

## POST - HARVEST EFFECT OF FOLIAR APPLICATION OF FUNGICIDES ON SEED GERMINATION AND SEED MYCOFLORA OF MUNG BEAN

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### ABSTRACT

The present study was conducted during year 2024 at section of Plant Pathology BTC, CARS, Bilaspur (C.G) to evaluate the post-harvest effect of foliar application of fungicides on seed germination and seed mycoflora. Seed samples from different fungicide-treated plots and untreated control were collected at crop harvest and stored at room temperature ( $25 \pm 2$  °C). The standard blotter paper method was used to assess seed germination and associated seed-borne fungi. Several seed-borne fungi, namely *Aspergillus* spp., *Fusarium* spp., *Cercospora* spp. and *Alternaria* spp. were detected in seeds from all treatments including the control. Results revealed that foliar application of fungicides significantly improved seed germination compared to untreated control. Seed germination ranged from 64.00% to 92.33% among treatments, whereas control recorded only 48.67%. The highest germination was observed in seeds from plots treated with Tebuconazole 50% + Trifloxystrobin 25% WG (92.33%), followed by Mancozeb 75% WP (91.33%), Hexaconazole 4% + Zineb 68% WP (90.00%), and Hexaconazole 5% SC (86.67%). However, Kasugamycin 3% SL, Validamycin 3% L and Tricyclazole 75% WP were comparatively less effective. The incidence of seed mycoflora was highest in untreated control (42.96%) and significantly reduced in fungicide-treated plots. The lowest mycoflora incidence was recorded in Tebuconazole 50% + Trifloxystrobin 25% WG (7.93%), followed by Tricyclazole 75% WP (11.63%), Propiconazole 25% EC (12.67%), Mancozeb 75% WP (14.00%) and Iprobenfos 48% EC (16.00%). Among the fungi detected, *Fusarium* spp. showed the highest frequency, followed by *Aspergillus* spp., *Cercospora* spp. and *Alternaria* spp. Overall, foliar application of fungicides effectively reduced seed-borne mycoflora and improved seed germination, indicating their potential role in improving seed health and quality.

(Keywords: Foliar fungicides, seed germination, seed mycoflora, seed-borne fungi, Blotter paper method, seed health)

### INTRODUCTION

Mung bean (*Vigna radiata* L. Wilczek), commonly known as green gram, belongs to the family Leguminosae. It is native to the India-Burma region of Southeast Asia. In India, mung bean is cultivated over an area of about 3.38 million hectares, with a total production of 1.61 million tonnes (Punse *et al.*, 2018). It serves as a valuable source of plant-based protein and plays a significant role in ensuring nutritional security in developing countries. The seeds contain approximately 20-24% protein along with carbohydrates, vitamins, minerals, dietary fibre, and bioactive compounds, which contribute to various health benefits and make mung bean an important component of human diets (Hou *et al.*, 2019; Ganesan and Xu, 2018).

Despite its nutritional and agronomic importance, mung bean productivity remains relatively low because of several biotic and abiotic stresses, including diseases, insect pests, drought, and poor crop management practices. Among these constraints, fungal diseases and other pathogens significantly affect plant growth, yield, and seed quality (Nair *et al.*, 2019; Pratap *et al.*, 2019)

Seed health is a critical factor influencing crop establishment and productivity. The presence of seed-borne pathogens in pulse crops often leads to reduced seed germination, poor seedling vigour, and weak plant stand in the field (Mathur and Kongsdal, 2003). Seed-borne fungi are capable of infecting seeds during crop growth, harvesting, and storage, and they may remain viable for a long period, acting as a source of infection for subsequent crops (Neergaard, 1979).

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Several seed-borne fungi have been reported to be associated with mung bean seeds. Among them, *Fusarium* spp., *Aspergillus* spp., *Alternaria* spp., and *Cercospora* spp. are considered the most common and economically important pathogens affecting seed quality and germination (Singh *et al.*, 2013). These fungi may cause seed discoloration, seed rot, and deterioration during storage, resulting in reduced seed viability and vigour. In severe cases, these pathogens may also lead to the transmission of diseases from infected seeds to healthy plants, thereby affecting crop productivity (Agarwal and Sinclair, 1997).

