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Morphological and pathogenic diversity among grain sorghum isolates of *Colletotrichum graminicola* in India+

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ABSTRACT : Thirty-three isolates of sorghum anthracnose pathogen *Colletotrichum graminicola,* collected during 1990-1994 from eight states of India were evaluated for morphological and pathogenic diversity. Colony morphology and growth rate of single-spore cultures of the isolates were compared by growing them on oat-meal agar (OMA) plates and incubating at 25°C for 10 days. The isolates showed considerable variation in colony morphology and significant (P <0.05) variations in growth rate. Pathogenic diversity of the isolates was evaluated by spray-inoculating (1 x 10^5 conidia ml^{-1}) on six differential sorghum lines in a greenhouse (25±2°C and r h >90%). The isolates significantly (P <0.001) varied in pathogenicity (latent period, virulence, and aggressiveness). "Virulence index" calculated as (virulence x aggressiveness) x latent $period^{-1}$ was derived to compare the pathogenic potential of the isolates. Hierarchical clustering of the isolates based on mean virulence index grouped the 33 isolates into 6 distinct clusters. Implication of these results in further virulence analysis and anthracnose management through host plant resistance is discussed.

Key words: *Sorghum bicolor, Colletotrichum graminicola,* Pathogenic diversity, Virulence index, Pathotypes.

Anthracnose of sorghum (*Sorghum bicolor* (L.) Moench), caused by *Colletotrichum graminicola* (Ces.) Wilson, is an important disease that damages leaves, stalks, peduncles, and panicles (Harris and Fisher, 1974) and causes substantial losses in both grain and forage production (Harris *et al.,* 1964; Powell *et al.,* 1977; Gorbet, 1977; Mishra and Siradhana, 1979; Thomas *et al.,* 1996). Management of this disease through host-plant resistance has often been unsuccessful because of wide genetic variation in the pathogen population (Ali and Warren, 1987; Cardwell *et al.,* 1989; Casela and Ferreira, 1995; Casela *et al.,* 1996; Ferreira and Casela, 1986; Pande *et al.,* 1991; Rao and Thakur, 1996; Thakur, 1995; Thomas, 1995) quickly resulting in matching the resistance genes of the host. Adequate knowledge of pathogen variability is not available for analysis of virulence genes in the pathogen and identification of resistance genes in the host genotypes. This study was aimed to determine the extent of morphological and pathogenic variability among *C. graminicola* isolates collected from different states of India.

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MATERIALS AND METHODS

The pathogen isolates

Thirty three isolates from eight Indian states (Andhra Pradesh-9; Karnataka-6; Gujarat-5; Maharashtra-8; Madhya Pradesh-2; Rajasthan-1; Tamil Nadu-1 and Uttar Pradesh-1) were collected as infected leaf samples (Table 1) from different sorghum cultivars and single-spore cultures were obtained on oat meal agar (OMA) plates at 25°C with 12 hr photoperiod for 10 days. The isolates were tested for infectivity on a anthracnose susceptible sorghum line IS 18442. These single spore isolates were maintained on OMA slants at 4°C.

Variation in growth characteristics

The isolates were grown on OMA plates at 25°C for 7 days and mycelial plugs (5 mm dia.) were removed from the margins of each culture isolate and transferred onto fresh OMA plates. Three replications (one plate/replication) were maintained for each isolate and incubated at 25°C for 20 days. The colony growth and morphology were observed 10 days after incubation-Appressoria-and-selerotia-m-the-eultures-were observed after 15 and 20 days, respectively, under a light microscope.

Pathogenicity

Pathogenic variability of the isolates was evaluted in a greenhouse using differential lines (Mathur *et al.,* 1997). Surface sterilized seeds of six differentials (A 2267-2, IRAT 204, IS 3758, IS 8354, IS 3089, IS 18442) were sown in sterilized black soil: sand: farm-yard manure mix (3:2:2 by volume) in 18cm square plastic pots in a glasshouse (25±2°C, rh <90%). The isolates were grown in 2% oat meal broth for 96 h at 25°C in an incubator shaker at 125 rpm with 12 h photoperiod for 10 days. Conidia were separated and a concentration of 1 x 10^5 conidia ml^{-1} was prepared using a hemocytometer, and two drops of Tween-20 were added to each suspension. Plants were spray-inoculated at the 5-6 leaf stage (21-day old) with each of the 33 isolates. The inoculated plants were air-dried and incubated at 25°C and >95% rh for 24 h. The plants were then moved to greenhouse benches at 25 ± 2°C.

The experiment was conducted in a completely randomized design involving 33 isolates x 6 host lines with two replications and 10 plants per replication, and was repeated once.

