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Varietal Susceptibility and Disease Resistance of Tomato Cultivars against *Meloidogyne incognita* in Greenhouse Condition

Mohd Ikram, Arshad Khan, Saba Fatima, Taruba Ansari, Mansoor A. Siddiqui, Moh Tariq

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ABSTRACT

Background: Root-knot nematodes (*Meloidogyne* spp.) are the most devastating pests of vegetables, especially in the tropic and subtropic regions. *Meloidogyne incognita* is among the major pathogens found in the tomato crop and causes a significant yield loss. This study evaluates the resistance and susceptible cultivar of tomato against root-knot nematodes (*Meloidogyne incognita*).

Methods: A greenhouse study was conducted to evaluate the resistance or susceptibility of nine tomato cultivars against *M. incognita*. Nine cultivars of tomato (Jyoti-4, Navuday, P-21, T9, Pusa-Rohini, Pusa-Sheetal, Pusa-Ruby, Tomato-Ped and Tomato-Round) were procured from Indian Institute of Agriculture Research (IARI), New Delhi and Chola seeds store Aligarh, Uttar Pradesh, India. Two weeks old seedlings of each cultivar were transplanted singly into each pot of 15 cm diameter (1 kg mixture of soil). Only one healthy seedling of each cultivar was maintained in each pot, including the control. Each pot was inoculated with 1500 freshly hatched second-stage juveniles (J2s) of *M. incognita*. This experiment was carried out in a completely randomized design (CRD) with five replications of each cultivar.

Result: According to the rating scale of galls and reductions in growth parameters, cultivars Pusa-Ruby and Tomato-Ped were found highly susceptible. Five cultivars, namely Jyoti-4, Navuday, Pusa-Sheetal, P-21 and Tomato-Round were found susceptible, while cultivar Pusa-Rohini was found moderately susceptible. Only one cultivar *viz*., T9, was found moderately resistant.

Key words: *Meloidogyne incognita*, Plant growth, Resistance, Susceptibility, Tomato.

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is a member of family Solanaceae. Tomato is a good source of vitamins (A, C and K), potassium and magnesium. It contains essential carotenoids such as lutein, lycopene and natural antioxidants.

Plant-parasitic nematodes caused significant losses in vegetable crops. They attack more than 3000 species of plants, including almost all cultivated plants and reduced world crop production (Abad *et al*., 2003). The estimated yield loss in tomato crop by plant-parasitic nematodes is about 23% in India (Kumar *et al*., 2020). Root-knot nematodes (*Meloidogyne* spp.) are one of the most economically destructive genera of nematodes in horticultural and field crops, causing an estimated annual loss of US$ 100 billion globally (Oka *et al*., 2000). They are sedentary endoparasites that affect the root system of plants (Mitkowski *et al*., 2003).

These nematodes are difficult to control due to their short life span, high population density and reproductive capacity (Sikora and Fernandez, 2005).

Various methods have been employed over the years to manage nematode infestation, including chemical, physical, organic amendments, biological control and cultural land management practices (Tariq *et al*., 2020). However, chemical nematicide usage is a common, effective and widespread practice to manage nematodes and minimize yield loss (Arita *et al.,* 2020; Ji *et al*., 2020). Nematicides use to check the development and reproduction of nematodes highly toxic to the environment. These chemicals enhance biodegradation and environmental pollution for a long time and have a negative impact on flora and fauna (Mahmood *et al.,* 2016). Because of the restrictions on chemical pesticide use and their adverse effects on the environment, as well as on human and animal health, therefore, alternative strategies for nematode management are highly desirable (Ntalli *et al.,* 2020). Nematode infestation levels could be minimized in the soil by using resistant cultivars with crop rotation (Ansari *et al*., 2018). The use of resistant cultivars is an environmentally friendly, safe, most comfortable and economically feasible alternative strategy to manage root-knot nematodes (Sujatha *et al.,* 2017). The cultivars which do not support disease development or resist the pathogens' growth and provide high yield called resistant cultivars. Numerous cultivars of different vegetable crops *viz*., tomato, carrot, okra, cucumber and brinjal have been recorded as resistant to *Meloidogyne* spp. (Tariq *et al*., 2022; Ahmad *et al*., 2018). The resistant

Department of Botany, Section of Plant Pathology and Nematology, Aligarh Muslim University, Aligarh-202 002, Uttar Pradesh, India.