Fungal pathogens can infect mung bean plants at various growth stages, particularly during flowering, pod formation and seed maturation. Environmental factors such as temperature and humidity play an important role in the development and spread of these pathogens. Under favourable environmental conditions, fungal spores may colonize pods and seeds, leading to contamination of harvested seeds with mycoflora (Agrios, 2005; Sharma *et al.*, 2020).

Foliar application of fungicides during crop growth can significantly reduce disease severity on leaves, stems, and pods, thereby limiting the infection of developing seeds. By reducing fungal infection during the later stages of crop development, fungicide treatments can improve seed health and reduce the incidence of seed-borne mycoflora after harvest (Mahal, 2016). Previous studies have reported that fungicidal treatments such as triazoles, strobilurins, and protectant fungicides effectively reduce fungal colonization and improve seed germination and seedling vigour in mung bean (Chatte *et al.*, 2022).

Detection and identification of seed-borne fungi are important aspects of seed health testing. Several laboratory techniques are used for detecting seed mycoflora, among which the standard blotter paper method is widely used due to its simplicity and reliability. This method allows the growth and identification of fungal colonies present on seeds under controlled laboratory conditions and is commonly recommended by the International Seed Testing Association for seed health testing (Anonymous, 2019).

## MATERIALS AND METHODS

### Experimental site and crop

The present investigation was carried out during the cropping season of 2024 at the experimental field of the section of Plant Pathology. Mung bean (*Vigna radiata* L.) was grown under field conditions following recommended agronomic practices. The crop was raised in different plots and subjected to foliar application of various fungicides along with an untreated control.

### Treatment details

There were seventeen treatments comprising sixteen fungicidal treatments and one untreated control, each replicated thrice. The treatments included T<sub>1</sub> - Metiram 70% WG, T<sub>2</sub> - Tricyclazole 75% WP, T<sub>3</sub> - Propiconazole 25% EC, T<sub>4</sub> - Azoxystrobin 18.2% + Difenoconazole 11.4% SC, T<sub>5</sub> - Validamycin 3% L, T<sub>6</sub> - Hexaconazole 5% SC, T<sub>7</sub> - Iprobenfos 48% EC, T<sub>8</sub> - Azoxystrobin 11% + Tebuconazole 18.3% SC, T<sub>9</sub> - Thifluzamide 24% SC, T<sub>10</sub> - Mancozeb 75% WP, T<sub>11</sub> - Propineb 70% WP, T<sub>12</sub> - Tebuconazole 50% + Trifloxystrobin 25% WG, T<sub>13</sub> - Kasugamycin 3% SL, T<sub>14</sub> - Tebuconazole 38.39% SC, T<sub>15</sub> - Hexaconazole 4% + Zineb 68% WP, T<sub>16</sub> - Metiram 55% + Pyraclostrobin 5% WG and T<sub>17</sub> - untreated control.

### Collection and storage of seed samples

Seeds from different fungicide-treated plots and the untreated control were collected at the time of crop harvesting. The collected seed samples were cleaned and stored in paper bags under room temperature conditions (25 ± 1 °C) until further laboratory analysis.

### Seed germination test

The seed germination test was conducted using the standard blotter paper method as described by (Anonymous, 2019). The experiment was laid out in a completely randomized design (CRD) with three replications. Sterilized blotter sheets of uniform diameter were moistened with sterilized distilled water and placed in sterilized Petri plates. Fifty seeds were placed in each Petri plate. The seeded plates from different treatments were incubated for seven days at 25 ± 1 °C in a BOD incubator with alternating cycles of 12 hours light and 12 hours darkness. The blotter paper method was adopted as it provides suitable moisture and environmental conditions for uniform seed germination and is widely recommended for seed health testing.

### Detection of seed mycoflora

Seed mycoflora associated with mung bean seeds were detected using the standard blotter paper method (Anonymous, 2019). Seeds were placed on moist blotter papers in sterilized Petri dishes and incubated at 25 ± 1 °C under alternating light and dark conditions for seven days. This method facilitates the growth and development of fungi present on seeds, enabling easy detection of seed-borne pathogens. After incubation, the seeds were examined under a microscope for the development of fungal colonies. The fungi growing on seeds were identified based on their morphological and microscopic characteristics using standard identification keys (Barnett and Hunter, 1998). The frequency of occurrence of different fungi such as *Fusarium* spp., *Aspergillus* spp., *Alternaria* spp., and *Cercospora* spp. was recorded. Observations i.e. per cent seed germination, number of seeds infected with seed mycoflora and per cent seed borne mycoflora was recorded from each treatment using following formula.

