

***In-vitro* inter-relationship between plant growth promoting rhizobacteria and root knot nematode (*Meloidogyne incognita*) and their effect on growth parameters of brinjal**

M. Inam-ul-Haq, J.A. Tariq, N. Javed, N.A. Khan, Imran-ul-Haq**
and H.U. Khan*

***Department of Plant Pathology, University of Agriculture, Faisalabad.*

**Pakistan Science Foundation, Islamabad.*

Abstract

The influence of rhizobacteria as the treatment on germination, migration and penetration of *Meloidogyne incognita* in brinjal was evaluated under laboratory conditions. The results obtained were highly significant and revealed that *Pseudomonas fluorescens* promotes germination 87.5% and was effective in reducing root penetration by *Meloidogyne incognita* i.e. 39.3 juveniles. Due to the effect of *P. fluorescens*, the plant height increased by 40.9%, number of leaves was maximum i.e., 50%, number of gall formation was also controlled i.e., 70.3%. It was concluded from the studies that rhizobacterium *Pseudomonas fluorescens* is a potential biocontrol agent and it has ability to increase the yield and suppress the attack of plant pathogen.

Keywords: Rhizobacteria, *Pseudomonas fluorescens*, *Meloidogyne incognita*, brinjal or eggplant, root-knot.

Introduction

Brinjal or eggplant belonging to family *Solanaceae* is one of the most common low priced, popular and principal vegetable crops grown in different parts of the world, including Pakistan, India, China, France, Italy and United States. It is said to be good for diabetic patients and for those suffering from liver complaints and is available in the market round the year.

The annual yield losses in eggplant due to root-knot nematode are said to be 16.9% (Sassar, 1989) or 32.37% (Dareker *et al.*, 1990). The nematode infested plants exhibit the symptoms of root galling, yellowing of leaves and stunting.

Use of chemical pesticide is a common method for pest control. But in view of environmental constraints, biological control is supposed to be one of the best alternate methods for plant disease control. Fungi and Bacteria have been described as a potential biological control agents (Laugtenberg *et al.* 1991) especially the fluorescent *Pseudomonas* (Schroth & Hancock, 1982).

The present study therefore aims at *in-vitro* interrelationship between *Pseudomonas fluorescens* and *Meloidogyne incognita* and their effect on development of brinjal plant.

Material and Methods

Diseased brinjal plants showing characteristic symptoms of root knot nematode *Meloidogyne* spp. alongwith soil were collected.

Meloidogyne incognita J-2S were obtained from severely infested roots of brinjal by Hemming tray method (Whitehead & Hemming, 1965).

After isolation the suspension of juveniles J-2S were bubbled with a pipette and three replications of 2 ml each of aliquots of J-2S were counted in a counting dish. Then they were picked with the help of bamboo needle. The nematodes after heat-killing, were fixed in the FAA fixative. Temporary and permanent slides were prepared for microscopic studies. Processing of root samples: includes staining of nematodes within roots with Lactophenol-acid fuchsin. The root knot nematodes were identified on the basis of perinial pattern (Hartman & Sasser, 1985). After identification, the root knot nematodes were cultured on eggplant var. Black-beauty. Rhizobacteria i.e., *Pseudomonas fluorescens* strain-5 which is a plant growth promoting bacterium was isolated from chickpea rhizosphere by dilution plate technique and identified (Hartman & Sasser, 1985). Experiment was done to determine the effects of *P. fluorescens* as seed treatment of brinjal on germination of seeds and effect of *P. fluorescens* treatment on the growth parameters of brinjal and development of root knot disease *in-vitro*.

Results

Effect of *P. fluorescens* as seed treatment of brinjal on germination percentage after 10

days of sowing: In this study experiment regarding germination of seed treated with *P. fluorescens* was mentioned as T₁ and F value in respect of germination percentage in T₁ 3rd, 4th, 5th, 6th, 8th, 9th and 10th day of sowing was significant showing germination percentage 3rd to 10th day 34.35%, 40.8%, 40.8%, 50%, 5%, 78.12%, 87.5% and 87.55 respectively and the seeds without treatment were mentioned as T₂ and germination percentage of T₂ was 21.8, 21.8, 28.12, 37.5, 46.87, 65.62 and 68.75% respectively. The results of treatment T₁ significantly differ from control.

Effect of *P. fluorescens* on the growth parameters of brinjal and development of root knot nematode disease was observed in pot experiment are shown below:

Plant Height: comparison of treatment means showed that plant height was maximum in T₃ i.e., 40.38% and minimum in case of T₂ which gave 21.9% decrease over control (Table 1).

Comparison of Treatment Means Fresh Weight of Shoot: Comparison of treatment means indicated that fresh weight of shoot was maximum in case of T₃ (*P. fluorescens*) which gave 40.28% increase over control and was minimum in T₂ (Nematode alone) which gave 27.12% decrease over control (Table 1).

Comparison of Treatment Means of Fresh Weight of Root: Comparison of treatment means showed that fresh weight of root was maximum in T₂ showing 24.18% increase over control and was minimum in T₃ gave 4.37% increase over control (Table 1).

