Assessment of anthracnose resistance (*Colletotrichum graminicola*) in sorghum (*Sorghum bicolor*) germplasm under field conditions in Nigeria

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(*Revised MS received 9 July 2001*)

SUMMARY

Twenty-one elite sorghum lines were assessed for resistance to foliar, peduncle, rachis, grain and overall panicle anthracnose under natural infection at Samaru and Bagauda in 1996 and 1997. Three genotypes (R 6078, IS 14384 and CCGM 1/19-1-1) were completely resistant to all forms of the disease while Nagawhite was completely resistant to foliar, peduncle and rachis but moderately resistant to panicle anthracnose. The data indicate vertical disease progression from leaves onto peduncle and the rachis. More genotypes were susceptible to grain than rachis anthracnose thus indicating that grain infection could be direct or through vertical progression and direct pathways.

INTRODUCTION

Sorghum (*Sorghum bicolor* [L.] Moench) is an important food crop grown in the savanna zones of Nigeria between latitude 8° and 14° N, where it occupies about 46 % of the total land area devoted to cereal production. The estimated current production is about 8 million metric tonnes (NAERLS 1996). Yields at farmers' levels average 1·2 t/ha while varieties that yield up to 4 t/ha have been released (Aliyu & Adedipe 1997).

Sustainable high yields from varieties have been limited by abiotic and biotic stresses in Nigeria (Zummo 1984; Akpa *et al.* 1996; Ogungbile *et al.* 1998) of which anthracnose caused by *Colletotrichum graminicola* (Ces.) G. W. Wilson (= *C. sublineolum* Henn. in Kab. & Bulba) is a serious disease (Pande *et al.* 1993; Marley *et al.* 2001). The fungus infects leaves, stalks, peduncles, panicle and the grain either separately or all together (Pastor-Corrales & Frederiksen 1980). In Mali and Nigeria, panicle anthracnose is reported to be prevalent on farmers fields (Hess *et al.* 2001; Marley 1996). Anthracnose is reported to cause considerable yield loss of up to 47 % under experimental conditions in Nigeria (Tyagi 1980; Marley

1997) and up to 67 % in other parts of West Africa (Thomas *et al.* 1996) and elsewhere (Ali *et al.* 1987).

The use of resistant sorghum germplasm continues to be the single most important method of control of the disease especially in developing countries where it is cultivated under subsistence levels (Pastor-Corrales & Frederiksen 1980). Further, the anthracnose pathogen has been shown to be highly variable in Nigeria (Ozolua *et al.* 1986; Marley *et al.* 2001). However, in West Africa, many local landraces and introduced varieties lack satisfactory resistance to anthracnose, hence the real need for continuous evaluation of sorghum germplasm within the region for use in breeding programmes to complement other management practices.

Twenty-one elite lines obtained from WCASRN were evaluated for anthracnose responses at two locations in Nigeria in 1996 and 1997. Based on the real need for identification of sources of resistance to anthracnose, we hypothesized that some of them could contain sources of resistance that could be used in our breeding programme. This paper reports results of field evaluation of this germplasm at two locations.

MATERIALS AND METHODS

Field management and experimental design

The experiment was conducted at two locations

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Table 1. *Sorghum genotypes used for evaluation of anthracnose resistance at Samaru and Bagauda in Nigeria in 1996 and 1997*

