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Waghunde RR
College of Agriculture, Navsari
Agricultural University,
Bharuch, Gujarat, India

Patel UT
College of Agriculture, Navsari
Agricultural University,
Bharuch, Gujarat, India

Vahunia B
College of Agriculture, Navsari
Agricultural University,
Bharuch, Gujarat, India

Morphological and cultural variability of *Alternaria Macrospora* causing leaf Blight in cotton

Waghunde RR, Patel UT and Vahunia B

Abstract

Cotton (*Gossypium hirsutum* L.) is an important cash crop which played a dominant role in the industrial and agricultural economy of India. *Alternaria* a major foliar fungal pathogen showed wide variability in morphology as well as in culture. The morphological study of four isolates of *A. macrospora* spores showed that the mean mycelial width (7.95 μ m), conidia size (70.11x21.845 μ m), length of beak (7.58 μ m), no. of transverse septa (3 to 7) and longitudinal septa (0 to 3) was maximum in Bharuch isolate and least mean mycelial width (5.09 μ m), conidia size (42.46x10.66 μ m), length of beak (5.02 μ m), no. of transverse (3 to 4) and longitudinal septa (0 to 1) was observed in Navsari isolate. In Surat and Narmada isolate the mean mycelial width (4.33 and 5.48 μ m), conidia size (46.51x12.88 and 48.51x12.90 μ m), length of beak (5.73 and 5.79 μ m), no. of transverse septa (3 to 8 and 3 to 5) and longitudinal septa (0 to 2 and 0 to 1), respectively. In case of cultural variability the colony color of Navsari, Surat, Bharuch and Narmada was grayish white, gray, grayish with sector and grayish black with sector, respectively. The excellent sporulation was observed in Bharuch isolate followed by Surat and Narmada. The Navsari isolate produced medium raised colony growth while other isolates showed raised pattern of mycelial growth.

Keywords: cotton (*Gossypium hirsutum* L.), *Alternaria Macrospora*, agricultural economy

Introduction

Cotton (*Gossypium hirsutum* L.) is a well-known fibre crop. Cotton is the most important cash crop of India. Cotton often referred as “white gold” because of its higher economical value among cultivable crop for quite a long period. India is the largest cotton growing country in the world with an area of around 105 lakh/ha with production of 351 lakh bales and productivity 568 kg/ha (Anonymous, 2017) [1]. In India, cotton is cultivated on a large scale in Maharashtra, Gujarat, Andhra Pradesh, Karnataka, Madhya Pradesh, Punjab, Rajasthan, Haryana, Tamil Nadu and Uttar Pradesh. In Gujarat, cultivated area of cotton is 24.00 lakh with production of 95 lakh bales and productivity 673 kg/ha (Anonymous, 2017) [1]. Rane and Patel (1956) [2] were the first to notice the disease from India, incited by *Alternaria* spp. The *Alternaria* species have a wide host range and they are reported to leaves, stem and fruits of the plant. The fungus *Alternaria* causes the leaf spot and blight symptoms on different plant parts.

The present study was undertaken to understand the variability in different isolates of *A. macrospora* with respect to its morphological characters like width of mycelium, size of conidia and number of transverse as well as longitudinal septa and beak length and cultural variability regarding colony color, sporulation, type of margin, radial growth and type of mycelial growth of four isolates.

Materials and Methods

The present investigation was carried out at P.G. Laboratory, Dept. of Plant Pathology, N.M.C.A, N.A.U, Navsari to study the morphological and cultural variability of *A. macrospora* causing leaf blight in cotton.

Collection and isolation of pathogen

Naturally infected diseased leaves of cotton plant showing the typical characteristics symptoms of *Alternaria* blight were collected from the Bharuch, Navsari, Surat and Narmada district of south Gujarat region during *kharif* 2017-18. Small pieces of diseased tissues along with adjoining healthy tissues cut and surface sterilized by dipping in (0.1%) NaOCl₂ solution for one minute followed by three successive washings with distilled sterile water. The sterilized pieces were then transferred aseptically under laminar air flow system (Klenzoid Contamination Control Ltd.) In sterilized Petri plates containing 20 ml. Potato Dextrose Agar (PDA) medium. The Petri plates incubated at 27±1 °C in BOD incubator for seven days. The

Correspondence

Waghunde RR
College of Agriculture, Navsari
Agricultural University,
Bharuch, Gujarat, India

well-isolated single spore was marked and then transferred to PDA slants separately under aseptic conditions for purification and further investigation.

