

Table 2. Incidence and intensity of grain mold in sorghum cultivars under two treatments of panicles (with and without bags)¹.

Cultivar	TGMR ²	Germination ¹	Incidence (%)	
			<i>Fusarium moniliforme</i> ³	<i>Curvularia lunata</i> ³
CSH 9	3.08	44.54	17.62	28.75
CSH 14	2.87	31.83	17.75	29.12
CSH 16	2.87	40.49	17.67	34.76
CSH 17	2.83	45.49	23.66	36.02
CSH 18	3.20	47.44	19.73	26.91
CSV 13	3.00	35.91	30.86	28.38
CSV 15	2.95	46.86	21.68	31.59
PVK 801	2.83	48.35	17.65	20.01
CD at 5%	NS ⁴	9.91	7.40	NS

1. Values are averages of two treatments.

2. TGMR = Threshed grain mold rating; observations recorded on 1 to 5 scale, where 1 = mold free and 5 = >50% threshed grains molded.

3. Figures are in arcsine transformed values.

4. NS = Not significant.

sorghum grains and thus data indicated that grain mold infection was reduced. Covering panicles with polythene bags during rains can reduce grain mold infection. The use of polythene bags to minimize the grain mold infection is reported for the first time.

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Sorghum Grain Mold: Variability in Fungal Complex

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Introduction

The grain mold complex in sorghum (*Sorghum bicolor*) involves a number of pathogenic and saprophytic fungi that vary in their frequencies and severities under different environmental conditions (Bandyopadhyay et al. 2000). To provide genetic management for grain mold in sorghum, a clear understanding of the major pathogenic fungi and their variability under different environments is critical. Among the major pathogenic fungi, *Fusarium moniliforme* (*F. verticilloides*) is known to produce fumonisins, a mycotoxin of concern for the use of molded sorghum grains as food and feed (Marasas 1996, Bhat et al. 1997). With the above objective we initiated a collaborative Sorghum Grain Mold Variability Nursery (SGMVN) between ICRISAT and the All India

Coordinated Sorghum Improvement Project (AICSIP) of the Indian Council of Agricultural Research (ICAR). The nursery was coordinated by ICRISAT and conducted at four locations in India during the rainy season 2002. The results of the trials are presented.

Materials and Methods

The SGMVN-2002 consisted of 10 sorghum lines that had shown moderate to high levels of tolerance to grain mold in previous field screenings at ICRISAT, Patancheru, India and possessed desirable agronomic traits, and two check lines (a resistant and a susceptible). The nursery was established at four locations, Akola and Parbhani in Maharashtra, and Palem and Patancheru in Andhra Pradesh. Each entry was grown in two rows, each 4 m long and in two replications. The recommended agronomic and cultural practices for raising a good sorghum crop were followed at each location.

Sprinkler irrigation was provided on dry days for 30 min in the evening during flowering to crop maturity to create high relative humidity (RH) (>95%) necessary for fungal infection and mold development. No artificial inoculation with any mold fungi was done. Damage by insect pests, particularly by shoot fly, stem borer, and head bug was minimized by timely application of pesticides.

Data Recording

Grain mold severity score. Five plants with uniform flowering were tagged in each row of the 2-row plot (10 plants plot⁻¹). At each location, the overall grain mold severity scores were taken on each panicle at physiological maturity (PM) and post-PM (PPM) (10 days after PM) stages on a progressive 1 to 5 scale (1 = no mold, 2 = 1-10%, 3 = 11-25%, 4 = 26-50%, and 5 = >50% grains molded on a panicle). In addition, at Patancheru grain mold infection was also recorded at the hard-dough stage. Since there were no visible grain mold symptoms at the hard-dough stage, grain samples taken from the tagged panicles in each replication, were surface sterilized and plated on moist blotter paper in glass petri dishes at 25 grains per petri dish and 50 grains per replication. These were incubated at 28°C for 3 days and grain colonization by specific fungi were recorded using the above scale.

