# **BIOLOGICAL CONTROL OF ROOT-KNOT NEMATODE (MELOIDOGYNE**

# **GRAMINICOLA) OF RICE AND THEIR MANAGEMENT**

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#### *Abstract*

*Basmati is one of the most popular aromatic rice cultivated in India and Pakistan. The basmati grains are identified by their unique aroma, cooking quality, and appearance. The typical fragrance of basmati rice is one of the most inescapable characteristics for consumer's acceptability. Root-knot nematodes, Meloidogyne spp., are sedentary, obligatory root endoparasites of great economic importance, and polyphagous species. Root-knot nematode (RKN) is considered as one of the most important damaging parasites for upland, lowland, and deep-water rice cultivation throughout the world, particularly in south and Southeast Asia. Biological control is regarded as a promising alternative and a wide array of microbial bio control agents (BCA) have been developed in the past decades for the management of pathogens causing the diseases in plants. Several Trichoderma species and isolates have been evaluated as bio control agents for the management of nematodes. Application* 

*of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil was found most effective to reduce the galls formation and enhance the growth parameters of rice plant.*

# *Key words: Basmati Rice, Root-Knot Nematode, Bioagents and Management* **1. INTRODUCTION**

Root-knot nematodes (RKNs) Meloidogyne spp. are worldwide plant pathogens playing a detectable role in limiting the productivity of economic agriculture crops in temperate regions. The root-knot nematode, Meloidogyne graminicola Golden et Birchfield is the most widely distributed serious pest of rice (Oryza sativa L.) in the sub-tropics and tropics and is considered economically important in all rice ecosystems **(Pankaj et al., 2010).** M. graminicola occurs across a range of different rice ecosystems, from completely aerobic upland systems to lowland rain-fed and deepwater rice systems that experience intermittent anaerobic and aerobic cycles. While it cannot penetrate roots in flooded soils, M. graminicola can survive long periods in anoxic environments and rapidly reinvade roots whenever soils are drained **(Bridge and Page 1982; Padgham et al., 2003).** Meloidogyne graminicola (which was first discovered by Golden and Birchfield in 1965 (Nematoda: Meloidogynidae)), commonly named as the rice RKN, is considered as one of the most important damaging parasites for upland, lowland, and deep-water rice cultivation throughout the world, particularly in South and Southeast Asia. The second juvenile stage  $(J_2)$  is the infective stage that hatches from the egg under favorable environmental conditions, finds the root, enters the meristematic zone, and induces the formation of giant galls by continuous feeding. Rice is the most important host for rice RKN, but this nematode has a wide range of alternative hosts, including many weeds commonly found in rice fields that may offer refuge to these nematodes.

Biological control and other eco-friendly disease control measures have gained increasing interest among researchers after the environmental restrictions on nematicidal use for controlling plant parasitic nematodes. Plant growth promoting rhizobacterium (PGPR) that belongs to Bacillus spp. is being exploited commercially for plant protection to induce systemic resistance against various pests and pathogens **(Mostafa et al. 2014).** Many species of nematode are free-living, but some nematodes have developed the ability to parasitize other organisms. The evolution of plant parasitism in nematodes has occurred independently on several occasions **(Van Megen et al., 2009**), giving rise to at least four different groups of plant-feeding nematodes, which include over 4100 species (**Decraemer** and **Hunt**, **2013**). Plant-parasitic nematodes (PPNs) are responsible for more than \$US80 billion losses in worldwide agriculture annually (**Nicol et al., 2011**). The most economically important species are the sedentary endoparasitic nematodes, including the root-knot nematodes (**Jones et al., 2013**).

#### **2. MATERIALS AND METHODS**

#### **2.1 Experimental site and location**

In the present investigations, field experiments were conducted at Crop Research Centre (CRC) of the Sardar Vallabhbhai Patel University of Agriculture & Technology, Meerut, (U.P.). Pot experiments were carried out in the premises of College of Agriculture and laboratory experiments were conducted in Nematology lab, Department of Plant Pathology, College of Agriculture, situated in the main campus of the Sardar Vallabhbhai Patel University of Agriculture & Technology, Meerut, (U.P.) India.

