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Variation among foliar isolates of *Colletotrichum sublineolum* of sorghum in Nigeria

P.S. Marley[a,*], R.P. Thakur[b], O. Ajayi[a]

[a]*International Crops Research Institute for the Semi-Arid and Tropics (ICRISAT), Kano, Nigeria*
[b]*International Crops Research Institute for Semi-Arid Tropics (ICRISAT), Patancheru, Andhra Pradesh 502324, India*

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Abstract

Foliar anthracnose, caused by *Colletotrichum sublineolum*, is a major disease of sorghum in Nigeria. Pathogenic diversity was studied among foliar isolates of *C. sublineolum* collected from sorghum cultivars in farmers fields in the Sahel, Sudan, northern Guinea and southern Guinea zones of Nigeria during the 1997 crop season. For the study a total of 50 isolates were identified based on typical symptom types and cultural characteristics. On the basis of growth in culture and morphological characteristics, the isolates were classified into nine morphological groups (MGs). Pathogenicity tests were done on a set of known sorghum differential lines by inoculating pot-grown seedlings in a greenhouse. On the basis of disease reaction and disease severity scores on the differential lines, the 50 isolates were classified into seven pathogenic groups (PGs). Sixteen representative isolates of the MGs and the PGs were further evaluated for virulence and aggressiveness on the differential lines and were classified into five distinct groups using the centroid method of cluster analysis. The existence of five races of *C. sublineolum* in major sorghum growing zones of Nigeria is suggested.

Keywords: *Sorghum bicolor*; *Colletotrichum sublineolum*; Pathogenic variability; Nigeria

1. Introduction

Sorghum (*Sorghum bicolor* (L.) Moench) is an important cereal crop grown in the savanna zones of Nigeria between latitude 8° and 14°N, where it occupies about 40% of the total land area devoted to cereal production. The estimated current annual production is about 8 Tg (NAERLS, 1996). Diseases constitute one of the most important constraints to sorghum production in Nigeria and other countries in West and Central Africa (Thomas, 1995). Anthracnose, caused by *Colletotrichum sublineolum* Ces. causes serious damage (Tyagi, 1980) and infects all above ground parts of the plant. Peduncle, inflorescence and grain infection is referred to as panicle anthracnose and is now prevalent on farmer's fields (Marley, 1996a).

Foliar anthracnose is the most widely distributed foliar disease of sorghum in the Sahel, Sudan, northern Guinea and southern Guinea zones of Nigeria. It is particularly destructive in the northern Guinea and Sudanian zones (Pande et al., 1993) and has been reported to cause up to 47% yield loss at Samaru (Marley, 1996b) under experimental conditions. Foliar anthracnose is characterised by small sunken circular

* Corresponding author. Present address: Faculty of Agriculture, Institute for Agricultural Research, Ahmadu Bello University, Samaru, Zaria, Nigeria.
E-mail address: psmarley@rcl.nig.com (P.S. Marley).

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to elliptical spots 3–6 mm in diameter with few or numerous fruiting bodies (acervuli) of the fungus on leaf lesions (Tarr, 1962; Zummo, 1984). However, differences in symptomology are common and may be attributed to variability in the pathogen, host reaction, the physiological state of the host, and environmental conditions (Pastor-Corrales and Frederiksen, 1980; Ferreira et al., 1985; Pande et al., 1991). In Nigeria, three distinct types of symptoms have been identified: patchy and diffused spots on the lamina, commonly referred to as patchy; lesions and discoloration on the midrib, referred to as midrib; and isolated pinpoint spots on the lamina. Patchy and midrib symptoms are the most prevalent (Alawode et al., 1983; Manzo, 1985). Using symptom type as well as morphological, cultural and pathological characteristics, Alawode et al. (1983) identified a distinct variety of *C. sublineolum* as *Colletotrichum graminicola var. isolatum*. Further studies by Bindawa (1987) indicated the existence of only patchy and midrib lesions as symptoms and that pinpoint symptoms were initials of patchy symptoms that later became diffused. This study contradicted the report by Alawode et al. (1983) and questioned the establishment of *C. graminicola var. isolatum* as distinct from *C. sublineolum*.

However, earlier work by King and Frederiksen (1976) and Ozolua et al. (1986) had suggested the existence of races in *C. sublineolum* attacking sorghum leaves in Nigeria. These studies were limited by the number of isolates considered and the area covered during sample collection. Given the large area under cultivation, the diversity of genotypes used, and the need to verify conflicting earlier reports, further, more-detailed studies of anthracnose pathogen variation in Nigeria were needed. This paper reports pathogenic, cultural and morphological diversity among 50 foliar isolates of *C. sublineolum* collected from different locations in the Nigerian savanna.

