REVIEW OF RESEARCH

DETECTION OF MYCOFLORA ASSOCIATED WITH PIGEONPEA SEEDS

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ABSTRACT:

The seed mycoflora of pigeonpea was carried out with the methods followed by ISTA (1985) viz. inspection of dry seeds, Seed washing test. Standard blotter method and agar plate method with potato dextrose agar which are described in "Material and Methods". Seeds of eleven varieties/lines were used, collected from farmer's fields, seed companies and pulse section of Narendra Dev University of Agriculture and Technology. Kumarganj, Faizabad (U.P) (table-1). Composite samples. Were prepared from the seed of above varieties/lines for the following studies.

KEYWORDS: *seed mycoflora , Material and Methods.*

1.(I) Inspection of dry seeds :

For this study, freshly harvested seed of pigeonpea varieties/lines (table-1) were taken randomly. The study was conducted by naked eyes and with the help of magnifying hand lens for the presence of fungal fructifications. Discolou - ration and deformities.

The result obtained are presented in table-3, it is clear from the results recorded in table that seeds if pigeonpea varieties, prabhat T21, Pant A-3 Pusa 33 and Bahar. Exhibited light to dark dicolouration at a range of 1.0 to 5.5 percent on the seed surface. However, the seeds of other varieties like; NDA, 91-2, T 7, T17, NDA 91-14, NAD 91-13 and NDA 91-1 did not show any discolouration on the seed surface, At the same time no fungal fructifications were observed no the seeds of any varieties/lines under study. Inert matter in the form of pieces of seeds, husk and leaves was found mixed with seeds. This inert matter when plated.

Table 1: Dry seed examination for discolouration and association of fungal bodies.

S. No.	Varieties/Lines	Percent Discolouration	Fungal bodles
1.	Prabhat	5.5	0
2.	T-21	5.0	0
3.	Pant A-3	3.5	0
4.	NDA 91-2	0	0
5.	T-7	0	0
6.	T-17	0	0
7.	PUSA-33	1.5	0
8.	NDA 91-14	0	0
9.	NDA 91-13	0	0
10.	Bahar	1.0	0
11.	NDA-91-1	0	0

on 2.0 percent potato dextrose agar medium after treatment with 1.0 percent chlorine solution for 10 minutes, appearance of colonies of *Alternria alternata, Curvularia lunata, Drechslera tetramera and Fusarium moniliforme* was observed. This indicated that such isolated fungi might be carried with the seed to the soil during sowing and may serve as primary source of infection.

1.(II) Seed washing test :

The pigeonpea seed from different samples were washed with sterilized distilled water. Thus obtained suspension was examined under compound microscope as per standard method followed for study. The results obtained are presented in Table 4 which clearly indicates that spores of five fungi viz. *Alternaria, Aspergillus, Curvularia, Drechslera* and *Fusarium* were found associated with seed samples of varieties/lines like Prabhat, T-21, Pant A-3, Puss 33, Bahar, NDA-91-2,T7, T17,NDA91-14, NDA 91-13 and NDA 91-1. Study also indicated that *Alternaria alternata* spores were dominant oin almost all the samples next predominant fungus was *Aspergillus* which was present in eight varieties out of eleven while five varieties viz, T 7, Bahar and NDA 91-1 showed no association of *Aspergillus, Curvularia lunata*. It was seen in the washings of only three varieties viz. prabhat, T 21 and pusa 33. The Spores of *Drechslera tetramera* was seen in the washings of varieties

TABLE 2 seed washing test for the detection of fungals speecies externally associated with pigeonpea seeds

S.No.	Fungal Species	VARIETIES HARBOURED											Total No. of Veritieties harboured
		Prabhat	T 21	PantA-3	NDA91-2	T 7	T17	Pusa33	NDA991-14	NDA19-13	Bahar	NDA91-1	
1.	*Alternaria alternata*	+	+	+	+	+	+	+	+	+	+	+	11
2.	Aspergilli	+	+	+	+	0	+	+	+	+	0	0	8
3.	Curvularia lunata	+	+	0	0	0	0	+	0	0	0	0	3
4.	Drechslera tetramera	0	0	0	0	0	0	0	0	0	0	0	1
5.	Fusarium sp.	+	+	+	+	+	0	0	0	+	0	0	6

\+ fungal species present
o fungal species absent

Only Prabhat. Macroconidia of *Fusarium* sp. Were observed in the washings of six varieties/lines out of eleven. These were Prabhat, T 21 Pant A-3, NDA 91-2, T 7, and NDA 91-13.

