MYCOFLORA OF COTTON PLANT (GOSSYPIUM HIRSUTUM L.) - WITH THREE NEW RECORDS OF DEUTEROMYCETES FROM BANGLADESH

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ABSTRACT

Fungal diseases of cotton plant (*Gossypium hirsutum* L.) were studied during July, 2011 to June, 2012. Eleven cotton varieties were selected for the experiment. *Alternaria alternata* (Fr.) Keissler, *Aspergillus niger* van Tiegh, *Colletotrichum gossypii* Sowthworth, *Colletotrichum dematium* (Pers.) Grove, *Curvularia clavata* Jain, *Fusarium* sp., *Gleotrichum* sp., *Hyalopycnis* sp., *Pestalotiopsis guepinii* (Desm.) Stay, and a sterile fungus were isolated from infected leaves of cotton varieties CB6 and CB11. *Colletotrichum gossypii* was isolated from both the varieties and *C. dematium* was exclusively isolated from CB6. Varieties CB6 and CB11 were more susceptible than other varieties. Cotton varieties CB1, CB2, and CB3 showed negligible infection on leaves and bolls. *Colletotrichum gossypii* and *Hyalopycnis* sp. are new record for Bangladesh. Moreover, association of *C. dematium*, *Gleotrichum* sp., *P. guepinii* and *Hyalopycnis* sp. with cotton is being reported first time. The three fungi *Colletotrichum. gossypii*, *C. dematium* and *Hyalopycnis* sp. are described and illustrated in this account as new Deuteromycetous records for Bangladesh.

Key words: Mycoflora, Cotton plant, Gossypium hirsutum, Deuteromycetes

INTRODUCTION

Cotton plant (*Gosypium* spp.) belongs to Malvaceae. The plant is affected by fungal disease along with bacteria, virus, nematodes and pests. So far 20 types of symptoms and 48 species of fungi have been reported on cotton plant from various cotton growing areas of the world (Wikipedia 2015). From Bangladesh seven types of symptoms were recorded on four varieties of cotton plant (Lutfunnessa and Shamsi 2011). The symptoms were Anthracnose, Alternaria leaf spot, Boll rot, Cercospora leaf spot, Rust, Sclerotium rot and Wilting. About 30 species of fungi belonging to 21 genera were found associated with four varieties of cotton plant. *Fusarium* spp. and *Colletotrichum* spp. were frequently associated with all the four cotton varieties examined. *Botryodiplodia* sp. was associated with Hill Cotton 1 (HC-1) and Hill Cotton 2 (HC-2). *Sclerotium rolfsii* was found associated with CB-3 cotton variety (Shamsi *et al.* 2008).

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Cotton diseases are less prevalent in Bangladesh compared to other cotton growing countries of the world, but with the changed environment and climate, presently cotton plants becoming susceptible and being attacked by various fungi and different types of symptom are expressed. In Bangladesh information regarding the cotton diseases, association of fungi and the causal agent is inadequate. Considering the importance of the plant and destructive attack of the fungi against its host, the present study was undertaken to investigate the fungal diseases of different cotton varieties, characterization and identification of the fungi associated with the diseased plant parts and to detect pathogenic potentialities of the fungi associated with the cotton varieties under Bangladesh conditions.

MATERIALS AND METHODS

Seeds of 11 varieties of cotton were collected from "Cotton Development Board" Bangladesh. Seedings were raised from the seeds in experimental plots of Botanic Garden, Curzon Hall Campus, University of Dhaka. Samples were collected during the period July, 2011 to June, 2012. At fruiting stage, the cotton plants that were grown in the experimental plots were severely infected by fungi showing anthracnose and blight on leaves (Fig. 1A, B). Fruits at maturity showed boll rot and plants gradually dried.

