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Potential of chemical fungicides against *Rhizoctonia solani* Kuhn. inciting web blight of mungbean [*Vigna radiata* (L.) Wilczek]

Manpreet Kaur, Prince Kumar Gupta and KPS Kushwaha

Abstract
Among the fungal diseases, Web blight, caused by *Rhizoctonia solani* Kuhn (Teleomorph: *Thanatephorus cucumeris* Donk), a ubiquitous opportunistic pathogen, is an important fungal disease of Mungbean. The potential losses due to web blight alone in India have been up to 20-60 percent. In present study an attempt was made in Pulse Pathology Laboratory, Department of Plant Pathology, GBPUAT, Pantnagar, to investigate the efficacy of five different fungicides (Hexaconazole, Propiconazole, Pyraclostrobin + Metiram, Carboxin + Thiram, and Carbendazim) against the mycelial growth of *R. solani* at 15, 25, 50, 100, 150 ppm concentration using poison food techniques. Significantly, all the tested fungicides inhibited the mycelial growth of the pathogen i.e., fungi but hexaconazole was found highly effective at lower concentration (15 ppm) inhibiting 100 percent mycelial growth. However, Propiconazole inhibit the growth by 100% at 100 ppm concentration. Carboxim +Thiram and Carbendazim were found least effective even at higher concentrations (150 ppm) over the control. The inhibitory effect of the tested fungicides was increased with increasing fungicide concentrations.

Keywords: Web blight, *Rhizoctonia solani*, mungbean, fungicide

Introduction
Vigna radiata (L.) Wilczek is the most economically significant pulse crops, belongs to the family Fabaceae commonly known as Mungbean and it is also known as "Golden gram" because of its nutritional richness.

In India, Mungbean is grown in almost all the states due to its triple use *i.e.*, food, fodder, and for improving soil fertility. Mungbean seeds have a protein content of about 20-24% and thus act as a good source of protein (Keatinge *et al*., 2011) [10]. The protein is high in essential amino acids such as leucine, lysine, and phenylalanine/tyrosine, valine, isoleucine and histidine which is lacking in cereals grains (Mubarak, 2005) [14]. Mungbeans also contain (56%) carbohydrates (Anisha and Prema, 2008) [1], (16.3%) Dietary fibre (Khatoon and Prakash, 2006; Lin and Lai, 2006) [11, 13], (1.3%) fat (Sathe, 1996) [20], phosphorus (124 mg/100 g), calcium (326 mg/100 g), minerals (3.5%), Iron (7.3 mg/100 g) and moisture of about 10% (Grewal and Jood, 2006) [6]. Mungbean can be used as green fodder or green manure once the pods are harvested. The extensive root structure of the Mungbean plant holds soil particles, preventing soil erosion, and being a legume crop can improve soil fertility by fixing atmospheric nitrogen in the soil (Graham and Vance, 2003) [5]. Intercropping as well as crop rotation of the Mungbean with the other crop enhances the production of successive cereal crops.

It is a fast-growing short-term pulse crop grown in summer and autumn with the least input requirement and performs well under heat and drought conditions. Mungbean is grown primarily in tropical and subtropical areas like India, China, Bangladesh, Myanmar, Indonesia, Thailand, and some parts of central and eastern Africa, the United States of America, and Australia (Westphal, 1974) [24]. Mungbean is cultivated on more than 6 million hectares of land worldwide (about 8.5% of global pulse cultivation area) and global annual production is about 3 million tons (5% of global pulse production). India is the largest producer and consumer of Mungbean (Nair *et al*., 2014) [4] holding about 35% area and production of about 25% across the globe with 2.17 million tonnes production followed by China with 0.98 million tonnes and Myanmar with 0.400 million tonnes production. Mungbean production in India has increased in recent years with an estimate of about 40.43 lakh hectares area, producing 19.48 lakh tonnes

of grains with a productivity of 483 kg per hectare. The largest Mungbean producing state are Rajasthan (15.16 lakh ha), Madhya Pradesh (34 lakh ha), Maharashtra (4.08 lakh ha), Bihar (1.71 lakh ha), Andhra Pradesh (1.55 lakh ha), Tamil Nadu (1.97 lakh ha) and Karnataka (3.70 lakh ha) which is about 80% of the total area. Among the Mungbean growing states, the highest production of about 7.66 lakh tonnes in Rajasthan followed by Madhya Pradesh (2.19 lakh tonnes) and Maharashtra (1.55 lakh tonnes) and the state with the highest productivity is Andhra Pradesh (670 kg/ha) followed by Bihar (652 kg/ha) and Madhya Pradesh (621 kg/ha).

