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Arbuscular mycorrhizal fungi association and root rot disease in mungbean (*Vigna radiata*) growing areas of Haryana

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ABSTRACT

The study was carried out during rainy (*kharif*) seasons of 2021 and 2022 at Chaudhary Charan Singh Haryana Agricultural University, Hisar, Haryana to provide a comprehensive understanding of how the presence and activity of arbuscular mycorrhizal (AM) fungi may influence the development of root rot in mungbean (*Vigna radiata* L.) crops in different mungbean-growing areas of Haryana. Root colonization by AM fungi in Hisar, Bhiwani, Mahendragarh, and Charkhi Dadri districts of Haryana ranged from 16.45 to 24.07%, 7.71 to 16.73%, 7.02 to 21.43% and 7.59 to 22.28%, respectively. This variability underscores the diverse interactions between mungbean plants and AM fungi in these distinct geographical regions, reflecting the influence of environmental factors and local soil conditions on mycorrhizal associations. Sporocarp numbers in these districts ranged from 76.5 to 131.5, 37 to 102, 54 to 119.5 and 65.5 to 118 per 100 g of soil, respectively. These findings emphasize the dynamic nature of AM fungal populations and their potential impact on the overall soil health and plant-microbe interactions in mungbean cultivation. Similarly, root rot incidence in these districts varied from 6.59 to 11.62%, 7.48 to 11.29%, 7.05 to 11.43% and 5.88 to 11.76%, while root rot intensity ranged from 2.77 to 4.82%, 2.70 to 4.61%, 2.06 to 4.61% and 2.43 to 4.85% in Hisar, Bhiwani, Mahendragarh, and Charkhi Dadri districts, respectively. This variability in root rot incidence and intensity highlights the susceptibility of mungbean crops to this detrimental disease and emphasizes the importance of region-specific strategies for disease management.

Keywords: Arbuscular mycorrhizal (AM) fungi, Disease incidence, Disease intensity, Mungbean, Per cent mycorrhizal colonization

Mungbean [*Vigna radiata* (L.) Wilczek] is one of the most important pulse crops in India. It has wider adaptability and is grown in different seasons under varied agro-climatic conditions in the country. India stands as the world's largest producer of pulses, contributing approximately 25–28% of global pulse production. Mungbean is grown across 0.55 million hectares in India, yielding a total production of 0.31 million tonnes. This crop thrives in various regions, including Madhya Pradesh, Rajasthan, Maharashtra, Karnataka, Gujarat, Bihar, Punjab, and Haryana. In Haryana, mungbean is cultivated across 79.24 thousand hectares, yielding a production of 52.38 thousand tonnes, with an average yield of 6.61 q/ha (Anonymous 2021–22).

Root rot disease in mungbean is a significant yield-limiting factor, primarily caused by *M. phaseolina* (Tassi) Goid (Pycnidial stage) and *Rhizoctonia bataticola* (Anamorphic stage). This disease leads to considerable annual yield losses about 10–44% in mungbean production in India, whereas Rhizoctonia root rot causes 33–44% yield losses (Nair *et al.* 2019). The predominant symptoms of root rot are yellowing of leaves, which drop off within 2–3 days. Within a week, the plants may shrivel, and dark lesions on the stem's bark may be observed at ground level. When plants are removed from the soil and their basal stems and main roots are examined, root rot signs become evident. Scattered sclerotial bodies may be observed on the altered tissues in the advanced stage (Kumar *et al.* 2020).

AM fungi is known as stress regulating organisms that help plants in nutrient uptake (Kadian *et al.* 2014, Boutaj *et al.* 2022), biotic (Singh 2016, Sarita and Chugh 2020) and abiotic stress management, plant protection and consequently enhancement of crop yields. Also plants can tolerate many difficult situations such as water problems, salt stress, heavy metals and temperature changes through AM fungi inoculation (Mitra *et al.* 2020). Therefore, present study was aimed to investigate the association between AM fungi and incidence/intensity of root rot in mungbean.