Observations for latent period were recorded every day for each isolate-sorghum line combination, beginning two days after inoculation. Plants were scored for disease reaction and disease severity 14 days after inoculation. Disease reaction was recorded as R = resistant (no symptoms or presence of chlorotic flecks); MR = moderately resistant (hypersensitive, necrotic lesions without sporulation); and S = susceptible (necrotic lesions with sporulation). Disease severity was recorded on a 1-9 scale on fourth and fifth inoculated leaves, where 1 = no lesions, 2 = 1-5%, 3 = 6-10%; 4 = 10-20%; 5 = 21-30%; 6 = 31-40%; 7 = 41-50%; 8 = 51-75%; and 9 = > 75% leaf area covered with lesions.

Table 1. Distribution of *Colletotrichum graminicola* isolates of sorghum from different locations in eight Indian states

State	District	No. of isolates	Isolates Nos.	Year of collection
Andhra Pradesh	Medak	3	Cg 001, 042, 044	1992
"	Mehaboobnagar	4	Cg 004, 007, 052, 084	1992 .
"	Warangal	1	Cg 008	1990
"	Karimnagar	1	Cg 009	1990
Karnataka	Bijapur	1	Cg 015	1992
"	Dharwad	3	Cg 017*	1992
			Cg 019	1991
			Cg 080	1993
"	Raichur	2	Cg 020, 083	1992
Maharashtra	Amaravathi	2	Cg 057, 064	1993
"	Jalgaon	2	Cg 070, 073	1993
"	Nanded	1	Cg 062	1993
"	Nagapur	1	Cg 088 **	1994
"	Pune	1	Cg 069	1993
"	Yavatmal	I	Cg 056	1993
Gujarat	Surat	5	Cg 031, 032, 035, 037, 040	1992
Madhya Pradesh	Indorc	2	Cg 047, 085	1992 ,
Rajasthan	Udaipur	1	Cg 029	1992
Tarailnadu	Tirunelveli	1	Cg 048	1993
Uttar Pradesh	Nainital	1	Cg 079	1993

*Isolated from cv. D-340; **Isolated from CSH-9; Remaining isolates collected from local landraces

Host-pathogen interaction model

Host-pathogen interaction model was determined on the basis of virulence - the potential of an isolate to infect a host line, measured qualitatively as disease reaction types (R, MR, S) and expressed quantitatively as R = 1; MR = 2 and S = 3, and aggressiveness - the amount of damage caused by a virulent isolate to the host line, measured quantitatively on a disease severity scale of 1-9. For designating virulence, isolates showing disease reaction of R/MR or MR on a sorghum line were considered as avirulent. Those showing R/MR/S or MR/S or S were considered as virulent 'Virulence index' was calculated as:

virulence index (VI) = [virulence (V) x aggressiveness (A)] x latent period $(L)^{-1}$

or $VI=(VA)\ L^{-1}$

Statistical analysis

Standard error of means was determined to compare significant differences in colony diameters among isolates. Analysis of variance was done using GENSTAT (1986) to determine the significant interactions of isolate and lines in two experimental runs separately. The error MS of two experimental runs was subjected to F-test. The error variances were considered homogenous as the highest error MS was not three-fold larger than the smallest error MS (Gomez and Gomez, 1984). The data from the two experimental runs were pooled and means were estimated through analysis of variance. Similarities of 33 isolates based on virulence index was determined through hierarchical clustering procedure using Euclidean test of average linkage cluster method (GENSTAT, 1986). A dendrogram for the 33 isolates was produced to determine the variation in virulence index.

RESULTS

Morphological variation

Significant (P<0.05) differences in colony growth were observed among the 33 isolates. Isolate Cg 017 had the minimum colony growth (4.0 cm) compared to Cg 032 which had the maximum growth (8.5 cm) on OMA plates (Table 2). There were significant variations in colony growth within the isolates of Andhra Pradesh, Karnataka, Maharashtra, and Madhya Pradesh, but no such variation was recorded for the isolates from Gujarat. Most of the isolates had raised white, dirty white or greyish white to grey growth except Cg 008, Cg 019 and Cg 088 which had submerged growth (Table 2). The mycelium was either felty or wooly. Isolates Cg 008, Cg 019, and Cg 088 had lilac-white grey to lilac grey mycelial growth. Acervuli development was distinct in most of the isolates, except Cg 001, 007, 009, 015, 057, 069, 073, 035, 037, and 048 in which acervuli were indistinct. In general distinct acervuli had more number of setae/acervulus compared to indistinct acervuli. Appressoria were observed in 21 isolates and sclerotia in 10 isolates, of which two-Cg 009 and Cg 044 produced abundant sclerotia (Table 2).

