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Survey of Fungal Diseases of Some Vegetables and Fruits in Aswan, EGYPT

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Abstract*: Fifteen species belonging to 9 terrestrial fungal genera were isolated from diseased fruits and vegetables on PDA media during this investigation. Aspergillus came in high incidence genera and represented by three species namely; A. flavus var colamnaris, A. niger and A. ochraceus. Another four fungal genera were came in the second position after Aspergillus and represented by two identified species these were; Acremonium, Alternaria, Fusarium and Penicillium. The remaining four fungal genera which isolated were representative by only one species were; Botryotrichum sp., Gilmaniela humicola, Mucor hiemalis and Torula sp. Solanum lycopersicum was yielded the highest number of genera and species (7 and 11, respectively). Psidium guava was yield the lowest number of fungal genera and species (1 and 1). All fungal which isolated in this investigation were screened for their ability to cellulose production on CMC agar plates within 3 days, among all tested isolates Aspergillus flavus and Fusarium proliferatum were the highest fungal isolates produced clear zone (3.65 mm) and (3.15 mm) respectively.*

Key Words: *Aspergillus, Fruit, Psidium guava, Solanum lycopersicum, Vegetables*

I. Intriduction

Fungi are universally present in all types of natural habitats and from one of the most important component of an ecosystem as decomposers. It has been known that vegetables and fruits plays a vital role in human nutrition by supplying the necessary growth factors such as vitamins and essential minerals in human daily diet and that can help to keep a good and normal health. Vegetables and fruits are widely distributed in nature, one of the limiting factors that influence the vegetables and fruits economic value is the relatively short shelf-life period by pathogens attacked. It is estimated that about 20 – 25% of the harvested vegetables and fruits are decayed by pathogens during post – harvest handling even in developed countries [1-2]. Fungal vegetables and fruits infection may occur during the growing season, harvesting, handling, transport, post – harvest storage, marketing conditions and after purchasing by the consumer. Vegetables and fruits containing high levels of nutrient elements and sugars and their low pH values make them particularly desirable to fungal decaying [3].

Cellulose is commonly degraded by an enzyme called cellulase. This enzyme is produced by several microorganisms, mainly by bacteria and fungi. Although a large numbers of microorganisms are capable of degrading cellulose, only few of these produce significant quantities of cell free enzymes capable of completely hydrolyzing crystalline cellulose in vitro. Fungi are the main cellulose producing microorganisms, although a few bacteria and actinomycetes have also been reported to yield cellulose activity [4].

Several works have been dealt with fungi which recovered from many vegetable and fruits from different parts of the world [5 – 6 – 7 – 8 – 9 – 10 – 11 - 12]. The production of cellulases by fungi was studied by many investigators such as [13 – 14 – 15].

The Aim of this investigation were represents an extension of a series of investigations in Upper Egypt, it is represents a survey of the terrestrial fungi from diseased fruit and vegetables which were collected from different markets in Aswan area and screening of some fungi for cellulose activity.

II. Materials and Methods

1. Collection and Isolation of fungi from fruits and vegetables

The infected nine fruits viz. *Allium cepa, Solanum lycopersicum, Solanum tuberosum, Citrus aurantifolia, Cucurnis sativus, Cucurbite pepo, Fragraria grandiflora, Psidium guava* and *Citrus reticulata* were collected from markets in Aswan city. Samples were put in separate sterilized plastic bags and transferred to the laboratory. The infected tissues transferred individually to Petri dishes containing 20 ml of PDA medium. Incubate the dishes at 30°C for 5 days then dishes were examined and identified.

2. Pathogenicity test

Some of isolated fungi were used to confirm their pathogenicity in their respective hosts. Some fresh healthy samples were brought in to the laboratory and surface sterilized with 0.1% Hgcl2. For inoculations, cork borers of (2mm) diameter were sterilized for used. The inoculated samples and their respective controls were kept under sterile conditions at room temperature under bell jars. The artificially inoculated samples were

examined daily & the extent of damage was recorded. The pathogens were re-isolated and disease symptoms were clearly evident, the culture and symptoms signs were compared with original.

