

NUTRITIONAL VALUE AND NATURAL OCCURRENCE OF MYCOBIOTA AND AFLATOXINS IN WHEAT and maize GRAINS

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ABSTRACT

The results of this study revealed that the values of moisture contents significantly increased while ash and carbohydrate were significantly decreased with storage. The change in the crude protein, fat and fiber contents of wheat grain samples were not significantly affected. *Alternaria alternata* (45.3% of the total propagules) followed by *Aspergillus* (13.3%) with *A. flavus* and *A. niger* (5.42% each) are the most common in freshly harvested wheat grains. Rarely occurred *aspergilla* include *A. candidus*, *A. fumigatus* and *A. ochraceus*. *Rhizopus* (1 species, 16.26%) and *Mucor* (2, 11.82%) followed by *Penicillium* (1, 4.93%) and *Fusarium* (3, 3.45%) were also dominant. On the other hand, *Aspergillus*, *Eurotium* and *Penicillium* dominated in stored grains on almost both isolation media. *A. flavus*, *A. niger*, *E. chevalieri*, *E. repens*, *E. rubrum*, *P. aurantiogriseum*, *P. chrysogenum* and *P. expansum* were the most common species. With respect to maize grains, *A. niger* followed by *A. flavus* were common on both freshly harvested and stored kernels, while *Eurotium* represented by *E. chevalieri* and *E. rubrum* on stored maize only. Total aflatoxin level in all local and imported samples tested was $\leq 10 \mu\text{g kg}^{-1}$ using both analytical methods (HPLC and Kits) in both freshly and stored wheat and maize grain samples which is below the permissible limit.

Keywords: Wheat, maize, aflatoxins, mycotoxins, fungi, nutritional values, storage.

INTRODUCTION

Wheat is one of the world's most important food crops. Foods made with wheat are a major part of the diet for over a third of the world's people. Wheat can be found in some form at almost every meal such as: bread, cookie, cake, macaroni and spaghetti (Dastmalchi 2008).

Colonization of cereal crops prior to harvest in the field (preharvest) by aflatoxigenic fungi often resulted in spoilage and aflatoxin accumulation in postharvest grains during storage (Resnik *et al.*, 1996, Nesci *et al.*, 2003). These fungi survive in a wide range of environments and can be found in soil, in plant and animal remains, and in grains and seeds such as maize, peanuts (Pitt 2000). These fungi are responsible for spoilage of stored grains around the world (Paster 1995). They contaminate various agriculturally important commodities including cereals and cereal products.

The Food and Agriculture Organization of the United Nations (FAO) estimated that at least 25% of the world's cereal grains are contaminated by mycotoxins, including aflatoxins (Dowling, 1997). Aflatoxins (AF) are highly toxic, mutagenic, carcinogenic and teratogenic metabolites and have been implicated as causative agents in human hepatic and extrahepatic carcinomas. They are produced by *Aspergillus flavus* and *A. parasiticus* with

A. flavus being the main cause of pre-harvest aflatoxin contamination in field crops (Bennett and Goldblatt 1973, Cleveland and Bhatnagar, 1992, Cotty, 1997, and Yu *et al.*, 2004). Aflatoxin B1 (AFB1) was evaluated as a Class 1 human carcinogen (IARC 1993). The incidence of AF in foods and feeds is relatively high in tropical and subtropical regions, where the warm and humid climate provides optimal conditions for the growth of moulds (Rustom 1997). A number of surveys and monitoring programs have been conducted in several countries attempting to obtain an overall pattern of the extent of food and/or feed contamination by AF (Liu *et al* 2006). Nevertheless, most countries are known to have some regulations limiting the maximum allowable amount of AF and other mycotoxins in food and feed (Xiao 1988, Hansen 1993). All countries with mycotoxin regulations have tolerances for AF in food and/or feed. Accordingly, maximum tolerated levels for total aflatoxins (AFT) in food e. g. cereal grains range from 4- 10 µg kg⁻¹ according to the European Commission maximum levels (Cheli *et al.*, 2009) and in most developing countries which is up to 50 µg kg⁻¹ (Dohlman 2003). Because of the food safety regulations, grains with higher level of aflatoxins are prohibited from trade domestically or internationally (Ellis *et al.*, 1991). Economic losses are significant to farmers. Comprehensive surveys were carried out in many parts of the world on aflatoxins in grains (Liu *et al.*, 1981, Ilago and Juliano 1982, Breckenridge *et al.*, 1986,

In Egypt, bread is a staple food and supplying people for energy, protein, minerals and vitamins. The purpose of this experimental study was to evaluate the nutritional value as well as the natural occurrence of fungi and aflatoxins in unstored and stored wheat and maize grains.

