

## MYCOFLORA ASSOCIATED WITH INFECTED PLANT PARTS OF *TAGETES ERECTA* L. AND *TAGETES PATULA* L.

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### Abstract

Twenty species of fungi associated with blight symptom of different plant parts of *Tagetes erecta* L. and *T. patula* L. have been described. The associated fungi were *Alternaria alternata* (Fr.) Keissler, *Aspergillus flavus* Link, *A. fumigatus* Fresenius, *A. niger* van Tieghem, *Bipolaris australiensis* (M.B. Ellis) Tsuda & Ueyama, *Chaetomium globosum* Kunze, *Cladosporium elatum* (Harz) Nannf., *Corynespora cambrensis* M. B. Ellis, *Curvularia brachyspora* Boedijn, *C. fallax* Boedijn, *C. lunata* (Wakker) Boedijn, *C. stapeliae* (du Plessis) Hughes & du Plessis, *Epicoccum purpurascens* Ehrenb. ex Schlecht, *Fusarium semitectum* Berk. & Rav., *Monochaetia ceratoniae* (Sousa da Camera) Sutton, *Nigrospora panici* Zimm., *Penicillium italicum* Wehmer, *Rhizopus stolonifer* (Ehrenb.: Fr.) Vuill, *Trichoderma viride* Pers. and *Trichothecium roseum* Link.

### Introduction

*Tagetes erecta* L. and *T. patula* (commonly known as Marigold) belonging to Asteraceae (Compositae) is native to North and South America, but some species now become naturalized around the world (Ahmed *et al.* 2008). No annual flower is more cheerful and easier to grow than marigolds. The floret of *Tagetes* spp. are rich in the orange, yellow carotenoid lutein and are used as a food colour. The essential oil of the flower contains antioxidants. Seeds of *T. erecta* are a natural pesticide. Leaves are used as blood clotting agents in Ayurvedic treatment. Plants have antifungal properties also and used against fever dysenteries, indigestions, ulcers and eczemas (Ghani 2003 and Yusuf *et al.* 2009). It is most effective against the nematode species *Pratylenchus penetrans* (Olabiyi and Oyedunmade 2000). Plants have also mosquitocidal potentiality (Rajasekaran *et al.* 2004). Marigold is now a profitable cultivated crop to the farmers, but socio-economic data and information of this flower are very little in Bangladesh. In Jashore and Jhenaidah district most of the farmers cultivated marigold. The gross margin and net return owing to marigold cultivation was Tk. 1,62,186 and 1,17,812 per hectare, respectively. The net return was 80% higher than lentil, 85% higher than mustard and 6% lower than potato cultivation (Hoque *et al.* 2012). Diseases were major constrain for marigold cultivation in Bangladesh, due to rapid expansion of commercial marigold cultivation many diseases appear on the plants. Leaf spot and blight are two common diseases of *T. erecta* and *T. patula* (Mukerji and Bhasin 1986). Hossain *et al.* (2010) reported powdery mildew and Sultana and Shamsi (2012) reported gray mold of *T. erecta*. Aktar and Shamsi (2014, 2015, 2016) reported blight disease of *T. erecta* and *T. patula*.

Present investigation was undertaken to find out the mycoflora associated with fungal blight symptoms of *T. erecta* and *T. patula*.

### Materials and Methods

Samples with blight symptom were collected from Bangladesh Agriculture Research Institute (BARI), Gazipur, Chottogram, Comilla, Dhaka city, Khulna, Pabna, Rajshahi, Sylhet and Rangpur during the period of 2009 to 2014. Severe blight symptom on leaves, bud and flowers of

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two species of *Tagetes* were recorded. Fungi associated with infected leaves, buds and flowers of *T. erecta* and *T. patula* were isolated following Tissue Planting and Blotter method (CAB 1968). Experiment was conducted in the Laboratory of Mycology and Plant Pathology, Department of Botany, University of Dhaka. Microscopic details of the associated fungi with *Tagetes* spp. were studied following standard techniques (Shamsi and Sultana 2008).

Microphotographs were taken with Nycon D 5000 digital camera. Identification of the isolates was determined by following the reports (Thom and Raper 1945, Raper *et al.* 1949, Booth 1971, Ellis 1976, Sutton 1980, Barnett and Hunter 2000). All the specimens were preserved in the Herbarium, Mycology and Plant Pathology section, Department of Botany, University of Dhaka, Bangladesh.

