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Biochemical mechanism of Lantana camara leaf extracts in the management of Meloidogyne incognita on tomato

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Abstract

An experiment was carried out on the mechanism of Lantana camara leaf extract in the management of Meloidogyne incognita on tomato. For this, leaf extract of L. camara (25gm/75ml w/v) were evaluated at 25, 50, 75 and 100 percent concentration through egg hatch inhibition and larval mortality test. Invitro efficacy test showed that maximum egg hatch inhibition and J2 mortality of M. incognita were recorded in the 100 percent concentration of L. camara leaf extract. A pot experiment study recorded maximum plant growth parameters were recorded in the 50 percent concentration whereas; minimum nematode multiplication was recorded in the 100 percent concentration of L. camara leaf extract after 35 and 45 days after inoculation. Further, biochemical analysis of tomato root revealed that maximum activity of the peroxodase, polypheno oxidase and total phenol content was recorded at 35 DAI while the same declined at 45 DAI.

Keywords: L. camara, M. incognita, peroxidase (PO), polyphenoloxidase (PPO), tomato

Tomato (Lycopersicon esculentum Mill.) is one of the most widely grown vegetable crops in the world and widely cultivated in tropical, sub-tropical and temperate climates (Valencia et al. [40]). Tomato fruits are rich in minerals (potassium, magnesium, calcium, iron and zinc), proteins (essential amino acids), citric acid, sugars, dietary fibers (pectin) and high levels of vitamin C, lycopene, and betacarotene which are anti-oxidants against oxygen free radicals that probably cause cancer, aging and arteriosclerosis Schieber *et al.* [30], Sogi *et al.* [35], Knoblich *et al.* [21], Naika *et al.* [26], Calvo *et al.* [14] and El-Adawy and Taha [17]. In Assam, tomato is one of the most important and profitable vegetable crops that give higher income to either large- or small-scale farmers. The total area under production reaches 18.18 (in thousands) hectares and annual production is estimated to be 415.41(in thousands) metric tons in Assam during 2016-17 Anonymous [7]. The production of tomato may be affected by abiotic factors like poor soil fertility, unreliable rainfall patterns etc. and biotic factors like pests and diseases. However, it is more prone to diseases than any other vegetables. In Assam, tomato is heavily infected by root-knot nematode (Meloidogyne incognita). In Assam, M. incognita has been reported to cause an avoidable loss in yield of 13.20 per cent on tomato Anonymous [8]. Although application of chemical nematicides have been found to be most effective measure for the control of this nematode, due to its high toxic residual effect and toxicity to beneficial flora and fauna in the soil, there is need to develop alternative and cost effective nematode control strategies Akhtar [4], Siddiqui and Alam [34]. One of the outstanding alternative control measures against the nematode is the application of plant extracts Oka et al. [28]. Plant extracts have been shown to contain various bioactive compounds (allelochemicals) like phenolics, organic acids, terpenes and terpenoids, coumarin-like compounds and other secondary metabolites and reported to have nematicidal activity Muller and Gooch [25] and Shaukat et al. [33]. These allelochemicals that inhibit the growth of some species at certain concentrations might in fact stimulate the growth of the same or different species at different concentrations Narwal [27]. It has been documented that allelopathy play an important role in plant-plant interference by those chemical compounds Turk and Tawaha [37]; Turk et al. [38] and Ashrafi et al. [9]. A wide variety of plant species, representing 57 families have been shownto have nematicidal compounds Sukul [36]. Among them, Lantana camara L. belonging to family the Verbinaceae have been shown to have allelopathy.

The biochemical assays indicate that *Lantana camara* root extracts increase the enzymatic activity in the root. The root of *L. camara* contains phenolic compounds and hydrogen cyanide which showed nematicidal activity Shaukat *et al.* [33]. However, allelopathic plants at higher concentrations may well produce phytotoxic symptoms. Before application of these plants under field conditions to suppress the plant parasitic nematodes, it is necessary to determine the optimal concentration that is toxic to nematodes but neither to the plant to be protected neither from nematodes nor to any associated beneficial microorganism such as those possessing bio-control and growth promoting properties. Keeping these in view, the present investigations have been undertaken on biochemical mechanism of *Lantana camara* leaf extracts in the management of *Meloidogyne incognita* on tomato.