Threshed grain mold severity score. Threshed grain (10 g) from each panicle, spread in a petri dish was scored for visual rating on a 1 to 5 scale using a magnifying lens under proper lighting. The threshed grain samples of each entry obtained from all the four locations were subjected

to the above mentioned blotter test (50 grains per replication) at Patancheru to determine the frequency of major mold fungi on the threshed grains.

Weather variables. Temperature, RH and rainfall from the flowering stage of an early-maturing line to PPM stage of a late-maturing line were recorded at all locations to determine the influence of weather variables on predominance of mold fungi.

Isolation of fungi. Fifty grains of each line obtained from the test locations were surface sterilized and plated on moist blotter paper. These were examined 3 days after incubation at 28°C with 12 h photoperiod. Molded grains were examined under stereobinocular, particularly for the presence of *Fusarium* species. The typical *Fusarium* colonies were aseptically transferred from the grains to potato-dextrose agar (PDA) plates and incubated at 28°C for 5 days for colony growth and further purification.

Results and Discussion

Variation in grain mold severity. The disease pressure as indicated by grain mold severity on the susceptible check SPV 104 at PM was highest (3.8) at Akola, followed by Palem (3.4) and Patancheru (3.0) (Table 1). However, at PPM the highest pressure was at Patancheru (5.0) followed by Parbhani (4.5) and the lowest at Akola and Palem (4.0). The overall field grain mold severity of the 12 sorghum lines at PPM ranged from 2.9 at Palem to 4.0 at Parbhani, and those of threshed grains from 2.8 at Akola to 4.3 at Patancheru. Significant ($P < 0.001$) variations of grain mold severity occurred at different stages for sorghum lines, locations and their interactions, and no sorghum line was found resistant (<2.5 score) at all locations across all stages. However, at PM five lines (Sepon/78-1, ICSV 95001, SPV 351, ICSV 91008 and IS 8545) showed mold severity of <2.5 across 3 locations (Table 1). These lines are likely to have resistance to pathogenic fungi that might have infected the grains during the flowering to milk stages of grain development. This needs further investigation.

Variation in mold fungi. Major fungi recorded on grains in the field at PPM were species of *Fusarium*, *Ahernaria*, *Curvularia*, *Cladosporium*, *Drechsleru* and *Phoma*. There were significant ($P < 0.05$) variations in severity of these fungi across locations and among sorghum lines at a particular location. *Fusarium moniliforme*, *Alternaria alternata* and *Curvularia lunata* were more prominent than others. The frequency of the fungi varied across locations: *F. moniliforme* from 32 to 64%, *A. alternata* from 19 to 38%, *C. lunata* from 18 to 23% and *P. sorghina* from 5 to 14% (Table 2). Other fungi appeared in relatively

Table 1. Grain mold severity of the Sorghum Grain Mold Variability Nursery-2002 entries at physiological maturity (PM), post-physiological maturity (PPM), and on threshed grain at four locations in India¹.