### Results and Discussion

A total of 20 species of fungi associated with infected plant parts of *T. erecta* and *T. patula* were *Alternaria alternata* (Fr.) Keissler, *Aspergillus flavus* Link, *A. fumigatus* Fresenius, *A. niger* van Tieghem, *Bipolaris australiensis* (M.B. Ellis) Tsuda & Ueyama, *Chaetomium globosum* Kunze, *Cladosporium elatum* (Harz) Nannf., *Corynespora cambrensis* M. B. Elli, *Curvularia brachyspora* Boedijn, *C. fallax* Boedijn, *C. lunata* (Wakker) Boedijn, *C. stapeliae* (du Plessis) Hughes & du Plessis, *Epicoccum purpurascens* Ehrenb. ex Schlecht., *Fusarium semitectum* Berk. & Rav., *Monochaetia ceratoniae* (Sousa da Camera) Sutton, *Nigrospora panici* Zimm., *Penicillium italicum* Wehmer, *Rhizopus stolonifer* (Ehrenb.: Fr.) Vuill, *Trichoderma viride* Pers. and *Trichothecium roseum* Link.

### Taxonomic treatment of fungal taxa

*Alternaria alternata* (Fr.) Keissler, 1912, Beih. Bot. Zbl. **29**: 434. (Fig.1A)

Colonies greenish black velvety. Reverse blackish. Hyphae pale to mid brown, smooth septate, 1-5  $\mu\text{m}$  in diameter. Conidiophores flexuous, septate, pale to mid brown, up to 85  $\mu\text{m}$  long, but usually much shorter (14-60)  $\mu\text{m}$  and 4-7(9)  $\mu\text{m}$  in diameter. Conidia straight, muriform, oblong, rounded at the base, pale to mid brown, 2-7 (mostly 5) septate, 20-55 (76)  $\times$  8-18 (13)  $\mu\text{m}$ . Beak 2-5  $\mu\text{m}$  thick.

*Specimen examined*: Isolates from infected leaves of *Tagetes erecta* L., Botanical Garden, University of Dhaka, Dhaka, 10 February 2009, M. Aktar 1.

*Aspergillus flavus* Link, in Obs. p.16. 1809; also, in Sp. Plant. **6**: 66. 1824. (Fig. 1B)

Colonies effuse, greenish. Reverse greenish. *Mycelium* well-developed, septate, profusely branched and hyaline. Conidiophores long, greenish brown variable mostly 440 -560  $\times$  4-8  $\mu\text{m}$ . Vesicles globose or subglobose, thick walled, commonly 20 - 30  $\mu\text{m}$ , usually fertile on the upper half only. Sterigmata in one series, 5-10  $\times$  2-3  $\mu\text{m}$ . Conidia dark green in mass, one celled globose, spinose, catenulate, mostly 2.5-3  $\mu\text{m}$  in diameter.

*Specimen examined*: Isolates from the infected leaves of *Tagetes patula* L., Botanical Garden, University of Dhaka, Dhaka, 14 February 2010, M. Aktar 36.

*Aspergillus fumigatus* Fresenius, in Beitragezur Mykologie, p.81, pl.10, Figs1-11. Frankfurt, 1850-53. (Fig. 1C)

Colonies of the fungus on PDA plates were grey-green, cottony and reverse side was off white. Conidiophores are aseptate, smooth, greenish up to 500  $\mu\text{m}$  in length and 2-8  $\mu\text{m}$  width. Vesicles flask shaped, 20 - 30  $\mu\text{m}$ , typically fertile over the upper half. Sterigmata in one series are crowded. Conidia are grey-green, one celled, globose, echinulate, catenate, 2-3  $\mu\text{m}$  diameter.

*Specimen examined:* Isolated from infected petals of *Tagetes patula* L., Botanical Garden, University of Dhaka, 7 April 2009, M. Aktar 25.

*Aspergillus nigervan* Tieghem, Ann. Sci. Nat. Bot., Ser. **58**:240. (1867). (Fig. 1D)

Colonies effuse, black. Reverse brownish. Mycelium well-developed, septate, profusely branched and brownish. Conidiophores brown 200 - 459 × 8 - 11 μm. Vesicles globose or subglobose, thick walled, commonly 22 - 54 μm. Sterigmata typically in two series, closely packed covering the vesicle usually 20 - 30 × 7 - 8 μm. Conidia dark brown, one celled globose, spinose, cattenulate. 2 - 4 (5.5) μm in diameter.