2. Materials and methods

2.1. Survey and sample collection

Diseased leaf samples were collected during the 1997 crop season from major areas of sorghum production in the Sahel, Sudan, northern and southern Guinea savanna zones. Samples were also collected from some locations in the derived savanna within the forest zone where sorghum is sparingly cultivated. This was achieved by undertaking two main surveys as indicated on Fig. 1. The first survey route was from Ibadan–Zaria–Sokoto–Minna–Keffi–Enugu–Lokoja–Abuja to Zaria, and covered about 3344 km. A total of 23 samples were collected. The second survey route from Zaria–Katsina–Kano–Nguru–Gashua–Damaturu–Maiduguri–Bama–Mubi–Yola–Jalingo–Wukari–Langtang–Jos to Zaria, covered over 3595 km with 20 samples collected.

During the survey, stops were made every 100–250 km and farmer's sorghum fields examined for symptoms of the disease. Stops were made every 50–100 km in areas where high cultivar diversity existed. At each stop, between one and five farmer's fields were inspected and diseased leaf pieces were cut, placed between blotter papers, labelled and packaged. They were then brought into the laboratory for examination. Samples were collected from one or more cultivars.

Seven samples were collected in Samaru and Bagauda, bringing the samples used in this study to 50 (Table 1). In general, sample collection emphasised cultivar diversity and collection from distinct zones in terms of temperature and rainfall conditions.

2.2. Cultural and morphological variability

From each leaf sample collected, diseased areas were cut into single-lesions and surface sterilised in 1% sodium hypochlorite solution for 1 min. The diseased tissue was then rinsed in three changes of sterile distilled water, plated on freshly prepared oatmeal agar (OMA), and incubated at 28°C for 7 days under constant cool-white fluorescent light to induce abundant sporulation. Pure cultures raised from single spore of each isolate were identified and subcultured for use in this study. The isolates were studied for various traits, including sporulation potential, growth rate, and morphology of conidia and setae.

Sporulation potential was determined from spore counts taken using 7-day-old cultures of each isolate. A 10 mm diameter disk of each isolate was cut from the centre (oldest portion of each culture) with a sterile cork borer and placed in test tubes containing 10 ml of sterile distilled water and shaken thoroughly to dislodge conidia. After filtering through a double-layered

