### Journal of Basic and Environmental Sciences



Research Paper

ISSN Online:2356-6388 Print:2536-9202

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### **Detection of Toxigenic Fungi Associated Some Dried Fruits**

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### Abstract

Contamination of agricultural and food products by some fungal species that produce mycotoxins can result in unsafe food and feed. Since dried fruits are good sources of sugars and other nutrients, they are susceptible to mold contamination and consequent mycotoxin production. The occurrence of toxigenic fungi in dried apricots, figs, and grapes was surveyed in this study. Fungal frequency and their ability to produce mycotoxin were studied. The obtained results presented that, the isolation of fungi from dried Apricots, Figs, and Grapes yielded 308 fungal isolates. Total fungal count isolated from dried apricot fruit samples yielded 128 fungal isolates equal to 41.6% followed by dried grape samples which gave 106 fungal isolates (34.4%) and dried fig fruit samples resulted in 74 fungal isolates equal to 24.0%. On the other hand, five fungal genera were identified from dried apricot fruits as Alternaria, Aspergillus, Fusarium, Penicillium, and Rhizopus, where Aspergillus niger had the highest fungal frequency (50.78%). Four fungal genera were identified from dried fig fruits as Aspergillus, Fusarium, Penicillium, and Rhizopus. Higher fungal frequency was recorded with A. niger (29.73%), while six fungal genera were identified with dried grape samples as Alternaria, Aspergillus, Fusarium, Penicillium, Rhizopus, and Trichoderma, where A. niger had the highest fungal frequency (49.06%). Test of mycotoxins production presented that, ten A parasiticus isolates from dried apricot fruit samples were aflatoxins producers. Eight Aspergillus isolates (A. flavus and A. parasiticus) from dried figs fruit samples produced aflatoxins, and six A parasiticus isolates from dried grape samples were positive producer of aflatoxins, whereas all Aspergillus niger, Fusarium sp., and Penicillium sp. isolates from dried fruit samples were negative producer of mycotoxins. It could be concluded that toxigenic fungi can attack the dried fruits and cause their deterioration.

Keywords: Dried apricot, Dried fig, Dried grape, Fungi, Mycotoxins

### **1. Introduction**

Approximately 25% of global food crops, including many essential foods, are believed to be contaminated with mycotoxin, according to the Food and Agriculture Organization (FAO). A more recent study suggests that mycotoxins are found in 60-80% of agricultural products [1, 2]. Fruits may be affected either by dry or soft rot. Fungal diseases during transportation and storage lead to a loss of 25 to 30 percent of fruits [3]. Dried fruits provide dietary fiber, minerals, vitamins, antioxidants, and other beneficial bioactive compounds that contribute to human health. Additionally, they have a glycemic index that ranges from low to moderate [4]. Dried fruits are a popular option globally for people looking for a long-lasting alternative to fresh fruit, overcoming typical obstacles that hinder fruit consumption [5]. Dried fruits are a popular and nutritious snack enjoyed worldwide for their extended shelf life and concentrated nutrients. However, these fruits can become contaminated with various harmful fungal species at different stages of cultivation, harvesting, processing, drying, and storage. The primary mycotoxins found in dried fruits are aflatoxins (AFs) and ochratoxin A (OTA), which can lead to health issues and financial losses. The level of mycotoxin contamination in dried fruits can vary based on their geographic origin, including vine fruits, figs, dates, apricots, prunes, and mulberries. The supply chain for dried fruits involves multiple important stages like harvesting, washing, sorting, drying, processing, and storage. Dried fruits are particularly susceptible to invasion by xerophilic fungi, like Aspergillus flavus, which can

produce mycotoxins at water activity levels as low as 0.73 and 0.85, respectively [6]. Dried fruits are prone to mold growth and contamination, leading to the production of mycotoxins. Several studies have shown significant levels of mycotoxin contamination in these products, potentially resulting in substantial financial losses [7]. Currently, the consumption of dried fruits is prevalent globally. Raisins make up almost half of the dried fruits sold worldwide, with apricots, figs, dates, prunes (dried plums), peaches, apples, and pears following. The presence of toxigenic molds and mycotoxins on these dried fruits can pose challenges in the Mediterranean region and other parts of the world, representing a health risk for the population and a trade concern for the export of local products [8]. If a small area on the surface is affected by mold, it can spread rapidly in a brief period. Additionally, the quantity of infected fruit may escalate quickly if the drying process is not carried out correctly [8, 9]. The primary concerns associated with sundrying fruits include potential contact with the ground and the increased risk of insect and pathogen infestations when drying outside for extended periods [10]. Species belonging to the genera Aspergillus, Penicillium, and Alternaria are key contributors to fruit spoilage. These fungi can produce mycotoxins, leading to notable financial losses in various food industry processes, including drying [11]. The presence of aflatoxins in dried figs primarily results from contamination by Aspergillus species, specifically A. flavus and A. parasiticus [12, 13]. While several fungi can contaminate grapes during their growth in the vineyard, the primary

regarding mycotoxin concern contamination stems from the black Aspergilli, specifically A. carbonarius and A. niger [14]. Mycotoxins are toxic metabolites produced by molds that can harm humans, domestic animals, food, animal feeds, as well as wild birds, and raw materials [15]. Aflatoxigenic fungi can be present on fig fruits during the stages of growth, ripening, and drying, with their growth particularly thriving during the ripening and over-ripening phases. The occurrence of aflatoxins in dried figs is primarily a result of contamination by Aspergillus species, notably A. flavus and A. parasiticus. The key periods for aflatoxin production in dried figs begin with the ripening of figs on the tree, continuing through the overripe phase where they dehydrate, wrinkle, and fall onto the ground, and persist until they are fully dried on drying surfaces. A. flavus predominantly produces aflatoxin B1 (AFB1) and aflatoxin B2 (AFB2), while A. parasiticus generates AFB1, AFB2. aflatoxin G1 (AFG1), and aflatoxin G2 (AFG2) [16, 17]. This study aimed to assess naturally occurring fungal contamination in samples of dried apricots, figs, and grapes collected from three different localities. Isolate and identify all detected fungal colonies, calculate the percentage of total fungal isolates and their frequencies, as well as quantify detect and the produced mycotoxins.