MATERIALS AND METHODS

Samples collection

Sampling was done from one of canvas covered stores with 5000 ton capacity. 10 samples (\geq 2kg) of wheat grains from those locally produced in Egypt or imported for human consumption from American and Asian countries were taken from storage silos located in Nazlet-Abdella, Assiut Governorate, Egypt during the month of June 2007. Samples were divided into two lots of subsamples. The first group of subsamples were refrigerated upon arrival in the laboratory until analysis within few days. The second group was kept at room temperature (\sim 30- 40 °c) for up to 6 month storage, thereafter were analyzed.

Determination of grain moisture content

The moisture content of the grain subsamples was determined. Three replicates of 50 g each were taken from each food sample and put on an aluminium foil dish. These were dried in an oven at 110 °C for 24 hours, and reweighed (Magan and Lacey 1985, Pitt and Hocking 1997). The moisture content of each subsample was expressed as the average percentage of weight loss of the three replicates.

Chemical composition

Crude fat, ash, crude protein, crude fiber contents were determined according to the standard methods of AOAC (1990). The total carbohydrates were calculated by difference.

Mycological analysis

The direct plate method was used for enumeration and isolation of fungi from the samples. All samples were analysed on plates of dichloran rose bengal chloramphenicol agar, DRBC (King *et al.*, 1979), and dichloran 18 % glycerol agar DG18 (Hocking and Pitt, 1980). Four plates for each medium were used for each sample. The inoculated plates were incubated at 25°C for 7–15 days. The growing fungi were enumerated, isolated and identified. The identification of fungi was carried out on the basis of their macroscopic and microscopic features using the keys by Raper and Fennell (1965), Booth (1971) Ellis (1971), Pitt (1979) and Pitt and Hocking (1997). The counts of fungi on both media were expressed as colony forming units (CFU/grains tested).

Aflatoxin analysis by HPLC and Rida^R Quick Aflatoxin test

All samples were analyzed for aflatoxins before and after storage using High performance liquid chromatography (HPLC), Kenour model 64, Germany. A fluorescence detector at 365 nm was used. A reverse phase column, Bandclone 10 C18, was used and the flow rate was at 1.5 ml/min.

Rida^R Quick aflatoxin test is a qualitative immunochromatographic test in strip format (R5204), R-Biopharm AG., Darmstadt, Germany was also used for the detection of aflatoxin. The basis of the immunochromatographic assay in test strip format is an antigen-antibody reaction. A specific antibody against aflatoxin recognises the aflatoxin molecules in the samples. The results are read visually by observing the development of colored bands.

Statistical analysis

Data were statistically analysed using paired T-test according to Steel and Torrie (1987).

RESULTS AND DISCUSSION

Gross chemical composition of wheat and maize grain samples

Moisture content of wheat samples was relatively low with the lowest value being recorded in the Russian wheat (3.58%) and the highest in the local (8.33%), however, the value was significantly increased with storage (9.29% to 11.96% in the Russian). In maize samples, moisture content significantly increased with storage from 5.56% to 12.04% (Table 1).

The change in the crude protein and fat contents of wheat grain samples were not significantly affected by storage, however, at the time that the most samples showed slight increase, others showed slight decrease with storage. Their values ranged from 8.63% to 10.11% before storage and from 9.25% to 10.13% after storage for protein and from 0.67% to 1.33% before storage and from 0.96% to 3.41% after storage for crude fat.

Table 1 . Mean values (\pm SE) of gross chemical composition of the wheat and maize grain before (b) and after (a) storage (6 month).