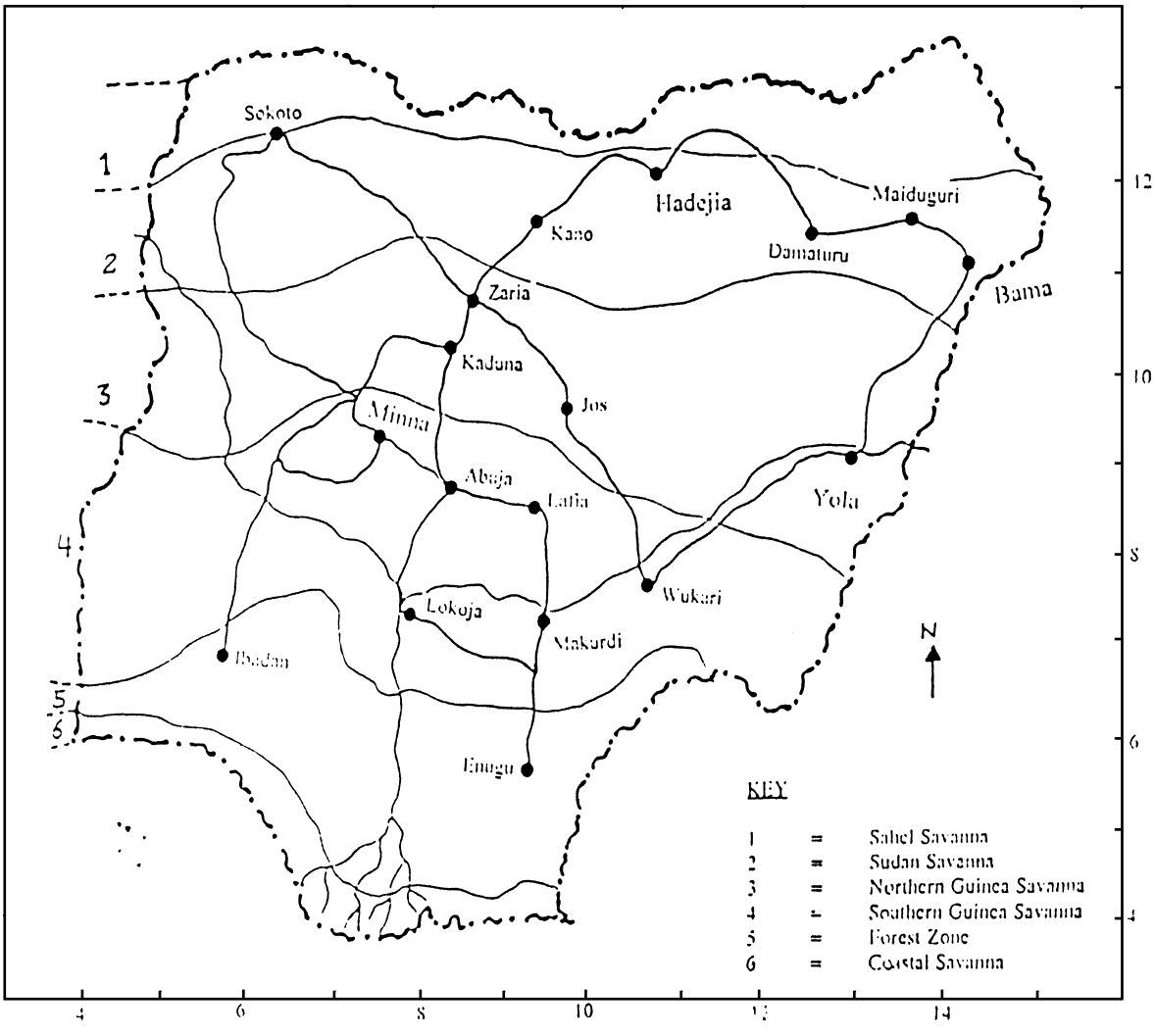

Fig. 1. Map of Nigeria showing the various routes of the survey in the five climatic zones where samples of *Colletotrichum sublineolum* were collected.

muslin cloth, spore and setae counts were taken using haemocytometer and the numbers of spores and setae per millilitre determined. Five replicate plates were used for each isolate.

Colony growth rate was measured by cutting 3 mm diameter disks of each isolate with a sterile cork borer, placing these onto OMA plates (care was taken to place the disks at the centre of the plates) and incubating as mentioned earlier. Seven days after inoculation, growth was measured and growth rate (mm per day) was calculated.

To record spore morphology; length and breadth of 50 spores of each isolate were measured using a stage micrometer/eye piece ocular scale and a compound microscope. Similarly, 50 setae obtained from each isolate were measured. For all the above studies five plates were maintained and each plate served as a replicate. Plates were arranged in a completely randomised design inside the incubator. The experiment was repeated once. The isolates were classified into different morphological groups (MGs) based on the above parameters.

Table 1
Locations, designations and symptom types of foliar anthracnose samples collected from different sorghum types in Nigeria during the 1997 crop season

Serial number	Location	Designation	Sorghum type/race	Symptom type[a]
1	Samaru	SM1	Kaura (local)	Patchy
2	Talata mafara	TM	BES	Patchy
3	Sokoto	SK1	BES	Patchy
4	Dadin kowa	DK	Caudatum (local)	Patchy
5	Kontogora	KT	Farafara (local)	Midrib
6	Bokani	BK	Red guinea (local)	Pinpoint
7	Ilorin1	IL1	White guinea (local)	Pinpoint
8	Ibadan	IB	Red guinea (local)	Midrib
9	Obbollo-affor	DA	Red guinea (local)	Midrib
10	Makurdi	MK	Red guinea (local)	Midrib
11	Bwari	BW	White guinea (local)	Patchy
12	Nguru	NG	Dungoya (local)	Patchy
13	Funtua	FT1	Farafara (local)	Patchy
14	Katsina	KT	ICSV III	Pinpoint
15	Sabon rafi	SR	Yantsantawa (local kaura)	Midrib
16	Damaturu	DMT1	Tunkwushe local	Midrib
17	Gezawa	GZ	Masakwa (post rainy season)	Pinpoint
18	Damaturu	DMT2	BES	Patchy
19	Pulka gwoza	PGZ	Sambul (local white)	Pinpoint
20	Fulka	PK	Masakwa (post rainy reason)	Pinpoint
21	Damaturu	DMT3	Yafimoro (local)	Patchy
22	Bama	BM	Merekime (local)	Midrib
23	Geidam	GD	Pramgram (kaura local)	Midrib
24	Numan	NM	Local	Midrib
25	Jalingo	JLG1	Kashongong (local)	Pinpoint
26	Amper	AP1	Amper (local)	Midrib
27	Pankshin	PK	Farafara (local)	Midrib
28	Jalingo	JLG2	Yarjingo (local)	Midrib
29	Gu-wukari	GWK	Local	Midrib
30	Langtang	LG	Local	Midrib
31	Amper	AP2	Trejen (local) (red)	Midrib
32	Funtua	FT2	SK 5912	Patchy
33	Gusau	GS	Farafara (local)	Midrib
34	Samaru kataf	SKT	ICSV III	Patchy
35	Buruku	BRK	Kaura (local)	Patchy
36	Tegina	TG	Kaura (local)	Patchy
37	Ikara	IK	Farafara (local)	Patchy
38	Makarfi	MKF	Kaura (local)	Pinpoint
39	Saminaka	SMK	Farafara (local)	Patchy
40	Samaru	SM2	ICSV 905 NG	Patchy
41	Dangora	DG	BES	Pinpoint
42	Ilorin	IL2	Local	Patchy
43	Sokoto	SK2	Yarsafe (local)	Patchy
44	Samaru	SM3	ICSV III	Patchy
45	Samaru	SM4	ICSV 400	Patchy
46	Samaru	SM5	Yarwasha (local)	Patchy
47	Arika	AK	Farafara (local)	Midrib
48	Bagauda	BG1	Farafara (local)	Patchy
49	Bagauda	BG2	IRAT 204	Patchy
50	Bagauda	BG3	ICSV 400	Patchy