Pathogenic variation

Latent period

The mean latent period of isolates varied from 2.5 to 9.5 days (Table 3). The isolate Cg 088 had the shortest mean latent period of 3.4 days across sorghum lines while Cg 069 had the longest (5.3 days). Among the sorghum lines, longest mean latent period (7.5 days) across the isolates was on A 2267-2, followed by 6.8 days on IRAT 204, and the shortest (3.3 days) on IS 8354 and IS 18442 (Table 3). Highly significant (P <0.001) effects of isolates, host genotypes and isolate x host genotype interactions were observed for latent period (Table 4).

Virulence

Five of the 33 isolates were virulent on A 2267-2; 12 on IRAT 204, and 30 on IS 8354 (Table 5). All the isolates were virulent on IS 3758, IS 3089 and IS 18442. Twenty eight, 21 and 3 isolates were avirulent on A 2267-2, IRAT 204 and IS 8354, respectively. Five isolates (Cg 042, Cg 044, Cg 015, Cg 035 and Cg 048) were more virulent than other isolates as these could infect all the six sorghum lines.

Among the nine isolates from Andhra Pradesh, two isolates Cg 042 and Cg 044 were virulent on all the six lines while others were avirulent on A 2267-2 and exhibited varied reactions on the remaining five lines (Table 5). Among the six isolates from Karnataka, Cg 015 was virulent on all the lines; Cg 019 was avirulent on A 2267-2; Cg 017 was avirulent on A 2267-2, IRAT 204 and IS 8354. Among the eight isolates from Maharashtra, all were avirulent on A 2267-2; two (Cg 073 and Cg 088) were virulent on five lines and one, Cg 057 was avirulent on A 2267-2, IRAT 204 and IS 8354. Among the five isolates from Gujarat, Cg 035 was virulent on all the six lines; two isolates (Cg 031 and Cg 032) were virulent on five lines. All the isolates from Andhra Pradesh. Karnataka Maharashtra and Gujarat were virulent on IS 3758, IS 3089 and IS 18442.

Table 2. Morphological characteristics of 33 isolates of *Colletotrichum graminicola* 10-20 days after incubation on oat meal agar at 25°C

Isolate desig-nation	Colony growth	Colour	Acervulus development	No. of setae/ acervulus	Appre-ssoria	Scle-rotia	Colony dia. (cm)[a]
Cg 001 (AP)	Raised, wooly	Dirty white-grey	Indistinct	1-2	+	-	8.3
Cg 004	Raised, felty	Grey	Distinct	3-5	-	-	7.7
Cg 007	Raised, felty	Dirty white-grey	Indistinct	1-2	-	-	8.4
Cg 008	Submerged, felty	Lilac White-grey	Distinct	5-6	+	-	8.0
Cg 009	Raised, wooly	Greyish-white	Indistinct	-	-	+	4.1
Cg 042	Raised, felty	White	Distinct	1-2	+	-	7.5
Cg 044	Raised, wooly	Whitish-grey	Distinct	1-2	+	+	6.2
Cg 052	Raised, wooly	Lilac-grey	Distinct	2-10	-	-	8.4
Cg 084	Raised, wooly	Whitish-grey	Distinct	2-10	-	+	8.0
Cg 015 (Kar)	Raised, felty	Greyish-white	Indistinct	-	-	-	7.5
Cg 017	Raised, wooly	Greyish-white	Distinct	-	+	+	4.0
Cg 019	Submerged, felty	Lilac, white-grey	Indistinct	2-5	+	+	8.1
Cg 020	Raised, wooly	Grey	Distinct	6-10	+	-	6.0
Cg 080	Raised, wooly	Dark grey	Distinct	1-2	+	+	6.9
Cg 083	Raised, wooly	Dirty-white	Distinct	3-4	-	-	7.6
Cg 056 (Mah)	Raised, wooly	Grey	Distinct	1	+	-	7.9
Cg 057	Raised, felty	Greyish-white	Indistinct	-	+	+	8.0
Cg 062	Raised, wooly	Whitish-grey	Distinct	>20	+	+	6.4
Cg 064	Raised, wooly	Whitish-grey	Distinct	6-10	+	-	8.0
Cg 069	Raised, wooly	Lilac-white grey	Indistinct	3-10	+	-	7.1
Cg 070	Raised, wooly	Whitish-grey	Distinct	1-10	-	-	4.1
Cg 073	Raised, wooly	Dirty white-grey	Indistinct	1-2	+	-	5.8
Cg 088	Submerged	Lilac-grey	Distinct	2-10	+	+	7.1
Cg 031 (Guj)	Raised, felty	Greyish-white	Distinct	>20	+	-	8.3
Cg 032	Raised, felly	Greyish-white	Distinct	10	-	-	8.5
Cg 035	Raised, felty	Grey	Indistinct	1	-	-	7.4
Cg 037	Raised, wooly	Grey	Indistinct	-	+	-	7.0
Cg 040	Raised, wooly	Greyish-white	Distinct	7-10	-	-	8.5
Cg 047 (MP)	Raised, felty	Greyish-white	Distinct	10-12	-	+	5.6
Cg 085	Raised, wooly	Lilac-grey	Distinct	2-10	+	-	7.3
Cg 029 (Raj)	Raised, wooly	Whitish-grey	Distinct	2-8	-	-	6.1
Cg 048 (TN)	Raised, felty	Greyish-white	Indistinct	2-5	+	+	8.0
Cg 079 (UP)	Raised, wooly	Whitish-grey	Distinct	1-7	+	-	7.1

+ = present; - = absent.

a = Mean of 3 replications.

