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ENTOMOLOGY

Effect of essential oils on mortality, hatching and multiplication of root-knot nematode, *Meloidogyne incognita* and its Impact on plant growth parameters

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Abstract

Essential oil from aromatic plants showed strong nematicidal activity *in vitro* experiments. Among six tested essential oils, percent juvenile mortality was observed in lemon grass (*Cymbopogon citratus*), 89, 51 as well as in palmarosa (*Cymbopogon martini*), 80 and 44 at the doses of 500 and 250 ppm respectively at 12 hour of exposure time. However, maximum mortality percentage was observed in *C. citratus* oil and it registered cent percent mortality at 500 ppm at 24 hours of exposure time. The hatching in both treatments started by 8th day and steeply increased in 10th day. Essential oils at 5 concentrations drastically reduced the total number of J_2 as both essential oils showed more than 50 % reduction in hatching over control. The minimum cumulative hatching was observed in 1000 ppm in *C. citrates* oil. The effect of root-dip treatments of tomato seedlings with *C. citratus* and *C. martini* significantly reduced total number of root knot galls/ per plant, per cent galled area and soil population as compared to control. The minimum number of *M. incognita* galls was found in *C. citratus* at 500 ppm it was significantly different from carbosulfan as well as *C. martini* treatments. The maximum shoot length was found in *C. martini* followed by *C. citratus* and carbosulfan at 500 ppm. All the treatments significantly improved the root length than the inoculated plants but they were not significantly different among themselves.

Highlights

- Essential oils from aromatic plants confirmed nematicidal potential
- *C. citratus* and *C. martini* oils may be used for the management of *M. incognita*

Keywords: Aromatic plants, nematicide, *Meloidogyne incognita*, *Lycopersicon esculentum*, carbosulfan, essential oil

Vegetable crops are the most essential for our daily diet as well as a high value crops for the farmers. Furthermore, they are rich in carbohydrates, proteins, vitamins, minerals, *fibre* and antioxidant compounds. The daily minimum requirement of vegetables in diet is about 284 g/person/day (Choudhury, 1980). According to National Horticulture Board, production of vegetables is about 162.897 million tonnes from an area of 9.396 million hectare with average productivity of 17.30 tonnes per hectare (NHB, 2014). Tomato (*Solanum lycopersicum*) is solanaceous vegetable crops grown worldwide (Maria *et al.*, 2014) and an important source of Vitamin A and C (Sekhar *et al.*, 2010) in addition to, it produces "Lycopene" as natural antioxidant that works successfully to slow the growth of cancerous cells (Bhowmik *et al.*, 2012). As per the National Sample survey conducted during 2011-2012 in India, per capita consumption of tomato in rural and urban area

is 586 and 806 grams per month, respectively (Anonymous, 2014).

Plant parasitic nematodes are major limiting factor affecting plant growth and yield of tomato. Theroot-knot nematode, *Meloidogyne* species are the most devastating (Williams-Woodward and Davis, 2001) and have extensive host range (Mitkowski and Abawi, 2003). Globally, over 90 species of the genus, *Meloidogyne* have been described (Sikoraand Fernandez, 2005). However, *M. incognita, M. javanica* and *M. arenaria* are of greatest economic importance, being responsible for at least 90% of all damage caused by root-knot nematodes (Castagnone-Sereno, 2002). The root-knot nematode species (*Meloidogyne* spp.) is the most devastating pest which causes 28.0-47.5 per cent yield losses in tomato (Gill and Jain, 1995). Currently, synthetic pesticides are the effective means of control but they are expensive as well as hazardous to environment. In the long term, indiscriminate use of pesticides can have repercussions on human health as well as on environment (Dinham, 1993).A wide range of pesticides are used for crop protection against pest infection during the cultivation of vegetables (Kalra, 2003), and the literature reveals that vegetables contain the residues of pesticides above their respective maximum residue limit (Taneja, 2005; Srivatsava *et al*., 2011) may pose health hazards to consumers (Flliion *et al*., 2000; Mukherjee and Gopal, 2003).

Therefore, there is an urgent need to replace these hazardous pesticides with other alternatives, which are less toxic and eco-friendly (Abhishek *et al*., 2013 and Geeta *et al*., 2015). One way of searching for such nematicidal compounds is to scree naturally occurring compounds in plants (Rajendra and Sarvjeet, 2014). Many compounds with nematicidal activity have been found inplants, including alkaloids, diterpenes, fattyacids, glucosinolates, isothiocyanates, phenols, polyacetylenes, sesquiterpenes and thienyls (Chitwood, 2002). Plant extracts containing volatile ecompounds, especially essential oils have been found to possess antimicrobial, insecticidal and nematicidal activity (Okoko, *et al*., 1999). Essential oils are active against a number of pests, are biodegradable from the environment and often have low toxicity to mammals (Bainard *et al*., 2006). Cloveoil has demonstrated toxicity to plant parasitic nematodes (Chitwood, 2002; Meyer, *et al*., 2008; Pandey and Dwivedi, 2000; Park *et al*., 2005; Salgadoand Campos, 2003a, 2003b). Use of pesticides of plant origin is among the several ecologically based alternatives available in nematode management (Mangala and Mauria, 2006). Essential oils of some plants and/or their components have been tested for nematicidal activity *in vitro* and in soil (Oka *et al*., 2000). Thus the current research efforts have been taken upto evaluate the nematicidal efficacy of some commercially available essential oils against *Meloidogyne incognita* in tomato.