Corresponding Author: Moh Tariq, Department of Botany, Lords University, Alwar-301 028, Rajasthan, India.
Email: aziztariq14@gmail.com

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cultivar may suppress the harmful nematode density by 10-50% (Oostenbrink, 1966). The susceptibility of different vegetable cultivars against nematode has been reported by researchers (Montasser *et al.,* 2019).

The present study was designed to evaluate the disease resistance in nine cultivars of tomato against *Meloidogyne incognita* in greenhouse conditions, which can be used for the sustainable management of nematodes.

MATERIALS AND METHODS

Collection of tomato cultivar

Nine cultivars of tomato (Jyoti-4, Navuday, P-21, T9, Pusa-Rohini, Pusa-Sheetal, Pusa-Ruby, Tomato-Ped and Tomato-Round) were procured from Indian Institute of Agriculture Research (IARI), New Delhi and Chola seeds store Aligarh, Uttar Pradesh, India.

Collection and maintenance of nematode inoculum

Infected roots were collected from the eggplant field from village Panjipur, district Aligarh. Eggmasses were detached from the infected roots and collected in a petri dish having distilled water. *Meloidogyne* spp. was identified by perineal patterns in the laboratory (Eisenback1985). After identification, single eggmass was cultured and maintained on eggplant in the greenhouse of the Department of Botany, Aligarh Muslim University, Aligarh. Eggmasses were hand-picked using sterilized forceps from heavily infected roots of eggplant. The second stage juveniles (J2s) were obtained from hatched eggs by incubating hand-picked egg masses in sterile distilled water at 27±2°C. The hatched juveniles were collected after every 24 hours and distilled water was added. The concentration of freshly hatched second-stage juveniles was standardized.

Screening of tomato cultivars

Screening of tomato cultivars for resistance against *M. incognita* was performed in clay pots. Surface sterilized seeds (1.0% NaOCl for 15 min) of each tomato cultivar were sown in clay pots containing 1kg autoclaved sandy loam soil and manure in the ratio 3:1 for preparation of nursery. Two weeks old seedlings of each cultivar were transplanted singly into each pot of 15 cm diameter (1 kg mixture of soil). Only one healthy seedling of each cultivar was maintained in each pot, including the control. Experiment was conducted in the year August, 2019 at section of Plant Pathology and Nematology, Department of Botany, Aligarh Muslim University, Aligarh, India.

Inoculation technique

Each pot was inoculated with 1500 freshly hatched second-stage juveniles (J2s) of *M. incognita* by making 3-4 holes around the rhizosphere of plant. This experiment was carried out in a completely randomized design (CRD) with five replications of each cultivar. The uninoculated plants were used as control. The plants were given the required amount of water and proper care was taken during the experiment period.

Data collection and observations

Plants were uprooted after three months. The roots of each tomato cultivar were gently uprooted from the pots and washed carefully to avoid egg mass losses and observations were recorded. The following morphological parameters, such as shoot and root length, fresh and dry weight of shoot and root were recorded. Physiological parameters like chlorophyll and carotenoid content were also estimated. Pathological parameters *viz*., number of egg masses/root, number of eggs/eggmass, nematode population per 250 g of soil and the number of galls were also noted. The percent reductions in various parameters over the control were calculated by using the formula.

$$\%\ \text{reduction} = \left(\frac{\text{Uninoculated-Inoculated}}{\text{Uninoculated}}\right) \times 100$$

Host rating status towards the nematode assessed by estimating the number of galls per root system.

Rating scale for the assessment of the level of resistance of plant cultivars against root-knot nematodes, based on the number of galls (Taylor and Sasser, 1978).

Root knot Index	Number of galls/roots	Resistance rating
0	0	Immune (I)
1	1-2	Resistant(R)
2	3-10	Moderately resistant (MR)
3	11-30	Moderately susceptible (MS)
4	31-100	Susceptible (S)
5	>100	Highly Susceptible (HS)

Statistical analysis

The experimental data were analyzed statistically by one-way analysis of variance (ANOVA) using SPSS-17 statistical software (SPSS Inc., Chicago, IL, USA). Mean values statistically compared differentiated by Duncan's multiple range test at $P \leq 0.05$.

