Girish, A.G., Singh, S.D., Chakrabarty, S.K., Prasada Rao, R.D.V.J.,
Surender, A., Varaprasad, K.S. and Bramel, P.J. (2001), *Seed Sci. & Technol.*, **29**, 429-443

Seed microflora of five ICRISAT mandate crops

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(Accepted September 2000)

Summary

International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) supplies seeds of sorghum, pearl millet, pigeonpea, chickpea, and groundnut for research globally. The export of seeds of these crops is channelized through the regional station of the National Bureau of Plant Genetic Resources (NBPGR), Rajendranagar, Hyderabad. However, the tests for quarantine clearance of seeds for export are done at the Export Certification Laboratory at the ICRISAT Center.

During the period from June 1989 to December 1997, ICRISAT exported 371,818 samples of its mandate crops to 136 countries. The largest number of exported samples were of sorghum (140,143) followed by chickpea (119,308). A total of 1786 samples (sorghum, 571; pearl millet, 120; pigeonpea, 311; chickpea, 199; Groundnut, 585) were detained due to heavy seed infection by fungi and/or bacteria (>80% seed infection). Pigeonpea appeared to be the most popular crop exported to 105 countries followed by sorghum (91 countries) and groundnut (88 countries). A total of 182 fungal spp. belonging to 71 genera were recorded. Largest number of fungi–132 fungal species across the years, were found associated with sorghum crop. The corresponding figures for pearl millet, chickpea, pigeonpea, and groundnut were 94, 91, 96, and 60, respectively. *Aspergillus* spp. were more on pulses and groundnut than on sorghum and pearl millet; however, *Curvularia* spp. showed the reverse trend. *Fusarium* and *Alternaria* spp. occurred most frequently on pigeonpea followed by on sorghum. Also, there was a total absence of three graminicolous fungi – *Dreschlera, Biopolaris* and *Exserohilum* spp. *on* groundnut. There were 31 fungi associated with all the five crops. *Aspergillus niger* (3.8%) and *Cladosporium* spp. (3.6%) were the most commonly occurring fungi being most predominant on groundnut and sorghum, respectively.

Introduction

International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) has a global mandate for the improvement of its five mandate crops – sorghum, pearl millet, chickpea, pigeonpea, and groundnut. In addition, ICRISAT also has the responsibility of conservation of world germplasm of these crops. This involves import of germplasm of these crops from countries world-wide. Inherent to these responsibilities are the supply of germplasm and breeding materials of all its mandate crops throughout the world. To achieve these objectives ICRISAT needed an easy system of germplasm movement – both import and export. In the early stage of the Institute both the import and export of all germplasm were done through the then Central Plant Protection and Training Institute, Rajendranagar, Hyderabad. However,

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keeping in view its commitment for a large scale and speedy exchange of germplasm, ICRISAT signed a Memorandum of Agreement (MoU) with the Government of India in 1972 to set up an Export Certification Laboratory, including a Post Entry Quarantine Isolation Area (PEQIA), and a greenhouse within its premises at Patancheru. This laboratory follows strict plant quarantine protocols established by the Government of India for export of seed. The work of the laboratory is supervised jointly by the scientists of National Bureau of Plant Genetic Resources (NBPGR), Regional Station, Rajendranagar, at Hyderabad and ICRISAT. The major activity during the quarantine clearance of seed is the detection and identification of seed-associated fungi and bacteria. This information on the seed that was exported between 1982 and May 1989 has already been published (Ravinder Reddy *et al.*, 1989). In this paper, we present information on seed-associated microflora on these crops between June 1989 and December 1997.

Materials and methods

A total of 371,818 seed samples of sorghum (140,143), pearl millet (29,414), chickpea (119,308), pigeonpea (36,204), and groundnut (46,749) including wild and cultivated germplasm accessions; breeding material; sources of disease, pest, drought, cold and chilling resistance/tolerance; and released, pre-released and unreleased cultivars of these crops from various storage conditions (short, medium and long terms) were processed (table 1). The microflora associated with the seed were recorded as percentage infection for each organism separately. The following seed health testing protocol was followed.

Cleaning. Soon after receival the seeds are visually examined. Seed mixtures, glumes, and other plant material are removed.

Fumigation. The first key operation is fumigation of all seed samples. Sorghum, chickpea, and pigeonpea seed are fumigated under vacuum at a pressure of 125 mm mercury with a methyl bromide dosage at 32 gm^{-3} for 4 h. Pearl millet and groundnut seed are fumigated at

Table 1. Germplasm and breeding materials of ICRISAT mandate crops supplied by ICRISAT to 136 countries during June 1989 to December 1997.