Entry	PM				PPM				Threshed grain				Mean across stages			
	AKL	PAR	PAL	PAT	AKL	PAR	PAL	PAT	AKL	PAR	PAL	PAT	AKL	PAR	PAL	PAT
IS 18758C-618-2	3.0	-	3.2	2.8	4.0	4.5	3.6	4.7	3.3	4.4	3.7	5.0	3.4	4.4	3.5	4.2
IS 18522	3.1	-	2.6	2.6	4.0	4.3	3.3	4.6	3.6	4.0	3.3	4.8	3.6	4.2	3.1	4.0
ICSV 96101	3.1	-	2.0	2.0	4.0	4.1	2.5	3.6	3.0	3.4	2.5	4.4	3.4	3.8	2.3	3.3
CS 3541	2.8	-	2.4	2.1	3.5	4.0	3.0	4.2	3.1	3.4	3.2	4.2	3.2	3.7	2.9	3.5
Sepon/78-1	2.2	-	2.1	1.4	3.3	3.7	2.1	2.6	2.4	3.4	2.7	4.1	2.6	3.6	2.3	2.7
ICSV 95001	2.0	-	1.8	1.8	3.0	3.9	2.1	3.4	2.0	3.4	2.3	4.4	2.3	3.7	2.1	3.2
IS 30469C-140	3.0	-	3.0	2.4	4.3	4.2	3.6	4.1	3.6	3.3	3.7	5.0	3.6	3.8	3.4	3.8
SPV 351 (ICSV 1)	2.3	-	2.3	2.0	3.2	4.0	3.1	3.7	2.3	3.5	3.2	3.9	2.6	3.8	2.9	3.2
ICSV 91008	2.3	-	2.1	1.4	3.0	3.8	2.8	3.5	2.0	3.3	3.0	4.3	2.4	3.6	2.6	3.1
CSH 9	3.1	-	2.6	2.3	4.0	4.1	3.5	3.9	2.6	3.3	3.6	4.4	3.2	3.7	3.2	3.5
SPV 104 ²	3.8	-	3.4	3.0	4.0	4.5	4.0	5.0	3.8	3.8	4.2	5.0	3.9	4.2	3.9	4.3
IS 8545 ³	2.2	-	1.1	1.1	3.0	3.4	1.2	1.7	2.3	3.2	1.2	2.6	2.5	3.3	1.2	1.8
Mean	2.7	-	2.4	2.1	3.6	4.0	2.9	3.7	2.8	3.5	3.0	4.3	3.1	3.8	2.8	3.4
LSD (<i>P</i> <0.05)	0.27	-	0.20	0.30	0.17	0.42	0.20	0.34	0.24	0.13	0.19	0.36	1.21	0.67	1.61	1.52

1. Each value is mean of 2 replications with 10 panicles per replication, based on 1-5 scale where 1 = no mold, 2 = 1-10% mold, 3 = 11-25% mold, 4 = 26-50% mold and 5 = >50% grains molded on a panicle.

AKL = Akola, PAR = Parbhani, PAL = Palem, PAT = Patancheru; - = Data not available.

2. Susceptible check.

3. Resistant check.

Table 2. Frequency of different mold fungi on threshed sorghum grains from the Sorghum Grain Mold Variability Nursery-2002 entries during rainy season 2002 at four locations in India¹.

Entry	Seeds (%) colonized by mold fungi															
	<i>Fusarium moniliforme</i>				<i>Alternaria alternata</i>				<i>Curvularia lunula</i>				<i>Phoma sorghina</i>			
	AKL	PAR	PAL	PAT	AKL	PAR	PAL	PAT	AKL	PAR	PAL	PAT	AKL	PAR	PAL	PAT
IS 18758C-618-2	47	91	29	40	10	19	30	23	17	12	22	31	10	22	10	10
IS 18522	43	85	44	60	12	12	28	18	21	10	19	12	15	3	4	5
ICSV 96101	56	55	54	26	15	40	10	41	15	31	15	17	15	9	0	8
CS 3541	34	93	49	38	12	9	13	29	34	9	17	14	25	16	4	6
Sepon/78-1	23	37	38	36	21	49	12	35	23	31	8	18	11	10	15	6
ICSV 95001	8	94	32	26	49	22	38	37	25	11	23	23	5	12	4	14
IS 30469C-140	56	70	32	36	11	34	36	46	19	19	33	15	22	19	8	8
SPV 351 (ICSV 1)	36	38	31	35	31	57	35	30	32	30	29	17	13	4	2	11
ICSV 91008	30	43	38	18	15	56	14	62	25	17	11	15	21	22	3	5
CSH 9	27	58	28	26	28	35	47	49	35	27	38	20	11	34	4	7
SPV 104 ²	40	70	34	30	11	39	43	39	24	13	27	21	21	7	6	4
IS 8545 ³	7	35	21	17	9	31	49	49	4	2	10	9	3	10	2	6
Mean	34	64	36	32	19	34	30	38	23	18	21	18	14	14	5	8
LSD (<i>P</i> <0.05)	17.1	12.1	14.9	15.4	9.9	10.3	12.3	17.1	15.5	8.8	9.1	11.8	6.9	7.9	6.9	7.0

1. Each value is mean of 2 replications; 50 sorghum grains per replication were tested using blotter technique at 28°C for 3 days.