Sample No.	Source	Moisture %		Crude protein %		Crude fat %		Crude fibre %		Ash %		Carbohydrate %	
		b	a	b	a	b	a	b	a	b	a	b	a
		1	Local(Beni-Suef)	6.29 \pm 0.02	11.15 \pm 0.01	9.25 \pm 0.01	9.30 \pm 0.00	0.94 \pm 0.01	1.30 \pm 0.00	3.71 \pm 0.02	3.06 \pm 0.00	3.51 \pm 0.03	0.65 \pm 0.02
2	Loca (Sids 1)	8.33 \pm 0.01	11.22 \pm 0.01	9.25 \pm 0.00	9.57 \pm 0.01	0.82 \pm 0.01	1.14 \pm 0.01	4.88 \pm 0.01	4.48 \pm 0.01	2.68 \pm 0.01	0.82 \pm 0.01	74.90 \pm 0.01	72.77 \pm 0.02
3	Local (Durum)	4.52 \pm 0.01	11.29 \pm 0.01	8.87 \pm 0.01	9.58 \pm 0.01	0.70 \pm 0.01	1.21 \pm 0.01	3.97 \pm 0.01	2.89 \pm 0.01	3.14 \pm 0.01	0.61 \pm 0.01	78.80 \pm 0.01	74.42 \pm 0.01
4	Local	5.12 \pm 0.01	10.69 \pm 0.01	10.11 \pm 0.01	9.59 \pm 0.01	0.75 \pm 0.02	0.83 \pm 0.02	3.69 \pm 0.03	3.33 \pm 0.01	2.29 \pm 0.02	0.73 \pm 0.01	78.04 \pm 0.01	74.83 \pm 0.01
5	Local (Sakha 93)	6.86 \pm 0.02	11.51 \pm 0.01	9.37 \pm 0.01	9.48 \pm 0.03	0.69 \pm 0.01	0.97 \pm 0.01	6.68 \pm 0.01	5.28 \pm 0.01	5.02 \pm 0.01	0.60 \pm 0.01	71.38 \pm 0.03	72.16 \pm 0.01
6	USA(White)	5.05 \pm 0.01	10.49 \pm 0.01	8.63 \pm 0.01	10.13 \pm 0.01	1.33 \pm 0.01	1.12 \pm 0.01	3.41 \pm 0.01	4.19 \pm 0.02	2.14 \pm 0.01	0.71 \pm 0.01	79.44 \pm 0.01	73.36 \pm 0.01
7	USA(Red)	7.70 \pm 0.01	11.27 \pm 0.01	9.25 \pm 0.00	9.25 \pm 0.01	1.30 \pm 0.01	0.99 \pm 0.01	2.25 \pm 0.01	3.79 \pm 0.01	2.22 \pm 0.01	0.67 \pm 0.02	77.28 \pm 0.01	74.03 \pm 0.01
8	Russian	6.89 \pm 0.01	11.96 \pm 0.01	9.20 \pm 0.01	9.87 \pm 0.01	1.31 \pm 0.01	3.41 \pm 0.01	3.91 \pm 0.01	5.02 \pm 0.01	4.15 \pm 0.01	1.02 \pm 0.01	74.54 \pm 0.01	68.72 \pm 0.01
9	Russian (Red)	3.58 \pm 0.01	9.29 \pm 0.01	9.90 \pm 0.00	9.41 \pm 0.01	1.31 \pm 0.02	1.04 \pm 0.01	3.96 \pm 0.01	3.91 \pm 0.01	2.90 \pm 0.02	0.61 \pm 0.03	78.35 \pm 0.00	75.74 \pm 0.01
10	Kazakhstan (Red)	6.76 \pm 0.01	11.31 \pm 0.01	9.38 \pm 0.01	9.48 \pm 0.01	0.67 \pm 0.01	0.96 \pm 0.01	6.58 \pm 0.01	5.29 \pm 0.00	5.32 \pm 0.01	0.62 \pm 0.01	71.29 \pm 0.01	72.70 \pm 0.01
11	Maize (Local)	5.56 \pm 0.00	12.04 \pm 0.01	8.93 \pm 0.01	9.22 \pm 0.00	3.21 \pm 0.02	3.85 \pm 0.03	2.77 \pm 0.00	3.88 \pm 0.01	1.97 \pm 0.01	0.67 \pm 0.01	77.56 \pm 0.01	70.34 \pm 0.01
	T test	-14.6**		-1.45		-1.73		0.2		7.07**		3.8**	

** Highly Significance at $P < 0.01$

Figures are number of CFUs calculated per 20 maize grains.