### 2. Materials and Methods

### 2.1. Collected samples

Three samples of dried fruits ( Apricots, Figs, and Grapes ) were collected from three different locations in Egypt. Each sample weighed 1 kg and was stored in a clear polyethylene bag with relevant information. The samples were then taken to a lab and stored at  $-4^{\circ}C$  for further analysis.

### **2.2. Isolation and identification of the fungal association**

The collected samples were analyzed with and without surface disinfection. Surface disinfection was carried out by immersing the samples in 0.3% sodium hypochlorite for 2 minutes followed by washing with sterile distilled water [3, 18]. The fruit sample pieces were then transferred onto agar plates containing Potato Dextrose Agar medium with streptomycin to prevent bacterial growth. The plates were incubated at  $25 \pm 10$  for 5 days using the direct plating method. After the incubation period, the sliced fruits were examined and any molds that had grown were isolated and purified on a PDA medium [3]. The fungal isolates, which were 5 days old, were identified in the Plant Pathology Department at the National Research Centre based on their cultural and morphological characteristics using specific media and references such as Raper & Funel [19] for Aspergillus, Booth [20] and Nelson et al [21] for Fusarium, and Barnett & Hunter [22] for other genera of imperfect fungi. The developing fungi were then cultured on PDA slants and stored in a refrigerator for future use.

### 2.3. Fungal frequency

The total fungal count and fungal frequency percentage of naturally-occurring fungi were calculated as:

The fungal frequency

The number of isolates of species

 $= \frac{1}{\text{The total number of fungal isolates}} \times 100$ 

### 2.4. Test of Mycotoxins production

All isolates of toxigenic Aspergilli, Fusaria, and Penicillia (such as Aspergillus niger, flavus. Α. Α. parasiticus, Fusarium sp., and Penicillium sp.) were examined for their ability to produce mycotoxins. The isolates of Aspergillus niger, Α. flavus, Α. parasiticus, and Penicillium sp. were cultured in 100 mL yeast extract sucrose (YES) medium as pure cultures to test for the production of Aflatoxins and Ochratoxin A according to A.O.A.C. [23], Koteswara Rao et al., [24]; Munimbazi and Bullerman [25]. Aflatoxins were extracted and determined by HPLC according to Kumar et al. [26], and Rubert et al. [27]. Ochratoxin A was extracted and determined by Highperformance liquid chromatography (HPLC) according to Abarca et al. [28], and Bragulat et al. [29]. Fumonisin B1 production was carried out by cultivating *Fusarium* sp. on a corn medium according to Bailly et al. [30]. The extraction and quantification of Fumonisin B1 using HPLC were conducted following the protocol outlined by Le Bars et al. [31], and Ndube et al. [32].

### **3. Results and Discussion**

### **3.1. Percentage of total fungal count isolated from dried fruits**

Isolation from dried Apricots, Figs, and Grapes yielded 308 fungal isolates. Location C gave higher fungal isolates compared to others which recorded 118 fungal isolates equal to 38.31%, whereas location A was moderate recording 110 fungal isolates equal to 35.71%. Location B was less and gave 80 fungal isolates with 25.97 %. Less total fungal counts were recorded with all sterilized samples

for all three localities compared to unsterilized as shown in Table (1). On the other hand data in the same table indicated that the percentage of total fungal count isolated from dried apricot fruit samples yielded 128 fungal isolates equal to 41.56 %. Non-sterilized samples were the most total fungal isolates which record 75 isolates equal 24.35 % whereas 53 isolates from sterilized samples equal 17.21 %. Whereas, isolation from dried fig fruit samples resulted 74 fungal isolates equal 24.03 %. Out of them 47 fungal isolates (15.26 %) from unsterilized dried fig fruit samples and 27 fungal isolates (8.77 %) from sterilized dried fig fruit samples. Also, data show that, dried grape samples gives 106 fungal isolates equal 34.42 %. Non sterilized samples gives higher total fungal isolate which record 64 isolates 20.78 equal % whereas, sterilized samples were less total fungal isolate which records only 42 isolates equal 13.64 %. These findings are consistent with the research of Sanchis and Magan [6] and Karaca et al. [7], who have reported that dried fruits are vulnerable to colonization by xerophilic fungi, such as Aspergillus flavus. Aspergilli from the Flavi section have the ability to thrive and produce mycotoxins at water activity levels as low as 0.73 and 0.85, respectively. Additionally, Curbelo and Kabak [17] and Ozer et al. [8] have that fruits can become noted contaminated with various toxigenic fungal species at different stages including cultivation, harvesting, processing, drying, and storage. The susceptibility of dried fruits to fungal infections and mycotoxin production is attributed to their high sugar content, as well as the methods of harvesting and

drying used [8, 33, 34]. The primary issues associated with sun-drying fruits are the risk of contact with soil and exposure to insects and pathogens during prolonged outdoor drying periods. If a small area of the fruit's surface becomes moldy, the mold can proliferate rapidly. Additionally, the number of infected fruits may increase quickly if the drying process is not conducted properly [8, 9, 10].

