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Pathogenicity of root-knot nematode, *Meloidogyne javanica* pt. 2 in groundnut

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Abstract
A study on the pathogenicity of root-knot nematode, *Meloidogyne javanica* pt. 2 to determine the economic threshold level in groundnut was conducted under pot condition at the Department of Nematology, BACA, AAU, Anand. An inoculum density of 1000 juveniles per plant per pot resulted in significant reduction of groundnut growth and development. Moreover, an increase in inoculum levels from 10 to 10,000 J_2 per plant led to a progressive increase in nematode population in both roots and soil, as well as the total nematode population. Conversely, the reproduction rate of the nematode declined with increasing inoculum levels, with the highest reproduction rate observed at 10 J_2 per plant and the lowest at 10,000 J_2 per plant.

Keywords: Groundnut, *Meloidogyne javanica* pt. 2, pathogenicity, pot condition

Introduction
Among the major oilseed crops of India, groundnut (*Arachis hypogaea* L.), also known as peanut, earthnut or monkey nut, is widely used in everyday culinary preparations. It holds a prominent position, contributing over 40-50 percent of the cultivated area and 60-70 percent of total production. China leads global groundnut production with 17.57 million tonnes, followed by India with 6.73 million tonnes (Anon., 2023) [1]. Groundnut seeds are composed of 44-56 percent oil and 22-30 percent protein on a dry seed basis (Savage and Keenan, 1994) [8]. However, despite its economic and nutritional importance, groundnut cultivation faces significant challenges from various diseases, pests and nematodes that adversely affect both yield and quality. Among these, plant parasitic nematodes particularly root-knot nematodes (*Meloidogyne* spp.) pose a significant threat to groundnut production worldwide. Their distribution and impact are influenced by favorable biological and environmental factors. In India, root-knot nematodes (*M. arenaria* and *M. javanica*) are responsible for an estimated 21.6 percent reduction in groundnut yield, resulting in financial losses of around Rs. 1911 million annually (Jain *et al.*, 2007) [4]. In Gujarat, *M. arenaria* and *M. javanica* are the predominant nematode species inflicting damage on groundnut crops, with avoidable yield losses ranging from 13-50 percent for both the species (Machal, 2023) [6]. The root-knot nematode, *M. javanica* (Pathotype 2) is first time recorded from Kapadwanj area of Kheda district in Gujarat causing considerable yield losses in pod yield of groundnut. Therefore, looking to the severe infestation problem of *M. javanica* in middle Gujarat, the current study was conducted to assess the pathogenic threshold level of *M. javanica* pt. 2 in groundnut.

Materials and Methods
An experiment was conducted to study the pathogenic effect of *M. javanica* pt. 2 on groundnut using Completely Randomized Design (CRD) with five repetitions. Earthen pots of 15 cm diameter were washed with tap water and disinfected with 4% formaldehyde (Formalin 40%) solution. After drying, pots were filled with steam sterilized sandy loam soil at 1 kg per pot. Three seeds of groundnut cv. GG 34 were sown in each pot. After germination, plants were thinned down to one plant/pot. The pots were inoculated with five inoculum levels *viz*., 0, 10, 100, 1000 and 10,000 juveniles of *M. javanica* pt. 2/pot with five repetitions of each treatment. Nematodes (J_2) were extracted from the egg masses collected from the pure culture maintained in micro plots and inoculated in the rhizosphere of each plant with the help of pippet by removing soil around plant stem by forceps.

The uninoculated plants were served as control. All plants were watered regularly to maintain optimum soil moisture and were uprooted carefully 60 days after nematode inoculation. The roots were washed with water and observations on plant growth and nematode multiplication were recorded and analysed. The observation of root-knot galling index in plants was recorded using the scale developed by Taylor and Sasser in 1978 where, 0 = No galls, 1 = 1-2 galls/root system, 2 = 3-10 galls/root system, 3 = 11-30 galls/root system, 4 = 31-100 galls/root system and 5 = >100 galls/root system. For assessment of root nematode population (number of females, no. of egg masses and no. of eggs), roots were cut in to 2-3 cm length and 3 g roots were stained in 0.05 percent acid fuchsin in lactophenol. The stained roots were washed with tap water to remove excess stain and kept overnight in lactophenol for destaining. Then, the roots were examined for nematode population. At the time of termination of experiment, final nematode population per 200 cc soil was recorded. All the observations were recorded and data were analysed using appropriate statistical procedure.