Thirty samples were examined from infected leaves and bolls of *G. hirsutum*. Fungi were isolated from infected samples on PDA (Potato Dextrose Agar) medium following "Tissue Planting" method. From a particular sample 30 inocula, each measuring 2 square mm were cut and kept in a sterile Petri plate. The inocula were washed in sterile water and then surface sterilized by dipping in 10% Chlorox for 3 - 5 minutes and transferred into a sterile Petri plate containing sterile blotting paper to remove the surface water. Ten Petri plates with PDA medium were inoculated with three inocula per plate. Inoculated Petri plates were incubated for 5 - 7 days at $25 \pm 1^{\circ}$ C. Fungi growing out of the inocula were transferred to separate plates and slants for further studies and storage. Percentage of association of the fungi was also recorded.

Microscopic details of the specimens were studied following standard techniques (Khan and Shamsi 1983, Shamsi and Sultana 2008). Identification of the isolates was determined following the standard literatures (Barnett and Hunter 2000, Ellis 1971, 1976, Ellis and Ellis 1997 and Sutton 1980).

All the isolated fungi were tested for their pathogenic potentiality following "Detached leaf technique" (Azad and Shamsi 2011).



Fig. 1. Anthracnose of cotton: A- B. Infected leaf; C. Colony of *Colletotrichum dematium*; D. Colony of *Colletotrichum gossypii*; E. Colony of *Hyalopicnis* sp.; F. Acervuli, setae and conidia of *C. dematium*; G. Acervuli and conidia of *C. gossypii*; H. Setae; I- J. Germinating conidia; K. Conidiophores and conidia of *Hyalopicnis* sp. (Bar = 50 µm).

The specimens were preserved in the Herbarium, Mycology and Plate Pathology Section, Department of Botany, University of Dhaka, Bangladesh.

RESULTS AND DISCUSSION

Three types of disease symptoms were recorded on 11 varieties of cotton plant. The symptoms were anthracnose and blight on leaves and boll rot, anthracnose and blight on fruits.

In total 9 species of fungi and a sterile mycelia colony were isolated from the different cotton varieties infected with anthracnose and blight symptoms. *Alternaria alternata* (Fr.) Keissler, *Aspergillus niger* van Tiegh, *Colletotrichum gossypii* Sowthworth, *C. dematium* (Pers.) Grove, *Curvularia clavata* Jain, *Fusarium* sp., *Gleotrichum* sp., *Hyalopycnis* sp., *Pestalotiopsis guepinii* (Desm.) Stay, and a sterile fungus were isolated from infected leaves of cotton varieties CB6 and CB11. *Colletotrichum gossypii* was isolated from both the varieties and *C. dematium* was exclusively isolated from CB6. Varieties CB6 and CB11 were more susceptible than other varieties. Cotton varieties CB6 and CB11 were more susceptible than other varieties.

The frequency percentage of fungi association with two cotton varieties are presented in Table 1. The percentage of association of fungi varied in two cotton varieties. Frequency percentage of association of *A. alternata* was highest 59 in CB6 followed by *C. gossypii* 50 in CB11 and 41 in CB6. The percentage of association of *C. dematium* was 30 in CB6. Frequency percentage of *A. niger* was 35.16 in cotton variety CB6. Frequency percentage of *C. clavata* was 25 in CB6. Frequency of percentage of association of *Fusarium* sp. was 26 in variety CB6. Frequency of percentage of *Hyalopycnis* sp was 8.33 in CB6. and *P. guepinii* was 8.8 in CB11. Lowest frequency percentage of association of fungi was recorded 5 in *Gleotrichum* sp. isolated from CB6. Frequency percentage of association of sterile fungus was 33 in CB6 and 33.33 in CB11.

Name of fungi	Cotton variety CB6	Cotton variety CB11
Alternaria alternata	59	41
Aspergillus niger	35.16	05
Colletotrichum gossupii	41	50
C. dematium	30	-
Curvulara clavata	25	-
<i>Fusarium</i> sp.	26	-
Geotrichium sp.	5	-
Hyalopycnis sp.	8.33	-
Pestalotiopsis guepinii	8.8	-
Sterile fungi	33	33.33

Table 1. Frequency percentage of associated fungi of two cotton varieties CB6 and CB11.

