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# Studies on the pathogenic potential of root-knot nematode, *Meloidogyne incognita* on urd bean

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**Abstract**
Investigation has been done on the relationships between the population density of *Meloidogyne incognita* and its effect on urd bean cv. T-9. Result of the pathogenicity test showed an initial inoculum level of 1000 $J_2$ of *Meloidogyne incognita* per kg soil was the damaging threshold level with which plant growth parameter was inversely proportional, except fresh and dry weight of root. With increasing inoculum densities reduction seen in the plant vegetative growth parameters and nematode population. An initial inoculum level of 100 nematodes per kg of pot soil caused significant reduction in plant growth parameters and proved to be pathogenic. Maximum galls per root system were recorded in 100 and 1000 $J_2$ per kg of soil, which suddenly decreases at 10000 inoculum level. Again the final nematode population increases with increase in inoculum levels but nematode multiplication rate was inversely proportional to the nematode inoculum level.

**Keywords:** Meloidogyne incognita, root-knot nematode, pathogenicity, urd bean

## Introduction
Pulse crops play an important role in improving the nutritional status of rural populations in India's semi-arid tropics. Improving availability and access to various and nutrient-dense foods such as pulses is crucial for ending malnutrition in India. India is the largest producer of urd bean accounting for more than 70% of the global production In India during *kharif* 2019-20, area is 35.53 lakh ha. Pulse crops provide a significant amount of protein to the Indian vegetarian population. Urd bean also known as Black gram (*Vigna mungo* (L.) Hepper) belongs to family Fabaceae sub family papilionaceae, is being grown as one of principle pulse crop. It has a high lysine, protein, Vitamin B, potassium, calcium, iron, niacin, thiamine and riboflavin and phosphoric acid content, making it an excellent source of balanced human nutrition. It is an N-fixing legume that upgrade soil fertility and soil physical properties (Parashar, 2006) [11]. The lowest yield was recorded in the state Odisha (326 kg/ha).Root-knot nematode (*Meloidogyne incognita*) is a common and significant group of plant parasitic nematodes that pose a threat to pulse crops such as urd bean. More than 2,000 plants are susceptible to infection, and their impact results in 5% of all global agricultural loss (Hussey & Janssen, 2002) [6]. They cause significant economic losses in agricultural crops all over the world. Keeping in view the importance of crop and less information available on pathogenicity of root knot nematodes on black gram, the study on the pathogenic potential of *M. incognita* was carried.

## Materials and methods
For the management of root knot nematode the basic information regarding the pathogenic behaviour and destructive potential of *M. incognita* is crucial. A pot culture experiment was conducted in the net house of department of Nematology, Orissa University of Agriculture and Technology, Bhubaneswar, Odisha, during the year 2016-17 to study the pathogenicity of *M. incognita* on urd bean between 21-35ºC temperature. Two weeks old healthy seedlings of urd bean var. T-9 grown in twenty earthen pots containing 1 Kg autoclaved sterilized soil + sand + farm yard manure (2:1:1) were separately inoculated with 0, 10, 100, 1000 and 10,000 second stage juveniles (J2) of root knot nematode *Meloidogyne incognita* suspension uniformly all around the exposed roots using sterilized pipette and exposed roots were immediately covered by levelling the sterilized soil.
Each five treatments having four replications including an un inoculated check arranged in Completely Randomized Design. The treatments were: T1= Check (No nematodes), T2= 10 nematodes (J2) / pot, T3= 100 nematodes (J2) / pot, T4= 1000 nematodes (J2) / pot, T5=

10000 nematodes (J2) / pot. Plant growth parameters (shoot length, root length, fresh shoot weight, fresh root weight, dry shoot weight and dry root weight) and nematode infestation parameter (number of galls, number of egg masses per plant, nematode population and multiplication factor) were recorded at 60 days after inoculation. Nematodes were isolated by Cobb's sieving and decanting method along with modified Baermann's funnel technique (Southey, 1986) and assessed. Reproduction factor was calculated by using formula, R = Pf/Pi, in which Pf represents final nematode population (root + soil) and Pi initial population of nematode. Various observations recorded from different treatments and analysed statistically.

