



# Impact of Integrative Management Strategies on the Reproduction of Root Knot Nematode, *Meloidogyne incognita*

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## ABSTRACT

Present investigation was planned to check the impact of bio (Cure, Azadirachtin) and synthetic (Cartap, Virtako) chemicals individually and concomitantly with fertilizers on the development of root knot nematode, *Meloidogyne incognita* and the growth of tomato (*Solanum lycopersicum*) plants. Tomato seedlings were transplanted in sterilized soil amended with Cartap, Virtako, Cure and Azadirachtin at their recommended doses and inoculated with 500 freshly hatched juveniles (J<sub>2s</sub>) of *M. incognita*. Data were recorded after 7, 14, 21 and 28 days to check the invasion and development stages. All the treatments significantly reduced invasion and subsequent development of *M. incognita* in roots. Different application methods regarding protective and curative were used to test the efficacy of Cartap, Virtako, Cure and Azadirachtin. Results revealed that Protective application of bio and synthetic chemicals was more effective at nematode suppression rather than being curative. Then the impact of bio and synthetic chemicals were tested in combination with NPK on nematode reproduction. Following treatments i.e. Virtako + NPK, Cartap + NPK, Cure + NPK, Azadirachtin + NPK, control (N) and control (H) with ten replications were applied in pots containing tomato seedlings. Inoculation of 2000 freshly hatched J<sub>2s</sub> of *M. incognita* was done. Results revealed that combined effect was more pronounced in reducing nematode reproduction and improving plant growth. Maximum number of J<sub>2s</sub> was recorded in nematode control (1241.0) while a dynamic decline was observed in Cartap+NPK, Virtako+NPK, Cure+NPK and Azadirachtin+NPK (144.4, 162.2, 198.1, 216.6) respectively. Combined application of selected chemicals with fertilizers not only reduced nematode reproduction but also improved plant growth. So, the integrated use of chemicals with fertilizers could be a successful tool in management strategies of root knot nematode.

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## Authors' Contribution

HA, NJ and MK planned, methodology and executed experimental work. SAK, AJ and HA writing-draft preparation. AH, AI and EH review, editing and data analyses.

## Key words

Root knot nematode, *Meloidogyne incognita*

## INTRODUCTION

Root-knot nematode was reported first time on cucumber (Berkeley, 1855). The type species of the genus *Meloidogyne exigua* was described by Goeldi in 1887 from where Chitwood found the name *Meloidogyne* which is currently used for root-knot nematodes. In Four new species were described within the genus *Meloidogyne* by Chitwood in 1949. Most of the species of *Meloidogyne* belong to tropical to sub-tropical climates however *M. hapla* and *M. chitwoodi* are well adjusted to temperate climates (Brodie et al. 1993). Management of root-knot nematodes is difficult due to their wide host range including more than 3000 plant species (Abad et al., 2003; Forghani and Hajihassani, 2020). Root-knot nematodes

cause severe losses in vegetables throughout the world. Estimated crop losses were 14% worldwide, equal to approximately \$ 100 billion dollars (Agris, 2005). The young ones of nematodes are defined as juveniles (Abd-Elgawad, 2020). Second Juvenile (J<sub>2</sub>) is the infective stage of RKN. J<sub>2</sub> hatch from eggs and invade into roots through root tips (Chen and Peng, 2019). These juveniles obtain their food by developing special cells called giant cells (Chen and Peng, 2019). A range of strategies are employed for the management of root-knot nematodes (Gamalero and Glick, 2020) including cultural practices, biological control, sanitation and host plant resistance (Tocco et al., 2020). But unfortunately, all these practices are unable to protect the crops under field conditions because these are not cost-effective and require extra labour (Kerry, 1990). However, chemical control is still considered as the main approach for the management of nematodes (Huang et al., 2020). The chemicals preferably used should possess the properties like high rate of nematode suppression in a

