POTENCY OF *Azadirachta indica* AND *Tithonia diversifolia* BASED BIO-PESTICIDE ROOT DIPS IN THE MANAGEMENT OF ROOT-KNOT NEMATODE INFECTION IN TOMATO (*Solanum lycopersicum* L.)

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SUMMARY

Root-knot nematodes, *Meloidogyne species*, remain very important economic threats to tomato cultivation. Screen-house experiment was carried out in 2014 and repeated in 2015 to test the efficacy of two plant-based pesticide slurries from Mexican sunflower (*Tithonia diversifolia*) and Neem (*Azadirachta indica*) at three different concentrations (0%, 0.5% and 1%), singly or in combination on root-knot nematodes disease on tomato. There were six treatments which included Roma VF tomato seedlings dipped for five minutes in Neem at 0%, 0.5%, and 1%, *Tithonia* at 0%, 0.5% and 1% slurries and transplanted in 7litrepots containing 6 kg sterilized soil. Treatments were arranged in Completely Randomized Design and replicated Eight times. At four weeks after transplanting, each treatment was inoculated with approximately 5000 eggs of *Meloidogyne incognita* except the control. Data were collected at 2, 4, 6 and 8 Weeks After Inoculation (WAI) on plant height, stem girth, leaf area and number of leaves. At 60 days after inoculation, the experiment was terminated to determine the final nematode population. Data collected were subjected to Analysis of Variance. Statistically different means were separated using Duncan Multiple Range Test (DMRT) at 5% level of probability. Results showed that dipping roots of Roma VF tomato seedlings slurries made from powdered leaves of *T. diversifolia* and *A. indica* for 5 minutes at 1% concentrations significantly reduced *Meloidogyne incognita* populations between 4% and 35% compared with the control in 2014 and 2015.In 2014, there was a significant reduction in *Meloidogyne* species populations in plant treated with *Tithonia* at 1% (3.84%) compared with Neem at 1% (16%). In 2015, plant treated with Neem at 1% reduced ($p \leq 0.05$) egg number from 50% to 35% while *Tithonia* at 1% caused significant ($p \leq 0.05$) reduction from 50% to 24%.The study concluded that, although, both *Tithonia diversifolia* and *Azadirachta indica* can be used to suppress nematode population and improve the Roma VF tomato plant growth. *T. diversifolia* proved to be more effective in nematode management strategy.

Keywords: Biopesticides, *Meloidogyne* species, Neem, Organic farming, Mexican Sunflower

TOMATO, *Solanum lycopersicun* L., is the world most highly consumed vegetable due to its status as a basic ingredient in large variety of raw, cooked or processed foods. It belongs to the family Solanaceae, Tomato is grown worldwide for local use or as an export crop. In 2014, the global area cultivated with tomato was 5 million hectares with a production of 171 million tonnes, the major tomato-producing country is Republic of China. (FAOSTAT, 2017). Tomato is a good source of vitamin which helps in the body development and maintenance of the scar tissue, blood vessels and cartilage. According to FAO report (2010), increasing concerns by the public in the past few years due to food borne diseases and the need to consume healthy food. Root-knot nematodes (*Meloidogyne* spp) are small-like worms, which overwinter in the roots of perennial weeds or as egg or larvae in the soil or affected debris. The larvae penetrate to the roots and excrete a substance that induces formation of root galls. Root- knot nematode (*Meloidogyne incognita)* cause stunting and wilting of the tomato plant. Root-knot nematodes *Meloidogyne* spp. problem have become a threat to tomato cultivation. The yield loss alone has been estimated to be 30 - 50% (Jain, 1991). Root-knot nematodes influence tomato plant causing the development of root-knot galls which drain the plant photosynthetic and nutrients and this leads to reduction in yield quantity and quality. In Nigeria a yield loss of between 28-68% was reported in tomato field (Adesiyan *et al*., 1990). Root-knot nematodes (*Meloidogyne* spp.) are among the most economically damaging genera of plant parasitic nematodes on horticultural and field crops, causing an estimated US$100 billion loss globally on an annual basis (Oka *et al*., 2000). Root-knot nematode *Meloidogyne incognita* (Kofoid & White) Chitwood, is one of the most harmful root-knot nematode species, which infects a wide range of vegetable crops in Egypt (Ibrahim *et al*., 2000). Severe damage has been documented as a result of its attack; its root infection makes a plant unfit to absorb water and nutrient from soil resulting in poor growth, yield loss and loss of market quality (Gowen *et al.*, 2005).Root-knot nematode damage result in poor growth and a decline in quantity and quality of the crop. (Makumbi *et al*., 2000). Root -knot nematodes can be controlled with bio control agents (Dama *et al*., 1999). They include the use of nematicides, organic amendments, resistance cultivars, soil solarization and biological controls, which have been used with different levels of success for the protection of tomato plants (Randhawa *et al*., 2001; Sakhuja and Jain, 2001).