RESULTS AND DISCUSSION

The statistical comparison of means of different parameters and the presence of galls revealed that none of the cultivars was found immune or highly resistant against *Meloidogyne incognita*. The nematodes displayed variable effects on the reproduction and plant growth of different tomato cultivars. According to the rating scale of galls and reductions in growth parameters, cultivars Pusa-Ruby and Tomato-Ped were found highly susceptible. Five cultivars, namely Jyoti-4, Navuday, Pusa-Sheetal, P-21 and Tomato-Round were found susceptible, while cultivar Pusa-Rohini was found moderately susceptible. Only one cultivar *viz*., T9, was found moderately resistant. All cultivars showed a significant reduction in growth parameters over their respective controls.

In the roots of highly susceptible cultivars *viz*., Pusa-Ruby and Tomato-Ped, the highest number of eggmasses were recorded as (166) and (137), respectively. Susceptible cultivars, namely, Jyoti-4, Navuday, Pusa-Sheetal, P-21 and Tomato-Round, also showed a high number of eggmasses but less than highly susceptible cultivar. Among susceptible cultivars, the number of eggmasses noted in P-21, Jyoti-4 and Tomato-Round was (118), (84) and (73), respectively, while cultivars Navuday (31), Pusa-Sheetal (59) recorded the lesser number of eggmasses. The number of eggmasses found in moderately susceptible cultivar Pusa-Rohini was (23). The minimum number of eggmasses (16) was observed in moderately resistant cultivar T9. A significant variation was also recorded in the number of eggs/eggmasses among all tested cultivars. Cultivars Pusa-Ruby and Tomato-Ped indicated the maximum number of eggs/eggmasses (253) and (218) respectively. Susceptible cultivars showed variation in the number of egg/eggmasses as P-21 and Jyoti-4 showed (152) and (121), respectively, while cultivars Pusa-Sheetal, Navuday and Tomato-Round showed (74), (57) and (98), respectively. In cultivar, Pusa-Rohini 34 eggs/ eggmasses were recorded whereas only (20) were found in cultivar T9.

All cultivars behaved differently regarding the formation of galls. The maximum number of galls was recorded in Cultivar Pusa-ruby. The second highly susceptible cultivar Tomato-Ped showed galls on the root. Among susceptible cultivars, P-21 and Jyoti-4 indicated higher number of galls in their group. The number of galls observed in other susceptible cultivars, namely, Tomato-Round, Pusa-Sheetal and Navuday. Among all cultivars, the minimum number of galls was found in moderately resistant cultivar T9. (Table 1b)

Variation in nematode populations was also observed in all tested cultivars. Cultivars Pusa-Ruby and Tomato-Ped showed the highest nematode population as (1649) and (1456), respectively. Among the susceptible cultivars, P-21 and Jyoti-4 showed higher nematode populations as (1302) and (1225), respectively, while Tomato-Round, Pusa-Sheetal and Navuday recorded as (942), (850) and (653), respectively. Nematode population observed in moderately susceptible cultivar Pusa-Rohini was (584). Least nematode population (268) was recorded in moderately resistant cultivar T9 (Table 1b).

All tested cultivars showed a significant reduction in total plant length (shoot and root) compared to their controls. Maximum percent reduction *i.e.*, 54.29% in total plant length was observed in highly susceptible cultivar Pusa-Ruby, whereas another highly susceptible cultivar Tomato-Ped showed 49.72% reduction. Minimum reduction of 18.16% in total plant length was found in moderately resistant cultivars T9. Among susceptible cultivars, the reduction in total plant length observed in P-21 and Jyoti-4 was 42.12% and 38.97%, respectively. The other three susceptible cultivars, *viz.*, Tomato-Round, Pusa-Sheetal and Navuday, showed reduction in total plant length as 34.57%, 31.13% and 30.38%, respectively. In comparison, the reduction in plant length observed in moderately susceptible cultivar Pusa-Rohini was 24.16% (Table 1a).

