Sources of Resistance to Sorghum Grain Molds

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ABSTRACT

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Selected accessions from the world collection of sorghum germ plasm were systematically screened in the field during the 1980-1985 rainy seasons to identify sources of grain mold resistance. Of 26,564 accessions in the collection, 7,132 that flowered and matured during the rainy season at ICRISAT Center were screened, and 156 genetically diverse lines were selected as resistant. All resistant accessions except one had colored pericarp. However, 24 accessions with colored pericarp had negligible amounts of tannin, and 14 of the 24 lacked the testa layer.

Sorghum grain mold caused by various nonspecialized fungi is an important disease wherever sorghum (Sorghum bicolor (L.) Moench) is grown if moist weather conditions prevail after flowering until grain maturity and before harvest (14). Traditional sorghum cultivars often escape grain molds because they are photoperiod-sensitive and of long duration, with flowering so timed that grain matures only after the rains have ceased (2). These cultivars are generally low to modest yielders, have limited adaptation outside their natural habitat, and often fail to produce grain if rains cease early. In order to increase yield potential and avoid terminal drought stress, most sorghum improvement programs are developing photoperiod-insensitive, short- to mediumduration cultivars that have wide adaptability and mature before the end of the rains. Under such conditions, the cultivars are often infected by grain molds that severely reduce the quantity and quality of marketable grain and make it unacceptable as food (12,14).

The only practical and economical method for control of sorghum grain molds is the use of mold-resistant cultivars. The status of the search for sources of grain mold resistance was reviewed in 1981 by Williams and Rao (14), who emphasized the need to search for better sources of resistance. In 1980, we began systematic screening of the world collection of sorghum germ plasm assembled at the Genetic Resources Unit (GRU) of the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru, India.

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MATERIALS AND METHODS

Selection of test material. Field experiments were conducted during the 1980–1985 rainy seasons at ICRISAT Center. Because of the limited rainy season (June-October) and photoperiod sensitivity of sorghum at Patancheru, we screened only those accessions that flowered and matured grain before the end of the rainy season. We selected for screening only those accessions from the germ plasm collection that flower within 80 days after planting in the rainy season at Patancheru. Flowering data on which

selections were based were obtained from the evaluation data recorded by GRU. The selected lines flowered before the first week of September, and their grains matured during frequent rains in September and October.

Screening method and grain mold rating. The following screening method was used unless indicated otherwise. Trials were sown before the onset of the rainy season in the first half of June so that grain maturing stages coincided with periods of frequent rainfall in September. The sorghum lines were classified into early (less than 66 days to 50% flowering) and medium (66-80 days to 50% flowering) maturity groups. Each maturity group was sown in a separate block to simplify data collection. The plots were sprinkled for 1 hr in the morning if it did not rain the previous night and same morning, and for an additional hour in the evening if it did not rain throughout the day. Overhead sprinkler irrigation was provided from

Table 1. Origin and number of accessions of world collection of sorghum germ plasm screened and found resistant to grain molds at ICRISAT Center, Patancheru, India

Origin	Number of accessions			
Geographical region*	Number of countries	In world collection ^b	Screened (%)	Resistant
East Africa	9	9,018	1,789 (19.8)°	35
West Africa	15	6,110	642 (10.5)	20
Southern Africa	11	2,339	1,103 (47.2)	43
North Africa and Middle East	11	1,737	124 (7.1)	15
Indian Subcontinent	6	4,215	1,524 (36.2)	6
Southeast Asia and Far East	8	258	168 (65.1)	9
North and Central America	8	2,131	1,342 (63.0)	26
South America	3	18	18 (100)	0
Europe	11	245	75 (30.6)	1
USSŘ	1	69	64 (92.8)	0
Australia and Oceania	2	29	22 (75.9)	0
Unknown	(****)	395	261 (66.1)	1
Total	85	26,564	7,132 (26.8)	156

East Africa: Burundi, Ethiopia, Kenya, Rwanda, Somalia, Sudan, Tanzania, Uganda, and Zaire. West Africa: Benin, Burkina Faso, Cameroon, Cape Verde Islands, Central African Republic, Chad, Gambia, Ghana, Ivory Coast, Mali, Niger, Nigeria, Senegal, Sierra Leone, and Togo. Southern Africa: Angola, Botswana, Lesotho, Malagasy Republic, Malawi, Mozambique, Namibia, South Africa, Swaziland, Zambia, and Zimbabwe. North Africa and Middle East: Algeria, Egypt, Iran, Iraq, Israel, Lebanon, Morocco, Saudi Arabia, Syria, Yemen Arabic Republic, and Yemen Democratic Republic. Indian Subcontinent: Afghanistan, Bangladesh, India, Nepal, Pakistan, and Sri Lanka. Southeast Asia and Far East: Burma, China, Indonesia, Japan, Philippines, South Korea, Taiwan, and Thailand. North and Central America: Cuba, El Salvador, Guatemala, Honduras, Mexico, Nicaragua, United States, and West Indies. South America: Argentina, Uruguay, and Venezuela. Europe: Belgium, Cyprus, France, German Democratic Republic, Greece, Hungary, Italy, Portugal, Spain, and Turkey. Australia and Oceania: Australia and Papua New Guinea.