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short time, no phytotoxic effects and long persistence in the soil that would be advantageous for subsequent crops. During recent years, the importance in the development of pesticides with natural origins has increased because of the adverse effects of synthetic chemical pesticides (Desaeger *et al.*, 2020), like environmental risk, toxicity to non target organisms including humans/animals and resistance development. Carbamates and organophosphates possess the ability to inactivate acetylcholinesterase (a crucial enzyme in nervous system of nematodes and insects), however, the inhibitory effect of carbamates on acetylcholinesterase is short living. Nematode locomotion depends on an array of approximately 90 motor neurons and interneurons that employ neurotransmitters acetylcholine and gamma aminobutyric acid (GABA) (Johnson and Stretton, 1980). Nematode species exhibit diversity in sensitivity of their acetylcholinesterase to organophosphates and carbamates (Lee and Hodsdon, 1963; Pree *et al.*, 1987). Different ways of application were used to assess the efficacy of nematicides against *M. incognita* (Bhosle *et al.*, 2012; Abbas *et al.*, 2015). Present investigation was conducted to check the effect of bio and synthetic chemicals on the developmental stages of *M. incognita*, efficiency of protective and curative application method and their compatibility with fertilizers to reduce the damage of nematode and to increase tomato (*Solanum lycopersicum*) growth.

## MATERIALS AND METHODS

### *Effect of bio and synthetic chemicals on the invasion and development of M. incognita*

Sterilized soil amended with Cartap, Virtako, Cure and Azadirachtin at their recommended doses was filled in 150 ml plastic pots except in control pots. Three weeks old seedlings of tomato cv. Moneymaker were transplanted in earthen pots. After two weeks when the plants established their root system, 500 freshly hatched  $J_{2s}$  of *M. incognita* (obtained from single egg mass culture through mass culturing of nematode in earthen pots) were inoculated in each pot. Experiment was accomplished in four sets of treatments with 10 replications under completely randomized design. Data were recorded after 7, 14, 21 and 28 days to check the invasion and development stages ( $J_2$ , developing (d) $J_2$ ,  $J_3$ /swollen,  $J_4$ /sausage, adult female and egg masses) of root-knot nematode. After seven days of inoculation, plants with their balls were soaked for 3-4 h in container. Then their roots were gently removed and washed free of soil under running water. The roots were carefully handled without damaging the root tips. Egg masses were counted by staining the roots with Phloxine B (Holbrook *et al.*, 1983) while the females attached to roots

were recorded after staining with acid fuchsin under the stereomicroscope (Olympus SZ 61) at 2.5X magnification.

### *Protective and curative effect of bio and synthetic chemicals on the development of M. incognita*

Selected bio and synthetic chemicals were used to check their protective effect against *M. incognita*. Sterilized soil amended with Cartap, Virtako, Cure and Azadirachtin at their S concentration was filled in earthen pots of (10 cm diam.). Three weeks old seedlings of tomato cv. Moneymaker were transplanted in earthen pots containing amended soil after 3 days. Sterilized soil was filled in earthen pots of (15cm diam.). Fifteen hundred freshly hatched  $J_{2s}$  of *M. incognita* were inoculated in each pot. Those seedling which were exposed to amended soil for 5 days were uprooted carefully and transplanted into these infested pots. Harvesting was done after 35 days of inoculation and data were recorded on nematode reproductive parameters. To check curative effect tomato seedlings were grown and maintained in small pots (10 cm diam.) containing sterilized soil, after one month these seedlings were inoculated with 1500 freshly hatched  $J_{2s}$  of *M. incognita*. Sterilized soil amended with Cartap, Virtako, Cure and Azadirachtin was filled in earthen pots of (15cm diam.). These pots were kept moistened. After 4 days the inoculated plants were uprooted very carefully and washed in plenty of water just by shaking to avoid damaging the roots. Then these plants were transplanted into amended soil in such a way that half of the amended soil was put in the pot then other half was put after planting the seedlings, this was done to avoid damaging to roots. Harvesting was done after 35 days of transplantation into amended pots and data were recorded on nematode reproductive parameters.

### *Impact of different fertilizers alone on nematode reproduction*

This experiment was carried out in two steps. Effect of different fertilizers was evaluated against *M. incognita* reproduction and plant growth. Impact of different fertilizers; Nitrogen, Phosphorus and Potassium was checked individually and concomitantly. Three weeks old seedlings of tomato cv. Moneymaker were transplanted in earthen pots (15 cm diam.) containing sterilized soil. Different treatments of fertilizer (N, P, K, N+P, N+K, P+K, NPK) and two control treatments healthy (H) and nematode (N) were applied in pots. After five days plants were inoculated with 2000 freshly hatched  $J_{2s}$  of *M. incognita* in each pot. After 35 days plants were carefully uprooted, washed and data were recorded on plant growth and nematode reproduction parameters.