- Colony-dia. SE(m)=+0.337; LSD (P<0.05) =0.95).

AP - Andhra Pradesh, Kar - Karnataka, Mah - Maharashtra, Guj - Gujarat, MP - Madhya Pradesh, Raj - Rajasthan, TN - Tamilnadu, UP - Uttar Pradesh.

Table 3. Latent period[a] (days) of 33 isolates of *Colletofrichum graminicola* on six sorghum lines

Isolate designation	Host genotypes						
	A 2267-2	IRAT 204	IS 3758	IS 8354	IS 3089	IS 18442	Mean
Cg 001 (AP)	9.5	7.0	4.0	3.0	3.5	3.0	5.0
Cg 004	-[b]	7.5	3.5	3.0	3.5	3.5	4.2
Cg 007	-	4.5	3.0	4.0	3.5	3.5	3.7
Cg 008	7.5	7.5	4.5	3.0	3.5	3.5	4.9
Cg 009	9.5	6.5	4.0	3.5	3.0	3.5	5.0
Cg 042	6.5	5.5	3.5	3.0	3.5	3.3	4.2
Cg 044	6.3	6.5	4.0	3.3	4.0	3.0	4.5
Cg 052	-	6.5	4.5	3.5	4.0	3.5	4.4
Cg 084	-	7.5	5.0	4.8	4.0	3.5	5.0
Cg 015 (Kar)	6.5	7.0	4.0	3.0	3.5	3.0	4.5
Cg 017	7.0	9.5	3.0	3.0	3.0	3.5	5.0
Cg 019	-	9.0	4.0	3.5	4.0	3.5	4.8
Cg 020	-	6.0	3.5	3.0	3.0	3.3	3.8
Cg 080	-	7.5	4.0	3.0	3.3	6.5	4.9
Cg 083	-	8.0	4.5	4.0	4.3	4.0	5.0
Cg 056 (Mah)	-	7.0	3.0	3.5	3.5	3.0	4.0
Cg 057	-	7.0	3.5	4.5	3.8	3.0	4.4
Cg 062	9.5	6.0	4.0	3.5	3.5	3.0	4.9
Cg 064	-	-	3.5	3.0	3.5	3.3	4.0
Cg 069	9.5	6.5	4.5	4.0	4.5	3.0	5.3
Cg 070	-	7.5	3.5	3.5	4.0	3.0	4.3
Cg 073	-	6.5	3.0	3.0	3.8	2.8	3.8
Cg 088	-	5.0	3.0	3.0	3.0	3.0	3.4
Cg 031 (Guj)	6.8	6.3	3.5	3.0	3.0	3.0	4.3
Cg 032	-	7.5	4.0	3.8	3.5	3.5	4.5
Cg 035	7.0	7.5	3.5	3.8	3.0	3.3	4.7
Cg 037	8.5	7.0	3.0	3.0	3.0	2.5	4.5
Cg 040	7.0	5.0	3.5	3.0	3.5	3.0	4.2
Cg 047 (MP)	7.3	6.0	3.5	3.0	3.5	3.0	4.4
Cg 085	-	8.5	5.5	3.5	3.3	3.5	4.9
Cg 029 (Raj)	-	-	4.3	3.0	3.3	3.8	3.6
Cg 048 (TN)	4.5	3.5	4.0	3.0	. 3.5	3.0	3.6
Cg 079 (UP)	-	8.3	3.5	3.0	4.0	3.5	4.5
Mean	7.5	6.8	3.8	3.3	3.5	3.3	-

SE(m) for isolate means = ± 0.06

SE(m) for host genotype means = ± 0.02

SE(m) for isolate x host genotype means = ± 0.14

a. Mean of 2 experimental runs.

b. No infection.

AP -. Andhra Pradesh, Kar - Karnataka, Mah - Maharashtra, Guj - Gujarat, MP - Madhya Pradesh, Raj - Rajasthan, TN - Tamil Nadu, UP - Uttar Pradesh.

