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In vitro Evaluation of Botanicals, Bio-agents and Fungicides against *Rhizoctonia solani* Causing Web Blight Disease of Mungbean

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Authors' contributions
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This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Mungbean [*Vigna radiata* (L.) Wilczek] is the important source of proteins, minerals, and vitamins of the predominantly vegetarian Indian diet. It belongs to the family Leguminaceae. Web blight caused by *Rhizoctonia solani* (Kuhn) is one of the most important fungal diseases which come every year

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with different intensity and causes huge losses in mungbean yield. The present investigations were carried out in the laboratory, Department of Plant Pathology Acharya Narendra Deva University of Agriculture and Technology, Kumarganj, Ayodhya to test the efficacy of different treatments *viz*., Neem, Garlic, Tulsi, onion, Ginger, *T. asperellum*, *T. harzianum,* Propiconazole and Hexaconazole against *Rhizoctonia solani* Kühn under *in vitro condition.* Botanicals and Fungicides were tested through Poisoned food technique and Bio-agents were tested through dual culture technique. Pathogen was isolated from diseased mungbean plant and further tested against different treatments. Radial growth and percent inhibition were recorded. Minimum radial growth and maximum percent inhibition were recorded in Propiconazole 1.45 mm, followed by Hexaconazole (3.65 mm), Garlic (10.18 mm), Ginger (11.43 mm), Neem (12.90 mm), Onion (15.42 mm), Tulsi (17.63 mm), *T. asperellum* (18.58 mm), *T. harzianum* (23.14 mm) as compared to Control (45.17 mm) at 24 hours of incubation. Similar trends were found at 36 and 48 hours intervals.

Keywords: Mungbean; neem; garlic; tulsi; onion; ginger; T asperellum; T harzianum; propiconazole; hexaconazole; Rhizoctonia solani.

1. INTRODUCTION

"Mungbean [*Vigna radiata* (L.) Wilczek] is the important source of protein, vitamins and minerals. It belongs to the family Leguminaceae. Among the pulses mungbean also called as green gram or golden gram. It is mainly grown in Rajasthan, Maharashtra, Karnataka, Andhra Pradesh, Orissa, Bihar, Tamil Nadu, Madhya Pradesh, and Uttar Pradesh. In Uttar Pradesh, it is cultivated on 93000 ha, with a production of 9480 tonnes. The productivity of mung bean in India and the U.P. is 567 kg/ha and 536 kg/ha, respectively, which is very low compared to the genetic potential of 1500–2000 kg/ha" [1] "In 1924, web blight was reported for the first time on mung bean from the Philippines" [2] While in India, Dwivedi and Saksena [3] first reported it on mung beans from Kanpur, Uttar Pradesh. Additionally, it has also been reported from Assam [4], Punjab [5], Madhya Pradesh [6], Bihar, Rajasthan, Haryana, Himachal Pradesh and Jammu and Kashmir [7]. Anonymous [8,9]. Web blight caused by *Rhizoctonia solani* (Kuhn) is one of the most important fungal disease which appear every year in varying intensity and causes heavy reduction in yield. Gupta et al. [10] reported "the losses in grain yield is more when the plants get infected earlier i.e. after 25 days after sowing (DAS) than 35 and 40 DAS. The losses in yield and lost weight were 33.40 to 37.80 per cent and 23.12 to 28.60 per cent respectively". Though, the web blight could be managed by the use of fungicide but due to the emergence of several problems like environmental pollution, residual effect, its use should be discouraged. Many botanicals and bio-agent have been found effective against *Rhizoctonia solani* in different crops, therefore keeping in view the importance of the crop and seriousness of diseases present research work carried.

2. MATERIALS AND METHODS

The present investigations were carried out in the laboratory, Department of Plant Pathology A.N.D.U.A. & T., Kumarganj, Ayodhya (Year 2022-23) to test the efficacy of different treatments against *Rhizoctonia solani* Kühn under *in vitro.* Pathogen was isolated from diseased mungbean plant parts (Leaf and Stem) and further tested against different treatments. In order of find out the efficacy of various plants extracts against *R. solani*, five plants extract viz., Neem (leaf), Garlic (bulb), Tulsi (leaf), onion (bulb) and Ginger (rhizome) were used. Fresh leaves, bulbs and rhizomes were collected and washed thoroughly in clean water. Equal amount of washed plant material was grinded in mortar and pestle by adding same amount of sterilized water (1:1 w/v) and boiled at 80 ^{0} C for 10 minutes in hot water bath . The extract was filtered by double layer muslin cloth followed by sterilized Whatman No.1 Filter paper. By adding the 20 ml of sterilized PDA medium and 2 ml Botanical extract, the concentrations of 10.0 percent were created. Two fungicides were tested *in vitro* against *R. solani* i.e., Propiconazole (20 ppm) and Hexaconazole (20 ppm). The flasks were thoroughly shaken to ensure an even mix of the extract and fungicide under aseptic conditions. Twenty ml of sterilized melted PDA was aseptically poured in sterilized Petri dishes and allowed to solidify. The efficacy of *T. asperellum* and *T. harzianum* against *Rhizoctonia solani* were assessed by using dual culture technique by measuring the radial growth of *R. solani* as well as that of *Trichoderma spp* [11]. Culture of Bio-agent were collected from