[a] Based on symptom description by Alawode et al. (1983) and Manzo (1985).

2.3. Pathogenicity test

Eight sorghum lines (KVS 8, BES, IS 3758, IS 6926, IRAT 204, IS 6958, IS 18442 and A 2267-2) selected for known differences in response in the International Sorghum Anthracnose Virulence Nursery (ISAVN) coordinated by ICRISAT — Patancheru (Thakur, 1995) were used in pathogenicity testing. Each was grown from seeds sown in 30 cm diameter plastic pots filled with sterilised soil, with the pots arranged in the glasshouse in a randomised complete block design comprising 50 isolates of *C. sublineolum* (Table 1), eight test lines and five plants/pot (each plant served as a replicate). Leaves of 21-day-old plants were inoculated with spore suspension (1×10^5 spores ml) of an isolate with a hand sprayer until runoff. Control plants were sprayed with sterile distilled water. Following inoculation, plants were maintained at high relative humidity (>90%) for 12 h for two consecutive days at an average temperature of 28°C. The isolates were classified into different pathogenic groups (PGs) based on their pathogenicity to specific sorghum lines.

2.4. Assessment of virulence and aggressiveness

Differences in virulence (the degree or measure of pathogenicity) amongst isolates was examined by comparing the number of genotypes on which isolates produced symptoms. The same eight sorghum lines were used in this study with plants grown and inoculated as described earlier. A total of 16 isolates, representing nine of the MGs and seven of the PGs were used. Inoculated plants were incubated in the greenhouse as described above and pots were arranged in a randomised complete block design. Each plant in a pot served as a replicate. The experiment was repeated twice.

Aggressiveness (ability to cause severe disease) among the isolates was measured by comparing the variation in the degree of pathogenicity (mean disease severity) on two lines (BES and IRAT 204), which were susceptible to all 16 groups of isolates.

2.5. Disease scoring

Two weeks after inoculation plants were rated for their reaction type as R, MR or S, where R (resistant) = no symptoms, or presence of chlorotic flecks; MR (moderately resistant) = hypersensitive lesions, red spots or necrotic spots without acervuli; and S (susceptible) = lesions with acervuli.

Disease severity (percentage of leaf area covered by lesions) was recorded using the 1–9 visual rating where 1 = no symptoms on leaf surface; $2 = 1-5\%$; $3 = 6-10\%$; $4 = 11-20\%$; $5 = 21-30\%$; $6 = 31-40\%$; $7 = 41-50\%$; $8 = 51-75\%$; $9 \geq 75\%$ of total leaf area of plant damaged by anthracnose.

2.6. Data analysis

Data were subjected to analysis of variance to determine significant differences among various treatments and their interactions. Disease severity data were subjected to cluster analysis using the centroid method on SPSS (Norman et al., 1984) to classify the isolates into different groups.

3. Results

3.1. Survey and sample collection

Geographic location, cultivar and symptom types of the samples used in the study are shown in Table 1.