With increasing population growth in India major constraints in the availability of Mungbean and its production is low soil fertility, abiotic stress, unavailability of resistant cultivars, insect pests, and diseases which are caused by different plant pathogenic microorganism like fungus, bacteria, virus, phytoplasma, nematodes, etc which causes significant yield loss across the globe. Among the biotic stress, major diseases are Mungbean Yellow Mosaic (MYMV), Cercospora leaf spot (*Cercospora canesens*), Bacterial leaf spot (*Xanthomonas phaseoli*), Powdery mildew (*Erysiphe polygoni*), Rust (*Uromyces phaseoli*), Web blight (*Rhizoctonia solani*), Dry rot (*Macrophomina phaseolina*) and Anthracnose (*Glomerella lindemuthianum*) causing yield losses upto 40-60%. Among all the pathogenic microorganism's fungi can infect the plant at various stages of development, including germination, the emergence of seedlings, and vegetative and reproductive stage.

Among the major disease, Web blight caused by *Rhizoctonia solani* is one of the major foliar disease-causing yield losses ranging from 20-60 percent across the globe. In India, the disease causeda reduction of 33 to 40 percent in grain yield and 28.6 percent in 1000 grain weight at a different level of disease severity (Gupta *et al.*, 2010) [7]. *Rhizoctonia solani* causes aerial symptoms on the lower portion of the plant that frequently occurs during the late vegetative growth stage. It is also responsible for pre- and post-emergence rot of Mungbeans and causing maximum mortality of seedlings. Initially, the symptoms on the leaf start as water-soaked, greyish-green lesions which turn tan to brownish at maturity. The pathogen may infect leaves, pods, and stems near the basal area of the plant. Due to its devastating nature, there is an urgent need to focus on remedies for its management. Hence present study investigates the *in-vitro* efficacy of different fungicides against *Rhizoctonia solani* causing web blight of Mungbean.

Material and Method

The standard laboratory techniques were used for the preparation of PDA media, cleaning and sterilization of glassware, isolation of fungus, inoculation, and maintenance of fungal culture, with modification whenever necessary.

Collection of disease specimen

Collection of the Mungbean leaves which showed the characteristic symptoms of the disease was collected from the Pulse Pathology Block of N. E. Borlaug Crop Research Centre, Pantnagar, Uttarakhand. The lesions, showing the initial and distinct characteristic symptoms, were selected for isolation of the pathogen.

Isolation of *Rhizoctonia solani*

For isolation, diseased leaves were washed first with fresh water and then sterile water to remove the extraneous soil and surface contaminants. These were cut into small bits of 2-3 mm dimension by tissue segmentation method (Bonmam *et al.*, 1987) [2]. These bits were surface sterilized by dipping in 0.1 percent mercuric chloride solution for 30 seconds followed by washing in 2 changes of sterilized water, then placed aseptically on PDA slants with the help of inoculating needle under aseptic condition. These were incubated at 28±2 °C. After 4 days of incubation, the fungus was transferred to sterilize Petriplates containing PDA medium and incubated in the same manner (Hemalatha *et al.*, 2018) [9]. After 6 days of incubation, a bit of hyphal growth from growing tips was transferred aseptically to fresh PDA slants. The fungus was purified by employing the single hyphal tip method (Singh, 1988) [21]. The fungus was identified following a mycological description (Ou, 1985) [18]. However, the culture is maintained by periodic transfer on PDA slants for further studies.

Evaluation of fungicides against *Rhizoctonia solani in vitro*

Ten fungicides were evaluated *in vitro* against *Rhizoctonia solani* using the poison food technique (Nene and Thapliyal, 1982) [16]. Their trade name, chemical name, formulation, are illustrated in (Table.1). A hundred ml sterilized Potato dextrose agar medium was fortified with 1.35, 2.5, 5, 10, and 15 mg of ten fungicides separately to get 15 ppm, 25 ppm, 50 ppm, 100 ppm, and 150 ppm concentrations. 20 ml of media of each concentration were gently poured intoa sterilized Petri plate and allowed to solidify. After solidification, a 5 mm disc of the seven-day-old culture of *Rhizoctonia solani* was cut with the help of cork borer and then placed in the centre of the Petri plates and incubated in a B.O.D incubator at 28±2 °C. There will be three replications of each treatment along with a control plate (without *Rhizoctonia solani* was measured with the help of a ruler scale after 48 hrs and subsequent observation was recorded at 48 hrs of intervals and continued till full growth of the pathogen in control (90 mm) i.e., 120 hrs. The percent inhibition of fungus over control was calculated by using the following formula as given by Vincent (1927) [22].