Table 4. Analysis of variance for latent period (LP), aggressiveness (AG) and Virulence index (VI)

	LP		MS		
Source of variation	df	MS	df	AG	VI
Experimental run (E)	1	101.48***	1	9.64***	4.23***
Isolates (I)	32	5.15***	32	16.37***	17.54***
Genotypes (G)	5	468.40***	5	736.08***	920.00**
E x I	32	3.47***	32	3.25***	4.72***
E x G	5	49.66***	5	5.94***	13.29***
I x G	140	2.31***	160	4.03***	5.13***
E x I x G	139	2.23***	160	0.81***	1.90***
Error	353	0.07	396	0.09	0.18

***Significant at *P* <0.001; **Significant at P<0.01.

Aggressiveness

All the isolates were more aggressive on IS 3758, IS 8354, IS 3089 and IS 18442 as compared to on A 2267-2 and IRAT 204 (Table 6). Fifteen isolates developed low to moderate disease severity (1.1- 4.0) on A 2267-2 and 31 on IRAT 204 (1.1- 6.0). Aggressiveness was significantly *(P* <0.001) influenced by host line, isolate, and their interactions (Table 4). Among the isolates, Cg 015 had the highest mean severity (6.0) across six host lines, while Cg 029 and Cg 085 had the lowest (2.9). Amongst the host lines, IS 3089 and IS 18442 had *6.6* and 6.8 mean severity scores, respectively, while A 2267-2 had lowest (1.2) across the 33 isolates (Table 6).

Virulence index

Significant (*P*<0.001) differences were observed between isolates, host lines, and their interactions for virulence index (Table 4). Isolate Cg 037 had the highest mean virulence index (6.3) across the sorghum lines, while Cg 085 had the lowest (2.5). Among genotypes, IS 18442 had highest virulence index (7.5), compared to the lowest on A 2267-2 (1.2) (Table 7).

Based on virulence index the nine isolates of Andhra Pradesh were grouped into two clusters: (i) Cg 042, 044, 009, 007, 001, 004, and (ii) Cg 052, 008 and 084; six isolates of Karnataka into three: (i) Cg 083 (ii) Cg 017, 019, 080, and (iii) Cg 015, Cg 020; eight isolates of Maharashtra into three: (i) Cg 064 and 088 (ii)Cg 069 (iii)Cg 056, 057,073,062, and 070; and five isolates of Gujarat into three: (i) Cg 031; (ii) Cg 037; and (iii) Cg 035, 032 and 040. However, based on pooled dendrogram the 33 isolates could be grouped into six clusters: (i). Cg 031, 052, 069, 079 (ii). Cg 029, 083, 085 (iii). Cg 015, 037, 047, 048 (iv). Cg 020, 035, 064, 088 (v). Cg 008, 017, 019, 056, 057, 062, 070, 073, 080, 084, and (vi). Cg 001, 004, 007, 009, 032, 040, 042, 044 (Fig. 1).

DISCUSSION

Isolates of *C. graminicola* collected from eight states of India during 1990-1994 exhibited significant diversity in colony morphology and growth rate and in pathogenic potential on a set of six sorghum differential lines. Variations within state were also evident as the isolates from the same state were assigned to different clusters. In the present study five isolates - Cg 042, 044, 015, 035 and 048 were more virulent on A 2267-2 and IRAT 204, the two known resistant lines, indicating matching virulence to resistance in these lines. However, there were mixed disease reactions (R/MR/S) on IRAT 204 , A 2267-2 and IS 8354. The variation in virulence on these specific lines in the two experiments conducted under controlled conditions indicate to some extent of intra-population variability due to heterogeneity, and also the significant line x isolate interactions. Evolution of such virulence is likely through somatic recombination. These results suggest the need for close monitoring for evolution of virulence in these states, particularly in the context of the current influx of popular high yielding hybrids. In a separate study in this lab, highly virulent forms have been isolated from popular sorghum hybrid CHS 9 from Maharashtra (Mathur *et al.,* 1997).

By using virulence index for determining the

Table 5. Virulence (Disease reactions)[a] of 33 isolates of *Colletotrichum graminicola* on six sorghum lines