Researchers have shown that extracts from plants such as *Azadirachta, Eucalyptus, Chrommelina, Sidaacuta* and *Targetis* have been found to be effective in the control. Although chemical nematicides are effective, easy to apply and show rapid effects, they have begun withdrawn from the market owing to concerns about public health and environmental safety (Rich *et al*., 2004). Attention is turning towards an integrated resistance, nematicidal or antagonistic plants, bio control agents and bio products to check the nematode population that they may remain below the level of economic threshold (Desaeager and Rao, 2000). The potential benefits to agriculture and public health programs through the use of bio-pesticides are considerable. Among the so called botanicals, derivatives from Neem (*Azadirachta indica* A. Juss) have been found to have potentials for controlling plant- parasitic nematodes (Khanna and Kumar 2006). *Tithonia diversilfolia* can be used as a green fertilizer on tomato plant that show stunted growth. It is a useful method to increase the crop yield in order to benefits the farmers wealth (Wanjiku *et al.*, 2006). Mexican sunflower (*Tithonia diversifolia*) is used in an *in vitro* experiment to manage plant parasitic nematode (Akinyemi *et al*., 2009). Therefore, the aim of this study is to determine the effect of *Azadirachta indica* and *Tithonia diversifolia* root-dip slurry on nematode population and agronomic parameters of tomato Roma VF.

MATERIALS AND METHODS

Experimental site

The Experiments were carried out at the screen house of the Department of Crop Protection, College of Plant Science and Crop Production, Federal University of Agriculture, Abeokuta (FUNAAB), Ogun State.

Soil sterilization and nursery establishment

Sandy loam soil was steam-sterilized for 3hours at approximately 100^{0} C. The sterilized soil was packed inside sacks and left for 6weeks before use to regain its stability. Fifteen grams (15 g) of the sterilized soil was weighed and loaded in the nursery trays and tomato seedlings were grown and nurtured for four (4) weeks before transplanting.

Experimental materials and transplanting

Tomato seed, Roma variety (Roma VF), were obtained from National Horticultural Research Institute (NIHORT), Jericho, Ibadan, Oyo State. The biopesticides used were Neem (*Azadirachta indica*), Mexican sunflower (*Tithonia diversifolia*), and was collected from FADAMA at Federal University of Agriculture Abeokuta (FUNNAB). The leaves of Neem (*Azadirachta indica*), Mexican sunflower (*Tithonia diversifolia*), were collected and air-dried, blend into powdery form using the blender. Six kilogram (6 kg) of soil was weighed into 7litres bucket and wet. After four weeks, the seedlings were transplanted to the experimental pots. The slurry was prepared using 1% volume of biopesticde by 1% volume of water. Seedlings were dipped in biopesticides slurries and were transplanted into experimental pots with sterilized soil.

Treatments and experimental design

Completely Randomized Design was used for both Screen house experiments. In 2014, the experiment consisted of Tomato Roma VF, five treatments (Control, Tomato+Tithonia, Tomato+Neem, Tomato+5000M.i+Tithonia, Tomato+5000M.i+Neem) and was replicated eight times. In 2015, the experiment consisted of Tomato Roma VF, six treatments (Control, Tomato+5000M.i, Tomato+5000M.i+Neem, Tomato+5000M.i+Tithonia, Tomato+5000M.i+Neem&Tithonia at 1%, Tomato+5000M.i+Neem&Tithonia at 0.5%. Tomato seedlings were dipped for five minutes in biopesicides slurries before been transplanted into the pots. Untreated control and check pots received no biopesticides.

