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Seedborne pathogens associated with soybean in Telangana State

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Abstract
Soybean [*Glycine max* (L.) Merill.] is an important oil seed and pulse crop grown in India. One of the major constraints that is causing significant reduction in seed quality and yield is due to seedborne diseases in soybean. It is very important to produce disease free seeds to maintain the quality and sustain the productivity of the crop. The present investigation was carried out to study the impact of seedborne mycoflora of soybean on seed quality using different health testing methods. In the present study, a total of eighty-six soybean seed samples were collected during *kharif* 2020-21 from the four major soybean growing districts of Telangana state *viz.,* Adilabad, Nizamabad, Kamareddy and Nirmal. All the seed samples were tested for seed quality parameters using rolled paper towel method, in which the seed samples of Nirmal district recorded significantly highest (74.50 and 1174) per cent seed germination and seedling vigour index-I (SVI-I) which was above IMSCS followed by Kamareddy (67.36 and 987), Adilabad (61.99 and 906) and Nizamabad (58.99 and 816). Seed associated mycoflora among the soybean seed sample were found to significantly reduce the seed germination and vigour. All the soybean seed samples were subjected to standard seed health testing methods (ISTA, 1996) *viz.,* standard blotter, agar plate, deep freeze and test tube agar methods to detect and isolate the mycoflora associated with seed samples. Of the four methods, standard blotter method was found superior in recovering the seed mycoflora over the other test methods. The fungi recovered from the seed health methods include *Colletotrichum truncatum, Macrophomina phaseolina, Fusarium* sp., *Alternaria* sp., *Phomopsis* sp., and storage fungi like *A. flavus* and *A. niger*. *Fusarium* sp. was the predominantly found in all the districts followed by *M. phasoina* while *Phomopsis* sp. was found least.

Keywords: *Kharif*, per cent germination, mycoflora, vigour, IMSCS

Introduction
Soybean [*Glycine max* (L.)] Merill. is a very important oil seed crop throughout the world. It is very rich source of protein for human beings as well as feed for livestock. This crop can be grown in tropical, sub-tropical as well as the temperate regions. It is primary source of vegetable oil and protein concentrates. It is an excellent source of major nutrients, about 40% of dry matter is protein and 20% fat (Lakshmeesha *et al*., 2013) [5]. Soybean protein is rich in valuable amino acid lysine (5%) which is deficit in most of the cereals. In addition to this it also contains a good amount of minerals, salts and vitamins (thiamine and riboflavin) and is cheapest source of protein, hence called 'poor man's meat. Its sprouting grains contain a considerable amount of vitamin A and vitamin C, which is present in the form of precursor carotene. Its oil is used for manufacturing vanaspati ghee, biodiesel and several other industrial products.

In soybean cultivation the major constraints is seed borne pathogens. Seed germination in soybean has considerably reduced due to presence of seedborne diseases. Seed borne pathogens have been involved in seed rots during germination and seedling mortality leading to poor crop stand reduction in plant growth and productivity of crops (Akranuchat *et al.,* 2007) [2]. Infected seeds also play major role in the establishment of economically important plant diseases in the field resulting in heavy reduction of crop yields. It has been reported that more than 100 plant pathogens affect soybean, several economically important diseases have been reported. Among them *Phakopsora pachyrhizi*, *Cercospora kikuchii*, *Colletotrichum*, *Fusarium* and *Macrophomina* are some of the very important. It also include *Alternaria*, *Cladosporium* and *Penicillium* which are secondary invaders on the injured pods. Thus the present review outlines and discusses the important seedborne diseases of soybean and their related aspects for better understanding of the issue.