#### *Concomitant effect of bio and synthetic chemicals with NPK on the development of M. incognita*

Bio and synthetic chemicals were tested against *M. incognita* in combination with NPK (which proved to be more effective from the above study in reducing nematode population and improving plant growth). To test the combined effect of NPK with chemicals, three weeks old seedlings of tomato cv. Moneymaker were transplanted in earthen pots (15 cm diam.) containing sterilized soil amended with chemicals and NPK. Pots were arranged in completely randomized design with following treatments i.e., Cartap + NPK, Virtako + NPK, Cure + NPK, Azadirachtin + NPK, Control (N) and Control (H) with ten replications for each treatment. After five days plants were inoculated with 2000 freshly hatched  $J_{25}$  of *M. incognita* in each pot. Harvesting was done after 35 days and data were recorded on plant growth and nematode reproduction parameters. Experiments were repeated to confirm the results.

The recommended doses of tested chemicals were as follows: Cartap (Thiocarbamate) 9 g /100 mL, Virtako (Thiamethoxam + chlorantraniliprole) 4 g /100 mL, Azadirachtin (Azadirachtin) 0.5 mL /100 mL, Cure (Abamectin) 0.4 mL /100 mL

## RESULTS

#### *Effect of bio and synthetic chemicals on the invasion and development of M. incognita*

Effectiveness of bio and synthetic chemicals was evaluated against invasion and developmental stages of *M. incognita* at different harvesting intervals; 7, 14, 21 and 28 days. Results revealed that significantly varied ( $P=0.05$ ) number of developmental stage of *M. incognita* were observed at different harvesting times (Table I). Cartap among all the chemicals caused minimum invasion of  $J_{25}$  after 7 days, maximum was observed in control treatment. After 7 days two stages were observed including  $J_{25}$  and  $dJ_{25}$ . Number of  $J_{25}$  and  $dJ_{25}$  was varied significantly in all the chemicals (Table I). At harvest after 14 days more developmental stages of *M. incognita* were observed as compared to 7 days. Number of  $dJ_{25}$  was increased in all the treatments as compared first harvest. A short lived  $J_3$  stage was noted after 14 days harvest interval. Maximum number of  $J_3$  (swollen) was recorded in control (48.4) while minimum was observed in Cartap (7.5). Due to the increased harvest interval,  $J_{25}$  were not observed though lesser number of  $dJ_{25}$  was recorded in comparison to other harvest intervals. Minimum development of  $J_3$  stage was observed in Cartap and Virtako (5.5, 11.4) as compared to Cure and Azadirachtin (22.6, 30.5) respectively. Initial stages of development were not recorded after 28 days,

only negligible number of  $J_{25}$  was observed. Among synthetic chemicals Cartap caused maximum reduction in  $J_4$  stage of *M. incognita* while among bio chemicals Cure caused maximum reduction (Table I).

#### *Protective and curative effect of bio and synthetic chemicals on the development of M. incognita*

Protective efficiency of bio and synthetic chemicals was observed on the basis of *M. incognita* reproductive parameters (Table II). Development of galls was significantly reduced in the protective treatment of all the chemicals. Maximum number of galls was observed in untreated control (377.6) while minimum was recorded in Cartap (87.4). Other treatments including Virtako, Cure and Azadirachtin caused significant reduction in number of galls (98.5, 117.7, 129.3) respectively. Due to the infectivity of *M. incognita* maximum number of numbers of egg masses were observed in control treatment. Significant reduction in the number of egg masses was observed in all the chemicals. Minimum number of egg masses were observed in Cartap (82.4) followed by Virtako and Cure (95.5, 107.7), respectively. In curative application of all the chemicals, number of galls was increased as compared to protective treatment. All the chemicals tested caused a significant ( $P=0.05$ ) reduction in the number of females but less as compared to protective application. Statistically significant reduction in the number of females was observed in Cartap (126.5). In all the chemicals number of eggs per root system of tomato decreased significantly. Maximum eggs were recorded in control (57210) while minimum were observed in Cartap and Virtako (17510, 20380) respectively. An increase in reproduction rate was observed in all the chemicals in comparison to protective application (Table III).

