

MYCOFLORA ASSOCIATED WITH MOMORDICA CHARANTIA L. AND THEIR PATHOGENIC POTENTIALITY

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Abstract

Momordica charantia L. (Bitter gourd) is one of the popular vegetable in Bangladesh as well as in the world. After harvesting the vegetables are contaminated with fungi within 3-4 days of short storage. An investigation was carried out to find out the fungi associated with fruits of two varieties of *Momordica charantia* during the tenure of May 2015 to June 2016. Nine species of fungi namely, *Aspergillus flavus* Link, *A. fumigatus* Fresenius, *A. niger* van Tiegh, *Curvularia brchyspora* Boedijn, *Fusarium* Link, *Mucor* Fresen, *Penicillium* Link, *Rhizopus stolonifer* (Ehrenb.:Fr.) Vuill and *Trichoderma viride* Pers. were found to be associated with the selected vegetable. *Aspergillus niger* was predominating fungus associated with both the varieties of bitter gourd. Among the isolated 9 fungi *A. niger*, *C. brchyspora*, *Fusarium* sp., *R. stolonifer* and *T. viride* were found to be pathogenic to both the varieties of *M. charantia*.

Key words: Mycoflora, *Momordica charantia*, Pathogenic potentiality

Introduction

Momordica charantia, known as bitter melon, bitter gourd, bitter squash or balsam-pear is a tropical and subtropical vine of the family Cucurbitaceae. Bitter gourd are available in a variety of shapes and sizes. The cultivar common to China is 20–30 cm long, oblong with bluntly tapering ends and pale green in color, with a gently undulating, warty surface. The bitter melon more typical of India has a narrower shape with pointed ends, and a surface covered with jagged, triangular "teeth" and ridges. It is green to white in color. Between these two extremes there are some intermediate forms. Some bear miniature fruit of only 6–10 cm in length, which may be served individually as stuffed vegetables. These miniature fruit are popular in Bangladesh, India (common name 'Korolla'), Pakistan, Nepal and other countries in South Asia.

Bitter gourd can be eaten as cooked and play an important role in human nutrition, being mostly low in fat and carbohydrates, but high in vitamins, minerals and fiber. Vegetables are rich and comparatively cheaper source of vitamins and minerals. Its consumption in sufficient quantities provides taste, palatability and increase appetite. Vegetables are currently reckoned as important adjunct for maintenance of food health and beneficial in protecting against some degenerative diseases. They also play a key role in neutralizing the acids that are formed during digestion (Ananya and Sarmistha 2010). Many people in

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urban and rural areas fully depend on vegetable cultivation and selling in the markets. But it is the matter of deep concern that a remarkable portion of harvested vegetables are being lost due to mismanagement of the vegetables during transit and storage, as a result fungal infection occurs and consequently vegetables are spoiled. The major diseases of *M. charantia* are Alternaria rot (*Alternaria alternata*), Belly rot (*Rhizoctonia solani*), Cottony leak (*Pythium* sp.), Rhizopus soft rot (*Rhizopus stolonifer*), Botryodiplodia rot (*Botryodiplodia theobromae*) (Wikipedia 1016) etc. Haque and Shamsi (2011) reported fungal association of five vegetables in Bangladesh. So far there is no report regarding the association of fungi with fresh fruit of *M. charantia* in storage. Present investigation was undertaken to find out the association of fungi with *M. charantia* and their pathogenic potentiality.

Materials and Methods

Fruits of two varieties of *Momordica charantia* L. (Korolla) were used in the present investigation. One is local variety (small in size, bitter in taste) and another is a hybrid variety (larger in size, less bitter in taste). The samples were collected from five different markets of Dhaka city namely, Ananda bazar, Hatirpul bazar, Kawran bazar, Palashi bazar and Siddique bazar. Five markets were visited three times to collect the samples. From each market sufficient amount of fresh vegetables were collected randomly. Samples were collected during May to December 2015. The fungi were isolated from samples following the 'Tissue Planting Method' on PDA medium following Islam and Shamsi (2016). Identification of the isolates were determined following the standard literature (Barnett and Hunter 1972, Booth 1971, Ellis 1971, 1976, Ellis and Ellis 1997, Thom and Rapper 1945 and Rapper *et al.* 1949).