*Specimen examined:* Isolates from infected leaves of *Tagetes erecta* L., Botanical Garden, University of Dhaka, Dhaka, 10 February 2009, M. Aktar 3.

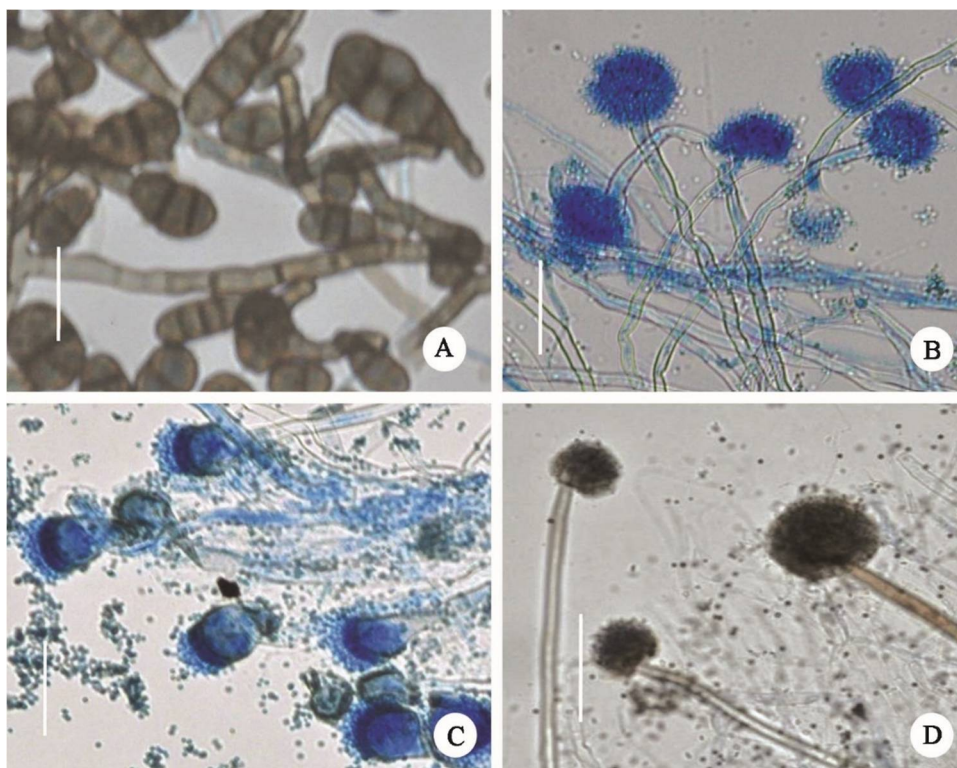


Fig.1. Conidiophores and conidia of *Alternaria alternata* (A), *Aspergillus flavus* (B), *A. fumigatus* (C) and *A. niger* (D). (Bar = 50 μm).

*Bipolaris australiensis* (Bugnicourt) Subram. & Jain ex M. B. Ellis; Subram. & Jain, 1966, Curr. Sci. **35**: 354. (Fig. 2A)

Colonies effuse, grey to dark blackish brown, velvety. Reverse blackish. Hyphae pale to dark brown, smooth, septate, 2-4 μ thick. Conidiophores solitary flexuous or geniculate, septate, reddish brown, sometimes up to 150 μ long but usually shorter, width 3-7 μm. Conidia straight, ellipsoidal or oblong, rounded at the ends, pale brown to mid reddish brown, 11.2- 26 × 6.6-9.8 μm.

*Specimen examined:* Isolates from the infected petals of *Tagetes patula* L., Botanical Garden, University of Dhaka, Dhaka, 18 March 2009, M. Aktar 19.

*Chaetomium globosum* Kunze ex Fr., *Systema Mycologicum* 3: 255 (1829). (Fig. 2B)

Colony cottony, greyish black, mycelia septate profusely branched. Perithecia globose with appendages. Ascospores lemon shaped and  $8-14.8 \times 3.4-6.8 \mu\text{m}$ .

*Specimen examined:* Isolates from infected leaves of *Tagetes erecta* L., Bangabandu Sheikh Mujibur Rahman Novo Theater, 17 January 2010, M. Aktar 28.

