant reduction on growth characteristics of tomato and had significant increase on nematode related parameters. Application of botanical had significant improvement on tomato growth and showed suppressive effect on galling intensity and nematode population when used alone and in combination with other treatments. Among the treatments, the combination of leaf extract of lantana and tomato-mustard intercropping was the most effective in reducing galling

intensity and nematode population; also, the combination of leaf extract of lantana and tomato-garlic intercropping was found the next effective treatment in reducing galling intensity and nematode population. Untreated inoculated tomato-sole cropping treatment was recorded the highest galling intensity and nematode population, thereby showing poor tomato growth. In conclusion, although this study was carried out in the greenhouse, it has clearly indicated that integrated use of botanicals and intercropping could be used as an alternative method for the management of root-knot nematode in tomato production. However, further research is needed to evaluate their efficacies under field conditions and improve their application technologies.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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