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Cultural and Morphological Variability among the Isolates of *Alternaria burnsii* (Uppal, Patel and Kamat) Causing Blight of Cumin

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A B S T R A C T

Cultural and morphological variability studies on three different media viz., Potato Dextrose Agar, Richard's Synthetic Agar and Czapek's Dox Agar revealed considerable variation among the isolates of *A. burnsii* indicated the existence of variability in the pathogen. Moreover, Potato Dextrose Agar and Czapek's Dox Agar were found as an excellent media to support the growth and spore formation of isolates of *A. burnsii*, respectively. Initially, the fungal growth was light green, sometimes whitish green septate mycelial growth and finally grey to black in color with dirty white to brownish colony margin and fluffy radial, plain irregular radial and fluffy knotting growth pattern on three different media. Among fifteen isolates of *A. burnsii*, distinct differences in terms of conidial length, breadth, beak length and number of septa were recorded. The average conidial length varied from 50.89 to 63.76µm and breadth ranges from 20.24 to 25.47 µm with beak length of 28.73 to 47.33 µm. The transverse septa varied from 1 to 6 and longitudinal septa varied from to 0 to 3, respectively. In the present studies, glaring differences in conidial size were noticed among the isolates even when same medium was used for the growth of the isolates. It can be assumed that variation in the isolates may be inherent since isolates were collected from diverse agroclimatic zones of Gujarat. Hence, these variations in the conidial size indicated the existence of variability in this pathogen

Keywords

Alternaria burnsii, Cumin blight, Cumin, Variability, Isolate

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Introduction

Cumin (*Cuminum cyminum* L.) popularly known as Jeera or Jiroo is the most important spice crop inIndia. Gujarat is second largest producer next to Rajasthan with 50-55% of total production of India. Cumin crop suffers with blight disease caused by *Alternaria burnsii*. Cumin seed is used as culinary for flavouring vegetables, pickles, soups, etc. The seeds contain 17.7 per cent protein, 23.8 per cent fat, 35.5 per cent carbohydrates and 7.7 per cent minerals (Chadha, 2006). Cumin suffers from important diseases, *viz*., blight (*Alternaria burnsii*), wilt (*Fusarium oxysporum* f.sp. *cumini*) and powdery mildew (*Erysiphe polygoni*) which causes considerable qualitative and quantitative yield losses in the crop. Blight (*A. burnsii)* is of common occurrence in the areas where cumin is grown. Cumin blight was recorded for the first time from Kaira district of Gujarat. The

causative agent was reported to be *Alternaria burnsii* by Uppal *et al.,* (1938). Later on, it was reported from Rajasthan by Joshi (1955). The disease usually appears at flowering stage. The infected plants show small, isolated, whitish necrotic areas on the aerial parts, especially on tips of young leaves (Sharma, 2010). Diseased seed are small, deshaped, shriveled, very light and turn black in colour (Gemawat and Prasad, 1972). Various species of *Alternaria* have been reported to show variability in terms of cultural, morphological and molecular characteristics (Pandey *et al.,* 2005; Raja *et al.,* 2007; Singh and Singh, 2007; Varma *et al.,* 2007; Pipaliya and Jadeja, 2008 and Kumar *et al.,* 2008). Variability studies are important to document the changes occurring in populations and individual as variability in morphological, cultural and molecular traits indicate the existence of different pathotypes. The variability is well known phenomenon in genus *Alternaria* and may be noticed as changes in spore shape and size, growth and sporulation, pathogenicity, etc. understanding pathogen population structure and mechanisms by which variation arises within a population is of paramount importance for developing a successful disease management strategy lead to the development of resistant genotypes to the given set of pathogenic races. An attemptwas made to study the cultural and morphological variability among the isolates of *Alternaria burnsii* (Uppal, Patel and Kamat) causing blight of cumin

Materials and Methods

Cultural variability among the isolates of *A. burnsii*

The blight infected samples were collected from major cumin growing areas of Gujarat and isolations were made to study the cultural and morphological variation among the different isolates (Table 1).

Growth characters on solid media

For cultural variability the isolates of the pathogen were studied on three different media *viz*., Potato Dextrose Agar, Richard's Synthetic Agar and Czapek's Dox Agar medium sterilized in autoclave for 20 minutes at 15 lbs p.s.i. The 5 mm disc of pure culture of isolates was inoculated at the center of the pre poured Petri plates from ten days old actively growing culture. All inoculated plates were incubated at 28±1 ºC temperature in BOD incubator. Three repetitions were taken for each isolate. The growth rate was measured and colony characters, growth habit and sporulation visually observed after 24 hrs of incubation till the complete growth of the pathogen in Petri plates.

Morphological variability

The morphological characters of different isolates of *A. burnsii* including size of conidia (length and breadth), beak length and number of septa (Transverse and longitudinal) were measured from 8 days old culture under high power magnification (45X). The photomicrograph were taken by using camera attachment binocular microscope to show the typical spore morphology of the isolates. The conidial measurement of different isolates were done by using SImage 2013 Beta software.