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^bAt ICRISAT Genetic Resource Unit as of June 1986.

e Percentage of accessions screened in parentheses.

flowering to grain maturity (black layer formation) and up to 2 wk later when panicles were harvested. Susceptible checks of variable maturities were sown for comparison. Flowering data were recorded as days to 50% flowering when 50% of the plants had 50% of the spikelets flowering. Panicles were harvested 14 days after maturity and threshed, and grains were evaluated for grain mold severity, called the threshed grain mold rating (TGMR). This is an estimate of the percentage of molded grain surface area and is recorded on a 1-5 scale where 1=no mold visible, 2 = 1-10%, 3 = 11-25%, 4 = 26-50%, and 5 = more than 50% of the grain surface molded. Germination was tested at least 1 mo after harvest by incubating 100 grains in petri dishes lined with wet filter paper at 30 C.

Initial screening. Test lines were sown as unreplicated 4-m row plots. Accessions with TGMRs of up to 3 and with less than 75% of the grain area covered by glumes were selected for further evaluation in advanced screening trials.

Advanced screening. Test lines were sown in two 4-m row plots in a split-plot design (two replications) with accessions assigned to main plots and two inoculation

treatments in subplots. Inocula of the major mold-causing fungi-Fusarium moniliforme Sheld., F. pallidoroseum (Cooke) Sacc., and Curvularia lunata (Wakker) Boedijn—were separately multiplied on autoclaved sorghum grain for 10-14 days at 30 C. Suspensions of spores and mycelium of the three fungi were prepared containing 1×10^6 spores of each fungus per milliliter. At the 50% flowering growth stage, 10 randomly selected panicles of one row in each plot were sprayed with the fungal suspension, tagged, and covered with kraft paper bags ($14 \times 6 \times 42$ cm) that were removed 7-10 days later. Ten panicles of the second row were tagged but not inoculated or bagged. TGMRs were recorded separately on the 20 tagged panicles, and a mean TGMR value for each inoculation treatment was calculated. Accessions with TGMRs of up to 3 and grain germination above 70% were repeatedly screened in subsequent years until 1985.

Presence of testa. All resistant genotypes were examined to determine if they possessed a testa layer. Grains were cut transversely with a sharp scapel near the tip of the embryo toward the stylar

end. Cavity slides were filled with 2% molten water agar, which was allowed to solidify. Then, three cut grains were pressed in the agar, which acted as a support so that the exposed grain surface faced upward parallel to the slide. The exposed grain surface was covered with a drop of immersion oil and examined for the testa layer at 200× magnification with epifluorescence optics of an Olympus fluorescence microscope equipped with HBO 200W mercury lamp, UG-1 exciter filter, and a DM 400 + L420 dichoric mirror-cum-absorption filter.

Estimation of tannin content. The amount of tannin in mature grain was determined by the vanillin assay method, and the values were calculated by substracting the blank with catechin as a standard (10). The results are reported as catechin equivalents (CE %).

RESULTS AND DISCUSSION

We screened 7,132 accessions (26.8%) of the world collection of sorghum germ plasm for resistance to grain mold and identified 156 accessions as resistant in repeated screenings for 2-6 yr during 1980-1985 (Tables 1 and 2). Grain mold pressure was high in all the years as

Table 2. Country of origin, presence of testa, grain tannin content, days to 50% flowering, and threshed grain mold rating (TGMR) of sorghum lines found resistant to grain mold in field screening at ICRISAT Center, Patancheru, India, during 1980–1985 rainy seasons