In maize grains crude protein and fat increased by storage from 8.93% to 9.22% for protein and from 3.21% to 3.85% for fat. Crude protein reported earlier were almost higher than those reported herein e, g. 11% in soft wheat and 13.5% in hard wheat (Ragaei *et al.*, 2006), from 12-39-15.83% in different wheat varieties (Coskuntuna *et al.*, 2008) or 19.8% (Widyaratne and Zijlstra 2007). Similar to the current results, crude fat in wheat analyzed by Widyaratne and Zijlstra (2007) was 1.8%, and in soft and hard wheat analyzed by Ragaei *et al.*, (2006) were 0.86 and 0.98%, respectively. Also slight increase (Bcyacioglu and Hettiarachchy 1995) or decrease (Wasonich 1991, Boyacioglu and Hettiarachchy 1995) in protein and fat contents of wheat when infected with different fungi had been also reported.

Crude fiber was not significantly affected by storage, however, only 3 samples out of the ten analyzed showed increase in the fiber content with storage. Their values ranged from 2.25% in the USA wheat to 6.68% in the local wheat. Crude fiber content increased in maize from 2.77% before storage to 3.88% after storage. Nearly similar values were reported for wheat (Widyaratne and Zijlstra 2007, Coskuntuna *et al.*, 2008), however higher level (15.8%) was reported in wheat kernels analysed by Boyacioglu and Hettiarachchy (1995).

Ash content significantly decreased with storage with the lowest value being recorded in the USA wheat (2.14%) and the highest in Kasahstan wheat (5.32%) before storage, whil after storage the lowest in the local (0.6%) and the highest in the Russian (1.02%). In maize grains ash content significantly decreased with storage from 1.97% to 0.67%. In most previous reports, ash content was lower than that reported in the current study e.g. 0.71% and 0.56% in soft and hard wheat (Ragaei *et al.*, 2006), 2.1% (Widyaratne and Zijlstra 2007) or 1.82-1.88% in different wheat varieties analysed by Coskuntuna *et al.* (2008).

Carbohydrate contents of wheat grain samples (values ranged from 71.29%-79.44% and 68.62%-75.74% before and after storage, respectively) were significantly increased with storage, however only 2 samples showed slight decrease in these contents with storage. Starch content in wheat analyzed by Ragaei *et al.* (2006) was within the range of 77.4-77.9%.

Mycobiota associated with freshly harvested wheat grains (recovered on DRBC).

From the results presented in Table (2), it is obvious that 18.4% of the total wheat grains tested were fungi-free. *Alternaria alternata* (45.3% of the total *propagules*) was found in all wheat samples tested. *Aspergillus* (13.3%) was found contaminating 90% of samples. *A. flavus* and *A. niger* (5.42% each) are the most common *Aspergilli* found in 60% of the samples. Rarely occurred aspergilla include *A. candidus*, *A. fumigatus* and *A. ochraceus*. In agreement with the current results, *A. alternata* was the most common field fungal species isolated from cereal grains including wheat in USA (Sauer *et al.*, 1982, 1984), Argentina (Broggi *et al.*, 2007), However, *Aspergillus niger*, *A.flavus*, *A.fumigatus*, *A.terreus* and *A. alternata* followed by *A. ochrceus* and *A.candidus* were the most common in wheat grains in Egypt (Mazen *et al.*, 1984).

Table 2. Fungal species found in wheat grain samples before and after storage on dichloran rose Bengal chloramphenicol agar (DRBC) and Dichloran glycerol 18 % agar (DG18) at 25 °C.