Preparation of nematode inoculum

The inoculum was extracted from galled root of Celosia argentea from the infested plants and was used to inoculate the tomato plants. Nematode eggs using the sodium hypochlorite (NaOCl) method. Galled roots was washed thoroughly, and chopped into 1-2 cm pieces and placed into a conical flask into which one litre of 0.5% sodium hypochlorite solution was poured suspension, and was shaken thoroughly for three (3) minutes. The suspension was poured through a 200-mesh sieve and was rested upon a 500-mesh sieve to separate the organic debris from the eggs. The 500-mesh sieve was rinsed gently under the flow of water and the eggs were collected and were counted under stereomicroscope. Suspension containing 5000 eggs was poured at the base of tomato seedling inside the pots and was used as inoculums for the experiment.

Inoculation of Tomato seedlings

Four weeks old seedlings were inoculated with *M. incognita* by pouring egg suspension containing approximately 5000 eggs into a shallow trench which was covered up and watered slightly. Roots of tomato plants were dipped into Neem and *Tithonia* slurry for five (5) minutes before transplanted and some left untreated.

Extraction of nematode

The nematode population was estimated using Whitehead and Hemming (1965) procedure through modified Bearman tray method nematode extraction. Each composite soil was thoroughly mixed, nematodes were assayed from 250g of sub sample. The soil samples were put together in different sieves for nematode extraction using Whitehead and Hemming (1965) Technique method. The set up was left undistributed for twenty-four hours after which the nematode suspensions were decanted into a (500 ml) nalgene bottles. Distilled water was added to the nematodes suspension to the fill level of (500 lm) nalgene bottles. The suspensions were left to settle for five hours then siphon to remove supernatant water and the concentrated suspension of (15 ml) were poured into Mc-Cartney bottle. The nematode suspensions were stored in a refrigerator for nematodes identification and census. The suspension were mixed and was drawn three times using a pipette from each suspension into a Doncaster's counting dish while swirling the flash and manually blowing air into it to ensure uniform suspension for the identification and quantification of the extracted nematodes under stereo microscope and compound microscope.

Data collection and analysis

Tomato plant was observed daily and data was collected. The data collected included:-Plant height (cm) using metre rule, stem girth (mm) using the vernier caliper, number of leaves, leaf area (cm). This was done every two weeks after inoculation till the eight week. At (8) eight weeks after inoculation, experiment was terminated to obtain the number of root galls, root fresh weight, and dry weight. Eggs were extracted from the root using Hussey and Barker (1973) and *Meloidogyne incognita* were extracted in the soil using Whitehead and Hemming (1965).Data obtained were subjected to analysis of variance (ANOVA) using statistical analysis system (SAS, 2003) and the means were separated using Duncan Multiple Range Test (DMRT) at 5% level of probability.

RESULTS

Table 1 shows the evaluation of *Meloidogyne incognita* inoculation on plant height and stem girth of tomato in 2014

Plant height

At two (2) weeks after inoculation the control pot (Tomato only) (41.43) produced the highest plant height and was not significantly different ($p \leq 0.05$) from Tomato + Tithonia slurry (34.33), while Tomato + *M.incognita* + Neem slurry (12.73) produced the lowest plant height. At four (4) weeks after inoculation Tomato + Tithonia slurry had the highest number of plant height (58.47) but was not significantly different ($p \leq 0.05$) from the control pot (Tomato only) (51.73) while Tomato+ *M.incognita* + Neem slurry had the lowest number of plant height (21.30). At six (6) weeks after inoculation Tomato + Tithonia slurry (71.00) produce the highest plant height but was not significantly different ($p \leq 0.05$) from the control pot (Tomato only) (65.67) and Tomato+ *M.incognita* + Neem slurry produced the lowest number of plant height. Also, at eight (8) weeks after inoculation the control pot (Tomato only) (78.24) had the highest number of plant height compared to Tomato+ *M.incognita* + Neem (40.44) with the lowest number of plant height.