All of the nine cultivars showed significantly variable reductions in fresh and dry weight of plant. The cultivars Pusa-Ruby and Tomato-Ped showed maximum reduction in fresh weight of the plant as 54.41% and 50.01%, respectively. Susceptible cultivars P-21, Jyoti -4, Tomato-Round, Pusa-Sheetal and Navuday showed plant fresh weight reduction as 39.09%, 39.29%, 34.02%, 33.66% and 30.11%, respectively. The reduction in plant fresh weight observed in moderately susceptible cultivar Pusa-Rohini was 25.14%, while the minimum reduction in plant fresh weight 16.18% was found in moderately resistant cultivar T9. A similar trend was also observed in the reduction of dry weight of plant in all cultivars. Cultivars Pusa-Ruby and Tomato-Ped noted higher reduction in the dry weight of the plant as 52.13% and 48.50%, respectively. Among susceptible cultivars, a significant reduction in dry weight of the plant observe in P-21 and Jyoti-4 was 40.11% and 42.00%, respectively, whereas other susceptible cultivars Tomato-Round, Pusa-Sheetal and Navuday showed reduction in plant dry weight as 32.67%, 33.61% and 30.08%, respectively. The reduction inplant dry weight observed in cultivar Pusa-Rohini was 22.70%. Minimum reduction in plant dry weight 18.11% was found in moderately resistant cultivar T9 (Table 1a).

The nematode infestation also caused reduction in plant yield. Highly susceptible cultivar *viz*., Pusa-Ruby and Tomato-Ped showed maximum reduction in yield. Susceptible cultivars also represent significant yield reduction compared to their respective control. Susceptible cultivars, namely, P-21, Jyoti-4, Tomato-Round, Pusa-Sheetal and Navuday showed reduction in yield. Moderately susceptible cultivar Pusa-Rohini showed reduction in yield. T9 showed a minimum reduction in yield (Table 1b).

There was a significant reduction in chlorophyll and carotenoid content due to nematode infestation in all tested cultivars. Cultivars Pusa-Ruby and Tomato-Ped showed maximum reduction in chlorophyll content as 51.43% and 49.33%, respectively. Susceptible cultivars P-21 Jyoti-4, Tomato-Round, Pusa-Sheetal and Navuday showed reduction in chlorophyll content as 40.00%, 37.88%, 34.98%, 32.90% and 27.40%, respectively. Cultivars Pusa-Rohini and T9 also showed reduction in chlorophyll content as 23.71% and 19.98%, respectively. Another biochemical parameter *i.e*., carotenoid content was also recorded in all tested cultivars. Reduction in carotenoid content observed in all tested cultivars is presented in Table 1b. The reduction in carotenoid content recorded in cultivars Pusa-Ruby and Tomato-Ped was 54.38% and 48.53%, respectively. Susceptible cultivars P-21, Jyoti-4, Tomato-Round, Pusa-Sheetal and Navuday showed reduction in carotenoid content as 39.50%, 39.90%, 35.12%, 30.29% and 27.19%, respectively. Moderately susceptible cultivar Pusa-Rohini showed 22.14%, while moderately resistant cultivar T9 showed 18.03%reduction in carotenoid content (Table 1b).

Positive and significant relationships were observed between number of root gall and the percent reductions in

Table 1a: Effect of *Meloidogyne incognita* on growth parameters of different tomato cultivars.

