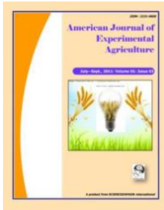

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The Impact of Weather Conditions on Response of Sorghum Genotypes to Anthracnose (*Colletotrichum sublineola*) Infection

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Authors' contributions

This work was carried out in collaboration between all authors. Author LKP conceived and designed the study, performed the statistical analysis, and wrote the first draft of the manuscript. Authors RP and GR assisted in inoculating the plants and data collection. Authors TI, CM and WLR assisted in field operations, including planting, inoculating, and disease assessments, data collection, and reviewed the experimental design and all drafts of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Rainfall is a major climatic factor influencing anthracnose development. In this study, 68 sorghum accessions were evaluated for anthracnose resistance under dry and wet growing conditions at the Texas A&M Agricultural Experiment Station, near College Station, Texas. Accessions, planted in a randomized complete block design with three replications, were inoculated with a mixture of

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Colletotrichum sublineola isolates 30 days after planting. Under dry growing conditions, three accessions showed a susceptible response across replications, whereas 41 accessions exhibited susceptibility under wet growing conditions. Also, 15 accessions that showed variation in susceptibility across replications under dry conditions were rated as susceptible across replications under wet growing conditions. Nineteen accessions consistently showed a resistant response under both dry and wet growing conditions. There was no significant correlation between weather variables and anthracnose development during the dry growing season, suggesting that climatic conditions were unfavorable for disease development. In contrast, there was a significant positive correlation between total rainfall and anthracnose infection and moderately significant relationships between number of days with rain and minimum relative humidity, with anthracnose infection during wet growing conditions. These results indicate that frequency and cumulative rainfall, as well as relative humidity are critical factors for disease development.

Keywords: Sorghum anthracnose; sorghum bicolor; foliar disease; Colletotrichum sublineola.

1. INTRODUCTION

Anthracnose, caused by *Colletotrichum sublineola* P. Henn, in Kabat and Bubák (syn. *C. graminicola* (Ces.) G. W. Wilson), is one of the most damaging diseases of sorghum and it is presently found in most growing areas [1-3]. The leaves, stalks, and panicles can be infected by the fungus although foliar infection is most common [4]. Foliar infection can occur at any stage of plant development, but symptoms are generally observed 40 days after seedling emergence. Symptom development on the leaves will depend on the cultivar and environmental conditions [4]. Symptoms may appear as small circular to elliptical spots or elongated lesions, and as the fungus sporulates, fruiting bodies (acervuli) appear as black spots in the center of the lesions [4]. Estimating grain yield losses due to foliar anthracnose infection can often be difficult [5], but losses as high as 50% have been reported in susceptible cultivars [6-8]. The occurrence of different pathotypes and levels of pathogenicity within the pathogen population require the identification of different sources of resistance [2,9-11]. Weather conditions, in particular rainfall, have been shown to play a critical role in the incidence and severity of sorghum anthracnose [4,12-16]. In this communication, we report the influence of weather conditions on the severity of anthracnose on sorghum accessions inoculated with a mixture of *C. sublineola* isolates.

2. MATERIALS AND METHODS

2.1 Experimental Materials and Design

Sixty-eight accessions were randomly selected from the Ethiopia, Mali, Sudan, and Uganda sorghum collections maintained by the US National Germplasm System. Seed samples for the anthracnose evaluation were provided by the USDA-ARS, Plant Genetic Resources Conservation Unit, Griffin, Georgia. Sorghum genotype BTx623 was included in the evaluation as the susceptile control while SC748-5 was included as a resistant control. Accessions were planted in a randomized complete block design with three replications in 2009 at the Texas A&M Agricultural Experiment Station, in Burleson county near College Station, Texas. During the 2009 growing season, summer (April-August) growing conditions were very dry followed by wet growing conditions in the fall (September-November), which provided a unique opportunity to evaluate the effect of weather variables on anthracnose disease development. Weather data during the two growing periods are presented in Table 1. Seed was planted in 6 m rows at 0.31 m spacing between rows. Field preparation included fall plowing and incorporation of NPK according to local recommendation. To control weeds and seedling insects, a pre-emergent insecticide ‘Counter 20 CR’ (BASF Group, Southfield, MI) and herbicide ‘Atrazine’ (Syngenta Crop Protection Inc. Greenboro, NC) were applied before planting. The trial was planted in April and disease assessment conducted in June and July and in late August, plants within each plot were cut about 15.2 cm above the ground, the debris removed, and plants allowed to tiller for the second disease assessment.