Stem girth

Stem girth at two (2) weeks shows that Tomato +Tithonia slurry (7.17) had the widest stem girth but was not significantly different from ($p \leq 0.05$) control pot (Tomato only) (6.10), while Tomato+ *M.incognita* + Neem (3.10) had the lowest stem girth but not significantly different from Tomato+ *M.incognita* + Tithonia (3.30). Four (4) weeks after inoculation the control pot (Tomato only) (7.67) had the widest stem girth but not significantly different ($p \leq 0.05$) from other treatments. Six (6) weeks after inoculation Tomato+ Tithonia slurry (9.17) had the widest stem girth but not significantly different ($p \leq 0.05$) from control pot (Tomato only) (8.43) while Tomato+ *M.incognita* + Neem (4.53) had the lowest number of stem girth. At eight (8) weeks after inoculation the control pot (Tomato only) (9.41) had the widest stem girth but not significantly different ($p \leq 0.05$) from Tomato+ Tithonia slurry (9.27), also Tomato+ *M.incognita* + Tithonia (7.48) was not significantly different ($p \leq 0.05$) from Tomato+ Neem slurry (5.84) while Tomato+ *M.incognita* + Neem (5.73) had the lowest number of stem girth.

Table 1: Evaluation of *Meloidogyne incognita* on plant height and stem girth of tomato plant in response to Neem and Tithonia as botanicals in 2014

Treatments Plant Height 2WAI	Plant Height 4WAI	Plant Height Girth 6WAI	Plant Height 8WAI	Stem 2WAI	Stem Girth 4WAI	Stem Girth 6WAI	Stem Girth 8WAI
Tomato only(Control) 41.43a	51.73ab	65.67a	78.24a	6.10ab	7.67a	8.43a	9.41a
Tomato+ Tithonia slurry 34.33a	58.47a	71.00a	73.41a	7.17a	7.17a	9.17a	9.27a
Tomato+ Neem slurry 16.37bc	36.57bc	34.33bc	45.60b	4.17bc	4.50a	4.67b	5.84b
Tomato+ M.incognita + 30.00ab Tithonia slurry	40.07b	44.33b	52.10ab	3.30c	5.23a	6.47ab	7.48ab
Tomato+M.incognita + 12.73c Neem slurry	21.30c	30.33c	40.44bc	3.10c	4.23a	4.53c	5.73bc

Means with the same letter(s) are not significantly different from each other. ($p \leq 0.05$)

P.H: Plant height (cm), S.G: Stem girth (mm), NOL: Number of leaves, NOB: Number of branches, WAI: Weeks after inoculating

Table 2 shows the evaluation of *Meloidogyne incognita* inoculation on number of branches and number of leaves of tomato in 2014

Number of branches

At two (2) weeks after inoculation the control pot (Tomato only) (1.67) had the highest number of branches but not significantly different ($p \leq 0.05$) from Tomato+ Tithonia slurry (1.33), Tomato+ *M.incognita* + Neem (0.33) had the lowest number of branches. At four (4) weeks after inoculation Tomato+ Tithonia slurry (2.33) and control pot (Tomato only) (2.00) were not significantly different ($p \leq 0.05$) from each other, while, Tomato + *M.incognita* + Neem (1.00) had the lowest number of branches. At six(6) weeks after inoculation Tomato+ Tithonia slurry (2.67) had the highest number of branches but not significantly different ($p \leq 0.05$) control pot (Tomato only) (2.33), Tomato+ *M.incognita* + Neem (1.33) had the lowest number of branches. At eight (8) weeks after inoculation control pot (Tomato only) (3.44) had the highest number of branches and was not significantly different ($p \leq 0.05$) from Tomato+ Tithonia slurry (3.21) while Tomato+ *M.incognita* + Neem (1.52) had the lowest number of branches.

