

Journal of Agriculture and Crops ISSN(e): 2412-6381, ISSN(p): 2413-886X Vol. 2, No. 10, pp: 101-106, 2016

URL: http://arpgweb.com/?ic=journal&journal=14&info=aims

Assessing the Vulnerability of Sorghum Converted Lines to Anthracnose and Downy Mildew Infection

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Abstract: A total of 59 converted sorghum lines and 6 checks were evaluated for resistance to two foliar fungal diseases, anthracnose and downy mildew (SDM) in 2008 and 2009 growing seasons at the Texas A&M AgriLife Research Farm, College Station, Texas. In 2008, 23 lines exhibited resistance (35%), 29 susceptible (45%) and 13 variable responses (20%) while 15 lines showed resistance (28%), 31 susceptible (57%), and 8 variable responses to anthracnose in 2009. Nine lines SC748, PI534101, PI534073, PI533950, PI534155, PI533802, PI533776, PI533911 and PI533759 exhibited anthracnose resistance response in both years. Significantly a wide range of 8 to 89% SDM incidence was observed in the study. None of the lines recorded SDM resistance reaction in both years. However, 15 lines PI534119, PI533983, PI597970, PI534160, PI570726, PI534161, PI534112, PI576374, PI533753, SC748, PI533991, PI569998, PI534050, PI534155 and PI533898 recorded moderate resistance to SDM incidence and recommended for use in further breeding programs. There was a positive significant correlation (P = 0.0392) between anthracnose and SDM, indicating that the lines showing higher SDM incidence favors higher anthracnose infection. Significant correlation between precipitation and SDM was also noted. SC748 and PI534155 exhibited resistance to anthracnose and downy mildew diseases and hold promise for utilization in breeding programs as potential checks.

Keywords: Sorghum anthracnose; Downy mildew; Colletotrichum sublineolum; Peronosclerospora sorghi; Fungal diseases.

1. Introduction

Sorghum (*Sorghum bicolor* L Moench) is the second most important feed grain in the United States. In Africa and Asia more than 500 million people are directly dependent on sorghum as a food crop, the major source of energy and protein [1]. Because of its tolerance to drought, sorghum is an excellent fit for production in semi-arid regions of the world where other cereals struggle. Sorghum lines/hybrids with improved biotic and abiotic stress resistance are required to increase profitability through increased yield (grain/forage/biomass) especially in light of increased production costs. Besides, the value and productivity of sorghum can be reduced greatly by diseases.

Sorghum anthracnose, caused by *Colletotrichum sublineolum* Hann. Kabát *et* Bub. (syn. *C. graminicola* (Ces.) G.W. Wilson and sorghum downy mildew (SDM), caused by *Peronosclerospora sorghi* (Weston & Uppal) C. G. Shaw, are two destructive diseases of sorghum worldwide [2, 3]. Anthracnose is detected in most sorghum growing regions worldwide and yield losses up to 50% may occur when susceptible lines are planted [4-6]. Sorghum anthracnose affects sorghum leaves, panicle, and stalk and significantly reduces grain quality [7]. *C. sublineolum* reproduces asexually by producing acervuli, a fruiting body with cushion like structure. Acervuli detection in sorghum foliar are often used as a diagnostic sign for anthracnose susceptibility [7-9].

SDM is reported in many sorghum producing regions worldwide with yield losses ranging from 11.7 to 78% reported in susceptible lines. [10-13]. *P. sorghi*, the pathogen of SDM is an oomycte that can survive for several years in soil and plant debris by producing soil borne oospores [14]. The oospores germinate in response to the germinating seed and are the primary source of inoculum for SDM [15]. Once in the plant, the foliar meristem tissues are colonized, resulting in the chlorotic appearance of the seedling leaf [15]. In these systemically-infected plants moisture promotes development of sporangiophores and sporangia on leaves and subsequent infection of nearby plants by the asexual sporangia [16]. Conidia from the sponrangia are short-lived, but play a significant role in spreading the disease [17] and in many sorghum growing regions they serve as the primary source of inoculum [18]. Later in the season, oospores subsequently develop in systemically-infected leaves, which fall onto the ground as the leaf shreds, completing the disease cycle. Six pathotypes of *P. sorghi* have been identified based on host differential [11, 19]. Several management strategies have been applied to control downy mildew, including metalaxyl seed treatment [19] which proved to be efficient for several years; however, due to the variable nature of *P. Sorghi*, new pathotype (P6) that is metalaxyl resistant emerged recently [19].