The objectives of this work were, to evaluate the effect of the essential oil on mortality and egg hatching of second stage juveniles of root-knot nematode *in vitro* studies; and to resolve the effects of essential oil as root dip treatment in the management of *M. incognita* infecting tomato.

Materials and Methods

The objectives of the present investigations were achieved by planning and conducting different experiments both in the laboratory and in the glass house at College of Horticulture and Forestry, CAU, Pasighat, Arunachal Pradesh at a longitude 28.1°N, latitude 95.4°E and altitude 155 m during year 2013.

Plant material and essential oils

The experiments were conducted with 5 essential oils *viz.* Citronella (*Cymbopogon nardus*), Eucalyptus (*Eucalyptus globulus*), Lemongrass (*Cymbopogon citratus*), Palmarosa (*Cymbopogon martini*) and Patchouli (*Pogostemoncablin*), which were commercially available. For making 1000ppm stock solutions, the required amounts of these essential oils were dissolved in small quantity of solvent, cyclohexanone (2% v/v) and 2 drops of surfactant, Tween 80 (0.5% v/v) were added and then volume was made up with distilled water. With these stock solutions, different concentrations of essential oils were prepared by diluting with double distilled water for various experiments.

Nematode inoculum

The nematode population used in the study was

Table 1: Effect of essential oils on second stage juveniles (J_2) of *M. incognita* at different concentration in *in vitro* trials

Sl. No.	Treatments	Doses (pm)	Exposure timing (% mortality of J_2)			
			6h	12h	24h	48h
1	*C. citratus*	500	0.00(4.05)*	89.25 (71.41)	100(85.95)	100(85.95)
		250	0.00(4.05)	50.75(45.71)	100(85.95)	100(85.95)
		100	0.00(4.05)	0.00(4.05)	28.25(32.37)	60.25(51.01)
		50	0.00(4.05)	0.00(4.05)	0.00(4.05)	33.75(35.89)
		25	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
2	*C. martini*	500	0.00(4.05)	80.00(63.80)	100(85.95)	100(85.95)
		250	0.00(4.05)	43.25(41.38)	91.00(75.14)	100(85.95)
		100	0.00(4.05)	0.00(4.05)	0.00(4.05)	44.00(41.83)
		50	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
		25	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
3	*C. nardus*	500	0.00(4.05)	0.00(4.05)	43.75(42.12)	80.25(64.08)
		250	0.00(4.05)	0.00(4.05)	0.00(4.05)	54.25(47.73)
		100	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
		50	0.00(4.05)	0.00(4.05)	0.00(4.05)	18.00(25.42)
		25	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
4	*E. globolus*	500	0.00(4.05)	0.00(4.05)	0.00(4.05)	29.00(31.29)
		250	0.00(4.05)	0.00(4.05)	0.00(4.05)	27.00(30.69)
		100	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
		50	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
		25	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
5	*P. cablin*	500	0.00(4.05)	0.00(4.05)	0.00(4.05)	28.25(32.35)
		250	0.00(4.05)	0.00(4.05)	0.00 (4.05)	18.50(25.76)
		100	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
		50	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
		25	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
6	Control (Water)	—	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
7	Control (Water + Solvent + Surfactant)	—	0.00(4.05)	0.00(4.05)	0.00(4.05)	0.00(4.05)
	EMS					
	Treatments	—	—	(0.170)	(0.478)	(0.423)
	Dose		—	(0.170)	(0.478)	(0.423)
	Interaction		—	(0.381)	(1.06)	(0.946)
	C.D. (0.05P)					
	Treatments	—	—	(0.48)	(1.34)	(1.19)
	Dose		—	(0.48)	(1.34)	(1.19)
	Interaction		—	(0.75)	(2.12)	(1.88)

*Figures in parenthesis are the angular transformed values.

obtained from roots of brinjal (cv. PusaUttam) grown in the experimental area of College of Horticulture and Forestry, CAU, Pasighat, Arunachal Pradesh. To establish and maintain cultures of *M. incognita*, a single egg mass from a single gall containing a single female was removed from a root, surface sterilized with 1% NaOCl for 4 minutes and rinsed through four series of sterilized water. To increase nematode populations, a single egg mass was inoculated into a pot containing 3 week old brinjal plants (cv. PusaUttam) in sterilized soil and kept for 3-4 months. Eggs inoculum was prepared according to procedures given by Hussey & Barker, 1973. Washed *M. incognita* infected brinjal plants roots