Number of leaves

At two (2) weeks after inoculation Tomato+ Tithonia slurry (73.00) but not significantly different ($p \leq 0.05$) from control pot (Tomato only) (68.67), also Tomato+ *M.incognita* + Neem (13.67) had the lowest number of leaves. Four (4) weeks after inoculation there was no significant difference ($p \leq 0.05$) between Tomato+ Tithonia slurry (129.00) and control pot (Tomato only) (107.3) compared to Tomato+ *M.incognita* + Neem (44.00) had the lowest number of leaves. At six(6) weeks after inoculation Tomato+ Tithonia slurry (144.00) but was not significantly different ($p \leq 0.05$) from control pot (Tomato only) (132.3) compared to Tomato + *M.incognita* + Neem (61.33) had the lowest number of leaves. At eight (8) weeks after inoculation, Control pot (Tomato only) (161.4) had the highest number of leaves but was not significantly different ($p \leq 0.05$) from

Tomato+ Tithonia slurry (158.20) while Tomato+ *M.incognita* + Neem (76.25) had the lowest number of leaves.

Table 2: Evaluation of *Meloidogyne incognita* on number of branches and number of leaves of tomato plant in response to Neem and Tithonia as botanicals in 2014

Treatments	Number of Branches 2WAI	Number of Branches 4WAI	Number of Branches 6WAI	Number of Branches 8WAI	Number of Leaves 2WAI	Number of Leaves 4WAI	Number of Leaves 6WAI	Number of Leaves 8WAI
Tomato only(Control)	1.67a	2.00a	2.33a	3.44a	68.67a	107.3a	132.3a	161.40a
Tomato+ Tithonia slurry	1.33a	2.33a	2.67a	3.21a	73.00a	129.00a	144.00a	158.20a
Tomato+ Neem slurry	1.00b	1.67ab	2.00ab	2.51ab	59.00ab	75.67ab	86.67ab	87.90ab
Tomato+M.incognita + Tithonia slurry	1.00b	1.10b	1.45b	1.92ab	26.67bc	71.00ab	80.33ab	95.30ab
Tomato+M.incognita + Neem slurry	0.33c	1.00c	1.33c	1.54b	13.67c	44.00c	61.33c	76.26c

Means with the same letter(s) are not significantly different from each other. ($p \leq 0.05$)

P.H: Plant height (cm), S.G: Stem girth (mm), NOL: Number of leaves, NOB: Number of branches, WAI: Weeks after inoculating

Table 3 shows the evaluation of *Meloidogyne incognita* on fresh root weight, nematode population, egg population and number of galls in 2014

Fresh root weight

Tomato + *M.incognita* + Tithonia (33.83) had the highest number of root weight but was not significantly different ($p\leq 0.05$) from Tomato+ Neem slurry (22.03) also, control (Tomato only) (19.50) and Tomato+ Tithonia slurry (13.37) were not significantly different($p\leq 0.05$) from each other, compared to Tomato + Neem slurry (10.20) with the lowest number root weight.

Nematode population

There was absence of nematode in Control (Tomato only) (0), Tomato +Tithonia slurry (0) and Tomato+ Neem slurry (0) because it was not inoculated with *Meloidogyne incognita*. There is statistically significance between Tomato+ *M.incognita* + Tithonia (384) and Tomato+ *M.incognita* + Neem (1640).

Egg population

There was absence of eggs in Control (Tomato only) (0), Tomato +Tithonia slurry (0) and Tomato+ Neem slurry (0) because it was not inoculated with *Meloidogyne incognita.* Application of Tomato+ *M.incognita* + Tithonia (77) significantly reduced the number of eggs than Tomato+ *M.incognita* + Neem (179).

Number of galls

There were no presence of galls in Control (Tomato only) (0), Tomato +Tithonia slurry (0) and Tomato+ Neem slurry (0) because it was not inoculated with *Meloidogyne incognita*. Application of Tomato+ *M.incognita* + Tithonia (15) significantly reduced the number of galls than Tomato+ *M.incognita* + Neem (52).