Results and Discussion

The findings of the present study as well as relevant discussion have been presented under the following heads.

Cultural variability among the isolates of *A. burnsii*

The study on cultural characteristics of isolates of *A. burnsii* was carried out on three different solid media *viz*., Potato Dextrose

Agar, Richard's Synthetic Agar, Czapek's Dox Agar as described in 'materials and methods'. These isolates showed significant differences in cultural characters *viz.*, colony color, colony margin, colony diameter and sporulation of the isolates of *A. burnsii* on three different solid media.

Growth characteristics of isolates of *A. burnsii* on different solid media

The result presented in Table 2 and depicted in Plate (1A-1C & 2A-2C) revealed that there was a considerable variations among the colony characteristics of the different isolates on three different solid media.

Colony diameter and sporulation of isolates of *A. burnsii* on different media

Colony diameter

The result presented in Table 3 and Plate (1A-1C) revealed significant differences between isolates and media and also interaction. Among the three different media tested, maximum colony diameter on fifth day of observation was recorded in Czapek's Dox Agar (36.28 mm).

The next media was Richard's Synthetic Agar (33.51 mm) followed by Potato Dextrose Agar (30.88 mm).

In case of different isolates, maximum colony diameter (40.77 mm) was recorded in Ahmedabad isolate (Ab-13) and Banaskantha isolate (Ab-5). The next was the Morbi isolate (Ab-7) and Surendranagar isolate (Ab-15) with colony diameter (35.66) which was at par with Ahmedabad isolate (Ab-8) (35.44 mm), Patan isolate (Ab-6) and Surendranagar isolate (Ab-14) with (35.22 mm), Ahmedabad isolate (Ab-10) and Banaskantha isolate (Ab-2) with (34.11mm), Patan isolate (Ab-1) and Banaskantha isolate (Ab-9) with (33.88 mm). The moderate growth was recorded in Morbi isolate (Ab-4) and Surendranagr isolate (Ab-12) with (28.55 mm) and least growth was observed in Patan isolate (Ab-3) and Morbi isolate (Ab-11) with (25.77 mm).

The interaction between media and isolates was found significant, which indicated the variation among the isolates in utilizing the media. Among them, maximum colony diameter (49.33 mm) was recorded in Banaskantha isolate (Ab-5) and Ahmedabad isolate (Ab-13) on Czapek's Dox Agar media which was at par with same isolates on Richard's Agar media (48.66 mm). The least colony diameter of (18.66 mm) was recorded in Patan isolate (Ab-3) and Morbi isolate (Ab-11) on Richard's Synthetic Agar media.

Overall, the excellent colony growth was recorded on Czapek's Dox Agar media. The moderate growth was recorded on Richard's Synthetic Agar media and least growth was recorded in Potato Dextrose Agar media.

Eighth day observation

The result presented in Table 4 and Plate (2A-2C) revealed significant differences between isolates and media and also interaction. Among the three different media tested, maximum colony diameter on eighth days of observation was recorded in Potato Dextrose Agar (70.73 mm). The next best media was Czapek's Dox Agar (67.08 mm) followed by Richard's Synthetic Agar (59.93mm).

In case of different isolates, maximum colony diameter (72.44 mm) was recorded in Ahmedabad isolate (AB-8) which was at par with Patan isolate (Ab-1) and Banaskantha isolate (Ab-9) with (72.00 mm), Morbi isolate (Ab-7) and Surendranagar isolate (Ab-15) with (69.11 mm). The moderate growth was recorded in Patan isolate (Ab-6) and with (65.66 mm) which was at par with

Banaskantha isolates (Ab-2, Ab-5) with (65.44 mm, 65.66 mm) respectively, Ahmedabad isolates (Ab-10, Ab-13) with (65.44mm, 65.66) respectively, Surendranagar isolates (Ab-12, Ab-14) with (65.22 mm, 66.11 mm) respectively and Morbi isolate (Ab-4) with (65.22 mm). The least growth was observed in Patan isolate (Ab-3) and Morbi isolate (Ab-11) with (54.55 mm).

The interaction between media and isolates was found significant, which indicated the variation among the isolates in utilizing the media. Among them, maximum colony diameter (78.66 mm) was recorded in Morbi isolate (Ab-4), Surendranagar isolate (Ab-12) on Potato Dextrose agar media and Morbi isolate (Ab-7), Banaskantha isolate (Ab-9) on Czapek's Dox agar media The least colony diameter of (46.33 mm) was recorded in Patan isolate (Ab-3) and Morbi isolate (Ab-11) on Richard's Synthetic Agar media.

Overall, the excellent colony growth was recorded on Potato Dextrose Agar media. The moderate growth was recorded on Czapek's Dox Agar media and least growth was recorded in Richard's Synthetic Agar media.

Sporulation

With regard to sporulation, the result presented in Table 5 revealed that abundant sporulation was recorded in Potato Dextrose Agar media (8.62) which was at par with Czapek's Dox Agar media (8.36 mm). The sporulation was recorded scanty in Richard's Synthetic Agar media (8.09 mm).