Table 2: Effect of essential oils on hatching of second stage juveniles (J_2) of *M. incognita* at different concentration in *in vitro* trials

Sl. No.	Treatments	Number of J_2 hatched out										
		Dose (ppm)	Day 2	Day 4	Day 6	Day 8	Day 10	Day 12	Day 14	Day 16	Total	% reduction over control
1	*C. citratus*	1000	0.25 (0.25)*	0.50 (0.50)	5.00 (2.22)	27.00 (5.17)	27.50 (5.18)	112.00 (10.57)	19.25 (4.37)	0.75 (0.60)	192.00 (13.85)	87
		500	0.00 (0.00)	0.75 (0.75)	7.00 (2.62)	27.00 (5.17)	60.50 (7.74)	168.50 (12.80)	34.50 (5.84)	1.00 (0.85)	295.00 (17.17)	81
		250	0.00 (0.00)	8.00 (2.78)	11.50 (13.37)	31.00 (5.50)	96.00 (9.78)	166.50 (12.89)	52.75 (7.25)	2.50 (1.06)	368.00 (19.18)	80
		100	1.50 (0.80)	23.00 (4.81)	34.00 (5.86)	26.25 (5.04)	140.00 (11.85)	206.00 (14.33)	44.00 (6.50)	2.75 (1.56)	472.00 (21.89)	69
		50	0.50 (0.35)	31.75 (5.59)	38.75 (6.19)	43.00 (6.50)	155.80 (12.47)	255.00 (15.95)	63.25 (7.87)	6.75 (2.45)	616.00 (24.37)	59
2	*C. martini*	1000	0.25 (0.25)	0.25 (0.25)	14.00 (3.70)	32.75 (5.69)	133.30 (11.54)	112.00 (10.57)	19.50 (4.39)	0.25 (0.25)	318.00 (17.66)	79
		500	0.00 (0.00)	1.75 (1.28)	17.00 (4.03)	31.50 (5.60)	156.30 (12.58)	163.00 (12.85)	48.00 (6.91)	0.75 (0.60)	421.00 (20.44)	72
		250	1.50 (1.00)	2.00 (1.39)	7.00 (2.62)	41.25 (6.40)	167.00 (12.90)	197.00 (14.05)	75.25 (8.65)	6.25 (2.49)	498.00 (22.31)	67
		100	7.25 (2.67)	28.25 (5.26)	23.00 (4.78)	26.50 (5.12)	146.50 (12.10)	275.50 (16.58)	84.25 (9.20)	7.25 (2.67)	537.00 (23.46)	64
		50	24.75 (4.93)	89.25 (9.43)	41.25 (6.37)	34.75 (5.27)	21.93 (14.78)	248.00 (15.76)	52.25 (7.20)	10.00 (3.14)	711.00 (26.45)	53
3	Control (Water)		370.00 (19.23)	385.00 (20.39)	287.00 (20.10)	376.00 (14.31)	121.00 (13.64)	11.25 (10.58)	0.00 (0.00)	0.00 (0.00)	1522.00 (39.01)	—
4	Control (solvent + surfactant + Water)		120.75 (10.98)	140.00 (20.39)	405.00 (20.10)	305.50 (14.31)	186.30 (13.64)	115.00 (10.58)	19.50 (4.30)	5.50 (2.02)	1498.00 (38.70)	—
	EMS											
	Treatments		(0.132)	(0.132)	(0.133)	(0.158)	(0.158)	(0.137)	(0.155)	(0.160)	(0.136)	—
	Dose		(0.208)	(0.209)	(0.210)	(0.250)	(0.250)	(0.217)	(0.245)	(0.253)	(0.215)	
	Interaction		(0.294)	(0.295)	(0.297)	(0.354)	(0.353)	(0.306)	(0.470)	(0.358)	(0.305)	
	C.D. (0.05P)											
	Treatments	(0.381)	(0.382)	(0.383)	(0.458)	(0.458)	(0.456)	(0.396)	(0.449)	(0.490)	(0.394)	—
	Dose	(0.301)	(0.603)	(0.606)	(0.723)	(0.723)	(0.722)	(0.626)	(0.709)	(0.730)	(0.623)	
	Interaction	(0.601)	(0.603)	(0.603)	(0.723)	(0.623)	(0.721)	(0.626)	(0.709)	(0.737)	(0.622)	

*Figures in parenthesis are square root transformed values.

were cut into small segments (1-2 cm long) and agitated for 3 minutes in 1% NaOCl. The suspension was passed through 75 and 5 µm sieves. The eggs and second stage juveniles (J_2) caught on the 5 µm sieve were washed several times with water, re-suspended and their concentration determined by dilution counts method.