Accession IS no.	Origin	Race ^a	Testa present (P) or absent (A)	Grain tannin content (CE %)	Number of years screened	Days to 50% flowering ^b	TGMR°
79	Mexico	С	P	3.6	5	62	2.3
529	USA	C	P	2.3	5	58	2.0
620	USA	C	P	1.4	5	52	2.6
621	USA	C	P	1.1	5	58	2.5
523	USA	C	P	1.8	5	60	2.0
524	USA	C	P	1.1	5	61	2.0
625	USA	C	P	3.3	5	56	2.2
715	USA	С	P	1.4	4	66	2.4
1222	China	KB	P	1.3	3	50	2.8
2284	USA	C	P	6.1	2	65	3.0
2333	Sudan	C	P	0.2	6	78	2.2
2453	USA	C	P	1.4	5	59	2.0
2454	USA	C	P	1.3	5	56	2.0
2560	USA	C	P	1.8	5	56	2.0
2821	Zimbabwe	С	P	1.9	5	55	2.0
2825	Zimbabwe	C	P	1.9	5	55	2.0
2867	South Africa	C	P	1.4	6	53	2.0
3413	Zaire	C	P	1.4	5	68	2.2
3547	Sudan	C	P	1.3	6	71	2.3
3789	Taiwan	DB	P	1.0	5	55	2.7
4006	India	C	P	2.5	5	62	2.3
5959	India	CB	P	2.4	4	58	2.5
5047	India	DC	P	2.6	5	59	2.4
5335	India	C	P	2.1	5	51	2.0
7072	Sudan	CB ·	P	2.2	5	63	2.2
7237	Nigeria	C	P	1.6	5	58	2.0
							(continued on next

^a B = bicolor, BC = bicolor-caudatum, C = caudatum, CB = caudatum-bicolor, D = durra, DB = durra-bicolor, DC = durra-caudatum, G = guinea, GB = guinea-bicolor, GC = guinea-caudatum, K = kafir, and KB = kafir-bicolor.

^bMean based on number of years tested.

^c Based on a 1-5 scale where 1 = no mold and 5 = more than 50% of grain surface molded. Values are the maximum of the mean scores of 10 panicles (either inoculated and bagged or not treated) recorded for any year of screening.

^dData not available.

^eMinimum grain mold rating for any year of screening.

Accession		,	Testa present (P) or	Grain tannin content	Number of years	Days to 50%	
IS no.	Origin	Racea	absent (A)	(CE %)	screened	flowering ^b	TGMRc
3219	Uganda	C	P	1.9	5	56	2.0
3385	Unknown	CB	P	4.5	2	51	2.4
3525	Ethiopia	C	P	2.1	5	51	2.2
3545	Ethiopia	C	P	1.9	5	56	2.0
8614	Uganda	C	P	1.2	5	55	2.3
3763	South Africa	C	P	1.1	5	56	2.0
3783	Kenya	Č	P	1.2	3	69	2.9
8848	Kenya	CB	P	2.8	5	56	1.9
058	Kenya	C	P	3.2	2	62	
308	South Africa	C	P		2		2.0
326				2.6	5	55	2.1
	South Africa	C	P	2.4	2	63	2.0
353	South Africa	K	P	1.6	5	61	2.2
470	South Africa	C	P	3.7	5	64	2.2
471	South Africa	C	P	1.2	5	59	2.0
482	South Africa	C	P	3.0	5	61	2.3
484	South Africa	C	P	2.4	5	61	2.1
487	South Africa	C	P	2.7	5	60	2.0
494	South Africa	C	P	3.4	5	57	2.2
498	South Africa	č	P	3.3	5	65	2.1
499	South Africa	Č	P	2.6	5	63	2.0
554	South Africa	Č	P	1.6	2	60	2.0
804	Sudan	СВ	P P	7.2	6	58	
0301	Thailand		P P				2.1
		K		3.3	5	57	2.1
0390	Uganda	CB	P	8.9	2	62	2.0
0646	USA	CB	P	6.9	2	61	2.4
0892	USA	CB	P	3.0	5	62	2.0
0942	USA	C	P	5.1	5	54	2.0
1227	Ethiopia	C	P	4.0	5	59	2.4
1234	Ethiopia	C	P	3.2	5	59	2.1
2718	USA	D	P	4.4	5	59	2.1
2732	China	D	P	5.9	5	59	2.0
2750	China	Č	P	d	4	62	2.5
2930	Ethiopia	СВ	P	1.5	5	59	
2932	China	CB	P				2.0
				2.1	5	58	2.2
3267	India	DC	P -	2.9	5	55	2.2
3615	Uganda	DC	P	2.6	5	56	2.4
3714	South Africa	DC	P	1.7	5	60	2.1
3798	South Africa	•••	P	1.6	5	55	2.1
3804	South Africa	CB	P	1.8	5	57	2.0
3817	South Africa	•••	P	3.9	3	62	2.0
3885	South Africa	G	Α	0.8	5	71	2.2
3934	South Africa	CB	P	4.0	5	55	2.2
3945	South Africa	C	P	5.1	5	70	2.0
3958	South Africa	č	P	3.0	5	62	
3965	South Africa	СВ	-		_		2.1
3969	South Africa		P P	2.8	5	56	2.1
4291		C		5.7	5 5	60	2.0
	Botswana	СВ	P	2.7		63	2.2
4375	Zimbabwe	G	A	0.3	6	71	2.1
4380	Zimbabwe	G	A	0.3	6	69	2.4
4384	Zimbabwe	G	Α	0.3	6	70	2.6
4385	Zimbabwe	G	P	6.5	5	71	2.7
4387	Zimbabwe	GB	P	4.2	6	68	2.0
4388	Swaziland	DC	P	3.4	6	58	2.0
4390	Swaziland	G	Α	0.1	5	68	2.4
4756	Ethiopia	CB	P	4.7	2	66	2.0
5106	Cameroon	CB	P	2.6	2	66	2.0
5119	Cameroon	C	P	3.9	2	74	
5948	Cameroon	C	P P				3.0
7141				2.7	2	79 70	3.0
7141 8079	Nigeria	DC GP	P	0.7	6	70	2.3
	Lebanon	CB	P	9.4	2	62	2.0
8135	Lebanon	•••	P	0.2	2	59	2.0
3139	Lebanon	•••	P	8.4	2	62	2.0
3140	Lebanon	•••	P	6.3	2	64	2.1
8141	Lebanon	•••	P	5.2	2	67	2.0
8144	Lebanon		P	10.7	2	64	2.0
8146	Lebanon		P	5.0	2	53	
8149	Lebanon	•••	P	5.1	2		3.0
8153		•••			2	55	2.4
	Lebanon		P	4.0	2	60	2.0
8154	Lebanon	•••	P	3.6	2	52	2.0
8155	Lebanon	•••	P	3.4	2	61	2.0
3165	Lebanon	•••	P	0.2	2	60	3.0
3175	Lebanon	•••	P	7.7	2	67	1.6