In case of isolates, good sporulation (9.48) was recorded in Banaskantha isolate (Ab-2) and surendranagar isolate (Ab-12) which was at par with Banaskantha isolate (Ab-9), Patan isolate (Ab-6), Morbi isolate (Ab-7) and Surendranagar isolates (Ab-15) with (9.44, 9.34, 9.19, 8.86) respectively. Banaskantha isolate (Ab-5) with (8.63) which was at par with Ahmedabad isolates (Ab-3, Ab-8, Ab-10) with (7.95, 8.23, 8.13) respectively. The moderate sporulation was recorded in Ahmedabad isolate (Ab-13) with (7.73) which was at par with Patan isolate (Ab-1) with (7.68), Surendranagar isolate (Ab-14) with (7.37) and Morbi isolate (Ab-11) with (7.35). The scanty sporulation was recorded in Morbi isolate (Ab-4) (6.48).

The interaction between media and isolates was found significant which indicated the variation among the isolates in utilizing the media. Among them, maximum sporulation (11.15) was recorded in Banaskantha isolate (Ab-2) and Surendranagar isolate (Ab-12) on Potato Dextrose Agar media which was at par with Patan isolate (Ab-6) with (10.34), Banaskantha isolate (Ab-9) with (10.33) and Ahmedabad isolate (Ab-10) with (10.14) on the same media. The least sporulation (5.04) was recorded in Morbi isolate (Ab-4) on Potato Dextrose Agar media.

Overall, the excellent sporulation was recorded on Potato dextrose agar media. Which was at par with Czapek's Dox Agar media and least growth was recorded in Richard's Synthetic Agar media.

Morphological variability among the isolates of *A. burnsii*

In all the isolates, the conidia were single, lateral, straight or curved, smooth walled with 1 to 6 septa, pale brown. However, the size of conidia varied among the isolates, some of them were very long and narrow, while some were fairly board. The result presented in Table 6-9 and Plate (1A-1C & 2A-2C) revealed that there were distinct variations among the morphological characters *viz*., length and breadth of conidia, conidial beak length and conidial septation of the isolates of *A. burnsii* on seven different solid media.

Table.1 List of *Alternaria burnsii* isolates collected from different locations

Sr. No.	Isolate designation	Location of isolate
1	Ab-1	Patan
2	Ab-2	Banaskantha
3	Ab-3	Patan
4	Ab-4	Morbi
5	Ab-5	Banaskantha
6	Ab-6	Patan
7	Ab-7	Morbi
8	Ab-8	Ahmedabad
9	Ab-9	Banaskantha
10	Ab-10	Ahmedabad
11	Ab-11	Morbi
12	Ab-12	Surendranagar
13	Ab-13	Ahmedabad
14	Ab-14	Surendranagar
15	Ab-15	Surendranagar

Ab - *Alternaria burnsii*

Table.2 Colony characteristics (colony color, colony margin and growth pattern) of isolates of *A. burnsii* on different media

Isolate designation	Potato Dextrose Agar	Richard's Synthetic Agar	Czapek's Dox Agar
Ab-1	Greyish black Light brown Fluffy radial	Greyish brown Light Brown Plain irregular radial	Whitish brown Brownish Fluffy radial
Ab-2	Greyish black Light brown Fluffy radial	Greyish white Dirty white Plain irregular radial	Whitish brown Brownish Plain irregular radial
Ab-3	Greyish white Dirty white Fluffy radial	Greyish brown Light brown Plain irregular radial	Greyish white Dirty white Fluffy radial
Ab-4	Greyish black Light brown Fluffy radial	Greenish brown Brownish Plain irregular radial	Greenish black Brownish Plain irregular radial
Ab-5	Greenish dark black Dirty white Fluffy radial	Greenish brown Brownish plain irregular radial	Greenish black Dirty white Plain irregular radial
Ab-6	Greenish dark black Dirty white Fluffy radial	Greyish white Dirty white Fluffy radial	Whitish brown Brownish Plain irregular radial
Ab-7	Greyish white Light brownish Fluffy radial	Greyish white Brownish Fluffy radial	Greyish white Dirty white Plain irregular radial
Ab-8	Greenish dark black Blackish Fluffy radial	Greyish white Dirty white Plain irregular radial	Greyish white Brownish Fluffy radial
Ab-9	Greenish dark black Blackish Fluffy radial	Greyish white Dirty white Fluffy radial	Greenish black Brownish Plain irregular radial
Ab-10	Greyish black Light brown Fluffy knotting	Greyish white Light brown Plain irregular radial	Greyish black brownish Fluffy radial
Ab-11	Greyish white Blackish Fluffy radial	Greenish brown Brownish Plain irregular radial	Greyish white Dirty white Fluffy radial
Ab-12	Greenish dark black Dirty white Fluffy knotting	Greyish white Dirty white Fluffy radial	Greyish black Brownish Fluffy radial
Ab-13	White Dirty white Fluffy knotting	Greyish brown Light brown Fluffy radial	Greyish white Dirty white Fluffy knotting
Ab-14	Greyish black Light brownish Fluffy radial	Greyish white Dirty white Plain irregular radial	Greyish black Brownish Fluffy radial
Ab-15	White Dirty white Fluffy knotting	Greyish white Dirty white Fluffy radial	Greyish white Dirty white Fluffy knotting