Table (1): Percentage of total fu	ungal count associated	with dried fruit samples
	angui count associated	with allow if all sumples

	Type of dried fruit						
T.C	Apricot		Fi	g	Grape	(zibib)	T.C
%	Non	St	Non	St	Non	St	-
T. C	33	24	18	5	17	13	110
%	10.71	7.79	5.84	1.62	5.52	4.22	35.71
T. C	21	17	16	11	15	0	80
%	6.82	5.52	5.19	3.57	4.87	0.00	25.97
T. C	21	12	13	11	32	29	118
%	6.82	3.90	4.22	3.57	10.39	9.42	38.31
T. C	75	53	47	27	64	42	308
%	24.35	17.21	15.26	8.77	20.78	13.64	100
T. C 128		74	1	106		308	
%		.56	24.	03	34	.42	100
	%           T. C           %           T. C           %           T. C           %           T. C           %	%         Non           T. C         33           %         10.71           T. C         21           %         6.82           T. C         75           %         24.35           C         12	T. CApricot%NonStT. C3324%10.717.79T. C2117%6.825.52T. C2112%6.823.90T. C7553%24.3517.21	T. CApricotFig $\%$ NonStNonT. C332418 $\%$ 10.717.795.84T. C211716 $\%$ 6.825.525.19T. C211213 $\%$ 6.823.904.22T. C755347 $\%$ 24.3517.2115.26C12874	T. CApricotFig $\%$ NonStNonStT. C3324185 $\%$ 10.717.795.841.62T. C21171611 $\%$ 6.825.525.193.57T. C21121311 $\%$ 6.823.904.223.57T. C75534727 $\%$ 24.3517.2115.268.77C1287474	T. CApricotFigGrape $\%$ NonStNonStNonT. C332418517 $\%$ 10.717.795.841.625.52T. C2117161115 $\%$ 6.825.525.193.574.87T. C2112131132 $\%$ 6.823.904.223.5710.39T. C7553472764 $\%$ 24.3517.2115.268.7720.78C128741616	T. CApricotFigGrape (zibib) $\%$ NonStNonStNonStT. C33241851713 $\%$ 10.717.795.841.625.524.22T. C21171611150 $\%$ 6.825.525.193.574.870.00T. C211213113229 $\%$ 6.823.904.223.5710.399.42T. C755347276442 $\%$ 24.3517.2115.268.7720.7813.64C12874106106106

T. C= Total Count

## **3.2.** Fungal frequencies isolated from dried apricot fruit samples in different localities

Fungal frequency associated with dried apricot fruit samples collected from three different localities were tabulated in Table (2). Data presented that seven fungal species under five fungal genera were identified. These are Alternaria, Asperigillus (A. flavus, A. parasiticus, and A. niger), Fusarium, Penicillium, and Rhizopus. Aspergillus niger was the most fungal frequency which recorded 65 isolates equal to 50.78%, followed by both Aspergillus parasiticus and Fusarium sp. with 19 isolates equal to 14.84% and Rhizopus stolnifer with 13 isolates recording 10.16%. Alternaria alternate was moderate fungal frequency and gave 6 isolates equal to 4.69 percent.

Whereas, Penicillium records 5 isolates with 3.91%. The less fungal frequency with Aspergillus flavus which recorded only one isolate equal 0.78%. These align with findings the research conducted by Heperkan [35], Ozer et al. [8], Samson et al. [36], and Serra et al. [37], who have observed that, within the Aspergillus species, those classified under the Nigri and Flavi sections are commonly identified in dried apricots, dates, prunes, and other dried fruits. The Nigri section of Aspergillus, also known as black Aspergilli, includes Aspergillus carbonarius and various members of the Aspergillus niger aggregate. Embaby et al. [38] isolated Aspergillus niger and A. parasiticus from decaying apricot fruits. Also, Ozer et al. [8] and Trucksess and have highlighted Scott [33] that

Aspergillus flavus and A. parasiticus are the predominant species contaminating food items, including dried fruits, due to their ability to produce aflatoxins. Moreover, Al Ghamdi et al. [39] isolated and identified seven different fungal genera and 13 species from forty samples of dried fruits using the direct plating method on Potato Dextrose Agar (PDA) medium, with Aspergillus (80.60%),Rhizopus (13.58%),and Penicillium (3.30%) being the most prevalent genera. Saadullah and Abdullah [34] have documented the isolation of 20 Aspergillus species from four varieties of dried fruits (apricot, fig, grapes, and plum), where Aspergillus niger, A. flavus, A. parasiticus, A. carbonarius, A. fumigatus, A. japonicas, A. awamori, A. ochraceus, and A. tubingensis were detected in all types of dried fruits.

 Table (2): Fungal frequencies isolated from dried apricot fruit samples in different localities

Fungi	Non Sterilized			S	Sterilized	Т. С.	%	
8-	Α	В	С	A	В	С		
Alternaria alternata	0	0	0	5	0	1	6	4.69
Aspergillus niger	15	10	13	10	9	8	65	50.78
Aspergillus flavus	0	0	1	0	0	0	1	0.78
Aspergillus parasiticus	3	4	5	4	3	0	19	14.84
<i>Fusarium</i> sp.	10	1	0	5	1	2	19	14.84
Penicillium sp.	3	0	1	0	0	1	5	3.91
Rhizopus stolnifer	2	6	1	0	4	0	13	10.16
Total	33	21	21	24	17	12		
%	25.78	16.40	16.41	18.75	13.28	9.38		
Total count for sterilization		75			53		128	100
%		58.59			41.41			