Table 3: Effect of Neem and Tithonia as botanicals on fresh root weight, number of galls, egg population, and nematode population in 2014

Treatments	Fresh Root Weight(g)	Nematode population	Egg population	Number of galls
Tomato only(Control)	19.50b	0	0	0
Tomato+ Ttithonia slurry	13.37b	0	0	0
Tomato+ Neem slurry	10.20c	0	0	0
Tomato+ M.incognita + Tithonia slurry	22.03a	384b	77ab	15b
Tomato + M.incognita Neem slurry	33.83a	1640a	179a	52a

Means with the same letter(s) are not significantly different from each other. ($p \leq 0.05$)

FRW: Fresh root weight

Table 4 shows the evaluation of *Meloidogyne incognita* inoculation on plant height and stem girth of tomato in 2015

Plant height

Plant height in the various treatments at two (2) weeks after inoculation showed no significant differences among each other ($p \leq 0.05$), while at four (4) weeks after inoculation the treatments showed significant differences between each other. Control (Tomato only) (39.25) produced the highest number of plant height but were not significantly different ($p \leq 0.05$) from Tomato + Tithonia slurry + *M. incognita* (32.42), also Tomato + Neem slurry + *M. incognita* (30.65) had higher number of plant height compared to Tomato + Tithonia +Neem + *M. incognita* at 1% (27.75), Tomato + Nematode (21.60) had the lowest number of plant height in all the treatments. At six (6) weeks after inoculation. Control (Tomato only) (40.59) had the highest number of plant height while Tomato + Nematode (27.71) had the lowest number of plant height. At eight (8) weeks after inoculation Control (Tomato only) (46.75) had the highest number of plant height but not significantly different p≤0.05 from Tomato + Tithonia slurry + *M. incognita* (43.45), while Tomato + Nematode (30.00) had the lowest number of plant height in all the treatments.

Stem girth

At two (2) weeks after inoculation Control (Tomato only) (1.23) had the widest stem girth, while Tomato + Tithonia slurry + *M. incognita* (1.80), also Tomato + Tithonia + Neem + *M. incognita* at 1% (1.08) was not significantly different from Tomato + Tithonia + Neem + *M. incognita* at 0.5 % (0.95) while), Tomato + Nematode (0.78) had the lowest number of stem girth. At four (4) weeks after inoculation there was no significant difference p≤0.05 among each treatment. At six (6) weeks after inoculation there was also no difference p≤0.05 among the treatments except for Tomato + Nematode (1.09) with the lowest number of stem girth. At eight (8) weeks after inoculation Control (Tomato only) (1.61) had the widest number of stem girth but was not significantly different from other except for Tomato + Nematode (1.12) with the lowest number of stem girth.

Table 4: Evaluation of *Meloidogyne incognita* of on plant height and stem girth of tomato in response to Neem and Tithonia as botanicals in 2015

Treatment	Plant height 2WAI	Plant height 4WAI	Plant height 6WAI	WAI Plant hei ght 8WAI	Stem girth 2WAI	Stem girth 4WAI	Stem girth 6WAI	Stem girth 8WAI
Tomato only(Control)	20.45a	39.25a	40.59a	46.75a	1.23a	1.05a	1.50a	1.61a
Tomato +Nematode	17.27a	21.60d	27.71c	30.00c	0.78c	1.20a	1.09 b	1.12c
Tomato+ Neem Slurry + M.incognita	17.77a	30.65b	35.10ab	41.32ab	1.8ab	1.30a	1.45a	1.52ab
Tomato+ Tithonia Slurry+ M.incognita	18.77a	32.42a	37.91a	43.45a	1.20ab	1.30a	1.49a	1.55a
Tomato+ Neem & Tithonia Slurry+ M.incognita 1%	17.73a	27.75c	33.82b	38.42ab	1.08a b	1.30a	1.41a	1.51ab
Tomato+ Neem & Tithonia Slurry+ M.incognita 0.5%	22.18a	26.82c	32.45b	37.65ab	0.95ab	1.28a	1.40a	1.50ab

Means with the same letter(s) are not significantly different from each other. ($p \leq 0.05$)

P.H: Plant height, S.G: Stem girth, NOL: Number of leaves, LA: Leaf area, WAI: Weeks after inoculating.

Table 5 shows the evaluation of *Meloidogyne incognita* inoculation on leaf area and number of leaves of tomato in 2015

Leaf area

Leaf area at two (2), four (4), six (6) and eight (8) weeks after inoculation all treatments were not significantly different $p \leq 0.05$ from each other.