3. Screening of cellulolytic fungi.

The isolated fungal cultures were screened for their ability to produce cellulases complex following the method of [16]. Czapek-Dox medium used in this method contained (g/l); sucrose – 30, NaNO3 – 2, K2HPO4 - 1, MgSO4– 0.05, Kcl – 0.5, FeSO4 – 0.01, carboxy- methyl cellulose – 1%, Agar agar - 20. pH of the medium was adjusted to 5. After autoclaving the medium was poured into Petri plates and allowed to solidify. Cavities of 6 mm size were made in the solidified medium and inoculated with 0.1ml of spore suspension prepared from 7 day old slants. The plates were incubated at room temperature (28 ± 2 °C) for three days to allow fungal growth, then again incubated for 18 h at 50 °C which is the optimum temperature for cellulases activity. After incubation, 10 ml of 1% Congo - Red staining solution was added to the plates that were shaken 15 min. The Congo - Red staining solution was then discarded, 10 ml of 1N NaOH was added to the plates and shaken again for 15 minutes. Finally 1N NaOH was also discarded and the staining of the plates was analyzed by noticing the formation of yellow zones around the fungal spore inoculated wells.

4. Identification of the Fungal genera and species

The following references were used for the identification of the recovered fungi [17 – 18 – 19 – 20 – 21 – 22 – 23 – 24 – 25 – 26 – 27 – 28 – 29 – 30 – 31].

III. Results And Discussion

Fifteen species belonging to 9 terrestrial fungal genera were isolated from diseased fruits and vegetables on PDA media during this investigation (Table, 1).

Aspergillus came in the first position with high incidence as highest fungal genera and represented by three species namely; *A. flavus var colamnaris, A. niger* and *A. ochraceus*. The genus *Aspergillus* was recovered from *Allium cepa, Solanum lycopersicum, S. tuberosum, Citrus lemon, Cucurnis sativus* and *Cucrbite pepo. A. flavus var colamnaris* was isolated from 5 fruits and vegetables, whereas *A. niger* was recovered from *Solanum lycopersicum* only. *Aspergillus ochraceus* was isolated from *Solanum lycopersicum* and *Cucurnis sativus* only.

Four fungal genera were came in the second position after *Aspergillus* and represented by two identified species these were; *Acremonium, Alternaria, Fusarium* and *Penicillium. Acremonium,* which represented by *A. butyri* and *A. strichtum,* were recovered from *Solanum lycopersicum* and *Solanum tuberosum* only, while not recovered from the remaining investigated plants. *Alternaria* were isolated from *Solanum lycopersicum, Citrus aurantifolia, Cucurnis sativus, Cucurbit pepo, Fragaria grandiflora* while absent in the other tested plants, this genus was represented by *A. alternate and A. brassicicola. Fusarium* was recovered from *Allium cepa, Solanum lycopersicum* and *Solanum tuberosum* only, *Fusarium* was represented by two identified species were; *F. moniliforme* and *F. proliferatum. Penicillium (P. janthinellum* and *P. italicum) was isolated from Solanum lycopersicum, Psidium guava* and *Citrus reticulate.*

The remaining four fungal genera which isolated were representative by only one species were; *Botryotrichum sp., Gilmaniela humicola, Mucor hiemalis* and *Torula sp*. These genera were isolated from 3 to one investigated plants.

Similar results on post – harvest fungi on storage fruits and vegetables were reported by many investigators as [32 – 33 – 34 – 35 – 36 – 37 – 38].

Table (1): Fungal genera and species which were recovered from different fruits and vegetables samples collected from different markets in Aswan.