# **3.3. Fungal frequencies isolated from dried fig fruit samples in different localities**

Isolation from dried fig fruits collected from three different localities resulted in 74 fungal isolates. Out of them, 47 isolates equal 63.51 % from nonsterilized fig fruit samples, and 27 isolates from sterilized fig fruit samples equal 36.49 % as shown in **Table (3)**. Locations B and C were the most infected by fungi followed by location A in sterilized isolates. In non-sterilized dry fig samples, location A had higher fungal frequency followed by B and C respectively. On the other hand, data presented that four fungal genera belonging to five species were identified as Aspergillus (Aspergillus flavus, A. parasiticus & A. niger), Fusarium sp., Penicillium sp., and Rhizopus stolnifer. Higher fungal frequency was recorded with Aspergillus niger which record 22 isolates equal 29.73% followed by Fusarium sp. which gives 18 isolates equal 24.32%. Each of A. parasiticus and Rhizopus stolnifer had moderate fungal frequency and gave 10 isolates equal to 13.51 percent followed by A. flavus

which recorded 8 isolates (10.81%), while Penicillium sp., which gave 6 isolates (8.12%). These results are in agreement with those obtained by Alghalibi Shater, and [40] who identified twenty-three species and one variety from fifteen genera in dried fruits, including raisins, dates, and figs. The most commonly isolated fungal species were Aspergillus niger, A. flavus, A. fumigatus, A. ochraceus, Penicillium chrysogenum, and Rhizopus stolonifer. Studies by **Basegmez and Heperkan** Heperkan et al. [12] [41], and Trucksess and Scott [33] have shown that the predominant fungal genera found in Mediterranean dried figs are Aspergillus, Fusarium, and Penicillium. Saadullah and Abdullah [34] reported that the most frequently isolated fungal species from dried figs were A. niger, followed by A. flavus, A. carbonarius, and A. parasiticus with respective

frequencies of 76.65%, percentage 66.6%, 33.1%, and 33.1%. Additionally, Embaby et al. [42] noted that, the common postharvest and storage fungi of fig fruits include Aspergillus spp. (A. flavus, A. fumigatus, A. niger), Fusarium spp., Alternaria alternata, Geotrichum sp., Penicillium spp., Rhizopus nigricans, *R. stolonifer*, and *Sclerotinia sclerotorum*. Pushparaj et al. [43] highlighted that among various fruits, figs are at high risk contamination by mycotoxigenic of species, specifically Aspergillus nigri, Fusarium sp., A. flavi, and Penicillium sp. Despite surface disinfection reducing the viable mold count in dried fruits, internal mold invasion remains a considerable concern. The prevalence of Aspergillus section Nigri in dried fruits is attributed to the ability of these fungi to withstand the drying process due to the black spores' resistance to sunlight and UV radiation [44].

Fungi	Noi	Non Sterilized			Sterilized	T.C	%	
i ungi	Α	В	С	Α	В	С		/0
Aspergillus niger	6	5	3	0	4	4	22	29.73
Aspergillus flavus	1	3	2	1	0	1	8	10.81
Aspergillus parasiticus	2	2	3	2	1	0	10	13.51
Fusarium sp.	1	4	5	0	4	4	18	24.32
Penicillium sp.	0	0	0	2	2	2	6	8.12
Rhizopus stolnifer	8	2	0	0	0	0	10	13.51
Total	18	16	13	5	11	11		
%	24.32	21.62	17.57	6.76	14.86	14.86		100
T. C		47			27		74	100
%		63.51			36.49			

Table (3): Fungal frequencies isolated from dried fig fruit samples in different localities

identified Aspergillus flavus, A. niger,

### 3.4. Fungal frequencies isolated from dried grape samples in different localities

Fungal frequency associated with dried grape samples collected from three different localities were tabulated in Table (4). Data presented that eight fungal species under six fungal genera were identified. These are Alternaria alternate, Aspergillus (A. flavus, A. parasiticus, and A. niger), Fusarium sp., Penicillium sp., Rhizopus stolnifer, and Trichoderma sp. Aspergillus niger was most fungal frequency which the recorded 52 isolates equal to 49.06%, followed by Penicillium sp., with 25 isolates equal to 23.58%, and Rhizopus stolonifer with 12 isolates recording 11.32%. Fusarium sp. had moderate fungal frequency and gave 8 isolates to 7.55 percent. Aspergillus equal parasiticus recorded 6 isolates with 5.66 percent. Less fungal frequencies were recorded with *Alternaria* alternate. Aspergillus flavus, and Trichoderma harzianum, all of them gave only one isolate equal to 0.94%. These findings align with the research conducted by Alghalibi and Shater [40], who isolated twenty-three species and one variety from fifteen genera in dried fruits such as raisins, dates, and figs using two types of media. The most commonly isolated fungal species included Aspergillus niger, A. flavus, A. fumigatus, A. ochraceus, Penicillium chrysogenum, and Rhizopus stolonifer. In addition, Alisa et al. [14] reported isolating 812 fungal isolates, including Aspergillus spp., B. cinerea, Alternaria Penicillium spp., spp., Cladosporium spp., Sphaeropsis spp., Trichoderma Rhizopus spp., spp., *Epicoccum* spp., and *Fusarium* spp., from grape samples. Sekar et al. [15]

Rhizopus sp., Mucor sp., and Penicillium sp. from diverse samples of figs, maize, dates, and grapes. Furthermore, Embaby et al. [45] isolated 60 fungal isolates from dried grape samples, representing five fungal genera known as Alternaria, Aspergillus, Botrytis, Mucor, and Rhizopus. Khashaba al. et [46] conducted an isolation of 102 fungal isolates from 30 dried grape samples, representing ten fungal genera, which included Alternaria, Aspergillus, Cladosporium, Curvularia, Emericella, Fusarium, Mocur, Penicillium, Rhizopus, and Trichoderma. Among these genera, Aspergillus was identified as the most predominant, followed by Penicillium, Alternaria, and Fusarium. Additionally, studies by Frisvad et al. [47] and Kizis et al. [48] highlighted the identification of various Penicillium species such as P. expansum, P. chrysogenum, and P. nordicum in grapes. Habib et al. [49] pointed out that grapes are susceptible to numerous fungal diseases both in the field and during storage, with Aspergillus, Penicillium, and Alternaria capable of causing decay and mycotoxin contamination. The epiphytic populations primarily consisted of wound pathogens, including Aspergillus spp. and Penicillium spp. In a study by Mohamed et al. [2], 119 fungal isolates were forty-nine obtained from samples encompassing eight grape varieties. The Penicillium and Aspergillus genera were present in all grape variety samples, with the most heavily infected grape varieties. Among the 25 Aspergillus strains identified, three strains (two A. flavus and one A. parasiticus) were capable of producing Aflatoxin during the veraison and maturity stages. Also, Bulut et al.