Material and Methods Nematode inoculums

Nematode inoculums were prepared according to method given by Annpurana et al. [6] with slight modifications. The egg masses of M. incognita were collected from pure culture plant i.e tomato, where the nematodes were maintained. Eggmasses were collected from infected tomato plant and surface sterilized with 0.4per cent sodium hypochlorite (NaOCl) for 60 seconds. Egg masses were washed thoroughly with distilled water until the traces of NaOCl is removed and kept in petriplate for further use. For dissolving gelatinous matrix of egg massess, sterilized egg masses were kept in a petridish and subjected to 0.5 % NaOCl solution for one minutes, with frequent stirring followed by a 40 seconds settleling. Further, eggs were washed thoroughly with distilled water to remove the traces of NaOCl. The egg suspension was prepared in such a way that 0.5 ml of it contained 50 eggs. For the extraction of juveniles (J₂), the sterilized eggs were kept in required quantity of distilled water at room temperature for hatching. Several such assemblies were maintained. The juveniles collected from these were mixed together at the time of inoculation in pot experiment as also in-vitro studies. The juvenile suspension was prepared in such a way that 0.5 ml of it contained 50 juveniles.

Preparation of Lantana camara leaf extract

The fresh green leaves of *Lantana camara* were collected from AAU Campus., Jorhat-13. The *L. camara* leaf extract was prepared according to method given by Ahmad *et al.* [1] For this, the 25 grams of fresh green leaves of *L. camara* were thoroughly washed, chopped and after grinding soaked in 75 ml of distilled water for 24 hrs. After filtering through two folds of muslin cloth, they were filtered through Whatmann No.1 filter paper. The filtrate was centrifuged at 2400 rpm for 10 min and clear supernatant was stored at 4 °C for experimentation. The supernatant was considered as standard solution 'S' and other different concentrations were prepared from the standard solution by adding requisite amount of sterilized distilled water at the time of experiment.

Effect of L. camara leaf extracts on juvenile (J_2) mortality of M. incognita

The mortality test was conducted under *in-vitro* conditions. For the test, desired concentrations (25, 50, 75 and 100 percent) of leaf extracts were poured on the sterile cavity blocks containing 100 juveniles (J₂) per cavity block. Observation on juvenile mortality was recorded at 24, 48, 72 and 96 hours of exposure. Apart from the treatments with

different concentration of *L. camara* leaf extracts and sterilized distilled water (SDW) were also maintained as controls. The test was replicated four times. For determining the dead nematodes revival test was conducted by transferring the immobile juveniles to distilled water and observed their activities after one day. The juveniles that showed no movement even when they were probed with bamboo splinter were considered dead. The percentage of juvenile mortality was calculated using the formula given below:

$$Mortality(\%) = \frac{\text{Number of dead juve niles in the treatment}}{\text{Total number of juve niles}} \times 100$$

Effect of L. camara leaf extracts on hatch inhibition of M. incognita eggs

The hatch inhibition test was conducted under *in-vitro* conditions. For the test, desired concentrations (25, 50, 75 and 100 percent) of leaf extracts were poured on the sterile cavity blocks containing 100 *M. incognita* eggs per cavity block. Observation on hatch inhibition was recorded at 24, 48, 72 and 96 hours of exposure. Apart from the treatments with different concentration of *L. camara* leaf extracts and sterilized distilled water (SDW) were also maintained as controls. The test was replicated four times. Observations were recorded after 7 days of exposure. The percentage hatch inhibition of eggs was calculated using the formula given below:

Egg hatch inhibition(%) =
$$\frac{\text{(Total number of eggs-Number of eggs hatched)}}{\text{Total number of eggs}} \times$$

Biochemical mechanism of leaf extracts of *Lantana* camara in the management of *Meloidogyne incognita* on tomato