Material and Methods

The present investigation was carried out in laboratory Seed Research and Technology Centre, Rajendranagar, Hyderabad during 2020-21. A total of eighty six soybean seed samples were collected from *kharif* harvested from major soybean growing districts of Telangana. (Table 1)

Table 1: List of seeds samples collected from major districts of Telangana

S. No.	Name of Districts	Number of seed samples collected
1.	Adilabad	30
2.	Nizamabad	13
3.	Kamareddy	14
4.	Nirmal	29
	Total	86

Seed quality and health testing methods

The seed quality parameters *i.e.*, germination, seedling vigor index-I and seed health parameters *i.e.*, seed infection, type and number of mycoflora associated with the seed samples were recorded by subjecting the seed samples to standard detection methods (ISTA, 1996) [4]. Four hundred seeds from each sample were evaluated for testing all the parameters and the detection methods used in the study. The following parameters were recorded.

Seed quality testing methods

Rolled paper towel method

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 ±10C 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 per cent 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) = Total seedling length (cm) x germination (%).

Seed health testing methods: Standard blotter method

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 ± 20C for 7 days under alternate cycles of 12 hrs 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.

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

Results and Discussion

Studies on seed quality parameters

A total of 86 soybean seed samples were collected from major soybean growing districts of Telangana state *viz.* Adilabad, Nizamabad, Kamareddy and Nirmal during *Kharif* 2020-21 at the time of harvest. The collected seed samples were subjected to the seed quality parameters using rolled paper towel method (ISTA, 1996) [4] for determining per cent seed germination and seedling vigour index (I).

In the present study, significant differences were observed in the seed quality parameters among the seed samples collected from different districts. The per cent seed germination ranged from 58.99 to 74.50. Among the districts, highest per cent seed germination was observed in Nirmal district (74.50%) which is above Indian Minimum Seed Certification Standard (IMSCS) i.e. >70 per cent and lowest was recorded in Nizamabad district (58.99%). While the districts Adilabad and Kamareddy recorded 61.99 and 67.36 per cent seed germination, respectively (Table 2).

Irrespective of the locations, the study also showed a significant difference in Seedling Vigour Index-I (SVI-I) among the seed samples of different districts which ranged from 816 to 1174. Maximum SVI-I of 1174 was exhibited by the seed samples from Nirmal and minimum of 816 was observed in Nizamabad district. However, Adilabad and Kamareddy districts recorded (SVI-I) of 906 and 987, respectively.

The study revealed that the soybean seed samples with the highest (74.50%) per cent germination recorded high (SVI-I) of 1174 and samples with the lowest (58.99%) per cent seed germination recorded least 816 (SVI-I).

Table 2: Seed germination (%) and Seedling Vigour Index-I of soybean seed samples

District	No. of Samples	Germination (%)	Seedling Vigour Index-I
Adilabad	30	61.99 (51.94)	906
Nizamabad	13	58.99 (50.18)	816
Kamareddy	14	67.36 (55.16)	987
Nirmal	29	74.50 (59.67)	1174
Total/Mean	86	65.71 (54.24)	970.93
		CV-1.87 CD@ 5%-1.90	CV-8.60 CD@ 5%-130.15

The lowest per cent germination of seed samples from Nizamabad district may be due to onset of maximum rainfall at the time of crop maturity/harvest stage. This might have increased moisture levels in seed and have further aided infections with seedborne mycoflora and that must have been affected the germination. The findings were in confirmatory with Penfield and MacGregor (2017) [7] who stated the percentage of seed germination has been affected by many factors including pathogen attack, genetic factors and the environment. Also the results are in agreement with Soesanto *et al.* (2020) that the increased percentage seed infection,

significantly decrease the germinability due to number of seed associated mycoflora of soybean seeds. Also with Ghangaokar and Kshirsagar (2013) [3] who stated that the seedborne microbes decreases seed germination capacity and plant vigor in soybean. Further the results of Venugopal (2013) [11], Ramesh *et al*. (2013) and Neelothpala (2018) [6] are in agreement with the present findings.

Studies on seed health parameters

Detection of seedborne pathogens associated with soybean seed samples

The collected soybean seed samples were subjected to standard seed health testing methods (ISTA 1996) [4] using standard blotter method. The seed borne pathogens that were found associated with seed samples were isolated, identified and purified for further studies.