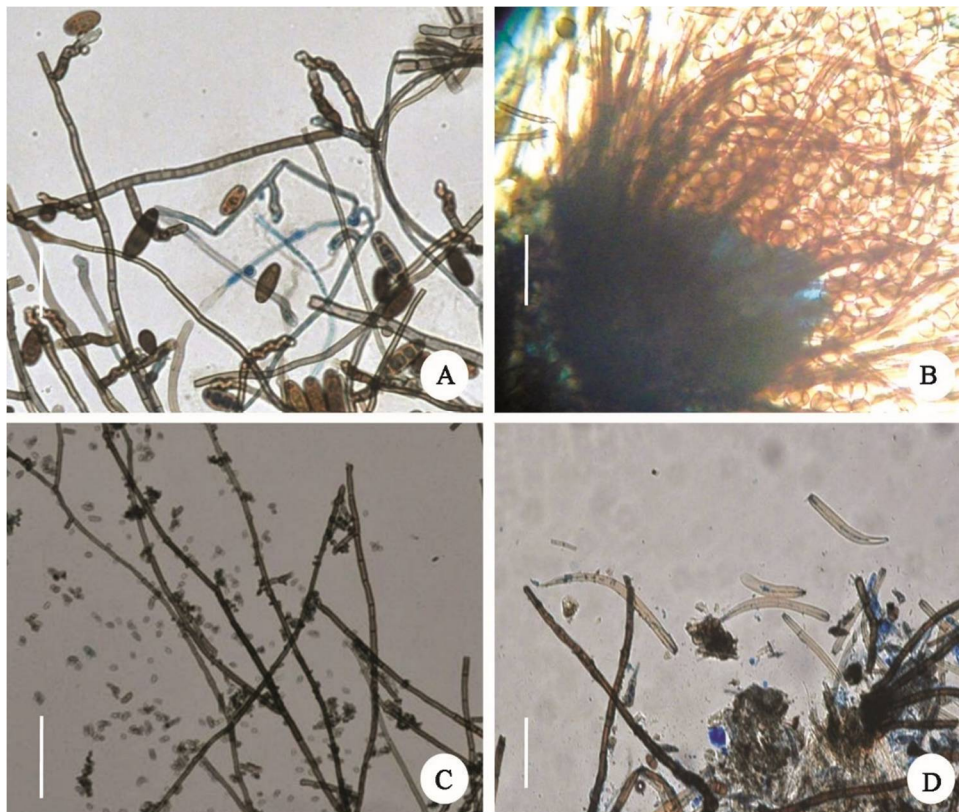


Fig.2. Conidiophores and conidia of *Bipolaris australiensis* (A), peritheceum, appendages and ascospores of *Chaetomium globosum* (B), conidiophores and conidia of *Cladosporium elatum* (C) and conidiophores and conidia of *Corynespora cambrensis* (D). (Bar = 50  $\mu\text{m}$ ).

*Cladosporium elatum* (Harz) Nannf., 1934, *Svenska SkogsvFo.Tidskr.* 32: 397. (Fig. 2C)

Colonies on medium effuse, olivaceous grey, reverse blackish olive. Conidiophores straight or flexuous, pale to mid brown or olivaceous brown, smooth, up to 87.6  $\mu\text{m}$  long, 5-6.8  $\mu\text{m}$  thick. Conidia in very long, branched chains forming wide, loose heads, fusiform, limoniform or subspherical tapered into a narrow tube or tubes at one or both ends, mostly 0 septate, very pale brown or olivaceous brown, smooth,  $8-16 \times 4.2-5.4$ , ramo-conidia present.

*Specimen examined:* Isolates from infected leaves of *Tagetes erecta* L., Botanical Garden, University of Dhaka, Dhaka, 16 February 2009, M. Aktar 5.

*Corynespora cambrensis* M. B. Ellis, 1971.

(Fig. 2D)

Colonies on medium effuse, reverse grayish, hyphae brown, smooth, septate, 2-4  $\mu\text{m}$  thick. Conidiophores arising singly or sometimes in fascicles, often proliferating terminally through the apical conidial scar, straight or flexuous, unbranched, brown or olivaceous brown, smooth. Conidia solitary often connected with the conidiophore with a hyaline isthmus or catenate, mostly obclavate, cylindrical., subhyaline, pale to darkly pigmented, pseudoseptate, smooth, 24.3-110.7  $\times$  5.4-10.8  $\mu\text{m}$ .