Toxon	DRBC				DG18	
	Freshly* harveste d	% F**	Stored* (6 month)	% F**	Stored* (6 month)	% F**
<i>Absidia cylindrospora</i>					2	10
<i>Acremonium</i> sp.			1	10		
<i>Alternaria alternata</i>	92	100	40	60	36	50
<i>Aspergillus</i> (Total)	27	90	107	100	69	70
<i>A. candidus</i>	2	20			4	10
<i>A. flavus</i>	11	60	54	90	26	40
<i>A. fumigatus</i>	2	20	2	20		
<i>A. niger</i>	11	60	47	80	39	70
<i>A. ochraceus</i>	1	10				
<i>A. sydowii</i>			4	10		
<i>Cladosporium</i> (Total)			6	20	30	50
<i>C. cladosporioides</i>			4	20		
<i>C. sphaerospermum</i>			2	10	25	50
<i>C. porophorum</i>					5	20
<i>Cochliobolus spicifer</i>			1	10		
<i>Elysiella athecica</i>					47	70
<i>Epicoccum nigrum</i>	1	10				
<i>Eurotium</i> (Total)	2		14	30	104	100
<i>E. armstelodami</i>			3	30	1	10
<i>E. chevalieri</i>	2	20	11	30	49	70
<i>E. repens</i>			1	10	34	40
<i>E. rubrum</i>					20	60
<i>Fusarium</i> (Total)	7	50	58	60		
<i>F. chlamydosporum</i>			3	10		
<i>F. nygamai</i>	2	10	2	10		
<i>F. proliferatum</i>			49	60		
<i>F. semitectum</i>			2	10		
<i>F. subglutinans</i>	2	20				
<i>F. verticillioides</i>	3	20	2	10		
<i>Geosmithia</i> sp.					1	10
<i>Mucor</i> (Total)	24	50	2	10		
<i>M. circinnelloides</i>	2	10				
<i>M. fuscus</i>	22	50				
<i>Mycocladus corymbiferus</i>	1	10				
<i>Nigrospora oryzae</i>	3	20				
<i>Penicillium</i> (Total)	10	60	67	100	50	60
<i>P. aurantiogriseum</i>			28	90	29	50
<i>P. chrysogenum</i>	10	60	25	30	16	10
<i>P. dudauxii</i>			5	20		
<i>P. expansum</i>			5	30	2	10
<i>P. griseofulvum</i>			2	20		
<i>P. pinophilum</i>			1	10	1	10
<i>P. spinulosum</i>			1	10	2	10
<i>Rhizopus</i> (Total)	33	70	47	80		
<i>R. oryzae</i>	33	70				
<i>R. stolonifer</i>			47	80		
<i>Stemphylium botryosum</i>					12	30
Yeast	1	10	3	20		
White sterile mycelia	2	10				
Total CFUs /250 grains	203	100	346	100	351	100
No. free grains (out of 250)	46	70	7	40	11	30

*Figures are number of CFUs (Colony-Forming Units) calculated per 250 wheat grains.

**%F: Frequency of occurrence out of 10 samples.

Rhizopus (1 species, 16.26%) and *Mucor* (2 species, 11.82%) were also dominant where they found in 70% and 50% of the samples. *R. oryzae* and *M. fuscus* were the dominant. These *mucoraceus* fungi were absent in wheat grains in the study of Mazen *et al.*, (1984), however, other species of these genera were detected but in low incidences and densities in the study of Broggi *et al.*, (2007).

Table 3. Fungal species found in maize grains on dichloran rose Bengal chloramphenicol agar (DRBC) and Dichloran glycerol 18% agar at 25°C.

Toxon	Freshly harvested		Stored (6 month)	
	DRBC	DRBC	DRBC	DG18
<i>Aspergillus</i> (Total)	28	42		9
<i>A. flavus</i>	3	17		
<i>A. niger</i>	25	25		9
<i>Eurotium</i> (Total)				26
<i>E. chevalieri</i>				25
<i>E. rubrum</i>				1
<i>Mycocladus corymbiferus</i>	2			
<i>Penicillium</i> (Total)	2	4		
<i>P. oxalicum</i>	1			
<i>P. duclauxii</i>	1	1		
<i>P. pinophilum</i>		3		
<i>Rhizopus stolonifer</i>		10		
Gross total	32	56		35

Penicillium (1 species, 4.93%) and *Fusarium* (3 species, 3.45%) occurred in 60% and 50% of the samples. They were represented by *P. chrysogenum*, *F. verticillioides*, *F. subglutinans* and *F. nygamai*. These species were also isolated from wheat grains in many studies but in different frequencies and densities in Egypt (Mazen et al., 1984) Argentina (Broggi et al., 2007), Iran (Gohari et al., 2007), Turkey (Askum 2007).

Other fungi with rare incidence include *Epicoccum nigrum*, *Eurotium chevalieri*, *Mycocladus corymbiferus*, *Nigrospora oryzae*, yeast and white-colored sterile mycelia.

Mycobiota associated with stored wheat grains (recovered on DRBC and DG18).