Owing to its tropical origin, sorghum is a short-day plant that requires day lengths of less than 12 h 20 min to induce flowering, and hence much of the world collection of 36,000 accessions flowers too late and tall, to be exploited for seed production in temperate-zone environments. These materials are potential new sources of desirable traits such as disease and insect resistance, drought resistance, and improved grain quality, and should be useful to breeders and other sorghum researchers in developing improved lines and hybrids. Hence, the sorghum cultivars were developed through a backcross procedure in which tall, late-maturing tropical sorghum cultivars were converted to early-maturing, combine height enhanced germplasm resources [20-22]. These lines represented new sources of germplasm from the World Sorghum Collection and are of a height and maturity to make them readily usable in the United States and other temperate-zone areas of the world. Due to the hypervariability of the anthracnose and downy mildew pathogens, there is a continuous need to identify new resistant sources. Thus, the aim of this study was to evaluate a subset of 60 exotic sources for resistance to anthracnose and downy mildew.

2. Materials and Methods

A subset of 59 sorghum converted lines of geographically diverse (USA, India, Ethiopia, Mali, Sudan, Honduras, Rhodesia, Nigeria, Uganda and Japan) origin were used along with checks SC748, SC719, RTx2911, BTx623, RTx2536 and Sureno in this study. The experiment was conducted during the 2008 and 2009 growing seasons at the Texas A&M AgriLife Research Farm, near College Station, Texas. Lines were planted in 6 m rows at 0.76 m spacing between rows in a randomized complete block design, and each accession replicated three times. Field preparation included fall plowing and incorporation of the compound fertilizer at 175, 116.5 and 116.5 respectively N, P_2O_5 and K_2O kg/ha. An additional 175 kg N/ha was applied as top dressing 5 weeks after planting. A pre-emergent insecticide 'Counter 20 CR' (BASF Group, Southfield, MI) and herbicide 'Atrazine' (Syngenta Crop Protection Inc. Greenboro, NC) were applied before planting to control weeds and protect against seedling insects.

The inoculation technique, disease assessment, and rating methods for the anthracnose trials have been previously described [8, 9]. Briefly, plants were inoculated 30 days after planting by placing C. sublineolumcolonized grain into plant whorls. Disease assessments were conducted 30 days post-inoculation and thereafter, on a weekly basis until the flowering stage. Ratings were based on a scale of 1 to 5, where 1 = no symptoms or chlorotic flecks on leaves; 2 = hypersensitive reaction (reddening or red spots) on inoculated leaves but no acervuli formation and no spreading to other leaves; 3 = lesions on inoculated and bottom leaves with acervuli in the center; 4 =necrotic lesions with acervuli on the bottom and middle leaves; and 5 = most leaves dead due to infection with infection on the flag leaf containing abundant acervuli. Symptom types were then categorized into two reaction classes, resistant rated as 1 or 2; and susceptible rated as 3, 4, or 5. The incidence of SDM systemic and local lesion symptoms were assessed 4 weeks after planting and again at flowering. Plants showing systemic and/or local lesions were counted as infected. Disease incidence was determined from the percentage of infected plants within each row, and at least 20 plants in each replication were evaluated for disease screening. Disease ratings were followed as described by Frederiksen [17]: <6 % incidence = resistant; 6–10 % = moderately resistant; 11–20 % = moderately susceptible; >20 % = susceptible. Disease incidence data were averaged across years (2008 and 2009), arcsine transformed and subjected to the analysis of variance using the command PROC GLIMMIX (SAS version 9.3, SAS Institute, Cary, NC) to determine the main effect of sorghum accessions. Mean comparisons among the lines were based on Tukey-Kramer at the 5% probability level.

3. Results and Discussion

The hypervariability in the populations of *C. sublineolum* and *P. sorghi*, the pathogens that incite anthracnose and SDM, respectively, requires continuous monitoring of changes in the pathogenic population and identification of new resistant sources [6, 11, 19, 23, 24]. Several studies have been conducted from different sorghum growing regions worldwide to identify new sources for anthracnose and SDM pathogens [3, 8, 25-30].