Year	Sorghum	Pearl millet	Chickpea	Pigeonpea	Groundnut	Total
1989	15,632	2,136	22,975	4,665	6,095	51,503
1990	18,645	4,467	14,722	5,526	4,675	48,035
1991	18,810	1,758	10,798	4,419	3,130	38,915
1992	19,205	2,391	14,580	8,691	4,551	49,418
1993	19,588	2,564	15,025	4,288	8,378	49,843
1994	15,928	5,356	13,619	3,328	6,367	44,598
1995	18,780	4,231	13,748	2,552	4,223	43,534
1996	6,664	3,952	11,342	2,289	3,992	28,239
1997	6,891	2,559	2,499	446	5,338	17733
Total	140,143	29,414	119,308	36,204	46,749	371,818

normal atmospheric pressure with aluminium phosphide at 3 g m^{-3} for 5 days (Varma and Ravi, 1984; Joshi,1988).

Dry seed examination. After fumigation, each seed sample is carefully examined under illuminated floating desk magnifier of (2 ×) to remove fungal bodies (e.g., sclerotia, galls, smut sori), discolored and mouldy seeds, and insects. Apparently healthy looking and clean seeds are selected.

Seed washing test. This test is done to remove invisible fungal bodies/spores that do not grow on the seeds after incubation on blotter in petri plates, such as oospores of downy mildews and teliospores of smut fungi on the surface of sorghum and pearl millet seeds. Fifty randomly selected seeds from each sample are taken in test tubes containing 10 ml distilled water and a few drops of 95% ethyl alcohol. The tubes are shaken in a mechanical shaker for 10 min and the suspension is centrifuged at 3000 rpm for 10 min. Discarding supernatant, the pellet is re-suspended in 2 ml distilled water. The suspension is examined under compound microscope for the detection of above bodies.

Blotter Test. Seed are sown on moist blotting paper lined in the lower lids of petriplates. The plates are incubated for 7 days at 22 ± 2°C under 12 h Near Ultra Violet light and 12 h light cycle. After incubation, each seed is examined under stereomicroscope of (60-500×) magnification for the presence of fungal growth. Fungi associated with seeds are identified based on their habitual characters (colony morphology, and sporulation).

A working sample is drawn from each bulk submitted for quarantine clearance for export. The working sample consists of 10-100 seeds, depending upon the size of the bulk. Ten seeds of cereal crops (pearl millet and sorghum) and five seeds of chickpea, pigeonpea, and groundnut are sown in one plate and each such plate serves as a replication. Number of replications depends on sample size. Fungal growth on seeds placed nearest to the wall of petri-plate (outer ring) is examined first followed by inner rings. Seeds showing development of even-single conidiophore with conidia of fungi like *Alternaria, Curvularia, Dreschlera* spp; single fruiting structure like pycnidium of fungi like *Ascochyta, Phoma, and Macrophomina* spp; and single acervulus of fungi like *Colletotrichum* and *Myrothecium* spp. are counted as infected. In certain cases, slides are prepared and observed under compound microscope for correct identity. The percentage of each fungi found associated with each seed lot in each plate (replicate) is recorded.

Enzyme-linked immunosorbent assay (ELISA). Groundnut accessions and cuttings submitted for export are tested by ELISA for the detection of Peanut Mottle Virus (PMV). At ICRISAT, the Direct Antigen Coating (DAC) system, standardized by Hobbs *et al.* (1987), is followed. The seeds which are found free from PMV were further tested through the blotter method for detecting seed microflora.

Sorghum, chickpea, and pigeonpea seeds are treated with Chlorpyriphos, Bavistin (WP), and Thiram (75 SD) in the ratio of 3:2.5:2 @ 7.5 g kg^{-1} seed. Seeds of pearl millet and groundnut are treated with Chlorpyriphos and Thiram (75 SD) in the ratio of 3:2 @ 5 g kg^{-1} seed. A mixture of 30% Benomyl and 30% Thiram has been used to eradicate *Rhizoctonia*

bataticola from pigeonpea seeds (Kannaiyan *et al.,* 1980). Benomyl with chlorpyriphos in the ratio 2.5:3 @ 5.5 g kg^{-1} seeds has also been effective in eradicating *R. bataticola* inoculum from groundnut seeds. (Girish *et al.,* 1998 Unpublished).