Fungal genera and species	fruits and vegetables samples								
	1	2	3	4	5	6	7	8	9
Acremonium:	-	+	+	-	-	-	-	-	-
A. Butyri	-	+	+	-	-	-	-	-	-
A. Strichtum	-	+	-	-	-	-	-	-	-
Alternaria:	-	+	-	+	+	+	+	-	-
A. Alternate	-	+	-	+	+	+	+	-	-
A. Brassicicola	-	-	-	-	+	+	-	-	-
Aspergillus:	+	+	+	+	+	+	-	-	-
A. flavus var colamnaris	+	+	+	+	-	+	-	-	-
A. niger	-	+	-	-	-	-	-	-	-
A. ochraceus	-	+	-	-	+	-	-	-	-
Botryotrichum sp.	-	+	+	-	-	-	+	-	-
Fusarium:	+	+	+	-	-	-	-	-	-
F. moniliforme	+	+	+	-	-	-	-	-	-
F. proliferatum	-	+	+	-	-	-	-	-	-

Gilmaniela humicola	-	-	-	+	-	-	-	-	+
Mucor hiemalis	-	-	+	-	-	-	-	-	-
Penicillium :	-	+	-	-	-	-	-	+	+
P. janthinellum	-	+	-	-	-	-	-	-	-
P. italicum	-	-	-	-	-	-	-	+	+
Torula sp.	-	+	-	-	-	-	-	-	-

1- Allium cepa 2- Solanum lycopersicum 3- Solanum tuberosum
4- Citrus aurantifolia 5- Cucurnis sativus 6- Cucurbit pepo
7- Fragaria grandiflora 8- Psidium guava 9- Citrus reticulate

Regarding to terrestrial fungal genera and species which recovered during this investigation (Fig. 1) showed that: *Solanum lycopersicum* was yielded the highest number of genera and species (7 and 11, respectively). *Psidium guava* was yield the lowest number of fungal genera and species (1 and 1). The remaining seven investigated fruits and vegetables were yield moderate numbers fungal genera and species during this investigation and ranging from (2 – 3 genera) and (2 – 4 species).

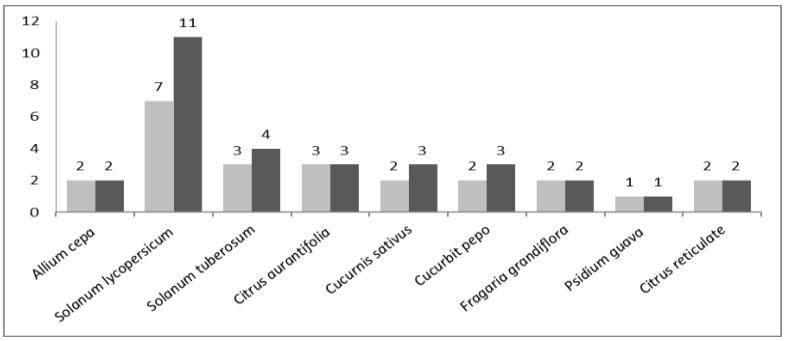

Fig (1): Numbers of fungal genera and species which were recovered from fruits and vegetables

Results in table (2) indicated that, all tested isolates were able to synthesize extracellular cellulose but in different levels. *Aspergillus flavus* and *Fusarium proliferum* were the highest investigated fungi produce clear zone of hydrolysis in CMC agar plates within 3 days. *Aspergillus flavus* was (3.65 mm) followed by *Fusarium proliferum* (3.15 mm); *Acremonium strictum* (1.9 mm); *Penicillium janthilinum* (1.65 mm); *Aspergillus ochraceus* (1.5 mm); *Alternaria alternata* (1.15 mm); *Fusarium moniliforme* (1.05 mm); *Botryotrichum sp.* (0.9 mm); *Acremonium butyri* (0.85 mm); *Torula sp*. (0.5 mm) and *Mucor hiemalis* (0.4 mm). Most of tested species for extracellular cellulose were previously examined in different part around the world by [39 – 40 – 41 – 42 – 43] when they isolated, screening, production and purification of cellulase by fungi.

Table (2): Biometric features of cellulolytic fungal cultures..

Tested fungal genera & species	Zone of hydrolysis on CMC agar plates (cm)
Penicillium janthilinum	1.65
Alternaria alternata	1.15
Acremonium butyri	0.85
Fusarium proliferum	3.15
Aspergillus ochraceus	1.5
Acremonium strictum	1.9
Botryotrichum sp.	0.9
Fusarium moniliforme	1.05
Aspergillus flavus	3.65
Mucor hiemalis	0.4
Torula species	0.5

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