In vitro experiments

Nematicidal activity of essential oils on J_2s

The effect of nematicidal activity of the essential oil of Citronella (*Cymbopogon nardus*), Eucalyptus (*Eucalyptus globulus*), Lemongrass (*Cymbopogon citratus*), Palmarosa (*Cymbopogon martini*) and

Patchouli (*Pogostemon cablin*) was evaluated against J_2s of *M. incognita.* Mature egg masses of the nematode were placed in sterilized double distilled water for 72 h in petridishes. The J_2s emerging were collected every 24 h, but those collected in the first 24 h were discarded. 1.0 ml of double strength (i.e. 1000 ppm if 500 ppm required) of each test concentration (25ppm to 500 ppm) of essential oil was put in a petridishes and 1.0 ml of nematode suspension containing 100 J_2 was added to it, so that required concentration was formed. Two controls were also kept such as distilled water alone and solvent i.e. cyclohexanone + surfactant (Tween 80 + water). All the treatments, concentrations and controls were replicated for four times. The petridishes were kept at 25°C for 6-48 hours of exposure time. After the scheduled time of exposure in different treatments, the nematodes were washed in fresh water by passing through 325 mesh sieve and kept in fresh water for 24 h to observe revival, if any. Then the number of active and inactive nematodes was counted by tacking the suspension in a counting dish under stereoscopic microscope. The inactive nematodes which did not move even with the touch of a pick were taken as dead and the mortality percentage was calculated.

Influence of essential oils on hatch

Hatching was monitored in 5cm glass petridishes containing sterile deionised distilled water (SDDW) covering the egg masses. Five mature uniformly sized egg masses, with mean viable 300 eggs were placed in each of four replicate petridishes in different concentration of each essential oil. For making stock solutions, the required amounts of these essential oils were dissolved in small quantity of solvent, cyclohexanone (2%v/v) and 2 drops of surfactant, Tween 80 (0.5% v/v) were added and then volume was made up with distilled water. With these stock solutions, different concentrations of essential oils were prepared by diluting with distilled water for various experiments. Egg masses of *M. incognita* were exposed to different concentrations (i.e. 1000, 500, 250, 100 and 50ppm) of the two essential oils *viz. C. citratus* and *C. martini* in glass petridishes for 48 hours of interval. Two controls, water and solvent +water were also kept and petridishes were kept at 25°C for 16 days or hatching almost stopped. The numbers of J_2s that emerged were recorded at 2 day intervals. After 16 days, egg masses from each petridish were separated to estimate the number of un-hatched eggs and the number of hatched J_2s was expressed as a cumulative percentage of viable J_2s.

In planta experiments

Effect of essential oils as root dip treatments

Effect of essential oils as root dip treatments of tomato seedlings were evaluated against *M. incognita* in pot trials. This experiment was carried out in 20 cm earthen pots containing 15,000g of steam sterilized soil and farm yard manure mixture (3:1) in green house. The twenty five days old tomato seedlings (Pusa Hybrid-1) were dipped in beakers for one hour in different concentrations of

Table 3: Effect of root dip treatments of essential oils against *Meloidogyne incognita* infecting tomato (*Lycopersicon esculentum L.*)

Sl. No.	Treatments	Dose (ppm)	No. of galls/plant	% galled area	No. of J_2/100gm soil
1	*C. citratus*	500	58.25(7.612)*	22.50(28.50)**	93.75(9.62)*
		250	75.25(8.60)	30.25(33.64)	137.50(11.71)
2	*C. martini*	500	72.25(9.19)	28.50(32.57)	125.00(11.03)
		250	100.00(9.98)	41.50(40.37)	131.25(11.41)
3		500	85.00(9.18)	30.00(33.49)	87.50(9.22)
4	Control (with nematode)	$2J_2$/gsoil	172.50(13.10)	80.00(63.97)	212.50(14.54)
	C.D. (0.05 P)		(1.096)	(2.47)	(1.836)
	EMS		(0.545)	(1.17)	(1.527)

*Figures in parenthesis are square root transformed values.