#### **2.2 Geographical situation**

The district Meerut is situated between 28° 59' 0" 'N latitude and 77º 45'E longitude at an altitude of 237 meters above the mean sea level. The district Meerut falls under north western plains. The district

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is spreaded over a geographical area of 2564  $km^2$ . The general climate of this district is semi-arid and sub-tropical, characterized by very hot summer and cold winters. The maximum temperature shoots up to 42 °C during summer whereas, minimum temperature remain 7- 8 °C and below during winter season. The average annual rainfall is 863 mm, 75-80 per cent of which is received through south west monsoon during the month of July to September. But few rain shower occasionally occur in the winter and summer season, also.

# **2.3 Culture medium and their preparations**

During the study, different fungal and bacterial bioagents were used. For cultivation of these microorganism different media viz., PDA, NAM and King's B were used. Composition and methods of preparation is as following:

# **2.3.1 Composition of Potato dextrose Agar**



# **2.3.2 PDA preparation procedure**

The required quantity of peeled potatoes were cut into small pieces and boiled in 500 ml of distilled water till the pieces became soft. The potato extract was filtered through muslin cloth and the filtrate was collected in the beaker. Rest of the 500 ml water was made warm and 20 g agar and 20 g dextrose was added properly by stirring through glass rod. Both solutions were mixed together in a beaker and volume was maintained up to 1000 ml by adding required amount of distilled water. 200 ml of this solution was dispensed in each conical flask of 250 ml capacity. Flasks were plugged with nonabsorbent cotton plugs and wrapped with silver foil. Flasks containing medium were sterilized at 121  $\degree$ C at 15 lbs pressure/inch<sup>2</sup> for 15 minutes in an autoclave. Medium was allowed to cool up to 40-42  $\degree$ C before pouring into petriplates.

#### **2.3.3 Composition of Potato dextrose broth**



#### **2.4.1 Potato dextrose broth preparation procedure**

Similar procedure was followed for the preparation of PDA broth except addition of Agar-agar powder.

#### **2.5.1 Composition of Nutrient Agar Medium**



## **2.5.2 Procedure for preparation of NAM medium**

All the ingredients were mixed properly in 500 ml distilled water with the help of glass rod and volume was made up 1000 ml by adding required amount of distilled water. 150 ml of this solution was poured in each 250 ml capacity of conical flasks. Flasks were tightly plugged with nonabsorbent cotton plug and wrapped with silver foil and autoclaved at  $121^{\circ}$ C temperature at 15 lbs pressure/inch<sup>2</sup> for 15 minutes. Sterilized medium was allowed to cool up to 40-42 °C before pouring into Petri plates.

#### **2.6.1 Composition of Nutrient Broth**



#### **2.6.2 Nutrient broth preparation procedure:**

Similar procedure was followed for preparation of Nutrient Agar Medium except addition of Agar-agar powder.

## **2.7.1 King's 'B' medium (Pseudomonas fluorescence selective medium)**

For culturing the Pseudomonas fluorescence, King's 'B' medium was used during the investigation.

#### **2.7.2 Composition of King's 'B' medium**



#### **2.7.3 King's 'B' medium preparation procedure**

All the ingredients of King's 'B' medium were mixed properly in distilled water with the help of glass rod and volume was made 1000 ml by adding required amount of distilled water. 150 ml of this solution was poured in each 250 ml capacity of conical flasks. Flasks were tightly plugged with nonabsorbent cotton plug and wrapped with silver foil and autoclaved at 121°C temperature at 15lbs pressure/inch<sup>2</sup> for 15 minutes. Sterilized medium was allowed to cool up to 40-42 °C before pouring into Petri plates.

#### **2.7.4 King's 'B' broth preparation procedure:**

Similar procedure was followed for the preparation of King's 'B' medium except addition of Agar.

#### **2.8 Pathogenicity test of M. graminicola:**

The experiment was conducted by using rice variety PB-1121 to test the pathogenicity. Two days prior to M. graminicola nematode inoculation, nematode infected rice plants were removed from rice fields. The roots of uprooted rice plants washed under running tap water. Their roots were cut into small pieces and grinded by grinder machine. After grinding of rice roots, eggs, second stage juveniles  $(J_2)$ ,

adult males and females of nematode came out in water. The  $J_2$  stage of M. graminicola were separated by spreading the suspension onto a double layer tissue paper placed over wire gauze and then submerged into water in petri plates.