During the 2008 field trial, 23 lines exhibited resistance to anthracnose (35%), 29 exhibited susceptible response (45%) and 13 showed variable responses (20%). Whereas, in the 2009 trial, 15 lines exhibited resistance response (28%), 31 were susceptible (57%) and 8 were variable responses (15%). Nine lines SC748, PI534101, PI534073, PI533950, PI534155, PI533802, PI533776 PI533911 and PI533759 exhibited resistance response in both years.

Whereas, 22 lines including PI534163, PI533934, PI533815, RTx2911, SC719, PI595699, PI534119, PI533983, PI533993, PI534022, PI534017Sureno, PI534160, PI533818, PI533755 and PI534112 exhibited susceptible response (Table 1). Hess, *et al.* [27] evaluated 19 sorghum lines at two locations in Mali and recorded 12 lines with high levels of resistance; however, none of the lines was completely resistant. Sharma, *et al.* [31] evaluated the sorghum minicore germplasm from India and found 13 accessions resistant.

In this study, the lowest SDM incidence ≤ 10 % infection was exhibited by 15 lines: PI534119, PI533983, PI597970, PI534160, PI570726, PI534161, PI534112, PI576374, PI533753, PI533991, SC748, PI569998, PI534050, PI534155 and PI533898 and are considered as moderately resistant. Six accessions: PI571342, PI534022, PI533892, PI533894, PI533802 and PI533776 recorded the highest SDM incidence of >80%. A positive significant correlation (*P*= 0.039) between anthracnose and SDM indicated that infection of SDM favors higher level of anthracnose infection. Significant positive correlation between precipitation and SDM indicated that high rainfall favors SDM infection (Table 2).

Two lines SC748 and PI534155 from Sudan and Ethiopian origin exhibited the resistance to anthracnose and moderately resistant to downy mildew diseases in two years (2008 and 2009) evaluation. These two converted lines and derived progenies were the most stable for anthracnose resistance across different environments [2, 11, 25, 29, 32, 33]. A dominant gene (*Cg1*) derived from cross between SC748 and BTx623 was mapped at the distal region of chromosome five in sorghum [34].

4. Conclusion

This work is significant because it has identified a number of potential new sources of anthracnose and downy mildew resistance in sorghum that can be utilized by breeders in the USA and abroad to develop new lines and hybrids.

Converted/	Origin	Anthracnose		SDM		
adapted lines	origini	2008 2009		Incidence*	Reaction	
PI534163	USA	S	S	19.0 abcd	MS	
PI533934	India	S	S	49.0 abcd	S	
PI533815	India	S	S	53.0 abcd	S	
RTx2911	USA	S	S	18.0 abcd	MS	
BTx623	USA	S	S	14.0 abcd	MS	
SC719-11E	Sudan	S	S	37.0 abcd	S	
PI595699	USA	S	S	70.0 abcd	S	
PI571342	Sudan	S	-	89.0 a	S	
PI534119	Ethiopia	S	S	8.0 d	MR	
PI533983	Sudan	S	S	9.0 d	MR	
PI533993	Sudan	S	S	13.0 bcd	MS	
PI534022	India	S	S	86.0 a	S	
PI570841	Sudan	S	-	71.0 abcd	S	
PI571012	Sudan	S	-	32.0 abcd	S	
PI534017	India	S	S	11.0 cd	MS	
PI576405	India	S	S	19.0 abcd	MS	
PI576360	India	S	-	71.0 abcd	S	
RTx2536	USA	S	S	58.0 abcd	S	
Sureno	Honduras	S	S	30.0 abcd	S	
PI597970	USA	S	-	8.0 d	MR	
PI534160	unknown	S	S	9.0 d	MR	
PI570726	Sudan	S	-	8.0 d	MR	
PI534053	Uganda	S	S	20.0 abcd	MS	
PI533818	Nigeria	S	S	46.0 abcd	S	
PI533755	Unknown	S	S	73.0 abc	S	
PI534096	Mali	S	V	76.0 ab	S	
PI534161	Nigeria	S	S	9.0 d	MR	
PI533975	Rhodesia	S	S	23.0 abcd	S	
PI534112	India	S	S	8.0 d	MR	
PI576374	India	V	S	8.0 d	MR	
PI595712	USA	V	R	17.0 abcd	MS	
PI534097	Japan	V	R	67.0 abcd	S	
PI534162	India	V	S	12.0 bcd	MS	
PI534116	Ethiopia	V	S	33.0 abcd	S	
PI569979	Sudan	V	-	38.0 abcd	S	
PI533892	Nigeria	V	S	84.0 a	S	