Cultivars		Plant length(cm)			% Reduction	Fresh weight (g)			% Reduction	Dry weight (g)			% Reduction
		Shoot	Root	Total	over control	Shoot	Root	Total	over control	Shoot	Root	Total	over control
Jyoti-4	Control	59.5a	20.3a	79.8a	38.97d	55.56bc	22.85a	78.41ab	39.29c	16.28b	2.62cd	18.9cd	42.00c
	Inoculated	36.4f	12.3ef	48.7g		34.20g	13.9hi	47.6f		9.61g	1.35i	10.96h	
Navuday	Control	50.2cd	16.3bc	66.5de	30.38g	48.53def	15.66fgh	64.19d	30.11d	12.51ef	1.85h	14.36g	30.08f
	Inoculated	35.1f	11.2f	46.3g		34.00g	10.86j	44.86fg		8.75g	1.29ij	10.04h	
T9	Control	54.6bc	17.0bc	71.6cd	18.16f	58.29ab	17.48de	75.77b	16.18f	18.20a	2.73bc	20.93ab	18.11h
	Inoculated	44.5e	14.1d	58.6f		48.94de	14.57gh	63.51de		14.92bcd	2.22fg	17.14def	
Pusa-Rohini	Control	59.0a	18.0b	77.0ab	24.16h	61.59a	21.54ab	83.13a	25.14e	19.32a	2.93b	22.25a	22.70g
	Inoculated	44.8e	13.6def	58.4f		46.19def	16.04efg	62.23de		14.94bcd	2.26efg	17.2def	
Pusa-Sheetal	Control	53.5bc	17.5bc	71cd	31.13g	45.27ef	17.2def	62.47de	33.66d	14.20cde	2.52cde	16.72ef	33.61e
	Inoculated	36.7f	12.2f	48.9g		30.60gh	11.47j	42.07gh		9.65g	1.45i	11.10h	
P-21	Control	54.0bc	15.8c	69.8cd	42.12c	47.13def	20.20bc	67.33cd	39.09c	16.13b	2.32efg	18.45cde	40.11d
	Inoculated	31.3g	9.1g	40.4h		28.80h	12.21ij	41.01h		9.67g	1.38i	11.05h	
Pusa-Ruby	Control	55.5ab	18.0b	73.5bc	54.29a	51.42cd	18.35d	69.77c	54.41a	15.60bc	3.85a	19.45bc	52.13a
	Inoculated	24.4h	9.2g	33.6i		23.74i	8.07k	31.81i		7.50hi	1.81h	9.31h	
Tomato- Ped	Control	46.0de	17.2bc	63.2ef	49.21b	43.20f	15.23gh	58.43e	50.01b	12.23f	2.12g	14.35g	48.50b
	Inoculated	23.5h	8.6g	32.1i		21.60i	7.61k	29.21i		6.30i	1.09j	7.39i	
Tomato-Round	Control	48.2de	14.0de	62.2ef	34.57e	49.16de	18.71cd	67.87cd	34.02d	13.62def	2.45def	16.07f	32.67e
	Inoculated	31.2g	9.5ga	40.7h		32.44gh	12.34ij	44.78cd		8.70gh	2.12g	10.82h	

Values are mean of five replicates; Means in each column followed by the same letters are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.

Table 1b: Effect of *Meloidogyne incognita* on physiological and pathological parameters of different tomato cultivars.

Cultivar		Chlorophyll content (mg/g)	% Reduction over control	Carotenoid content (mg/g)	% Reduction over control	yield (g)	% Reduction over control	Eggmasss/ root	No. of eggs/ eggmasses	Nematode Population (250g soil)	No. of galls	Reaction
Jyoti-4	Control	2.484cd	37.88d	0.872c	39.90c	310cd	38.70d	-	-	--	-	Susceptible
	Inoculated	1.543h		0.348l		190hi		84c	121d	1225c	81d	
Navuday	Control	2.653abc	27.40f	0.853cd	27.19e	280de	28.57f	-	-	-	-	Susceptible
	Inoculated	1.931g		0.621h		201gh		31g	57g	653f	41f	
T9	Control	2.732a	19.98g	0.904ab	18.03g	313cd	19.80h	-	-	-	-	Moderately
	Inoculated	2.186f		0.741f		251ef		16h	20i	268g	10h	resistant
Pusa-Rohini	Control	2.539bcd	23.71fg	0.918a	22.14f	345abc	24.92g	-	-	-	-	Moderately
	Inoculated	1.937g		0.697g		259ef		23f	34h	584f	29g	susceptible
Pusa-Sheetal	Control	2.398de	32.90e	0.835de	30.29e	355ab	32.95e	-	-	-	-	Susceptible
	Inoculated	1.609h		0.582i		238fg		59e	74f	850e	57e	
P-21	Control	2.684ab	40.00c	0.813e	39.50c	365a	41.91c	-	-	-	-	Susceptible
	Inoculated	1.610h		0.491j		212gh		118c	152c	1302c	98c	
Pusa-Ruby	Control	2.726a	51.43a	0.912ab	54.38a	372a	52.15a	-	-	-	-	Highly
	Inoculated	1.324i		0.416k		178i		166a	253a	1649a	136a	susceptible
Tomato-Ped	Control	2.572abcd	49.33b	0.882bc	48.53b	381a	50.26b	-	-	-	-	Highly
	Inoculated	1.303i		0.453j		190hi		137b	218b	1456b	123b	Susceptible
Tomato-Round	Control	2.298ef	34.98de	0.857cd	35.12d	319bc	36.36d	-	-	-	-	susceptible
	Inoculated	1.494h		0.556i		203gh		73d	98e	942d	66e	