Genotypes	Origin	Race	Grain colour	Days to 50% flowering
ICSV 905	ICRISAT	Caudatum	White	70
M943208-1	ICRISAT	Caudatum	White	90
Malisor 84-7	MALI	Caudatum	White	80
87W810	ICRISAT	Caudatum	White	82
91 W113-2-1	ICRISAT	Caudatum	White	85
82 Sel 1-Grain dur	ICRISAT	Caudatum	White	60
R 6078	ICRISAT	Caudatum	White	87
87-SB-F4-54-2	ICRISAT	Caudatum	White	71
IS 30469C-1526-4	ICRISAT	Caudatum	White	74
IS 30469C-1518T-5	ICRISAT	Caudatum	White	82
IS 14384	ICRISAT	Guinea	RB	82
IS 21658	ICRISAT	Caudatum	White	80
CEM 328/1-1-1-2	CIRAD/ICRISAT	Caudatum	White	75
CEM 328/3-3-1-1	CIRAD/ICRISAT	Caudatum	White	83
CCGM 1/19-1-1	ICRISAT	Caudatum	White	71
CGM 39/17-2-2	ICRISAT	Caudatum	White	75
ICSV 1079	ICRISAT	Caudatum	White	79
CEM 326/11-5-1-1	CIRAD/ICRISAT	Caudatum	White	74
S 34	ICRISAT	Caudatum	White	80
Nagawhite	GHANA	Caudatum	White	70
NR 71182	NIGERIA	Caudatum	White	81
Gaya Early	NIGERIA	Caudatum	White	80

during the 1996 and 1997 cropping seasons. One trial was planted at ICRISAT Research fields, Bagauda, Kano (11° 40′ N; 8° 30′ E) in the Sudan zone and the second trial was planted at IAR Research fields, Samaru, Zaria (11° 11′ N; 7° 38′ E). Twenty sorghum accessions (Table 1) obtained from WCASRN, Mali were used while NR 71182 and Gaya Early were planted as local checks at Samaru and Bagauda, respectively. Each site of the trial during each year was harrowed twice and ridges made 0·75 m apart. The trial was planted in a split-plot design with three replications. Main plots were treated with three applications of EC cypermethrin (0·02%) at weekly intervals starting from complete anthesis using hand sprayer to control headbugs, or left untreated; subplots were varieties. The trial was conducted under natural infection by anthracnose.

Individual plots were made up of 2 rows of 5 m each with an inter row spacing of 0·75 m. In Bagauda, sowing was done on 8 July 1996 and 2 July 1997. Seed of each line was planted 30 cm apart and thinned to 2 plants per hill, 4 weeks after crop emergence. Split application of fertilizer with 64 kg N, 30 kg P and 30 kg K/ha of N:P:K (20:10:10) was carried out. First application containing half of N and all of P and K were applied at harrowing while the second application was carried out 6 weeks after crop emergence. At Samaru, sowing was carried out on 26 July 1996, and 17 July 1997, and planting done as indicated above. Split application of fertilizer at similar rates as above was carried out with the first application carried out immediately after first weeding at 3 weeks after sowing, while the second application was carried out 10 weeks after sowing. Manual weeding was done at 3 and 6 weeks after sowing while moulding up (earthening up of existing ridges) was carried out immediately after the second fertilizer application.

Disease assessment

Six weeks after sowing, 5 plants per plot were tagged randomly and used subsequently for disease assessment. Foliar anthracnose was assessed when the plants had reached physiological maturity. Two weeks later, panicles of the tagged plants were harvested with their peduncles intact and assessed for peduncle, rachis and grain anthracnose damage. In addition, overall panicle anthracnose damage was determined (i.e. damage to peduncle, rachis and glumes, and grain combined). Disease severity on leaves, peduncle and rachis was assessed using a 1–9 visual rating scale (Thakur *et al.* 1998). In this scale, 1 = no symptoms of disease on plant part; 2 = 1–5% damaged by disease; 3 = 6–10% damaged by disease; 4 = 11–20% damaged by disease; 5 = 21–30% damaged by disease; 6 = 31–40% damaged by disease; 7 = 41–50% damaged by disease; 8 = 51–75% damaged by disease and 9 = > 75% damaged by disease.