# **2.9 Field experiment**

# **2.9.1 Land Preparation**

The land was thoroughly ploughed with tractor driven disc plough followed by harrowing. Land was puddled thoroughly by ploughing and then leveling of the field. Weeds and stubbles were removed from the field. Layout of experimental field was done according to the design. The land was ready for seed sowing.

# **2.9.2 Effect of different fungal and bacterial bio-agents on the management root knot nematode of rice**

Bio control agent's viz., Trichoderma isolates-7, Trichoderma isolates-13, Paecilomyces lilacinus, Pochonia chlamydosporia, Bacillus subtilis and Pseudomonas fluorescens obtained from, Nematology Laboratory, Department of Plant Pathology, Sardar Vallabhbhai Patel University of Agriculture and Technology, Meerut, (U.P.), India. These cultures were used for the management study conducted under field condition.

# **2.9.3 Application of different bio-agents**

A field experiment was conducted to know the effect of bioagents (Trichoderma isolate S-7, Trichoderma isolate S-13, Paecilomyces lilacinus, Pochonia chlamydosporia, Bacillus subtilis and Pseudomonas fluorescens). All the bioagents on the management of root knot nematode in nursery were applied as soil application. Three replications were maintained for each treatment. Randomized Block Design (R.B.D.) was used in the field experiment. One untreated check was maintained as control for comparison.

# **3. RESULTS**

# **3.1 Effect of bio-agents against the root knot nematode in rice nursery**

#### **3.1.1. Effect on number of galls/plant:**

Data presented in **"Table 1, Figure 1 and 2** revealed that all the treatments reduced the number of galls/plant over the control. In the year 2020, minimum 11.54, 10.56 galls/plant and maximum 64.63, 63.99 % gall reduction was recorded in (T<sub>7</sub>) Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil followed by 12.58, 13.11 galls/plant and 61.44, 55.30 % gall reduction in  $(T_2)$  Trichoderma isolate S13 @ 20 g/ m<sup>2</sup> of soil. Whereas, maximum 21.38, 19.00 galls/plant and minimum 34.47, 35.21 % gall reduction was recorded in (T<sub>5</sub>) Pseudomonas fluorescens @ 20 g/ m<sup>2</sup> of soil in case of control 32.63, 29.33 galls/plant was recorded at 30 and 60 day after sowing, respectively.

Similarly, in the year 2021, minimum 10.54, 14.78 galls/plant and maximum 72.38, 59.17 % gall reduction was observed in (T<sub>7</sub>) Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil followed by, 13.21, 15.22 galls/plant and 65.39, 60.06 % gall reduction in (T<sub>2</sub>) Trichoderma isolate S13 @ 20 g/ m<sup>2</sup> of soil. Whereas maximum 15.17, 19.22 galls/plant and minimum 60.25, 49.42 % gall reduction was recorded in (T5) Pseudomonas fluorescens @ 20 g/  $m^2$  of soil which was least effective among all the treatments as compare to untreated control 38.17, 38.11 galls/plant were recorded at 30 and 60 day of seed sowing, respectively.

# **3.1.2 Effect on root length:**

Data present in **Table 2 & 3** and **Figure 3, 4, 5 & 6** revealed that all the treatments were effective increasing the root length over the untreated control. In the year 2020, maximum root length 6.96, 9.25 cm was recorded in treatment  $(T_2)$  soil application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil followed by 6.63, 8.45 cm in (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil at 30 and 60 days after sowing, respectively. Whereas, minimum 5.52 cm in  $(T_5)$  Pseudomonas fluorescens @ 20 g/m<sup>2</sup> of soil and 6.75 cm in (T<sub>4</sub>) Bacillus subtilis @ 20 g/m<sup>2</sup> of soil as compare to control 5.21, 6.50 cm root length was recorded at 30 and 60 days after sowing, respectively.