#### *Impact of different fertilizers on nematode reproduction*

Effect of different fertilizers was evaluated against *M. incognita* reproduction and plant growth individually and concomitantly. All the treatments varied significantly ( $P=0.05$ ) in their response toward plant growth and nematode reproduction. Impact of different fertilizers; N, P, K, N+P, N+K, P+K, NPK varied significantly on the reproduction of *M. incognita* (Table IV). Minimum number of galls was observed in concomitant application of NPK (160.6) while maximum galls were observed in control treatment having only nematodes (386.5). Fresh and dry weight of root was higher in healthy control followed by the plants that received fertilizer treatments (Table V). A progressive increase in shoot length was observed in fertilizer treatments. Maximum increase in shoot length was observed in NPK in comparison to nematode control.

**Table I. Effect of bio- and synthetic-chemicals on the invasion and development of *M. incognita***

Days	Treatment	$J_{2s}$	$dJ_{2s}/\text{sausage}$	$J_3$	$J_4$	Adult female	Egg mass
7 Days	Cartap	65.6 <sup>1</sup> e	13.1 e	-	-	-	-
	Virtako	87.1 d	20.4 d	-	-	-	-
	Abamectin	102.3 c	33.3 c	-	-	-	-
	Azadirachtin	122.2 b	39.2 b	-	-	-	-
	Control	176.5 a	69.5 a	-	-	-	-
	LSD	0.65	0.59				
14 Days	Cartap	12.3 e	25.4 e	7.5 e	28.5 e	-	-
	Virtako	19.1d	34.1 d	15.1 d	35.3 d	-	-
	Abamectin	30.4 c	52.5 c	29.2 c	49.1 c	-	-
	Azadirachtin	38.5 b	60.4 b	38.6 b	57.6 b	-	-
	Control	57.7 a	75.6 a	48.4 a	85.4 a	-	-
	LSD	0.56	0.51	0.56	0.47		
21 Days	Cartap	-	8.40 e	5.5 e	41.1 e	12.3 e	-
	Virtako	-	11.6 d	11.4 d	62.6 d	15.2 d	-
	Abamectin	-	17.5 c	22.6 c	90.6 c	21.5 c	-
	Azadirachtin	-	21.4 b	30.5 b	104.4 b	26.4 b	-
	Control	-	27.6 a	45.5 a	138.3 a	48.5 a	-
	LSD		0.46	0.47	0.54	0.44	
28 Days	Cartap	-	-	-	10.4 e	71.5 e	58.6 e
	Virtako	-	-	-	13.6 d	93.5 d	72.4 d
	Abamectin	-	-	-	18.5 c	104.3 c	85.5 c
	Azadirachtin	-	-	-	21.7 b	121.6 b	98.7 b
	Control	-	-	-	26.5 a	220.8 a	214.3 a
	LSD				0.46	0.44	0.45

<sup>1</sup>Means within a column sharing the same letter are not significantly different from each other at  $P = 0.05$  according to Bartlett's test

**Table II. Protective effect of bio- and synthetic-chemicals on the development of *M. incognita***

Treatments	No. of galls	No. of females	No. of egg masses	$J_{2s}/100\text{cm}^3$ of soil	eggs/ root system	RF <sup>2</sup>
Cartap	87.4 <sup>1</sup> e	102.4 e	82.4 e	195.5 e	15310 e	0.59 e
Virtako	98.5 d	118.6 d	95.5 d	223.7 d	18090 d	0.67 d
Abamectin	117.7 c	130.7 c	107.7 c	247.4 c	23340 c	0.74 c
Azadirachtin	129.3 b	144.5 b	122.5 b	262.5 b	25320 b	0.79 b
Control	377.6 a	393.6 a	370.6 a	1230 a	54100 a	3.69 a
LSD	0.45	0.46	0.46	0.46	154.6	0.005

<sup>1</sup>Means within a column sharing the same letter are not significantly different from each other at  $P = 0.05$  according to Bartlett's test; <sup>2</sup>Reproduction factor (RF) = final population/initial population.