Morphological variability

The seven days old isolates of *Alternaria* culture grown on PDA plates were used to study the morphological characters like width of mycelium, size of conidia and number of transverse as well as longitudinal septa and beak length. The size of conidia and mycelium width measured under light microscope at 40X using software Scope photo 3.0. Twenty observations were recorded for conidial measurement, beak length, number of transverse as well as longitudinal septa and for mycelium measurement and mean values were calculated.

Cultural variability

The cultural characters of four isolates of *Alternaria* were recorded from culture grown on PDA. Twenty ml of sterilized PDA was poured in Petri plates. Five mm disc was cut through sterilize cork borer from the seven days old fungal culture grown in Petri plates. One disc was placed in the center of each plate and incubated at 27 ± 1 °C for seven days. The differences between observations regarding colony color, sporulation, type of margin, radial growth and type of mycelial growth of four isolates were recorded 7 days after inoculation.

Results and Discussion

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

Morphological variability

To study the morphological variability in *A. macrospora*, the

diseased samples were collected from cotton field of Navsari, Surat, Bharuch and Narmada districts of South Gujarat and subjected for tissue isolation in laboratory. Seven days old isolates of *A. macrospora* grown on PDA was used to study morphological characters. The results obtained are presented in Table 1.

The mean mycelial width (7.95 μ m), conidia size (70.11x21.845 μ m), length of beak (7.58 μ m), no. of transverse septa (3 to 7) and longitudinal septa (0 to 3) was maximum in Bharuch isolate and least mean mycelial width (5.09 μ m), conidia size (42.46x10.66 μ m), length of beak (5.02 μ m), no. of transverse septa (3 to 4) and longitudinal septa (0 to 1) observed in Navsari isolate. In Surat isolate mean mycelial width (46.514 μ m), conidia size (46.51x12.88 μ m), length of beak (5.73 μ m), no. of transverse septa (3 to 8) and longitudinal septa (0 to 2) and mean mycelial width (5.48 μ m), conidia size (48.51x12.90 μ m), length of beak (5.79 μ m), no. of transverse septa (3 to 5) and longitudinal septa (0 to 1) observed in Narmada isolate.

Result similar to the present investigation was reported by Kaur and Aggarwal (2015) that size of conidia ranged from 25-57.5x12.5-25 μ m with 1 to 6 transverse and 0-2 longitudinal septa with beak length ranged between 5-15x5-7.5 μ m in pathogen *A. macrospora* infecting cotton. Venkatesh and Darvin (2016) found similar result to the present study on morphological characteristics of *A. macrospora* and recorded conidia size ranged between 43.4 x 13.3 μ m with 1 to 8 transverse septa. Sangeetha *et al.* (2016) [5] maximum conidial length with 90-180 (μ m). Conidia recorded 1-5 vertical and 4-9 horizontal septa and beak length was equal or twice of the length of conidia.

Table 1: Morphological variability of *A. macrospora*

Name of isolate	Mycelial width (μ m)	Conidia size		Beak length (μ m)	No. of transverse septa	No. of longitudinal septa
		Length (μ m)	Width (μ m)			
Navsari	5.09	42.46	10.66	5.02	3-4	0-1
Surat	4.33	46.51	12.88	5.73	3-8	0-2
Bharuch	7.95	70.11	21.84	7.58	3-7	0-3
Narmada	5.48	48.51	12.90	5.79	3-5	0-1