*Specimen examined:* Isolates from infected petals of *Tagetes patula* L., Chottogram road, 13 February 2010, M. Aktar 32.

*Curvularia brachyspora* Boedijn. Ellis, M.B., Mycol. Pap.106:2-43, 1966.

(Fig. 3A)

Colonies effuse, black, reverse blackish. Hyphae brown, smooth, septate. Conidiophore solitary, branched, straight, mostly flexuous geniculate, mid brown, septate, up to 62  $\mu\text{m}$  long and 6.0-8.2  $\mu\text{m}$  thick. Conidia mostly 3-septate, dark brown, mostly curved, smooth, 22-28  $\times$  9-13  $\mu\text{m}$ .

*Specimen examined:* Isolates from infected leaves of *Tagetes erecta* L., Botanical Garden, University of Dhaka, Dhaka, 14 February 2010, M. Aktar 33.

*Curvularia fallax* Boedijn. Ellis, M.B., Mycol. Pap., 106:2-43, 1966.

(Fig. 3B)

Colonies effuse, black, hairy. Reverse blackish. Hyphae brown, smooth, septate. Conidiophores solitary, branched, straight, mostly flexuous geniculate, mid brown, septate, up to 62  $\mu\text{m}$  long and 6.8-7.2  $\mu\text{m}$  thick. Conidia mostly 4-septate, dark brown, mostly curved, smooth, (16) 22-40  $\times$  (7) 9-15  $\mu\text{m}$ .

*Specimen examined:* Isolates from infected petals of *Tagetes patula* L., BARI, 24 February 2011, M. Aktar 47.

*Curvularia lunnata* (Wakker) Boedijn. Ellis, M.B., Mycol. Pap.106:2-43, 1966.

(Fig. 3C)

Colonies effuse, dark black. Reverse blackish. Hyphae brown, smooth, septate, branched. Conidiophores solitary, mostly branched, straight, mostly flexuous geniculate, pale to dark brown, septate, 24-60.4 (70.6)  $\mu\text{m}$  long, 8-10.4  $\mu\text{m}$  thick, often swollen at the base. Conidia 3 septate, olivaceous black to dark brown, almost always curved at the third cell from the base which is larger and darker than the others, end cells subhyaline or pale brown, smooth, 20 - 34  $\times$  9-15  $\mu\text{m}$ .

*Specimen examined:* Isolates from infected buds and petals of *Tagetes erecta* L., Botanical Garden, University of Dhaka, Dhaka 16 February 2009, M. Aktar 6.

*Curvularia stapeliae* (du Plessis) Hughes & du Plessi. Ellis, M.B., Mycol. Pap.106:2-43, 1966.

(Fig. 3D)

Colonies effuse, grayish black. Reverse blackish. Hyphae brown, smooth, septate, branched, straight or slightly undulating, often geniculate, pale to dark brown, septate, 20-64.4 (83.6)  $\mu\text{m}$  long, 6-9.4  $\mu\text{m}$  thick, often swollen at the base. Conidia 3 septate, olivaceous black to dark brown, almost always curved at the third cell from the base which is larger and darker than the others, end cell subhyaline or pale brown, smooth, 30-45  $\times$  11-17  $\mu\text{m}$ .

*Specimen examined:* Isolates from the infected petals of *Tagetes patula* L., Khagrachari, 24 May 2012, M. Aktar 61.

