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OCCURRENCE AND DISTRIBUTION OF CHARCOAL ROT DISEASE AND SEED MYCOFLORA IN SOYBEAN CROPPING SYSTEMS OF TELANGANA STATE, INDIA

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ABSTRACT

Charcoal rot, caused by *Macrophomina phaseolina* (Tassi) Goid., is a major soil-borne disease limiting soybean (*Glycine max*) productivity in India. A roving survey was conducted during Kharif 2024 across major soybean-growing districts of Telangana to assess disease incidence, seed health, and seed quality parameters. The overall mean disease incidence was 15.3%, with the highest incidence recorded in Adilabad Rural (36.9%). The disease was more severe in sandy loam and sandy clay loam soils, especially under monocropping systems and moisture-stressed conditions. Seed health testing of 120 samples revealed the highest infection in Adilabad (35.7%), followed by Nirmal (30.69%), Kamareddy (29.91%), Nizamabad (28.8%) and Sangareddy (25.4%). *M. phaseolina* (2.7–3.21%) was the predominant pathogen, followed by *Fusarium* spp., *Colletotrichum truncatum*, and *Alternaria* spp., while *Aspergillus* spp. occurred at low levels. The highest germination and SVI-I were recorded in Nirmal and Sangareddy (73% and 6188.2), while Nirmal showed maximum SVI-II (1113.4). Adilabad samples exhibited the lowest seed quality parameters, likely due to higher pathogen load and moisture stress during harvest. The findings highlight the need for integrated disease management and improved seed systems for sustainable soybean production in Telangana.
Keywords **:** charcoal rot, *Macrophomina phaseolina*, seedborne pathogens, seed quality, Soybean, Telangana.

Introduction

Soybean (*Glycine max* L. Merrill) is a vital legume crop valued for its high protein (36–42%) and oil (18–20%) content. Telangana has emerged as a key soybean-producing state in India, yet the crop faces severe yield constraints due to biotic and abiotic stresses, particularly charcoal rot disease caused by *Macrophomina phaseolina*. This necrotrophic fungus, with a broad host range exceeding 500 species, infects roots and basal stems, often under high-temperature and drought-prone conditions, causing premature plant death and severe yield loss (Wrather *et al.,* 2010; Gupta *et al.,* 2012; Sagarika *et al.,* 2023).

Despite its widespread occurrence, systematic data on the field-level incidence and seedborne association of *M. phaseolina* in Telangana remains limited. This study was therefore undertaken to assess the incidence and distribution of charcoal rot in different agro-climatic zones of Telangana, characterize the associated seed mycoflora and evaluate seed health and quality status using standard protocols.

Materials and Methods

Field Survey

A roving survey was conducted during *Kharif* 2024 in five major soybean-growing districts: Adilabad, Nirmal, Nizamabad, Kamareddy and Sangareddy. A total of 57 fields spanning 11 villages were surveyed at the R_7 physiological maturity stage.

Disease incidence was recorded from five randomly selected 1m² quadrats per field. Per cent Disease Incidence (PDI) was calculated using the formula:

$$\text{PDI}(\%) = \frac{\text{No. of diseased plants}}{\text{Total number of plants observed}} \times 100$$