Table 1. List of botanicals and their scientific names

S. No.	Common Name	Scientific Name	Plant part used
1.	Neem	*Azadirachta indica*	Leaves
2.	Garlic	*Allium sativum*	Bulb
3.	Tulsi	*Ocimum snactum*	Leaves
4.	Onion	*Allium cepa*	Bulb
5.	Ginger	*Zingiber officinale*	Rhizome

laboratory Department of plant pathology A.N.D.U.A. & T., Kumarganj, Ayodhya. Three-day-old *R. solani* culture discs were cut into five mm pieces with a sterilized cork borer and positioned in the centre of Petri dishes with plant extracts and fungicides added. Five mm disc of each antagonist and *R. solani* cut with the help of sterilized cork borer from the age of three days old culture and were placed in Petri dishes having solidified PDA in such a manner that they lie opposite to each other 60 mm apart. "Control (Check) Petri dishes were inoculated only with *R. solani* bits. Each treatment and control was repeated four times to make four replications. These Petri dishes were kept in BOD incubator at 28^0 C. The observations on radial growth were made at 24, 36, and 48 hours of incubation in Petri dishes amended with different treatments as well as in control. Per cent growth inhibition was calculated by using formula" [12]. The conclusion was arrived at after statistical analysis of the data. The Completely Randomized Design (CRD) method recommended by Goon *et al.* was used to conduct the statistical analysis of laboratory experiments (1931). The variance ratio test at the 5% level of probability was used to determine the significance of treatment differences. The observation of per cent inhibition of mycelial growth, were transformed in to "Arc sin Transformation" = $\sin^{-1}\sqrt{p}/100$ used for statistical analysis.

$$I = \frac{C - T}{C} \times 100$$

Where,

I = Per cent inhibition of fungal growth.
C = Radial growth of control.
T = Radial growth in treated Petri dish.

3. RESULTS AND DISCUSSION

- **At 24 Hours of Incubation**

Minimum radial growth was obtained in Propiconazole (1.45 mm) followed by Hexaconazole (3.65 mm), Garlic (10.18 mm), Ginger (11.43 mm), Neem (12.90 mm), Onion (15.42 mm), Tulsi (17.63 mm), *T. asperellum* (18.58 mm), *T. harzianum* (23.14 mm) as compared to Control (45.17 mm). Propiconazole, Hexaconazole, Garlic, Ginger, Neem, Onion and *T. harzianum* were significantly different to each other while Tulsi and *T. asperellum* were found at par to each other. (Table 2, Fig. 1).

- **At 36 Hours of Incubation**

Minimum radial growth was obtained in Propiconazole (1.85 mm) followed by Hexaconazole (4.85 mm), Garlic (14.04 mm), Ginger (16.07 mm), Neem (18.63 mm), Onion (20.00 mm), Tulsi (22.00 mm), *T. asperellum* (23.24 mm), *T. harzianum* (31.58 mm) as compared to Control (71.56 mm). Propiconazole, Hexaconazole, *T. harzianum* were significantly different to each other while Neem, Onion Tulsi and *T. asperellum,* Garlic and Ginger, were found at par to each other (Table 2, Fig. 1).

- **At 48 Hours of Incubation**

Minimum radial growth was obtained in Propiconazole (2.16 mm) followed by Hexaconazole (5.93 mm), Garlic (15.49 mm), Ginger (18.00 mm), Neem (19.95 mm), Onion (22.33 mm), Tulsi (24.66 mm), *T. asperellum* (25.60 mm), *T. harzianum* (39.55 mm) as compared to Control (85.42 mm). Propiconazole, Hexaconazole, Garlic, Ginger, Neem, Onion and *T. harzianum* were significantly different to each other while Tulsi and *T. asperellum* were found at par to each other (Table 2, Fig. 1).