Number of leaves

At two (2) weeks after inoculating number of leaves were not significantly different from each other, while at four (4) weeks after inoculating Control (Tomato only) (66.75) produced the highest number of leaves compared to Tomato + Nematode (27.75) had the lowest number of leaves. Also, at six (6) weeks after inoculating Control (Tomato only) (72.10) gave the highest number of number of leaves but was not significantly p≤0.05 different from Tomato +Tithonia slurry + *M. incognita* (68.05) while Tomato + Nematode (32.00) had the lowest number of leaves. The control (Tomato only) (72.10) produced the highest number of leaves but was also not different from Tomato +Tithonia slurry + *M. incognita* (72.80) compared to Tomato + Nematode (45.57) had the lowest number of leaves at eight (8) weeks after inoculation.

Table 5: Evaluation of *Meloidogyne incognita* of on leaf area and number of leaves of tomato in response to Neem and *Tithonia* as botanicals in 2015

Treatment	Leaf area 2WAI	Leaf area 4WAI	Leaf area 6WAI	WAI Leaf area 8WAI	Number of leaves 2WAI	Number of leaves 4WAI	Number of leaves 6WAI	Number of leaves 8WAI
Tomato only(Control)	99.17a	106.50a	111.60a	116.91a	46.00a	66.75a	72.10a	74.22a
Tomato + M.incognita	95.95a	92.80a	98.70a	100.1a	36.50a	27.75e	32.00c	45.57c
Tomato+ Neem Slurry + M.incognita	99.00a	102.70a	106.00a	111.00a	41.00a	42.75b	56.10ab	64.80ab
Tomato+ Tithonia Slurry+ M.incognita	99.10a	106.40a	110.24a	117.50a	44.80a	49.00ab	68.05a	72.80a
Tomato+Neem & Tithonia Slurry+ M.incognita 1%	98.70a	102.50a	105.91a	110.35a	38.80a	40.50bc	53.55ab	60.70ab
Tomato+Neem & Tithonia Slurry+ M.incognita 0.5%	98.72a	102.00a	101.41a	107.30a	38.80a	37.00bc	46.30ab	55.00abc

Means with the same letter(s) are not significantly different from each other. (p≤0.05)

P.H: Plant height, S.G: Stem girth, NOL: Number of leaves, LA: Leaf area, WAI: Weeks after inoculating.

Results of the evaluation of *Meloidogyne incognita* on fresh root weight, nematode population, egg population and number of galls in 2015 is presented in Table 6.

Fresh root weight

Tomato + Nematode (26.50) had the highest number of fresh root weight compared to Tomato+ Tithonia slurry + *M.incognita* (14.80) with the lowest number of fresh root weight.

Nematode population

There was absence of nematode in Control (Tomato only) (0) because it was not inoculated with *M. incognita*, Tomato+ *M. incognita* (61.0) had the highest nematode population and was significantly different $p \leq 0.05$ from Tomato + Tithonia slurry+ *M. incognita* (44.0) with the lowest nematode population.

Egg Population

There was absence of eggs in Control (Tomato only) (0) because it was not inoculated with *M. incognita*, Tomato + *M. incognita* (5798) had the highest egg population in all the treatment, while the application of Tomato + Tithonia slurry+ *M. incognita* (2378) significantly reduced the number of eggs than Tomato+ Neem + *M. incognita* 3458).

Number of Galls

There were no presence of galls in Control (Tomato only) (0) because it was not inoculated with *M. incognita*, Tomato + Tithonia slurry + *M. incognita* (52) had the highest number of gall. Tomato+ *M. incognita* + Tithonia (18.8) significantly reduced the number of galls in all treatments.