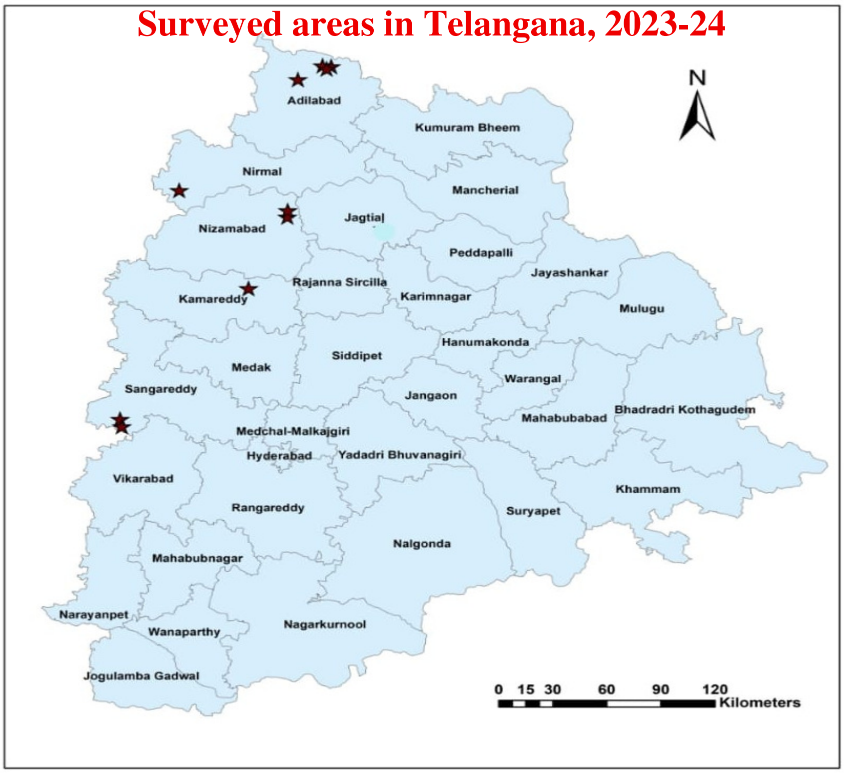

Fig. 1 : Map showing sample collection sites in different soybean growing districts of Telangana during *Kharif*- 2023-24

Seed Collection and Health Testing

A total of 120 freshly harvested soybean seed samples were collected from the same surveyed locations and tested at the lab using Standard Blotter Method (ISTA, 2022) for mycoflora detection and Paper towel method for seed germination and vigour index assessment. Seed borne fungi were identified morphologically and recorded in percentage terms.

Seed health testing methods:

Standard blotter method (ISTA, 2022)

Three sterilized blotter paper discs of 9 cm diameter were placed in sterile petri plates and moistened with sterile distilled water. The excess water was drained off from the plates. Ten soybean seeds were placed at equidistance with nine seeds in the periphery and one in the centre. The plates were incubated at 25 ± 2 ºC for 7 days under alternate cycles of 12hrs light and 12 hrs darkness in the BOD incubator. Using the stereo binocular microscope, the seeds were examined on the seventh day of incubation. The total number and type of fungal colonies were recorded and expressed in percentage. Four replications were maintained for each seed sample.

$$\text{Seed Infection}(\%) = \frac{\text{No. of seeds infected by fungi}}{\text{Total no. of seed in each plate}} \times 100$$

Seed quality testing methods:

Rolled paper towel method (ISTA, 2022)

Germination: The paper towels were initially soaked in sterile distilled water. After draining out excess water, one moistened paper towel was placed on sterile platform. One hundred seeds from each seed sample were randomly taken and placed on it at equidistance spacing and covered with another moistened paper towel and rolled. The rolled paper towels were placed vertically in cabinet of seed germinator and maintained constant temperature of 25 ±10 ºC and relative humidity of 95±2 per cent. Four replications for each seed sample were maintained. The germination percentage was recorded on the 8th day based on normal seedlings. The percent seed germination was calculated as per the following formulae.

$$\text{Germination}(\%) = \frac{\text{Number of normal seedlings}}{\text{Total number of seeds}} \times 100$$

Seedling vigour index (SVI-I): In each sample, ten seedlings were selected randomly for measuring the seedling length. The shoot length was measured from the cotyledonary node to the tip of the apical bud. The root length was measured from the cotyledonary node to the tip of the primary root. The mean root and shoot lengths were expressed in centimeters (cm).The seedling vigour index-I was calculated as per the formula suggested by Abdul and Anderson (1973).

Seedling vigour index (SVI-I) = Germination (%) x Total seedling length (cm).

Seedling vigour index (SV-II): The randomly selected ten seedlings from each seed sample were allowed for drying in hot air oven for about 24hrs at a temperature of 80°C. The seedling vigour index II is computed by following the method suggested by Abdul Baki and Anderson (1973) and calculated as per the formula given below and expressed in whole number.

SVI- II = Germination (%) x Seedling dry weight (g)

Results and Discussion

Disease Incidence in Fields

Charcoal rot was observed in all surveyed districts, with significant variation in incidence across all the locations (Table 1). The disease incidence ranged from 2.5 to 36.9 per cent, with an overall mean of 15.39 per cent. Highest incidences were recorded in Adilabad Rural (A4: 36.9%, A5: 33.4%, A2: 32.5%), while the lowest was seen in Jainad- J5(2.5%), J2(3.4%). District-wise mean incidence was: Adilabad (17.5%) > Nizamabad (14.6%) > Kamareddy (13.3%)> Sangareddy (12.9%) > Nirmal (11.2%).