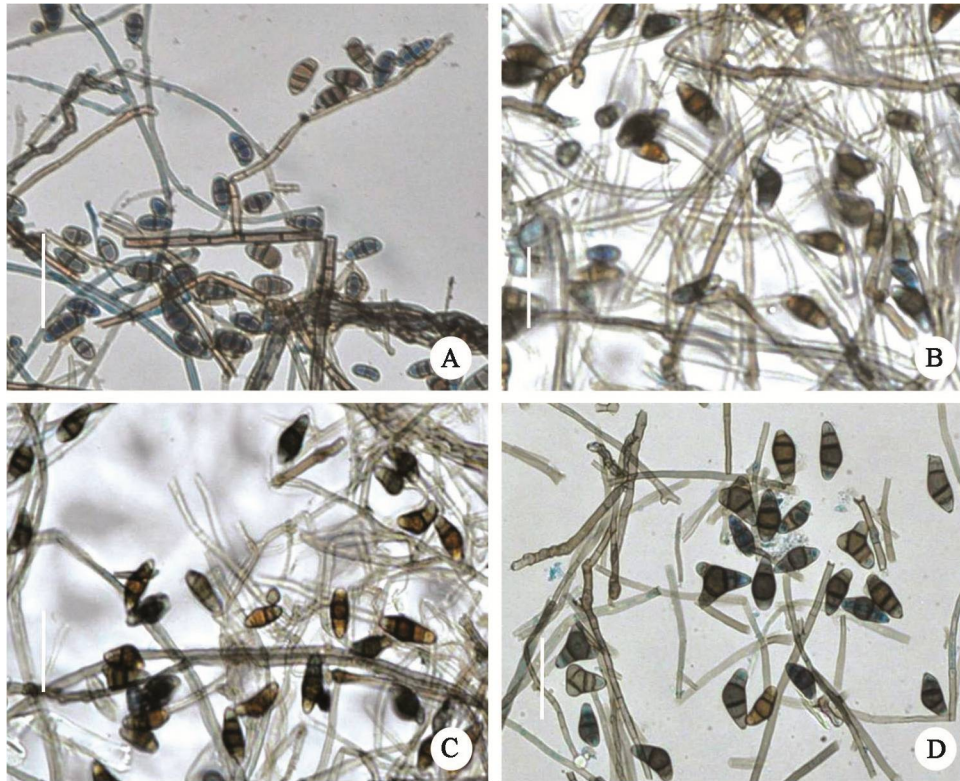


Fig.3. Conidiophores and conidia of *Curvularia brachyspora* (A), *C. fallax* (B), *C. lunata* (C) and *C. stapeliae* (D). (Bar = 50  $\mu$ m).

*Epicoccum purpurascens* Ehrenb. ex Schlecht., 1824, Synop. Pl. crypt.: 136. (Fig. 4A)

Colonies grayish green, reverse blakish green. Hyphae brown, septate, profusely branched. *Sporodochia* up to 2.5 mm diam. Conidiophores 5-16  $\times$  3-6  $\mu$ m. Conidia brown muriform, 14 - 27 (51)  $\mu$ m diam.

*Specimen examined*: Isolates from the infected bud of *Tagetes patula* L., Botanical Garden, University of Dhaka, Dhaka, 14 February 2010, M. Aktar 34.

*Fusarium semitectum* Berk. & Rav. in Berkeley, Grevillea 3: 98, 1875. (Fig. 4B)

Colonies at first white and gradually becomes pink in colour. Mycelia hyaline, septate, profusely branched. *Sporodochia* absent, *Phialide* present. Microconidia 6-11.2  $\times$  1.6-2.8  $\mu$ m and macroconidia 17.2-25.2  $\times$  1.8-3.4  $\mu$ m.

*Specimen examined*: Isolates from infected leaves of *Tagetes patula* L., Botanical Garden, University of Dhaka, Dhaka, 16 February 2009, M. Aktar 7.

*Monochaetia ceratoniae* (Sousa da Camera) Sutton, Mycol. Pap. 88: 42 (1963). (Fig. 4C)

Colonies white, cottony reverse off white. Mycelium immersed, branched, pale brown, septate. Acervulus black shinnig. Conidiophores hyaline short, cylindrical, straight or curved, sparsely branched, septate only at the base. Conidia brown, 4 septate, end cells hyaline, 19-23  $\times$

5.6-7.4  $\mu\text{m}$ , median cells thin-walled, smooth, 13-14 $\mu\text{m}$  long; apical appendage 5-6  $\mu\text{m}$  long, basal appendage 2.5-4  $\mu\text{m}$  long.

*Specimen examined:* Isolates from the infected petals of *Tagetes erecta* L., Botanical Garden, University of Dhaka, Dhaka, 16 February 2009, M. Aktar 8.

*Nigrospora panici* Zimm. M.B. Ellis 1971.

(Fig. 4D)

Colonies brownish black with shining black conidia on the surface. Mycelium partially superficial, branched, brown, septate. Conidiophores semi-macronemous, mostly unbranched, hyaline. Conidia solitary, sphaerical or broadly ellipsoidal, compressed dorsiventrally, aseptate, black shining, smooth 0-septate. 22.4-33.6  $\mu\text{m}$  diam.

*Specimen examined:* Isolates from the infected petals of *Tagetes erecta* L., Chottogram, 27 January 2010, M. Aktar 30.