$$\text{Percent Growth inhibition} = C - T / T \times 100$$

Where,

I = Percent growth inhibition =Colony diameter in control Petri plate;

T = Colony diameter in the treated Petri plate. The percent inhibition data were analyzed statistically using a completely randomized design (C.R.D).

Table 1: The list of fungicides tested against mycelial radial growth of fungal phytopathogens.

S. No	Trade name	Chemical composition	Formulations	Manufacturing Company
1.	Contaf	Hexaconazole 5%	SC	Hyderabad Chemical Supplies Ltd., Hyderabad
2.	Tilt	Propiconazole 25%	EC	Hyderabad Chemical Supplies Ltd., Hyderabad
3.	Vitavex	Carboxin (37.5%) + Thiram (37.5%)	DS	Dhanuka Agritech Limited,
4.	Commando	Carbendazim (50%)	WP	Ram Shri Chemical, Mumbai
5.	Cabrio Top	Pyraclostrobin (55%) + Metiram (5%)	WG	BASF India Ltd. Mumbai

Data analysis

Data were statistically analyzed using statistical analysis software (SAS) packages. Critical differences were calculated at a 5% level of significance for comparison of treatment mean. The Microsoft Excel (2010) computer software package was used to prepare all the graphs.

Result and Discussion

The mycelial growth inhibition of *Rhizoctonia solani*, causing web blight in Mungbean has been observed at various concentrations of fungicides under *in-vitro* conditions and was recorded (Table 2). The radial growth of *Rhizoctonia solani* was measured after 24 hrs of inoculation and subsequent observation was recorded at three days intervals till full growth of the pathogen in control *i.e.*, 120 hrs, and percent inhibition was calculated based on final observation. The perusal of the result showed that (Table 2) (Fig1and Plate 1) among all the five fungicides tested, hexaconazole was found effective at all concentrations (15, 25 50 100, and 150 ppm) inhibiting mycelial growth of *Rhizoctonia solani* by 100%. Whereas Propiconazole inhibit the mycelium growth by 100% @ 100 and 150 ppm. Vitavax (Carboxin + Thiram) and Carbendazim were found less effective against *R. solani* at 15, 25, 50, 100, and 150 ppm concentration over the control. The inhibitory effect of the test fungicides was increased with increasing fungicide concentrations. This finding is also in consonance with the results of Neelam *et al.* (2017) [15] that showed that Hexaconazole, Propiconazole and Carbendazim at 20 ppm were inhibiting the mycelial growth of *R. solani* by 91.48- 93.33% over check. Similarly, several researchers have reported similar results on the efficacy of hexaconazole against *R. solani.* This efficacy of the hexaconazole is may because of the demethylation of C-14 during ergosterol biosynthesis that leads to the accumulation of C- 14 methyl sterols (mode of action) (Dinakaran *et al.*, 2011; Gupta *et al.*, 2013; Kushwaha and Yadav, 2016; Yadav and Kushwaha, 2016) [3, 8, 12. 26]. Poussio *et al.*, (2021) [19] also reported Nativo (Tebuconazole + Trifloxystrobin) showed the highest mycelium growth inhibition of the *F. oxysporum* f.sp. *lycopersici i.e.*, 95.55, and 88.88%at 1000 and 500ppm respectively. Niwas *et al.* (2020) [17] reported carbendazim, at 500 and 750ppm completely inhibited the growth of *F. oxysporum* f. sp. *cubense* followed by Azoxystrobin *i.e.*, 32.96, 11.30, 8.12, and 7.16 mm growth observed in 100, 250, 500 and 750 ppm, respectively.