Table 2. *Mean anthracnose severity for 21 sorghum genotypes under field conditions at Samaru, 1996 and 1997*

Genotype	Anthracnose severity*				
	Foliar	Peduncle	Rachis	Grain	Panicle
ICSV 905	7·6	4·1	5·2	4·2	2·6
M943208-1	8·2	2·4	1·5	4·0	2·2
Malisor 84-7	7·2	3·7	1·4	4·9	2·7
87W810	7·4	3·0	2·7	5·3	3·3
91W113-2-1	8·9	4·6	5·2	6·2	4·6
82 Sel 1-Grain dur	8·5	4·4	1·8	2·4	2·7
R 6078	1·0	1·0	1·0	1·0	1·0
87-SB-F4-54-2	8·1	3·5	2·0	3·7	2·2
IS 30469C-1526-4	6·9	3·9	1·8	4·8	2·5
IS 30469C-1518T-5	8·5	4·4	2·8	4·9	2·6
IS 14384	1·0	1·0	1·0	1·0	1·0
IS 21658	8·4	2·7	1·1	2·9	2·1
CEM 328/1-1-1-2	8·7	4·9	1·3	4·7	3·2
CEM 328/3-3-1-1	7·8	3·8	3·9	5·8	3·2
CCGM 1/19-1-1	1·0	1·0	1·0	1·0	1·0
CGM 39/17-2-2	3·2	2·5	1·2	5·0	3·2
ICSV1079	8·2	4·2	1·6	3·4	2·0
CEM 326/11-5-1-1	4·9	3·9	3·2	4·2	3·6
S 34	8·6	4·8	4·0	5·6	3·7
Nagawhite	1·0	1·0	1·0	4·4	2·2
NR 71182 (local)	5·8	4·9	5·2	5·7	3·5
Mean	6·23	3·34	2·41	4·09	2·62
S.E.	0·97	1·29	0·80	1·17	0·83
C.V. (%)	21·3	38·5	33·1	28·7	31·6

* Using a 1–9 rating scale where 1 = no disease symptoms, 9 = > 75 % plant part damaged by disease.

Grain anthracnose was assessed after threshing using the Threshed Grain Mould Rating (TGMR) visual scale of 1–9 (Bandyopadhyay & Mughogho 1988) where 1 = no mould, 2 = 1–5 % of grain surface area mouldy, 3 = 6–10 % grain surface area mouldy, 4 = 11–20 % grain surface area mouldy, 5 = 21–3 % grain surface area mouldy, 6 = 31–40 % grain surface area mouldy, 7 = 41–50 % grain surface area mouldy, 8 = 51–75 % grain surface area mouldy and 9 = > 75 % grain surface area mouldy.

Rainfall and relative humidity data were collected during the crop growth period in each year for each location.

Data analysis

Data were subjected to analysis of variance (ANOVA) to determine significant differences among the various genotypes and their interactions using GENSTAT 5, (Release 3.2; Rothamsted Experimental Station, Harpenden, UK). Further, Pearson correlation coefficients were determined using multiple regression analysis to investigate the relationship among the various disease symptoms.

RESULTS

Rainfall quantity and distribution were good at both locations to support crop growth and disease development. Total cumulative rainfall at Samaru between June and October was 667·2 mm with mean relative humidity (RH) at 77·5 % in 1996 and 928·2 mm with mean RH at 81·6 % in 1997. For the same years and period, rainfall at Bagauda was 852 mm with mean RH = 68·6 % and 829·3 mm with mean RH at 60·8 % respectively. Samaru is located in the wetter northern Guinea savanna while Bagauda is located in the drier Sudan savanna.

The reactions of sorghum genotypes to foliar, peduncle, rachis, grain and overall panicle anthracnose at Samaru in 1996 and 1997 are shown in Table 2. The response of the genotypes to anthracnose was not affected by insecticide treatment as there were no significant ($P < 0.05$) differences between plots treated with insecticides and the untreated plots. Three genotypes, R6078, IS 14384 and CCGM 1/19-1-1 were completely resistant to the disease while Nagawhite was resistant to foliar, peduncle and rachis but

Table 3. *Mean anthracnose severity for 21 sorghum genotypes under field conditions at Bagauda in 1996 and 1997*