- **At 24 hours of incubation**

Results clearly indicated that maximum percent inhibition was found in Propiconazole (96.80%) followed by Hexaconazole (91.92%), Garlic (77.45 %), Ginger (74.67 %), Neem (71.43 %), Onion (65.79 %), Tulsi (60.95 %), *T. asperellum* (58.87 %) and *T. harzianum* (48.74%) as compared to control (0.00%). Propiconazole, Hexaconazole, Garlic, Ginger, Neem, Onion and *T. harzianum* were significantly different to each other while Tulsi and *T. asperellum* were found at par to each other (Table 3, Fig. 2).

Table 2. Effect of botanicals, bio-agents and fungicides against *R. solani* on mycelial growth at 24, 36 and 48 hours

Treatment	Mycelial Growth (mm) Duration in Hours		
	24 Hours	36 Hours	48 Hours
Neem	12.90	18.63	19.95
Garlic	10.18	14.04	15.49
Tulsi	17.63	22.00	24.66
Onion	15.42	20.00	22.33
Ginger	11.43	16.07	18.00
T. asperellum	18.58	23.24	25.60
T. harzianum	23.14	31.58	39.55
Propiconazole	1.45	1.85	2.16
Hexaconazole	3.65	4.85	5.93
Control	45.17	71.86	85.42
CD at5%	1.18	2.27	1.30
SE(m)	0.39	0.76	0.44

Table 3. Effect of botanicals, bio-agents and fungicides against *R. solani* on mycelial growth at 24, 36 and 48 hours

Treatment	Percent inhibition Duration in Hours		
	24 Hours	36 Hours	48 Hours
Neem	71.43(57.67)	74.09(59.40)	76.52(61.07)
Garlic	77.45(61.62)	80.42(63.76)	81.89(64.79)
Tulsi	60.95(51.31)	69.39(56.40)	71.08(57.48)
Onion	65.79(54.20)	72.20(58.20)	73.81(59.23)
Ginger	74.67(59.77)	77.63(61.75)	78.98(62.64)
T. asperellum	58.87(50.08)	67.65(55.31)	70.82(57.28)
T. harzianum	48.74 (44.26)	56.04(48.51)	54.30(47.45)
Propiconazole	96.80(79.67)	97.42(80.74)	97.47(80.81)
Hexaconazole	91.92 (73.46)	93.25(74.91)	93.05(74.70)
Control	0.00(0.00)	0.00(0.00)	0.00(0.00)
CD at 5%	2.34	2.88	1.47
SE(m)	0.78	0.96	0.49

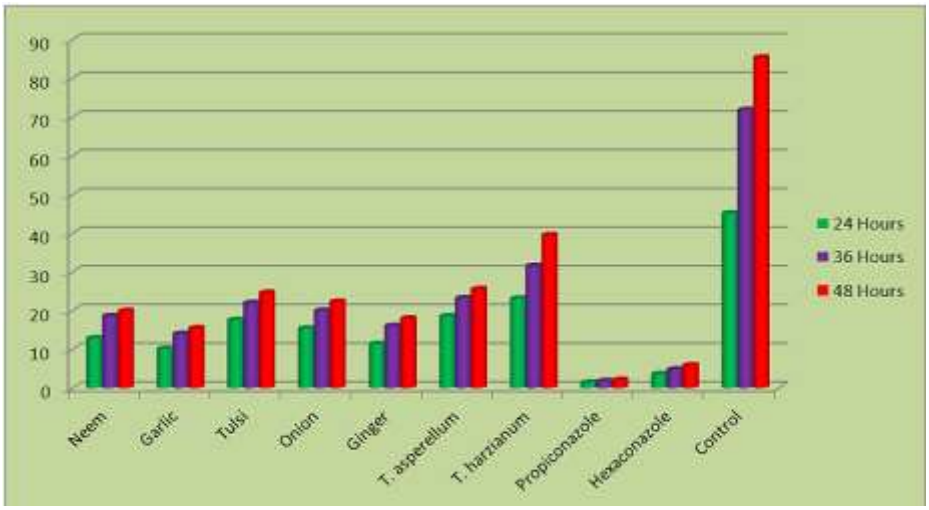


Fig. 1. Effect of botanicals, bio-agents and fungicides against *R. solani* on mycelial growth at 24, 36 and 48 hours