1. (III-a) Standard blotter method (Untreated seeds) :

Observations recorded in table 5. Indicated that twelve fungal species belonging to ten genera were recorded on the Pigeonpea seeds of different varieties with varying degree of incidence. These were *Alternaria alternata, Aspergillus flavus, A. niger, A. fumigatus, Chaetomium globosum, Curvularia lunata, Drechslera tetramera Fusarium moniliforme, Mucor sp., Penicillium oxalicum, Rhizoctonia solani and Rhizopus nigricans.*

Observations recorded in Table 5 further indicated that pigeon pea variety Prabhat was found to harbour maximum number of fungal species and their colonies followed by prabhat, T 21, Pant A-3, Pusa 33, NDA 91-13, T 7, NDA 91-1, T17, NDA 91-14,Bahar, NDA 91-2.

It is also clear from the results presented in the same table that *Alternaria alternata* was seen on the seeds of all varieties/lines in the range of 5.0 to 22.0 percent. Its incidence was maximum in Prabhat followed by T 21 and minimum on NDA 91-2 *Aspergillus* niger was the

TABLE -3 : Percent incidence of fungal species associated with seeds of different varieties of pigeonpea in standard method (No. of seed tested – 400)

S. Fungal No. Species	VARIETIES											No. of Range Vers. of in- har cedence	
	Pra-bhat	T 21	Pant A-3	NDA 91-2	T7	T17	Pusa 33	NDA 91-14	NDA 19-13	Bahar	NDA 91-1		
1.Alternaria alternata	22	13	12	5	8	7	12	6	10	6	7	11	5-22
2. *Aspergillus flavus*	12	9	9	3	0	4	5	4	8	0	0	8	3-12
3. *A. niger*	12	6	5	1	2	7	3	3	5	2	0	10	1-12
4. *A. Fumigatus*	2	3	0	1	0	0	0	0	0	0	0	3	1-3
5.Chaetomium *globosum*	5	0	0	0	0	0	0	0	0	0	0	1	1-5
6.*Curvularia Lunata*	5	3	0	0	0	0	1	0	0	0	0	3	1-5
7. *Drechslera tetramera*	7	0	0	0	0	0	0	0	0	0	0	1	1-7
8. *Fusarium moniliforme*	8	8	2	2	1	0	0	0	4	0	0	6	1-8
9. *Mucor sp.*	2	1	0	0	1	1	0	0	0	0	1	5	1-5
10. *Pencillium oxalicum*	3	0	0	0	1	1	0	0	0	0	0	3	1-3
11. *Rhizoctonia solani*	5	0	4	0	0	0	0	0	0	0	1	3	1-5
12. *Rhizopus nigricans*	3	2	0	0	0	0	0	0	0	1	0	3	1-3
Total no. of fungal species	12	8	5	5	5	5	4	3	4	3	3		

Next important fungal species recorded on 10 varieties, while its presence was recorded maximum in variety prabhat and minimum in NDA 91-2 where range of incidence of 1.0 to 12.0 percent. Colonies of *A. flavus* were recorded on 3.0 to 12.0 per cent seed of 8 varieties/lines with maximum incidence in variety Prabhat and minimum in varieties NDA 91-2. Colonies of *Fusarium moniliforms* were observed on 1.0 to 8. 0 percent seed of 6 varieties with maximum incidence in Prabhat and minimum in T 7. The fungal species *A. fumigatus C. globosum, C. lunata, D. tetramera, Mucor sp., P. oxalicum, R. soloni and R. nigricans* showed their presence in small number on the seeds.

1. (Ill-b) standard blotter method (pretreated seed):

In this experiment, pretreated seeds of all the varieties/lines of pigeonpea were tested by standard blotter method as described in "Materials and Methods".

Results recorded in Table-6, revealed that only eight species of fungi belonging to six genera were isolated from pretreated seeds, which showed distinct effect of chlorine pretreatment on the prevalence of fungi associated with pigeonpea seeds whereas, twelve fungal species were recorded on untreated seeds (Table-5). The treatment reduced considerably the average number of colonies of all the fungal species.