An experiment was conducted in the net house of the Department of Nematology, AAU Jorhat during winter season of 2018-2019 to know the biochemical changes influenced by L. camara leaf extracts on tomato infected by M. incognita under pot conditions. For that required amount of soil was collected from upland near the nematode culture house, Department of Nematology, Assam Agricultural University, Jorhat. The soil was mixed thoroughly after removing unwanted materials like stones and roots etc. Then the soil was mixed homogenously with finely dried cow dung and sand in the ratio of 2:1:1, respectively. The soil mixture was put in a gunny bag and sterilized in an autoclave at 121°C for half an hour. Plastic pots of 1 kg capacity were selected, cleaned and sterilized in sunshine for conducting the experiment. Few broken pieces of bricks were placed at the bottom of the pots before filling up with sterilized soil mixture. Proper labelling of each pot was done. The pots (1kg capacity) were arranged in a completely randomized design with five replications for each treatment. All the pots were transplanted with 25 days old seedlings of tomato. The pots were inoculated with freshly hatched second stage juveniles of M. incognita @ 1J₂/cc soil and thereafter drenched with different concentrations of L. camara leaf extract viz., 25, 50, 75 and 100 percent. Two control treatments viz., M. incognita alone (@ 1J₂/cc soil) and uninoculated and untreated control were maintained. Tomato seeds of the variety Pusa Ruby (highly susceptible to M. incognita) were obtained from the market. Tomato seeds were washed with sterilized distilled water and sown in tray containing sterilized soil. The seedlings were watered regularly and 25 days old seedlings of tomato were transplanted in pot. The pots were inoculated with freshly hatched second stage juveniles of M.

incognita @ 1J₂/cc soil and thereafter drenched with different concentrations of L. camara leaf extract viz., 25, 50, 75 and 100 percent. Two control treatments viz., M. incognita alone (@ 1J₂/cc soil) and uninoculated and untreated control were maintained. The treatments are as follows:T₁- Uninoculated and Untreated control, T₂- M. incognita @ 1J₂/cc soil, T₃-M. incognita @ 1J₂/cc soil+ Soil drenching of L. camara leaf extracts @ 25 %, T₄- M. incognita @ 1J₂/cc soil+ Soil drenching of L. camara leaf extracts @ 50 %, T₅- M. incognita @ 1J₂/cc soil+ Soil drenching of L. camara leaf extracts @ 75 % and T₆- M. incognita @ 1J₂/cc soil+ Soil drenching of L. camara leaf extracts @ 100 %. After 35 and 45days from nematode inoculation, the plants were uprooted and the soil adhering to the roots was removed (250 g/pot) by gentle agitation in water. The observations on plant growth parameters viz., shoot and root length (cm), fresh shoot and root weight (gm), dry shoot weight (gm) were recorded. Further, nematode multiplication in terms of number of galls per root system, egg masses per root system, and nematode populations of J₂ were extracted and counted. The fresh roots from each treatment were utilized for the analysis of enzyme activity viz., peroxidase (PO), polyphenol oxidase (PPO) and total phenol content were carried out at Department of Biochemistry and Agricultural Chemistry. Each treatment replicated five times in completely randomized design.

Enzyme extraction

Root samples (1gm) were collected from the tomato plants and immediately homogenized with liquid nitrogen. One gram of powdered sample was extracted with 2 ml of sodium phosphate buffer, 0.1 M (pH 7.0) at 4 °C. The homogenate was centrifuged at 16,000 rpm at 4 °C for 20 min at 4000×g protein extract prepared from the roots were used to estimate peroxidase(PO) and polyphenol oxidase (PPO).

Peroxidase activity

The reaction mixture contained of 1.5 ml of 0.05 M pyrogallol, 0.5 ml of 1% H_2O_2 and 0.5 ml of enzyme extract. The reaction mixture was incubated at 28 ± 1 °C temperature. Changes in absorbance at 420 nm were recorded at 60s intervals for 3 min and the enzyme extract served as a blank. Enzyme activity was expressed as the change in the absorbance of the reaction mixture $min^{-1}\mu g^{-1}$ on a fresh weight basis Hammerschmidt $et\ al.$, [19].

Polyphenol oxidase (PPO) activity

PPO activity was determined following the methods of Mayer *et al.*, ^[24]. The reaction mixture contained of 1.5 ml of 0.1 M sodium phosphate buffer (pH 6.5) and 200 μ l of the enzyme extract which was centrifuged at 16,000 rotations per minute for 15 min at 4 °C and the supernatant was used as enzyme source. To start the reaction, 200 μ l of 0.01 M catechol was added and the activity was expressed as changes in absorbance at 495 nm min⁻¹ μ g⁻¹ fresh weight of root.