**Figures in parenthesis are the angular transformed values.

Table 4: Effect of root dips treatments of essential oils against *Meloidogyne incognita* infecting tomato (*Lycopersicon esculentum L.*)

Sl. No.	Treatments	Dose (ppm)	Shoot length (cm)	Root length (cm)	Shoot fresh weight (g)	Root fresh weight (g)
1	*C. citratus*	500	41.00*	23.25	127.25	9.12
		250	33.25	20.00	119.75	10.75
2	*C. martini*	500	43.25	25.00	105.50	11.50
		250	37.25	23.00	81.75	11.75
3	Carbosulfan	500	40.25	22.00	122.75	8.50
4	Control (without nematode)		51.00	31.00	133.75	16.75
5	Control (with nematode)	$2J_2$/g soil	30.50	20.25	73.25	8.97
	C.D. (0.05 P)		6.02	4.16	20.84	2.45
	EMS		16.76	8.02	201.02	2.87

(Average of 4 replications)

essential oils and insecticide such as *C. citrates* (500 and 250 ppm), *C. martini* (500 and 250 ppm) and carbosulfan 500 ppm. The time of exposure for root dip treatment and doses were selected based on preliminary trials. The two controls inoculated (with nematodes) and uninoculated (without nematodes) were kept for comparisons. The plantation was done @ one seedling/pot. The inoculation of J_2 of *M.incognita* was done by pouring the suspension in the root zone of the transplanted seedlings of tomato @ 30,000 J_2/pot (2 J_2/g soil).

Plants were watered daily with tap water to maintain sufficient moisture in pots. The experiment was terminated 45 days after transplanting of tomato seedling and nematode inoculation. After termination of transplanting, the shoot of each plant was cut off at soil level and the roots were uprooted thoroughly and washed with tap water. The following variables were assessed i.e. fresh shoot and fresh root weight, shoot length and root length recorded as plant parameters. Nematode population density was determined by extracting eggs and J_2s from soil samples. Severity of nematode galling of the root system was assessed such as number of galls/plant, per cent galled area as nematode parameter.

Statistical analysis

The data of all experiments was statistically analyzed. In general, two factorial randomized designs were used for the analysis. The hatching of juveniles and number of J_2s/100g of soil and number of galls/plant were analyzed by square root transformed values while the per cent galled area and percent mortality of J_2 were analyzed through angular transformed value.

Results and Discussion

The viability of J_2s of *M. incognita* was influenced by the time of exposure as well as concentrations of essential oils throughout the experiment (Table 1). Among six tested essential oils percent juvenile mortality was observed in *C. citratus* 89, 51 as well as in *C. martini* 80, 44 at the doses of 500 and 250 ppm respectively with 12 hour of exposure time. However, maximum mortality percentage was observed in *C. citratus* followed by *C. martini*, *C. nardus*, *E. globules* and *P. cablin*. *C. citratus* registered cent percent mortality at 500 ppm at 24 hours of exposure time. As concentrations of *C. citratus* increased from 50 to 500 ppm there was a corresponding increase in J_2 mortality at 48 hours of exposure time. Among the doses, 500 and 250 ppm showed nematicidal potential with effective toxicity (≥50%) in *C. citratus*, *C. martini* and *C. nardus* at 48 h of exposure. *In vitro* experiments clearly demonstrated that viability of J_2 of *M. incognita* was significantly reduced by the essential oils *viz. C. citratus* and *C. martini*. The effect of essentials oils on juveniles mortality supported by several other workers. Essential oils from several plant species have been shown to have nematicidal activity on root-knot nematodes *in vitro* and in soil (Soler-Serratosa *et al*., 1996). Bhatti (1988) reported that essential oils of *Cymbopogon flexuosus*, *Cymbopogon martini* and *Cymbopogon nardus* and their major constituents, geraniol, citral, citronellol

and citranellal exhibited nematicidal activity against *Anguinatririci, Tylenchulus semipenetrans, Meloidogyne javanica* and *Heterodera avenae*. Salagado *et al.,* (2003) evaluated the essential oils of *Eucalyptus camaldulensis, E. saligma, E. urophylla* and *C. nardus* against Juveniles of *Meloidogyne exigua* and found toxic to nematode after immersion in for 24 hours of exposure time. The *Cymbopogon* species produce essential oils rich in monoterpenes such as citral, citronellal, citronellol, linalool, elemol, 1,8-cineole, limonene, geraniol, β-carophyllene, methyl heptenone, geranyl acetate and geranyl formate (Ganjewala *et al.,* 2008). *Cymbopogon citratus* is widely used in nutraceutical industries due to its strong lemony odor for its high content of the aldehyde citral and small quantities of geraniol, geranyl acetate and monoterpene olefins (Debashis *et al.,* 2014). Studies on extracts from *C. citratus* leaves have demonstrated the presence of antioxidant, anti-microbial and anti-fungal activities (Oloyede, 2009; Pereira *et al.,* 2009). The results of the present studies confirm the nematicidal activity of *C. citratus, C. martini* and *C. nardus* against *M. incognita*.

The effect of different concentration of two essential oils on the hatching of J_2s at different days is shown in Table 2. The hatching was almost nil in all the treatment on 2nd day while large number of juveniles coming out from the eggs in both control. In the majority of treatments with essential oils, the greatest reduction in hatching occurred in the first 8 days. Rate of hatching was inversely proportional with concentration of essential oil as it was decreased with increase in concentration.