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MATERIALS AND METHODS

Survey and sample collection: To assess root colonization, spore population and the incidence, and intensity of root rot, a survey was conducted during rainy (*kharif*) seasons of 2021 and 2022 at various mungbean-growing areas of Haryana, viz. Hisar, Bhiwani, Mahendragarh and Charkhi Dadri. The survey encompassed 10 randomly selected fields from each district. From each site, 10 plants were chosen at random and uprooted for the subsequent calculation of root rot incidence and intensity. For the assessment of sporocarp numbers in the soil, 250 g of soil samples were collected, following the method outlined by Gerdemann and Nicolson (1963). To estimate mycorrhizal colonization in the roots, plant root samples were collected from each location and placed in polythene bags, following the procedure outlined by Phillips and Hayman (1970).

Estimation of mycorrhizal colonization: The estimation process involved several steps: The roots were cleaned under running water and then cutted into 2 cm segments. The segments were boiled in a 10% KOH solution at 90°C for 1 h. After boiling, the roots were rinsed with fresh 10% KOH solution and submerged in alkaline hydrogen peroxide (H_2O_2) for 30 min. Subsequently, the roots were acidified with 5 N HCL for 30 min before being washed with distilled water to remove any excess H_2O_2. The roots were then immersed in a solution of lactophenol (0.05%) and trypan blue for 5 min. After soaking in lactophenol, the roots were inspected under a microscope to remove any excess dye colour. Root colonization was assessed using a rating scale ranging from 0 to 5, and the calculation was performed as:

$$\text{Mycorrhizal colonization (\%)} = \frac{\text{No. of root bits showing colonization}}{\text{Total number of roots bits observed}} \times 100$$

Mycorrhizal root colonization rating scale:

Rating	Number of mycorrhizal spore in root
0	0
1	1–25
2	25–50
3	50–75
4	75–90
5	>90

Source: Jalali and Domstch (1975)

Estimation of sporocarp numbers in soil: The soil sample was meticulously mixed, and 100 g of soil was suspended in a pan filled with 1 litre of water. The mixture was thoroughly blended and left to settle for 30 sec. Subsequently, the filtrate was carefully collected in pan B, with the material in pan A being discarded. The suspension in pan B was gently agitated by hand for a few seconds and then passed through a 60-mesh sieve, leading to the collection of the filtrate in pan C. In pan C, the suspension underwent further filtration, this time using a 100-mesh sieve. This step allowed for the retrieval of the most mature sporocarps, which were washed to remove excess soil and other particles. The residue from the 100-mesh sieve was collected into a beaker. A 1 ml sample of this solution was placed in a counting plate, and the sporocarp population in the soil was quantified using a stereomicroscope.

Estimation of disease incidence and intensity: A total of 40 fields were chosen for the survey to assess the incidence and intensity of root rot disease. In each field, four 1 m^2 quadrants were randomly selected. Within each quadrant, the numbers of infected plants were tallied to determine the disease incidence (Belkar and Gade 2016).

Disease intensity was scored on 10 plants randomly selected per field using a disease intensity rating scale ranging from 1 to 9, and calculated as:

$$\text{Disease intensity (\%)} = \frac{\text{Sum of all numerical rating}}{\text{Total no. of sample observed} \times \text{maximum scale}} \times 100$$

Disease intensity rating scale:

Grade	Disease intensity	Disease reactions
1	0–10%	No infection on roots
3	10–25%	Very few small lesions on roots
5	25–50%	Lesions on roots clear but small, new roots free from infection
7	50–75%	Lesions on roots many, new roots generally free from lesions
9	>75%	Roots infected and completely discolored

Source: Nene *et al.* (1981)

RESULTS AND DISCUSSION

Mycorrhizal root colonization and spore population: A survey was conducted at the mungbean-growing districts of Haryana, including Hisar, Bhiwani, Mahendragarh, and Charkhi Dadri, to record mycorrhizal colonization and sporocarp numbers in 100 g of mungbean soil. The data (Table 1) exhibited a pooled average range for root colonization (13.44 to 20.23%) and sporocarp numbers (64.06 to 96.70) during the *kharif* seasons of 2021 and 2022.