- = No fungal growth.

Pathogenicity test following detached leaf technique revealed that *A. alternata*, *C. gossypii*, *C. dematium*, *Hyalopycnis* sp. and *P. guepinii* were pathogenic to cotton plant.

Description of the taxa

Colletotrichum dematium (Pers.) Grove

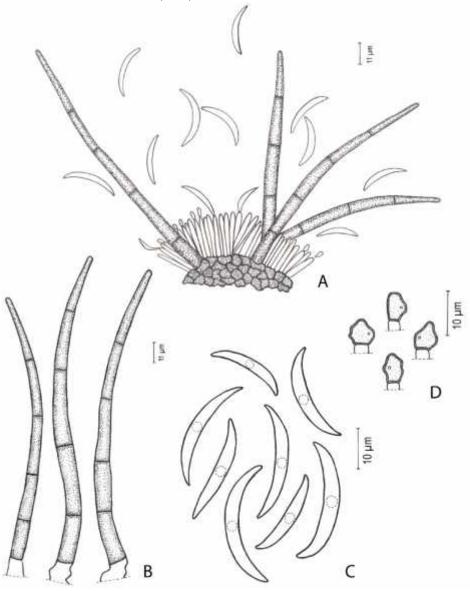


Fig. 2. Colletotrichum dematium on cotton: A. Acervulus; B. Setae; C. Conidia; D. Appressoria.

Colonies dark, cottony, with black shinning aervuli. Hyphae brown, septate, branched superficial. Setae dark brown. 3 - 4 septate, 62 - 72 μ m × 3.5 - 4.0 μ m. Conidiophores pale brown, 14 - 22 × 1.8 - 2.7 μ m. Conidia solitary, curved, hyaline, 20 - 34 × 3 - 4 μ m. Appressorium present, brown, small irregular, 4.8 - 6.0 × 4.0 - 48 μ m.

Specimens examined: Isolated from infected leaves of cotton varieties CB6, Botanic garden, Dhaka University, Dhaka.

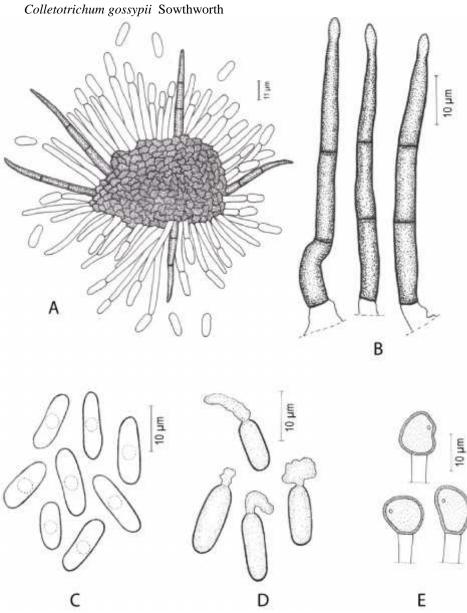
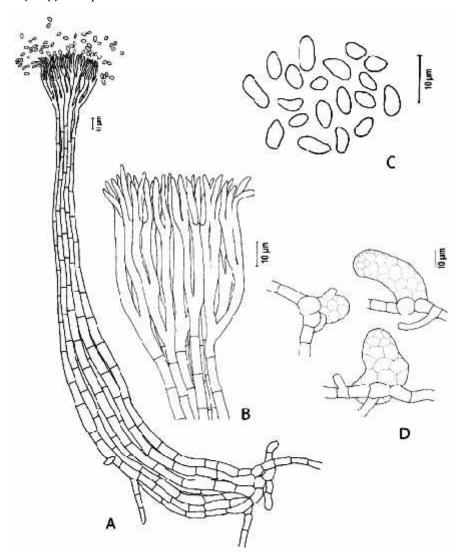


Fig. 3. Colletotrichum gossypii on cotton: A. Acervulus; B. Setae; C. Conidia; D. Germinating conidia; E. Appressoria.