Isolate designation	Host line					
	A 2267-2	IRAT 204	IS 3758	IS 8354	IS 3089	IS 18442
Cg 001 (AP)	R/MR	R/MR/S	S	MR/S	S	S
Cg 004	R	MR/R	S	S	S	S
Cg 007	R	MR/R	S	MR	S	S
Cg 008	R/MR	R/MR	S	MR/S	S	S
Cg 009	R/MR	MR	S	S	S	S
Cg 042	R/MR/S	R/MR/S	S	MR/S	S	S
Cg 044	R/MR/S	R/MR/S	S	S	S	S
Cg 052	R	MR/S	S	MR/S	S	S
Cg 084	R	R/MR	S	S	S	S
Cg 015 (Kar)	R/MR/S	S/MR	S	S	S	S
Cg 017	R/MR	R/MR	S	MR	S	S
Cg 019	R	R/MR/S	S	MR/S	S	S
Cg 020	R	MR/R	S	MR/S	S	S
Cg 080	R	R/MR	S	S	S	S
Cg 083	R	MR/R	S	S	S	S
Cg 056 (Mah)	R	MR/R	s	MR/S	S	S
Cg 057	R	MR/R	s	MR	S	S
Cg 062	R/MR	MR/R	MR/S	S/MR	S	S
' Cg 064	R	R	S	MR/S	S	S
Cg 069	R/MR	R/MR	MR/S	MR/S	S	S
Cg 070	R	R/MR	S/MR	S	S	S
Cg 073	R	MR/S	S	MR/S	S	S
Cg 088	R	S/MR	s	MR/S	S	S
Cg 031 (Guj)	R/MR	R/MR/S	MR/R/S	MR/S	S	S
Cg 032	R	R/MR/S	S	S	S	S
Cg 035	R/MR/S	MR/R/S	S	S	S	S
Cg 037	R/MR	MR	S	S/MR	S	S
Cg 040	MR/R	MR/R	S	MR/S	S	S
Cg 047 (MP)	R/MR	MR/R	S	S	S	S
Cg 085	R	R/MR	S	MR/S	S	
Cg 029 (Raj)	R	R	R/MR/S	MR/S	S/MR	S
Cg 048 (TN)	S/MR/R	S/MR	S	S/MR	S	S
Cg 079 (UP)	R	R/MR	S/MR	S	S	S

a.From 2 experimental runs.

AP - Andhra Pradesh, Kar - Karnataka, Mah - Maharashtra, Guj - Gujarat, MP - Madhya Pradesh, Raj - Rajasthan, TN - Tamil Nadu, UP - Urtar Pradesh.

Table 6. Aggressiveness[a] (disease severity on a 1-9 scale) of 33 isolates of *Colletotrichum graminicola* on six sorghum lines

Isolate designation	Host line						
	A 2267-2	IRAT 204	IS 3758	IS 8354	IS 3089	IS 18442	Mean
Cg 001 (Ap	1.3	2.0	6.1	3.3	7.1	7.0	4.5
Cg 004	1.0	1.7	5.7	4.3	7.7	6.6	4.5
Cg 007	1.0	1.8	6.0	4.5	8.4	7.1	4.8
Cg 008	1.2	1.7	*5.8*	*3.8*	6.7	7.4	*4.4*
Cg 009	1.2	2,3	6.2	4.9	6.3	7.6	4.7
Cg 042	2.2	2.7	5.6	5.0	6.6	7.8	5.0
Cg 044	2.0	3.9	6.5	5.6	5.8	7.9	5.3
Cg 052	1.0	2.2	4.8	4.5	5.9	*6.6*	4.0
Cg 084	1.0	1.8	7.6	5.9	6.9	6,6	5.0
Cg 015 (Kar)	1.8	4.0	7.1	6.6	8.4	8.5	6.0
Cg 017	1.1	1.3	7.5	4.6	7.6	3.4	4.2
Cg 019	1.0	1.7	6.5	4.7	7.3	6.4	4.6
Cg 020	1.0	1.6	6.2	5.7	8.8	7.3	5.1
Cg 080	1.0	1.2	5.9	6.2	8.0	3.2	4.2
Cg 083	1.0	1.6	3.7	3.5	4.6	5.3	3.3
Cg 056 (Mah)	1.0	1.9	5.9	3.6	5.7	6.5	4.1
Cg 057	1.0	2.1	6.1	3.8	5.7	6.9	4.3
Cg 062	1.4	2.1	4.3	4.7	7.1	6.2	4.3
Cg 064	1.0	1.0	7.2	5.5	8.1	8.2	5.2
Cg 069	1.1	1.4	3.1	3.1	7.0	6.8	3.8
Cg 070	1.0	1.7	3.9	4.2	6.3	7.3	4.1
Cg 073	1.0	2.1	5.2	4.0	3.7	7.2	3.8
Cg 088	1.0	3.0	5.4	4.0	8.2	7.2	4.8
Cg 031 (Guj)	1.3	1.6	4.0	3.9	5.0	7.2	3.8
Cg 032	1.0	1.7	6.3	6.1	7.1	8.8	5.1
Cg 035	1.3	1.7	6.9	6.7	6.8	8.2	5.2
Cg 037	1.5	2.2	8.0	4.9	8.7	8.2	5.6
Cg 040	1.9	2.2	6.9	3.8	6.9	7.5	4.9
Cg 047 (MP)	1.2	2.0	8.1	5.1	8.5	8.3	5.5
Cg 085	1.0	1.3	3.5	2.1	3.4	2.6	2.9
Cg 029 (Raj)	1.0	1.0	1.9	3.9	2.5	7.3	2.9
Cg 048 (TN)	2.9	4.6	6.2	6.7	6.9	7.4	5.8
Cg 079 (UP)	1.0	1.5	5.6	4.3	4.5	3.5	3.4
Mean	1.2	2.0	5.7	4.6	6.6	6.8	

SE(m) for isolate means = ±0.06

SE(m) forhost genotype means = ± 0.03

SE(m) for isolate x host genotype means = ± 0.15

a.From 2 experimental, runs.