Disease severity was highest around 70–75 DAS, especially during pod-setting and maturity stages. Fields with sandy loam and sandy clay loam soils, and those with poor crop rotation practices (especially Bengal gram or cotton as preceding crops), reported the highest disease severity. Similar findings were reported by Wrather *et al.* (1997), Gupta and Chauhan (2005), and Amrate *et al.* (2024) in other Indian states.

Seed Health and Seed Quality Analysis

A total of 120 soybean seed samples collected from the *Kharif*, 2024 harvest across five districts *viz.,* Adilabad, Nirmal, Nizamabad, Kamareddy and Sangareddy were evaluated for seed health and seed quality. The analysis was conducted using standard protocols laid out by the (ISTA, 2022).

Per cent Seed Infection

The per cent seed infection determined using the Standard Blotter Method, varied significantly among districts. The overall mean seed infection across samples was 30.16%. The highest infection rate was observed in Adilabad (35.7%), followed by Nirmal (30.69%), Kamareddy (29.91%), Nizamabad (28.8%) and the lowest in Sangareddy (25.4%) (Table 2).

The most prevalent pathogens were *M. phaseolina* (2.7–3.21%), *Fusarium sp.*(1.76–2.9%), *C. truncatum* (0.75–0.90%) and *Alternaria* (<0.8%). Storage fungi like *Aspergillus*, present in lower percentage, indicating good post-harvest handling but their potential to increase under improper storage conditions. These results align with those reported by Rao *et al.* (2015) and Iranna *et al.* (2021), who observed *M. phaseolina*, *F. oxysporum*, and *C. truncatum* as major seedborne fungi in soybean seeds across Telangana.

Seed Germination and Seedling Vigour I and II

Germination percentage and Seedling Vigour Index-I and II were evaluated using the rolled paper towel method. The results are presented in Table 3. The highest germination and SV-I were recorded in Nirmal and Sangareddy district (73% and 6188.2), respectively and SV-II in Nirmal(1113.4), despite a moderately high seed infection percentage, possibly due to higher seed viability and better harvest management. Adilabad showed the lowest germination (51%), SVI-I (4456.32) and SV-II (828), likely due to higher infection by *M. phaseolina* and increased moisture stress during harvest.

Seedling vigour-I positively correlated with germination percentage and negatively with total seed infection, confirming the adverse impact of seedborne pathogens on seed quality. These trends are supported by findings from Singh *et al.* (2016), Soesanto *et al.* (2020) who emphasized that both biotic (pathogen load) and abiotic (harvest conditions) factors play a critical role in seed viability and quality, where the increased per cent seed infection, significantly decreased the germinability due to number of seed associated mycoflora of soybean seeds and they have concluded that pathogen infestation significantly reduces seed viability and vigour.

Table 1 : Collection of charcoal rot diseased samples from soybean growing fields of Telangana state during 2024

