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REPORT ON ALTERNARIA BLIGHT OF TAGETES ERECTA AND TAGETES PATULA CAUSED BY ALTERNARIA ALTERNATA (Fr.) KEISSLER

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Abstract

Severe blight symptom was recorded on *Tagetes erecta* L. and *Tagetes patula* L. during the tenure of January to April 2009, 2010, 2011, 2012 and 2013. Samples were collected from Chittagong, Comilla, Dhaka, Gazipur, Khulna, Pabna, Rajshahi and Sylhet. Fungi associated with infected samples were isolated following "Tissue Planting" and "Blotter" methods at temperature 25-28^{0}C and pH 6.0. *Alternaria alternata* (Fr.) Keissler was isolated from diseased leaves, buds, calyx and petals of both selected plant species. The pathogenicity of the isolated fungus was tested following modified "detached leaf technique" and spraying of spore suspension methods. *Alternaria alternata* was found to be pathogenic to *Tagetes* spp. This is the first report on *Alternaria* blight of *T. erecta* and *T. patula* from Bangladesh.

Key words: Report, Alternaria blight, *Alternaria alternata*, *T. erecta, T. patula,* Bangladeshs

Introduction

Tagetes erecta and *Tagetes patula* belong to Asteraceae (Compositae) family and it is native to North and South America, but some species now become naturalized around the world. *Tagetes erecta* are the tallest, at three to five feet. Flowers are golden yellow, orange or cream coloured. They are sometimes known as American or African marigolds. *Tagetes patula* is bushy, somewhat smaller plant as compare to *T. erecta* and known as French marigold. They are brick red, orange red, yellowish or brownish yellow in colour. No annual flower is more cheerful and easier to grow than marigolds. French Marigolds are commonly planted in butterfly gardens as a nectar source. The florets 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* is a natural pesticide. Leaves are used as blood clotting agents in Ayurbedhic treatment. Plants has antifungal properties also. Plant is also used against fever dysenteries, indigestions, ulcers and eczemas. It is most effective against the nematode species Pratylenchus penetrans (Olabiyiand and Oyedunmade 2000 and Politi *et al.* 2012). Plant has also mosqutocidal potentiality (Rajasekaran *et al.* 2004). Marigold is now a profitable cultivated crop to the farmers, but socioeconomic data and information of this flower are very scare in Bangladesh. In Jessore and Jhenaidah districts 95% farmers cultivated T-004 line and only 5% farmers cultivated T-003 line of marigold.

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The yield of marigold was 2,650,447 flowers per hectare. The gross margin and net return 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 problems for marigold cultivation.

In Bangladesh, rapid expansion of commercial marigold cultivation many diseases appear on the plants. In many cases, disease occurrence is an important threat for commercial cultivation of marigold. However, reports on the occurrence of diseases of marigold in Bangladesh are scanty (Aktar and Shamsi 2012). Reports on the yield loss of marigold due to diseases are not available in the country. Present study was undertaken to find out the presence of pathogenic fungi with marigold in Bangladesh.

Materials and Methods

In the present study fungi were isolated from infected leaves, buds and flowers of *Tagetes erecta* and *Tagetes patula.* Samples were collected from BARI, Chittagong, Comilla, Dhaka, Gazipur, Joydebpur, Khulna, Pabna, Rajshahi and Sylhet during the period of January to April 2009, 2010, 2011, 2012 and 2013. All these samples showed severe blight symptom on leaves, bud and flowers. Sixty samples were examined from infected plant parts of both species. Fungi associated with healthy and infected samples were isolated following "Tissue Planting" and "Blotter" methods(CAB 1968). Experiment was conducted in the Laboratory of Mycology and Plant Pathology, Department of Botany, University of Dhaka. In case of Tissue Planting" method, 30 inocula each measuring 2 square mm. were cut with a sterilized scalpel from a particular specimen and kept in a sterile Petri plate. The inocula were washed in sterile water and then surface sterilized by dipping in 10% Clorox for 3-5 minutes. Then the inocula were transferred in to a sterile Petri plate containing sterile blotting paper to remove the surface water. Three inocula were placed in each plate containing sterilized Potato Dextrose Agar (PDA) medium in pH 6 and incubated for 5-7 days at temperature 25 to 28^0 C. In "Blotter" method, moist chambers were made by placing two layers of filter paper on the bottom of the Petri plates, moistened with sterilized water, covered with upper lids and were sterilized (autoclaved at 15 lbs pressure and 120°C temperature). Four square mm. sized inocula were prepared from the samples, surface sterilized with 10% chlorox for 3-5 minutes. A total number of 30 inocula was transferred in 10 Petri plates and incubated for 5-7 days at temperature 25 to 28^0 C. The fungi growing out of the inocula were transferred to separate plates and slants for further studies and storage. Percentage association of the fungi was also recorded.