Estimation of Total phenols

Total phenol was estimated by Folin – ciocalteu method given by Bray and Thorpe [13]. For estimation of total phenol content, weighed 1 gm of tomato root and grinded it in 10 times volume of 80.00 percent ethanol with help of grinder. Centrifuged the homogenate at 10,000 rotations per minute for tewnty (20) minutes and saved the supernatant. Reextracted the residue with five times the volume of 80.00 percent ethanol, centrifuged and pooled the supernatants. Then evaporated the supernatant to dryness and dissolved the

residue in 5ml of distilled water. Then pipetted out different aliquots (0.2 to 2 ml) into test tube and made up the volume in each tube to 3 ml with and add 0.5 ml of Folin – ciocalteu reagent. After 3 minutes, 2 ml of 20% Na₂CO₃ solution added to each tube and placed the tubes in a boiling water bath for exactly 1 minute, cooled and measured the absorbance at 650 nm against a reagent blank. The standard curve was prepared using different concentration of catechol (ug).

Results and Discussion

Bioefficacy of Lantana camara leaf extracts against Meloidogyne incognita

The data on egg hatch inhibition and juvenile (J₂) mortality of M. incognita in different concentration of L. camara leaf extracts are presented in Table 1. All the concentrations of leaf extract showed juvenile (J₂) mortality and inhibition of egg hatching of M. incognita as compared to control. No egg hatch inhibition and juvenile (J₂) mortality was recorded in control (SDW). It was observed that with increase in concentrations there was increase in the egg hatch inhibition and juvenile (J₂) mortality of M. incognita. At 25 percent concentration of L. camara leaf extracts the minimum egg hatch inhibition as well as juvenile (J₂) mortality followed by 50 per cent and 75 per cent concentration of L. camara. Maximum egg hatch inhibition and juvenile (J₂) mortality was recorded with 100 per cent concentration of L. camara. It was observed that there was an increasing trend in mortality of J₂ with increase in concentration of *L. camara* leaf extracts and time of exposure. The maximum (91.60%) mortality of M. incognita J₂ was recorded in the 100 percent concentration of L. camara leaf extracts at 96 hours of exposure time whereas minimum (43.80%) was recorded at 25 percent concentration after 96 hours of exposure time. Similarly, egg hatch inhibition was also found to be more with increase in the concentration. These results are in agreement with the findings of Akhtar and Mahmood [3], Shaukat et al. [33], Ali et al. [5] and Bhuyan [12] who reported that highest concentrations of leaf extracts of L. camara caused maximum mortality of J₂ and egg hatch inhibition of M. incognita under invitro conditions. The plant extracts contain different chemical compounds like aldehydes, phenolics, fatty acids, alcohols and terpinoids etc. which showed nematicidal activities like inhibition of egg hatching and/or repulsion of J₂ nematodes Chitwood ^[15]. Apart from this, plant extracts also contain bioactive principles like phenolics, organic acids, terpenes and terpenoids, caumarin-like compounds and other secondary metabolites that show nematicidal activity against root knot nemaotdes Muller and Gooch [25] and Shaukat et al. [33]. Similarly, Begum et al. [11] isolated chemical compound like pomolic acid, lantanolic acid, lantonic acid, camarin, lantacin, camarinin and urosolic acid from L. camara leaf and reported that these compounds exhibit 100 percent mortality of M. incongita J₂ under in-vitro condition. However, nematodes eggs are semi-permeable in nature and allow certain molecules or ions to pass through it. So toxin had permanent adverse effect on eggs or there appears to be physical retention of toxins in eggs Clark and Perry [16], as hatching was not resumed on transfer of eggs in water after one day. The reason behind showing the nematicidal properties by various plant extracts might be attributed to the lipophilic properties of oxygenated compounds which they dissolve the cytoplasmic membrane of nematodes, thus interfere with enzyme protein structure Knoblock et al. [22]. The application of plant extracts suppressed the enzyme acetylcholine esterase activity to the tune of more than 75

percent in *Helicotylenchus dihystera* as reported by Koraytem *et al.* ^[23]. Similar kind of mechanism might be operative in the present investigation leading to mortality of juveniles and egg hatch inhibition of *M. incognita*.