## Observations recorded

$$\text{Per cent seed germination} = \frac{\text{Number of seeds germinated}}{\text{Total number of seed examined}} \times 100$$

$$\text{Per cent seed mycoflora} = \frac{\text{Number of seeds with seed borne fungi}}{\text{Total number of seed examined}} \times 100$$

$$\text{Seed mycoflora frequency} = \frac{\text{Seed borne fungus associated with number of seeds}}{\text{Total number of seed borne fungi}} \times 100$$

## Statistical analysis

The data obtained from the laboratory experiments on seed germination and seed mycoflora were statistically analysed following appropriate statistical methods. Treatment means were compared to determine the effectiveness of different fungicides in improving seed germination and reducing the incidence of seed mycoflora.

## RESULTS AND DISCUSSION

### Post-harvest effect of foliar application of fungicides on seed germination

Data recorded from Table 1 on post-harvest effect of fungicide indicated that there was significant increase in seed germination compared to seeds collected from the untreated plot. Seed germination recorded from different treatments was ranged between 64.00%-92.33%. The highest seed germination was recorded from Tebuconazole 50% + Trifloxystrobin 25% WG (92.33%), closely followed by Mancozeb 75% WP (91.33%), Hexaconazole 4% + Zineb 68% WP (90.00%) and Hexaconazole 5% SC (86.67%). Other fungicides i.e. Tebuconazole 38.39% w/w SC (82.67%), Iprobenfos 48% EC (82.00%) and Propineb 70% WP (81.00%) were also effective in enhancing the per cent seed germination over untreated control (48.67%). Whereas Kasugamycin 3% SL (71.33), Validamycin 3% L (69.63%) and Tricyclazole 75% WP (64.00%) was found less effective. Higher seed germination in treated plots might be due to less incidence of seed borne mycoflora at the time of maturity of crop. It is also indicated from the present study that the significant reduction on seed germination from control plot might be due to heavy incidence of seed borne mycoflora (Chatte *et al.*, 2022).

### Post-harvest effect of foliar application of fungicides on seed mycoflora

Result from Table 1 indicated that seed mycoflora i.e. *Alternaria* spp., *Aspergillus* spp, *Cercospora* spp. and *Fusarium* spp., were found to be associated with the seeds taken from different treatments. However, the highest frequency was observed for *Fusarium* spp., followed by *Aspergillus* spp., *Cercospora* spp. and *Alternaria* spp.

Results indicated that the seeds from untreated control were found to be colonised with the highest percentage (42.96) compared to seeds taken from different treatments.

The lowest incidence of seed mycoflora was observed in Tebuconazole 50% + Trifloxystrobin 25% WG (7.93), followed by Tricyclazole 75% WP (11.63), Propiconazole 25% EC (12.67), Mancozeb 75% WP (14.00) and Iprobenfos 48% EC (16.00). Other fungicides i.e. Validamycin 3% L (16.23), Azoxystrobin 18.2% + Difenconazole 11.4% SC (17.33), Propineb 70% WP (17.99), Hexaconazole 4% + Zineb 68% WP (20.00), Hexaconazole 5% SC (21.67) and Metiram 55% + Pyraclostrobin 5% WG (22.68) were also found significantly effective in checking the per cent seed mycoflora over control. Azoxystrobin 11% + Tebuconazole 18.3% SC (25.33), Kasugamycin 3% SL (27.66), Metiram 70% WG (31.33), Tebuconazole 38.39% w/w SC (32.00) and Thifluzamide 24% SC (34.29) were found less effective in checking incidence of seed mycoflora compared to other fungicides.

***Alternaria* spp.** Amongst the fungicide treated plot the lowest incidence of *Alternaria* spp. was recorded from Metiram 70% WG (1.96) followed by Thifluzamide 24% SC (2.74), Propiconazole 25% EC (3.29), Mancozeb 75% WP (3.71), Tebuconazole 38.39% w/w SC (4.10), Validamycin 3% L (4.16), Azoxystrobin 18.2% + Difenconazole 11.4% SC (4.39), Tebuconazole 50% + Trifloxystrobin 25% WG (4.41), Propineb 70% WP (4.48), Hexaconazole 4% + Zineb 68% WP (4.73) and Hexaconazole 5% SC (4.84) over control (20.54). Other fungicides i.e. Iprobenfos 48% EC (6.16), Metiram 55% + Pyraclostrobin 5% WG (8.44), Kasugamycin 3% SL (8.55), Tricyclazole 75% WP (10.95) and Azoxystrobin 11% + Tebuconazole 18.3% SC (11.92) were less effective.