#### *Combined effect of bio and synthetic chemicals with NPK on the development of M. incognita*

All the chemical treatments in combination with NPK were varied significantly ( $P=0.05$ ) on reproduction of *M. incognita*. Number of galls was minimum in the combined treatment of Cartap+NPK (72.1) followed by

Virtako+NPK (86.3). Cure+NPK treatment was proved to be more effective in reducing number of females (119.2) from Azadirachtin+NPK (127.4). Maximum number of egg masses was observed in nematode control plants (365.4). Cure+NPK caused more reduction in number of egg masses as compared to Azadirachtin+NPK (Table VI).

Recovery of  $J_{2s}$  from soil was significantly suppressed in all the combined treatments. Maximum number of  $J_{2s}$  was recorded in nematode control (1241.0) while a dynamic decline was observed in Cartap+NPK, Virtako+NPK, Cure+NPK and Azadirachtin+NPK (144.4, 162.2, 198.1, 216.6) respectively. Plant growth responses were increased

significantly in combined treatment of chemicals with NPK (Table VII). Tomato growth was most affected in control treatment having only nematodes. A variable effect of combined treatments was significant on all the growth parameters recorded.

**Table III. Curative effect of bio- and synthetic-chemicals on the development of *M. incognita***

Treatments	No. of galls	No. of females	No. of egg masses	$J_{2s}/100\text{ cm}^3$ of soil	Eggs/ root system	RF <sup>2</sup>
Cartap	105.5 <sup>1</sup> e	126.5 e	97.5 e	230.3 e	17510 e	0.69 e
Virtako	116.4 d	140.4 d	112.7 d	248.5 d	20380 d	0.74 d
Abamectin	136.5 c	161.7 c	133.4 c	272.7 c	27330 c	0.82 c
Azadirachtin	145.2 b	172.8 b	146.5 b	287.4 b	29520 b	0.86 b
Control	380.6 a	395.5 a	368.4 a	1237 a	57210 a	3.71 a
LSD	0.49	0.44	0.46	0.45	204.5	0.006

<sup>1</sup>Means within a column sharing the same letter are not significantly different from each other at  $P = 0.05$  according to Bartlett's test; <sup>2</sup>Reproduction factor (RF) = final population/initial population

**Table IV. Impact of different fertilizers on nematode reproduction.**

Treatments	No. of galls	No. of females	No. of egg masses	$J_{2s}/100\text{ cm}^3$ of soil	RF <sup>2</sup>
N	190.5 e	202.1 e	186.2 e	328.5 e	0.98 e
P	222.3 c	232.6 b	217.5 b	376.4 b	1.13 b
K	225.3 b	230.0 c	215.0 c	372.0 c	1.11 c
N+P	176.1 f	188.4 f	172.6 f	308.3 g	0.92 g
N+K	174.5 g	185.0 g	169.0 g	312.6 f	0.94 f
P+K	204.2 d	216.4 d	198.4 d	344.5 d	1.03 d
N+P+K	160.6 h	172.4 h	152.7 h	282.6 h	0.85 h
Control (N)	386.5 a	392.9 a	374.7 a	1253.0 a	3.75 a
Control (H)	0.00 i	0.00 i	0.00 i	0.00 i	0.00 i
LSD	0.564	0.517	0.533	0.471	0.003

<sup>1</sup>Means with in a column sharing the same letter are not significantly different from each other at  $P = 0.05$  according to Bartlett's test; <sup>2</sup>Reproduction factor (RF) = final population/initial population