3.2. Morphological variability

The 50 isolates were categorised into nine MGs on the basis of various growth and cultural characteristics (Table 2). The number of isolates identified in each MG varied; MGs 2, 5 and 9 had, respectively 9, 10 and 15 isolates, while MGs 6 and 8 had only one isolate each. Significant differences ($P < 0.05$) were found in all parameters evaluated (Table 3). MGs 2 and 3 had the highest growth rate of 4.3 mm per day. Conidia produced were falcate or spindle shaped, hyaline and aseptate. MG 9 produced conidia profusely with a spore count of 191.7 ml^{-1} while MGs 4 and 6 had the lowest count of 41.7 ml^{-1}. MG 6 had the highest mean conidial length and width of 27.76 and 9.54 μm, respectively. The representative isolates selected from each of nine groups were: isolates 2, 3, 11, 16, 25, 33, 40, 46 and 47.

Table 2
Cultural and morphological grouping of 50 isolates of *Colletotrichum sublineolum* into cultural and morphological groups[a]

Morphological group	Isolate numbers	Isolate characteristics	Representative isolate selected
MG 1	9, 10, 11, 13 (4)	Dark brown colour with light tan top, most prominent at middle of culture. Surface of culture with many concentric rings with zonations which are very prominent towards the middle	11 (P)[b]
MG 2	4, 7, 14, 15, 16, 19, 24, 26, 29 (9)	Gray coloured cottony or fluffy cultures with only one concentric ring close to point of inoculation	16 (M)
MG 3	2, 27, 49 (3)	Dark coloured, appressed cultures with many concentric rings evenly distributed	2 (P)
MG 4	32, 44, 47, 48 (4)	Dark coloured but fluffy cultures with no concentric rings	47 (M)
MG 5	6, 8, 17, 21, 22, 39, 41, 45, 46, 50 (10)	Dark brown, appressed cultures with faint concentric rings	46 (P)
MG 6	3 (1)	Dark coloured, appressed cultures with no concentric rings	3 (P)
MG 7	33, 34, 35 (3)	White base with gray coloured top. Gray mycelium forms faint concentric rings	33 (M)
MG 8	40 (1)	Dark with gray coloured middle, fluffy with evenly distributed concentric rings	40 (P)
MG 9	1, 5, 12, 18, 20, 23, 25, 28, 30, 31, 36, 37, 38, 42, 43 (15)	White compact with no concentric rings	25 (P)

[a] Figures in bracket are total number of isolates comprising a morphological group.
[b] P: patchy; M: midrib; PP: pinpoint.

3.3. Pathogenicity test

On the basis of infective performance with eight selected sorghum lines (R/MR/S), the 50 isolates were separated into seven pathogenicity groups (Table 4). Pathogenicity group (PG) 4 had the highest number of isolates (11) while PGs 2 and 7 had the least five isolates each. The representative isolates from each of the seven PGs were: isolates 6, 8, 10, 15, 20, 24 and 30. Two sorghum lines IRAT 204 and BES were susceptible to all the seven isolates, while KSV 8 was highly resistant.

Table 3
Cultural and morphological characteristics of nine representative isolates of *Colletotrichum sublineolum* from sorghum growing areas of Nigeria

Group	Growth rate (mm)	Spore count	Conidial length (mm)		Conidial width (mm)		Setae length (μm), mean
			Mean	Range	Mean	Range	
MG 1	3.7	62.0	22.0	21.8–31.7	9.30	8.0–10.4	141.4
MG 2	4.3	104.3	20.98	20.6–21.3	8.32	7.2–9.5	151.2
MG 3	4.3	69.3	27.46	24.7–30.4	9.20	8.4–10.0	127.5
MG 4	3.7	41.7	24.76	20.4–30.7	8.60	7.7–9.8	132.1
MG 5	3.4	48.0	25.14	20.7–30.4	9.02	8.7–9.9	136.2
MG 6	4.3	41.7	27.76	24.5–30.5	9.54	8.7–9.9	119.9
MG 7	3.7	66.3	26.42	23.9–29.8	9.00	8.5–9.8	118.2
MG 8	3.7	53.7	24.36	21.1–27.8	8.28	7.6–9.7	148.0
MG 9	3.2	191.7	17.46	15.3–19.4	4.06	3.0–5.2	87.3
Mean	3.8	75.41	24.04	–	8.37	–	129.1
S.E.M.	0.1	5.90	1.42	–	0.34	–	1.7
CV(%)	4.0	13.6	12.90	–	9.00	–	2.9
Error DF	32	16	32	–	32	–	32