Biochemical mechanism of leaf extracts of *Lantana* camara in the management of *Meloidogyne incognita* on tomato

A pot experiment was conducted to explore the biochemical mechanism of leaf extracts of L. camara in the management of M. incognita on tomato. For this the activity of defence related enzymes viz., peroxidase (PO) and polyphenoloxidase (PPO) and total phenol content were observed at 25, 50, 75 and 100 percent concentration of L. camara leaf extract after 35 and 45 DAI of M. incognita in tomato. Among the different concentrations, highest activity of peroxidase (PO), polyphenoloxidase (PPO) and total phenol content was observed at 100 percent concentration while the minimum activity was recorded in the 25 percent concentration both after 35 and 45 DAI. However, maximum activity of the biochemical compounds was recorded at 35 DAI while the same declined at 45 DAI. The lower concentration of L. camara leaf extract viz., 25 and 50 percent showed stimulatory effect on plant growth parameter whereas higher concentrations viz., 75 and 100 percent concentration of L. camara leaf extract showed inhibitory effect on plant growth. Ullah et al. [39] observed the application of Phytolaccalatbenia extract significantly increased the catalase activity and malondialdehyde content in Brassica napus and Triticum the increasing concentrations with aestivum Phytolaccalatbenia extracts. Bano and Naz et al. [10] evaluated at higher concentration of L. camara leaf extracts significantly increases the enzyme activity like peroxidase (PO), polyphenoloxidase (PPO) and total phenol in root of maize as compared to control. Further, they reported that higher concentration of extracts had strong inhibitory effects; whereas the lower concentration showed stimulatory effect in maize root. These findings are in agreement with the results of the present investigation.

Effect of *L. camara* leaf extracts on plant growth parameters on tomato infected by *M. incognita*

The data on plant growth parameters in different treatments have been presented in table 3. There was a significant increase in plant growth parameters viz., shoot height, fresh shoot weight, root length, fresh root weight and dry shoot weight from 25 to 50 percent concentration of L. camara leaf extract and thereafter declined at 75 and 100 percent concentration at 35DAI. At 35 DAI, the maximum shoot height (38.42 cm), fresh shoot weight (11.25 gm), root length (19.52 cm), fresh root weight (8.26 gm) and dry shoot weight (8.16 gm) was recorded in the treatment T₁ (untreated and uninoculated control) and it was significantly different from rest of the treatments. Among the L. camara leaf extracts, maximum shoot height, (38.42 cm), fresh shoot weight, fresh root length, root weight and dry shoot weight was recorded in the treatment, M. incognita + Soil drenching of L. camara leaf extract at 50 % (T_4) , followed by the treatment M. incognita + L. camara leaf extract at 25% (T₃), T₅ (M. incognita +L. camara leaf extract at 75%) and T₆ (M.

incognita +L. camara leaf extract 100%). Similar trend of improvement in shoot height, fresh shoot weigh, root length, fresh root weight and dry shoot weight was recorded at 45 DAI and all treatments were found significantly different from each other. Similar effects of L. camara extract on plant growth were also reported by Inderjit et al. [20] in Raphanus sativus, Shaukat et al. [33] in Vigna mungo and Ahmed et al. [2] in B. juncea, Cucumis sativus, Phaseolus mungo, Raphanus sativus, V. unguiculata and Cicer arietinum and Bano and Naz et al. [10] reported that at higher concentration of L. camara extract there was reduction in plant growth parameters whereas at lower concentration had the stimulatory effect on plant growth parameters. In the present investigation although there was significant reduction in number of galls, number of eggmass on roots and nematode population in soil at higher concentrations (75% and 100%) corresponding increase in plant growth parameters were not observed. This might be due to fact that L. camara is an allelopathic plant and at higher concentration, its leaf extract might have phytotoxic effects.

Effect of *L. camara* leaf extracts on infection and multiplication of *M. incognita* on tomato.