Table.3 Colony diameter of isolates of *A. burnsii* on different media on 5th day

Isolates/ Media	Colony diameter (mm)			Mean (mm)
	Potato Dextrose Agar	Richard's Synthetic Agar	Czapek's Dox Agar	
Ab-1	34.66	33.00	34.00	33.88
Ab-2	31.66	38.00	32.66	34.11
Ab-3	33.00	18.66	25.66	25.77
Ab-4	24.66	22.33	38.66	28.55
Ab-5	24.33	48.66	49.33	40.77
Ab-6	35.66	38.33	31.66	35.22
Ab-7	31.66	33.00	42.33	35.66
Ab-8	32.00	38.67	35.66	35.44
Ab-9	34.66	33.00	34.00	33.88
Ab-10	31.66	38.00	32.66	34.11
Ab-11	33.00	18.66	25.66	25.77
Ab-12	24.66	22.33	38.66	28.55
Ab-13	24.33	48.66	49.33	40.77
Ab-14	35.66	38.33	31.66	35.22
Ab-15	31.66	33.00	42.33	35.66
Mean	30.88	33.51	36.28	

	S.Em. ±	C.D. at 5%	C.V.%
Media	0.309	0.870	6.18
Isolates	0.691	1.944	
Isolates x Media	1.197	3.368	

Table.4 Colony diameter of isolates of *A. burnsii* on different media on 8th day

Isolates/ Media	Colony diameter (mm)			Mean (mm)
	Potato Dextrose Agar	Richard's Synthetic Agar	Czapek's Dox Agar	
Ab-1	73.00	64.33	78.66	72.00
Ab-2	73.00	64.00	59.33	65.44
Ab-3	64.00	46.33	53.33	54.55
Ab-4	78.66	48.33	68.66	65.22
Ab-5	74.00	59.33	63.66	65.66
Ab-6	70.33	59.66	68.66	66.22
Ab-7	58.33	70.33	78.66	69.11
Ab-8	78.33	74.33	64.66	72.44
Ab-9	73.00	64.33	78.66	72.00
Ab-10	73.00	64.00	59.33	65.44
Ab-11	64.00	46.33	53.33	54.55
Ab-12	78.66	48.33	68.66	65.22
Ab-13	74.00	59.33	63.66	65.66
Ab-14	70.33	59.66	68.33	66.11
Ab-15	58.33	70.33	78.66	69.11
Mean	70.73	59.93	67.08	72.00

	S.Em. ±	C.D. at 5%	C.V.%
Media	0.608	1.711	6.19
Isolates	1.360	3.826	
Isolates x Media	2.355	6.627	

Table.5 Relative amount of sporulation of isolates of *A. burnsii* on different media

Isolates/ Media	Sporulation on media(n x $10^{4)}$			Mean
	Potato Dextrose Agar	Richard's Synthetic Agar	Czapek's Dox Agar	
Ab-1	7.24	7.90	7.90	7.68
Ab-2	11.15	8.15	9.15	9.48
Ab-3	7.18	7.84	8.84	7.95
Ab-4	5.04	7.70	6.70	6.48
Ab-5	8.75	9.08	8.08	8.63
Ab-6	10.34	8.34	9.34	9.34
Ab-7	9.64	8.64	9.30	9.19
Ab-8	8.25	8.25	7.91	8.13
Ab-9	10.33	8.66	9.33	9.44
Ab-10	10.14	5.42	9.14	8.23
Ab-11	7.24	7.90	6.90	7.35
Ab-12	11.15	8.15	9.15	9.48
Ab-13	7.18	8.51	7.51	7.73
Ab-14	7.04	7.70	7.37	7.37
Ab-15	8.75	9.08	8.75	8.86
Mean	8.62	8.09	8.36	

	S.Em.±	C.D. at 5%	C.V.%
Media	0.122	0.343	9.78
Isolates	0.273	0.767	
Isolates x Media	0.472	1.329	

Table.6 Conidial length of isolates of *A. burnsii* on different media

Isolates/ Media	Conidial length (µm)			Mean (µm)
	Potato Dextrose Agar	Richard's Synthetic Agar	Czapek's Dox Agar	
Ab-1	67.30	56.57	60.24	61.37
Ab-2	60.90	62.70	60.88	61.49
Ab-3	59.32	53.48	56.88	56.56
Ab-4	64.11	56.27	53.16	57.85
Ab-5	60.13	56.87	62.66	59.89
Ab-6	62.66	64.93	63.70	63.76
Ab-7	59.81	50.89	59.58	56.76
Ab-8	62.41	63.43	60.36	62.07
Ab-9	59.43	55.43	59.63	58.16
Ab-10	59.23	62.20	59.88	60.44
Ab-11	59.99	56.53	60.43	58.98
Ab-12	57.44	56.27	54.23	55.98
Ab-13	59.13	56.77	61.10	59.00
Ab-14	61.66	64.93	63.70	63.43
Ab-15	59.94	51.43	60.31	57.23
Mean	60.90	57.91	59.78	

