
OCCURRENCE OF AFLATOXINS IN TABLE EGGS SOLD IN DAMIETTA CITY REGARDING ITS HEALTH SIGNIFICANCE

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SUMMARY

Forty random table chicken eggs samples (20 each of “brown” and “balady” eggs) were collected from different groceries and supermarkets at Damietta City. The collected samples were submitted to analysis for isolation of viable moulds and qualitative detection of aflatoxin (AFs) residues. The isolation of moulds revealed that 3 (15%) and 20 (100%) mould species belonging to three genera were isolated from examined samples respectively. *Aspergillus* was the less prevalent genus encountered comprising 5% while; *Trichophyton* comprised 10% in table eggs “brown”. Otherwise, *Cladosporium* was the most prevalent genus encountered comprising 80% while; *Aspergillus* comprised 20% in table eggs “balady”. On the other hand, different percentages of aflatoxin contamination AFB1 (45%), AFB2 (20%) and AFG2 (15%) were detected in table eggs “brown” samples. While, lower percentage of AFB2 (10%) was detected in table eggs “balady” although AFB1, AFG1 and AFG2 were failed to detectable in the same samples. The obtained results and public health implications and suggestive measures for improving table chicken eggs were discussed.

INTRODUCTION

Avian eggs are familiar, versatile, nutritious economical and quick and easy to prepare food, as they provide a unique well balanced source of nutrients for persons of all ages. Moreover, their high quality, low caloric value and ease of digestibility make eggs valuable in many therapeutic diets for adults **Burley & Vadehra (1989)** and **Bufano (2000)**. Worldwide, there are two kinds of eggs produced, primary or chicken eggs and other eggs (excluding hens). Chicken eggs production is the most significant, amounting to 91-96% of worldwide totals during 1961-2001. Also, is a large-scale activity, more important commercially than production of other types of eggs. In 2001, poultry production in Egypt and other middle-income countries totaled 37.5 million tons, compared with 29.3 million tons in high-income countries and only 3.6 million tons in low-income countries. Egypt's poultry industry is highly dependent on feed imports, with a dependency rate approaching 100 percent for soybean and 48 percent for yellow corn. In the early 21st century, Egypt like many other middle-income countries faces the potential challenge of greater liberalization under the World Trade Organization (WTO), and the efficiency of its domestic poultry industry will influence domestic consumers, producers, and exporters competing to sell either products or intermediate inputs into this growing market **Fawzi (2003)**.

Both man and animals live under a certain degree of “biological hazard” from natural toxicants that occur in food and foodstuffs **Crosby (1969); Armbrrecht (1971); Allam et al. (1999-a; 1999-b & 2002)** and **Abdelhamid et al. (2002)**.

Naturally occurring toxins such as mycotoxins pose profound challenges to food safety widespread in many countries, especially in tropical and subtropical regions where temperature and humidity conditions are optimum for growth of moulds and production of toxins, so they are found in a wide variety of agricultural products (such as corn, wheat, soybean, barley and rice) **FAO (1991)**, and animal feeds as well as meat products and chicken products including eggs, as a result of carry over from contaminated animal feed **Mossel & Shennan (1976); Blunden et al. (1991); Moustafa (1994); Adams and Moss (1995); Orriss (1997); Palli et al. (1999); Sayed et al. (2000); Takahashi-Ando et al. (2004); Cavaliere et al. (2006)** and **Trucksess et al. (2006)**. Mycotoxins contamination of food and feeds remains a worldwide problem, the United Nation Food and Agriculture Organization (FAO) has estimated that up to 25% of the world’s food crops are significantly contaminated with mycotoxins **Jelinek et al. (1989); Smith et al. (1994)** and **WHO (1999)**. Mycotoxins are unavoidable food contaminants even when good agricultural practices are applied. Crop transfers through international trade have made aflatoxins contaminated food a worldwide problem **Sherif (2003)**.

Nowadays the main mycotoxins of interest are aflatoxins (AFs), ochratoxins, trichothecenes, zearalenone, fumonisins, ergot alkaloids and deoxynivalenol **Hussein & Brasel (2001); Bhat & Vasanthi (2003)** and **Cleveland et al. (2003)**. AFs are a group of polyketide-derived furanocoumarins, with at least 16 structurally related toxins that have been characterized. These toxins are produced by a number of different *Aspergillus* species **CAST (1989); FAO (1991); Abdelhamid (1993); Goto et al. (1996); Klich et al. (2000); Ito et al. (2001)** and **Peterson et al. (2001)**. However, in the agriculture commodities, they are primarily produced by *Aspergillus flavus* and *Aspergillus parasiticus*. There are four major AFs (AFB1, AFB2, AFG1, AFG2) all of which occur naturally **Anonymous (1998)** AFB1 is the most commonly occurring of the mould producing compounds **Bhat & Miller (1991)** and **Brera et al. (1998)**. AFB1 has been included in category 1A of active carcinogenic compound **IARC (1993)**. Meanwhile, AFB1 had assumed the biosynthetic precursor of the other aflatoxins **Dutton et al. (1985)**. Other significant member of the AF family, M1 and M2 are 4-hydroxy derivatives of AFB1 and AFB2 respectively. The crud AFs are heat tolerant (melting point up to 250 °C) and their breakdown during cooking or processing is almost unlike **Fink (1989)**, these toxins were found in the food in spite of disappearing fungal organisms **Mislivec (1981); Fraizer (1983)** and **Macdonald & Castle (1996)**.