Identification of the isolates was determined by following the standard literatures (Ellis 1971). All the specimens were preserved in the Herbarium, Mycology and Plant pathology section, Department of Botany, University of Dhaka, Bangladesh.

The pathogenicity of all the isolated fungi was tested following modified "detached leaf technique" (Azad and Shamsi 2011). Moist chamber was prepared by placing small cotton bar at the corner of Petri plate and autoclaved. Six treatments with three replications for each fungi were used as follows: T_1 = (control) dorsally uninoculated leaflets, T_2 = (control) ventrally uninoculated leaflets, T_3= dorsally unpricked inoculated leaflets, T_4 = ventrally unpricked inoculated leaflets, T_5 = dorsally pricked inoculated leaflets and T_6 = ventrally pricked inoculated leaflets.

Healthy seedlings of *Tagetes erecta* and *T. patula* were separately transplanted in earthen pots (10 inch diameter.) containing sterilized soil at three seedlings per pot and allow to grow for one month in net house providing necessary water and nutrients. Identified fungus were purified and its pathogenicity was examined by inoculating fresh healthy plants following spraying of spore suspension method. Conidia from seven days old culture of test fungus were taken in 250 ml conical flask with sterilized water. Ten ml water suspension of test fungus at 10^4 ml conc. were taken in a hand sprayer and sprayed on healthy potted plant. Control received only sterilized water without fungal inoculum. Five pots were inoculated for each treatment. The inoculated and control plants were placed in a clean bench in net house following completely randomized design.

The plants were examined daily and continued for 10 days for recording the development of symptoms. Symptom produced on artificial inoculated plants was recorded and compared with those observed on naturally infected plants. The fungus was reisolated from the inoculated plants of *Tagetes* spp. on PDA medium to fulfill Koch's postulates.

Results and Discussion

Severe blight symptom was noticed on leaves, buds and flowers of *Tagetes erecta* and *T. patula.* Healthy and infected plants of *T. erecta and T. patula* are presented in Plate I.

During the period of this study *Alternaria alternata*, was frequently isolated from different parts of *Tagetes* spp. Morphological characters of the fungi were recorded on PDA. Colonies of *A. alternata* were black velvety. Hyphae were pale to mid brown, smooth septate, 1-5 µm in diameter. Conidiophore was solitary, flexous, septate, pale to mid brown, up to 85 µm long, but usually much shorter (14-60) µm and 4-7(9) µm in diameter. Conidia were straight, muriform, oblong, rounded at the base, pale to mid brown, 2-7 (mostly 5) septate, 20-55 (76) × 8-18 (13) µm. Beak was 2-5 µm thick (Plate 2.).

Plate 1. *Tagetes erecta*: A. Healthy plants, B-D. Infected plants
Tagetes patula : E. Healthy plants, F. Infected plants

The morphological characters recorded in the present study were compared with those reported by Ellis 1971. *Alternaria alternata* is opportunistic pathogen on numerous host causing leaf spots, rots and blight on many plant parts. It was recorded causing leaf spot and other diseases on over 380 host species (Wikipedia 2013). It is a member of the

imperfect fungi and is one of the most important amongst the allergenic fungi. The fungus has been isolated from plants, soil, food, indoor and outdoor air. (Wikipedia 2013).