Results

The details of seed samples dispatched and the fungi associated with seed of different crops are presented in table 2 and a comparison of number of genera, species, and most frequently occurring fungi on different crops, fungi of quarantine significance, and seed-borne fungi obtained between 1982 to May 89 (period I) and June 1989 to December 97 (period II) are presented in table 3. During period II, ICRISAT exported 371818 seed samples of its five mandate crops to 136 countries. Sorghum topped the list with 140143 samples followed by chickpea with 119,308 samples. However, pigeonpea appeared to be the most favorite crop with export to 105 countries followed by sorghum to 91 countries. A total of 1786 samples of five crops were detained due to severe fungal and/or bacterial infection (>80% seed infection and loss in viability).

Crop-wise distribution of fungi

Sorghum. Sorghum seed samples yielded the largest number of fungi – 132, belonging to 51 genera. The largest number of fungi (21) belonged to the genus *Curvularia.* However, *Cladosporium* spp. were the most frequently occurring fungi associated with 17.6% of the samples. Sorghum samples yielded eight fungi of quarantine significance and there were 27 seed-borne fungi (tables 2 and 3).

Pearl millet. Pearl millet samples yielded 94 fungi belonging to 39 genera. The largest number of fungi (16) occurring on seed of this crop belonged to the genus *Bipolaris.* The largest number of pearl millet samples were infected with *Fusarium* spp. (3.9%) followed by *Rhizopus* spp. and *Cladosporium* spp. (3.4%). The samples yielded four fungi of quarantine significance and there were 12 seed borne fungi (tables 2 and 3).

Chickpea. Chickpea samples yielded 91 fungi belonging to 46 genera. Fungi belonging to genus *Aspergillus* were more common on chickpea seed with *A. niger* being the most predominant (17.8% samples infected). Like sorghum, chickpea hosted largest number of fungi (11) belonging to genus *Curvularia.* There were six fungi of quarantine significance and nine fungi were found to be seed-borne (tables 2 and 3).

Pigeonpea. Pigeonpea samples were infected with 96 fungi belonging to 50 genera. *A. niger* attacked the largest number of pigeonpea samples (17%) followed by *Rhizopus* spp. (12.6%). There were four fungi of quarantine significance and seven fungi were seed-borne (tables 2 and 3).

Groundnut. Groundnut harboured the least seed-borne and seed-carried pathogens compared to the four other crops; only 60 fungi belonging to 36 genera were obtained. *A. niger* was the most frequently occurring fungus (21.5% samples infected), followed by *Rhizopus* spp. (10%

seed samples infected) on this crop. No fungus belonging to *Bipalaris, Dreschlera,* and *Exserohilum* was found associated with groundnut seed. There were only three fungi of quarantine significance and 14 fungi were seed-borne (tables 2 and 3).

Distribution of bacteria and actinomyces

Gram positive bacteria ranging from 0.13 to 0.5% of the seed samples were found associated with ICRISAT crops. However, gram negative bacteria were found only on sorghum seed. *Bacillus* spp. was found only on 0.03% of the chickpea seeds and Actinomyces were observed only on sorghum and pearl millet seeds.

A comparison of data obtained during the two periods – period I and period II, shows some shift in the distribution of fungi. There were less number of fungal genera associated with sorghum, pearl millet, and chickpea in period II than in period I, but the reverse was the trend with respect to number of fungal species. However, for pigeonpea and groundnut, the number of both, the genera and the species, were more in period II than period I. Interestingly, period II also has more fungi of quarantine significance and also seed-borne fungi on all the crops than period I. *Fusarium* was the most commonly occurring genus on chickpea, pigeonpea, and groundnut, but *A. niger* was the most common fungus on these crops. The two cereal crops did not show such trend.

Discussion

Five ICRISAT mandate crops harbor a wide range of fungal flora. Sorghum, supporting 132 fungal spp. belonging 51 genera, is the most preferred host to fungi among these crops. Groundnut, on the other hand, supported the least number of fungi (60). This may probably be due to the fact that sorghum is grown in much more diverse range of environment, latitudes, longitudes, soil types etc., than groundnut, which exposes the crop to a wide range of mycroflora.

The increase in the number of fungi associated with all the five crops along with increases in the number of fungi of quarantine importance and seed-borne fungi during period II is a cause for concern. Although, several factors including crop accession, their origin, and growing season etc., may play role in the association of fungal flora, equally important fact probably has been the availability of more information on these crops during this period as has been well documented in case of chickpea and pigeonpea (Nene *et al.,* 1996).