AKL = Akola, PAR = Parbhani, PAL = Palem, PAT = Patancheru.

2. Susceptible check.

3. Resistant check.

Table 3. Infection severity of fungi on the Sorghum Grain Mold Variability Nursery-2002 entries at hard-dough (HD) stage, physiological maturity (PM), and post-physiological maturity (PPM) under field conditions at Patancheru, India¹.

Entry	<i>Fusarium moniliforme</i>			<i>Curvularia lunata</i>			<i>Phoma sorghina</i>		
	HD	PM	PPM	HD	PM	PPM	HD	PM	PPM
IS 18758C-618-2	2.2	1.7	2.9	1.7	2.0	3.4	2.1	2.3	2.4
IS 18522	1.9	2.3	3.0	1.2	1.9	2.7	1.0	2.6	2.4
ICSV 96101	1.5	1.5	2.4	1.1	1.4	2.4	1.1	2.5	2.6
CS 3541	1.9	1.9	3.1	1.2	1.4	2.7	1.0	2.5	2.4
Sepon/78-1	1.0	1.6	2.2	1.0	1.2	2.3	1.0	1.5	2.0
ICSV 95001	2.2	1.9	2.6	1.3	1.9	2.6	1.0	1.9	2.1
IS 30469C-140	1.4	1.6	2.6	1.4	2.0	3.1	1.2	2.4	2.5
SPV 351 (ICSV 1)	1.2	1.3	2.8	1.1	1.2	2.7	1.3	2.2	2.4
ICSV 91008	1.4	1.3	2.2	1.3	1.3	2.8	1.0	1.7	2.4
CSH 9	1.6	1.8	2.8	1.4	1.7	2.4	1.0	2.5	2.8
SPV 104 (susceptible check)	2.1	2.1	3.2	1.3	2.3	3.9	1.1	2.4	2.4
IS 8545 (resistant check)	1.1	1.0	1.4	1.1	1.0	1.1	1.0	1.0	1.4
Mean	1.6	1.6	2.6	1.2	1.6	2.7	1.1	2.1	2.3
LSD ($P < 0.05$)	0.31	0.29	0.33	0.26	0.27	0.34	0.17	0.27	0.29

1. Each value is mean of 2 replications with 10 panicles per replication, and is based on 1-5 scale where 1 = no mold, 2 = 1-10% mold, 3 = 11-25% mold, 4 = 26-50% mold and 5 = >50% grains molded on a panicle.

low frequencies and may not be of much consequence to grain molding. Among locations, based on frequency mean, *F. moniliforme* was most predominant at Parbhani (64%) and least at Patancheru (32%); *A. alternata* most at Patancheru (38%) and least at Akola (19%); *C. lunata* most at Akola (23%) and least at Parbhani and Patancheru (18%); and *P. sorghina* most at Akola and Parbhani (14%) and least at Palem (5%) (Table 2).

Frequency of mold fungi at different grain development stages. At the hard-dough stage, *F. moniliforme* was most dominant (mean score 1.6), followed by *C. lunata* (mean score 1.2), and *P. sorghina* (mean score 1.1) while other fungi were either absent or in traces on certain sorghum lines at Patancheru (Table 3). Grain mold severity scores at PM and PPM were almost high for all fungi. These results reveal the pathogenic nature of *F. moniliforme*, *C. lunata* and *P. sorghina* that might have infected the developing grains during the flowering to milk stages, while other fungi may be weakly pathogenic or saprophytic. We suggest that screening and evaluation for grain mold resistance should focus primarily on the major pathogenic fungi.