Table-1. Response of exotic sorghum lines to anthracnose and downy mildew

PI533787	Unknown	V	R	17.0 abcd	MS
PI533753	Unknown	V	S	8.0 d	MR
PI533991	Nigeria	V	R	8.0 d	MR
PI533894	Nigeria	V	R	85.0 a	S
PI533903	Ethiopia	V	R	56.0 abcd	S
PI533885	Nigeria	V	S	52.0 abcd	S
PI597971	USA	R	V	37.0 abcd	S
SC748	Sudan	R	R	8.0 d	MR
PI597977	USA	R	V	40.0 abcd	S
PI576418	Nigeria	R	V	66.0 abcd	S
PI576343	Nigeria	R	S	46.0 abcd	S
PI576409	Sudan	R	-	11.0 cd	MS
PI571184	Sudan	R	-	12.0 bcd	MS
PI569853	Sudan	R	-	61.0 abcd	S
PI569998	Sudan	R	-	9.0 d	MR
PI534101	Japan	R	R	12.0 bcd	MS
PI534164	Uganda	R	V	25.0 abcd	S
PI534145	Rhodesia	R	S	77.0 ab	S
PI534050	Burkina Faso	R	V	10.0 cd	MR
PI534073	Nigeria	R	R	48.0 abcd	S
PI533950	Sudan	R	R	49.0 abcd	S
PI534155	Ethiopia	R	R	8.0 d	MR
PI534088	Nigeria	R	S	69.0 abcd	S
PI534042	Sudan	R	V	58.0 abcd	S
PI533802	Unknown	R	R	84.0 a	S
PI533776	Unknown	R	R	86.0 a	S
PI533911	Sudan	R	R	21.0 abcd	S
PI533898	Nigeria	R	V	9.0 d	MR
PI533759	Unknown	R	R	11.0 cd	MS

Anthracnose: Ratings were based on a scale of 1 to 5 by Prom, *et al.* [9], where 1 = no symptoms or chlorotic flecks on leaves; 2 = hypersensitive reaction (reddening or red spots) on inoculated leaves but no acervuli formation; 3 = lesions on inoculated leaves and bottom leaves with acervuli in the center; 4 = necrotic lesions with acervuli on the bottom and middle leaves; and 5 = most leaves dead due to infection with infection on the flags leaf containing abundant acervuli. A rating of 1 or 2 is considered a resistant (R) response; a rating of 3, 4, or 5 is considered a susceptible (S) response and a variable resistant and susceptible response (V) between replications.

SDM: Percent incidence was based on at least 15 plants per accession per row. Reaction: <6% = resistant, R; 6–10 % = moderately resistant, MR; 11–20 % = moderately

susceptible, MS; >20 % = susceptible, S

* Means combined over two years within a column followed by the same letter(s) are not significant at the 5% probability level based on Tukey Kramer adjustment for multiple comparisons.

	Anthrac		Tmax		Tmin		Precip	
	r	Р	R	Р	r	Р	r	Р
SDM	0.188	0.039**	-0.069	0.448	0.012	0.907	0.234	0.010***
Anthrac			-0.078	0.396*	0.133	0.100	0.111	0.227
Tmax					0.890	0.001*	-0.292	0.001***
Tmin							-0.220	0.016**

Table-2. Correlation coefficients among anthracnose (Anthrac), downy mildew (SDM), and weather parameters during the two growing seasons in College Station, TX

Tmax = Maximum daily temperature during the evaluation period.

Tmin = Minimum daily temperature during the evaluation period.

Precip = Daily precipitation during the evaluation period.

*, **, *** significance at the 10%, 5% or 1% probability level, respectively.

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