	S.Em.±	C.D. at 5%	C.V.%
Media	0.547	1.541	6.17
Isolates	1.224	3.445	
Isolates x Media	2.120	5.968	

Table.7 Conidial breadth of isolates of *A. burnsii* on different media

Isolates/ Media	Conidial breadth (µm)			Mean (µm)
	Potato Dextrose Agar	Richard's Synthetic Agar	Czapek'sDox Agar	
Ab-1	25.43	24.13	23.78	24.45
Ab-2	22.93	22.38	21.92	22.41
Ab-3	22.41	22.00	21.43	21.94
Ab-4	21.81	20.40	21.17	21.13
Ab-5	22.53	20.73	23.40	22.22
Ab-6	25.08	23.25	24.35	24.23
Ab-7	23.11	20.74	22.82	22.22
Ab-8	23.07	22.34	22.83	22.75
Ab-9	26.35	24.83	25.22	25.47
Ab-10	22.93	25.08	22.25	23.42
Ab-11	23.41	23.90	21.34	22.88
Ab-12	23.44	19.60	23.17	22.07
Ab-13	21.25	19.07	22.10	20.80
Ab-14	23.25	22.85	23.75	23.28
Ab-15	19.44	18.17	23.12	20.24
Mean	23.09	21.96	22.84	

	S.Em. ±	C.D. at 5%	C.V.%
Media	0.216	0.609	6.41
Isolates	0.483	1.361	
Isolates x Media	0.837	2.357	

Table.8 Beak length of conidia of isolates of *A. burnsii* on different media

Isolates/ Media	Conidial beak length (µm)			Mean (µm)
	Potato Dextrose Agar	Richard's Synthetic Agar	Czapek's Dox Agar	
Ab-1	41.92	38.25	40.93	40.37
Ab-2	36.36	34.70	37.70	36.25
Ab-3	34.69	31.01	34.01	33.23
Ab-4	33.10	31.44	33.44	32.66
Ab-5	30.85	28.16	34.50	31.17
Ab-6	46.33	40.12	45.68	44.04
Ab-7	29.87	26.86	32.20	29.64
Ab-8	41.74	40.07	42.07	41.29
Ab-9	41.77	40.43	41.10	41.10
Ab-10	34.73	37.73	35.40	35.95
Ab-11	33.47	32.81	33.81	33.36
Ab-12	33.86	33.20	34.16	33.74
Ab-13	29.16	32.50	32.46	31.37
Ab-14	48.80	48.10	45.10	47.33
Ab-15	27.84	31.18	27.18	28.73
Mean	36.30	35.10	36.65	

	S.Em.±	C.D. at 5%	C.V.%
Media	0.3283	0.924	6.11
Isolates	0.7341	2.066	
Isolates x Media	1.271	3.578	

Table.9 Conidial septation of isolates of *A. burnsii* on different media

Isolates/ Media	Conidial septation (No.)					
	Potato Dextrose Agar		Richard's Synthetic Agar		Czapek's Dox Agar	
	T*	L*	T	L	T	L
Ab-1	0-3	0-1	0-2	0-1	0-2	0-1
Ab-2	1-3	0-2	0-2	0-2	0-3	0-2
Ab-3	1-3	0-1	0-2	0-1	0-3	0-2
Ab-4	1-4	0-1	0-1	0-1	1-2	0-1
Ab-5	1-5	0-2	0-2	0-1	1-2	0-1
Ab-6	1-4	0-2	0-2	0-1	1-3	0-1
Ab-7	0-5	0-2	0-3	0-2	0-2	0-2
Ab-8	1-6	0-3	0-2	0-2	0-3	0-2
Ab-9	1-5	0-2	0-2	0-1	1-3	0-1
Ab-10	1-5	0-1	0-2	0-1	1-3	0-2
Ab-11	1-3	0-1	0-2	0-2	1-4	0-2
Ab-12	1-4	0-2	0-3	0-1	1-2	0-1
Ab-13	1-5	0-1	0-2	0-1	1-3	0-2
Ab-14	1-3	0-2	0-2	0-1	0-4	0-2
Ab-15	1-3	0-1	0-2	0-1	1-3	0-1