***Aspergillus* spp.** Amongst the fungicide treated plot the lowest incidence was observed in Tebuconazole 50% + Trifloxystrobin 25% WG (1.42), followed by Tricyclazole 75% WP (2.80), Thifluzamide 24% SC (3.13), Validamycin 3% L (4.42), Mancozeb 75% WP (4.48), Iprobenfos 48% EC (4.49), Propineb 70% WP (4.85), Hexaconazole 4% + Zineb 68 %WP (4.73), Azoxystrobin 11% + Tebuconazole 18.3% SC (4.93), Azoxystrobin 18.2%

**Table 1. Post - harvest effect of foliar application of fungicides on seed germination and seed mycoflora**

Treatments		Per cent seed germination	Per cent seed mycoflora	<i>Aspergillus</i> spp.	<i>Fusarium</i> spp.	<i>Cercospora</i> spp.	<i>Alternaria</i> spp.
T <sub>1</sub>	Metiram 70% WG	77.33	31.33	5.35	8.23	3.79	1.96
T <sub>2</sub>	Tricyclazole 75% WP	64.00	11.63	2.80	9.52	4.10	10.95
T <sub>3</sub>	Propiconazole 25% EC	73.33	12.67	7.30	4.01	2.38	3.29
T <sub>4</sub>	Azoxystrobin 18.2% + Difenoconazole 11.4% SC	78.00	17.33	5.09	9.83	3.53	4.39
T <sub>5</sub>	Validamycin 3% L	69.63	16.23	4.42	10.45	7.66	4.16
T <sub>6</sub>	Hexaconazole 5% SC	82.67	21.67	7.73	7.25	5.52	4.84
T <sub>7</sub>	Iprobenfos 48% EC	86.00	16.00	4.49	3.24	5.14	6.16
T <sub>8</sub>	Azoxystrobin 11% +Tebuconazole 18.3% SC	75.92	25.33	4.93	21.77	7.70	11.92
T <sub>9</sub>	Thifluzamide 24% SC	78.47	34.29	3.13	8.69	3.67	2.74
T <sub>10</sub>	Mancozeb 75% WP	91.33	14.00	4.48	2.93	5.16	3.71
T <sub>11</sub>	Propineb 70% WP	81.00	17.99	4.85	4.52	7.81	4.48
T <sub>12</sub>	Tebuconazole 50% + Trifloxystrobin 25% WG	92.33	7.93	1.42	0.00	3.72	4.41
T <sub>13</sub>	Kasugamycin 3% SL	71.33	27.66	9.46	6.59	11.37	8.55
T <sub>14</sub>	Tebuconazole 38.39% w/w SC	82.67	32.00	6.61	3.51	8.91	4.10
T <sub>15</sub>	Hexaconazole 4% + Zineb 68% WP	90.00	20.00	4.73	4.72	9.47	4.73
T <sub>16</sub>	Metiram 55% + Pyraclostrobin 5% WG	78.00	22.68	10.21	6.41	6.28	8.44
T <sub>17</sub>	Control	48.67	42.96	25.10	32.27	21.99	20.54
	SE(m) ±	1.31	0.58	0.53	0.42	0.49	0.34
	CD at 5%	3.79	1.66	1.53	1.20	1.43	1.00
	Coefficient of Variation (CV)	2.93	4.57	10.93	8.53	11.34	9.33

Figure 1. Post-harvest effect of foliar application of fungicides on seed germination and seed mycoflora