	Anthracnose severity				
Genotype	Leaves	Peduncle	Rachis	Grain	Panicle
ICSV 905	5·3	1·9	2·2	1·0	1·1
M943208-1	4·9	3·4	2·3	1·0	1·1
Malisor 84-7	5·4	3·6	2·4	2·8	2·4
87W810	6·7	4·3	1·0	4·8	5·3
91W113-2-1	4·8	8·0	1·0	1·0	1·0
82 Sel 1-Grain dur	4·5	3·9	4·2	7·6	5·0
R 6078	1·0	1·0	1·0	1·0	1·0
87-SB-F4-54-2	6·7	7·1	4·6	7·7	6·3
IS 30469C-1526-4	6·4	4·3	1·0	3·3	3·8
IS 30469C-1518T-5	4·3	2·6	3·2	1·0	1·3
IS 14384	1·0	1·0	1·0	1·0	1·0
IS 21658	4·7	3·2	2·1	6·5	5·8
CEM 328/1-1-1-2	5·5	1·0	1·0	4·1	3·9
CEM 328/3-3-1-1	4·5	3·3	2·0	1·9	2·5
CCGM 1/19-1-1	1·0	1·0	1·0	1·0	1·0
CGM 39/17-2-2	1·0	4·1	3·8	1·0	1·3
ICSV 1079	5·5	3·5	4·1	5·1	3·1
CEM 326/11-5-1-1	1·0	6·5	1·0	2·9	1·2
S 34	6·3	6·1	3·1	2·2	2·2
Nagawhite	1·0	1·0	1·0	6·1	3·1
Gaya Early (local)	1·0	1·0	1·0	1·0	1·0
Mean	3·93	3·40	2·08	3·12	2·66
S.E.	0·83	1·17	1·01	1·24	0·82
C.V. (%)	14·6	34·6	48·3	39·6	30·9

susceptible to grain anthracnose. Results further indicate that no genotype was moderately resistant to foliar anthracnose but 10 genotypes were moderately resistant to overall panicle anthracnose. Generally, foliar anthracnose was most severe followed by grain anthracnose. Peduncle anthracnose was observed to be high while rachis anthracnose was least severe among the genotypes.

At Bagauda, four of the tested genotypes (R 6078, IS 14384, CCGM 1/19-1-1 and Gaya Early) were found to be resistant to foliar, peduncle, rachis, grain and overall panicle anthracnose in the two years (Table 3). Seven genotypes (R 6078, IS 14384, CCGM 1/19-1-1, CGM 39/17-2-2, CEM 326/11-5-1-1, Nagawhite and Gaya Early) were completely resistant to foliar anthracnose, while 14 other genotypes were susceptible. Nine genotypes were moderately resistant to overall panicle anthracnose. Generally, foliar anthracnose was most severe, followed by peduncle and grain while rachis anthracnose was the least severe. At both locations, three genotypes (R 6078, IS 14384 and CCGM 1/19-1-1) were completely resistant to foliar and panicle anthracnose while Nagawhite was completely resistant to foliar anthracnose but moderately resistant to panicle anthracnose.

There was a strong, significant ($P < 0.001$) correlation between severity of foliar, peduncle, rachis,

Table 4. *Correlation coefficients of disease severities of sorghum genotypes infected by* Colletotrichum graminicola *in Nigeria in 1996 and 1997*

Correlation between	Correlation coefficient
Foliar and peduncle	0·53*
Foliar and rachis	0·32*
Foliar and grain	0·37*
Foliar and panicle	0·49*
Peduncle and rachis	0·44*
Peduncle and grain	0·44*
Peduncle and panicle	0·45*
Rachis and grain	0·51*
Rachis and panicle	0·47*
Grain and panicle	0·81*

* $P < 0.001$.
$n = 252$.