## Results

The findings of pathogenicity experiment stated that increase in nematode inocula was associated with progressive reduction in plant growth parameters which gave conclusive evidence that *M. incognita* is potential pathogens for urd bean. The rate of multiplication was inversely proportional to the population density. An increase in level of inoculum resulted in increase in root-knots there by increasing nematode population but this increase was inversely proportional with the nematode population which was density dependent that causes competition for nutrition among the developing nematodes within available root system and also due to inability of juveniles of subsequent generation to find infection sites. The observed data were compiled in a tabular form and were subjected to statistical analysis in order to test the significance of various inoculum on plant growth and the nematode population. The effect of *M. incognita* at different inoculums was estimated on the basis of the differential changes in. plant growth parameters (shoot length, root length, fresh and dry shoot weight, fresh and dry root weights) and nematode infection parameter

**Table 1:** Influence of different inoculums level of *M. incognita* on urd bean growth parameter

| Treatments | Shoot length (cm) | % Change over control | Root length (cm) | % Change over control |
|---|---|---|---|---|
| 10N* | 43.85±0.39$^{c}$ | -0.97 | 29.8±0.34$^{c}$ | -1.65 |
| 100N | 38.30±0.62$^{b}$ | -13.5 | 27.2± 0.39$^{b}$ | -10.2 |
| 1000N | 34.13±0.50$^{a}$ | -22.9 | 23.3±0.16$^{a}$ | -23.1 |
| 10000N | 33.73±0.52$^{a}$ | -23.8 | 22.7±0.16$^{a}$ | -25.0 |
| Un-inoculated Check | 44.28±0.40$^{c}$ | | 30.3±0.34$^{c}$ | |
| SE(m)± | 0.49 | | 0.29 | |
| CD (0.05) | 1.48 | | 0.89 | |

*N; Nematode population

$^{\#}$ Similar alphabet in the superscript indicates the statistically at par. (LSD-5%) and number after ± is standard error

**Table 2:** Influence of different inoculums level of *M. incognita* on urd bean growth parameter

| Treatments | Fresh shoot weight (g) | % Change over control | Fresh root weight (g) | % Change over control | Dry shoot weight (g) | % Change over control | Dry root weight | % Change over control |
|---|---|---|---|---|---|---|---|---|
| 10N | 28.1±0.13$^{c}$ | -1 | 6.7±0.25$^{b}$ | +8 | 3.3±0.17$^{b}$ | -4 | 1.2±0.13$^{a}$ | -20 |
| 100N | 27.0±0.21$^{b}$ | -5 | 10.0±0.14$^{c}$ | +61 | 6.2±0.13$^{a}$ | -27 | 1.3±0.0a | -30 |
| 1000N | 23.1±0.15$^{a}$ | -19 | 10.9±0.31$^{d}$ | +76 | 2.5±0.09$^{a}$ | -49 | 1.7±0.18$^{b}$ | -70 |
| 10000N | 22.6±0.13$^{a}$ | -21 | 11.4±0.11$^{d}$ | +84 | 2.2±0.19$^{a}$ | -55 | 1.8±0.13$^{b}$ | -80 |
| Un-inoculated Check | 28.4±0.31$^{c}$ | | 4.7±0.07$^{a}$ | | 4.9±0.15$^{c}$ | | 1.0±0.08$^{a}$ | |
| SE(m)± | 0.19 | | 0.20 | | 0.14 | | 0.12 | |
| CD (0.05) | 0.59 | | 0.06 | | 0.42 | | 0.39 | |

*N; Nematode population

$^{\#}$ Similar alphabet in the superscript indicates the statistically at par. (LSD-5%) and number after ± is standard error

**Table 3:** Influence of different inoculums level of *M. incognita* on nematode multiplication of urd bean

| Treatments | No of galls plant$^{-1}$ | Final Nematode population |
|---|---|---|
| 10N | 18.5 (1.27 ± 0.02)$^{b}$ | 536 (2.72* ± 0.04)$^{b}$ |
| 100N | 41.3 (1.62 ± 0.05 )$^{c}$ | 2142 (3.33 ± 0.01)$^{c}$ |
| 1000N | 168.8 (2.23 ± 0.11)$^{d}$ | 5525 (3.66 ± 0.11)$^{d}$ |
| 10000N | 222 (2.35 ± 0.16)$^{e}$ | 12748 (3.87 ± 0.18)$^{d}$ |
| Un-inoculated Check | 0.0 (0.0)$^{a}$ | 0.0 (0.0)$^{a}$ |
| SE(m)± | 0.006 | 0.005 |
| CD (0.05) | 0.019 | 0.017 |

*Similar alphabet in the superscript indicates the statistically at par. (LSD-5%)

