

Original Research Article

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Studies on Pathogenicity of Potato cyst Nematode, *Globodera rostochiensis* and *G. pallida* on Potato

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ABSTRACT

The pot culture experiments were conducted under polyhouse condition to study the pathogenicity of potato cyst nematode, *Globodera rostochiensis* and *G. pallida* on potato cv. Kufri Jyoti at different inoculum levels of 0, 10, 100, 1000 and 10,000 J₂ per plant. The results revealed that, the potato cyst nematode Index was 4.0 at inoculum level of 10,000 J₂/plant and 1.75 at 10 J₂ per plant. However, the reproduction factor was highest 215.61 with the lowest inoculum level of 10 J₂/ plant where as 118.42, 47.71 and 22.02 respectively with the initial inoculum levels of 100, 1000 and 10,000 J₂ / plant. A gradual increase in the nematode inoculum levels declined the plant growth parameters progressively from 100 to 10,000 second stage juveniles (J₂) per plant. The highest reduction in plant growth was recorded at a level of 10,000 J₂/plant and the minimum was recorded at a level of 100J₂/plant, where as 1000 J₂/plant recorded as an Economic Threshold Level. Inoculation of 1000 J₂/plant showed 40.60, 32.00, 40.07 and 70.20 per cent reduction in plant height, fresh weight, dry weight and yield per plant compared to uninoculated control. The maximum of 87.35 % yield reduction was recorded, at an inoculum level of 10,000 J₂/plant followed by 70.20 and 27.00 % at a levels of 1000 and 100 J₂/plant respectively.

Keywords

Pathogenicity, *G. rostochiensis*, *G. pallida*, Kufri Jyoti

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Introduction

Potato (*Solanum tuberosum* L.) is one of the most important cash crops in the Nilgiris and Kodaikanal hills of Tamil Nadu. The major constraints in potato production are pests, nematodes and diseases which account for nearly 37% yield loss throughout the Globe,

of which the share of diseases and nematodes alone is 23% (Sasser and Freckman, 1987). Primarily, the potato cyst nematode and root knot nematode are important among plant parasitic nematodes associated with the potato rhizosphere in India. Among the plant parasitic nematodes, the potato cyst nematodes (PCN), *Globodera* spp. occupied a

position of high economic importance (Trudgill *et al.*, 2003). The demand of nutrition increases with the reproductive phase of nematode. This causes a huge yield losses ranging from 10% to 75% (Trudgill, 1986). Because of their adverse effect on the quality and quantity of tubers, easy transmission and difficulties in eradication has made this pest as the big hurdle in potato cultivation across the world. Thus both of these species are therefore included in the list of domestic as well as International Quarantine pathogens of many countries including India (Mhatre *et al.*, 2019).

Several studies showed the presence of potato cyst nematodes in soil from potato cultivated fields around the Nilgiris and Kodaikanal hills of Tamil Nadu, India (Jones 1964; Thangaraju, 1983; Seenivasan *et al.*, 2017). Also the presence of potato cyst nematodes from North Indian state, Himachal Pradesh was reported first time by Singh *et al.*, (2010). Now potato cyst nematodes became an important biotic constraint in sustainable production of potato as very recently (12th October 2018) govt. of India imposed a domestic quarantine on Himachal Pradesh, Uttarakhand, Jammu and Kashmir along with Tamil Nadu and restricted the sale of seed potatoes outside these states due to presence of potato cyst nematodes (Mhatre *et al.*, 2019).

The association of potato cyst nematodes usually showed stunting of plants, chlorosis of leaves, poorly developed root system, small sized tubers and presence of potato cyst nematodes cysts in soil. In spite of these widespread distributions, symptoms caused and potential of damage. Hence, the the present investigation was undertaken to study the pot culture experiment was conducted to pathogenicity of different inoculum levels of potato cyst nematodes on potato under polyhouse condition.