In Hisar district, the maximum root colonization (25.09%) was recorded in Nalwa followed by Bheria (23.99%), Kanwari (23.20%), Muklan (22.60%), HAU Farm (20.10%), Chandan nagar (19.90%), Deva (18.90%), Balsamand (17.90%), Mirkan (17.80%), and Chaudhariwas (14.60%) villages during the *kharif* season of 2021. Similarly, during the *kharif* season of 2022, the maximum colonization (23.05%) was observed in Nalwa followed by 23.00%, 22.50%, and 22.10% in Bheria, Muklan and HAU Farm village. The minimum colonization was recorded in Chaudhariwas (14.60%) and Mirkan (15.80%) villages during the *kharif* seasons of 2021 and 2022, respectively.

In Bhiwani district, the maximum root colonization (19.34%) was found in Tosham Rural followed by 17.22%, 15.67%, 15.31%, 13.11%, 12.56%, 11.67%, 10.54%, 8.38%, and 6.30% in Siwani, Dulheri, Kharkhari, Dhani

Mansukh, Jhumpa Kalan, Pahari, Surpura Kalan, Khanak, and Bithan villages during the *kharif* season of 2021. Similarly, during the *kharif* season of 2022, the maximum colonization (17.78%) was recorded in Dulheri followed by 17.67%, 16.17%, 15.32%, and 15.19% in Surpura Kalan, Dhani Mansukh, Kharkhari, and Siwani villages. The minimum colonization was recorded in Bithan (6.30% and 9.11%) village during the *kharif* seasons of 2021 and 2022, respectively.

Furthermore, in Mahendragarh district, the maximum root colonization (22.65%) was observed in Patharwa followed by 19.21%, 18.43%, 16.34%, and 14.27% in Bawania, Khatod, Jat Guwana, and Beri villages during the *kharif* season of 2021. Similarly, during the *kharif* season of 2022, the maximum colonization (20.21%) was recorded in Patharwa followed by 19.52%, 16.33%, 15.28%, and 15.27% in Khatod, Jat Guwana, Bawania, and Beri villages. The minimum colonization was recorded in Siana (7.51% and 6.53%) village during both the seasons.

In Charkhi Dadri district, the maximum root colonization (23.21%) was found in Jhojhu Kalan followed by 21.46%, 19.24%, 15.26%, and 14.55% in Sehlanga, Badhra, Dadri, and Charkhi villages during the *kharif* season of 2021. Similar trend was observed during *kharif* 2022. The minimum colonization was recorded in Birhi Kalan (8.53% and 6.64%) village during both the seasons.

Similar results were observed by Molla and Solaiman (2009), who found variations in root colonization and spore numbers in different leguminous crops in various agro-ecological zones in Bangladesh. Hindumathi and Reddy (2012) reported mycorrhizal root colonization ranging from 36.74 to 90.68% in Mungbean plants from Andhra Pradesh districts. Singh (2018) observed varying root colonization in different districts of Punjab during different seasons.

The highest spore population was recorded in soil samples collected from Muklan village (128 spores in 2021 and 135 spores in 2022) in Hisar district during both *kharif* seasons, while the minimum spore populations were recorded in Surpura Kalan (35 spores) and Kharkhari (36 spores) villages in Bhiwani district during *kharif* seasons of 2021 and 2022, respectively. The pooled average of per cent colonization and spore population was highest in Hisar (20.23% and 96.70), followed by Charkhi Dadri (14.72% and 86.30) and Mahendragarh (14.07% and 82.60), while the lowest colonization and spore population were recorded in Bhiwani district (13.44% and 64.06).

Similarly, Molla and Solaiman (2009) found variations in spore numbers in different leguminous crops across various agro-ecological zones in Bangladesh, with spore populations ranging from 89.4 to 495, 40 to 390, 89 to 390, 99.6 to 298, and 100 to 295/100 g of soil in AEZs Rangpur, Jessore, Faridpur, Noakhali, and Gazipur, respectively. Hindumathi and Reddy (2012) collected rhizosphere soil samples from Mungbean fields in Adilabad, Nizamabad, and Karimnagar districts of Andhra Pradesh and found spore populations ranging from 12 to 89/10 g of soil. Singh (2018) recorded varying spore populations ranging maximum 800 to minimum 125 spores/100 g of rhizosphere soil in different districts of Punjab during different seasons from Ferozepur and Bathinda in spring season, respectively. Similarly, in *kharif* season the maximum spore population was recorded from Ferozepur (780 spores) districts and it was minimum in Patiala (450 spores). The number of mycorrhizal spores of *Glomus* spp. varied from 76.72 to 440.29 in Ferozepur and Bathinda during both seasons, respectively.