Per cent frequency of the occurrence of the fungal isolates was calculated by adopting the following formula (Spurr and Welty 1972):

$$\% \text{ frequency} = \frac{\text{Total number of inocula from which a fungal isolate was observed}}{\text{Total number of inocula}} \times 100$$

All the fungi isolated from fruits of *M. charantia* were tested for their pathogenic potentiality following detached fruit assay (Shamsi *et al.* 2016).

Pure isolates from *M. charantia* were individually inoculated into healthy fruits of *M. charantia* at the same stage of maturity. The fungi that were isolated from *M. charantia* were grown separately on PDA medium at pH 6.0 and temperature 25 to 28°C for five to seven days. The healthy vegetables were surface sterilized with 10% Chlorox and washed thrice in changes of sterilized distilled water. Then the inoculated vegetable were kept for one hour for drying.

When the vegetables were fully air dried, a cork borer (5 mm diameter) was driven to a depth of 4 mm into the vegetables to make a groove where the isolated organism could be put. The cultured fungal isolates were also cut into round block with the help of

another sterile cork borer (4 mm diameter) from pure PDA culture medium. Some fresh PDA blocks were also cut for the control set. Two sets of fruits of *M. charantia* were prepared where one set is control i.e, fruits inoculated with PDA blocks and another set is treated i.e, fruits inoculated with particular test pathogens. Two round grooves in local variety and three grooves in hybrid variety of *M. charantia* in suitable distance were made. Fresh PDA blocks were placed aseptically into the groove of the control set and the mycelial blocks were placed into the grooves of the treatment set. Then both the control set and the treatment set of vegetables were incubated into separate sterilized plastic boxes of suitable size. Inoculated fruits were incubated for 5 to 7 days at $25 \pm 2^\circ\text{C}$. Regular observations were made and re-isolation of any pathogenic test fungi were carried out for comparison with the original fungi.

Results and Discussion

A total of nine species of fungi namely *Aspergillus flavus*, *A. fumigatus*, *A. niger*, *Curvularia brachyspora*, *Fusarium* sp., *Mucor* sp., *Penicillium* sp., *Rhizopus stolonifer* and *Trichoderma viride* was isolated from the fresh fruits of two varieties of *M. charantia*.

Table 1 shows eight species of fungi namely *Aspergillus flavus*, *A. niger*, *A. fumigatus*, *Curvularia brachyspora*, *Fusarium* sp., *Penicillium* sp., *Rhizopus stolonifer* and *Trichoderma viride* which were isolated from *M. charantia* (Local variety). Four species of fungi (*Aspergillus niger*, *A. flavus*, *Penicillium* sp., *Rhizopus stolonifer*) were isolated from the samples of Ananda bazar where frequency percentage of association of *A. niger* was the highest (44.) followed by *Rhizopus stolonifer* (26.7) and *Penicillium* sp. (24.3). *A. flavus* showed lowest mean frequency (15.7). Four species of fungi were isolated from Hatirpul bazar where frequency percentage of association of *A. niger* was the highest (31) followed by *A. flavus* (22.3) and *Penicillium* sp. (21). Frequency percentage of association of *Fusarium* sp. was lowest (15.7). Five species of fungi were isolated from the samples of Karwan bazar where frequency percentage of association of *A. niger* was the highest (40) followed by *Trichoderma viride* (20), *Penicillium* sp. (17.7) and *Fusarium* sp. (13.3). Frequency percentage of association of *A. fumigatus* was lowest (09). Five species of fungi were isolated from the samples of Palashi bazar where frequency percentage of association of *Aspergillus niger* was the highest (49) followed by *Penicillium* sp. (42), and *A. flavus* (15.7). Lowest count of *C. brachyspora* and *T. viride* (11). Five species of fungi were isolated from the samples of Siddique bazar where frequency percentage of association of *Aspergillus niger* was also found the highest (40) followed by *A. fumigatus* (20), *Fusarium* sp. (15.7) and *Penicillium* sp. (13.3%). Frequency percentage of association of *C. brachyspora* was lowest (11%).