TABLE -4 : Percent incidence of fungal species associated with chlorine pretreated seeds of different varieties of pigeonpea in standard method (No. of seed tested – 400)

S. Fungal No. Species	VARIETIES											No. of Range Vers. of in- har dence	
	Pra-bhat	T 21	Pant A-3	NDA 91-13	NDA 91-12	Pusa 33	NDA 91-14	T7	T17	Bahar	NDA 91-1		
1.Alternaria alternata	16	11	8	8	4	9	3	8	8	7	6	11	4-16
2. *Aspergillus flavus*	10	5	10	5	2	6	2	0	5	0	0	8	2-10
3. *A. niger*	8	4	0	6	0	4	2	0	6	3	0	7	2-8
4. *A. Fumigatus*	3	0	0	0	1	0	0	0	0	0	0	2	1-3
5. Curvularia *lunata*	4	2	0	0	0	2	0	0	0	0	0	3	2-4
6. *Drechslera tetramera*	5	0	0	0	0	0	0	0	0	0	3	2	3-5
7. *Fusarium moniliforme*	9	5	1	3	1	0	0	2	0	0	0	6	1-9
8. *Rhizoctonia Solani*	6	0	2	0	0	0	0	0	0	0	0	2	2-6
Total no. of fungal species	8	5	4	4	4	4	3	2	3	2	2		

Associated with pigeonpea seeds. Four fungal species viz. chaetomium *globosum*, Mucor sp., *Penicillium oxalicum and Rhizopus nigricans* were totally absent due to pretreatment. This might be due to suppression or and elimination of fast growing fungi like C. *globosum*, *Mucor* sp. and R. nigricans. Consequently, slow growing fungal species faced less competition and developed in more number of seeds.

Results also revealed that *A. alternata* was detected from seeds of all the varieties followed by *A. flavus* on eight varieties. It was not, however, detected in varieties Bahar, NDA 91-1and T 7. *Aspergillus*

niger was observed in seven varieties, such as Prabhat, T 21 NDA 91- 13, Pusa 33, NDA 91-14, T 17, and Bahar. *Fusarium moniliforme* was detected from seeds of six varieties/lines, viz. Prabhat, T 21, Pant A-3. NDA 91-13, NDA 91-2, and T 7. The other fungal species shown in table 5 were recorded on seeds of 2 to 3 varieties.

Out of twenty varieties/lines tested, variety Prabhat harboured all fungal species on pretreated seeds followed by T 21 which yielded five fungal species. other varieties/line such as Pant A-3, NDA 91-13. Pusa 33 and NDA 91-2 carried four fungal species on the pretreated seeds.

1 (IV) Agar plate method:

Seeds of all the varieties of Pigeonpea were also tested by agar plate method with potato dextrose medium for observing the presence of *mycoflora* associated with them as described in "Materials and Methods". The observations were recorded after 7 days of incubation under sterioscopic binocular and compound microscope as summarized in Table 7.

The results presented in Table 7 revealed that seven fungal species belonging to five genera were detected from Pigeonpea seeds on PDA; *Alternaria alternata, Aspergillus fluvus*, *A. niger* and *F. moniliforme* were detected from six to eleven varieties. some fungal species i.e. A. *fumigatus*, *C. lunata* and *R. solani* were not observed on seeds of most of the varieties. It indicated that either surface disinfection or the method is responsible for their elimination of the twenty varieties Seeds of variety Prabhat were found to yield maximum number of fungal species (seven fungal species) followed by T21, Pant, A-3, NDA 91-13, NDA 91-2 and Pusa 33 which exhibited five and four fungal species.

CONCLUSION:

Seeds of eleven varieties/lines were used, collected from farmers fields, seed companies and pulse section of Narendra Dev University of Agriculture and Technology.

The result obtained are presented in table-3, it is clear from the results recorded in table that seeds if pigeonpea varieties, prabhat T21, Pant A-3 Pusa 33 and Bahar.

However, the seeds of other varieties like; NDA, 91-2, T 7, T17, NDA 91-14, NAD 91-13 and NDA 91-1 did not show any discolouration on the seed surface, At the same time no fungal fructifications were observed no the seeds of any varieties/lines under study.

Alternaria, Aspergillus, Curvularia, Drechslera and Fusarium were found associated with seed samples of varieties/lines like Prabhat, T-21, Pant A-3, Puss 33, Bahar, NDA-91-2,T7, T17,NDA91-14, NDA 91-13 and NDA 91-1.

Study also indicated that Alternaria alternata spores were dominant oin almost all the samples next predominant fungus was Aspergillus which was present in eight varieties out of eleven while five varieties viz, T 7, Bahar and NDA 91-1 showed no association of Aspergillus, Curvularia lunata.

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