Root rot disease incidence and intensity: The data on root rot incidence and intensity (Table 2) revealed that pooled average root rot incidence ranged from 8.65 to 9.14% and root rot intensity ranged from 3.34 to 3.50% during *kharif* 2021 and 2022, respectively. In Hisar district, the maximum root rot incidence (12.33%) was recorded in Muklan followed by 10.99%, 9.57%, 9.13%, and 9.10% in Kanwari, Mirkan, Bheria, and Deva villages during the *kharif* season of 2021. Similarly, during the *kharif* season of 2022, the maximum root rot incidence (10.91%) was observed in Muklan followed by 10.75%, 10.10%, 9.30%, and 8.94% in HAU Farm, Balsamand, Chandan Nagar, and Kanwari villages. The minimum root rot incidence was recorded in Chaudhariwas (4.35%) and Bheria (4.78%) villages during the *kharif* seasons of 2021 and 2022, respectively. The maximum root rot intensity was recorded in Muklan (5.21% and 4.43%), while the minimum was recorded in HAU farm (2.05%) and Bheria (2.39%) villages during the *kharif* seasons of 2021 and 2022, respectively.

In Bhiwani district, the maximum root rot incidence was 13.14%, followed by 12.14%, 10.00%, 9.76%, and 9.47% in Siwani, Surpura Kalan, Bithan, Dulheri, and Tosham rural villages during *kharif* season of 2021. Similarly, during *kharif* season of 2022, the maximum root rot incidence was 10.77%, followed by 10.40%, 9.86%, 9.44%, and 8.46% in Khanak, Surpura Kalan, Dulheri, Siwani, and Pahari villages. The minimum root rot incidence was 7.25% and 6.35% in Pahari and Bithan villages during the *kharif* seasons of 2021 and 2022, respectively. The maximum root rot intensity was recorded in Siwani (5.53%) and Khanak (4.36%), while the minimum was recorded in Jhumpa Kalan (2.58%) and Bithan (2.15%) villages during the *kharif* seasons of 2021 and 2022, respectively.

Furthermore, in Mahendragarh district, the maximum root rot incidence was 13.06%, followed by 12.83%, 10.94%, 10.86%, and 8.97% in Jat Guwana, Siana, Khatod, Kheri Talwana, and Baghot villages during *kharif* season of 2021. Similarly, during *kharif* season of 2022, the maximum root rot incidence was 10.84%, followed by 10.14%, 10.02%, 9.55%, and 9.39% in Kheri Talwana, Khatod, Siana, Dalanwas, and Jat Guwana villages. The minimum root rot incidence was recorded in Bawania (6.15%) and Jhagroli (5.96%) villages during the *kharif* seasons of 2021 and 2022. The maximum root rot intensity was recorded in Jat Guwana (5.54%) and Kheri Talwana (4.38%), while the minimum was recorded in Bawania (2.03% and 2.09%) village during the *kharif* seasons of 2021 and 2022, respectively.

The pooled average of the maximum root rot incidence and intensity was highest in Bhiwani district (9.14%

Table 1 Mycorrhizal root colonization and spore population of AM fungi in different districts of Haryana in mungbean during *kharif* seasons of 2021 and 2022