Colonies grayish, cottony with pinkish orange acervuli. Hyphae brown, partly superficial, septate. acervuli are 100 to 153 µm in diameter. Setae olive or dark brown, straight, curved, flexuous, or rarely branched, 46 - 65 μ m × 4.0 - 5.8 μ m. Occasionally spores are produced upon the ends of the setae. Conidiophores pale brown, 12 - 39 × 3.4 - 4.5 μ m. Conidia solitary, straight, hyaline, 10.5 - 16.8 μ m × 4.2 - 5.2 μ m. The conidia have only one nucleus. Appressorium present, brown, subcircular or oblong, 12 - 12.8 × 8 - 0.4 μ m.

Specimen examined: Isolated from infected leaves of cotton varieties CB11, Botanic garden, Dhaka University, Dhaka.



Hyalopycnis sp. Hone.

Fig. 4. *Hyalopicnis* sp. on cotton (*Gossypium hirsutum*). A. Pycnidium, B. Conidiophores, C. Conidia and D. Initiation of pycnidia.

Colonies grayish black, pycnidia superficial, light-colored (shiny white) membranous, with a globos base and a long, sub cylindrical neck, fimbriate at the apex, wall of pycnidium and neck composed of parallel hyphae fused laterally; conidiophores long, simple or branched 14 - 23.2 μ m × 2.2 - 3 μ m. Conidia 1- celled, hyaline, cylindrical or ovoid 3.4 - 7.6 μ m × 2.4 - 3.2 μ m, on other fungi.

Specimens examined: Isolated from infected leaves of cotton varieties CB6, Botanic garden, Dhaka University, Dhaka.

Colletotrichum gossypii and C. dematium were isolated from leaves and bolls of cotton showing anthracnose symptoms. *Hyalopinis* sp. was exclusively isolated from leaves showing anthracnose symptoms. *Rhizopus oryzae* was also isolated from cotton bolls (Shamsi and Naher 2014).

Lutfunnessa and Shamsi (2011) reported seven types of symptom on four varieties of cotton plant. The symptoms were Anthracnose, Alternaria leaf spot, Boll rot, Cercospora leaf spot, Rust, Sclerotium rot and Wilting. They reported 30 species of fungi belonging to 21 genera associated with four varieties of cotton plant. Shamsi *et al.* (2008) also reported *Sclerotium rolfsii* associated with cotton variety CB-3.

Present study differed from the results of Lutfunnessa and Shamsi (2011). In this experiment 11 cotton varieties were screened for fungal associates. *Colletotrichum gossypii* and *Hyalopycnis* sp. are new record for Bangladesh. Association of *C. dematium, Gleotrichum* sp., *P. guepinii* and *Hyalopycnis* sp. with cotton is being reported for the first time.

Atkinson (1891) first time reported Anthracnose of cotton caused by *Colletotrichum gossypii* in USA. He examined the field symptoms of the disease beginning in 1890. At the same time Sowthworth was working with same problem of cotton plants. Her work focused on boll infections and morphological descriptions of the fungus (Southworth 1890, Ristaino and Peterson 2002).

CONCLUSION

From Bangladesh this is the first report of *C. gossypii*, the causal agent of antracnose of cotton. Moreover, the association of *C. dematium, Gleotrichum* sp., *P. guepinii* and *Hyalopycnis* sp. with cotton for the first time is being reported. All the fungi were found pathogenic to cotton plant. Cotton varieties CB6 and CB11 are more susceptible than other varieties.

The results of present research focused on new addition regarding the diseases of cotton and its causal agent. This finding will be also helpful for designing of proper management of cotton diseases.

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