grain and overall panicle anthracnose (Table 4). Significant interactions ($P < 0.001$) between genotype × location, genotype × year and location × year were observed to affect foliar and panicle anthracnose (Figs 1 and 2).
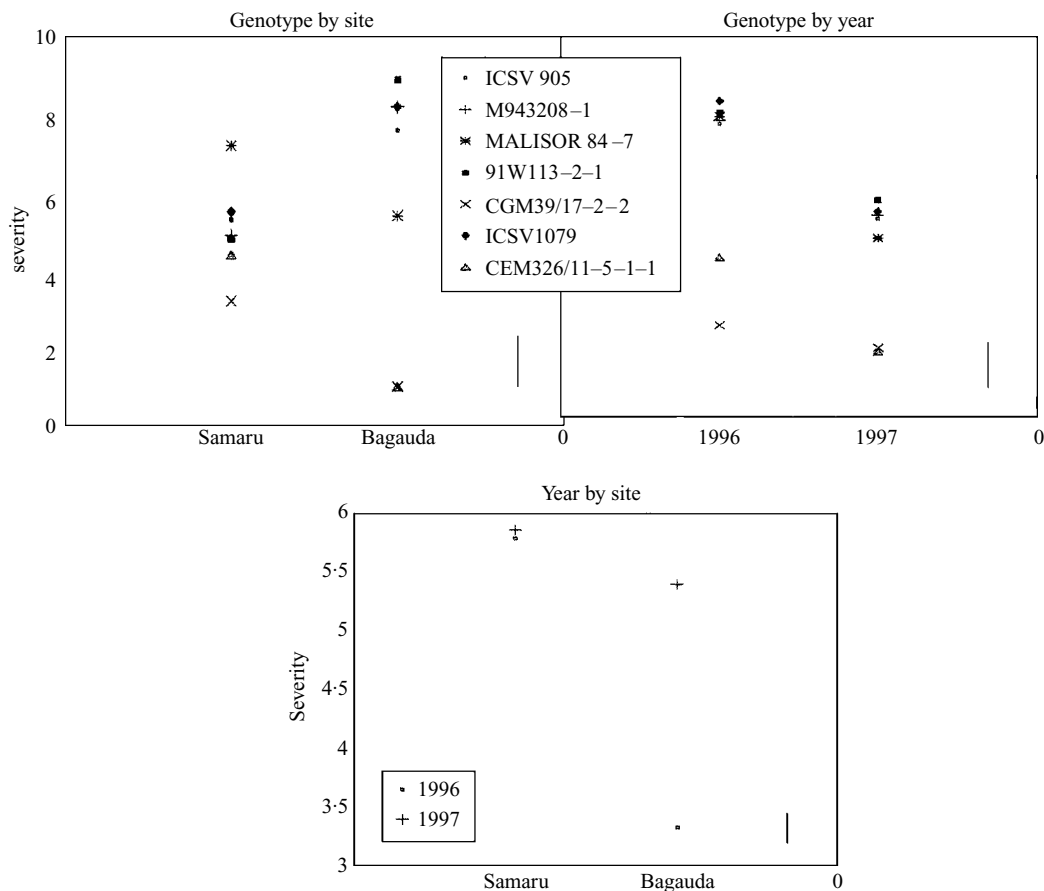
Fig. 1. Effect of sorghum genotype, location and year on foliar anthracnose severity. Bars represent s.e.

DISCUSSION

The objective of the multilocational study was to identify sources of resistance to anthracnose in the northern Guinea and Sudan savanna zones of Nigeria where most of the country's sorghum is cultivated. Although the study was conducted under natural infection, disease pressure was high enough to identify resistance at Samaru and Bagauda. As the weather data indicate, favourable environmental conditions prevailed for disease development during the critical stages of host-pathogen interaction. Further, field screening for resistance to sorghum anthracnose must be carried out during the rainy seasons at locations where warm and humid weather prevails (Tinline *et al.* 1989; Pande *et al.* 1994).

The first signs of the disease on leaves were observed at Samaru on susceptible genotype NR 71162 at 30 and 28 days after planting in 1996 and 1997, respectively, while at Bagauda, first disease appearance occurred at 35 and 38 days respectively on susceptible genotype 91W113-3-1. Higher disease

severity to foliar, peduncle and grain anthracnose was observed at Samaru than Bagauda. This may be attributed to higher relative humidity and longer duration of leaf wetness which resulted in higher disease pressure. Furthermore, a more diverse pathogen population exists at Samaru than Bagauda (Marley *et al.* 2001), attributed to the fact that studies on the pathogen have been carried out on Samaru research farms since 1960 (King 1970).