Fungi belonging to many genera were found associated with all the crops. The most commonly occurring fungi are the species of *Alternaria, Aspergillus, Cladosporium, Curvularia, Fusarium, Macrophomina, Penicillium, Phoma and Rhizopus.* Of these, *Fusarium* spp. were generally equally distributed on all the crops. However, spp. of *Bipolaris, Curvularia,* and *Cladosporium* were more on sorghum and pearl millet, while spp. of *Aspergillus* and *Rhizopus* were more on groundnut, pigeonpea and chickpea. Irrespective of the crop-wise distribution, the result shows that these are the most important fungi of ICRISAT mandate crops. In order to make our germplasm health test a realistic one, we have to study the potential of these fungi including their survivability under different storage conditions and their ability to reduce seed viability.

Table 2. Fungi, bacteria, and actinomyces found associated with seed of sorghum (Sor), pearl millet (Pm), chickpea (Cp), pigeonpea (Pp), and groundnut (Gn) in the Export Certification Laboratory at ICRISAT from June 1989 to December 1997.

Fungi/Bacteria/Actinomyces	Number and percentage of infected samples									
	Sor		Pm		Cp		Pp		Gn	
	1`	A	2	B	3	C	4	D	5	E
	No	%	No	%	No	%	No	%	No	%
Absidia spp.	-	-	-	-	1	<0.01	3	0.02	-	-
Acremonium spp.	8	0.01	4	0.03	-	-	1	0.01	8[Q]	0.03
A. strictum	1[QS]	<0.01	-	-	-	-	-	-	-	-
Alternaria spp.	6,080[S]	10.73	171	1.21	1,214	6.07	1,522	10.97	22	0.07
A. alternata	2,091	3.69	119[S]	0.84	457	2.28	553[S]	3.98	12	0.04
A. dauci	-	-	-	-	-	-	2	0.01	-	-
A. longipes	8	0.01	4	0.03	-	-	5	0.04	-	-
Alternaria longissima	3	0.01	-	-	3	0.01	7	0.05	-	-
Alternaria tenuissima	1	<0.01	-	-	-	-	-	-	-	-
Aspergillus spp.	113	0.20	97	0.69	375	1.87	329	2.37	254	0.81
A. glaucus	-	-	-	-	-	-	-	-	29	0.09
A. candidus	-	-	3	0.02	1	<0.01	2	0.01	-	-
A. flavus	207	0.37	99	0.70	486	2.43	614	4.42	719[S]	2.29
A. fumigatus	-	-	-	-	5	0.02	2	0.01	3	0.01
A. nidulans	-	-	-	-	-	-	-	-	1	<0.01
A. niger	936[S]	1.65	369	2.62	3,560	17.79	2,354	16.96	6,748[S]	21.53
A. parasiticus	1	<0.01	-	-	1	<0.01	1	0.01	5	0.02
Arthrobotrys spp.	1	<0.01	-	-	-	-	3	0.02	-	-
Ascochyta rabiei	-	-	-	-	4[QS]	0.02	-	-	-	-
Ascochyta sorghina	2[QS]	<0.01	-	-	3	0.01	1	0.01	-	-
Ascochyta subalphina	-	-	-	-	-	-	1	0.01	-	-
Aureobasidium spp.	2	<0.01	-	-	9	0.04	-	-	-	-
Beauveria spp.	-	-	2	0.01	-	-	1	0.01	1	<0.01
Bipolaris spp.	252	0.44	3	0.02	-	-	-	-	-	-
B. bicolor	3	0.01	-	-	-	-	-	-	-	-

Table 2. Continued.