**Table V. Impact of different fertilizers on tomato growth.**

Treatments	Leaf count/ plant	Fresh root weight (g)	Fresh shoot weight (g)	Dry root weight (g)	Dry shoot weight (g)	Root length (cm)	Shoot length (cm)
N	31.0 e	5.48 e	38.86 e	1.88 e	8.83 e	9.78 e	25.53f
P	25.0 f	3.75 g	35.80 g	0.96 g	6.72 g	7.60 g	22.80g
K	24.0 g	3.69 h	35.40 h	0.93 h	6.68 h	7.42 h	22.21h
N+P	39.0 c	6.37 c	40.62 c	2.78 b	9.57 c	10.81c	26.67c
N+K	39.0 c	6.34 d	40.58 d	2.75 c	9.52 d	10.52d	26.30d
P+K	36.0 d	4.86 f	37.92 f	1.34 f	7.94 f	8.34 f	25.76e
N+P+K	41.0 b	7.48 b	42.44 b	2.47 d	10.34 b	11.56b	27.62b
Control (N)	20.0 h	2.92 i	26.71 i	0.52 i	5.18 i	6.80 i	19.56i
Control (H)	63.1 a	12.18 a	60.28 a	4.58 a	17.43 a	17.42a	37.35a
LSD	0.662	0.008	0.0397	0.007	0.007	0.0073	0.06292

<sup>1</sup>Means within a column sharing the same letter are not significantly different from each other at  $P = 0.05$  according to Bartlett's test

**Table VI. Concomitant effect of bio- and synthetic-chemicals with NPK on the development of *M. incognita***

Treatments	No. of galls	No. of females	No. of egg masses	J <sub>2s</sub> /100cm <sup>3</sup> of soil	RF <sup>2</sup>
Cartap + NPK	72.1 e	78.1 e	66.2 e	144.4 e	0.43 e
Virtako + NPK	86.3 d	95.0 d	80.2 d	162.2 d	0.48 d
Abamectin + NPK	108.0c	119.2c	97.1 c	198.1 c	0.59 c
Azadirachtin + NPK	120.3b	127.4 b	107.5 b	216.6 b	0.65 b
Control (N)	390.7a	390.7 a	365.4 a	1241.0 a	3.72 a
Control (H)	0.00 f	0.00 f	0.00 f	0.00 f	0.00 f
LSD	0.612	0.610	0.532	0.569	0.003

<sup>1</sup>Means with in a column sharing the same letter are not significantly different from each other at  $P = 0.05$  according to Bartlett's test. <sup>2</sup>Reproduction factor (RF) = final population/initial population at  $P = 0.05$  according to Bartlett's test.

**Table VII. Concomitant effect of bio- and synthetic-chemicals with NPK on tomato growth.**

Treatments	Leaf count/ plant	Fresh root weight (g)	Fresh shoot weight (g)	Dry Root weight (g)	Dry shoot weight (g)	Root length (cm)	Shoot length (cm)
Cartap + NPK	60.0b	11.64b	57.64 b	4.44 a	16.24 b	16.61b	35.62b
Virtako + NPK	55.0c	10.37c	55.84 c	4.12 c	15.97 c	15.43c	33.22c
Abamectin + NPK	46.0d	8.86 d	50.76 d	3.52 d	13.86 d	14.74d	29.40d
Azadiractin + NPK	43.0e	8.22 e	49.23 e	3.46 e	13.38 e	13.43e	28.50e
Control (N)	19.0f	2.95 f	27.13 f	0.57 f	5.22 f	6.73f	20.24f
Control (H)	64.0a	12.24a	60.72 a	4.30 b	17.53 a	17.61a	37.52a
LSD	0.668	0.028	0.040	0.028	0.006	0.0634	0.0634

<sup>1</sup>Means with in a column sharing the same letter are not significantly different from each other at  $P = 0.05$  according to Bartlett's test.

## DISCUSSION

Various life stages of nematode viz., J<sub>2</sub>, d J<sub>2</sub>, J<sub>3</sub>, J<sub>4</sub>, adult female and egg masses were observed by harvesting at different time intervals. Suppression of nematode invasion was varied in Cartap, Virtako, Cure and Azadirachtin and subsequently development of nematode was also reduced. Invasion of nematode was decreased due to the nematicidal activity of chemicals (Tobin *et al.*, 2008). Absorption of chemicals by root system acts as barrier in penetration of nematodes into the roots. Feeding and development of penetrated nematodes was also affected by chemicals (Hague and Gowen, 1987). Invasion of J<sub>2</sub> was lowest in Cartap, as nematode locomotion depends upon motor neurons and interneurons that use a neurotransmitter acetylcholine whose activity is stopped by acetylcholinesterase (Johnson and Stretton, 1980) which was inactivated by Cartap. Some plant metabolites activated in results of nematode invasion i.e., phenolics and phytoalexins that caused suppression in further invasion (McCarter, 2008).