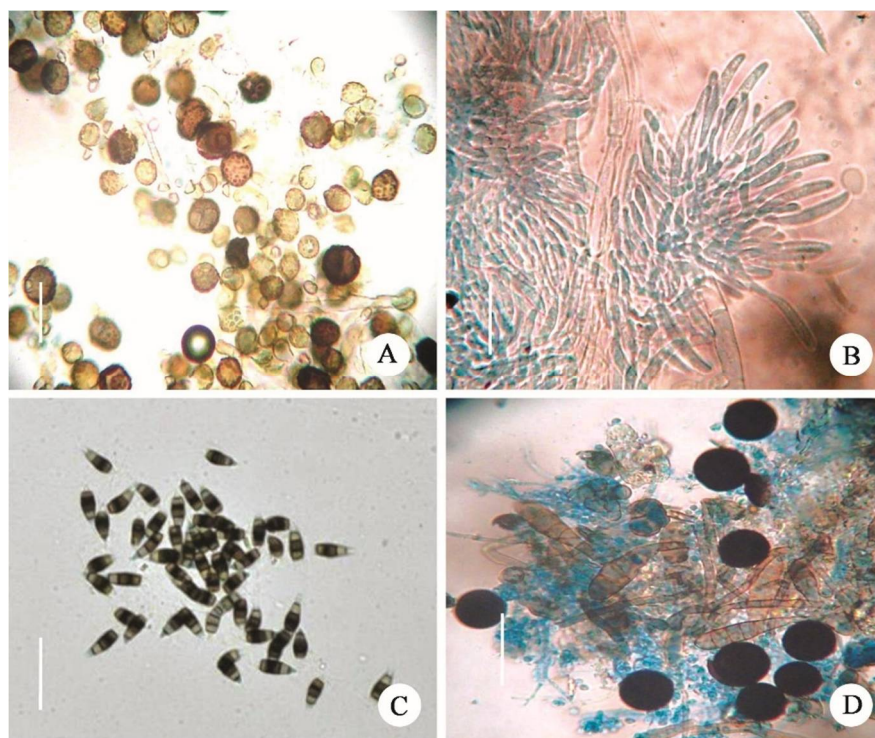


Fig.4. Sporodochia, conidiophores and conidia of *Epicoccum purpurascens* (A), *Fusarium semitectum* (B), Acervulus, conidiophores and conidia of *Monochaetia ceratoniae* (C) and conidiophores and conidia of *Nigrospora panici* (D). (Bar = 50  $\mu\text{m}$ ).

*Penicillium italicum* Wehmer, Hedwigia 33: 211-214. 1894.

(Fig. 5A)

Colonies on PDA medium growing restrictedly, often marked by a few shallow furrows, with margins usually its inner surface pale gray-green shades. Penicilli asymmetric, often comparatively long up to 50-70  $\mu\text{m}$  and 3-5  $\mu\text{m}$  width, bearing tangled chains of conidia. Strigmata 3-6 in a whole 8-12  $\times$  3.5  $\mu\text{m}$ . Conidia one celled with greenish tinge, 4-5  $\times$  25-35  $\mu\text{m}$ .

*Specimen examined:* Isolated from the infected calyx of *Tagetes erecta* L., Botanical Garden, University of Dhaka, Dhaka, 12 April 2009, M. Aktar 9.

*Rhizopus stolonifer* (Ehrenb.) Vuill., Revue Mycologique Toulouse **24**: 54 (1902). (Fig. 5B)

*Mycelium* coenocytic, well-developed, branched and fluffy. Mycelium produces many aerial stolones that develop rhizoids at certain points. Directly above the rhizoids one or more sporangiophores are produced. The top of each sporangiophores becomes swollen as the latter reaches maturity, and a sporangium is developed. The central portion of sporangium becomes highly vacuolated and it eventually surrounded by a wall that separates it's from the peripheral zone. The central portion in the columella. Sporangium produces non-motile sporangiospores.

*Specimen examined:* Isolates from the infected leaves of *T. patula* L., Mushroom Centre, Savar, 21 April 2011, M. Aktar 51.