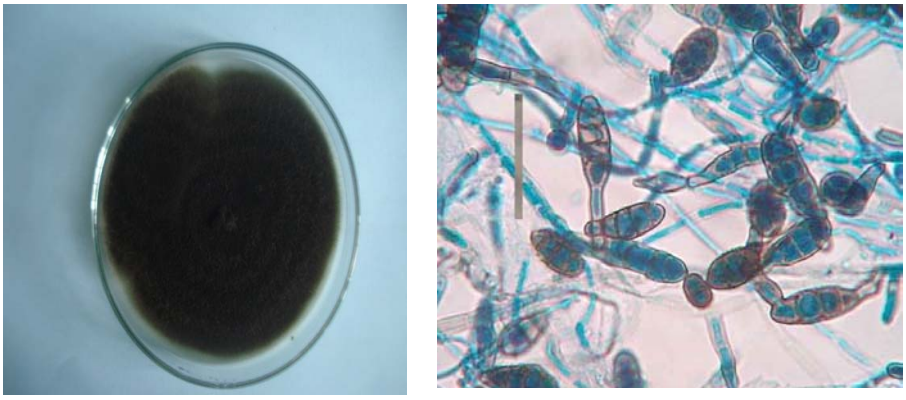

Plate 2. *Alternaria alternata* : A. 10 days culture on PDA medium,
B. Mycelia, conidiophores and conidia.

Leaf spot and blight are two common diseases of *Tagetes erecta* and *T. patula*. Mukerji and Bhasin (1986) reported *Alternaria alternata* (Fr.) Keissler, *A. tagetica* (Shome and Mustaffe), *A. tenuissima* (Kze. Ex Pers) Wilt.), flower and bud rot (*A. dienthii* Stevens & Hae), leaf and inflorescens blight (*A. zinnia* Pape), and head blight and grey mold (*Botrytis cinerea* Pers.), diseases of *T. erecta* and *T. patula* plant from India. Sultana and Shamsi (2011) reported gray mold of *T. erecta* caused by *Botrytis cineria* from Bangladesh. Dhiman and Arora (1990) reported Leaf spot and flower blight of marigold (*Tagetes erecta* L.) caused by *Alternaria tagetica* in Punjab. Due to disease average reduction of 28.2 and 53.53% was in seed weight and germination respectively. The seeds obtained from diseased flowers produced 2-5% sickly seedlings.

In the present study *A. alternata* was found to be the causal agent of blight disease of *Tagetes* spp. In case of *T. erecta* highest frequency percentage of association of the fungus was recorded (100%) in 2013 on flower parts followed by leaves (90%). Frequency percentage of association of the fungus was recorded 100% on leaves in 2010 and on calyx in 2013. During the year 2011 and 2012 *T. patula* did not show symptom in sampling areas (Table 1).

Table 1. Frequency (%) of association of *Alternaria alternata* with symptomatic plant parts of *Tagetes erecta* and *T. patula* during 2009-2013.

Years	*T. etrecta*				*T. patula*			
	Plant parts							
	Leaf	bud	calyx	petal	Leaf	bud	calyx	petal
2009	60.00	20.00	33.33	56.66	60.00	16.66	46.66	16.66
2010	13.33	50.00	66.66	63.33	100.00	16.66	16.66	50.00
2011	16.66	80.00	90.00	23.33	-	-	-	-
2012	-	-	-	40.00				
2013	90.00	100.00	100.00	100.00	60.00	50.00	100.00	33.33

"- " = plants did not show symptom in sampling area

Alternaria alternata showed symptom on all the inoculated leaflets and plants of *Tagetes* spp. *in vitro* and *in vivo* except control leaflets and plants. The fungus was successfully reisolated from inoculated leaflets and plants (Plates 3 and 4). This is the first report of Alternaria blight of *Tagates* spp. from Bangladesh. Present investigation will be helpful for designing successful control measures of the disease.

Plate 3. *Tagetes erecta*: : A. T_1 = (control) dorsally uninoculated leaflets, B. T_2 = (control) ventrally uninoculated leaflets, C. T_3 = dorsally unpricked inoculated leaflets, D. T_4 = ventrally unpricked inoculated leaflets, E. T_5 = dorsally pricked inoculated leaflets and F. T_6 = ventrally pricked inoculated leaflets. *Tagetes patula:* G. T_1 = (control) dorsally uninoculated leaflets, H. T_2 = (control) ventrally uninoculated leaflets, I. T_3 = dorsally unpricked inoculated leaflets, J. T_4 = ventrally unpricked inoculated leaflets, K. T_5 = dorsally pricked inoculated leaflets and F. T_6 = ventrally pricked inoculated leaflets.

Plate 4. *Tagetes erecta*: A. control, B. treated plant
Tagetes patula: C. control, D. Infected plants.

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