Materials and Methods

Collection of root exudates

The quality tubers of a potato cyst nematodes susceptible potato cv. Kufri Jyoti was sown in pots of 1 kg capacity filled with sterilized pot mixture soil (1:1:2 Farm yard manure: Sand: Red soil) in a polyhouse.

After establishment of the seedlings 15 – 20 days after sowing the seedling was pulled out and washed thoroughly and transferred into 500 ml beaker containing 200 ml of tap water for incubation 24 hrs at 20-25° C in the dark (Only the root system is immersed in the water).

Then the root exudates were filtered through a Whatman No.1 filter paper for getting a clear suspension and consider as a 100% concentration. Stored at 4° C and used as hatching agent described by European and Mediterranean Plant Protection Organization (2013).

Collection of second stage juveniles from cysts

For extraction of cysts, the soil samples were collected from potato fields having previous history of mixed population of potato cyst nematode, *G. rostochiensis* and *G. pallida* infestation. Then the soil samples were shade dried and then cysts were extracted using Fenwick can method (Fenwick 1940).

For hatching of potato cyst nematodes the root exudates were collected from 25-30 day old potato plants of cv. Kufri Jyoti grown in pots. Second stage juveniles were obtained from cysts pre soaked for one week in distilled water and then cysts were placed in the root exudates and kept at 18-22°C up to the hatching. The freshly harvested juveniles were used for further studies.

Pathogenicity of *Globodera* spp. on potato cv. Kufri Jyoti

The experiment was conducted in polyhouse facility of Horticulture Research Station, Udhamandalam, Tamil Nadu at a temperature of 17-25 ° C. The present study was conducted during 2018. The experimental site is located at an altitude of 2140 m above MSL at 11°24'N latitude and longitude 74°4'E.

The pot culture experiments were conducted under polyhouse condition with five treatments and four replications in a Completely Randomized Design. Plastic pots of five kg capacity were selected and filled with sterilized pot mixture soil (1:1:2 Farm yard manure: Sand: Red soil). The equal size of quality tubers were obtained from ICAR-CPRS, Muthurai, Udhamandalam. Healthy tubers of susceptible cv. Kufri Jyoti were sown in pots. After establishment of the seedlings at 20 days after sowing different populations (0, 10, 100, 1000 and 10,000) of freshly hatched second stage juveniles (J_2) of *Globodera* spp. per plant were inoculated through making 4–5 holes near the rhizosphere of each seedling. After inoculation, the holes were closed with sterilized soil followed by light irrigation and uninoculated plants were treated as control. The plants were regularly watered until their harvesting.

The experiment was terminated after 90 days of inoculations and observations on plant growth parameters, tuber yield, potato cyst nematode index and nematode population per plant was recorded. For interpretation of results, the reduction in plant growth was calculated in terms of percentage dry weight reduction. During the experimental period external symptoms of nematode infestation on plants were also recorded. Potato cyst nematodes index was determined using 1-4

grades, grade 1 = no females/inch root, 2 = 1 females/inch root, 3 = 5 females/inch root and 4 = > 10 females/inch root was given as per Grainger's test (Grainger, 1952). Potato cyst nematodes were extracted using Fenwick can method (Fenwick 1940) whereas numbers of juveniles were extracted from soil using Cobb's sieving and Decanting method followed by Modified Baermann's funnel technique. The nematode population in roots was determined by staining roots with acid fuchsin lactophenol stain (Beth *et al.*, 1941). The multiplication rate of nematode was determined by calculating the reproduction factor ($R_f = P_f/P_i$) where P_f represented the final population and P_i is the initial population of nematode. The data was analyzed statistically by applying the technique of analysis of variance (ANOVA). Mean values was calculated at 5% significance level.