Table 1. Mean frequency (%) of association of fungi with fruits of *M. charantia* (Local variety) samples collected from five different markets.

Name of isolates	Frequency (%) of fungi					Total	Mean
	A	H	K	P	S		
<i>Aspergillus flavus</i>	15.7	22.3	0	15.7	0	53.7	10.74
<i>A. fumigatus</i>	0	0	09	0	20	29	5.80
<i>A. niger</i>	44.3	31	40	49	40	204.3	40.86
<i>C. brachyspora</i>	0	0	0	11	11	22	4.40
<i>Fusarium</i> sp.	0	15.7	13.3	0	15.7	44.7	8.94
<i>Penicillium</i> sp.	24.3	21	17.7	42	13.3	118.3	23.66
<i>Rhizopus stolonifer</i>	26.7	0	0	0	0	26.7	5.34
<i>Trichoderma viride</i>	0	0	20	11	0	31	6.20

A = Ananda bazar, H= Hatirpul bazar, K = Karwan bazar, P = Palashi bazaar,S= Siddique bazar.

Nine species of fungi namely *Aspergillus niger*, *A. flavus*, *A. fumigatus*, *Curvularia brachyspora*, *Fusarium* sp., *Mucor* sp., *Penicillium* sp., *R. stolonifer* and *T. viride* were isolated from fruits of *M. charantia* Hybrid variety (Table 2). Five species of fungi were isolated from the samples of Ananda bazar where frequency percentage of association of *Aspergillus niger* was the highest (40) followed by *Penicillium* sp. (26.7), *Fusarium* sp. (15.3), and *A. fumigatus* (13.3). Frequency percentage of association of *Curvularia brachyspora* was lowest (11). Five species of fungi were isolated from Hatirpul bazar where frequency percentage of association of *A. niger* was highest (35.7) followed by *Penicillium* sp. (22.3), *Fusarium* sp. (15.7) and *A. flavus* (13.3). Frequency percentage of association of *R. stolonifer* was lowest (13.3). Five species of fungi were isolated from the samples of Karwan bazar where frequency percentage of association of *A. niger* was highest (40) followed by *Penicillium* sp. (15.7), *Mucor* sp. (13.3), and *Fusarium* sp. (11). Frequency percentage of association of *A. fumigates* was lowest (6.7). Six species of fungi were isolated from the samples of Palashi bazar where frequency percentage of association of *A. niger* was the highest (49) followed by *Penicillium* sp. (46.7), *T. viride* (22.3), *Mucor* sp. (11) and *A. spergillus flavus* (9). Frequency percentage of association of *R. stolonifer* was lowest (6.7). Six species of fungi were isolated from the samples of Siddique bazar where frequency percentage of association of *A. niger* was also found to be highest (46.7) followed by *Fusarium* sp. (20), *Penicillium* sp. (18), *A. fumigatus* (15.7), *C. brachyspora* (15.7) and *Fusarium* sp. (15.7). Frequency percentage of association of *T. viride* was lowest (11).

Table 2. Mean frequency (%) of association of fungi with fruits of *M. charantia* (hybrid variety) samples collected from five different markets.

Name of isolates	Frequency (%) fungi					Total	Mean
	A	H	K	P	S		
<i>Aspergillus flavus</i>	0	13.3	0	9	0	22.3	4.46
<i>A. fumigatus</i>	13.3	0	6.7	0	15.7	35.7	7.14
<i>A. niger</i>	40	35.7	40	49	46.7	211.4	42.28
<i>C. brachyspora</i>	11	0	0	0	15.7	26.7	5.34
<i>Fusarium</i> sp.	15.3	15.7	11	0	20	62	12.4
<i>Mucor</i> sp.	0	0	13.3	11	0	24.3	4.86
<i>Penicillium</i> sp.	26.7	22.3	15.7	46.7	18	129.4	25.88
<i>Rhizopus stolonifer</i>	0	13.3	0	6.7	0	20	4.00
<i>Trichoderma viride</i>	0	0	0	22.3	11	33.3	6.66

A = Ananda bazar, H= Hatirpul bazaar, K = Karwan bazar, P = Palashi bazar, S= Siddique bazar.