Detection of seedborne pathogens following standard blotter method

The seedborne mycoflora found detected with standard blotter method was given in the table 3. In the present study, significant differences were observed in the recovery of seedborne mycoflora from the collected seed samples. The results indicated that, a total of 7 fungal species belonging to 6 genera were found associated with the samples tested for seed health. The mycoflora identified include *Colletotrichum* sp., *Macrophomina* sp., *Fusarium* sp., *Alternaria* sp., *Phomopsis* sp., *Aspergillus flavus* and *A. niger*. Of the seed samples tested, the per cent seed infection ranged from 0.13 to 18.29. It was observed that, the highest mean per cent seed infection was recorded from Nizamabad (5.62) district and lowest with Nirmal (4.19) district. The districts, Adilabad and Kamareddy recorded 5.43 and 4.50 mean per cent seed infection, respectively.

The results also revealed that out of the seven fungal genera, the most predominantly occurring fungi was *Fusarium* sp. followed by *Macrophomina* sp., while the fungi *Alternaria* sp. and *Phomopsis* sp. recorded were low from the samples tested. It was noticed that, irrespective of the locations, occurrence of *Fusarium* sp. ranged from 11.19 to 18.29 per cent with highest in Nizamabad district (18.29%) followed by Adilabad (16.76%) and lowest in Nirmal district (11.19%). In case of *Macrophomina* sp. the per cent seed infection ranged from 7.65 to 12.26 per cent where its infection was found maximum in Nizamabad district (12.26%) followed by Kamareddy district (10.29%) with lowest in Nirmal district (7.65%). The per cent infection of *Colletotrichum* sp. ranged from 0.48 to 3.59 per cent with highest infection in Adilabad district (3.59%) followed by Nizamabad (0.81%) and lowest in Nirmal district (0.48%). While the per cent infection of *Alternaria* sp. and *Phomopsis* sp. have recorded in a range of 0.21 to 0.97 per cent and 0.13 to 1.23 per cent, respectively. Whereas, the occurrence of storage fungi *viz. A. flavus* and *A. niger* ranged from 3.61 to 6.89 per cent and 0.36 to 2.60 per cent, respectively. However, the per cent infection of *Alternaria* sp. and *Phomopsis* sp. was found to be low compared to the other fungi in the study (Table 3).

The findings of the present study were in similarity with earlier results of Sewedy *et al.* (2019) [9] who isolated nineteen fungal species comprising 13 genera *i.e. C. dematium, F. solani, F. moniliforme, F. oxysporum, Alternaria* sp., *Aspergillus niger, Aspergillus flavus, Botryodiplodia* sp., *Cladosporium* sp., *Colletotrichum lindemuthianum, Macrophomina phaseolina, Alternaria alternata, Aspergillus ochraceus, Botryodiplodia*.

Table 3: Seed health status of soybean seed samples using Standard blotter method

Districts	Pathogens observed/Per cent Seed Infection (%)							
	C. truncatum	***M. Phaseolina***	***Fusarium* sp.**	***Alternaria* sp.**	***Phomopsis* sp.**	***A. flavus***	***A. niger***	**Mean**
Adilabad	3.59 (10.92)	8.63 (17.08)	16.76 (24.17)	0.60 (4.44)	0.13 (4.01)	5.79 (13.92)	2.50 (9.11)	5.43
Nizamabad	0.81 (5.15)	12.26 (20.49)	18.29 (25.32)	0.54 (4.25)	1.23 (6.37)	3.61 (10.92)	2.60 (9.27)	5.62
Kamareddy	0.52 (4.20)	10.29 (18.70)	14.38 (22.28)	0.21 (4.06)	0.00 (4.06)	5.79 (13.90)	0.36 (4.25)	4.50
Nirmal	0.48 (4.12)	7.65 (16.05)	11.19 (19.54)	0.97 (5.64)	0.19 (4.06)	6.89 (15.21)	2.00 (8.12)	4.19
Mean	1.35 (6.10)	9.70 (18.08)	15.15 (22.82)	0.58 (4.63)	0.39 (4.59)	5.52 (13.49)	1.86 (7.69)	
	CV-5.57 CD@5%-Districts: 0.15 Pathogens: 0.19 Interaction: 0.39							