Results and Discussion

After eight weeks of inoculation, *Globodera* spp. exhibited some visible symptoms were observed in infested plants inoculated with 1000 and 10,000 J_2 /plant. The result showed that, the potato cyst nematode Index was highest 4.0 at inoculum level of 10,000 juveniles and 1.75 at 10 juveniles per plant. The results of host infestation were recorded in terms of potato cyst nematode index which was directly proportional to the level of inoculum. The final root population of *Globodera* spp. was highest 905.40 females/plant at the highest inoculum level of 10,000 J_2 . However, the reproduction factor was highest 215.61 with the lowest inoculum level of 10 J_2 per plant. The reproduction factor was 118.42, 47.71 and 22.02 respectively with the initial inoculum levels of 100, 1000 and 10,000 J_2 per plant. Our findings confirm earlier results reported by Chitwood (1948) showed that a heavily infested plant with 50 g of root would support

60,000 females affect the growth and yield. Differences in environmental conditions, length of the potato growth cycle and differences in the cultivars may reflect on differences in the reproduction rates of the nematode. The results of our study revealed that, the total population was highest in the plants inoculated with 10,000 J₂/plant and lowest in those inoculated with 100 J₂/plant Table 1. A significant linear relationship was found between the initial and final population of *Globodera* spp. whereas, reproduction factor was significantly reduced with an increased inoculum levels. This decline trend of reproduction factor suggested a density dependent phenomenon.

The data presented in Table 2 clearly showed that with an increase in inoculum levels (except 0 and 10J₂/plant) of *Globodera* spp. there was a corresponding reduction in all the plant growth parameters viz. plant height, fresh weight, dry weight and yield, however, the inoculum level of 10 J₂/plant did not show any significant variation in plant growth characters and all the characters were found on par with uninoculated plants. The highest reduction in plant growth was recorded at a level of 10,000 J₂/kg soil and the minimum was recorded at a level of 100J₂/kg soil were as 1000 J₂/plant recorded as an Economic Threshold Level. Inoculation of 1000 J₂/plant showed 40.60, 32.00, 40.07 and 70.20 per cent reduction in plant height, fresh weight, dry weight and yield per plant respectively compared to uninoculated control.

The results of our study was in agreement with Sivakumar (1994) who observed that, the, economic threshold level at 1000 J₂/pot, which caused significant yield reduction by *Meloidogyne hapla* on carrot. In our study the plant growth was significantly different between 100 and 1000 J₂ inoculated plants. The maximum of 87.35 per cent yield reduction was recorded, at an inoculum level

of 10,000 J₂/plant followed by 70.19 and 26.99 per cent at a levels of 1000 and 100 J₂/plant respectively (Fig. 1). The relationship between the initial population and root weight was highly significant and negatively related.

The nematode infected plants above inoculum level of 1000 J₂/ plant was poor in vigor, unhealthy in appearance and showed severe stunted growth. Affected plants also exhibited yellowing, premature drying, shedding of leaves like symptoms with reduced size and number of tubers. The root system becomes less developed and plants produce a greater amount of lateral roots, leading to overall decreased plant growth and premature death of plant. The presence of sphere shaped white and yellow females were also noticed on the surface of root hairs.

The progressive decrease in plant growth with increasing inoculum levels of nematodes has also been reported by Trudgill *et al.*, (1975). The results of our study was in agreement with Grainger (1951) who observed that potato cyst nematode decreased the growth and weight of new tubers produced by potato cv. Epicure by more than 70% and infested plants were smaller and senesced earlier than uninfected plant.

Trudgill (1968) showed that growth of potato roots invaded by nematode larvae is markedly slower than uninoculated plants as Evans (1969) showed that roots were invaded before shoots emergence, recording more than 2000 larvae per g of root at only 60% emergence i.e.33 days after planting.

Infested plants were not only smaller, but also senesced earlier than lightly infested plants, as observed by Grainger (1951) but conflicting with the observations of Seinhorst and Den (1971) who suggested that nematode infestation delayed senescence.