The data on effect of L. camara leaf extracts on infection and multiplication of *M. incognita* in tomato at 35 and 45 DAI are presented in Table 4. All the treatments with L. camara leaf extracts significantly reduced the number of galls and eggmasses per root system and final nematode population in soil as compared to control (nematode alone) at 35 and 45 DAI. The minimum number of galls and eggmasses per root system and final nematode population in soil was recorded in the treatment T_6 (M. incognita + L. camara leaf extract at 100%) and maximum was recorded in T₂ (nematode alone). All the treatments were found to be significantly different from each other. The results show that among the L. camara leaf extracts, T₆was found to be most effective in reducing nematode infection and multiplication followed by $T_5(M.\ incognita + L.\ camara\ leaf\ extract\ at\ 75\%)$, $T_4(M.\ incognita + L.\ camara\ leaf\ extract\ at\ 75\%)$ incognita +L. camara leaf extract at 50%) and T₃ (M. incognita + L. camara leaf extract at 25 %). It means that less number of eggmasses, number of galls and final nematode population in soil were recorded in treatment receiving 25 percent concentration of L. camara leaf extract. Similar type of effect of L. camara extract on M. incognita were recorded by Ali et al. [5] and Shaukat and Siddiqui [31] in V. mungo, Radwan et al. [29] in tomato and El-Nagdi et al. [18] in V. uniguiculata. They observed that application of L. camara extract in soil significantly reduced the number of root galls, eggmasses per root system and nematode population of M. incognita in soil. Shaukat and Siddiqui [32] reported L. camara extract contains phenolic compounds like caffeic acid, p-hydroxybenzoic acid and p-coumaric acid which inhibited M. javanica in the soil. Shaukat et al., [33] observed that L. camara root leachate released the hydrogen cynide (HCN) in soil and that affect on the multiplication of M. *javanica* either in plant or in soil. The *L. camara* also releases the camaric acid, camarinic acid, Lantanolic acid, linaroside, Oleanonic acid and showed nematicidal activity (Begum et al. [11]).

Table 1: In-vitro efficacy of Lantana camara leaf extracts on egg hatch inhibition and juvenile mortality of Meloidogyne incognita

	Concentration	Egg hatch inhibition (%)	Larval mortality (%)						
	Concentration (%)	Exposure time							
		7 day	24 hr	48 hr	72 hr	96 hr			
L. camara	25	21.40 (4.67) ^d	19.40 (4.45) ^d	25.60 (5.10) ^d	37.20 (6.13) ^d	43.80 (6.65) ^d			
	50	29.40 (5.46) ^c	24.80 (5.02) ^c	33.20 (5.80) ^c	43.20 (6.60) ^c	52.80 (7.30)°			
	75	48.40 (6.99) ^b	38.80 (6.26) ^b	56.00 (7.51) ^b	66.20 (8.16) ^b	78.60 (8.89) ^b			
	100	66.40 (8.17) ^a	52.80 (7.30) ^a	71.80 (8.50) ^a	80.00 (8.97) ^e	91.60 (9.59) ^a			
SDW	-	0.00 (0.70) ^e							
S.Ed±		1.67	1.25	1.24	1.17	1.30			
CD@0.05		3.33	2.49	2.48	2.33	2.59			

The values in the parenthesis are square root transformation before analysis

Table 2: Induction of defense enzyme activity of Lantana camara leaf extracts in tomato affected by Meloidogyne incognita

Treatment		nge in absorbance m ⁻ ¹ protein)	Polyphenol o absorbance	xidase (change in m ⁻¹ μg ⁻¹ protein)	Total Phenol (µg/g fresh root)		
	35DAI	45DAI	35DAI	45DAI	35DAI	45DAI	
T_1	2.56a	2.15 ^a	0.33a	0.10^{a}	2.18 ^a	1.20a	
T ₂	2.77 ^b	2.45 ^b	0.38 ^b	0.16 ^b	5.26 ^b	4.81 ^b	
T ₃	7.08 ^c	4.32°	0.49 ^c	0.29 ^c	6.67 ^c	5.70°	
T ₄	7.50 ^d	7.42 ^d	0.56 ^d	0.33 ^d	8.05 ^d	6.22 ^d	
T ₅	7.86 ^e	7.63 ^e	0.62e	0.39e	11.90e	9.27e	
T ₆	9.13 ^f	8.27 ^f	0.68 ^f	0.45 ^f	13.09 ^f	10.43 ^f	
SE.d±	0.03	0.04	0.01	0.01	0.06	0.01	
CD (0.05%)	0.07	0.08	0.02	0.02	0.12	0.31	

Table 3: Effect of Lantana camara leaf extracts on plant growth parameters of tomato infected by Meloidogyne incognita