Chemicals also caused reduction in the development of nematodes (Miller, 1971). In present findings, lesser

number of nematodes was able to enter into successive stages of their life cycle due to the nematicidal effects of chemicals. Our results are in line with (Rehman *et al.*, 2009; Safdar *et al.*, 2012) who observed the suppressive effect of chemical on the subsequent development of *M. incognita* after invasion, that reduce invasion rate by causing paralysis of nervous system (Radwan *et al.*, 2019).

Behavioral changes have a significant effect on the development of nematodes. Inhibition in nematode development was also reported as disturbance in feeding behavior of nematodes due to nematicidal effect of oxamyl. Refusal to feed was also caused by Azadiractin due to negative effects on chemoreceptors (Aerts and Mordue, 1997). Suppression in the development was reported by several researchers (Rehman *et al.*, 2009) due to chemicals impact on nematodes. Reduction in *M. incognita* population was high in protective treatment of chemicals as compared to curative (Garabedian and Van Gundy, 1983). Maximum reduction in number of galls and J<sub>2s</sub> was recorded in protective treatment as compared to curative. These findings are in conformity with the results of other workers. Safdar *et al.* (2012) reported that nematode population was decreased in protective

application.

Reduction in number of egg masses and eggs/egg mass was reported by (Rehman *et al.*, 2009) in protective treatment of chemicals as compared to curative. Reproduction rate was also lower in protective treatment of chemicals. Decrease in nematode population was reported by (Khalil, 2012; Radwan *et al.*, 2012) under the influence of chemicals. Nematicidal potential of chemicals was featured by inactivation of enzyme of nervous system (Batiha *et al.*, 2020; Sasanelli *et al.*, 2020), blockage of electrical conductivity in muscle and nerves cells (Bloomquist, 2003), impaired muscle cells and paralysis (Cordova *et al.*, 2006), phenolic compounds (Mohan, 2011), formaldehyde and fatty acids (Khan *et al.*, 1974) nimbin, nimbidine and limonoid. In protective application, penetration of nematodes decreased as chemicals were absorbed by the root system that prevent entry of nematodes (Javed *et al.*, 2007). Fertilizers caused an increase in plant growth, seed germination, uptake of water and tolerance to diseases as they caused physiological changes in plant that hinder penetration of pathogens (Huber, 1980; Vejan *et al.*, 2016). Suppressive effects of fertilizers were recorded on nematode population (Dawar *et al.*, 2007; Hu and Qi, 2010).

An increase in fresh root and shoot weight, root length, shoot length and leaf/ plant and decrease in number of galls, females, egg masses and  $J_2/100\text{ cm}^3$  was observed in individual treatment of N as compared to P and K, similarly concomitant application of N+P was more effective N+K and P+K while NPK was most efficient in nematode suppression and plant growth improvement from all treatments. N application was more effective as involved in cellular functions and production of metabolites that caused an increased yield and reduced nematode reproduction (Bado *et al.*, 2011). Efficiency of P and NPK (Irshad *et al.*, 2006) was reported on plant growth and nematode reproduction, while K was effective only in reducing nematode population when applied at low levels (Badra and Yousuf, 1979).

The most effective fertilizer NPK was selected and further tested with Cartap, Virtako, Cure and Azadirachtin, to determine their combined impact on plant growth and nematode reproduction. Suppression in nematode population was increased in combined application of chemicals with NPK. Plant growth responses also increased in combined application due to dual effect of fertilizer on plant growth and nematode suppression (Akhtar and Mahmood, 1996). These findings are comparable with (Hemmatia and Saeedizadehb, 2020) who reported the increased efficacy of chemicals with NPK and compost.

## CONCLUSIONS

From these findings it is summarized that protective application of chemicals is more effective in reducing nematode population under field conditions as compared to curative. Consequently, fertilizers are required to enhance plant growth but their role in nematode management could not be neglected. Their integrated use with chemicals might also a successful tool in management strategies as internal effect due to fertilizers and external by chemicals caused maximum reduction in nematode population.

### Statement of conflict of interest

The authors have declared no conflict of interest.

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