Results and Discussion

The data presented in Tables 1 and 2 indicate that increasing inoculum levels of *M. javanica* pt. 2 resulted in a significant reduction in plant height (cm) and fresh shoot weight (g) whereas fresh root weight (g), root-knot index, root nematode population, soil nematode population and total nematode population showed a progressive increase with higher inoculum levels.

Maximum plant height (31.70) was observed in the control treatment followed by inoculum level of 10 J_2/plant (31.14) and 100 J_2/plant (30.22). Minimum plant height was obtained at inoculum level of 10,000 J_2/plant (17.80) which was found at par with 1000 J_2/plant. Significant reduction in plant height was observed at 1000 J_2/plant (19.00) which was statistically differed from other lower inoculum levels (0, 10 and 100 J_2/plant). The lowest fresh shoot weight was observed in the treatment of 10,000 J_2/plant (3.46) which was at par with the inoculum level of 1000 J_2/plant (3.53) whereas the highest fresh shoot weight (5.34) was observed in control treatment. Significant reduction in fresh root weight was found at inoculum level of 1000 J_2/plant (2.37) which was found at par with 10,000 J_2/plant (Table 1).

Regarding root-knot index, all the treatments were significantly differed from each other. It increased with an increase in inoculum levels. Maximum root-knot index was recorded in the treatment of 10,000 J_2/plant (3.79) while minimum was found in the treatment of 10 J_2/plant (1.00). Control plants showed '0' index as it was not inoculated with nematodes (Table 2). Similarly, the reproduction of *M. javanica* pt. 2 increased with higher inoculum levels, as reflected by a greater number of females, egg masses, and total eggs per plant in the roots. All the treatments exhibited statistically significant differences in the number of females, egg masses and eggs per plant. Maximum number of females (126), no. of egg mass (113) and no. of eggs/plant (40350) was observed in the highest inoculum level of 10,000 J_2/plant. As expected, there was no female in control plants as nematodes were not inoculated (Table 2). With respect to soil nematode population, all the treatments significantly differed from each other and increased with an increase in inoculum levels. Maximum soil nematode population (11841) was recorded in the level of 10,000 J_2/plant whereas minimum (47) was observed at the level of 10 J_2/plant followed by the level of 100 J_2/plant. Overall, the final nematode population per plant increased proportionally with the level of nematode inoculum applied. The rate of reproduction (P_f/P_i) was observed maximum at an inoculum level of 10 J_2/plant (126.90) while minimum reproduction rate (5.25) was observed at 10,000 J_2/plant (Table 2).

The overall data indicated that significant reduction in plant growth parameters such as plant height and fresh shoot weight was observed at inoculum levels of 1000 J_2/plant of *M. javanica* pt. 2 in groundnut. In contrast, root weight was increased at each increasing levels due to gall formation on roots. This observation aligns with the findings of Patel and Dash (2021) [7] who reported that 1000 J_2 of *M. incognita* was the threshold for significant plant damage in urd bean, with all growth parameters (except root weight) decreasing as nematode density increased. Similarly, Chaudhary and Bhatt (2020) also revealed that the higher inoculum levels (1000 and 10,000 J_2) of *M. incognita* on pea caused the most severe damage, resulting in the highest number of galls, no. of egg masses and no. of eggs per egg mass. The inoculum levels of 1000 J_2/plant of *M. javanica* and above significantly reduced the growth of groundnut cv. GG 2, while root weights increased due to galling (Joshi, 1989) [5]. Likewise, Dhruj and Vaishnav (1981) [3] reported that inoculum levels of 1000 J_2 and above of *M. arenaria* significantly reduced growth in groundnut cv. GAUG 10, with marked declined in plant development at this threshold level.

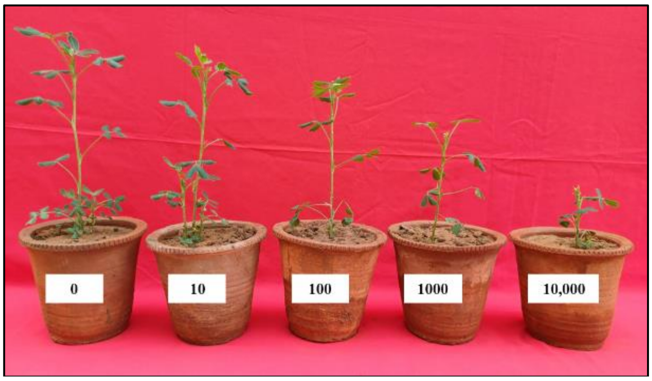
Table 1: Effect of different inoculum levels of *M. javanica* pt. 2 on plant growth of groundnut

