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The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2023; 12(1): 363-365

www.thepharmajournal.com
Received: 04-12-2022
Accepted: 13-01-2023

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Symptomatology of leaf spot disease of cotton caused by *Curvularia lunata* (Wakker) Boedijn

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DOI: https://doi.org/10.22271/tpi.2023.v12.i2e.18444

Abstract
The Curvularia leaf spot is the most important and destructive disease of cotton. The research was conducted to prove *Curvularia lunata* as an Incitant of Curvularia leaf spot of cotton. Infected leaves with typical symptoms were subjected to isolation and purification. As a result, brown to brownish-black fungal colonies were found. The morphological study was carried out by using microscope, the spores of fungus found slightly curved or straight; mycelium was septate and brown to black in colour, and conidia was found 25-27 × 8-10 µm in size. The pathogenic nature of incitant was proved and typical symptoms such as small, brown to black in coloured spots were observed. Further, re-isolated pure culture resembled with original culture on the basis of their cultural characteristics. Hence it is identified as *Curvularia lunata* (Wakker) Boedijn - an Incitant of Curvularia leaf spot of cotton.

Keywords: Curvularia leaf spot, *Curvularia lunata*, Cotton

Introduction
Cotton is the world's most widely grown fibre crop, which belongs to the genus *Gossypium* in the family of Malvaceae. It is oldest among the commercial crops of the world and also known as "The King of fibers" and "White Gold". It has delicate, white, soft and fluffy fibre that is made of about 87 to 90% of cellulose (Proto *et al*., 2000) [1]. It is grown in 75 countries across the world out of which United States, China and India contribute 80% of the total yield in the world. India is the second largest cotton producer in the world (22% of the global cotton production) with largest cultivation area (30% of global cotton area) among the major cotton growing countries. In India, Gujarat, Maharashtra, Haryana, Punjab, Rajasthan and Madhya Pradesh are the major cotton growing states.
The cotton crop is prone to several fungal, bacterial and viral diseases; among these infections the fungal infections are predominant on cotton and cause the major yield loss. In India, the various fungal diseases reported on cotton includes Fusarium wilt, Verticillium wilt, Grey mildew, Alternaria leaf spot, Myrothecium leaf spot, Root rot, Anthracnose and Curvularia leaf spot. Among them, Curvularia leaf spot is minor disease of cotton gaining such importance. *Cochliobolus lunatus* Nelson and Haasis (*Curvularia lunata*) has been known to cause a leaf spot of cotton in India (Sharma and Chauhan, 1985) [2]. The present study was carried out on isolation, pathogenicity and symptomatology of Curvularia leaf spot pathogen.

Material and Methods
Collection of Disease Samples
Cotton plant showing typical diseased symptoms were collected from different regions and farmer fields of South Gujarat. The infected samples were brought to the laboratory and subjected to microscopic examination of spores to identify on the basis of morphological characters.

Isolation of Pathogen
Collected diseased samples such leaves were subjected to isolation of causal agent with standard isolation technique (Yadav, 2013) [3]. The isolation was done by following the standard tissue isolation method. The diseased samples were chopped into small pieces and then surface sterilized with 0.5% sodium hypochlorite for 1 minute, after that rinsed in to distilled water for three times then placed on to blotter paper for removal of access water. Those pieces were placed on to Petri plate containing Potato Dextrose Agar (PDA) media and kept into incubator at 25±2 °C for 5-6 days. After incubation period is over small colonies of pathogen was appeared, which were picked up and kept in to PDA media.

Purification, Maintenance and Identification of Pathogen
The isolated culture was further purified by hyphal tip method under aseptic condition. When the mycelial growth was observed in Petri plates, advancing hyphal tips growing out of tissue segments cut off with sterilized inoculation needle and transferred to PDA slants for further growth.