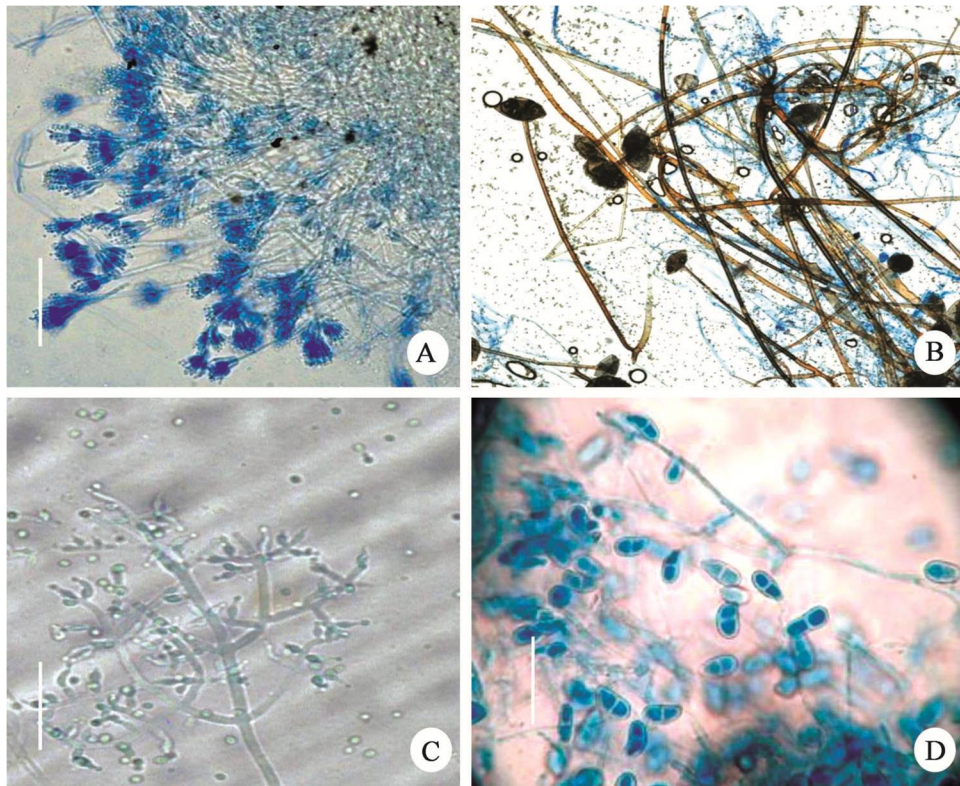


Fig.5. Conidiophores and conidia of *Penicillium italicum* (A), *Rhizopus stolonifer* (B), *Trichoderma viride* (C) and *Trichothecium roseum* (D). (Bar = 50  $\mu$ m).

*Trichoderma viride* Pers., Neues Magazin für die Botanik **1**: 92 (1794). (Fig. 5C)

*Colony* effuse, light green. Conidiophores hyaline, much branched, bearing phiallides single or in groups. Conidia hyaline, powdery mass, 1-celled, ovoid, borne in small terminal clusters 3-4  $\mu$ m.

*Specimen examined:* Isolates from the infected petal of *Tagetes erecta* L., Chottogram road, 8 February 2010, M. Aktar 38.



*Trichothecium roseum* (Pers.) Link, Magazin der Gesellschaft Naturforschenden Freunde Berlin **3**(1): 18, t. 1:27 (1809). **(Fig. 5D)**

Colonies effuse, at first white but soon turning rosy pink. Conidiophores up to  $147 \times 3-4.5 \mu\text{m}$ , hyaline, often slightly swollen at their tips. Conidia hyaline, pink in mass, 1-septate, thick walled, each with a flattened protuberance at the base,  $12-18.4 \times 5.4-7.2 \mu\text{m}$ , often clustered.

*Specimen examined*: Isolates from the infected petal of *T. erecta* L., BARI, 14 March 2011, M. Aktar 49.

An extensive survey of the literature revealed that *Corynespora cambrensis* M. B. Elli, *Monochaetia ceratoniae* (Sousa da Camera) Sutton and *Nigrospora panici* Zimm has not been reported in any relevant literature of Bangladesh (Talukar 1974, Siddiqui *et al.* 2007, Shamsi and Hosen 2016, Shamsi 2017 and 2018 and Momtaz *et al.* 2018). Hence, *Corynespora cambrensis* M. B. Elli, *Monochaetia ceratoniae* (Sousa da Camera) Sutton and *Nigrospora panici* Zimm. are reported here as a new fungal record for Bangladesh.

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