Table 2: *In vitro* effect of fungicides on the radial growth of the *Rhizoctonia solani* after the 5th day of incubation

Treatments		Concentration (ppm)									
		15 ppm	Inhibition over control (%)	25 ppm	Inhibition over control (%)	50 ppm	Inhibition over control (%)	100 ppm	Inhibition over control (%)	150 ppm	Inhibition over control (%)
		Mycelial growth (mm)									
T_1	Carboxin and Thiram	90	00	85.6	4.88	70.1	22.1	50.4	44.0	32.5	63.8
T_2	Hexaconazole	00	100	00	100	00	100	00	100	00	100
T_3	Propiconazole	42.5	52.7	28.6	68.2	23.4	74.0	00	100	00	100
T_4	Pyraclostrobin and Metiram	35.7	60.3	30.6	66.0	24.6	72.6	16.5	81.6	13.7	84.7
T_5	Carbendazim	90	00	82.6	8.22	67.4	25.1	47.4	47.3	28.8	68.0
T_6	Control (Check)	90.0	--	90.0	--	90.0	--	90.0	--	90.0	-
		Fungicide (A)		Concentration (B)				Interaction (A x B)			
CD @ 0.5%		1.07		0.76				2.21			
S.Em±		0.45		0.26				0.82			

*Mean of three replications; ppm: part per million

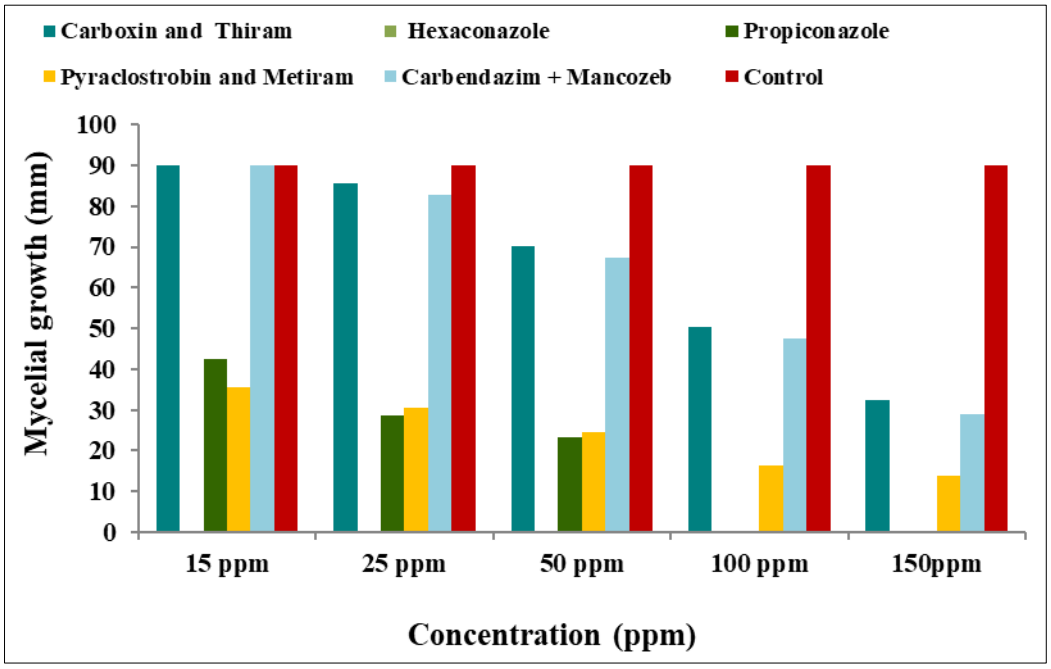


Fig 1: Effect of different fungicides on radial growth of *R. solani* under the *in-vitro* condition.

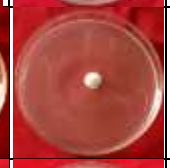
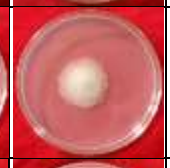
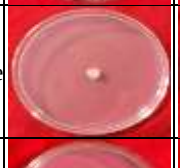
Treatments	15ppm	25ppm	50ppm	100ppm	150ppm	Control
Vitavex						
Metiram						
Carbendazim						
Hexoconazole						
Propiconazole						

Plate 1: The mycelium growth of *R. solani* against different fungicidal concentrations

Conclusion

From the foregoing result, it can be concluded that the tested fungicides significantly inhibited the web blight disease of Mungbean caused by *Rhizoctonia solani.* Among all the tested fungicides, Hexaconazole (Contaf) was found quite effective in cent percent inhibition of fungus mycelial growth followed by Propiconazole over control. Vitavax (Carboxin + Thiram) Carbendazim (Commando) showed the least effective against the pathogen based on antifungal efficiency.

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