Similarly, in the year 2021, maximum root length 7.50, 9.83 cm was recorded in treatment  $(T_2)$  soil application of Trichoderma isolates S-13 @ 20  $g/m^2$  of soil followed by 7.29, 9.00 cm (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5  $g/m^2$  of soil at 30 and 60 days after sowing, respectively. Whereas, minimum 6.17, 7.52 cm in ( $T_5$ ) Pseudomonas fluorescens @ 20 g/ m<sup>2</sup> of soil as compare to control 6.02, 7.33 cm root length was recorded at 30 and 60 days after sowing, respectively.

#### **3.1.3 Effect on shoot length:**

Data present in the **Table 2 & 3** and **Figure 5 & 6** indicate that all the treatments were effectively increasing the shoot length over the untreated control. In year 2020, maximum shoot length 33.94, 62.66 cm was recorded in treatment  $(T_2)$  soil application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil followed by 33.78, 60.55 cm (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil at 30 and 60 days after sowing, respectively. Whereas, minimum 31.06, 53.22 cm in  $(T_5)$  Pseudomonas fluorescens  $\omega$  20 g/ m<sup>2</sup> of soil as compare to control 29.33, 50.44 cm shoot length was recorded at 30 and 60 days after sowing, respectively.

Similarly, in the year 2021, maximum shoot length 38.89, 68.89 cm was recorded in treatment ( $T_2$ ) soil application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil followed by 37.33, 67.89 cm in (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5  $g/m^2$  of soil at 30 and 60 days after sowing, respectively. Whereas, minimum 36.00, 64.78 cm in (T<sub>5</sub>) Pseudomonas fluorescens @ 20 g/ m<sup>2</sup> of soil as compared to control 33.67, 66.11 cm shoot length was recorded at 30 and 60 days after sowing, respectively.

#### **3.1.4 Effect on fresh root weight**

Result **Table 2 & 3** and **Figure 7 & 8** revealed that all the treatments used were increasing the fresh root weight over the control. In year 2020, maximum fresh root weight 0.76, 1.85 g/plant was recorded in treatment (T<sub>2</sub>) soil application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil followed by 0.56, 1.60 g/plant in (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil at 30 and 60 days after sowing, respectively. Whereas, minimum 0.40 g/plant in  $(T_5)$  Pseudomonas fluorescens @ 20 g/ m<sup>2</sup> of soil and 1.00 g/plant in (T<sub>4</sub>) Bacillus subtilis @ 20 g/ m<sup>2</sup> of soil as compare to control 0.38, 0.95 g/plant fresh root weight was recorded at 30 and 60 days after sowing, respectively.

Similarly, in the year 2021, maximum fresh root weight 0.88, 2.03 g/plant was recorded in treatment (T<sub>2</sub>) soil application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> followed by 0.74, 1.88 g/plant in (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5  $g/m^2$  of soil at 30 and 60 days after sowing, respectively. Whereas, minimum 0.45 g/plant in (T<sub>5</sub>) soil application of Pseudomonas fluorescens @ 20 g/ m<sup>2</sup> of soil and 1.26 g/plant in (T<sub>6</sub>) Paecilomyces lilacinus @ 20 g/ m<sup>2</sup> of soil as compare to control 0.42, 1.13 g/plant fresh root weight was recorded at 30 and 60 days after sowing, respectively.

# **3.1.5. Effect on dry root weight:**

Result revealed from the **Table 2 & 3** and **Figure 9 & 10** that all the treatments used were increasing the dry root weight over the control. In year 2020, maximum dry root weight 0.25, 0.37 g/plant was recorded in treatment (T<sub>2</sub>) soil application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil followed by 0.18, 0.32 g/plant in (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil at 30 and 60 days after sowing, respectively. Whereas, minimum 0.13 g/plant in the treatments i.e.  $(T_4)$  Bacillus subtilis @ 20 g/ m<sup>2</sup> of soil, (T<sub>5</sub>) Pseudomonas fluorescens @ 20 g/ m<sup>2</sup> of soil and (T<sub>6</sub>) Paecilomyces lilacinus @ 20 g/  $m^2$  of soil and 0.22 g/plant in (T<sub>4</sub>) Bacillus subtilis @ 20 g/ m<sup>2</sup> of soil as compare to control 0.12, 0.20 g/plant dry root weight was recorded at 30 and 60 days after sowing, respectively.