Fungi/Bacteria/Actinomyces	Number and percentage of infected samples									
	Sor		Pm		Cp		Pp		Gn	
	1	A	2	B	3	C	4	D	5	E
	No	%	No	%	No	%	No	%	No	%
B. carbonum	1	<0.01	4	0.03	-	-	-	-	-	-
B. cynodontis	5	0.01	77	0.55	6	0.03	4	0.03	-	-
B. dactyloctenii	1	<0.01	-	-	-	-	-	-	-	-
B. hawaiiensis	88[s]	0.16	26	0.18	7	0.03	6	0.04	-	-
B. maydis	7	0.01	2	0.01	-	-	-	-	-	-
B. neergaardii	1	<0.01	-	-	-	-	-	-	-	-
B. oryzae	10	0.02	1	0.01	-	-	-	-	-	-
B. papendorfii	-	-	2	0.01	-	-	-	-	-	-
B. perotidis	1	<0.01	-	-	-	-	-	-	-	-
B. rostrata	5	0.01	6	0.04	-	-	-	-	-	-
B. sacchari	15	0.03	6[s]	0.04	2	0.01	7	0.05	-	-
B. setariae	6	0.01	15[s]	0.11	1	<0.01	-	-	-	-
B. sorghicola	36[s]	0.06	2	0.01	-	-	-	-	-	-
B. sorokiniana	-	-	3[Q]	0.02	-	-	-	-	-	-
B. spicifera	21[s]	0.04	2[s]	0.01	1	<0.01	2	0.01	-	-
B. triticicola	4	0.01	1	0.01	-	-	-	-	-	-
B. zeae	9	0.02	1	0.01	-	-	-	-	-	-
Botryodiplodia spp.	2	<0.01	-	-	5	0.02	15	0.11	1	<0.01
B. theobromae	2	<0.01	-	-	2	0.01	7[s]	0.05	9	0.03
Botryosphaeria ribis	-	-	2	0.01	-	-	-	-	1	<0.01
Botrytis spp.	-	-	-	-	13	0.06	4	0.03	-	-
Botrytis cinerea	-	-	-	-	9[QS]	0.04	-	-	-	-
Cephalosporium spp.	9	0.02	4	0.03	1	<0.01	3	0.02	2	0.01
Cercospora spp.	6	0.01	-	-	-	-	40	0.29	-	-
C. sorghi	2[QS]	<0.01	-	-	-	-	2	0.01	-	-
Chaetomium spp.	10	0.02	8	0.06	61	0.30	17	0.12	4	0.01

Table 2. Continued.

Fungi/Bacteria/Actinomyces	Number and percentage of infected samples									
	Sor		Pm		Cp		Pp		Gn	
	1	A	2	B	3	C	4	D	5	E
	No	%	No	%	No	%	No	%	No	%
Choanephora spp.	-	-	-	-	-	-	2	0.01	-	-
Chaetophoma spp.	1	0.00	-	-	-	-	-	-	1	<0.01
Circinella spinosa	-	-	-	-	3	0.01	-	-	2	0.01
Cladosporium spp.	9,964	17.58	473	3.36	1,755	8.77	1,140	8.21	82	0.26
Cochliobolus lunatus	3	0.01	1	0.01	-	-	-	-	1	<0.01
C. setariae	-	-	1	0.01	-	-	-	-	-	-
Colletotrichum cajani	-	-	-	-	-	-	15[Q]	0.11	-	-
C. gloeosporioides	1	<0.01	-	-	-	-	2	0.01	1[S]	<0.01
C. dematium	-	-	-	-	4[S]	0.02	1	0.01	5[QS]	0.02
C. graminicola	16[S]	0.03	-	-	-	-	-	-	-	-
Corynespora spp.	-	-	-	-	1	<0.01	-	-	-	-
Cunninghamella spp.	2	<0.01	1	0.01	-	-	1	0.01	-	-
Curvularia spp.	1,730	3.05	310	2.20	185	0.92	104	0.75	5	0.02
C. andropogonis	8	0.01	-	-	-	-	-	-	-	-
C. clavata	44	0.08	17	0.12	12	0.06	3	0.02	1	<0.01
C. cymbopogonis	1	<0.01	-	-	-	-	-	-	-	-
C. eragrostidis	7	0.01	2	0.01	4	0.02	1	0.01	-	-
C. geniculata	5[S]	0.01	-	-	2	0.01	-	-	-	-
C. halodes	1	<0.01	-	-	-	-	-	-	-	-
C. ischaemi	6	0.01	1	0.01	-	-	1	0.01	-	-
C. intermedia	6	0.01	-	-	-	-	-	-	-	-
C. lunata	463[S]	0.82	79[S]	0.56	32	0.16	16[S]	0.12	4	0.01
C. oryzae	10	0.02	-	-	2	0.01	-	-	-	-
C. ovoidea	3	0.01	-	-	2	0.01	2	0.01	-	-
C. pallescens	40	0.07	6	0.04	20	0.10	2	0.01	2	0.01
C. penniseti	7	0.01	6[S]	0.04	-	-	-	-	-	-

Table 2. Continued.