Weather variables and grain mold severity. The period of flowering to PPM varied from 34 days at Patancheru to 59 days at Palem, and considerable variations in temperature and RH were recorded across locations (Table 4). The mean temperature varied from 12°C (minimum at Palem) to 39°C (maximum at Akola); RH from 21% (minimum at Akola) to 100% (maximum at

Parbhani); the number of rainy days from 5 (at Parbhani) to 12 (at Akola); and rainfall from 22 mm (at Parbhani) to 166 mm (at Akola). The threshed grain mold severity score on the susceptible check SPV 104 was maximum (5.0) at Patancheru and minimum (3.8) at Akola and Parbhani; it was 4.2 at Palem. In general, higher RH seems to have positive correlation with mold severity. A detailed analysis of individual weather variables in relation to infection by individual mold fungi at each location is required to better understand the weather-mold relationships.

Isolation of fungi. We made about 500 isolations of *Fusarium* spp from the molded sorghum grain samples from different locations. These cultures would be studied for speciation and their potential for fumonisins production under the ICRISAT-USAID collaborative project at Iowa State University, Ames, Iowa, USA.

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Table 4. Weather variables at four locations in India in the Sorghum Grain Mold Variability Nursery-2002 during rainy season 2002.

Location	No. of days ²	Temperature ¹ (°C)		Relative humidity ¹ (%)		No. of rainy days	Total rainfall (mm)
		Minimum	Maximum	Minimum	Maximum		
Akola	48	18-24	25-39	21-98	53-98	12	166
Parbhani	42	17-33	29-37	29-76	68-100	5	22
Palem	59	12-22	26-36	26-85	66-95	6	92
Patancheru	34	14-27	26-38	25-85	79-99	6	69

1. Range.

2. From flowering to PPM; sprinkler irrigation was provided at all locations except Palem during this period.

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Sorghum Grain Mold: Resistance Stability in Advanced B-lines

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Introduction

Grain mold resistance breeding in sorghum (*Sorghum bicolor*) at ICRISAT and in Indian national programs has focused on developing varieties, restorer lines, and hybrid seed parents utilizing resistance from germplasm

lines of diverse geographical origin. During the past few years, ICRISAT has developed a large number of high-yielding, grain mold resistant B-lines using pedigree breeding with single- and three-way crosses and selecting the progenies under high disease pressure in field screenings (Reddy et al. 2000). Resistance stability of some selected elite B-lines was tested through a collaborative Sorghum Grain Mold Resistance Stability Nursery (SGMRSN) established in 2002. The results of trials conducted at diverse locations in India are presented.

Materials and Methods

The nursery and its management. The SGMRSN is a collaborative nursery between ICRISAT and the All India Coordinated Sorghum Improvement Project (AICSIP) of National Research Centre for Sorghum (NRCS) under the Indian Council of Agricultural Research (ICAR), coordinated by ICRISAT. The nursery was established at Akola, Parbhani, Palem and Patancheru in India. It included 43 F₆ to F₈ male-sterility maintainer progenies from 17 crosses involving 20 B-lines, 8 inbred lines and one B-line population that had shown desirable agronomic traits and grain quality, moderate grain yield potential and moderate to high level of grain mold resistance at Patancheru, and two resistant and two susceptible check lines.

Each entry was grown in 2 rows, 4 m long in 2 replications. The recommended agronomic and cultural practices were followed at each location. Sprinkler irrigation was provided on dry days for 30 min per day in the evening during the flowering to post-physiological maturity (PPM) stages to maintain high relative humidity (RH) (>95%). No artificial inoculation was done with any mold fungi. Damage by insect pests, particularly by shoot fly, stem borer and head bug was minimized by timely application of pesticides.