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Efficacy of Some Bacterial Biocontrol Agents as Seed Treatment against Root Knot Nematode, *Meloidogyne incognita* on Tomato

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A B S T R A C T

Pot experiment was conducted to evaluate the efficacy of two formulations (talc formulation and vermi formulation) of four bacterial nematode biocontrol agents *viz., Bacillus subtilis, Bacillus pumilus, Bacillus megaterium* and *Pseudomonus flourescens* as seed treatment against root knot nematode, *Meloidogyne incognita* on tomato. All the bioagents were tries at 10 and 20g / kg of seed. The experiment was terminated at 60 days after sowing the seed. The results revealed that all the treatments significantly increased the plant growth parameters and reduced the nematode multiplication over untreated control. However, maximum plant growth parameters and minimum galls per root system, eggmasses per root system, eggs per egg masses and final nematode population was recorded when seeds of tomato (var. Pusa Ruby) was treated with talc formulation of *Pseudomonus flourescens* @20g/kg followed by seed treatment with vermi formulation of *Bacillus subtilis* @ 20g/kg of seed.

Introduction

In recent years, management of plant parasitic nematodes using biocontrol agents (fungal as well as bacterial) is gaining importance in the light of increased awareness of environmental and human health hazards associated with nematicidal chemicals application. Biological control promises to be one of these alternatives. Biological agents are environment-friendly, hence cause no side effect, they are affordable compared to agrochemicals (pesticides, insecticides, nematicides) and easy to use. Among the biological control agents that have been assessed against nematodes are antagonistic bacteria, nematophgous fungi and yeasts (Das and Borgohain, 2018; Jayakumar, 2019). A good number of bacterial biocontrol agents have been identified for their nematicidal action on root knot nematodes and becomes promising source of biopesticides. The present investigation aims to evaluate the efficacy of few bacterial biocontrol agents as

seed treatment against root knot nematode, which is responsible for 27.21 per cent annual yield loss of tomato.

Materials and Methods

Pot experiment was laid out at 4CRD in the Net House of Department of Nematology, Assam Agricultural University, Jorhat with thirteen treatments, *viz.*, T_1 : Seed treatment with *Bacillus subtilis* (1×10^9cfu/gm of talc formulation) @10gm/kg of seed; T_2: Seed treatment with *B. subtilis* (1×10^9cfu/gm of talc formulation) @20gm/kg of seed; T_3: Seed treatment with *B. subtilis* (1×10^9cfu/gm of vermi formulation) @ 10 gm/kg of seed; T_4: Seed treatment with *B. subtilis* (1×10^9cfu/gm of vermi formulation @ 20 gm/kg of seed; T_5: Seed treatment with *B. pumilus* (1×10^9cfu/gm of talc formulation) @10gm/kg of seed; T_6: Seed treatment with *B. pumilus* (1×10^9cfu/gm of talc formulation) @20gm/kg of seed; T_7: Seed treatment with *B. megaterium* (1×10^9cfu/gm of talc formulation) @ 10gm/kg of seed; T_8: Seed treatment with *B. megaterium* (1×10^9cfu/gm of talc formulation) @ 20gm/kg of seed; T_9: Seed treatment with *B. megaterium* (1×10^9cfu/gm of vermi formulation) @ 10gm/kg of seed; T_{10}: Seed treatment with *B. megaterium* (1×10^9cfu/gm of vermi formulation) @20gm/kg of seed; T_{11}: Seed treatment with *Pseudomonas fluorescence* (1×10^9cfu/gm of talc formulation) @ 10gm/kg of seed; T_{12}: Seed treatment with *P. fluorescence* (1×10^9cfu/gm of talc formulation) @20gm/kg of seed and T_{13}: Untreated control. Seeds of susceptible tomato variety (Pusa Ruby) were pre soaked in water for 12 hours, treated with biocontrol agents with calculated amount and then incubated at 25-30^{o}C for 48 hours. Treated seeds were raised in sterilized pot mixture of sand, field soil and cowdung at 1:2:1 in 1kg earthen pots. Seedlings were thinned to keep one healthy seedling per pot at 3 leaf stage. Second stage juveniles of root knot nematode *Meloidogyne incognita* were inoculated around the root zone of tomato seedlings at $1J_2$ /1g of soil. Seedlings were maintained following package of practices. The experiment was terminated at 60 days after sowing the seeds and observations on plant height, root length, fresh and dry shoot weight, fresh and dry root weight, number of galls per root system; numbers of egg masses per root system and final soil nematode population were recorded.

Results and Discussion

Results revealed that all the treatments significantly increased the plant growth parameters and reduced the infestation of root knot nematodes in tomato. However, maximum plant growth parameters and minimum number of galls, eggmass per root system, final soil nematode population were recorded when tomato seeds (var. Pusa Ruby) were treated with *Pseudomonas fluorescens* @ 20g/kg seed (T_{12}), followed by seed treatment with *Bacillus subtilis* (vermi formulation) @20g/kg seed (T_4); whereas, minimum was recorded in untreated control (T_{13}) (Table 1).