Pathogenicity Test of Pathogen
The pathogenicity test was carried out by using standard spore suspension spray method (Yadav, 2013) [3] and find out whether the isolated fungus is capable to producing typical symptoms of disease under artificial inoculation condition.
The isolated pathogen from diseased sample was multiplied in PDA plates. The spore suspension was prepared from the sporulating seven days old culture of pathogen by using sterilized distilled water. Total thirty cotton plants were raised in steam sterilized potting mixture with soil, sand and FYM in 2:1:1 ratio. Two month old cotton plants were pre-incubated for 24 hours in humid tent made up of plastic sheets in which humidity was maintained between 60 to 80 per cent before the inoculation. Among them, fifteen plants were slightly injured by a sterilized pin and spore suspension wassprayed on to the surface of leaves and stems with low pressure sprayer. Similarly, total fifteen plants were injured and sprayed with sterile water which was constituted as control. All the plants were kept in the plastic tent in which high humidity was maintained. Observations were made for the development of symptoms of Curvularia leaf spot.
The Percent Disease Incidence (PDI) was calculated by using the following formula given by Wheeler (1969),

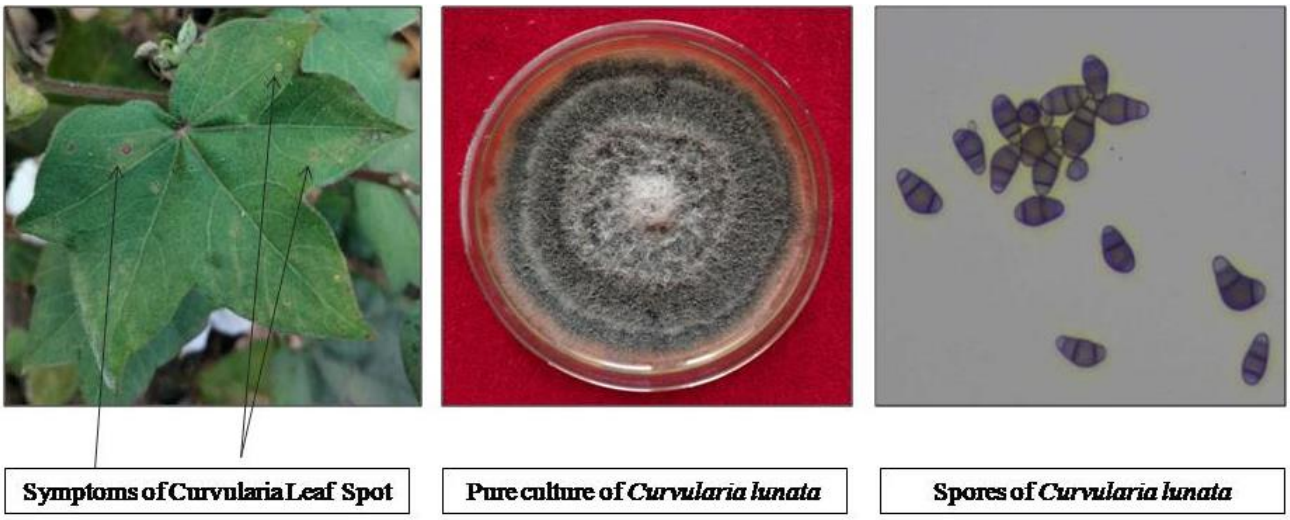
$$PDI = \frac{\text{Total No. of infected leaves}}{\text{Total number of leaves}} \times 100$$

After the development of the typical symptoms of the disease, the pathogen was reisolated and compared with original culture to prove the pathogenicity.

Morphological characters
The morphological characteristics such as a mycelia characters, septation and size of conidia was observed by using microscope. The hypha and spores of pathogen were observed under microscopes to identify.

Results and Discussion
Collection of disease samples: The infected plant parts such as leaves showing typical symptoms were collected from nearby region of Bharuch district and brought to the laboratory. The symptoms appears initially as small circular brown to brownish black spot surrounding with yellow hallow, later it became dark yellow to brown hallow surrounding to brownish black spots.