*T= Transverse, L= Longitudinal

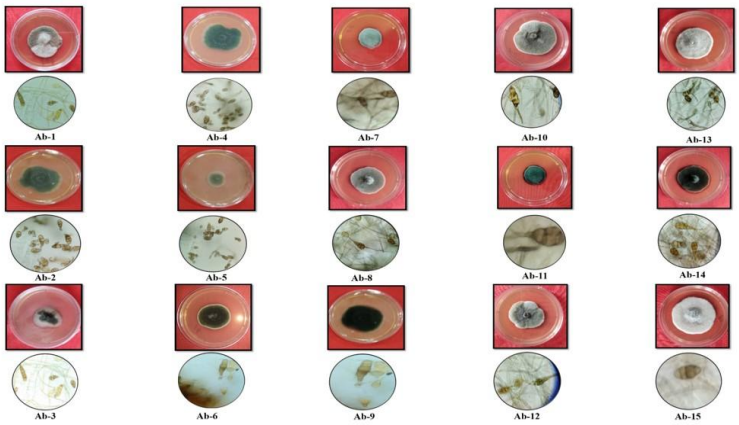

Plate 1A Mycelial growth and spore morphology of *Alternaria burnsii* on different media on 5th day
Media : Potato dextrose agar
Isolates : Ab-1=Patan; Ab-2=Banaskantha; Ab-3=Patan; Ab-4=Morbi; Ab-5=Banaskantha; Ab-6=Patan; Ab-7=Morbi; Ab-8=Ahmedabad Ab-9=Banaskantha; Ab-10=Ahmedabad; Ab-11=Morbi; Ab-12=Surendranagar; Ab-13=Ahmedabad; Ab-14=Surendranagar; Ab-15= Surendranagar

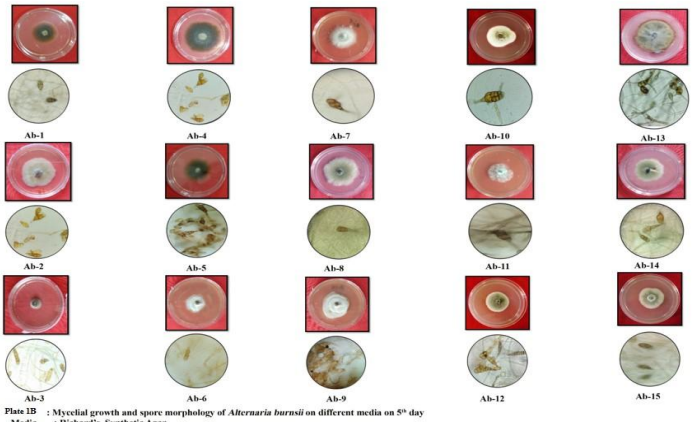

Plate 1B : Mycelial growth and spore morphology of ***Alternaria burnsii*** on different media on 5th day
Media : Richard's Synthetic Agar
Isolates : Ab-1=Patan; Ab-2=Banaskantha; Ab-3=Patan; Ab-4=Morbi; Ab-5=Banaskantha; Ab-6=Patan; Ab-7=Morbi; Ab-8=Ahmedabad Ab-9=Banaskantha; Ab-10=Ahmedabad; Ab-11=Morbi; Ab-12=Surendranagar; Ab-13=Ahmedabad; Ab-14=Surendranagar; Ab-15= Surendranagar

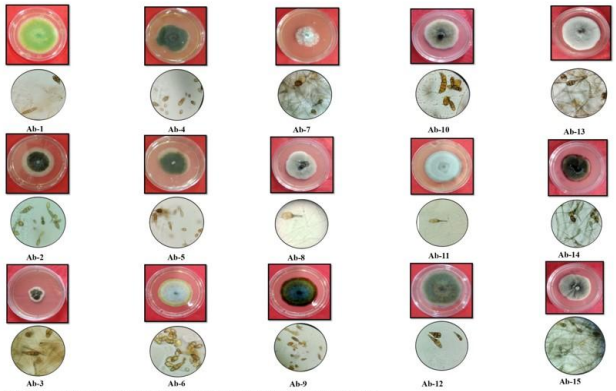

Plate 1C : Mycelial growth and spore morphology of ***Alternaria burnsii*** on different media on 5th day
Media : Czapek's Dox Agar
Isolates : Ab-1=Patan; Ab-2=Banaskantha; Ab-3=Patan; Ab-4=Morbi; Ab-5=Banaskantha; Ab-6=Patan; Ab-7=Morbi; Ab-8=Ahmedabad Ab-9=Banaskantha; Ab-10=Ahmedabad; Ab-11=Morbi; Ab-12=Surendranagar; Ab-13=Ahmedabad; Ab-14=Surendranagar; Ab-15= Surendranagar

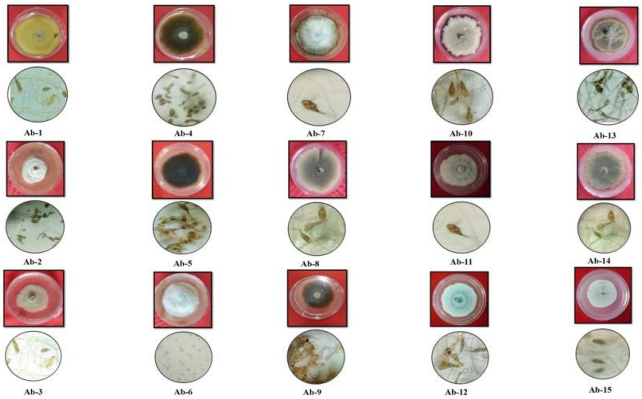

Plate 2B : Mycelial growth and spore morphology of ***Alternaria burnsii*** on different media on 8th day
Media : Richard's Synthetic Agar
Isolates : Ab-1=Patan; Ab-2=Banaskantha; Ab-3=Patan; Ab-4=Morbi; Ab-5=Banaskantha; Ab-6=Patan; Ab-7=Morbi; Ab-8=Ahmedabad Ab-9=Banaskantha; Ab-10=Ahmedabad; Ab-11=Morbi; Ab-12=Surendranagar; Ab-13=Ahmedabad; Ab-14=Surendranagar; Ab-15= Surendranagar