Inoculum levels	Plant height (cm)	Fresh weight (g)	
		Shoot	Root
0	31.70	5.34	1.14
10	31.14	5.22	1.40
100	30.22	4.96	1.59
1000	19.00	3.53	2.37
10000	17.80	3.46	2.53
S. Em.±	0.45	0.13	0.10
CD at 5%	1.55	0.38	0.29
CV%	3.85	6.40	12.22

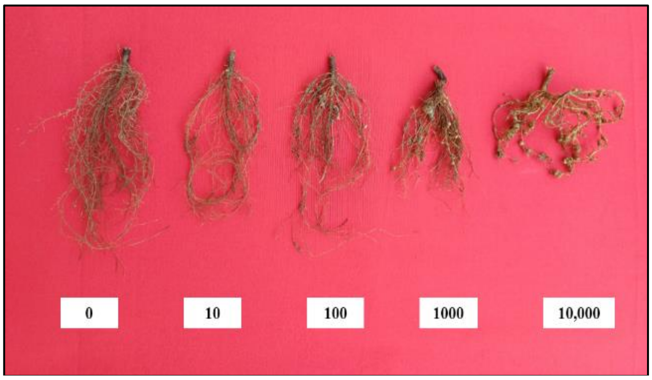
Table 2: Effect of different inoculum levels on reproduction of *M. javanica* pt. 2 in groundnut

Inoculum levels	RKI (0-5)*	Nematode population/plant				Final nematode population	R_f (P_f/P_i)
		Root			Soil		
		No. of females	No. of egg mass	No. of eggs/egg mass			
0	0.71 (0.00)**	0.00 (0)***	0.00 (0)***	0.00 (0)***	0.00 (0)***	0.00 (0)***	0.00
10	1.22 (1.00)	1.31 (19)	0.95 (08)	3.08 (1194)	1.68 (47)	3.10 (1269)	126.90
100	1.51 (1.57)	1.67 (46)	1.29 (19)	4.05 (11139)	2.21 (162)	4.06 (11370)	113.70
1000	1.97 (3.00)	1.93 (84)	1.69 (48)	4.19 (15378)	3.43 (2697)	4.26 (18214)	18.21
10000	2.21 (3.79)	2.10 (126)	2.06 (113)	4.61 (40350)	4.07 (11841)	4.72 (52479)	5.25
S. Em.±	0.05	0.04	0.02	0.02	0.01	0.02	-
CD at 5%	0.14	0.11	0.07	0.05	0.04	0.05	-
CV%	7.14	5.74	4.32	1.24	1.24	1.13	-

Note: 1. *0 = Free; 5 = Maximum disease intensity
2. **Figures in parentheses are re-transformed values of $\sqrt{x + 0.5}$
3. ***Figures in parentheses are re-transformed values of log X + 1
4. P_f = Final nematode population
5. P_i = Initial nematode population



(A) Shoot



(B) Root

Fig 1: Effect of various inoculum levels of *Meloidogyne javanica* pt. *2* on growth and nematode multiplication in groundnut

Conclusion

The pathogenicity of *M. javanica* pt. 2 at different inoculum levels 0, 10, 100, 1000 and 10,000 J_2/plant were tested to determine the economic threshold level on groundnut. The results indicated that significant reduction in plant growth parameters such as plant height and fresh shoot weight was observed at inoculum levels of 1000 J_2/plant of *M. javanica* pt. 2 in groundnut. Plant growth declined as inoculum levels increased, showing an inverse relationship between nematode inoculum density and plant development. In contrast, fresh root weight increased progressively with higher inoculum levels, primarily due to gall formation induced by nematode infestation. The root-knot index is positively correlated with nematode population density. As more J_2 infest the plant, the severity of root damage escalates and the RKI increased with an increasing nematode levels. The number of females, egg masses, eggs, soil and final nematode population per plant also increased with increasing inoculum levels whereas the nematode reproduction rate declined as inoculum levels increased. Hence, inoculum level of 1000 J_2/plant/pot and above had a detrimental impact on the growth and development of groundnut, signifying a critical threshold for economic damage.

Conflict of interest

All authors declared that they have no conflicts of interest.

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