S. No.	Sample ID	District	Mandal	Village	Latitude Nº	Longitude Eº	Soil type	Previous crop grown	Variety	Stage of the crop (DAS)	% disease incidence
1	SP1	Sangareddy	Zahirabad	Shaikapur	17.6041	77.62173	Sandy loam	BG, M	Karishma, Vikranth	75 DAS	**19.70** 25.935*
2	SP2	Sangareddy	Zahirabad	S9haikapur	17.6052	77.6224	Sandy loam	BG, M	Karishma, Vikranth	75 DAS	**19.4** 26.103*
3	SP3	Sangareddy	Zahirabad	Shaikapur	17.6043	77.6218	Sandy loam	BG, M	Karishma, Vikranth	75 DAS	**19.4** 26.122
4	SP4	Sangareddy	Zahirabad	Shaikapur	17.6067	77.6256	Sandy loam	BG, M	Karishma, Vikranth	75 DAS	**10.4** 18.831
5	SP5	Sangareddy	Zahirabad	Shaikapur	17.6036	77.6248	Sandy loam	BG, M	Karishma, Vikranth	75 DAS	**13.8** 21.734
6	GP1	Sangareddy	Zahirabad	Gotigarpally	17.5606	77.63196	Sandy clay loam	BG, C	Omkar, DSB-34	75 DAS	**17.9** 24.987
7	GP2	Sangareddy	Zahirabad	Gotigarpally	17.57437	77.65737	Sandy clay loam	BG, C	Omkar, DSB-34	75 DAS	**13.4** 21.460
8	GP3	Sangareddy	Zahirabad	Gotigarpally	17.5758	77.65712	Sandy clay loam	BG, C	Omkar, DSB-34	75 DAS	**22.4** 28.206
9	GP4	Sangareddy	Zahirabad	Gotigarpally	17.5629	77.63204	Sandy clay loam	BG, C	Omkar, DSB-34	75 DAS	**11.7** 19.947
10	GP5	Sangareddy	Zahirabad	Gotigarpally	17.5634	77.63187	Sandy clay loam	BG, C	Omkar, DSB-34	75 DAS	**12.8** 20.962
11	ML1	Nirmal	Modhul	Modhul	18.97661	77.90657	Sandy loam	BG,C	Heeramoti, Malavika	70 DAS	**16.8** 24.187
12	ML2	Nirmal	Modhul	Modhul	18.97756	77.90684	Sandy loam	BG,C	Heeramoti, Malavika	70 DAS	**9.2** 17.652
13	ML3	Nirmal	Modhul	Modhul	18.97651	77.90712	Sandy loam	BG,C	Heeramoti, Malavika	70 DAS	**12.2** 20.403
14	ML4	Nirmal	Modhul	Modhul	18.97688	77.90623	Sandy loam	BG,C	Heeramoti, Malavika	70 DAS	**12.7** 20.833
15	ML5	Nirmal	Modhul	Modhul	18.97714	77.90698	Sandy loam	BG,C	Heeramoti, Malavika	70 DAS	**4.9** 12.828
16	MR1	Nizamabad	Morthad	Morthad	18.826766	78.477818	Sandy clay loam	BG,C	JS-9305, JS-335	65 DAS	**12.0** 20.273
17	MR2	Nizamabad	Morthad	Morthad	18.826666	78.477801	Sandy clay loam	BG,C	JS-9305	65 DAS	**7.3** 15.671
18	MR3	Nizamabad	Morthad	Morthad	18.826624	78.477795	Sandy loam	BG,C	JS-335	65 DAS	**8.7** 17.174
19	MR4	Nizamabad	Morthad	Morthad	18.826709	78.477809	Sandy clay loam	BG,C	JS-335	65 DAS	**7.5** 15.928
20	MR5	Nizamabad	Morthad	Morthad	18.824376	78.474781	Sandy clay loam	BG,C	JS-9305	65 DAS	**17.2** 24.507
21	T1	Nizamabad	Morthad	Thimmapur	18.864657	78.479215	Sandy clay loam	BG,C	JS-9305, Karishma	65 DAS	**20.7** 27.023
22	T2	Nizamabad	Morthad	Thimmapur	18.864646	78.479221	Sandy clay loam	BG,C	Karishma	65 DAS	**12.0** 20.180
23	T3	Nizamabad	Morthad	Thimmapur	18.86459	78.479165	Sandy clay loam	BG,C	JS-335	65 DAS	**12.6** 20.767
24	T4	Nizamabad	Morthad	Thimmapur	18.864663	78.479322	Sandy clay loam	BG,C	Karishma	65 DAS	**15.6** 23.250
25	T5	Nizamabad	Morthad	Thimmapur	18.86467	78.47932	Sandy clay loam	BG,C	JS-335	65 DAS	**8.6** 17.007
26	V1	Nizamabad	Velpur	Velpur	18.763126	78.381698	Sandy clay/clay	BG,C	JS-335, Greengold-3344	70 DAS	**15.2** 22.893
27	V2	Nizamabad	Velpur	Velpur	18.763028	78.381711	Sandy clay	BG,C	JS-335, Unnath	70 DAS	**14.4** 22.307
28	V3	Nizamabad	Velpur	Velpur	18.763043	78.381712	Sandy clay	BG,C	Omkar	70 DAS	**19.7** 26.352
29	V4	Nizamabad	Velpur	Velpur	18.763159	78.381683	Sandy clay loam	BG,C	Unnath	70 DAS	**20.6** 27.011