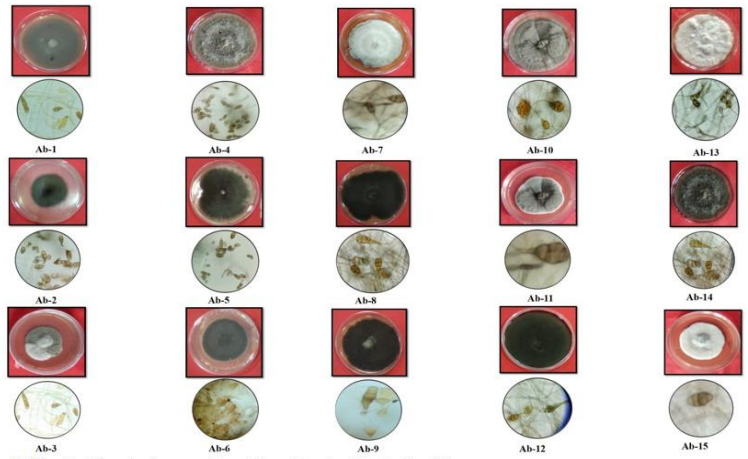

Plate 2A : Mycelial growth and spore morphology of *Alternaria burnsii* on different media on 8th day
Media : Potato dextrose agar
Isolates : Ab-1=Patan; Ab-2=Banaskantha; Ab-3=Patan; Ab-4=Morbi; Ab-5=Banaskantha; Ab-6=Patan; Ab-7=Morbi; Ab-8=Ahmedabad Ab-9=Banaskantha; Ab-10=Ahmedabad; Ab-11=Morbi; Ab-12=Surendranagar; Ab-13=Ahmedabad; Ab-14=Surendranagar; Ab-15= Surendranagar

Plate 2C : Mycelial growth and spore morphology of *Alternaria burnsii* on different media on 8th day
Media : Czapek's Dox Agar
Isolates : Ab-1=Patan; Ab-2=Banaskantha; Ab-3=Patan; Ab-4=Morbi; Ab-5=Banaskantha; Ab-6=Patan; Ab-7=Morbi; Ab-8=Ahmedabad Ab-9=Banaskantha; Ab-10=Ahmedabad; Ab-11=Morbi; Ab-12=Surendranagar; Ab-13=Ahmedabad; Ab-14=Surendranagar; Ab-15= Surendranagar

Conidial length

The result presented in Table 6 revealed significant differences between isolates and media and also interaction Among the three different media tested, maximum conidial length was recorded in Potato Dextrose Agar (60.90 µm) which was at par with Czapek's Dox agar (59.78 µm). While, the minimum conidial length was recorded in Richard's Synthetic Agar (57.91 µm).

In case of different isolates, maximum (63.76 µm) conidial length was noticed in Patan isolate (Ab-6) which was at par with Patan isolate (Ab-1) with (61.37 µm), Banaskantha isolate (Ab-2) with (61.49 µm), Ahmedabad isolate (Ab-8, Ab-10) with (62.07 µm, 60.44 µm) respectively and Surendranagar isolate (Ab-14) with (63.43 µm). The minimum conidial length was recorded in Surendranagar isolate (Ab-12) with (55.98 µm).

The interaction between media and isolates was found significant, which indicated the variation among the isolates in utilizing the media. Among them, maximum (67.30 µm) conidial length was recorded in Patan isolate (Ab-1) in Potato Dextrose Agar media which was at par with Morbi isolate (Ab-4) with (64.11 µm), Patan isolate (Ab-6) with (62.66 µm), Ahmedabad isolate (Ab-8) with (62.41 µm) amd Surendranagar isolate (Ab-14) with (61.66 µm) same media. Banaskantha isolate (Ab-2) with (62.70 µm), Patan isolate (Ab-6) with (64.93 µm), Ahmedabad isolates (Ab-8, Ab-10) with (63.43 µm, 62.20 µm) respectively and Surendranagar isolate (Ab-14) with (64.93 µm) on Richard's Synthetic Agar media. Banaskantha isolate (Ab-5) with (62.66 µm), Patan isolate (Ab-6) with (63.70 µm) and Surendranagar isolate (Ab-14) with (60.31 µm) on Czapek's Dox Agar media. The minimum conidial length was noticed in Morbi isolate (Ab-7) (50.89 µm) in Richard's Synthetic Agar media.