District	Village	Soil type	Mycorrhizal colonization (%)			Sporocarp numbers/100 g soil		
			2021	2022	Pooled	2021	2022	Pooled
Hisar	Hau Farm	Loamy sand	20.10	22.10	21.10	110	100	105.0
	Chandan nagar	Loamy sand	19.90	17.80	18.85	102	112	107.0
	Deva	Sandy loam	18.90	20.10	19.50	87	89	88.0
	Muklan	Sandy loam	22.60	22.50	22.55	128	135	131.5
	Bheria	Sandy loam	23.99	23.00	23.50	94	98	96.0
	Chaudhariwas	Sandy loam	14.60	18.30	16.45	100	105	102.5
	Balsamand	Sandy loam	17.90	18.70	18.30	91	92	91.5
	Nalwa	Sandy loam	25.09	23.05	24.07	80	78	79.0
	Kanwari	Sandy loam	23.20	19.20	21.20	96	83	89.5
	Mirkan	Sandy loam	17.80	15.80	16.80	73	80	76.5
Average					20.23			96.70
Bhiwani	Siwani	Sandy loam	17.22	15.19	16.21	107	98	102.5
	Jhumpa Kalan	Sandy loam	12.56	13.43	13.00	95	102	98.5
	Surpura Kalan	Sandy loam	10.54	17.67	14.11	35	39	37.0
	Bithan	Sandy loam	6.30	9.11	7.71	66	70	68.0
	Pahari	Sandy loam	11.67	12.23	11.95	53	55	54.0
	Kharkhari	Sandy loam	15.31	15.32	15.32	40	36	38.0
	Dhani Mansukh	Sandy loam	13.11	16.17	14.64	43	43	43.0
	Dulheri	Sandy loam	15.67	17.78	16.73	56	58	57.0
	Tosham Rural	Loamy sand	19.34	11.56	15.45	69	72	70.5
	Khanak	Loamy sand	8.38	10.25	9.32	74	80	77.0
Average					13.44			64.06
Mahendragarh	Patharwa	Sandy loam	22.65	20.21	21.43	81	85	83.0
	Dalanwas	Sandy loam	12.23	14.26	13.25	95	92	93.5
	Khatod	Sandy loam	18.43	19.52	18.98	118	121	119.5
	Beri	Sandy loam	14.27	15.27	14.77	107	103	105.0
	Jat Guwana	Sandy loam	16.34	16.33	16.34	84	84	84.0
	Bawania	Sandy loam	19.21	15.28	17.25	67	71	69.0
	Jhagroli	Sandy loam	10.52	11.62	11.07	55	53	54.0
	Kheri Talwana	Sandy loam	9.35	8.28	8.82	59	60	59.5
	Baghot	Sandy loam	11.21	12.36	11.79	74	76	75.0
	Siana	Sandy loam	7.51	6.53	7.02	81	86	83.5
Average					14.07			82.60
Charkhi Dadri	Jhojhu Kalan	Sandy loam	23.21	21.34	22.28	84	81	82.5
	Ladawas	Sandy loam	11.32	12.28	11.80	86	89	87.5
	Sehlanga	Sandy loam	21.46	20.37	20.92	114	122	118.0
	Makrana	Sandy loam	13.18	12.23	12.71	108	111	109.5
	Dadri	Sandy loam	15.26	16.26	15.76	79	81	80.0
	Badhra	Sandy loam	19.24	20.28	19.76	65	66	65.5
	Atela Khurd	Sandy loam	10.61	10.61	10.61	92	92	92.0
	Charkhi	Sandy loam	14.55	15.55	15.05	64	67	65.5
	Dohki	Sandy loam	11.36	10.12	10.74	88	92	90.0
	Birhi Kalan	Sandy loam	8.53	6.64	7.59	71	74	72.5
Average					14.72			86.30

Table 2 Mungbean root rot disease incidence and intensity in different districts of Haryana during *kharif* seasons of 2021 and 2022