Fungi/Bacteria/Actinomyces	Number and percentage of infected samples									
	Sor		Pm		Cp		Pp		Gn	
	1	A	2	B	3	C	4	D	5	E
	No	%	No	%	No	%	No	%	No	%
C. robusta	31	0.05	16	0.11	2	0.01	-	-	-	-
C. senegalensis	9	0.02	-	-	2	0.01	1	0.01	-	-
*C. siddiquii***	1	<0.01	-	-	-	-	-	-	-	-
C. sorghina	6	0.01	3	0.02	-	-	-	-	-	-
C. trifolii	7	0.01	1	0.01	3	0.01	1	0.01	-	-
C. tuberculata	2[s]	<0.01	1	0.01	-	-	-	-	-	-
C. verruculosa	30	0.05	2	0.01	-	-	-	-	-	-
Cylindrocladium crotalariae	-	-	-	-	-	-	-	-	1	<0.01
Diplodia spp.	1	<0.01	-	-	-	-	1	0.01	1[s]	<0.01
Drechslera spp.	90	0.16	19	0.13	5	0.02	3	0.02	-	-
*D. bicolor**	2	<0.01	1[s]	0.01	-	-	-	-	-	-
D. carbonum	1	<0.01	-	-	-	-	-	-	-	-
D. eragrostidis	1	<0.01	-	-	-	-	-	-	-	-
D. holmii	2	<0.01	2	0.01	-	-	3	0.02	-	-
D. longirostrata	23	0.04	6	0.04	-	-	-	-	-	-
*D. maydis**	6[s]	0.01	1	0.01	-	-	-	-	-	-
*D. oryzae**	12	0.02	-	-	-	-	-	-	-	-
D. rostrata	97	0.17	24	0.17	-	-	1	0.01	-	-
*D. sacchari**	1	<0.01	1	0.01	-	-	-	-	-	-
*D. setariae**	-	-	1	0.01	-	-	-	-	-	-
*D. sorghicola**	3	0.01	-	-	-	-	-	-	-	-
D. sorokiniana	-	-	-	-	-	-	1	0.01	-	-
D. tetramera[#]	19	0.03	4	0.03	8	0.04	2	0.01	-	-
D. victoriae	1[s]	<0.01	1[s]	0.01	-	-	-	-	-	-
Epicoccum spp.	359	0.63	25	0.18	96	0.48	20	0.14	-	-
E. purpurascens	-	-	-	-	1	<0.01	-	-	-	-

Table 2. Continued.

Fungi/Bacteria/Actinomyces	Number and percentage of infected samples									
	Sor		Pm		Cp		Pp		Gn	
	1	A	2	B	3	C	4	D	5	E
	No	%	No	%	No	%	No	%	No	%
Exserohilum spp.	28	0.05	15	0.11	7	0.03	3	0.02	-	-
E. gedarefense	1	<0.01	-	-	-	-	-	-	-	-
E. holmii	5	0.01	6	0.04	1	<0.01	3	0.02	-	-
E. longirostratum	74[S]	0.13	20	0.14	-	-	1	0.01	-	-
E. oryzae	1	<0.01	-	-	-	-	-	-	-	-
E. oryzicola	1	<0.01	-	-	-	-	-	-	-	-
E. oryzinum	2	<0.01	-	-	-	-	-	-	-	-
E. rostratum	236[S]	0.42	93	0.66	29	0.14	6	0.04	-	-
E. turcicum	39[QS]	0.07	6	0.04	-	-	-	-	-	-
E. curvatum	-	-	1	0.01	-	-	-	-	-	-
Fusarium spp.	1,700	3.00	553	3.92	458	2.29	791	5.70	460	1.47
F. acuminatum	7	0.01	2	0.01	-	-	2	0.01	-	-
F. chlamydosporum	12	0.02	4	0.03	1	<0.01	6	0.04	3	0.01
F. decemcellulare	4	0.01	-	-	-	-	-	-	-	-
F. dimerum	-	-	-	-	-	-	1	0.01	2	0.01
F. equiseti	31	0.05	21	0.15	10	0.05	23	0.17	4[S]	0.01
F. graminearum	-	-	1	0.01	2	0.01	-	-	-	-
F. lateritium	5	0.01	-	-	-	-	-	-	-	-
F. moniliforme	1,954[S]	3.45	201[S]	1.43	23	0.11	72	0.52	15	0.05
F. oxysporum	22	0.04	8	0.06	61[QS]	0.30	148[Q]	1.07	155[S]	0.49
F. pallidoroseum	1	<0.01	-	-	1	<0.01	-	-	1	<0.01
F. proliferatum	2	<0.01	-	-	1	<0.01	1	0.01	-	-
F. semitectum	158	0.28	60	0.43	37	0.18	148	1.07	46	0.15
F. solani	25	0.04	29	0.21	19[QS]	0.09	139[S]	1.00	122[S]	0.39
F. sporotrichioides	-	-	-	-	-	-	2	0.01	-	-
F. udum	-	-	1	0.01	6	0.03	14[QS]	0.10	1	<0.01

Table 2. Continued.