2.2 Inoculation Technique

The inoculation technique and disease assessment method followed the procedures described by Prom et al. [17]. Briefly, sorghum plants were inoculated 30 days after planting by placing 10 *C. sublineola*-colonized grains onto the leaf whorls. A mixture of anthracnose isolates was used. For the evaluation conducted under wet growing conditions, tillers were inoculated 30 days after the plants were cut. Disease assessments were conducted 30 days post-inoculation and thereafter, on a weekly basis for four weeks. Ratings were based on a scale of 1 to 5 [14,17], 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 lesion development on other leaves; 3 = lesions on inoculated and bottom leaves with acervuli in the center; 4 = necrotic lesions with acervuli on inoculated leaves and infection spreading to bottom and middle leaves; and 5 = most leaves dead with abundant acervuli on the flag leaf. Accessions were considered resistant if plants in the row were rated as 1 or 2 and susceptible if rated as 3, 4, or 5.

2.3 Statistical Analysis

Pearson's correlation coefficients were calculated between anthracnose rating and weather parameters such as maximum average temperature, minimum average temperature, maximum average relative humidity, minimum average relative humidity, total rainfall, and number of days with rain for the two seasons. All these weather parameters were noted for the dry and wet growing conditions starting a week before inoculation and ending 8 weeks post-inoculation. Using PROC REG (SAS Institute, SAS version 9.2, Cary, NC) with stepwise as the selection option, the best model for the dry and wet periods anthracnose score were determined.

3. RESULTS AND DISCUSSION

In this study, anthracnose response was significantly affected by accession (P<0.001) under both dry and wet growing conditions. For the dry growing conditions, three accessions showed a susceptible response with 15 accessions showing a variation in susceptibility (Table 2).

While under the wet growing conditions, 41 accessions exhibited susceptibility across replications and eight showed a variation in susceptibly across replications. Nineteen accessions that were rated as resistant and three accessions that were rated as susceptible showed a similar disease response across the two evaluations. Erpelding and Prom [14] evaluated 270 accessions from the Mali working collection in Isabela, Puerto Rico and noted higher number of susceptible accessions during the wet season than the dry season. Studies have shown that the environmental conditions during evaluation of sorghum germplasm have profound influence on anthracnose infection response [4,12,13,15]. Néya and Le Normand [12] reported a higher number of susceptible genotypes and higher anthracnose severities in locations with favorable weather conditions, such as high relative humidity, warmer temperature, and higher total cumulative rainfall. Higher anthracnose severities on sorghum germplasm were also noted in wetter regions of Nigeria [13]. In this study, 23 accessions with a resistant response to anthracnose under dry growing conditions exhibited susceptibility when evaluated under wet conditions (Table 2). Five accessions (PI563301, PI568373, PI570743, PI217674 and PI570873) from Sudan, 7 accessions (PI609009, PI609044, PI608974, PI608986, PI609991, PI609947, and PI609008) from Mali, and 7 accessions (PI297093, PI154758, PI330977, PI154966, PI584214, PI584284, and PI330985) from Uganda exhibited resistant responses in both the dry and wet periods (Table 3). Further, 15 accessions with a variable response across replication under dry conditions were found to be susceptible under wet growing conditions.

Table 1. Weekly averaged weather data for the dry (summer) and wet (fall) growing conditions in 2009[a]

Weather variable	Dry	Wet
21 days after planting (DAP)		
Total rainfall (mm)	0	3.81
Number of days with rain	0	4
Maximum temperature (ºC)	32.6	28.3
Minimum temperature (ºC)	18.4	19.4
Maximum relative humidity (%)	89.2	91.8
Minimum relative humidity (%)	29.2	56.9