Table 6: Evaluation of *Meloidogyne incognita* on fresh root weight, number of galls, egg population, nematode population and gall index in response to Neem and *Tithonia* as botanicals in 2015

Treatment	FRW(g)	Number of galls	Egg population	Nematode population	Gall index
Tomato only (Control)	20.50a	0d	0d	0d	0d
Tomato + M.incognita	26.50a	52.0a	5798a	61.0a	4.0a
Tomato+ Neem Slurry+ M.incognita	19.20b	24.5bc	3458b	50.00b	3.75ab
Tomato+ Tithonia Slurry + M.incognita	14.80b	18.8c	2378c	44.0c	3.0c
Tomato+Neem &Tithonia Slurry+ M.incognita at 1%	23.50a	32.5b	3015b	52.25b	3.25ab
Tomato+ Neem &Tithonia Slurry+ M.incognita at 0.5%	22.50a	27.0bc	3172b	56.75a	3.25ab

Means with the same letter(s) are not significantly different from each other. ($p \leq 0.05$)

FRW: Fresh root weight

DISCUSSION

Plant pesticide can suppress the root-knot nematodes and also result in positive changes in plant growth. The application of botanical slurry improved growth parameters (plant height, stem girth, number of leaves and number of branches). This finding of this research are also in agreement with those of Aiyadurai *et al.* (2018) who suggested that plant parasitic nematodes can be controlled with the application of botanicals. There was no significant different in terms of fresh shoot and root weight. This result was also similar to those obtain by Wondimeneh *et al*. (2013). As far as the plant growth characters are concerned, the applications of botanicals improved the plant height. The plant height, stem girth, number of leaves, leaf area and number of branches recorded in uninoculated plant with Tithonia slurry and uninoculated plant with neem slurry increased significantly. This results of this study agree with those by Rajesh *et al*. (2017) who reported that crude extracts from Neem leaf extracts at 100ml/L and Tithonia 100 ml/L were most effective and increased the yield significantly untreated plants decreased due to the reproduction of the nematode at the root and no plant extract to reduce the nematode population. The addition of botanicals to soil leads to a better environment for the growth of the roots. This enhances the utilization of soil nutrients, as a consequence of which the nematode damage might have been markedly reduced (Abubakar *et al*., 2004).

Number of egg extracted from the root and nematode from the soil were extremely lower than the number of eggs used for inoculation. Inoculated plant with Tithonia gave lower egg and lower *Meloidogyne incognita* while inoculated plants with Neem gave higher number of eggs and number of *Meloidogyne incognita.* This suggest that *Tithonia* proved to be more effective in term of growth than Neem and clearly showed that Tithonia have nematicidal growth inhibitory effect than Neem. Number of galls on the treated plant was reduced while the number of galls of the untreated gave increase, also it was observed that tomato plant treated with *Tithonia* extract slows down the nematode population than tomato plant treated with Neem extract according to Coyne *et al*. (2005). The combination of both *Neem* and *Tithoni*a at 1% also slows down the nematode population than the combination of both *Neem* and *Tithonia* at 0.5% because of the high concentration. Chitwood (2002) suggested that the nematicidal properties of plant species vary considerably with plant species and cultivar, the plant tissue used, plant growth stage, application method and nematode species tested. Also the combination of both Neem and *Tithonia* at 1% controls the nematode population than the combination of both Neem and *Tithonia* at 0.5%. The importance of organic horticultural production, which avoid synthetic nematicides applications, increased the research on botanical pesticides with potential use for nematode management (Mishra *et al* 2018). The toxicity of extract increases with increase in their concentration as well as exposure time. Various neem products including neem cake, its oil and nimin (containing neemtriterpenes) as urea coating agents, and root-dip or seed treatment with neem extracts, have been found to be nematicidal against several species of parasitic nematodes (Alam, 1991) attacking vegetables and legumes (Haseeb *et al*., 2005).Chemical analysis of Neem (*Azadicrachta indica*) contains tannins and azadirachtin as active ingredient (Akhtar, 2000, Fatoki, 2001) which are detrimental to nematode survival.

CONCLUSION

The study concluded that, although, both *Tithonia diversifolia* and *Azadirachta indica* can be used to suppress nematode population and improve the Roma VF tomato plant growth, *T. diversifolia* is effective in nematode management strategy. The combination of both *Tithonia* and *Neem* at 1% reduced nematode (*Meloidogyne incognita*) population than the combination of both *Tithonia* and *Neem* at 0.5%. Thus, this study has clearly enlightened that Neem and Mexican sunflower appeared to be promising botanicals. However, further research is needed to evaluate their efficacy under field condition of different agro-ecologies and improve their application technology.

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