[18] noted that the most prevalent species isolated from dried fruits like grapes, cherries, and cranberries was *Aspergillus niger*. These variations in fungal species may be attributed to climatic conditions such as rainfall and temperature.

Temperature and humidity are crucial factors influencing the growth of fungal pathogens in fruits and vegetables, ultimately impacting the occurrence and severity of fungal diseases in plants **[50]**.

Table (4): Fungal frequencies isolated from dried	l grape samples in different localities
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Funci	No	on- Steril	ize	Sterilize				%
Fungi	A	В	С	Α	В	С	T.C	70
Alternaria alternata	1	0	0	0	0	0	1	0.94
Aspergillus niger	10	9	11	10	0	12	52	49.06
Aspergillus flavus	0	0	1	0	0	0	1	0.94
Aspergillus parasiticus	2	0	1	2	0	1	6	5.66
<i>Fusarium</i> sp.	0	3	1	0	0	4	8	7.55
Penicillium sp.	3	3	17	1	0	1	25	23.58
Rhizopus stolnifer	0	0	1	0	0	11	12	11.32
Trichoderma harzianum	1	0	0	0	0	0	1	0.94
Total	17	15	32	13	0	29		
%	16.04	14.15	30.19	12.26	0	27.36		
T. C		64			42		106	100
%		60.38			39.62			

#### 3.5. Test of mycotoxin production

Fruits are highly susceptible to infection by toxic molds and microorganisms. So, some toxigenic fungi were tested for mycotoxin production as Aflatoxins, Fumonisin B1, and Ochratoxin A (OTA). Data were recorded in Table (5). Data isolates presented that. some of Aspergillus were positive producers of aflatoxins while other isolates were negative producers. The most producing aflatoxins were detected with some isolates of Aspergillus fluvs and A. parasiticus. whereas all Aspergillus niger, Fusarium sp., and Penicillium sp. isolates

from dried fruit samples were negative producers of mycotoxins. According to Barkai-Golan and Paster [51], a variety of Aspergillus species can produce harmful mycotoxins that pose risks to both humans and animals. The primary mycotoxins linked to Aspergillus species present in fruits and vegetables include aflatoxins, primarily generated by strains of A. flavus and A. parasiticus, as well as ochratoxin A (OTA), produced by A. carbonarius and other ochratoxigenic aspergilla. Khashaba al. et **[46]** conducted a study examining ten different isolates of filamentous fungi obtained from diverse grape products, with eight

isolates belonging to Aspergillus and two isolates to Penicillium, to evaluate their potential for mycotoxin production. All the Aspergillus isolates tested, except A. *niger*, displayed the ability to produce mycotoxins, while the A. niger isolates did not exhibit mycotoxin production. Two Aspergillus flavus isolates from dried and fresh grape samples were found to produce aflatoxin B1 and aflatoxins B1 plus B2, respectively, whereas Α. parasiticus isolated from fresh grape samples produced aflatoxins B1, B2, G1,

Al Ghamdi et al. [39] and G2. discovered that only A. flavus and A. parasiticus were toxigenic fungi capable of aflatoxin production. Abbas et al. [52] reported that 14% of the Aspergillus flavus strains isolated from dried apricots and almonds demonstrated aflatoxigenic activity. The production of mycotoxins can be impacted by a range of factors, such as the virulence of the strain and its capability withstand different to environmental nutritional and circumstances [53].

				Туре	of drie	d fruit			
Tested toxigenic fungi	Apricot			Fig			Grape (zibib)		
	AFs	FB <sub>1</sub>	OTA	AFs	FB <sub>1</sub>	OTA	AFs	FB <sub>1</sub>	OTA
Aspergillus niger	-	-	ND	-	-	ND	-	_	ND
Aspergillus flavus	+	-	-	+	-	-	+	-	-
Aspergillus parasiticus	+	-	-	+	-	-	+	-	-
Fusarium sp.	-	ND	-	-	ND	-	-	ND	-
Penicillium sp.	-	-	ND	-	-	ND	-	-	ND

#### Table (5): Test of Mycotoxin production

ND = Not detected, AFs = Aflatoxin,  $FB_1 = Fumonisin B_1$ , OTA = Ochratoxin A

# **3.5.1.** Determination of mycotoxins production by toxigenic fungi isolated from dried apricot fruit samples

Data in **Table (6)** and **Fig. 1 (a-k)** presented that ten *A parasiticus* isolates from dried apricot fruit samples were aflatoxins producers. Higher concentration was found in *A. parasiticus* isolate No. 5 with 743.53 ng/ml **Fig. 1 (f)**, followed by *A. parasiticus* isolate No. 8 with a concentration of 29.84 ng/ml. **Fig. 1 (i)**. *A. parasiticus* isolate No. 7 was found to produce AFB<sub>1</sub>, AFB<sub>2</sub>, AFG<sub>1</sub> and AFG<sub>2</sub> which produce 9.34 ng/ml **Fig. 1** (**h**), followed by *A. parasiticus* isolate