Weather variable	Dry	Wet
37 DAP (First week post-inoculation)		
Total rainfall (mm)	0	19.8
Number of days with rain	0	4
Maximum temperature (ºC)	33.2	24.7
Minimum temperature (ºC)	18.8	17.3
Maximum relative humidity (%)	89.7	94.4
Minimum relative humidity (%)	33.8	71.8
44 DAP (Second week post-inoculation)		
Total rainfall (mm)	0	0
Number of days with rain	0	0
Maximum temperature (ºC)	36.1	26.7
Minimum temperature (ºC)	24.0	14.4
Maximum relative humidity (%)	87.5	92.9
Minimum relative humidity (%)	36.0	46.7
51 DAP (Third week post-inoculation)		
Total rainfall (mm)	0	13.7
Number of days with rain	0	4
Maximum temperature (ºC)	36.9	22.6
Minimum temperature (ºC)	23.2	10.8
Maximum relative humidity (%)	90.0	95.3
Minimum relative humidity (%)	29.9	53.3
58 DAP (Fourth week post-inoculation)		
Total rainfall (mm)	0	0.3
Number of days with rain	0	3
Maximum temperature (ºC)	39.2	24.6
Minimum temperature (ºC)	24.3	8.4
Maximum relative humidity (%)	82.1	94.3
Minimum relative humidity (%)	24.8	38.5
1st Disease assessment week [65 DAP (5th week post-inoculation)]		
Total rainfall (mm)	0.23	1.27
Number of days with rain	1	4
Maximum temperature (ºC)	36.4	25.3
Minimum temperature (ºC)	24.3	11.6
Maximum relative humidity (%)	86.4	94.4
Minimum relative humidity (%)	34.0	47.2
2nd Disease assessment week [72 DAP (6th week post-inoculation)]		
Total rainfall (mm)	0.10	3.6
Number of days with rain	2	5
Maximum temperature (ºC)	38.9	23.6
Minimum temperature (ºC)	24.7	9.2
Maximum relative humidity (%)	86.6	95.8
Minimum relative humidity (%)	24.8	46.9
3rd Disease assessment week [79 DAP (7th week post-inoculation)]		
Total rainfall (mm)	0.76	4.1
Number of days with rain	2	5
Maximum temperature (ºC)	37.9	19.4
Minimum temperature (ºC)	23.5	10.6
Maximum relative humidity (%)	88.5	91.4
Minimum relative humidity (%)	28.1	58.0
Final disease assessment week [86 DAP (8th week post-inoculation)]		
Total rainfall (mm)	0.13	0.13
Number of days with rain	1	1
Maximum temperature (ºC)	36.1	17.8
Minimum temperature (ºC)	24.3	2.3
Maximum relative humidity (%)	88.2	94.7
Minimum relative humidity (%)	31.5	25.7

[1a] *Data obtained from: http://researchfarm.tamu.edu/test.txt*

A significant positive correlation between total rainfall and anthracnose infection was noted during the wet growing conditions, indicating that rainfall plays a critical factor in the disease development (Table 4). The number of days with rain and minimum relative humidity also appear to be vital in anthracnose development as indicated by the moderately significant relationships. In the study, 1.22 mm of total rain on 6 rainy days were noted during the dry season, while 46.7 mm of rain on 30 rainy days were noted during the wet season (Table 1). Modeling disease score for the dry season showed that none of the measured variables met the significant level for entry into the model. While in the wet season, minimum average temperature with $R^2 = 0.94$ and precipitation with $R^2 = 0.99$ were the two variables with significant levels for entry into the model. This would suggest that minimum average temperature and rainfall are critical elements in anthracnose development within sorghum fields. Several studies have shown the importance of rainfall in creating high levels of anthracnose infection in sorghum [4,12,13,15,16,18,19]. Rain plays an integral part in the dispersal and subsequent germination of *C. sublineola* conidia [4]. Thus, anthracnose is more severe in areas with higher annual rainfall [4,12,13,15]. Hess et al. [18] observed higher anthracnose severity on susceptible sorghum genotypes in regions of higher annual rainfall in Mali. Ngugi et al. [19] noted that higher rainfall and humidity are important factors in creating epiphytotics in sorghum. Rainfall has also been shown to be a critical factor in other sorghum diseases. Tarekegn et al. [20] noted significant relationship between total rainfall and frequency of rainfall with the incidence of grain mold fungi in sorghum seeds. Under dryland conditions, Hennessy et al. [21] observed little or no leaf blight infection on sorghum. Similarly, no significant correlations between the weather variables and anthracnose development were observed under the dry growing conditions in this study.

Table 2. The number of anthracnose resistant and susceptible sorghum accessions observed during the dry (summer) and wet (fall) growing conditions in 2009[a]

Reaction type	Number of accessions	Number of accessions
	Dry	Wet
Resistant	50	19
Susceptible	3	41
Variable response*	15	8

*[a]Sorghum accessions were planted at the Texas A&M University Experiment Station near College Station, Texas. Ratings were based on a scale of 1 to 5 (Erpelding and Prom, 2004; Prom et al. 2009), 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 with abundant acervuli on the flag leaf. Accessions were considered resistant if plants in the row were rated as 1 or 2 and susceptible if rated as 3, 4, or 5. *At least one replication was resistant*

Table 3. Disease reaction of 68 sorghum accessions and two controls inoculated with *Colletotrichum sublineolum* and evaluated under dry and wet growing conditions in 2009 at the brazos bottom near college station, texas[a]

Accession	Country	Dry	Wet
PI569076	Sudan	4[b]	5[b]
PI297192	Uganda	3	4
PI305056	Ethiopia	3	3
PI330794	Ethiopia	3/3/2*	4
PI305022	Ethiopia	3/3/2	4
PI568288	Sudan	2/3/3	5
PI297218	Uganda	3/2/3	4
PI568388	Sudan	2/3/3	4
PI568485	Sudan	2/3/3	5
PI568403	Sudan	3/2/3	4
PI276797	Ethiopia	2/3/2	4
PI297212	Uganda	3/2/2	3
PI152687	Sudan	2/3/2	4
PI154804	Uganda	2/2/3	3
PI568477	Sudan	3/2/2	4
PI563321	Sudan	2/3/2	4
PI350287	Uganda	2/2/3	5
PI305044	Ethiopia	2/2/3	4
PI609009	Mali	2	2