Conidial breadth

The result presented in Table 7 revealed significant differences between isolates and media and also interaction. Among the three different media tested, maximum conidial breadth (23.09 µm) was noticed in Potato Dextrose Agar which was at par with Czapek's Dox Agar (22.84 µm). While, the minimum conidial breadth (21.96 µm) was noticed in Richard's Synthetic Agar media.

With respect to different isolates, maximum (25.47 µm) conidial breadth was noticed in Banaskantha isolate (Ab-9) which was at par with Patan isolates (Ab-1, Ab-6) with (24.45 µm, 24.23 µm) respectively. Followed by Ahmedabad isolate (Ab-10) with (23.42 µm) which was at par with Banaskantha isolates (Ab-2, Ab-5) with (22.41 µm, 22.22 µm) respectively, Morbi isolate (Ab-7, Ab-11) with (22.22 µm, 22.88 µm) respectively, Surendranagar isolates (Ab-12, Ab-14) with (22.07 µm, 23.28 µm) respectively and Ahmedabad isolate (Ab-8) with (22.75 µm). The minimum conidial breadth was recorded in Surendranagar isolate (Ab-15) with (20.24 µm).

The interaction between media and isolates was found significant. Among them, maximum conidial breadth was recorded in Banaskantha isolate (Ab-9) (26.35 µm) in Potato Dextrose Agar media which was at par with Patan isolates (Ab-1, Ab-6) with (25.43 µm, 25.08 µm) on same media. Ahmedabad isolate (Ab-10) with (25.08 µm), Patan isolate (Ab-1) with (24.13 µm), Banaskantha isolate (Ab-9) with (24.83 µm) on Richard's Synthetic Agar media. Patan isolate (Ab-6) with (24.35 µm) and Banaskantha isolate (Ab-9) with (25.22 µm) on Czapek's Dox Agar media. The minimum conidial breadth was recorded in Surendranagar isolate (Ab-15) (18.17 µm) in Richard's Synthetic Agar media.

Conidial beak length

The result presented in Table 8 revealed significant differences between isolates, media and their interaction. Among the three different media tested, longest conidial beak was recorded in Czapek'sDox Agar (36.65 µm) which was at par with Potato Dextrose Agar (36.30µm). While, the smallest conidial beak length was recorded in Richard's Synthetic Agar (35.10µm).

In case of different isolates, longest (47.33 µm) conidial beak length was recorded in Surendranagar isolate (Ab-14). The minimum conidial beak length was recorded in Surendranagar isolate (Ab-15) with (28.73 µm).

The interaction between media and isolates was found significant. Among them, longest (48.80 µm) conidial beak length was recorded in Surendranagar isolate (Ab-14) on Potato Dextrose Agar media which was at par with Patan isolate (Ab-6) with (46.33 µm) on same media. Surendranagar isolate (Ab-14) with (48.10 µm) on Richard's Synthetic Agar media and Patan isolate (Ab-6) (45.68 µm) on Czapek's Dox Agar media. The smallest conidial beak length was recorded in Morbi

isolate (Ab-7) (26.86 µm) on Richard's Synthetic Agar media.

Conidial septation

The result presented in Table 9 revealed significant differences among the isolates with respect to transverse and longitudinal septation of conidia in different media. The maximum numbers of transverse septa were associated with the Ahmedabad isolate (Ab-8) in the range of 1to 6 in Potato Dextrose Agar media and least septation was recorded in Morbi isolate (Ab-4) in the range of 0 to 1 in Richard's Synthetic Agar media. The number of longitudinal septa was higher in the Ahmedabad isolate (Ab-8) 0-3 on Potato Dextrose Agar media. Overall, the average number of septation among the isolates varied from 1to 6 transverse and 0 to 3 longitudinal septa (Table-9).

These observations are in conformity with the findings of earlier workers Singh *et al.,* (2016); Mali *et al.,* (2017); Pipaliya and Jadeja, 2008; Shekhawat *et al.,* (2013); Paul *et al.,* (2015) who reported differences among the isolates of *A. burnsii* in terms of conidial length, breadth and number of septation. Several workers Singh and Prasada (1973); Kaul and Saxena (1988); Singh *et al.,* (2001); Gopinath (2002); Pandey *et al.,* 2005; Rahmatzai *et al.,* 2016; Kumar *et al.,* (2003); Mehta *et al.,* (2003); Jadhav *et al.,* (2011); Bassadat *et al.,* (2014) also observed diversity in cultural and morphological characters of different isolates of *Alternaria* spp.

Thus, the present findings tallied with the studies carried out by earlier workers. Morphological variation i.e. conidial size in the isolates of *A. burnsii* could be due to nutrition rather than a characteristic pathological variation. However, in the present studies glaring differences in conidial size were noticed among the isolates even when same medium was used for the growth of the isolates. It can be assumed that variation in the isolates may be inherent since isolates were collected from diverse agro climatic zones of Gujarat. Hence, these variations in the conidial size indicated the existence of variability in this pathogen.

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