It is noted that 4.4% of the grains tested were fungi-free. *Aspergillus* (5 species) was common on both DRBC and DG18 (Table 2). It was isolated from 100% and 70% of the samples, comprising 30.9% and 19.66% of total fungi, on both media, respectively. *A. flavus* (90% and 40% of the samples; and 15.61% and 6.4% of total fungi) and *A. niger* (80% and 70%; and 13.58% and 11.11%) were the most common on both media. Less common species were also recovered on DRBC (*A. sydowii*) and DG18 (*A. candidus*).

Eurotium (4 species) was also common and was isolated from 30% and 100% of the samples, constituting 4.01% and 29.63% of total fungi, on both media, respectively. The four species were encountered on DG18 while only two on DRBC with *E. chevalieri* being the most common on both media while *E. repens* and *E. rubrum* on DG18.

Penicillium (7 species) was encountered in 100% and 60% of the samples, accounting for 19.36% and 14.25% of total fungi on both media, respectively. *P. aurantiogriseum* (90% and 50% of the samples) followed by *P. chrysogenum* and *P. expansum* (30% and 10% each) were the most common *penicillia* found in stored wheat, accounting for 8.1% and 8.3%; 7.23% and 4.56%; and 1.45% and 0.57% of the total propagules on both media, respectively. The other four *Penicillium* species were less frequent (Table 2).

Other common fungi were also isolated on either DG18 (the xerophilic species *Edyullia athecia*), DRBC (*Rhizopus stolonifer* and *Fusarium*, 5 species with *F. proliferatum* being the most common), or both media (*Alternaria alternata*).

Less frequently encountered fungi include *Absidia cylindrospora*, *Cladosporium porophorum*, *Geosmithia sp.* and *Stemphylium botryosum* on DG18; and *Acremonium sp.*, *Cladosporium cladosporioides*, *Cochliobolus spicifer*, *Mucor* and yeast on DRBC; and *C. sphaerospermum* (Table 2).

Table 4. Aflatoxins, Aflatoxigenic species and total fungal propagules in wheat and maize grain samples before and after storage.

Sample No.	Freshly harvested				Stored (6 month)					
	Aflatoxins (μgkg^{-1})		Aflatoxigenic (Total) *		Aflatoxins (μgkg^{-1})		Aflatoxigenic (Total) *			
	HPLC	KITS	(DRBC)		HPLC	KITS	(DRBC)	(DG18)		
1	10	9	-	(13)	5	5	2	(23)	1	(27)
2	8	8	1	(29)	10	10	23	(50)	21	(69)
3	6	6	-	(28)	7	7	-	(43)	-	(44)
4	6	6	1	(15)	6	6	7	(49)	3	(37)
5	9	9	4	(21)	7	7	8	(31)	-	(31)
6	8	8	-	(21)	5	6	2	(38)	1	(35)
7	8	8	1	(12)	10	9	2	(19)	-	(18)
8	7	6	-	(26)	8	7	7	(48)	-	(41)
9	7	6	3	(26)	8	8	1	(26)	-	(31)
10	8	7	1	(12)	10	9	2	(19)	-	(18)
11	9	9	3	(32)	4	4	17	(56)	-	(35)

*On dichloran rose Bengal chloramphenicol agar (DRBC) and Dichloran glycerol 18 % agar (DG18) at 25 °C.

Mycobiota associated with freshly harvested and stored maize.

Five genera represented by 9 species were recovered from maize grains before (3 genera and 5 species) and after storage (4 and 7). *Aspergillus* (*A. niger* followed by *A. flavus*) was common on DRBC in both freshly harvested and stored maize. *A. flavus* was found frequently in U. S. export corn samples, Sauer *et al.*, 1982, 1984) and Uganda (Ismail *et al.*, 2003). *A. flavus* can invade corn in the field, so our and the previous data reflect both preharvest and postharvest invasion (Ismail *et al.*, 2003). Also in Nigeria, Amadi and Adeniyi (2009) found that *Aspergillus* (represented by *A. terreus*, *A. flavus*, *A. niger* and *A. oryzae*) was associated with maize in storage.

Eurotium, with *E. chevalieri* followed by *E. rubrum* being the most commonly encountered, was isolated only from the stored maize grains and on DG18. *Aspergillus glaucus* (Teleomorph: *Eurotium*) was the most common fungus found in all (Sauen *et al.*, 1982) or 84% (Sauer *et al.*, 1984) of the farm-stored corn samples in USA.