30	V5	Nizamabad	Velpur	Velpur	18.7631	78.381749	Sandy clay loam	BG,C	Omkar	70 DAS	**24.6** 29.702
31	A1	Adilabad	Adbd Rural	Rampur	19.647876	78.52066	Sandy clay loam	BG, J	JS-335, Unnath	75 DAS	**31.8** 34.342
32	A2	Adilabad	Adbd Rural	Rampur	19.647783	78.520582	Sandy clay loam	BG, J	JS-335, Unnath	75 DAS	**32.5** 34.711
33	A3	Adilabad	Adbd Rural	Rampur	19.647798	78.520556	Sandy clay loam	BG, J	JS-335	75 DAS	**30.0** 33.198
34	A4	Adilabad	Adbd Rural	Rampur	19.648164	78.520375	Sandy loam	BG, J	JS-335, Unnath	75 DAS	**36.9** 37.369
35	A5	Adilabad	Adbd Rural	Rampur	19.648826	78.51938	Sandy loam	BG, J	Omkar, Malavika	75 DAS	**33.4** 35.281
36	A6	Adilabad	Adbd Rural	Rampur	19.647898	78.520077	Sandy clay loam	BG, J	Unnath	75 DAS	**25.7** 30.413
37	A7	Adilabad	Adbd Rural	Rampur	19.647883	78.520125	Sandy clay loam	BG, J	Unnath	75 DAS	**20.1** 26.638
38	A8	Adilabad	Adbd Rural	Rampur	19.647872	78.520207	Sandy clay loam	BG, J	Unnath, Karishma	75 DAS	**24.0** 29.349
39	A9	Adilabad	Adbd Rural	Rampur	19.647832	78.520267	Sandy clay loam	BG, J	Unnath, Karishma	75 DAS	**32.1** 34.466
40	A10	Adilabad	Adbd Rural	Rampur	19.648602	78.51911	Sandy clay loam	BG, J	Unnath, Karishma	75 DAS	**24.1** 29.365
41	B1	Adilabad	Jainad	Belgum	19.72791	78.69642	Sandy loam	BG, C	Karishma, Vikranth	75 DAS	**18.6** 25.562
42	B2	Adilabad	Jainad	Belgum	19.72756	78.69694	Sandy loam	BG, C	Vikranth	75 DAS	**14.9** 22.635
43	B3	Adilabad	Jainad	Belgum	19.72748	78.69652	Sandy loam	BG, C	Vikranth	75 DAS	**19.4** 26.151
44	B4	Adilabad	Jainad	Belgum	19.72783	78.69685	Sandy loam	BG, C	JS-335	75 DAS	**0.0** 0.000
45	B5	Adilabad	Jainad	Belgum	19.72737	78.69673	Sandy loam	BG, C	Karishma	75 DAS	**10.3** 18.486
46	J1	Adilabad	Jainad	Jainad	19.73259	78.65059	Sandy loam	BG, C	Malavika, Karishma	75 DAS	**0.0** 0.000
47	J2	Adilabad	Jainad	Jainad	19.73264	78.65024	Sandy clay loam	BG, C	Karishma	75 DAS	**3.4** 10.144
48	J3	Adilabad	Jainad	Jainad	19.73242	78.65045	Sandy clay loam	BG, C	Karishma	75 DAS	**0.0** 0.000
49	J4	Adilabad	Jainad	Jainad	19.73263	78.65069	Sandy clay loam	BG, C	Vikranth	75 DAS	**4.1** 11.665
50	J5	Adilabad	Jainad	Jainad	19.73245	78.65038	Sandy loam	BG, C	karishma	75 DAS	**2.5** 7.416
51	K1	Kamareddy	Sadasivnagar	Sadasivnagar	18.39581	78.28001	Sandy loam	BG,C	Tejaswini, Greengold-3344	65 DAS	**11.7** 19.877
52	K2	Kamareddy	Sadasivnagar	Sadasivnagar	18.39581	78.28001	Sandy loam	BG,C	Karishma	65 DAS	**16.1** 23.602
53	K3	Kamareddy	Sadasivnagar	Sadasivnagar	18.39581	78.28001	Sandy loam	BG,C	Greengold-3344	65 DAS	**11.5** 19.678
54	K4	Kamareddy	Sadasivnagar	Sadasivnagar	18.39581	78.28001	Sandy loam	BG,C	Karishma	65 DAS	**15.0** 22.586
55	K5	Kamareddy	Sadasivnagar	Sadasivnagar	18.39581	78.28001	Sandy loam	BG,C	Tejaswini	65 DAS	**12.5** 20.507
56	Mk2	Adilabad	Jainad	Makoda	19.71332	78.67214	Sandy loam	BG, J	Omkar	75 DAS	**5.7** 11.330
57	Mk2	Adilabad	Jainad	Makoda	19.71346	78.67257	Sandy loam	BG, J	JS-335	75 DAS	**5.5** 11.126
CD (P=0.05)											**4.213**
Se (m)											**1.500**
Se (d)											**2.122**
C.V.											**11.959**