	Shoot Length(cm)		Fresh Shoot weight(g)		Fresh Root weight(g)		Root length (cm)		Dry Shoot Weight (g)	
Treatments	35DAI	45DAI	35DAI	45DAI	35 DAI	45DAI	35 DAI	45DAI	35 DAI	45 DAI
T1	40.42a	42.58a	11.25a	15.38a	8.26a	10.22a	19.52a	22.86a	8.16 ^a	10.52a
T2	21.94 ^f	24.98 ^f	7.28 ^f	10.22 ^f	3.24 ^f	6.12 ^f	11.32 ^f	13.38 ^f	4.56 ^f	6.34 ^f
T3	33.00°	36.00 ^c	10.00°	13.40°	6.00°	7.00 ^e	14.24e	15.28e	7.18 ^c	8.20°
T4	36.40 ^b	40.00^{b}	10.36 ^b	14.62 ^b	6.48 ^b	8.30 ^b	17.40 ^b	18.26 ^b	7.64 ^b	8.72 ^b
T5	27.50 ^d	30.28 ^d	9.22 ^d	12.88 ^d	5.70 ^d	7.70 ^c	16.18 ^c	17.26 ^c	6.52 ^d	7.46 ^d
T6	24.76 ^e	27.56 ^e	8.44e	12.10e	5.20 ^e	7.26 ^d	15.26 ^d	16.26 ^d	5.34 ^e	6.94 ^e
S. Ed (±)	1.0	0.61	0.16	0.23	0.10	0.15	0.14	0.17	0.15	0.16
CD(0.05)	2.26	1.35	0.36	0.51	0.23	0.33	0.30	0.39	0.30	0.31

T₁- Uninoculated and Untreated control, T₂- *M. incognita* @ 1J2/cc soil, T₃-*M. incognita* @ 1J2/cc soil + Soil drenching of *L. camara* leaf extract @ 25 %, T₄- *M. incognita* @ 1J2/cc soil + Soil drenching of *L. camara* leaf extract @ 50 %, T₅- *M. incognita* @ 1J2/cc soil + Soil drenching of *L. camara* leaf extract @ 75 % and T₆- *M. incognita* @ 1J2/cc soil + Soil drenching of *L. camara* leaf extract @ 100 %.

Table 4: Effect of Lantana camara leaf extracts on infection and nematode multiplication on tomato infected by Meloidogyne incognita

Treatment	No	of Galls	No. of egg ma	isses N	Nematode population /200 cc soil			
	35 DAI	45 DAI	35 DAI	45 DAI	35 DAI	45 DAI		
T_1	$0.00(0.70)^{f}$	$0.00 (0.70)^{\rm f}$	$0.00(0.70)^{\rm f}$	$0.00(0.70)^{\rm f}$	0.00 (0.70) ^f	$0.00 (0.70)^{\rm f}$		
T_2	93.00 (9.66) ^a	112.80 (10.64) ^a	51.40 (7.20) ^a	74.40 (8.65) ^a	352.40 (18.77) ^a	493.20 (22.21) ^a		
T ₃	45.40 (6.77) ^b	56.00 (7.51) ^b	35.00 (5.95) ^b	43.00 (6.59) ^b	192.40 (13.89 ^b	226.60 (15.06) ^b		
T ₄	41.20 (6.45) ^c	51.00 (7.17) ^c	30.80 (5.59) ^c	37.00 (6.12) ^c	162.40 (12.75) ^c	192.80 (13.90) ^c		
T ₅	38.20 (6.22) ^d	42.60 (6.56) ^d	25.20 (5.06) ^d	33.20 (5.80) ^d	139.00 (11.81) ^d	144.60 (12.04) ^d		
T_6	33.80 (5.85) ^e	38.80 (6.26)e	20.40 (4.57) ^e	25.80 (5.12) ^e	124.40 (11.17) ^e	132.80 (11.54) ^e		
SE.d±	0.05	0.11	0.15	0.10	0.12	0.14		
CD (0.05%)	0.13	0.25	0.34	0.21	0.27	0.32		

T₁- Uninoculated and Untreated control, T₂- *M. incognita* @ 1J2/cc soil, T₃-*M. incognita* @ 1J2/cc soil + Soil drenching of *L. camara* leaf extract @ 25 %, T₄- *M. incognita* @ 1J2/cc soil + Soil drenching of *L. camara* leaf extract @ 50 %, T₅- *M. incognita* @ 1J2/cc soil + Soil drenching of *L. camara* leaf extract @ 75 % and T₆- *M. incognita* @ 1J2/cc soil + Soil drenching of *L. camara* leaf extract @ 100 %

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