Table 2. (continued from preceding page)

Accession			Testa present (P) or	Grain tannin content	Number of years	Days to 50%	
IS no.	Origin	Racea	absent (A)	(CE %)	screened	flowering ^b	TGMR ^c
18219	Lebanon	•••	P	5.0	2	61	2.0
18528	India	•••	P	6.6	6	59	2.0
18759	Nigeria	•••	P	2.0	5	56	2.0
20620	USA	C	P	3.0	5	56	2.3
20639	USA	•••	P	5.4	2	57	2.0
20708	USA	В	Α	0.4	2	56	3.0
20720	USA	В	P	2.0	2	68	2.0
20721	USA	BC	P	1.1	2	69	2.0
20757	USA	C	P	9.2	2	60	2.0
20758 20768	USA	GC 	P	3.9	2	64	2.0
20708	USA USA	GC	P	1.1	2	52	2.8
20835	USA		P P	8.1 3.4	2 2	63	2.0
20843	South Korea	C	r P	2.3	2	53 61	2.0
20844	China	Ċ	P	2.0	2	53	2.0
20861	Iran	GC	P	3.7	2	63	2.4 2.3
20884	Hungary		A	0.2	2	55	2.5
21454	Malawi	G	P	3.1	6	56	2.3
21498	Malawi	DC	P	6.1	6	68	2.1
21509	Malawi	GC	A	0.2	5	64	2.0
21599	Malawi	G	A	0.2	6	72	2.1
22617	Burma	СВ	P	6.6	5	60	2.1
23585	Ethiopia	GC	P	2.2	3	73	2.0
23599	Ethiopia	GC	P	0.1	3	73	2.1
24903	Zambia	GC	P	1.8	3	77	2.6
24911	Zambia	GC	P	2.0	3	78	2.2
24931	Zambia	В	P	6.8	3	70	2.0
24938	Zambia	GC	P	9.8	3	79	2.0
24941	Zambia	СВ	P	9.4	3	78	2.0
24981	Sudan	GC	P	3.0	3	76	2.5
24988	Sudan	GC	P	2.3	3	78	2.4
24989	Sudan	C	P	2.8	3	77	2.1
24992	Sudan	GC	P	1.2	3	75	2.1
24995 24996	Sudan	GC	P	1.8	3	77 22	2.0
24996 25008	Sudan Sudan	GC	P	0.3	3	77	2.0
25008 25010	Sudan	GC CB	P P	0.1	3	73 75	2.9
25010	Sudan	GС	P P	3.0 2.7	3	75 78	2.8
25015	Sudan	GC	r P	2.7	3 3	78 76	3.0
25017	Sudan	C	A	0.2	3	73	3.0 3.0
25022	Sudan	GC	P	2.0	3	73 78	2.0
25025	Sudan	DC	P	0.9	3	75	2.0
25032	Sudan	GC	P	0.2	3	79 79	2.0
25038	Sudan	GC	P	1.5	3	75	2.1
25060	Ghana	GC	P	3.5	3	75	2.0
25069	Ghana	GC	A	0.5	3	73	2.3
25070	Ghana	G	A	0.4	3	74	2.0
25074	Ghana	GC	P	2.9	3	75	2.9
25075	Ghana	GC	P	3.6	3	75	2.2
25084	Ghana	GC	P	2.8	3	74	2.0
25085	Ghana	DC	P	1.8	3	76	2.3
25095	Ghana	GC	P	0.2	3	71	2.4
25098	Ghana	G	A	0.6	3	74	2.1
25100	Ghana	G	A	0.7	3	73	2.0
25102	Ghana	C	P	3.0	3	66	2.5
25103	Ghana	GC	P	4.8	3	65	2.3
25104	Ghana	C	P	2.1	3	72	2.0
25105 Eusaantibla ah	Ghana	GC	P	4.1	3	71	2.1
Susceptible ch			A	0.1	,	67	
CSH 1 SPV 104	India India	•••	A	0.1	6	57	5.0°
102	India USA	•••	A A	0.1 0.1	6 5	62 53	5.0
+02 417	USA		A A	0.1	5 5	52 56	5.0
18758	Ethiopia		A	0.1	6	78	4.8 5.0