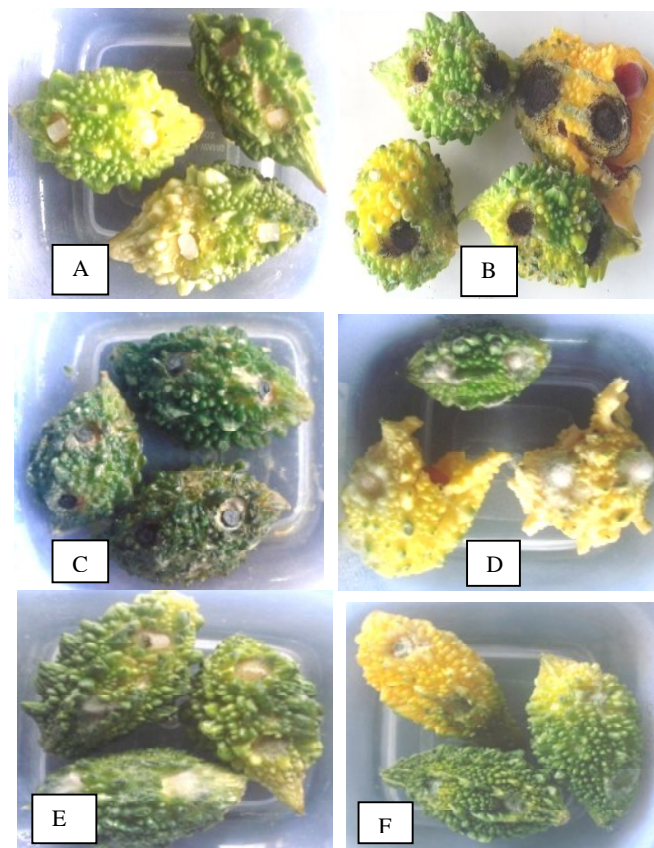


Plate 1. Fruits of *M. charantia* (Local variety) :A. control fruits; B. fruits inoculated by *A. niger*, *C. brachyspora*; D. *Fusarium* sp.; E. *R. stolonifer* and F. *T. viride*.