Table.1 Effect of different inoculum levels of *G. rostochiensis* and *G. pallida* on nematode multiplication

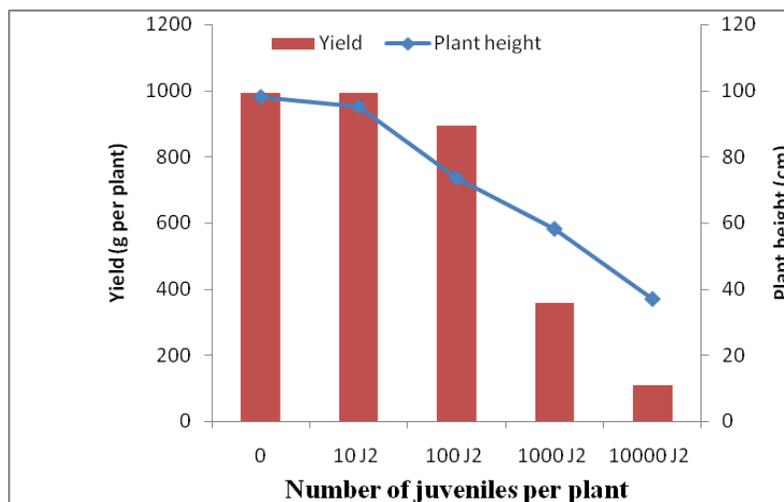
Inoculum level (Pi)	Nematode population				RF = (Pf/Pi)	PCN Index as per Grainger's test No. of females/linear inch root
	Soil population/ 5 kg (a)	Total root population/ plant (b)	No. of eggs per cyst (c)	Final Population (Pf) [a+(b×c)]		
0 uninoculated control	0	0	0	0	000-	1.00
10 second stage juveniles/plant	2.50	7.10	303.33	2156.14	215.61	1.75
100 second stage juveniles/plant	47.45	40.30	213.56	11842.32	118.42	2.75
1000 second stage juveniles/plant	450.40	325.5	145.20	47713.00	47.71	5.00
10,000 second stage juveniles/plant	778.50	905.40	176.42	220189.80	22.02	5.00
SE(d)	0.0026	4.3019	3.9110	-	-	0.2236
CD (p = 0.05)	0.0055	9.1693	8.5213	-	-	0.4766

Table.2 Effect of different inoculum levels of *G. rostochiensis* and *G. pallida* on the plant growth characters

Inoculum level (Pi)	Plant height (cm)			Plant fresh weight (g)			Plant dry weight (g)			Yield (g) per plant
	Shoot	Root	Total	Shoot	Root	Total	Shoot	Root	Total	
0 uninoculated control	62.47	35.72	98.19	210.15	24.04	234.19	25.87	6.70	32.57	692.64
10 second stage juveniles/plant	60.86 (2.58)	34.41 (3.67)	95.27	205.72 (2.10)	22.48 (6.49)	228.20	24.17 (6.57)	5.02 (25.07)	29.19	650.73 (6.05)
100 second stage juveniles/plant	51.92 (16.89)	21.69 (39.28)	73.61	177.61 (15.48)	17.60 (26.79)	195.21	20.09 (22.34)	4.01 (40.15)	24.10	505.65 (26.99)
1000 second stage juveniles/plant	44.56 (28.66)	13.76 (61.48)	58.32	143.49 (31.72)	15.76 (34.44)	159.25	15.76 (39.08)	3.76 (43.89)	19.52	206.50 (70.19)
10,000 second stage juveniles/plant	29.76 (52.36)	7.37 (79.37)	37.13	53.10 (74.73)	7.14 (70.29)	60.24	7.30 (71.78)	2.06 (69.25)	9.36	87.65 (87.35)
SE(d)	0.0647	0.0681	-	0.1747	0.0721	-	0.0552	0.0248	-	0.2380
CD (p = 0.05)	0.1380	0.1452	-	0.3723	0.1536	-	0.1177	0.0528	-	0.5073

(Figures in parentheses indicate the per cent decrease over uninoculated control)

Fig.1 Effect of different inoculum levels of *Globodera* spp. on the plant height and yield



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