The 616 number of juveniles was observed in 50ppm and only 192 juveniles were observed in 1000ppm of *C. citratus* oil. Generally, hatching inhibition occurred at the highest concentration level. The cumulative numbers of J_2 after 16 days revealed that maximum hatching was observed in controls although among treated one, minimum hatching was observed in 1000 ppm in *C. martini* oil. Essential oils at 5 concentrations (1000, 500, 250, 100 and 50 ppm) drastically reduced the total number of J_2s as both essential oils showed more than 50% reduction in hatch over the control, even at 50 ppm very adverse effect was noted on hatching. The data for the nematicidal activity of essential oils agree with results of other researchers, who found that the egg hatch of *Meloidogyne* spp. was reduced when it was exposed to different concentration of essential oils. Oka *et al.,* (2000) reported that eight number of essential oils at a concentration of 1,000 µl/l reduced hatching to less than 5%, while hatching was 32.5% in the control after 7 days. The main components of essential oils were revealed to be thymol, carvacrol, and *t*-anethole. The results of present studies, however, indicated an inhibition of hatching J_{2S} of *M. incognita* with the *C. citratus* and *C. martini* oils after incubation of 48 h at 25°C.

The data presented in Table 3 and 4 showed the effect of rootdip of tomato for 1.0 h in two essential oils *viz. C. citratus* and *C. martini* at two concentrations in comparison with carbosulfan and inoculated and uninoculated controls on *M. incognita*. A perusal of data presented in Table 3 indicated that all the treatments significantly reduced total number of root knot galls/plant, percent gall area and soil population as compared to the control. The minimum number of root knot nematode galls was found in *C. citratus* at 500 ppm which was significantly different from carbosulfan.

The most effective treatment for reducing the per cent galled area were *C. citrates* at 500 ppm followed by *C. martini* at 500 ppm. The minimum number J_2s was counts in carbosulfan followed by *C. citratus* at 500 ppm and these treatments were also found at par with each other. The maximum shoot length was found in *C. martini* followed by *C. citratus* and carbosulfan at 500 ppm but it was not found significantly different from each other. *C. martini* at 500 ppm resulted in maximum shoot length though it was at par with *C. citratus* at 500 ppm. The maximum fresh shoot weight was recorded in *C. citratus* followed by carbosulfan.

All the plants treated with essential oils and carbosulfan significantly improved the root length than the inoculated plants but they were not significantly different among themselves. The maximum fresh shoot weight was observed in *C. martini* at 250 ppm which was found significantly different as compared to the carbosulfan. Results from the present *in planta* experiments indicated that essential oil compounds directly affect nematode biology by interfering with nematode hatching and J_2 viability. The use of various parts of indigenous plants as botanical extracts has been reported as important component in pest management

(Mangala and Mauria. 2006). The effect of essentials oils as root dip treatments on nematode parameters as well as plant growth parameters are supported by other workers. Walker and Melin (1996) have reported that number of root knot galls reduced due to application of essential oils of *Menthapiperita* and *Menthaspicata* in soil. Hasabo and Noweer (2005) found that water extract of basil was effective in reducing populations of root-knot nematodes in eggplant to a moderate degree at 5% concentration.

Additionally, as essential oils from *C. coronarium* have also been reported to have insecticidal and fungicidal activities (Pérez *et al*., 1999; Alvarez-Castellanos *et al*., 2001), treatment of soil with essential oils or their components could serve as a means of soil disinfection. In the present investigations, effect of rootdip treatment on root knot nematodes *M. incognita* was confirmed that the nematicidal potential of *C. citratus* and *C. martini* could be used as soil drenching for nematode management in tomato. Rootdip treatment was found to be effective though care had to be taken for the adverse effect on plant by adjusting the dose and the dip time.

Conclusion

Essential oils showed nematicidal potential in the management of *M. incognita* in present study. However, effective toxicity (>50 % mortality) was found in *C. citratus* followed by *C. martini* and *C. nardus*. Among the tested essential oils *C. citratus* was the most effective followed by *C. martini*. The use of essential oils may be one of the efficient alternatives and cheap methods of nematode control that are need of the hour and safe to farmers as well as environment.

If low concentrations can be effective in nematode management, as demonstrated by this study, then a given quantity of plant material can be better utilized over a larger area. Therefore, the use of indigenous essential oils should be considered in integrated nematode management strategies. It is suggested that further trials be conducted in the field on the basis of the promising results from these studies.

References

Abhishek, W., Preeti, M., Anjali, C. and Shirkot, C.K. 2013. Antagonistic Activity of Plant Growth Promoting Rhizobacteria Isolated from Tomato Rhizosphere Against Soil Borne Fungal Plant Pathogens. *International Journal of Agriculture, Environment and Biotechnology* **6**(4): 571-580.

Alvarez-Castellanos, P.P., Bishop, C.D. and Pascual-Villalobos, M.J. 2001. Antifungal activity of the essential oil of flower heads of garland chrysanthemum (*Chrysanthemum coronarium*) against agricultural pathogens. *Phytochemistry* **57**: 99-102.