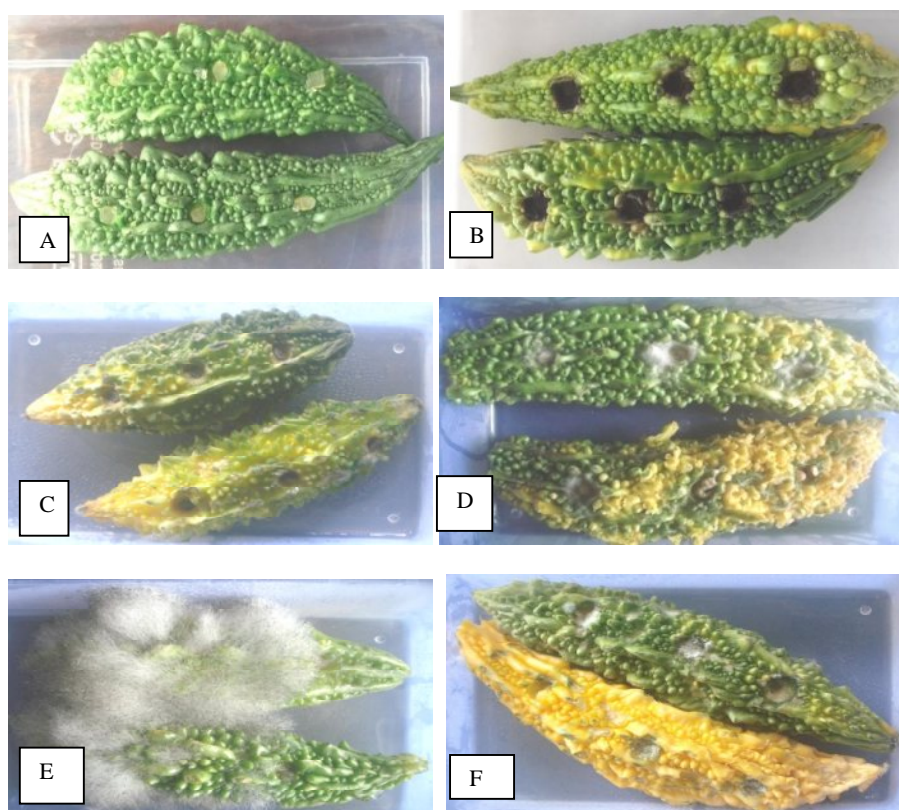


Plate 2. Fruits of *M. charantia* (Hybrid variety): A. control fruits, B. fruits inoculated by *A. niger*, C. *brachyspora* ; D. *Fusarium* sp. E. *R. stolonifer* and F. *T. viride*.

All the isolated fungi from local and hybrid variety of *M. charantia* were tested for their pathogenic potentiality. Five isolated fungi viz, *A. niger*, *C. brachyspora*, *Fusarium* sp., *R. stolonifer* and *T. viride* were found to be pathogenic to both the varieties of *M. charantia*. Control plate remained fresh without any fungal growth (Plates 1 and 2).

From India Mukerji and Bhasin (1986) reported fruit rot of *M. charantia* caused by *Alternaria tenuissima* (Kunze. ex Pers.), *Colletotrichum lagenarium* (Pers.) Ell. & Halst, *Fusarium oxysporum* Schlecht ex fr., *Pythium aphanidermatum* (Exd.) Fitzp. and *Synchytrium wurhii* Rytz.

From Pakistan Sultana and Gaffar (2007) reported 29 species of fungi namely *Alternaria alternata* (Fr.) Keisler, *A. tenuissima* Kunze ex Pers. *A. candidus* Link., *A. flavus* Link & Pers., *A. niger* Van Tiegh., *A. tamarii* Kita, Centr., *A. terreus* Thom, *A. wentii* Wehmer, *Chaetomium funicola* Cooke, *C. globosum* Kunze ex Fr., *C. murorum*, *C. olivaceum* Cook & Ellis, *Cladosporium cladosporioides* (Fr.) de Vries, *C. oxysporum* Schlecht.

Emend. Snyder & Han., *C. sphaerospermum* Penz., *Drechslera* state of *Cochliobolus spicifer* Nelson, *F. moniliforme*, *F. oxysporum*, *F. semitectum*, *F. oxysporum*. *Memnoniella echinata* (Riv.) Galloway, *M. verrucaria* (Alb. & Schw.) Ditm. ex Fr., *Nigrospora oryzae* (Berk & Br.) Petch, *Penicillium purpurogenum* Stoll, *Rizoctonia solani* Kuhn., *Rhizopus* sp., *Stachybotrys atra* Corda, *Stemphylium* sp., and *Trichurus spiralis* as seed borne pathogen of *M. charantia*.

The isolated fungal pathogens reported by Mukerji and Bhasin (1986) are not found in the present investigation in Bangladesh.

Sultana and Gaffar (2007) isolated 29 species of fungi associated with seeds of *M. charantia* among which two species are same as reported in the present investigation.

In the present investigation nine fungi were associated with two varieties of *M. charantia* of which five fungi were found to be pathogenic. This is the first report on association of *C. brachyspora*, *Fusarium* sp., *Mucor* sp., *Rhizopus stolonifer* and *Trichoderma viride* with *M. charantia* in storage. This finding will be helpful for designing effective control measures of *M. charantia* for post harvest handling and storage.

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