Similarly, in the year 2021, maximum dry root weight 0.29, 0.40 g/plant was recorded in treatment  $(T_2)$  soil application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil followed by 0.26, 0.37 g/plant in  $(T<sub>7</sub>)$  soil application of Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil at 30 and 60 days after sowing, respectively. Whereas, minimum 0.16 g/plant and 0.25 g/plant in both (T<sub>4</sub>) Bacillus subtilis @ 20 g/ m<sup>2</sup> of soil & (T<sub>5</sub>) Pseudomonas fluorescens @ 20 g/  $m^2$  of soil as compare to control 0.14, 0.23 g/plant dry root weight was recorded at 30 and 60 days after sowing, respectively.

# **3.1.6 Effect on number of tillers:**

Results present in the **Table 2 & 3** and **Figure 11 & 12** indicate that all the treatments were effectively increasing the number of tillers over the control. In the year 2020, maximum number of tiller 0.22, 1.89 were recorded in treatment (T<sub>2</sub>) soil application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil followed by 0.11 in (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil and (T<sub>3</sub>) soil application of Pochonia chlamydosporia @ 20 g/m<sup>2</sup> of soil and 1.78 in (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5  $g/m<sup>2</sup>$  of soil at 30 and 60 days after sowing, respectively. Whereas, no tillering was recorded in (T<sub>1</sub>) soil application of Trichoderma isolates S-7 @ 20 g/m<sup>2</sup> of soil, (T<sub>4</sub>) Bacillus subtilis @ 20 g/ m<sup>2</sup> of soil, (T<sub>5</sub>) Pseudomonas fluorescens @ 20 g/ m<sup>2</sup> of soil and (T<sub>6</sub>) Paecilomyces lilacinus @ 20 g/ m<sup>2</sup> of soil, respectively and 1.33 in (T<sub>4</sub>) Bacillus subtilis @ 20 g/ m<sup>2</sup> of soil and (T<sub>5</sub>) Pseudomonas fluorescens @ 20 g/  $m^2$  of soil as compare to control 0.00, 1.33 number of tillers were recorded at 30 and 60 days after sowing, respectively.

Similarly, in the year 2021, maximum number of tillers/plant 0.33, 2.48 were recorded in treatment (T<sub>2</sub>) soil application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil followed by 0.22 tillers/plant in (T<sub>7</sub>) soil application of Phorate 10-G  $\qquad \varpi$  2.5 g/m<sup>2</sup> of soil and (T<sub>3</sub>) soil application of Pochonia chlamydosporia @ 20 g/m<sup>2</sup> of soil and 2.33 in (T<sub>7</sub>) soil application of Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil at 30 and 60 days after sowing, respectively. Whereas, no tillering was recorded in treatments i.e. (T4) Bacillus subtilis @ 20 g/ m<sup>2</sup> of soil, (T<sub>5</sub>) Pseudomonas fluorescens @ 20 g/ m<sup>2</sup> of soil, and (T<sub>6</sub>) Paeciliomyces lilacinus @ 20 g/ m<sup>2</sup> of soil and 2.11 tiller/plant in (T<sub>4</sub>) Bacillus subtilis @ 20 g/ m<sup>2</sup> of soil and (T<sub>5</sub>) Pseudomonas fluorescens @ 20 g/ m<sup>2</sup> of soil as compare to control 0.00, 2.00 number of tiller were recorded at 30 and 60 days after sowing, respectively.

#### **4. DISCUSSION**

The result revealed that all the treatments reduced the number of galls/plant over the control in both the year 2020 & 2021. In the year 2020, minimum 11.54, 10.56 galls/plant and maximum 64.63, 63.99 % gall reduction was recorded in (T<sub>7</sub>) Phorate 10-G @ 2.5 g/m<sup>2</sup> of soil. Whereas, maximum 21.38, 19.00 galls/plant and minimum 34.47, 35.21% gall reduction was recorded in  $(T_5)$  Pseudomonas fluorescens@ 20 g/  $m^2$  of soil in case of control 32.63, 29.33 galls/plant were recorded at 30 and 60 day after sowing, respectively. Similarly, in the year 2021, minimum 10.54, 14.78 galls/plant and