Disease scores of susceptible genotypes (e.g. 82 Sel 1-Grain dur, 91W113-2-1, 87-SB-F4-54-2, CEM 328/3-3-1-1 and S 34) ranged from 3·0 (6–10%) on the rachis to 7·4 (41–50%) on leaves. Among the genotypes evaluated, some have been identified with good levels of resistance to foliar and panicle anthracnose. These include R 6078, IS 14384, CCGM 1/19-1-1, CEM 326/11-5-1-1, CGM 39/17-2-2 and Nagawhite which could be grown in zones to which they are well adapted. A report by Hess *et al.* (2001) shows that IS 14384 and Nagawhite had good levels of resistance to both foliar and panicle anthracnose in Mali. In addition, genotypes ICSV
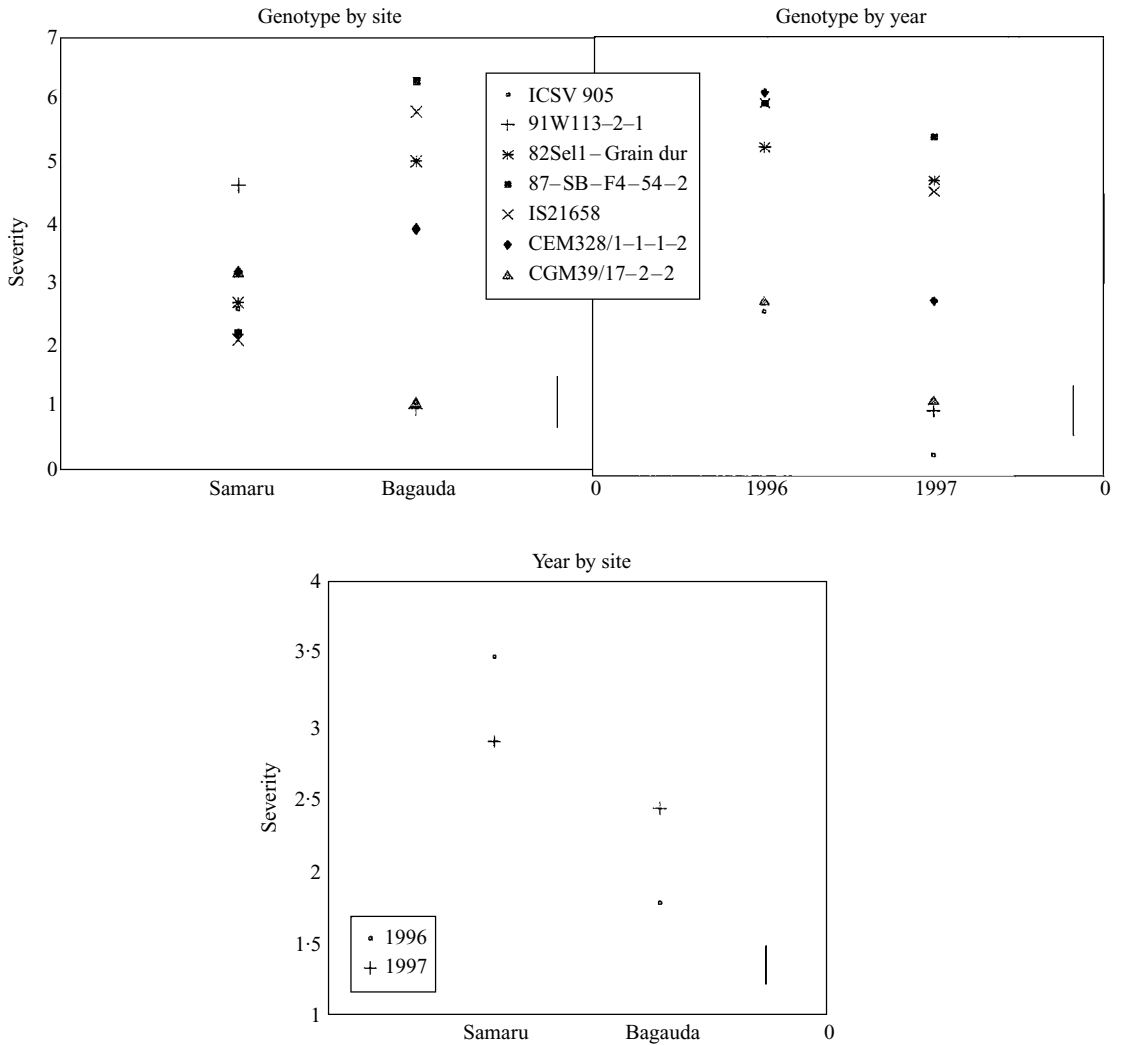
Fig. 2. Effect of sorghum genotype, location and year on panicle anthracnose severity. Bars represent S.E.