Accession	Country	Dry	Wet
PI154973	Uganda	2	3
PI297093	Uganda	2	2
PI154901	Uganda	2	3
PI217891	Sudan	2	3
PI305034	Ethiopia	2	4
PI196054	Ethiopia	2	5
PI297204	Uganda	2	4
PI152634	Sudan	2	5
PI297215	Uganda	2	2/3/3
PI154758	Uganda	2	2
PI330977	Uganda	2	2
PI154802	Uganda	2	3
PI297196	Uganda	2	3
PI454096	Ethiopia	2	3/3/2
PI454164	Ethiopia	2	4
PI563145	Sudan	2	4
PI563301	Sudan	2	2
PI330819	Ethiopia	2	4
PI563328	Sudan	2	5
PI568284	Sudan	2	4
PI305035	Ethiopia	2	4
PI568300	Sudan	2	4
PI568373	Sudan	2	2
PI297128	Uganda	2	4
PI297139	Uganda	2	2/2/3
PI297144	Uganda	2	3/2/2
PI568406	Sudan	2	3
PI570743	Sudan	2	2
PI568660	Sudan	2	4
PI154966	Uganda	2	2
PI609044	Mali	2	2
PI217674	Sudan	2	2
PI570211	Sudan	2	2/2/3
PI570873	Sudan	2	2
PI571386	Sudan	2	3
PI584214	Uganda	2	2
PI584284	Uganda	2	2
PI608974	Mali	2	2
PI608986	Mali	2	2
PI608992	Mali	2	2/3/2
PI609991	Mali	2	2
PI267624	Ethiopia	2	4
PI330983	Uganda	2	2
PI609008	Mali	2	2
PI569049	Sudan	2	4
PI609947	Mali	2	2
PI569033	Sudan	2	2/3/3
PI608990	Mali	2	2/3/2
PI569066	Sudan	2	4
BTx623	USA	3	5
SC748-5	USA	2	2

[a]Sorghum accessions were planted at the Texas A&M University Research Farm, Brazos Bottom near College Station, Texas. Ratings were based on a scale of 1 to 5 (Prom et al. 2009; Erpelding and Prom, 2004), 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. A rating of 1 or 2 is considered a resistant response, whereas a rating of 3, 4, or 5 is considered a susceptible response. [b]Rows with a single value indicate no variation for disease response within an experiment. An accession with more than one disease response value indicates variation within an experiment (data presented for the replications) or within-accession variation (*)

Table 4. Pearson's correlation coefficients between anthracnose rating and weather parameters for the dry (summer) and wet (fall) growing conditions: maximum average temperature (MaxT), minimum average temperature (MinT), maximum average relative humidity (MaxRH), minimum average relative humidity (MinRH), total rainfall (TRF) and number of days with rain (NDR)[a]

Season	**MaxT**		**MinT**		**MaxRH**		**MinRH**		TRF		NDR		WetS	
	r	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*
DryS[b]	0.08352	0.9165	-0.67201	0.3280	0.75531	0.2447	-0.19313	0.8069	0.85584	0.1442	0.79143	0.2086	0.93091*	0.0691
WetS[c]	-0.83929	0.1607	0.39525	0.6048	-0.50945	0.4905	0.94365*	0.0564	0.96889**	0.0311	0.94497*	0.0550		

*[a]Anthracnose score for the two growing seasons (dry and wet) is based on the sum of the scores across the accessions from each disease assessment date. The data for the weather parameters were based on the weekly average starting at the 4th week post-inoculation and ending at the 7th week post-inoculation. [b]DryS=anthracnose score for the dry season. [c]WetS=anthracnose score for the wet season.**or * = denotes significant at the 5% or moderately significant at the 10% probability level, respectively*

4. CONCLUSION

From these results, cumulative rainfall, frequency of rainfall and relative humidity are critical factors for disease development. Thus, the lack of rainfall during critical periods of disease development under the dry growing conditions would be responsible for the lack of susceptibility observed for some of the accessions in the study. Nineteen accessions from the Mali, Sudan, and Uganda collections were found to be resistant to anthracnose when evaluated during the dry and wet periods. These accessions could be used as parental lines in anthracnose breeding programs.

DISCLAIMER

Mention of trade names or commercial products in this manuscript is solely for the purpose of providing specific information and does not imply recommendations or endorsement by the U.S. Department of Agriculture.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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