Table 4
Pathogenicity reaction of seven representative isolates of pathogenicity groups (PG) of *Colletotrichum sublineolum* on eight sorghum differential lines in a screenhouse[a]

Sorghum line	Race	Origin	PG 1 (24)	PG 2 (6)	PG 3 (10)	PG 4 (30)	PG 5 (15)	PG 6 (20)	PG 7 (8)
IS 6958	Durra caudatum	Sudan	S	R	MR	S	MR	S	R
A 226-2	Caudatum	India	MR	R	R	MR	S	S	MR
IS 18442	Guinea-durra	India	MR	MR	MR	S	S	S	S
IS 6928	Kafir caudatum	India	S	R	S	S	MR	S	MR
IS 3758	Caudatum bicolor	USA	S	S	S	S	S	S	MR
IRAT 204	Durra caudatum	Burkina Faso	S	S	S	S	S	S	S
BES	Caudatum	Nigeria	S	S	S	S	S	S	S
KSV 8	Caudatum	Nigeria	R	R	R	R	R	R	R

[a] R: resistant (no symptoms); MR: moderately resistant (reddening or red spots, necrosis); S: susceptible lesions with acervuli; PG 1: 7, 16, 19, 24, 26, 34, 44, 48 (8); PG 2: 6, 9, 13, 22, 39 (5); PG 3: 2, 10, 27, 32, 47, 49, 50 (7); PG 4: 12, 18, 25, 30, 31, 33, 35, 38, 40, 42, 53 (11); PG 5: 3, 4, 14, 15, 29, 45, 46 (7); PG 6: 1, 5, 20, 23, 28, 36, 37 (7); PG 7: 8, 11, 17, 21, 41 (5); the representative isolate number of each PG.

3.4. Assessment of virulence and aggressiveness

The reaction types and disease severity of the test isolates of MGs and PGs presented in Table 5. Disease reaction was quite variable amongst the isolates and the host lines. Isolates of MGs 7, 9 and PG 6 were the most virulent on seven of the eight lines evaluated susceptible, while isolate MG 1 was the least virulent producing symptoms on only two lines. Significant differences ($P < 0.05$) were evident among isolates and lines (Table 6). Disease severity was also highly variable amongst the isolates and lines. Aggressiveness among the isolates, measured by comparing the variation in the degree of mean disease severity on the

Table 5
Reaction class (RC) and disease severity (DS) of 16 isolates of *Colletotrichum sublineolum* on eight sorghum lines in a screenhouse[a]

Isolate	Sorghum line																MDS[b]	NSG[c]
	IS 6958		A 2267-2		IS 18442		IS 6928		IS 3758		IRAT 204		BES		KSV 8			
	RC	DS	RC	DS	RC	DS	RC	DS	RC	DS	RC	DS	RC	DS	RD	DS		
MG 1	R	1.0	MR	2.5	MR	2.6	R	1.0	MR	2.2	S	3.2	S	4.3	R	1.0	2.2	2
MG 2	R	1.0	MR	2.7	S	5.4	R	1.0	S	4.0	S	6.9	S	7.2	R	1.0	3.7	4
MG 3	MR	2.0	MR	2.6	MR	2.8	R	1.0	S	6.9	S	6.0	S	5.3	R	1.0	3.5	3
MG 4	R	1.0	R	1.0	R	1.0	R	1.0	S	5.9	S	5.1	S	6.9	R	1.0	2.9	3
MG 5	R	1.0	MR	1.8	S	5.6	R	1.0	MR	2.8	S	3.9	S	7.6	MR	1.2	3.1	3
MG 6	MR	2.8	S	4.1	S	6.4	MR	2.3	S	5.1	S	5.4	S	6.4	R	1.0	4.2	5
MG 7	S	7.1	S	3.9	S	7.0	S	5.2	S	6.0	S	3.9	S	7.5	R	1.0	5.2	7
MG 8	S	6.7	S	6.6	S	5.6	S	5.5	MR	2.6	S	7.8	S	5.4	MR	1.3	5.2	6
MG 9	S	4.4	S	5.4	S	5.9	S	4.5	S	5.0	S	5.8	S	8.4	R	1.0	5.1	7
PG 1	S	5.1	MR	1.3	MR	2.6	S	4.5	S	5.3	S	6.9	S	7.4	R	1.0	4.3	5
PG 2	R	1.0	R	1.0	MR	2.1	R	1.0	S	4.7	S	5.3	S	5.4	R	1.0	2.7	3
PG 3	MR	1.4	R	1.0	MR	1.3	S	5.3	S	5.1	S	6.2	S	6.3	R	1.0	3.5	4
PG 4	S	4.7	MR	1.3	S	5.0	S	5.1	S	5.2	S	6.1	S	8.6	R	1.0	4.6	6
PG 5	MR	1.2	S	4.1	S	4.2	MR	2.4	S	4.3	S	5.8	S	6.0	R	1.0	3.6	5
PG 6	S	7.0	S	3.8	S	5.0	S	4.1	S	4.8	S	6.2	S	8.0	R	1.0	5.0	7
PG 7	R	1.0	MR	1.2	S	4.6	MR	2.1	MR	2.9	S	6.1	S	7.6	R	1.0	3.3	3
Mean		3.0		2.8		4.2		2.9		4.6		5.7		6.8		1.0		