Anonymous. 2014. Household consumption of various goods and services in India 2011-12. Ministry of Statistics and Programme Implementation, Government of India, pp. 1-1143.

Bainard, L.D., Isman, M.B. and Upadhyaya, M.K. 2006. Phytotoxicity of clove oil and its primary constituent eugenol and the role of leaf epicuticular wax in the susceptibility to these essential oils. *Weed Science* **54**: 833-837.

Bhatti, D.S. 1988. Utilization of Toxic Plants for the Control of Nematode Pests of Economic Crops. *Final Technical Report, PL 480 Project*. Haryana Agriculture University, Hisar, p. 231.

Bhowmik, D., Kumar, K.P.S., Paswan, S. and Srivastava, S. 2012. Tomato-A Natural Medicine and Its Health Benefits. *Journal of Pharmacognosy and Phytochemistry* **1**(1): 33.

Castagnone–Sereno, P. 2002. Genetic variability in parthenogenetic root–knot nematodes, *Meloidogyne* spp. and their ability to overcome plant resistance genes. *Nematology* **4:** 605-608.

Chitwood, D.J. 2002. Phytochemical based strategies for nematode control. *Annual Review of Phytopathology* **40**: 221-249.

Choudhury, B. 1980. Horticultural crops - Vegetables. In: Handbook of Agriculture (Eds. S.N. Tata and A.M. Wadhwani), ICAR, New Delhi, pp. 1049-1127.

Debashis, D., Pawan, K., Nath, A., Verma, N. and Gangwar, B. 2014. Qualities of lemongrass (*Cymbopogan citratus*) essential oil at different drying conditions. *International Journal of Agriculture, Environment & Biotechnology* **7**(4): 903-909.

Dinham, B. 1993. The Pesticides Hazard*: A Global health and Environmental Audit*. Zed Books, London UK, p. 228.

Fillion, J., Sauve, F. and Selwyn, J. 2000. Multiresidue method for the determination of residues of 251 pesticides in fruits and vegetables by gas chromatography/mass spectrometry and liquid chromatography with fluorescence detection. *Journal of AOAC International* **83**(3): 698-713.

Ganjewala, D. 2008. RAPD Characterization of three selected cultivars OD-19, GRL-1 and krishna of east Indian lemongrass (*Cymbopogon flexuosus Nees ex Steud*) Wats. *American-Eurasian Journal of Botany* **1**: 53-57.

Geeta, S., Sujoy, S., Ruchi, G., Bineet, K.S., Awadhesh, B.R. and Rana, P.S. 2015. Evaluation of suitable antagonists in the management of early blight of tomato cultivar CO-3. *International Journal of Agriculture, Environment and Biotechnology*: **8**(1): 127-133.

Gill, J.S. and Jain, R.K. 1995. Nematode problems of vegetable crops in India. In: *"Nematode Pest Management - An Appraisal of Eco-friendly Approaches"* (Eds. Gopal Swarup, Dasgupta, D.R. and Gill, J.S.) Nematological Society of India, New Delhi, pp. 166-178.

Hasabo, S.A. and Noweer, E.M.A. 2005. Management of Root-Knot Nematode, *Meloidogyne incognita* on eggplant with some Plant Extracts. *Egyptian Journal of Phytopathology* **33**(2): 65-72

Hussey, R.S. and Barker, K.R. 1973. A comparison method of collecting inocula for *Meloidogyne* species, including a new technique. *Plant Diseases Rep.* **57**: 1052-1058.

Kalra, R.L. 2003. Assessment of human exposure of pesticide residues through food, and water and other sources in India. In: Proceedings of symposium on risk assessment of pesticide residues in water and food, ILSI Washington DC, ITRC Lucknow and ICMR, New Delhi, India, pp. E1-9.

Mangala, R. and Mauria, S. 2006. Handbook of Agriculture Fact and figure for teacher, students and all interested farmers. Indian Council of Agricultural Research, New Delhi, p. 1346.

Maria, R., Guzman, M.G.D., Amanda, M.W., Frusciante, L. and Barone, A. 2013. Production of Pharmaceutical Proteins in Solanaceae Food Crops. *International Journal of Molecular Science* **14**: 2753-2773.

Meyer, S.L.F., D.K. Lakshman, I.A., Zasada, B.T.V. and Chitwood, D.J. 2008. Dose-response effects of clove oil from *Syzygium aromaticum* on the root knot nematode *Meloidogyne incognita*. *Pest Management Science* **64:** 223-229.

Mitkowski, N.A. and Abawi, G.S. 2003. Root-knot nematodes. Doi: 10.1094/PHI-1-2003-0917-01.