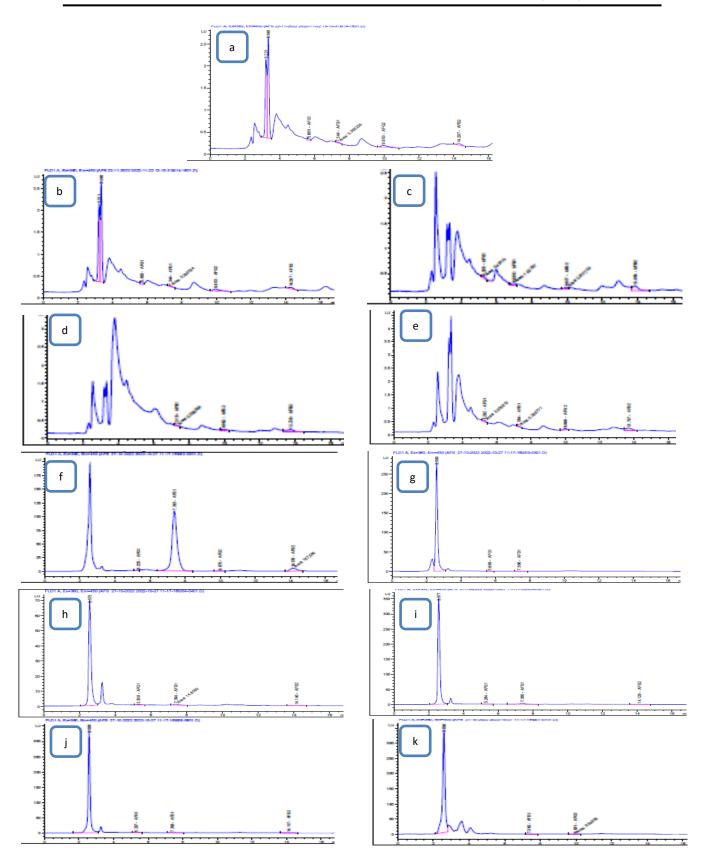
No. 9 with concentration 6.80 ng/ml Fig. 1 (j) and A. parasiticus isolate No.10 with 3.87 ng/ml Fig. 1 (k) and A. parasiticus isolate No.2 with 0.81 ng/ml Fig. 1 (c) and A. parasiticus isolate No. 4 with 0.71 Fig. 1 (e) followed by A. parasiticus isolate No. 6 with 0.69 ng/ml, Fig. (g) and A. parasiticus isolate No.1 with 0.66 ng/ml Fig. 1 (b). Less aflatoxins were produced by A. parasiticus isolate No. 3 which gave only 0.59 ng/ml Fig. 1 (d). Similar findings were reported by Celik and Ozturk [54], Ozer et al. [8] and Zohri and Abdel-Gawad [55] in their analysis of dried apricot samples that had been dried on soil and tarp to determine

the presence of aflatoxins and OTA. These studies revealed contamination of samples with Aflatoxin B1 (AFB1) and Aflatoxin G1 (AFG1) ranging between 0.10-1.47 µg kg-1 and 0.35-1.27 µg kg-1. respectively. Additionally. OTA contamination was noted in dried apricots within the range of 50-110 µg kg-1. Jianlan et al. [56] observed similar results, reporting that three isolates of A. flavus from apricot fruit rots produced aflatoxin B1 and B2, while one A. parasiticus isolate produced B1, B2, G1, and G2. Embaby et al. [38] also that isolates documented two of Aspergillus parasiticus from rotted apricot fruits were capable of producing

one or more aflatoxins, namely B, B, G, and G. Also Peter and Jones [57] detected aflatoxins in a single dried apricot sample (10%), but the levels detected were minimal (total aflatoxins less than 1 µg/kg). Hosseini and Bagheri [58] and Luttfullah and Hussain [59] highlighted contamination levels of aflatoxins in various dried fruit samples, with dried apricots showing a 5% contamination rate, dried figs at 30%, raisins at 10%, and apricot kernels at 6%. Furthermore, Abbas et al. [52] found that 14% of the total Aspergillus flavus strains isolated from dried apricots and almonds were aflatoxigenic.

	e (6): Determination I dried apricot same	v	production by toxigenic fungi isolated from
No	Type of fungi	Type of	Mycotoxin conc. (ng/ml)

N	T-m o of from at	Type of		Mycoto	oxin conc.	(ng/ml)	
No.	Type of fungi	Mycotoxin	AFB1	AFG1	AFB2	AFG2	Total
STD		Aflatoxins	40	40	12	12	104
1	A. parasiticus	AFs	0.05	0.18	0.10	0.33	0.66
2	A. parasiticus	AFs	0.17	0.16	0.33	0.15	0.81
3	A. parasiticus	AFs	0.05	0.00	0.31	0.23	0.59
4	A. parasiticus	AFs	0.05	0.17	0.34	0.15	0.71
5	A. parasiticus	AFs	692.02	4.77	44.26	2.48	743.53
6	A. parasiticus	AFs	0.39	0.30	0.00	0.00	0.69
7	A. parasiticus	AFs	4.54	4.23	0.57	0.00	9.34
8	A. parasiticus	AFs	24.17	4.23	1.44	0.00	29.84
9	A. parasiticus	AFs	3.83	2.54	0.43	0.00	6.80
10	A. parasiticus	AFs	0.38	0.00	0.00	3.49	3.87



**Figure 1. a-** HPLC chromatograms of standard aflatoxins AFG<sub>1</sub>, B<sub>1</sub>, G<sub>2</sub>& B<sub>2</sub>, HPLC chromatograms of Aflatoxin produced by **b-** *A. parasiticus* No.1, **c-** *A. parasiticus* No.2. **d-** *A. parasiticus* No.3, **e-** *A. parasiticus* No.4, **f-** *A. parasiticus* No.5, **g-** *A. parasiticus* No.6, **h-** *A. parasiticus* No.7, **i-** *A. parasiticus* No.8, **j-** *A. parasiticus* No.9, **k-** *A. parasiticus* No.10.