Maximum plant height (39.23 cm), root length (16.29cm), fresh shoot and root weight (15.59g and 9.70g), dry shoot and root weight (5.88g and 4.81g) were recorded when seeds were treated with *Pseudomonas fluorescens* @ 20g/kg seed. All plant growth parameters were minimum at control, where only *M. incognita* were inoculated. Minimum galls per root system (12.00), eggs per egg mass (16.25) and final nematode population per 200 cc of soil (110.25) were recoded when seeds were treated with *Pseudomonas fluorescens* @ 20g/kg seed (T_{12}), followed by seed treatment with *Bacillus subtilis* (vermi formulation) @20g/kg seed (T_4); whereas, minimum was recorded in untreated control (T_{13}) (Table 2). The result of the present

investigation clearly showed that bacterial bio-agents can effectively manage the infestation of root knot nematode, *M. incognita* in tomato.

Table.1 Effectiveness of bacterial bioagents as seed treatment on plant growth parameters of tomato (var.- Pusa Ruby)

Treatment	Plant Height	Fresh Shoot weight	Dry shoot weight	Root Length	Fresh Root weight	Dry root weight
T1	31.40	10.90	3.96	11.55	5.29	2.33
T2	31.78	10.95	3.73	13.29	6.17	2.94
T3	32.48	12.93	3.74	12.57	6.01	2.65
T4	37.13	14.69	4.23	14.51	7.90	3.83
T5	29.90	9.54	2.67	10.77	5.15	1.99
T6	33.33	11.38	3.27	11.99	5.62	2.42
T7	29.98	9.03	2.59	10.96	5.17	1.79
T8	29.10	8.47	2.53	11.30	6.90	3.32
T9	31.20	9.085	2.41	11.56	5.80	1.16
T10	31.73	10.44	2.92	11.63	5.61	1.34
T11	33.18	12.36	3.24	12.25	6.62	1.88
T12	39.23	15.59	5.88	16.29	9.70	4.81
T13	21.63	7.11	1.49	9.22	4.65	1.13
SEd	3.37	1.32	0.7	0.88	0.89	0.56
CD(0.05)	5.65	2.21	1.18	1.48	1.49	0.93

Table.2 Effectiveness of bacterial bioagents as seed treatment on infestation of root knot nematode, *Meloidogyne incognita* on tomato

Treatment	Gall/root system	Egg mass/root system	Final nematode population (200 cc of soil)
T1	34.0	36.50	145.25
T2	20.50	26.00	138.25
T3	33.00	31.50	149.00
T4	19.00	21.00	131.00
T5	34.50	34.50	162.25
T6	29.25	27.50	155.50
T7	36.50	36.50	168.25
T8	35.00	30.00	143.00
T9	39.00	31.50	164.50
T10	32.50	25.00	147.00
T11	29.50	20.50	130.50
T12	12.00	16.25	110.25
T13	50.50	41.00	231.25
SEd	3.16	2.89	10.65
CD(0.05)	5.29	3.84	17.84

The present findings are in the line of findings reported by Verma *et al*., (1998), Ali *et al.*, (2002), Hashem and Abo-Elyousr (2011) and Roy *et al*., (2015). Verma *et al*., (1998) reported that application of *Pseudomonas fluorescens* @ 10g/kg seed was effective in reducing the menace of root-knot nematode, *Meloidogyne incognita* in tomato. Hashem and Abo-Elyousr (2011) conducted an experiment to evaluate the nematicidal activity of *Pseudomonas fluorescens*, *Paecilomyces lilacinus*, *Pichia guilliermondii* and *Calothrix parietina* against root-knot nematode (*Meloidogyne incognita*) on tomato. Tomato seeds were immersed in the biocontrol agent suspensions, which contained separately 10^8 cell ml^{-1} of *P. fluorescens*, 10^5 CFU ml^{-1} of *P. lilacinus*, 10^8 CFU ml^{-1} of *P. guilliermondii* and 10^5 cell ml^{-1} of *C. Parietina*. Sterile distilled water (DW) was used as control. Results revealed that all treatments significantly reduced the population of *M. incognita* and root galling on tomato along with increased percentage of germination and vigor index (VI) except, *C. parietina*, which has no significant difference from the control. Among all the treatments the most vigorous root system was obtained in the treatment with *P. fluorescens*. Sonkar *et al.*, (2018) observed that *P. fluorescens* was able to reduce nematode parameters at different concentration when applied either as seedling root dip treatment or soil drench around tomato. Siddiqui and Shaukat (2002) revealed that these rhizobacteria have the ability to induce systemic resistance against root knot nematode, which may paly role in the present study.

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