Isolation and identification of pathogen
Collected diseased samples such leaves were subjected to isolation of causal agent with standard isolation technique (Yadav, 2013) [3]. Isolation of pathogen was done by following the standard tissue isolation method as described in material and methods. After incubation period is over small colonies of pathogen was appeared, which were picked up and kept in to PDA media for purification of fungal colonies of pathogen.
As a result of isolation, brown to brownish-black fungal colonies with grayish center were found from the infected tissue. The pure cultures of the isolates were maintained by repeated sub culturing at interval of fifteen days for further studies.



Pathogenicity test
The pathogenicity test was carried out to know the ability of pathogen to cause the disease by using standard spore suspension spray method as described in material and methods. Where, total fifteen plants were inoculated with spray inoculation technique, among them eleven plants were found infected with 73.33 Percent disease incidence. The pathogenic nature of incitant was proved and typical symptoms such as small, erratic, brown to brownish-black in coloured spots were observed on the leaves. Where, total fifteen untreated plants (control) having no symptoms.
The pathogen was reisolated and compared with original culture to prove the pathogenicity. On the basis of cultural characteristics, re-isolated pure culture resembled the original.

Table 1: Pathogenicity test of *Curvularia lunata* causing Curvularia leaf spot of cotton

Sr. No.	Inoculation technique	Total No. of plants	No. of plants infected	Per cent disease incidence	Symptoms
1.	Spray inoculation technique	15	11	73.33	Small, erratic, brown to brownish-black in Coloured spots were observed on the leaves
2.	Control (Without inoculation)	15	00	00	No symptoms observed

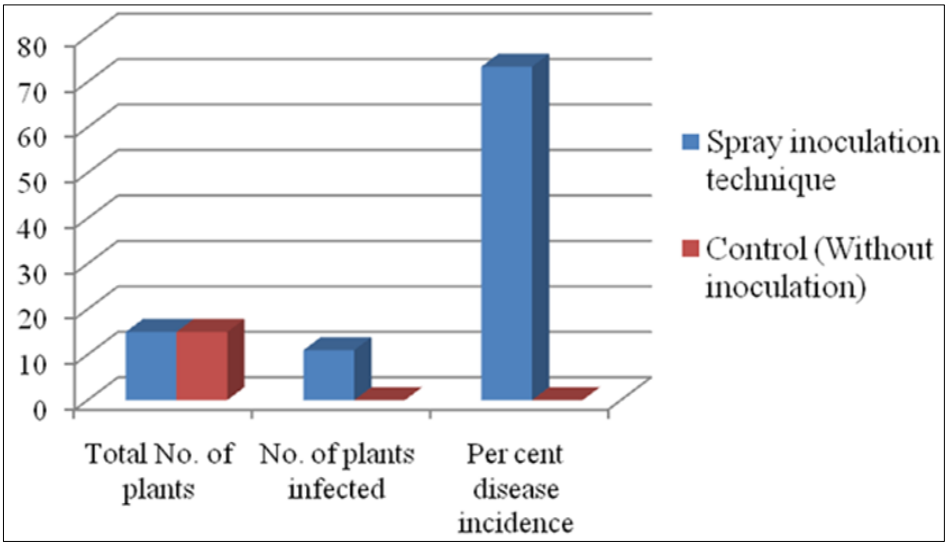


Fig 1: Pathogenicity test of *C. lunata* causing leaf spot of cotton

Morphological characters of pathogen

In morphological study, the spores of fungus showed two or three septa, slightly curved or straight morphology; mycelium was septate and brown to black in colour, and size of conidia ranged from 25-27×6-8 µm.

References

1. Proto M, Supino S, Malandrino O. Cotton: A flow cycle to exploit. Industrial Crops and Products. 2000;11:173-178.
2. Sharma BK, Chauhan MS. Studies on the chemical control of foliar diseases of cotton in Haryana state. Agriculture Science Digests. 1985;5:153-157.
3. Yadav ML. Investigations on the epidemiology and management of *Curvularia lunata* (Wakker) boedijn, causing leaf spot of cotton (*Gossypium hirsutum* L.) M.Sc. Thesis, Maharana Pratap University of agriculture and technology, Udaipur, India; c2013.