*Figures in parentheses are log transformed values

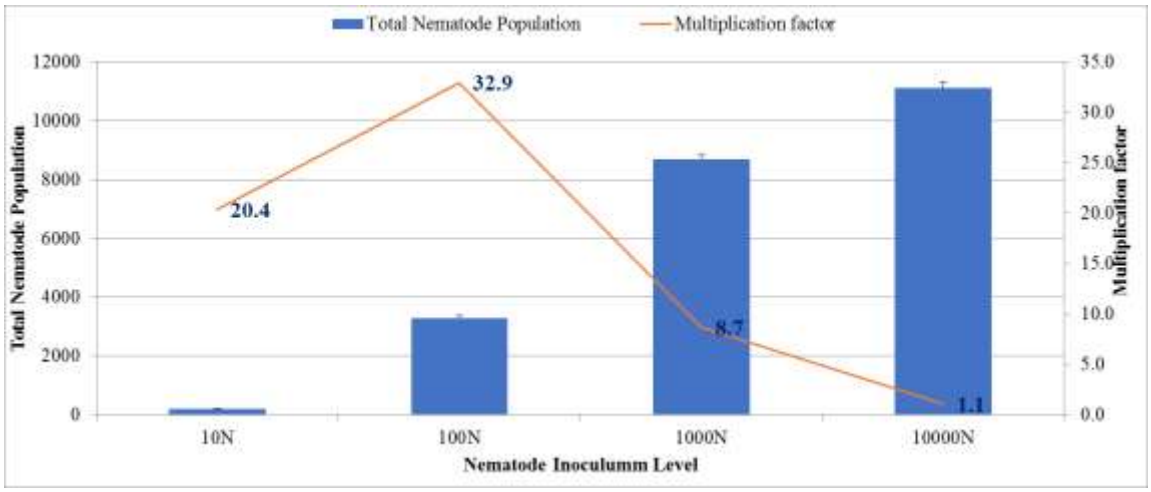


**Fig 1:** Influence of different level of inoculation on total nematode population and multiplication factor

**Table 3:** Influence of different inoculums levels of *M. incognita* pod and nodular characteristics of urd bean

| Treatments | Nodular Characteristics | | | | | |
|---|---|---|---|---|---|---|
| | Total no of nodule plant$^{-1}$ | % Change over control | No of effective nodule plant$^{-1}$ | % Change over control | Effective nodule Percentage | % Change over control |
| 10N | 144±2.74$^{c}$ | -8.28 | 119±3.20$^{c}$ | -15 | 83±1.19$^{bc}$ | -6.74 |
| 100N | 133±2.50$^{b}$ | -15.28 | 105±4.19$^{b}$ | -25 | 79±2.00$^{b}$ | -11.23 |
| 1000N | 110±1.87$^{a}$ | -29.93 | 80±2.40$^{a}$ | -42.8 | 72±2.05$^{a}$ | -19.10 |
| 10000N | 104±2.25$^{a}$ | -33.75 | 72±3.68$^{a}$ | -48.5 | 69±3.51$^{a}$ | -22.47 |
| Un-inoculated Check | 157±2.35$^{d}$ | | 140±2.06$^{d}$ | | 89±0.23$^{c}$ | |
| SE(m)± | 2.35 | | 3.20 | | 2.09 | |
| CD (0.05) | 7.11 | | 9.65 | | 6.31 | |

*Similar alphabet in the superscript indicates the statistically at par. (LSD-5%)
*Figures in parentheses are log transformed values

Although significant differences was noticed among treatment means of these parameters the mean data revealed that there were no significant differences in the shoot growth parameters with initial inoculum level of 10 and 100 J2 per pot in comparison to un inoculated treatment (T1) and between 1000 and 10000 J2 per pot. The shoot length (43.85 cm) was the highest followed by 38.30 cm, 34.13 cm, 33.73 cm and 44.28 cm in inoculated plant at 10, 100, 1000 and 10000 J2 s per pot with 0.97 percent, 13.5 per cent, 22.9 per cent and 23.8 per cent decrease, respectively over control. The tabulated data revealed that, there were significant differences in the root length with initial inoculum level of 1000 and 10000 J2 per pot in comparison to un inoculated treatment. The root length was 30.3 cm at control and reductions in root length were recorded 29.80 cm, 27.2 cm, 23.30cm and 22.70 cm at 10, 100, 1000 and 10000 J2s with 1.65, 10.2, 23.10 and 25.0 per cent decrease respectively.