Values are mean of five replicates; Means in each column followed by the same letters are not significantly different according to Duncan's multiple range test at P≤0.05.

total plant length (R^2 = 0.97), total fresh weight (R^2 = 0.97), total dry of plant (R^2 = 0.98), chlorophyll (R^2 = 0.99), yield (R^2 = 0.98) and carotenoids content (R^2 = 0.99) represented by Fig 1.

The use of resistant cultivar is considered as an important strategy to manage nematode disease in the plants. In the present study, nine tomato cultivars' response was assessed based on root galls and reduction in plant growth characters. *M. incognita* caused significant damage in all the tested cultivars. According to Taylor and Sasser scale (1978), none of the cultivars was found immune or highly resistant against *M. incognita*. However, one cultivar was found moderately resistant. The present findings showed significant variations in the number of galls, eggmasses, eggs and nematode populations of *M. incognita* in all tested cultivars. The number of galls, eggmass, eggs and nematode population was significantly higher in highly susceptible and susceptible tomato cultivars than moderately resistant. While minimum was observed in moderately resistant cultivars. Resistance and susceptibility of plants against parasitic nematodes also depend on the nematode's ability to reproduce (Cook and Evans, 1987). The high number of root galls and increased population density observed in susceptible cultivars might be due to the higher invasion of nematodes preferred by susceptible cultivar. On the other hand, moderately resistant cultivar permitted few juveniles to enter in the root. Resistant host stops or slows down the invasion and growth of nematode juveniles (Williamson *et al.,* 1996). Sasser (1954) also., reported that resistant cultivars showlow nematode invasion rate than susceptible cultivars.

In the present study, the growth of tomato cultivars was negatively correlated with the number of galls. Maximum reduction in plant growth parameters was observed in highly susceptible cultivars. Susceptible cultivar showed a moderate level reduction in pant growth while moderately resistant cultivar indicated minimum reduction. Many researchers have also reported reductions in plant height and weight, yellowing of leaves as a result of root-knot nematodes infection (Azam *et al.,* 2011). Infective second-