Fungi/Bacteria/Actinomyces	Number and percentage of infected samples									
	Sor		Pm		Cp		Pp		Gn	
	1	A	2	B	3	C	4	D	5	E
	No	%	No	%	No	%	No	%	No	%
Gliocladium spp.	-	-	-	-	-	-	-	-	3	0.01
Gloeocercospora sorghi	1[s]	<0.01	-	-	2	0.01	4	0.03	-	-
Glomerella spp.	1	<0.01	-	-	-	-	2	0.01	-	-
G. cingulata	-	-	-	-	-	-	1	0.01	-	-
Gonatobotrys spp.	14	0.02	4	0.03	2	0.01	8	0.06	-	-
Leptosphaerulina spp.	3	0.01	-	-	21	0.10	-	-	1	<0.01
L. craciassca	1	<0.01	-	-	4	0.02	1	0.01	5[s]	0.02
Macrophomina phaseolina	25[s]	0.04	4[s]	0.03	23[s]	0.11	364[s]	2.62	216[s]	0.69
Memnoniella spp.	3	0.01	-	-	1	<0.01	1	0.01	-	-
Melanospora spp.	1	<0.01	1	0.01	-	-	-	-	-	-
Metarrhizium spp.	-	-	3	0.02	-	-	-	-	-	-
Mucor spp.	8	0.01	-	-	15	0.07	46	0.33	3	0.01
Myrothecium spp.	7	0.01	3	0.02	4	0.02	1	0.01	1	<0.01
Nigrospora spp.	57[s]	0.10	21	0.15	25	0.12	13	0.09	9	0.03
Oedocephalum spp.	20[s]	0.04	17	0.12	8	0.04	26	0.19	2	0.01
Papularia spp.	-	-	-	-	1	<0.01	-	-	-	-
Penicillium spp.	54	0.10	83	0.59	207	1.03	134	0.97	121	0.39
Periconia spp.	21	0.04	1	0.01	2	0.01	7	0.05	-	-
Periconia byssoides	-	-	-	-	-	-	2	0.01	-	-
Peronosclerospora sorghi	5[QS]	0.01	-	-	-	-	-	-	-	-
Pestalotia spp.	2	<0.01	1	0.01	5	0.02	11	0.08	1	<0.01
Phoma spp.	1,003	1.77	357	2.53	289	1.44	270	1.95	57	0.18
P. medicaginis	-	-	-	-	1[s]	<0.01	-	-	-	-
P. sorghina	142[s]	0.25	20	0.14	1	<0.01	-	-	-	-
Phomopsis spp.	-	-	-	-	2[Q]	0.01	144[QS]	1.04	-	-
Phyllosticta cajani	-	-	-	-	-	-	5[s]	0.04	-	-

Table 2. Continued.