# **3.5.2.** Determination of mycotoxins production by toxigenic fungi isolated from dried figs fruit samples

Data in Table (7) and Fig. 2 (a-h) presented that Eight Aspergillus isolates (A. flavus and A. parasiticus) from dried figs fruit samples produced aflatoxins. A higher concentration was found with Aspergillus flavus No. 1 which gave 281.89 ng/ml Fig. 2 (a), followed by Aspergillus parasiticus isolate No. 5 with a concentration of 37.91 ng/ml Fig.2 (e). Aspergillus parasiticus isolate No.8 was found to produce 8.50 ng/ml of aflatoxin Fig.2 (h). A. parasiticus isolate No. 6 sample produced 2.61 ng/ml Fig.2 (f), followed by Aspergillus flavus isolate No. 3 with concentration 1.66 Fig.2 (c) and Aspergillus flavus isolate No.4 with 1.06 ng/ml Fig.2 (d) and Aspergillus flavus isolate No.2 with 0.69 ng/ml Fig.2 (b). Aspergillus parasiticus isolate No.7 was less producer which gave 0.56 only ng/ml Fig.2 (g). Similar findings were reported by Ozay and Alperden [60], who noted that Aflatoxin (AFB1, AFB2, AFG1, and AFG2) was detected in 29% of the examined contaminated dried fig samples. Jones et al. [61] stated that Aflatoxins are commonly found in dried figs and raisins globally, with significant levels reported up to 550 µg/kg and 63 µg/kg, respectively. Peter and Jones [57] and Stanton [62] identified Aflatoxins in 30% (3/10) of the analyzed dried fig samples, with the highest total aflatoxin concentration recorded at 6.7 µg/kg. Ioannou-Kakouri et al. [63] indicated that sixty-one percent of Turkish dried fig samples showed contamination by Aspergillus and Penicillium strains and

contained aflatoxins, with levels ranging up to 340 ng g-1 (14%). Some samples of the local fig crop were found to be contaminated with aflatoxin, but the detected levels (up to 5 µg kg-1) were within the permitted maximum levels (10 µg kg-1 total aflatoxins, with AFB1 not exceeding 5  $\mu$ g kg-1). Lewis and Goodrich [64] and Shenasi et al. [65] highlighted that aflatoxin contamination in individual figs had exceeded 600  $\mu g/kg-1$  in 83% of figs infected by A. parasiticus and in 38% from A. flavus colonization, with contamination levels ranging from 35 to 11,610 µg/ kg-1. According to Ashiq, [66], Ghazanfari et al. [67] and Zulfigar et al. [68], dried apricots, figs, plums, dates, and dried nuts are particularly susceptible to fungal infections mycotoxins. and These preserved fruits are at risk of mycotoxin exposure during the drying process in trays and while drying on trees. Alghalibi and Shater [40] identified two samples that were naturally of dried figs contaminated with aflatoxin B1, with aflatoxin concentrations ranging from 120 to 250 µg/kg in dried figs. Embaby et al. [38] observed that some isolates of Aspergillus from dried figs were capable of producing aflatoxins. Senvuva et al. [69] detected Aflatoxin B1 in 49 fig samples, with levels ranging from 0.7 to 222 ng g-1, and 40 individual figs containing more than 2 ng g-1. Additionally, Heperkan et al. [12] concluded that the formation of aflatoxins in dried figs is primarily attributed to contamination by Aspergillus species, particularly A. flavus and A. parasiticus.

Table (7): Determination of mycotoxins production by toxigenic fungi isolated from dried fig samples

NI-	T	Type of	Mycotoxin conc. (ng/ml)						
No.	Type of fungi	Mycotoxin	AFB1	AFG1	AFB2	AFG2	Total		
STD	-	Aflatoxins	40	40	12	12	104		
1	Aspergillus flavus	AFs	263.52	0.00	18.37	0.00	281.89		
2	Aspergillus flavus	AFs	0.69	0.00	0.00	0.00	0.69		
3	Aspergillus flavus	AFs	0.91	0.00	0.75	0.00	1.66		
4	Aspergillus flavus	AFs	0.93	0.00	0.13	0.00	1.06		
5	Aspergillus parasiticus	AFs	25.85	11.33	0.73	0.00	37.91		
6	Aspergillus parasiticus	AFs	0.43	0.00	0.29	1.89	2.61		
7	Aspergillus parasiticus	AFs	0.45	0.11	0.00	0.00	0.56		
8	Aspergillus parasiticus	AFs	6.34	1.01	1.15	0.00	8.50		

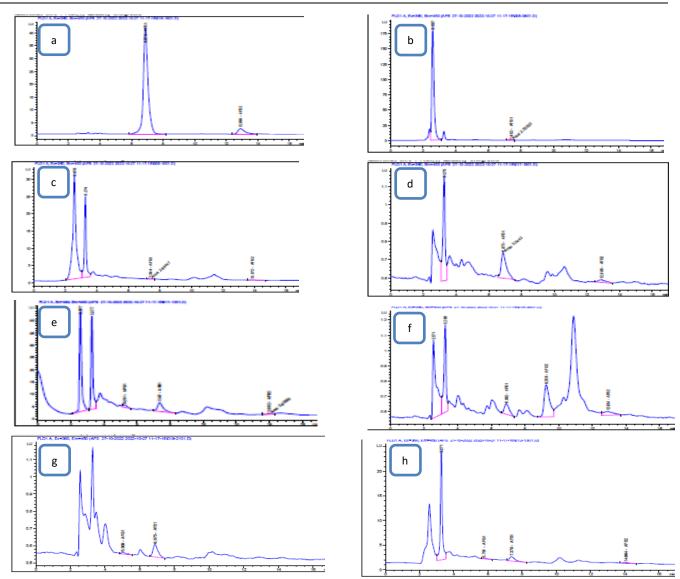


Figure 2. HPLC chromatograms of Aflatoxin produced by a- *Aspergillus flavus* No. 1. b- *A. flavus* No. 2., c- *A. flavus* No. 3., d- *A. flavus* No. 4., e- *A. parasiticus* No. 5., f- *A. parasiticus* No.6, g- *A. parasiticus* No.7, h- *A. parasiticus* No.8.