AP - Andhra Pradesh, Kar - Karnataka, Mah - Maharashtra, Guj - Gujarat, MP - Madhya Pradesh, Raj - Rajasthan, TN - Tamil Nadu, UP - Uttar Pradesh.

Table 7. Virulence index[a] [(Virulence x aggressiveness) x latent period^{-1}] of 33 isolates of *Colletotrichum graminicola* on six sorghum lines

Isolate designation	Host line						
	A 2267-2	IRAT 204	IS 3758	IS 8354	IS 3089	IS 18442	Mean
Cg 001 (AP)	1.2	1.6	6.0	4.2	7.2	8.0	4.7
Cg 004	1.0	1.4	5.9	5.3	7.8	6.9	4.7
Cg 007	1.0	1.7	7.0	3.5	8.2	7.3	4.8
Cg 008	1.2	1.3	4.8	4.1	6.8	7.5	4.3
Cg 009	1.1	1.7	6.1	5.4	7.3	7.8	4.9
Cg 042	1.8	2.2	5.7	5.5	6.7	8.3	5.0
Cg 044	1.7	2.5	5.9	6.2	5.5	8.9	5.1
Cg 052	1.0	1.8	4.3	3.8	5.4	,6.8	3.8
Cg 084	1.0	1.4	5.9	4.8	6.1	6.9	4.3
Cg 015 (Kar)	1.5	2.7	6.3	7.6	8.4	9.4	6.0
Cg 017	1.1	1.2	8.5	4.0	8.6	3.8	4.5
Cg 019	1.0	1.3	5.8	4.3	7. I	6.7	4.4
Cg 020	1.0	1.5	6.6	5.9	9.8	8.0	5.4
Cg 080	1.0	1.2	5.4	6.3	8.5	3.7	4.3
Cg 083	1.0	1.4	3.4	3.7	4.3	4.9	3.1
Cg 056 (Mah)	1.0	1.5	6.9	3.6	5.9	7.5	4.4
Cg 057	1.0	1.5 .	6.4	3.0	5.6	7.9	4.2
Cg 062	1.2	1.6	3.4	4.9	7.2	7.5	4.3
Cg 064	1.0	1.0	7.4	5.3	8.1	8.6	5.2
Cg 069	1.1	1.3	3.0	2.7	5.8	8.9	3.8
Cg 070	1.0	1.3	4.3	4.6	6.3	8.3	4.3
Cg 073	1.0	1.7	6.1	3.9	4.1	8.9	4.3
Cg 088	1.0	2.6	6.4	3.9	9.2	8.1	5.2
Cg 031 (Guj)	1.3	1.3	3.5	3.8	6.0	8.2	4.0
Cg 032	1.0	1.4	5.a	6.0	7.1	8.7	5.0
Cg 035	1.2	1.4	7.0	6.7	7.8	8.6	5.4
Cg 037	1.2	1.6	9.0	5.3	9.7	11.0	6.3
Cg 040	1.4	1.8	7.0	4.0	7.0	8.5	4.9
Cg 047 (MP)	1.2	1.7	8.1	6.1	8.5	9.3	5.8
Cg 085	1.0	1.2	3.0	2.4	4.2	3.2	2.5
Cg 029 (Raj)	1.0	1.0	2.0	3.7	3.2	7.1	3.0
Cg 048 (TN)	2.7	5.1	5.6	6.8	6.9	8.4	5.9
Cg 079 (UP)	1.0	1.2	5.9	5.3	4.5	4.2	3.7
Mean	1.2	1.7	5.71	4.7	6.8	7.5	

SE(m) for isolate means = ± 0.086
SE(m) for genotype means = ± 0.037
SE(m) for isolate x genotype means = ± 0.21

a.From 2 experimental runs.

AP - Andhra Pradesh, Kar - Karnataka, Mah - Maharashtra, Guj - Gujarat, MP - Madhya Pradesh, Raj - Rajasthan, TN - Tamil Nadu, UP - Uttar Pradesh.