District	Village	Root rot incidence (%)			Root rot intensity (%)		
		2021	2022	Pooled	2021	2022	Pooled
Hisar	Hau Farm	6.14	10.75	8.45	2.05	4.30	3.18
	Chandan nagar	7.53	9.30	8.42	2.81	3.69	3.25
	Deva	9.10	7.40	8.25	3.53	2.70	3.12
	Muklan	12.33	10.91	11.62	5.21	4.43	4.82
	Bheria	9.13	4.78	6.96	3.61	2.39	3.00
	Chaudhariwas	4.35	8.83	6.59	2.13	3.41	2.77
	Balsamand	8.89	10.10	9.50	3.45	3.99	3.72
	Nalwa	8.02	8.12	8.07	3.01	3.23	3.12
	Kanwari	10.99	8.94	9.97	4.43	3.54	3.99
	Mirkan	9.57	7.79	8.68	3.79	2.89	3.34
Average				8.65			3.43
Bhiwani	Siwani	13.14	9.44	11.29	5.53	3.69	4.61
	Jhumpa Kalan	7.32	7.63	7.48	2.58	2.81	2.70
	Surpura Kalan	12.14	10.40	11.27	4.08	4.18	4.13
	Bithan	10.00	6.35	8.18	4.00	2.15	3.08
	Pahari	7.25	8.46	7.86	2.60	3.21	2.91
	Kharkhari	9.01	7.66	8.34	3.50	2.83	3.17
	Dhani Mansukh	8.02	8.26	8.14	3.00	3.14	3.07
	Dulheri	9.76	9.86	9.81	3.90	4.01	3.96
	Tosham Rural	9.47	8.33	8.90	3.71	3.13	3.42
	Khanak	9.46	10.77	10.12	3.56	4.36	3.96
Average				9.14			3.50
Mahendragarh	Patharwa	6.28	7.81	7.05	2.12	2.89	2.51
	Dalanwas	8.77	9.55	9.16	3.33	3.73	3.53
	Khatod	10.94	10.14	10.54	4.52	3.07	3.80
	Beri	8.60	6.03	7.32	3.30	2.01	2.66
	Jat Guwana	13.06	9.39	11.23	5.54	3.67	4.61
	Bawania	6.15	6.21	6.18	2.03	2.09	2.06
	Jhagroli	8.33	5.96	7.15	3.15	2.97	3.06
	Kheri Talwana	10.86	10.84	10.85	4.44	4.38	4.41
	Baghot	8.97	7.55	8.26	3.49	2.79	3.14
	Siana	12.83	10.02	11.43	3.39	3.96	3.68
Average				8.91			3.34
Charkhi Dadri	Jhojhu Kalan	11.01	12.50	11.76	4.50	5.20	4.85
	Ladawas	9.61	7.65	8.63	3.81	2.86	3.34
	Sehlanga	5.01	6.75	5.88	2.51	2.35	2.43
	Makrana	10.76	8.19	9.48	4.38	3.09	3.74
	Dadri	8.84	8.25	8.55	3.43	3.10	3.27
	Badhra	8.47	7.11	7.79	3.21	2.49	2.85
	Atela Khurd	8.05	11.90	9.98	2.98	4.94	3.96
	Charkhi	10.86	9.06	9.96	4.48	3.52	4.00
	Dohki	8.05	6.38	7.22	3.02	2.17	2.60
	Birhi Kalan	9.52	10.06	9.79	3.80	4.04	3.92
Average				8.90			3.49

and 3.50%), while the minimum root rot incidence was recorded in Hisar district (8.65%), and the lowest root rot intensity was observed in Mahendragarh district (3.34%). Similar findings were reported by Khaire *et al.* (2020) who found that the average occurrence of root rot incidence in mungbean was highest in Jalna (28.33%) and Aurangabad (22.24%) districts, with a range of (7.34 to 40.00%) and (3.18 to 40.00%), respectively. Similarly, the percent disease intensity ranged from 3.46 to 33.24% in Jalna and 1.23 to 35.56% in Aurangabad district. Root rot disease was more prevalent in sandy soil, followed by clay loam soil, and least prevalent in clay soil. The disease incidence was higher in areas where farmers used susceptible varieties without seed treatment and in regions where irrigation was less frequent, as reported by Mallaiah and Rao (2016) and Karibasappa *et al.* (2018). These earlier findings support the present study's results.

The current investigation highlights the significant damage caused by dry root rot, which affects mungbean crops across various soil types and cultivars. AM fungi helps host crops in biotic and abiotic stressful situations by mediating complicated communication events between the plant and the fungus. AM fungi may boost plant growth and development by altering important hormonal pathways that minimize the negative impacts of stress.

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