Tebuconazole 50%+  
Trifloxystrobin 25% WG



Mancozeb 75% WP



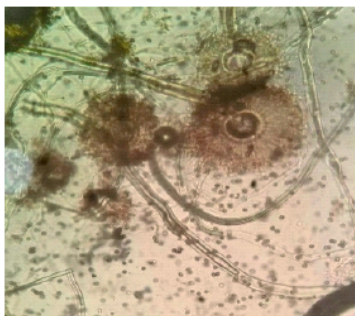
Hexaconazole 4% +  
Zineb 68% WP



Hexaconazole 5% SC



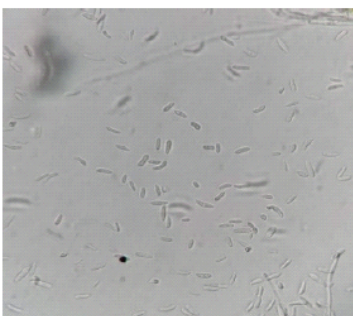
Control



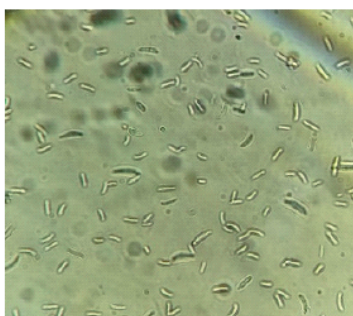
*Aspergillus* spp.



*Cercospora* spp.



*Fusarium* spp.



*Alternaria* spp.



+ Difenconazole 11.4% SC (5.09) and Metiram 70% WG (5.35) over control (25.10). Other fungicide i.e. Tebuconazole 38.39% w/w SC (6.61), Propiconazole 25% EC (7.30), Hexaconazole 5% SC (7.73), Kasugamycin 3% SL (9.46) and Metiram 55% + Pyraclostrobin 5% WG (10.21) were less effective.

**Cercospora spp.** Amongst the fungicide treated plot the lowest incidence was observed in Propiconazole 25% EC (2.38) followed by Azoxystrobin 18.2% + Difenconazole 11.4% SC (3.53), Thifluzamide 24% SC (3.67), Tebuconazole 50% + Trifloxystrobin 25% WG (3.72), Metiram 70% WG (3.79), Tricyclazole 75% WP (4.10), Iprobenfos 48% EC (5.14), Mancozeb 75% WP (5.16) and Hexaconazole 5% SC (5.52) over control (21.99). Other fungicides i.e. Metiram 55% + Pyraclostrobin 5% WG (6.28) Validamycin 3% L (7.66), Azoxystrobin 11% + Tebuconazole 18.3% SC (7.70), Propineb 70% WP (7.81), Tebuconazole 38.39% w/w SC (8.91), Hexaconazole 4% + Zineb 68% WP (9.47) and Kasugamycin 3% SL (11.37) were less effective.

**Fusarium spp.** Amongst the fungicide treated plot the lowest incidence was observed in Tebuconazole 50% + Trifloxystrobin 25% WG (0.00) followed by Mancozeb 75% WP (2.93), Iprobenfos 48% EC (3.24), Tebuconazole 38.39% w/w SC (3.51), Propiconazole 25% EC (4.01), Propineb 70% WP (4.52) and Hexaconazole 4% + Zineb 68% WP (4.73) over control (32.27). Other fungicide i.e. Metiram 55% + Pyraclostrobin 5% WG (6.41), Kasugamycin 3% SL (6.59), Hexaconazole 5% SC (7.25), Metiram 70% WG (8.23), Thifluzamide 24% SC (8.69), Tricyclazole 75% WP (9.52), Azoxystrobin 18.2% + Difenconazole 11.4% SC (9.83), Validamycin 3% L (10.45) and Azoxystrobin 11% + Tebuconazole 18.3% SC (21.77) were less effective.

The lower incidence of seed mycoflora and higher seed germination from the seeds collected from different plots treated with fungicides indicated that the foliar application of fungicides might have provided the protective layer around the seeds which significantly prevented the attack of seed borne fungi during the maturity of crop. The higher efficacy of Tebuconazole 50% + Trifloxystrobin 25% WG, Mancozeb 75% WP, Hexaconazole 4% + Zineb 68% WP and Hexaconazole 5% SC in checking incidence of seed mycoflora and increasing the per cent seed germination under the present study indicated that these fungicides are not only effective in reducing the foliar diseases of mung bean but also may protect the seed from the infestation of seed borne mycoflora during the storage period (Kavyashree *et al.*, 2016).

The above results are in accordance with Ali *et al.* (2011), who also recorded the incidence of *Aspergillus* spp., *Fusarium* spp. and *Alternaria* spp. in seed samples of mung bean.

The higher efficacy of Mancozeb in checking the incidence of seed mycoflora under the present study is in accordance with the findings of Singh *et al.* (2014), who reported the maximum reduction of seed mycoflora and highest per cent of seed germination by the application Mancozeb.

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