905, M 94-3208-1, Malisor 84-7, 91W113-2-1, CEM 329/1-1-1-2 and ICSV 1079 had moderate resistance to panicle anthracnose which is attributed to slow disease development and could serve as good sources of resistance in breeding programmes. Slow anthracnose development has previously been reported in sorghum as a useful component of host resistance (Casela *et al.* 1993).

In this study, foliar anthracnose was most severe, followed by grain, peduncle and rachis in that order. Reports by Bandyopadhyay *et al.* (1996) and Marley (1996) indicate that apart from foliar symptoms, panicle anthracnose is also prevalent on farmers' fields in West Africa. However, much attention has been paid to the former symptom of the disease by earlier workers as this phase is considered the most

important (Tarr 1962; King 1970; Pande *et al.* 1993). Thomas *et al.* (1996) observed that foliar anthracnose appeared early especially on susceptible sorghums in the field (about 4 weeks after sowing) and disease development could be very rapid and top leaves killed before grain reached physiological maturity. This may account for high disease scores on the leaves in susceptible genotypes when evaluation is done at grain physiological maturity as is the case in our study.

Although disease progress on leaves was not monitored during the course of this study, severity data obtained for all forms of the disease give an overall picture of the possibility of disease progression from leaves to peduncle and to rachis as was evident by progressively lower disease severity scores from

foliar to peduncle to rachis in most genotypes, e.g. ICSV 905, M 943208-1, Malisor 84-7, 87W810, IS 21658 and CEM 328/1-1-1-2, amongst others. Although there were significant correlations between panicle and grain symptoms, the reaction of genotypes, e.g. Nagawhite, S 34, CEM 328/1-1-1-2, IS 21658, 87-SB-F4-54-2 and 87W810 to grain anthracnose (which have higher severity of grain anthracnose than rachis) indicates clearly the possibility of direct grain infection by *C. graminicola*. The higher number of genotypes showing susceptibility to grain anthracnose than rachis further underscores the possibility of direct grain infection. Primary infection by the anthracnose pathogen is commonly from mycelium and conidia on crop residue (Dickson 1956) and on crop plants. In wet weather, spores ooze out from acervuli and are spread by rain splash (Edmunds *et al.* 1970). This clearly indicates that direct grain infection can occur by spore splash up the plant. This mode of direct infection appears to occur along with vertical progression rather than by the latter method alone as suggested by Hess *et al.* (2001). They observed significant correlations between severity of disease on the grain, rachis, peduncle and leaves. Further, high correlations between anthracnose on the peduncle and rachis were observed. These indicated vertical disease progression as they observed disease progression up the plant from lower infected leaves to upper plant parts. They concluded that high correlations between disease on the rachis and panicle and grain anthracnose therefore indicated disease progression from panicle branches and glumes onto the grains. Furthermore, evidence provided by Basu Chaudhary & Mathur (1979) and Sannoussi *et al.* (in press) show that sorghum seeds infected with *C. graminicola* could develop into infected seedlings by extra-embryonic infection followed by systemic infection. This serves as a source of primary inoculum showing vertical disease progression.

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