sp., *Myrothecium* sp., *Penicillium* sp., *Rhizoctonia solani, Stemphylium* sp., *Trichoderma* sp. and *Trichothecium* sp. from soybean and common bean seed samples using standard blotter. The mycoflora *Macrophomina phaseolina, Fusarium oxysporum, A. flavus, A. niger, Phoma* sp. and *Sclerotinia sclerotiorum, F. solani, F. moniliforme, Rhizopus* S., *Botrytis cinerea* and *Cercospora kikuchii.* (Ramesh *et al*. 2013). The soybean seed mycoflora *viz. Colletotrichum dematium, Macrophomina Phaseolina, Fusarium oxysporum A. niger, A flavus* and Purple seed stain (Harne *et al.* 2017).

Conclusion

Of the soybean seed samples collected from major soybean growing districts of Telangana state, maximum number of seedborne mycoflora with reduced seed germination and seedling vigour was observed in Nizamabad district followed by Adilabad, Kamareddy and Nirmal districts. Fungal genera *C. truncatum, M. phaseolina, Fusarium* sp., *Phomopis* sp., *Alternaria* sp., *A. flavus* and *A. niger* were the mycoflora associated with soybean seeds. Among the different seed health testing methods, standard blotter method was found superior in recovering higher number mycoflora.

References

1. Abdul Baki AA, Anderson JD. Relationship between decarboxylation of glutamic acid and vigour in soybean (*Glycine max* L.). Crop Science. 2013;13:227-232.
2. Akranuchat P, Noimanee P, Krittigamas N, Horsten DV, Vearasilp S. Control of seed borne fungi by radio frequency heat treatment as alternative seed treatment in barely (*Hordeum vulgare*). In Conference on International Agricultural Research for Development, 2007, 9-11.

3. Ghangaokar NM, Kshirsagar AD. Study of seed borne fungi of different legumes. Trends in Life Sciences. 2013;2(1):32-35.
4. ISTA (International Seed Testing Association). International rules for seed testing rules. Seed Science and Technology. 1996;13:299-513.
5. Lakshmeesha TR, Sateesh MK, Vedashree S, Mohammad SS. Antifungal activity of some medicinal plants on Soybean seed-borne Macrophomina phaseolina. Journal of Applied Pharmaceutical Science. 2013;3(2):84-87.
6. Neelothpala M. Evaluation of Soybean Genotypes against Seedborne Mycoflora and Its Management. M.sc (Ag.) Thesis. Professor Jayashankar Telangana State Agricultural University, Hyderabad, India, 2018.
7. Penfield S, MacGregor DR. Effects of environmental variation during seed production on seed dormancy and germination. Journal of experimental botany. 2017;68(4):819-825.
8. Ramesh CH, Avitha KM. Presence of external and internal seedmycoflora on sunflower seeds. Journal of Mycology and Plant Pathology. 2005;35(2):362-364.
9. Sewedy ME, Atia MM, Zayed MA, Ghonim MI. Molecular detection and controlling of seed-borne *Colletotrichum* spp. In common bean and soybean. Zagazig Journal of Agricultural Research. 2019;46(6):1919-1935.
10. Shovan LR, Bhuiyan MKA, Sultana N, Begum JA, Pervez Z. Prevalence of fungi associated with soybean seeds and pathogenicity tests of the major seed-borne pathogens. International Journal of Sustainable Crop Production. 2008;3(4):24-33.
11. Venugopal RT. Studies on seedborne fungi of soybean and its management. M.Sc. (Ag). Thesis. Acharya N.G. Ranga Agricultural University, Hyderabad, India, 2013.