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maximum 72.38, 59.17 % gall reduction was observed in ( $T_7$ ) Phorate 10G @ 2.5 g/m<sup>2</sup> of soil. Whereas, maximum 15.17, 19.22 galls/plant and minimum 60.25, 49.42 % gall reduction was recorded in  $(T_5)$ Pseudomonas fluorescens@ 20 g/  $m^2$  of soil which was least effective among all the treatments as compare to untreated control 38.17, 38.11 galls/plant were recorded at 30 and 60 day of seed sowing, respectively. **Mukesh et al. (2016)** evaluated the effectiveness of 20 isolates of Trichoderma spp. against root knot nematode (M. graminicola) of rice in a pot experiment. All the isolates of Trichoderma were found effective. Minimum galls per plant (2.67) were observed in S12 and maximum in S7 and S9 (8.60).

In case of growth parameters, all the treatments were most effective in increasing the growth parameter over the untreated control. Application of Trichoderma isolates S-13 @ 20 g/m<sup>2</sup> of soil was found most effective to increase the root length, shoot length, fresh root weight, dry root weight, increasing the number of tillers comparison to all the tested bio-agents in both the year 2020 and 2021. **Anitha and Rajendran (2005)** revealed that the integration of P. fluorescens (2.5 kg/ha), neem cake (1 ton/ha) and carbofuran (1 kg a.i./ha) was highly effective in improving plant growth and yield in the nursery as well as in main field. **Narasimhamurthy et al. 2017** revealed that the combination of Pseudomonas fluorescens @ 20g/m² + carbofuran 3G recorded highest plant height (83.26 cm), root length (20.60 cm), maximum grain yield (44.1 q/ha) and least nematode population (132.67/200 g soil) with reduction of 79.34 % nematode population followed by T. viride @  $20g/m^2$  + carbofuran with plant height (81.67cm), root length (18.50 cm), grain yield (43.6 q/ha) with least nematode population (198.00/200 g soil) with reduction of 69.17% nematode population. Similarly, **Singh and Mahanta (2013)** also reported that application of bio-agents with vermicompost increase the growth parameters and significant reduction of final population of nematode in soil. Maximum growth parameters were recorded in (T. harzianum @ 2.5 kg/ha + G. fasciculatum @ 300 spores/m<sup>2</sup> + carbosulfan ST @ 1.5% w/w + vermicompost 1.5 t/ha) followed by the treatment with integration of T. harzianumand G. fasciculatum.

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# **Table 1 Effect of different bio-agents on number of galls/plant in rice nursery**

#### **Table 2 Effect of different bio-agents on plant growth parameters at 30 DAS**



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#### **Table 3 Effect of different bio-agents on plant growth parameters at 60 DAS**





![](_page_10_Figure_2.jpeg)

**Figure 2 Effect of different bio-agents on number of galls/plant in rice nursery at 60 DAS**

**Figure 3 Effect of different bio-agents on root length at 30 DAS**

![](_page_10_Figure_5.jpeg)

![](_page_11_Figure_2.jpeg)

**Figure 4 Effect of different bio-agents on root length at 60 DAS**

![](_page_11_Figure_4.jpeg)

![](_page_11_Figure_5.jpeg)

![](_page_12_Figure_2.jpeg)

**Figure 6 Effect of different bio-agents on shoot length at 60 DAS**

**Figure 7 Effect of different bio-agents on fresh root weight at 30 DAS**

![](_page_12_Figure_5.jpeg)

![](_page_13_Figure_2.jpeg)

**Figure 8 Effect of different bio-agents on fresh root weight at 60 DAS**

![](_page_13_Figure_4.jpeg)

![](_page_13_Figure_5.jpeg)

![](_page_14_Figure_2.jpeg)

**Figure 10 Effect of different bio-agents on fresh root weight at 60 DAS**

**Figure 11 Effect of different bio-agents on number of tillers/plant at 30 DAS**

![](_page_14_Figure_5.jpeg)

![](_page_15_Figure_2.jpeg)

**Figure 12 Effect of different bio-agents on number of tillers/plant at 60 DAS**