Penicillium (3 species) was less common with *P. dauclexii* in both cases, *P. oxalicum* in fresh grains and *P. pinophilum* on stored ones. On the other hand, *Mycocladus corymbifera* was isolated only from fresh grains and *Rhizopus stolonifer* from only stored grains on DRBC. Species of *Penicillium*

and other fungi were also reported from freshly harvested or stored corn in Spain (Sanchis *et al.*, 1982), USA (Sauer *et al* 1982, 1984), Uganda (Ismail *et al.*, 2003), Cameroon (Ngoko *et al.*, 2008), Nigeria (Amadi & Adenigi 2009).

Aflatoxins in wheat and maize grains before and after storage

However, all samples were contaminated with aflatoxin in range of 6-10 μgkg^{-1} in freshly harvested and of 4-10 μgkg^{-1} in stored grains using both analytical methods (HPLC and Kits) in both wheat and maize grain, the total aflatoxin level in all local and imported samples tested was below the permitted limit according to the European Commission fixed maximum levels which is 10 μgkg^{-1} for total aflatoxins (Cheli *et al.*, 2009) and in most developing countries which is up to 50 $\mu\text{g kg}^{-1}$ (Dohlman 2003) . Aflatoxin analysis of 10 maize grain samples in Uganda revealed that only 5 were naturally contaminated with aflatoxins in the range of 0-10 ppb (Ismail *et al.*, 2003); however, the study of Liu *et al* (2006) in china revealed only 0.84-1.17 $\mu\text{g /kg}$ aflatoxins in 69 out of 71 stored corn samples tested.

The CFUs of aflatoxigenic species *A. flavus* were generally low in freshly harvested grain samples (found in 60% of the samples and with a range of 3.45%- 19.1% of the total fungal propagules on DRBC), however it was higher in stored samples (90% of the samples and with a range of 3.85%-46.0% on DRBC, and 40% of the samples and with a range of 2.86%-30.44% on DG18).

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القيمة الغذائية ومدى تواجد الفلورا الفطرية والأفلاتوكسينات في حبوب القمح و الذرة

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أشارت نتائج الدراسة الحالية على حبوب القمح المحلية والمستوردة للاستهلاك الأدمى عن زيادة معنوية في نسبة الرطوبة أثناء عملية التخزين بينما كان هناك انخفاض معنوي في نسبة الرماد والكربوهيدرات أثناء عملية التخزين ولم تظهر أي اختلافات معنوية في كل من البروتين الخام والدهون والألياف. وتم عزل اجناس مختلفة من الفطريات من أهمها. *Alternaria alternate* ونسبته ٤٥,٣%، *Aspergillus* ١٣,٣%، ساد منها كل من *A. niger* و *A. flavus* (بنسبة ٥,٤٢% لكل) في حبوب القمح الطازجة تم عزل بعض الانواع الاخرى من جنس *Aspergillus* ولكنها كانت أقل سيادة مثل *A. candidus* و *A. fumigatus* و *A. ochraceus*. كما ساد أيضا فطر *Rhizopus* (ممتلا بنوع واحد ونسبته ١٦,٢٦%) و *Mucor* (نوعين ونسبته ١١,٩٢%) وفطر *Penicillium* (نوع واحد ونسبته ٤,٩٣%) وكذلك فطر *Fusarium* (ثلاث انواع ونسبته ٣,٥٤%). ومن الحبوب المخزنة تم عزل كل من *Aspergillus* و *Eurotium* و *Penicillium* ومن الانواع الشائعة التي تم عزلها *A. niger* و *flavus* و *E. chevalieri* و *E. repens* و *E. rubrum* و *P. expansum* و *P. chrysogenum* و *aurantiogriseum* عزلها من الذرة الطازجة والمخزنة كان *A. niger* و يليه *A. flavus*، كما تم عزل فطر *Eurotium* ومن الانواع التي تم عزلها *E. chevalieri* و *E. rubrum* من حبوب الذرة المخزنة فقط. كما أظهر تحليل العينات أن المستوى الكلى للتوكسينات في العينات المحلية والمستوردة المختبرة ≥ 10 ملليجرام/كجم بطريقتي التقدير المستخدمين في كل من الحبوب الطازجة والمخزنة والتي تعتبر في داخل الحد المسموح بتواجده في الغلال.