[a] Each value represents the mean of 15 replications (five replications per repetition).
[b] Mean disease severity of an isolate on eight genotypes.
[c] Number of lines showing susceptible reaction to each isolate.

Table 6
Analysis of variance for disease severity of 16 isolates of *Colletotrichum sublineolum* on eight sorghum lines grown in a screenhouse

Source of variation	DF	Mean square	*F*-value
Replications	4	0.194	0.03
Isolates	15	9.664	1.67**
Sorghum genotypes	7	26.864	4.64**
Isolates × sorghum genotypes	105	4.096	
Residual (error)	508	5.785	

two sorghum genotypes (BES, IRAT 204) that were susceptible to all 16 isolates. Isolates of PGs 1, 4, 6, and MGs 2 and 9 were identified as most aggressive while isolate MG 1 was the least aggressive (Table 7).

Cluster analysis of 16 representative MG and PG isolates categorised them into five groups (Fig. 2). Group 1 comprised of Isolates MGs 1, 3, 4, 5 and PGs 3 and 7 that caused S reaction on BES and IRAT 204; R or MR reaction on KSV 8, IS 6928, IS 6958 and A2267-2, and were identified as least virulent. Group 2, which comprised a single isolate from MG 2, caused S reaction on four lines (IS 18442, IS 3758, IRAT 204 and BES) and R reaction in KSV 8, IS 6928 and IS 6958. Group 3 also comprised a single isolate, in this case from PG 2, caused S reaction in three lines (IS 3758, IRAT 204, BES) and R in KVS 8 and IS 6928. Group 4 included isolates of MG 6 and PGs 1 and 5, that caused S reaction in five lines and R in one (KSV 8). Group 5 included isolates of MGs 7, 8 and 9, and PGs 4 and 6 causing S reaction in six lines and R reaction in two lines (KSV 8, IS 6958). Group 5 was identified as most virulent group.

Table 7
Relative aggressiveness of 16 isolates of *Colletotrichum sublineolum* as measured by disease severity on two susceptible sorghum lines in a screenhouse

Isolate	Disease severity		
	IRAT 204	BES	Mean[a]
MG 1	3.2	4.3	3.8
MG 2	6.9	7.2	7.1
MG 3	6.0	5.3	5.7
MG 4	5.1	6.9	6.0
MG 5	3.9	7.6	5.8
MG 6	5.4	6.4	5.9
MG 7	3.9	7.5	5.7
MG 8	7.8	5.4	6.6
MG 9	5.8	8.4	7.1
PG 1	6.9	7.4	7.2
PG 2	5.3	5.4	5.4
PG 3	6.2	6.3	6.3
PG 4	6.1	8.6	7.4
PG 5	5.1	6.0	5.6
PG 6	6.2	8.0	7.1
PG 7	6.1	7.6	6.9
Mean	5.7	6.8	

[a] High mean value indicates high aggressiveness level of isolate.