shown by the TGMRs of susceptible checks (Table 2). Grain mold ratings of resistant accessions were 3 or below and germination was greater than 70%. Resistance in these accessions was

maintained for 2 wk after maturity, when panicles were harvested. Mold severity in susceptible sorghums usually increases after maturity, and the disease achieves serious proportions when harvesting is delayed after maturity in wet and humid conditions. All resistant lines except IS 25017 had colored pericarp. Under the fluorescence microscope, the testa layer was clearly identified as a dark brown

layer, often with cross-walls, above the aleurone cell layer. The testa layer was absent in 14 resistant accessions. Tannin content in these 14, and in 10 other accessions with testa, was negligible (less than 1.0 CE %). The range of tannin content in the resistant accessions was 0.1–10.7 CE %.

The resistant accessions had wide morphological variability and diversity in taxonomic races (Table 2). The number of resistant accessions of the various races based on the classification by Harlan and de Wet (7) were: caudatum, 56; guinea, 11; bicolor, 3; durra, 2; kafir, 2; guinea-caudatum, 28; caudatum-bicolor, 20; durra-caudatum, 9; kafir-bicolor, 1; bicolor-caudatum, 1; durra-bicolor, 1; and guinea-bicolor, 1. Twenty-one accessions have not been identified to race. The resistant accessions came from 25 countries, suggesting diversity in geographic origin.

Sources of grain mold resistance have been reported from the United States (1,4-6), Senegal (3), and India (9,11), including ICRISAT Center (13). The majority of the mold-resistant accessions reported from the United States have grain characteristics similar to the ones we report in this paper. Grains of these lines have colored pericarp; some have testa and some do not. Tannins in the testa layer have been reported to confer grain mold resistance (8), but tannins are not always associated with grain mold resistance because several resistant accessions reported in this paper had negligible levels of tannin. Sorghums with colored grain usually have poor consumer acceptance in most parts of the world where the grain is used as nonbeverage food. However, the purpose of the present research was not to identify mold-resistant sorghums for direct introduction as cultivars, but to identify sources of mold resistance that can be further used in breeding programs. ICRISAT sorghum breeders are utilizing some of the resistant sources, particularly those without testa and with negligible tannin, to develop mold-resistant sorghums with good grain quality.

Although data are not presented in this paper, our screening results showed that contrary to previous reports in India (9,11,13), the majority of the accessions had low levels of resistance, if any. The screening techniques used by Rao and Williams (13) and by us were similar (inoculation and bagging of panicles), but we harvested the panicles 10 days later than they did. Inoculation and bagging did not offer significant advantages over using untreated panicles, the method used by others (9,11).

Length of the rainy season (mid-June to mid-October) at ICRISAT Center in relation to photoperiod sensitivity of the germ plasm limited screening of the majority of the accessions. The photoperiod-sensitive accessions flowered and filled grains after the rains ceased, thus escaping grain mold. Such lines can be screened at a location where grain filling and postmaturity stages coincide with grain mold-conducive environment.

Seed of the mold-resistant accessions described are available on request from the Genetic Resources Unit of ICRISAT.

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