### **3.5.3. Determination of mycotoxins** production by toxigenic fungi isolated from dried grape samples

Data in Table (8) and Fig. 3 (a-f) presented that six A parasiticus isolates from dried grape samples were positive producers of aflatoxins. A higher concentration was found in Aspergillus parasiticus sample No. 4 with 472.94 ng/ml Fig.3 (d), followed by Aspergillus parasiticus sample No. 5 with a concentration of 82.54 ng/ml Fig.3 (e). Aspergillus parasiticus sample No.1 was found to produce 46.63 ng/ml of aflatoxin AFB<sub>1</sub> Fig.3 (a). Aspergillus parasiticus No. 3 sample was found to produce AFB<sub>1</sub>, AFB<sub>2</sub>, AFG<sub>1</sub>, and AFG<sub>2</sub> which produce 34.67 ng/ml Fig.3 (c), followed by Aspergillus parasiticus No. 6 with a concentration of 23.97 ng/ml Fig.3 (f). Less aflatoxins quantity were produced by Aspergillus parasiticus No.2 which recorded only 13.63 ng/ml Fig.3 (b). These findings align with those of Jones et al. [61], who highlighted that Aflatoxins are most commonly reported in dried raisins and figs on a global scale, with notable levels reaching up to 550 μg/kg and 63 μg/kg, respectively. Özay and Alperden [70] examined 103 samples from various orchards and stages of grape processing, including dried grapes, and revealed the presence of Aflatoxins B1, B2, G1, and G2 in 29% of

the samples, while Ochratoxin A was detected in only 3% of the samples. Li et al. [71] indicated that the formation of mycotoxins in dried grapes primarily stems from contamination by Aspergillus species, particularly A. flavus and A. parasiticus. Additionally, Khashaba et al. [46] noted that all tested Aspergillus isolates from different grape products, excluding A. niger, exhibited mycotoxinproducing capabilities. Specifically, two Aspergillus flavus isolates from dried and fresh grape samples produced aflatoxin aflatoxins **B**1 and **B**1 plus B2, respectively, while A. parasiticus isolated from fresh grape samples generated aflatoxins B1, B2, G1. and G2. Furthermore, they found seven out of ten dried grape samples were naturally contaminated with ochratoxin A and aflatoxins B1, B2, and G1. Furthermore, Kizis et al. [48], Lorenzini et al. [72], and Susca et al. [73], determined that the primary factors leading to grape contamination with mycotoxins are fungi from the saprophytic genus Aspergillus, notably species within the Aspergillus section Nigri, and Penicillium. Various Aspergillus species, such as A. flavus, A. parasiticus, and A. nominus, are recognized as significant producers of aflatoxins in various food items, including grapes.

No.	Type of fungi	Type of	Mycotoxin conc. (ng/ml)					
190.	Type of fungi	Mycotoxin	AFB1	AFG1	AFB2	AFG2	Total	
STD	-	Aflatoxins	40	40	12	12	104	
1	Aspergillus parasiticus	AFs	38.96	7.21	0.46	0.00	46.63	
2	Aspergillus parasiticus	AFs	11.60	0.33	1.56	0.14	13.63	
3	Aspergillus parasiticus	AFs	32.95	0.11	1.61	0.00	34.67	
4	Aspergillus parasiticus	AFs	451.40	5.03	16.51	0.00	472.94	
5	Aspergillus parasiticus	AFs	19.17	8.89	43.16	11.32	82.54	
6	Aspergillus parasiticus	AFs	4.78	10.73	6.77	1.69	23.97	

 Table (8): Determination of mycotoxins production by toxigenic fungi isolated from

 dried grape samples

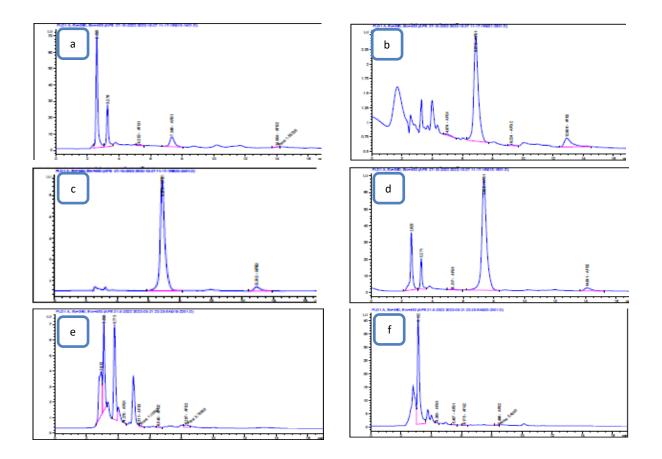


Figure 3. HPLC chromatograms of Aflatoxin produced by a- Aspergillus parasiticus No. 1. b- A. parasiticus No. 2.,
c- A. parasiticus No. 3.. d- A. parasiticus No. 4., e- A. parasiticus No. 5., f- A. parasiticus No.6.

### 4. Conclusion

Fruits, due to their high sugar and nutrient content, provide an optimal environment for microbial growth, including mold. Some of these molds have the potential to produce mycotoxins. The analysis of tested dried fruit samples in this study revealed contamination by toxigenic fungi like *A. flavus* and *A. parasiticus*. The principal mycotoxins found in dried apricots, figs, and grapes, namely AFs

(B1, B2, G1, and G2), are primarily produced by fungi from the Aspergillus genus. These fungi have the potential to pose health risks to consumers. Therefore, all fruits need to undergo more stringent hygiene processes to ensure their suitability for human consumption.

Acknowledgments The authors wish to thank to Botany and Microbiology Department, Faculty of Science, Benha University, and Plant Pathology Dept., as well as the Food Toxicology and Contamination Dept. in the National Research Center (NRC), Egypt for their help and encouragement during this study.

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