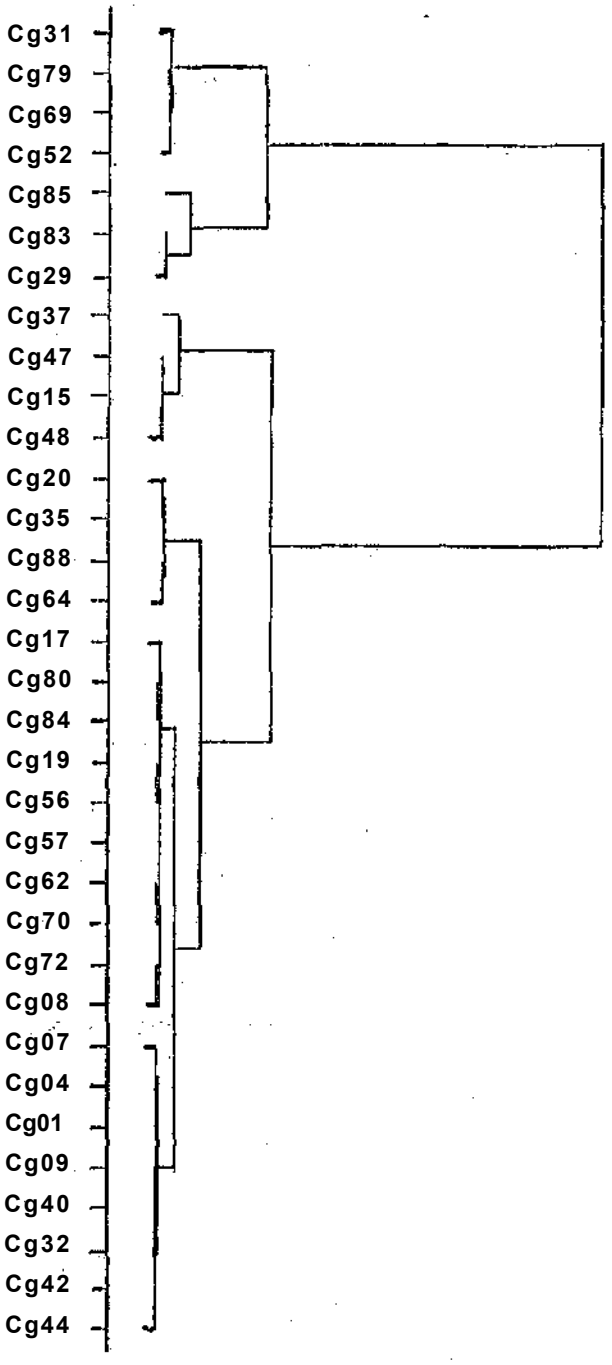

Fig. 1. Dendrogram showing successive dissimilarity between 33 isolates of *Colletotrichum graminicola* based on mean virulence index on six sorghum lines.

overall pathogenic potential of an isolate it was also possible-to-correlate the three independent pathogenicity parameters-latent period, virulence and aggressiveness. The analysis of variance indicated that significant differences for latent period in isolates, sorghum lines and their interactions were more attributed to the host lines than to isolates or to line-isolate interactions. From the results it appears that A 2267-2 and IRAT 204 have different genes for anthracnose resistance, and IS 8354 has partial resistance. In sorghum, anthracnose resistance is known to be governed by dominant genes (Frederiksen and Rosenow, 1971), but there is no information on the number of resistance loci as well as for the genes in lines showing partial (moderate) resistance or delayed disease development. The availability of the pathogen isolates infecting more than one resistant line may be of value for determining the resistance loci in the host (Crute, 1992).

Pande *et al.* (1991) observed that the nine monoconidial isolates of *C. gramimcola* from various sorghum cultivars from different locations in India varied in pathogenicity on 17 sorghum lines, and designated these as nine different races. However, in our studies all the 33 monoconidial isolates were grouped into six clusters with some variations within a cluster. These could be designated as six pathotypes. It would be more useful to assign lineage of the isolates based on virulence on resistant lines like A2267-2 and IRAT 204. Most of the lines used in this study have shown susceptibility to isolates of C. *graminicola.* Induction of more number of resistant lines, and studying virulence of these isolates would be useful for assigning lineage based on resistant lines, and also for characterization of resistant genes in these lines.

Many isolates tested in this study readily produced sclerotia in culture. These sclerotia might act as primary source of infection in the field (Casela and Frederiksen, 1993). However, a clear role of sclerotia in survival and pathogenic fitness of *C. graminicoh* is yet to be demonstrated. Morphological variations could not be correlated with pathogenic variability and these may be independent of each other as reported by Thomas *at al.* (1995). Pathogenicity is known to be independent of other features, including DNA polymorphism and virulence analysis based on phenotype using differential host lines seems useful in determining race structures of plant pathogens (Casela and Ferreira, 1995; Chen *at al.,* 1993). Further studies on virulence analysis, using more number of differentials for deter--mining-finer level of variability (Kelemu *et al.,* 1996) would further add to the knowledge of race structure of *C. gramimcola.*

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