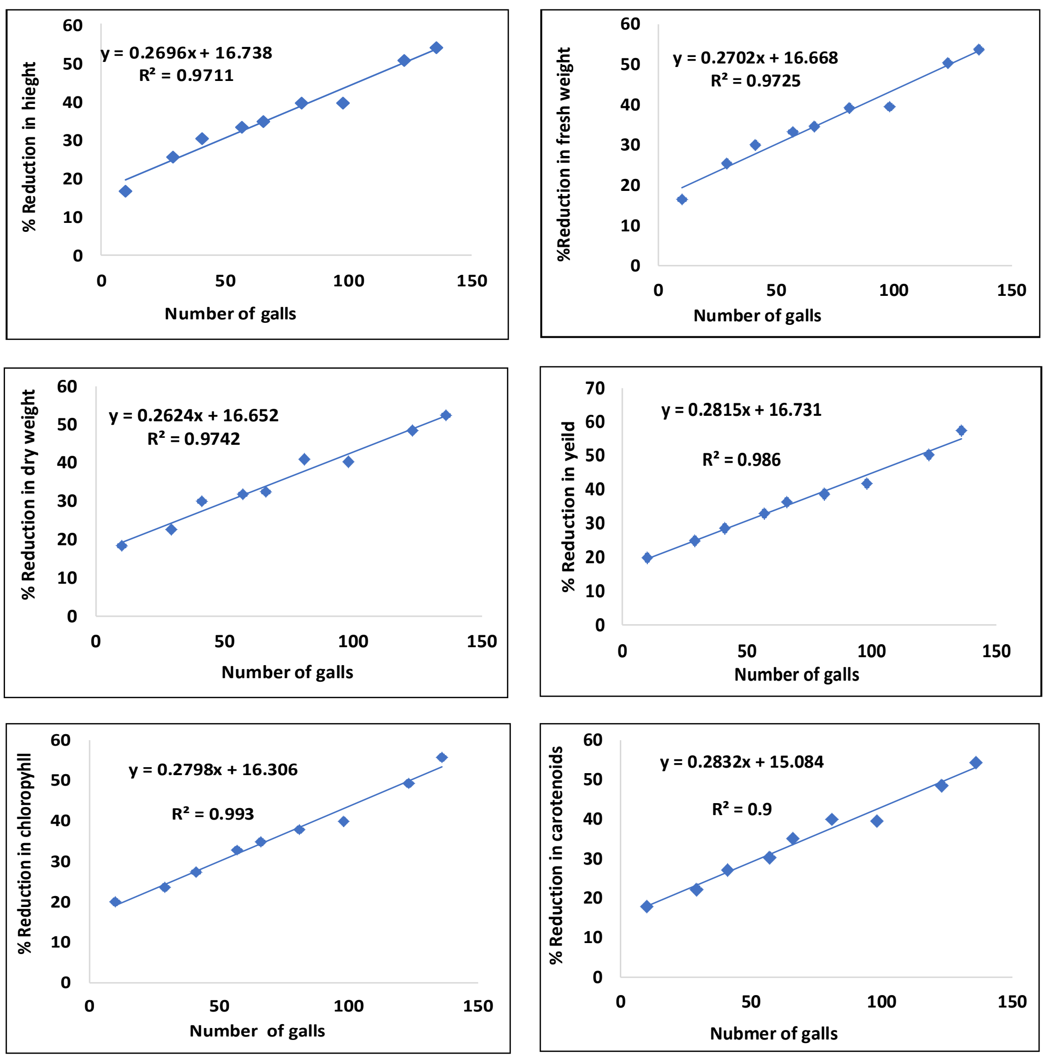

Fig 1: Relationship between number of root gall and % reduction in different parameters of tomato cultivars.

stage juveniles enter the host roots by the epidermis and move through the root cortex to establish a feeding site, called giant cells in the vascular tissues. This feeding site provides adequate nutrition for the growth and reproduction of nematodes (Abad *et al.*, 2010). These giant cells receive more nutrients, so the decreased amount of nutrients translocate to the normal cells causes reduced fruit growth (Escobar *et al*., 2015). Reduction in shoot lengths, root length and fresh weight of plant was likely a result of nematode feeding on giant cells (Siddiqui *et al.,* 2014). The formation of galls and eggmasses in the root system ceases the absorption and translocation of water and minerals in the plant, due to which plant growth is reduced. Similarly, all tested cultivars showed reduction in plant yield due to nematode infestation. Similar results were also observed by other workers that resistant cultivars yield comparatively better than susceptible cultivars (Ansari *et al*., 2018). In addition to plant growth reduction, some biochemical changes were also found in all cultivar of tomato. The reductions in chlorophyll and carotenoid content were observed, which was also supported by Melakeberhan *et al*. (1986). These reductions in chlorophyll and carotenoid contents are the results of nutrient deficiency in the plant.

CONCLUSION

This study demonstrates that the cultivation of resistant or moderately resistant cultivars could minimize nematode infestation and minimize yield losses. The use of resistant cultivars is an efficient and environmentally acceptable approach to manage root-knot nematode. Resistant cultivars are the low-cost strategy because there is no extra cost to spend as nematicide or pesticide. Furthermore, resistant cultivars can be used in breeding programs for developing new resistant cultivars against the nematode. Further study is necessary to search out nematode-resistant cultivars of tomatoes.

Conflict of interest: None.

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