Fungi/Bacteria/Actinomyces	Number and percentage of infected samples									
	Sor		Pm		Cp		Pp		Gn	
	1 No	A %	2 No	B %	3 No	C %	4 No	D %	5 No	E %
Pithomyces spp.	250	0.44	52	0.37	111	0.55	42	0.30	2	0.01
Pithomyces chartarum	2	<0.01	-	-	-	-	-	-	-	-
Puccinia penniseti	-	-	1[Q]	0.01	-	-	-	-	-	-
Pyricularia spp.	-	-	-	-	-	-	1	0.01	1	<0.01
Pyricularia penniseti	-	-	1[QS]	0.01	-	-	-	-	-	-
Pyrinochaeta spp.	-	-	3	0.02	-	-	1	0.01		0.00
Rhizoctonia spp.	-	-	-	-	2	0.01	-	-	29	0.09
R. bataticola	4[S]	0.01	-	-	4	0.02	6	0.04	279[S]	0.89
R. solani	-	-	-	-	6[S]	0.03	7	0.05	25[S]	0.08
Rhizopus spp.	1,161	2.05	495	3.51	1321	6.60	1,744	12.56	3,057	9.75
Sclerotium rolfsii	-	-	-	-	1[QS]	<0.01	-	-	30[QS]	0.10
Sclerospora graminicola	-	-	1[QS]	0.01	-	-	-	-	-	-
Spegazzinia spp.	1	<0.01	8	0.06	-	-	-	-	-	-
Sphacelotheca cruenta	1[QS]	<0.01	-	-	-	-	-	-	-	-
Stachybotrys spp.	10	0.02	17	0.12	10	0.05	15	0.11	3	0.01
Stemphylium spp.	1[Q]	<0.01	-	-	8	0.04	-	-	-	-
Sterile fungus	-	-	-	-	3	0.01	-	-	-	-
Tolyposporium ehrenbergii	1[QS]	<0.01	-	-	-	-	-	-	-	-
Torula spp.	-	-	1	0.01	11	0.05	-	-	-	-
Trichoconis spp.	1	<0.01	-	-	-	-	-	-	-	-
Trichoderma spp.	10	0.02	-	-	2	0.01	6	0.04	-	-
Tricothecium spp.	51	0.09	19	0.13	43	0.21	115	0.83	9	0.03
Ulocladium spp.	-	-	-	-	1	<0.01	1	0.01	-	-
U. botrytis	-	-	-	-	1	<0.01	-	-	-	-
Verticillium spp.	7	0.01	1	0.01	5	0.02	6	0.04	1[S]	<0.01
Bacteria (Gram positive)	82	0.14	76	0.54	95	0.47	38	0.27	41	0.13

Table 2. Continued.

Fungi/Bacteria/Actinomyces	Number and percentage of infected samples									
	Sor		Pm		Cp		Pp		Gn	
	1	A	2	B	3	C	4	D	5	E
	No	%	No	%	No	%	No	%	No	%
Bacteria (Gram negative)	47		-		-		-		-	
Actinomycetes	1	<0.01	23	0.16	-	-	-	-	-	-
Bacillus spp	-	-	-	-	6	0.03	-	-	-	-
Streptomyces spp.	3	0.01	63	0.45	1	<0.01	1	0.01	1	<0.01

Sor : Sorghum, Pm: Pearl millet, Cp: Chickpea, Pp: Pigeonpea, Gn: Groundnut
Q : Quarantine significance pathogen
S : Seedborne fungi
< : Less than
* : *Drechslera* converted into *Bipolaris*
** : *Curvularia siddiquii* converted into *Bipolaris papendorfii*
# : *Drechslera tetramera* converted into *Bipolaris specifera*
No. : Number of seed samples despatched

Table 3. Details of different fungus genera/species, their frequencies, fungi of plant quarantine significance and seed-borne fungi detected on seeds of five ICRISAT mandate crops in the Plant Quarantine laboratory during 1982 to May 1989 (period I) and June 1989 to December 1997 (period II).

Crop*	Total genera (No.)		Total fungal species (No.)		Most frequent genera		Most frequent fungal species		Fungi of Quarantine importance		Seed-borne fungi (No.)		New seed-borne fungi
	Period I	Period II	Period I	Period II	Period I	Period II	Period I	Period II	Period I	Period II	Period I	Period II	89-97
Sor	61	51	128	132	*Curvularia*	*Curvularia*	*Alternaria spp.*	*Cladosporium* spp.	5	8	26	27	*Phoma sorghina*
Pm.	46	39	89	94	*Curvularia*	*Bipolaris*	*Cladosporium* spp.	*Fusarium* spp.	3	4	11	12	*Pyricularia penniseti*
Cp.	49	46	67	91	*Fusarium*	*Curvularia*, Fusarium.	*A. niger*	*A. niger*	3	6	8	9	*P. medicaginis*
Pp.	35	50	52	96	*Fusarium*	*Fusarium*	*A. niger*	*A. niger*	1	4	4	7	*Alternaria alternata* *Phyllosticta cajanii* *Botryodiplodia theobromae*
Gn.	26	36	40	60	*Fusarium*	*Fusarium*	*A. niger*	*A. niger*	0	3	9	14	*Aspergillus flavus, A.niger* *Fusarium equiseti* *F. oxysporum* *Leptosphaerulina crassiasca*

* Sor = Sorghum, Pm = Pearlmillet, Cp = Chickpea, Pp = Pigeonpea, Gn = Groundnut

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