Table 2 : Seed mycoflora associated with soybean seed samples using standard blotter method

S. No.	Location / District	No. of seed samples	Seed Infection (%)	Seed mycoflora/ Pathogens observed (%)					
				M. phaseolina	*Fusarium sp.*	*C. truncatum*	*Alternaria sp.*	*Aspergillus sp.*	Mean
1.	Adilabad	25	35.70 (36.66)*	2.89 (9.77)*	1.82 (7.72)*	0.87 (5.291)*	0.44 (3.78)*	0.35 (3.35)*	1.27 (6.02)*
2.	Nirmal	20	30.69 (33.63)	2.7 (9.38)	2.3 (7.85)	0.9 (5.3)	0.8 (4.9)	0.5 (4.3)	1.44 (6.503)
3.	Nizamabad	30	28.8 (32.46)	3.21 (10.33)	1.76 (7.6)	0.75 (4.9)	0.63 (4.52)	0.63 (4.52)	1.40 (6.401)
4.	Sangareddy	25	25.4 (30.3)	3.04 (10.03)	2.03 (8.12)	0.82 (5.12)	0.43 (3.76)	0.51 (4.09)	1.37 (6.25)
5.	Kamareddy	20	29.91 (33.3)	3.12 (10.14)	2.9 (8.12)	0.83 (5.16)	0.73 (3.76)	0.58 (4.09)	1.62 (6.892)
	Total / mean	120	30.16 (33.3)	3.02 (6.015)	1.88 (6.503)	0.84 (6.401)	0.56 (6.254)	0.52 (6.312)	
			C.V.: 1.718	C.V. : 10.79					
			CD (P=0.05) : 0.76	CD (P=0.05) Districts :0.54 Pathogen :0.59 Interaction :0.99					

Table 3 : Seed quality parameters of soybean seed samples using paper towel method.

S.No.	District	No. of samples	Germination %	Seedling Mortality	Seedling Vigour-I	Seedling Vigour-II
1.	Adilabad	25	51.00 (45.7)*	8.31 (16.78)*	4456	828
2.	Nirmal	20	73.00 (58.69)	6.32 (14.53)	5587	1113
3.	Nizambad	30	55.00 (47.87)	10.12 (18.49)	5306	1025
4.	Sangareddy	25	59.00 (50.18)	6.612 (14.94)	6188	1015
5.	Kamareddy	20	53.00 (46.72)	7.13 (15.54)	5316	943
	Total/Mean	120	56.4 (48.68)	7.7 (16.03)	5370	985
	CV		1.174	2.01	1.79	2.105
	CD(P= 0.05)		1.040	18.45	177.06	38.22

Conclusion

A roving survey was conducted among different major soybean growing areas to assess the charcoal rot incidence in Telangana State. Among eleven mandals of five districts surveyed, disease incidence varied from district to district and within the district ranging from 2.5per cent to 36.9 per cent. The highest disease incidence was observed in Adilabad Rural, with a maximum of 36.9 per cent, while the overall mean incidence across surveyed fields was 15.39 per cent. The disease was more severe in sandy and sandy clay loam soils and under moisture stress conditions, particularly in fields with poor crop rotation and use of privately sourced seed.

Seed health studies using the standard blotter method identified *M. phaseolina*, *Fusarium sp.*, and *C. truncatum* as the predominant seed borne fungi, with *M. phaseolina* consistently showing the highest infection across all districts. Germination and seedling vigour indices were adversely affected in seed lots with higher levels of seed infection, confirming the negative impact of seedborne mycoflora on seed quality. An integrated approach involving timely disease monitoring, use of certified disease-free seeds, crop rotation and soil health management is crucial to mitigate the impact of charcoal rot and improve soybean productivity in the region.

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Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the views of their affiliated institutions. The authors are responsible for the accuracy and completeness of the information provided, but do not accept any liability for any direct or indirect losses resulting from the use of this content.

Informed consent

The survey conducted in this study involved collection of field and soil-related information only. No personal or identifiable human data were collected. Farmers/field owners were informed about the purpose of the study, and their voluntary cooperation was sought before accessing fields and collecting observations. Verbal consent was obtained, as no personal participation or sensitive information was required. All data were recorded anonymously and used solely for research purposes.

Conflict of interest

All the authors have declared and confirmed that they have no conflict of interest.

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