There was maximum reduction in fresh shoot weight recorded at an inoculums level of 10000 J2. The reductions in shoot weight caused at a level of 1000 J2s was statistically at par at 10000 J2s. But there was progressive increase in reduction of fresh shoot weight over check recorded as 1.0, 5.0, 19.0 and 2.0 per cent at 10, 100, 1000 and 10000 J2s, respectively. The tabulated data revealed that, inoculated plants resulted 28.1, 27.0, 23.1, 22.6 per cent reduction in fresh weight of root with 10, 100, 1000 and 10000 larval inoculums, respectively. The fresh root weight at control was 4.7 g and on inoculated levels were 6.70 g., 10.0 g., 10.9 g. and 11.40 g., respectively. There was gradually decrease in the dry weight of shoot of urd bean with an increase in the inoculum density of nematode. The dry shoot weight at control was 4.90 g. and the reductions in inoculated were 3.3 g., 6.2 g., 2.5 g. and 2.2 g with 4, 27, 49 and 55 per cent at 10,100, 1000 and 10000 inoculum density of nematode. There was significant reduction of root dry weight was observed at and above 1000J2 per kg soil. The inoculated plant resulted 1.2g., 1.30 g., 1.7 g. and 1.8 g. dry root weight at 10, 100, 1000 and 10000 inoculum density of nematode with 20, 30, 70, and 80 per cent reduction respectively over check (1.0 g.) It was evident from the observation that the reduction in plant growth character of black gram was directly proportional to the inoculum level of *Meloidogyne incognita* with increasing level of the inoculums from 10 to 10000 J2 of *M. incognita* with non-significant between the inoculum levels of 1000 and 10000 J2/ pot (Table 1&2). But the significance reduction in plant growth was noticed at and above 1000 J2/ pot. An increase in nematode inoculum was associated with progressive reduction in plant growth parameters of urd bean which gave conclusive evidence that *M. incognita* is potential pathogen for urd bean. Statistical analysis on number of galls, egg masses and nematode population in soil indicated significant difference among the treatments. The number of galls per plant varied from 18.5 to 222 with the increase of inoculum level from 10 to 10,000 juveniles per kg of soil. Same trend was noticed in case of final nematode population per plant varying from 536 to 12748 with the increase of inoculum level from 10 to 10,000 juveniles per kg of soil. Similarly total no of nodule plant-1, No of effective nodule plant-1, Effective nodule Percentage also decrease with increasing level of the inoculums.

## Discussion

In the present investigation the multiplication factor of *M. incognita* showed a declining trend with increasing initial

inoculum levels, as root surface area for both the lower and higher inoculum level remained the same. Crowding of nematode at high inoculum densities cause competition for root surface among nematodes resulted not finding a site for penetration, natural death for lack of nutrition and reduced multiplication. High rate of multiplication factor at low level of inocula on the other hand, may be possibly be due to positive factors like abundance of food, lack of competition and ability of host to support population levels. The rate of multiplication was inversely proportional to the population density which was in conformity with Seinhorst (1970) [12]. An increase rate of reproduction also observed which proved are good host for root-knot nematode, *M. incognita*. Vovlas and Di Vito (1991) [15] reported relationship between initial and final population densities on coffee seedlings in a glasshouse experiment. Agwu and Ezigbo (2005) [1], Anwar *et al*. (2007) [3], El-Sherif *et al*. (2007) [5] and several other also workers reported that root galls increased progressively and significantly with increased levels of inoculum. Plant growth in relation to inoculums levels have been reported by other workers on different crops (Mani and Sethi, 1984; Mahapatra *et al*., 1999; Khan, 2003; Khan and Hussain, 1989; Mucksood *et al*., 2011; Singh *et al*., 2012; Mukhtar *et al*., 2013 and Anamika, 2015) [8, 7, 9, 13, 10, 2]. Significant reduction in the number of nodules was also observed at the inoculum densitv of 1000 $J_2$/kg soil. Such adverse effect on nodulation was reported earlier by Chahal *et al.,* (1988) [4] and Verdejo *et al*. (1958) [14] in urd bean. The rate of nematode multiplication was inversely proportional to the inoculumn level.

## Conclusion

conclusive evidence for *M. incognita* is potential pathogens for urd bean was confirmed by increase in nematode inoculums with progressive reduction in plant growth parameters of urd bean. Rate of multiplication was inversely proportional to the population density is due to competition for nutrition among the developing nematodes within root system and inability of juveniles of subsequent generation to find infection sites.

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