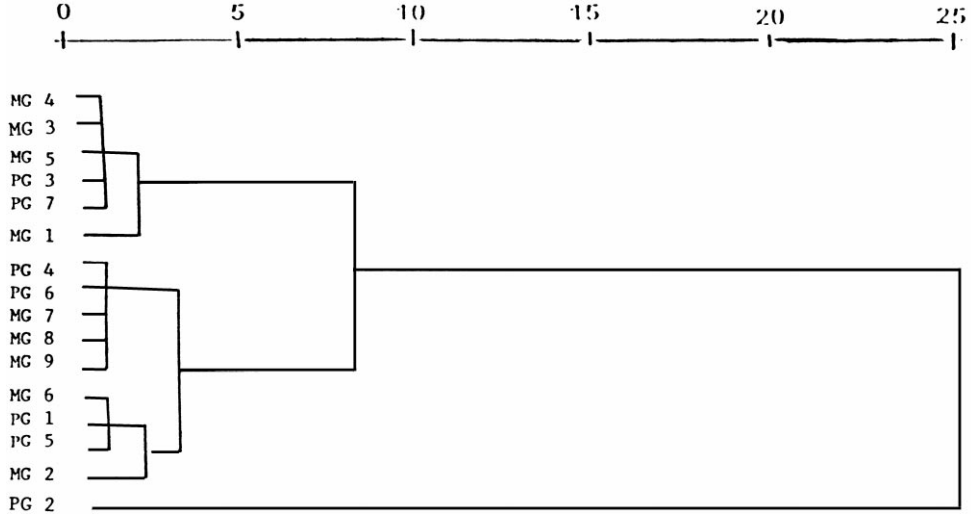

Fig. 2. Dendrogram showing the clustering of the nine morphological and seven pathogenic groups of *Colletotrichum sublineolum* isolates into five physiologic races.

4. Discussion

The 50 isolates of *C. sublineolum* collected from major sorghum growing areas of Nigeria showed considerable variation in cultural, morphological and pathogenic characteristics. Using cultural and morphological characteristics, we established nine groups among the isolates; while on the basis of pathogenicity, seven distinct groups were identified. This evidence of physiological races within *C. sublineolum* population supports earlier work in Nigeria by Ozolua et al. (1986). Reports by Ali and Warren (1987), Cardwell et al. (1989), Ferreira et al. (1985) and Pande et al. (1991) have variously shown the existence of variation among foliar anthracnose isolates in other parts of the world. A recent report by Thakur and Rao (1998) based on the results of ISAVN from 1992 to 1997 also indicated the existence of variation among populations of the pathogen in the different parts of the world. It is desirable to compare between isolate groupings identified in this study with those by other workers, e.g. Ali and Warren (1987), Pande et al. (1991) from other parts of the world, but to do so using standard differential lines. There is the need to identify an international set of differentials (e.g. using the ISAVN set) for use by future workers to enable evaluation of populations and establishment of race groups as in other pathogens, e.g. *Spacelotheca sorghi* (Frowd, 1980).

MG 9 and PG 6 were identified as the most virulent isolates in this study. None of the 16 selected MG or PG isolates induced an MR reaction with a high disease severity. Pande et al. (1991) also reported that in the hypersensitive reaction (HR), chlorotic flecks did not enlarge and sometimes disappeared 7 days after inoculation. It may therefore not be possible to have resistant phenotypes with a high disease severity rating. The level of aggressiveness was observed to vary among the isolates within groups and did not directly correlate with virulence. Isolates MG 7 and MG 8 were more aggressive than others. Pande et al. (1991) ascribed the differential relationship between virulence and aggressiveness or degree of pathogenicity as observed in this trial to isolate-host genotypic interaction. However, when mean disease severity on the two most susceptible cultivars was used (Table 7), virulence in isolates MG 9 and PG 6 correlated positively with aggressiveness. Because isolates in these two groups were both widely infective and caused highest level of disease severity, they may be the most widespread and damaging in Nigeria.

Cluster analysis of disease severity data obtained for the 16 isolates show that they belong to five distinct classes or physiologic races. Alawode et al. (1983) proposed three races, based on symptom type, but the results of this study show that isolates obtained from pinpoint lesion isolates (MG 9 in Table 2) are not distinctly different from those obtained from leaves exhibiting patchy or midrib symptoms. Isolates from pinpoint lesions had similar cultural and morphological characteristics to other isolates in this group. This supports an earlier report by Bindawa (1987), who indicated that isolates from patchy and pinpoint lesions differed only on the basis of symptom expression. Thus, there may be only five physiologic races of the pathogen present on leaves of sorghum in Nigeria. An on-going study is examining the variability of the pathogen in isolates obtained from different plant parts and may add to the number of races obtained from the current study. With increasingly diverse sorghum cultivars in use, and as the makeup of populations of *C. sublineolum* changes, appearance of new races is likely. There is therefore an apparent need to routinely monitor and study the make up of the pathogen population through virulence surveys over time. As the existence of races among pathogen populations present challenging problems to breeders and pathologists trying to develop resistant cultivars, efforts to identify sources of resistance to new races should receive priority in order to breed resistant cultivars of sorghum. It is expected that the results of this study will contribute to on-going breeding programmes targeted at reducing the rate of disease development with all races. This has been achieved for KSV 8 which is resistant to anthracnose and high yielding. It has since been released to Nigerian farmers as SAMAORG 14 (Aliyu et al., 1996).

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