Mukherjee, I. and Gopal, M. 2003. Pesticide Residues in Vegetables. In: Proceedings of symposium on risk assessment of pesticide residues in water and food, ILSI Washington DC, ITRC Lucknow and ICMR, New Delhi, India, pp. A1-8.

Oka, Y., Nacar, S., Putieusky, E., Ravid, U., Zohara, Y. and Spiegal, Y. 2000. Nematicidal activity of essential oils and their components against the root knot nematode. *Phytopathology* **90**: 710-715.

Okoko, F.J., Nwafor, O.E. and Ejechi, B.O. 1999. Growth inhibition of tomato-rot fungi by phenolic acids and essential oil extracts of pepper fruit (*Dennetiatripelata*). *Food Research International* **32**: 395-399.

Oloyede, O.I. 2009. Chemical profile and anti-microbial activity of *Cymbopogon citratus* leaves. *Journal of Natural Products* **2**: 98-103.

Pandey, R.C. and Dwivedi, B.K. 2000. Comparative study of different plant extracts for their nematicidal potential. *Current Nematology* **11**: 39-43.

Park, I.K., Park, J.Y., Kim, K.H., Choi, K.S., Choi, I.H., Kim, C.S. and Shin, S.C. 2005. Nematicidal activity of plant essential oils and components from garlic (*Allium sativum*) and cinnamon (*Cinnamomum verum*) oils against the pine wood nematode (*Bursaphelenchus xylophilus*). *Nematology* **7**: 767-774.

Pereira, P.P., Puntel, R.L., Boschetti, T.K. and Morel, A.F. 2009. Antioxidant effects of different extracts from *Melissa officinalis, Matricaria recutita* and *Cymbopogon citratus*. *Neurochemical Research* **34**(5): 973-983.

Pérez, M.P. and Pascual-Villalobos, M.J. 1999. Efectos del aceiteesencial de inflorescencias de *Chrysanthemum coronarium* L. enmoscablanca y plagas de almacén. Investigación Agraria: *Producción y Protección Vegetal* **14**: 249-58.

Rajendra, K.M. and Sarvjeet, K. 2014. Screening of *Bacillus thuringiensis* Isolates Recovered from Diverse Habitats in India for the Presence of Insect and Nematode-active *cry* Genes. *International Journal of Agriculture, Environment and Biotechnology* **7**(1): 55-62.

Salgado, S.M.L. and Campos, V.P. 2003a. Hatching and mortality of *Meloidogyne exigua* in extracts and in natural products. *Fitopatologia Brasileira* **28:** 166-170.

Salgado, S.M.L. and Campos, V.P. 2003b. Effects of natural extracts on pathogenicity and reproduction of *Meloidogyne exigua* in coffee and *Meloidogyne incognita* race 3 in the common bean. *Nematologia Brasileira* **27**: 41-48.

Salgado, S.M.L., Campos, V., GracasCa, M.D. and Salgado, A.P.S. 2003. Hatching and mortality of second stage juveniles of *Meloidogyne exigua* in essential plant oils. *Nematologia Brasileira* **27**: 17-22.

Sekhar, L., Prakash, B.G., Salimath, P.M., Channayya, P., Hiremath, Sridevi, O. and Patil, A.A. 2010. Implication of heterosis and combining ability among productive Single cross Hybrids in tomato. *Electronic Journal of Plant Breeding* **1**(4): 706-711.

Sikora, R.A. and Fernandez, E. 2005. Nematode parasites of vegetables. In: Plant Parasitic Nematodes in Subtropical and Tropical Agriculture (Second edition). (Eds. Luc M, Sikora RA, Bridge J). CAB International Wallingford, UK, pp. 319-392.

Soler-Serratosa, A., Kokalis-Burelle, N., Rodríguez-Kábana, R., Weaver, C.F. and King, P.S. 1996. Allelochemicals for control of plant-parasitic nematodes. 1. In vivo nematicidal efficacy of thymol and thymol/benzaldehyde combinations. *Nematropica* **26**: 57-71.

Srivastava, A.K., Trivedi, P., Srivastava, M.K., Lohani, M., and Srivastava, L.P. 2011. Monitoring of pesticide residues in market basket samples of vegetable from Lucknow City, India: QuEChERS method. *Environmental Monitoring and Assessment* **176**: 465–472.

Taneja, A. 2005. Monitoring of organochlorine pesticide residues in vegetables from Agra, India – a case study. *Environmental Monitoring and Assessment* **110**: 341-346.

Walker, J.T. and Melin, J.B. 1996. *Mentha × piperita, Mentha spicata* and effects of their essential oils on *Meloidogyne* in soil. *Journal of Nematology* **28**: 629-635.

Williams-Woodward, J.L. and Davis, J.F. 2001. *Meloidogyne incognita* and *M. arenaria* reproduction on Dwarf hollies and Lantana. *Supplement Journal of Nematology* **33**: 332-337.