Comparison of Treatment Means of Dry Weight of Shoot: Comparison of treatment means indicated that dry weigh of shoot was maximum in T₃ which gave 41.3% increase over control and was minimum in T₂ which gave 48% decrease over control (Table 1).

Comparison of Treatment Means of Dry Weight of Root: Comparison of treatment means indicated that weight was maximum in T₂ 99.7% increase over control, which was best (Table 1).

Comparison of Treatment Means of Number of Galls: There was no gall formation in case of T₃ and T₄ because sterilized soil was used. In treatment T₁ number of galls were 146. It means *P. fluorescens* in T₂ controlled gall formation (Table 1).

The results demonstrate that the application of *P. fluorescence* controls *M. incognita* and have potential for possible management of root knot nematode in brinjal.

Table 1: Effect of *P. fluorescens* on the growth parameters of brinjal and development of root knot nematode disease

| Treatments | Mean Plant Height | Mean Fresh weight of shoot | Mean Fresh weight of Root | Mean Dry weight of Shoot | Mean Dry Weight of Root | Mean Number of Galls |
|----------------|-------------------|----------------------------|---------------------------|--------------------------|-------------------------|----------------------|
| T ₃ | 18.25A | 3.507A | 1.480B | 1.750 A | 0.2625C | 0 |
| T ₁ | 16.47B | 2.840B | 1528B | 1.602 B | 0.6760B | 146 |
| T ₄ | 13.00C | 2.500C | 1.418C | 1.238 | 0.4175 | 0 |
| T ₂ | 10.15D | 1.822D | 1.761A | 1.180D | 0.8338A | 0 |

Discussion

Chemicals, though are in extensive use for control of plant diseases, but besides their high cost of application create air pollution and health hazard problems in the biological life.

In the experiment regarding germination, seeds treated with *P. fluorescens* was mentioned as T₁ and seeds without treatment were taken as T₂. The results of treatments T₁ significantly differ from control. Similar results were obtained in green house tests on cotton, tomato, peanut and sugarbeet treated with *Bacillus subtilis* to control *M. incognita* and *M. arenasica* (Sikora, 1988).

Nematodes seemed to be optimum targets for biological control with rhizobacterial system. Only short period of rhizobacteria activity at the root

surface would be necessary to reduce nematode root damage since juvenile survival in soil after hatch is of short duration (Sikora, 1988).

Pseudomonas fluorescens produces Siderophores and antibiotic like phenazine (Brisbane *et al.*, 1987) and other antibiotics which suppress the pathogenic attack and enhance development.

It is believed that antagonistic reactions observed in these studies may have been based on mechanisms that alter, hatch, attract and cause host recognition. Similar results by seed treatments were also obtained by Oostendorp & Sikora (1989).

The results of different parameters of growth, revealed that application of biocontrol agent *P.*

fluorescens enhance plant growth characteristics such as plant height, fresh and dry weight of shoot and significantly reduces the root knot incidences. Moreover, ethylene production is reported to increase weight of root infested with *M. incognita* due to gall formation (Glazer *et al.*, 1983).

The results demonstrate that these applications of *P. fluorescens* control *M. incognita* and have potential for possible management of root knot nematode in brinjal.

References

- Brisbane PG, Jonik LJ, Tate ME, 1987. Revised structure for Phenazine antibiotic from *P. fluorescens*. *Antimicrob-Agents Chemother.*, **31**: 1967-1971.
- Dareker KS, Mhase NL, Sheke SS, 1990. Effect of green gram seed treatment with certain nematicides on root knot nematode and crop yield. *Int. Nematol. Network. Newsletter*, **7**(3): 4-5.
- Glazer I, Orion D, Apelbaum A, 1983. Interrelationship between ethylene production, gall formation and root knot nematode development in tomato plants infected with *M. javanica*. *J. Nematol.*, **15**: 539-544.
- Hartman KM, Sasser JN, 1985. Identification of *Meloidogyne* spp on the basis of different host tests and perinéal pattern morphology. In: *Advanced Treatise on Meloidogyne Methodology* (K.R. Barker, C.C. Carter and J.N. Sasser, eds.), Vol.II. Publ. Dept. Pl. Path. NCSU and USAID Raligh.
- Laugtenberg BJJ, de Weger LA, Bennet JW, 1991. Microbial stimulation of growth and protection from diseases. *Curr. Top. Biotechnology*, **2**: 457-464.
- Oostendorp M, Sikora RA, 1989. Seed treatment with antagonistic rhizobacteria for the suppression of *Heterodera schachtii* early root infection of sugar beet. *Revued. Nematology*, **12**: 77-83.
- Sassar JN, 1989. Plant parasitic nematode, the farmer's hidden economy. Deptt. Pla. Path. North Carolina State University U.S.A., P-13.
- Schroth MN, Hancock JG, 1982. Disease suppressive soil and root colonizing bacteria. *Science*, **216**: 1376-1381.
- Sikora RA, 1988. Interrelationship between plant health promoting rhizobacteria. Plant parasitic nematodes and soil microorganism. *Med. Fac. land bouw. Rijk suniugent.*, **53/2b**: 867-878.
- Whitehead AG, Hemming JK, 1965. Comparison of some quantitative methods of extracting small vermiform nematodes from soil. *App. Biol.*, **55**: 25-38.