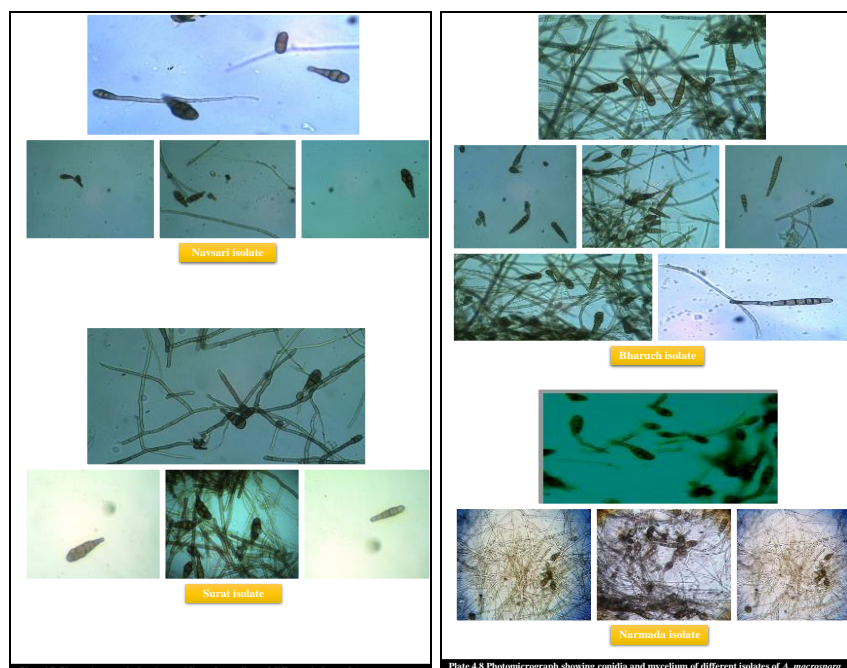


Plate 1 Photomicrograph showing conidia and mycelium of different isolates of *A. macrospora*

Cultural variability

The four isolates of *A. macrospora* infecting to cotton were grown on PDA to study the cultural characteristics. The 5 mm disc of *A. macrospora* culture was placed on the PDA media and incubated at 27 ± 2 °C for 7 days. The colony characters and radial growth were recorded after 7 days of incubation. The results are presented in Table 2 and Plate 2.

It was clear from the present results that colonies varied in respect to their color (gray to grayish white; grayish black with sector), sporulation {Excellent sporulation >30 spores/microscopic field (40X), good sporulation 21-30 spores/microscopic field (40X) and moderate sporulation 11-20 spores/microscopic field (40X), poor sporulation 6-8 spores/microscopic field (40X)}, type of margin irregular to smooth, radial growth ranged between 71 to 85 mm with medium raised to raised mycelial growth.

The colony color of Navsari, Surat, Bharuch and Narmada isolates were grayish white, gray, grayish with sector and grayish black with sector, respectively.

The excellent sporulation was observed in Bharuch isolate

followed by Surat and Narmada. The poor sporulation was recorded in Navsari isolate. The highest radial growth (85 mm) was recorded in Narmada isolate followed by Surat (84 mm), Bharuch (78mm) and lowest in Navsari isolate (71 mm). The Navsari colony showed medium raised colony growth while other isolates showed raised pattern of mycelial growth.

Similar result to the present investigation reported by Sangeetha and Ashtaputre (2015) [6]. They observed that colony color of *A. macrospora* varied from gray to black with white to black colony margin with irregular or smooth and mycelial growth with sectoring. Perane *et al.* (2015) [7] recorded similar result and indicated that growth characteristics of *A. macrospora* exhibited 72 mm colony diameter after seven days of incubation. Hosagoudar (2012) [8] studied on cultural variability of seventeen isolates of *Alternaria* spp. and recorded colony diameter upto 60-80 mm, colony color gray to blackish, smooth irregular margin with flat to medium raised mycelial growth.

Table 2: Cultural variability of *A. macrospora*

Name of Isolate	Cultural Variability				
	Colony color	Sporulation	Type of margin	Radial growth (mm)	Type of mycelial growth
Navsari	Grayish white	++	Irregular	71	Medium raised
Surat	Gray	+++	Smooth	84	Raised
Bharuch	Grayish with sector	++++	Smooth	78	Raised
Narmada	Grayish black with sector	+++	Smooth	85	Raised

++++ Excellent sporulation, +++ Good sporulation, ++ Moderate sporulation, + Poor sporulation



Plate 2: Cultural characteristics of *A. macrospora*

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