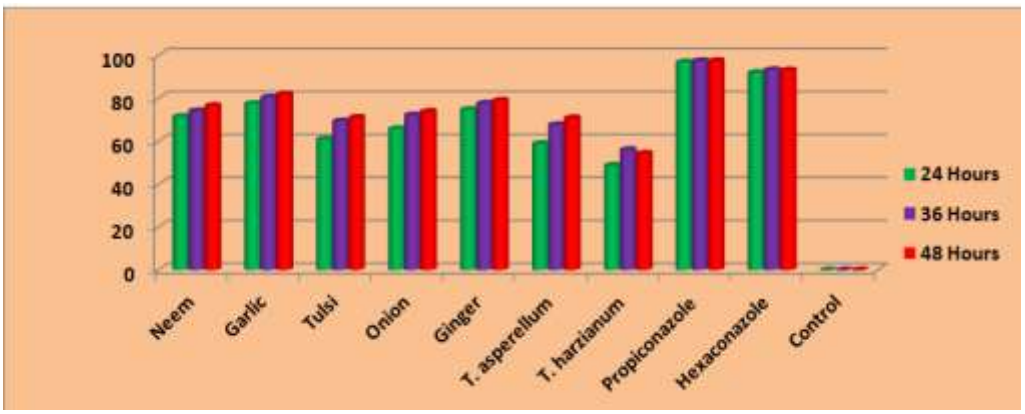


Fig. 2. Effect of botanicals, bio-agents and fungicides against *R. solani* on percent inhibition at 24, 36 and 48 hours

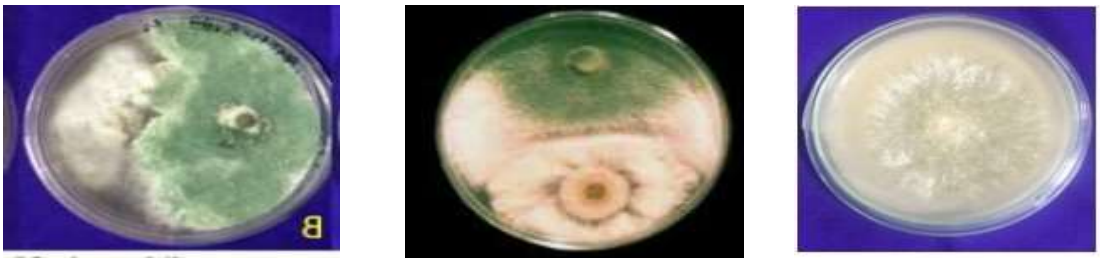
Plate 1. Response of botanicals against *R. solani* on mycelial growth

- **At 36 hours of incubation**

Maximum percent inhibition was recorded in Propiconazole (97.41%) followed by Hexaconazole (93.25%), Garlic (80.42 %), Ginger (77.63 %), Neem (74.09 %), Onion (72.20 %), Tulsi (69.39 %), *T. asperellum* (67.65 %) and *T. harzianum* (56.04 %) as compared to control (0.00%). Propiconazole, Hexaconazole and *T. harzianum* were found statistically differed to each other while Tulsi, *T. asperellum,* Onion, Neem and Ginger and Garlic were found at par to each other (Table 3, Fig. 2).

- **At 36 hours of incubation**

Maximum percent inhibition was recorded in Propiconazole (97.47%) followed by



Trichoderma asperellum ***Trichoderma harzianum*** **Control**

Plate 2. Response of bio-agents against *R. solani* on mycelial growth

Propiconazole **Hexaconazole** **Control**

Plate 3. Response of fungicides against *R. solani* on mycelial growth

Hexaconazole (93.25%), Garlic (81.89 %), Ginger (78.98 %), Neem (76.52 %), Onion (73.81 %), Tulsi (71.08 %), *T. asperellum* (70.82 %) and *T. harzianum* (54.3 %) as compared to control (0.00%). Propiconazole, Hexaconazole, Garlic, Ginger, Neem Onion, and *T. harzianum* were found statistically differed to each other while Tulsi and, *T. asperellum* were found at par to each other (Table 3, Fig. 2).

Similar findings of botanicals, and fungicides on mycelial growth and percent inhibition were reported by Meena et al [13] and Vipin et al. [7], Findings of botanicals and bio- agents were supported by Singh et al. [14], Shinde and Patel [15] and Dubey et al. [16,17].

4. CONCLUSION

Our study very well demonstrated that, Propiconazole demonstrated the highest efficacy against *Rhizoctonia solani*, followed by Hexaconazole, while garlic and ginger extracts exhibited notable inhibitory effects. *T. asperellum* and *T. harzianum* bio-agents also displayed significant antifungal activity. These findings support the potential integration of botanicals and bio-agents for web blight disease in mungbean. Many environmental concerns linked to chemical

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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