In Egypt, the increased demand of animal protein presents a serious problem and among the planned projects to resolve such problem is the concentration on poultry industry as a rapid and more economic source of protein (meat and eggs). Freshly laid eggs are generally sterile, however they may constitute, if contaminated, a public health hazard, leading to losses from economic point of view through spoilage. Although foodborne hazards may be of physical, chemical or microbiological origin, there is currently widespread classification that microbial foodborne hazards represent the greatest risk to consumers **Hafez (1999)**. Sources of this contamination are numerous as egg may be infected before it is laid, also outside eggs by faecal matter, the lining of the nest, wash water if the eggs are washed, handling and perhaps by the material in which eggs are packed **Board & Fuller (1994)** and **Cox et al. (2000)**. Furthermore, according to the

Centers for Disease Control and Prevention (CDC), eggs are responsible for an estimated 230,000 case of foodborne illness each year **Bufano-Nancy (2000)**.

Researchers from USDA Division of Epidemiology and Surveillance have articulated that the ingestion of mycotoxin-contaminated animal-based food products could pose a concern to public health **Hollinger & Ekperigin (1999)**. Mycotoxins occur in small amount in the foods, however their continuous intake even in microdoses can result in their accumulation in the human and animal bodies, during the long term consumption which can cause a variety of ill effects in humans from allergic responses to immunosuppression and cancer, probably underestimates the effect of mycotoxins as a cause of human mortality **Varman & Evans (1991)**; **El-Shinawy et al. (1994)**; **Ramasatry et al. (2000)**; **Pitt (2000)** and **Kovacs (2004)**.

To secure the safety of foodstuffs, regular monitoring of mycotoxin is necessary. For this purpose the following work is to provide a qualitative overview about aflatoxin residues in marked table chicken eggs.

MATERIAL AND METHODS

Sampling:

A total of 40 random table chicken eggs samples, representing 20 of each of table eggs “brown” and table eggs “balady”, were collected from different groceries and supermarkets at Damietta City. The samples were labeled and taken to the laboratory then kept in refrigeration till analysis:

1. **Isolation of viable mould:** using Sabouraud’s dextrose agar (**Mislivec et al., 1992**). The isolated fungi were identified according to different guidelines adopted by **Raper and Thom (1949)**; **Raper & Fennel (1965)**; **Domasch et al. (1980)** and **Nirenberg (1989)**.
2. **Mycotoxin analysis:** qualitative detection of aflatoxins was adopted by the technique described by **Jonsy et al. (1995)** and **Truksess (2000)**.

RESULTS AND DISCUSSION

Table, (1): Prevalence of isolated moulds in examined table eggs “brown” and table eggs “balady” samples (n= 20 each):

Types of isolates	Table eggs “brown” (20 sample)		Table eggs “balady” (20 sample)	
	Positive samples		Positive samples	
	No.	%	No.	%
1- <i>Aspergillus</i> spp.	1.00	5.00	4.00	20.00
2- <i>Cladosporium</i> spp.	00.00	00.00	16.00	80.00
3- <i>Trichophyton</i> spp.	2.00	10.00	00.00	00.00
total	3.00	15.00	20.00	100.00

It is noticeable from the results outlined in **table 1** that, 1 (5%) and 2 (10%) mould species belonging to *Aspergillus* and *Trichophyton* genera were presented in examined samples of table eggs “brown” respectively. While, 4 (20%) and 16 (80%) mould species belonging to *Aspergillus* and *Cladosporium* genera were presented in examined

samples of table eggs “balady” respectively. As the mould quality reflects the care with which eggs were produced, handled and stored APHA (1992). Otherwise, different types of moulds were isolated by several investigators as, El-Essawy et al. (1989); Bekhit et al. (1992); Aman et al. (1993); Hassan (1995); Martins et al. (1998) and Abdel-Latif (2001) these moulds including, Penicillium, Cladosporium, Aspergillus, Alternaria alternata, Mucor and Rhizopus genera from examined avian eggs.

Table, (2): Incidence of aflatoxins (AFs) residues in examined table eggs “brown” and table eggs “balady” samples (n= 20 each):

Types of aflatoxins	Table eggs “brown” (20 sample)		Table eggs “balady” (20 sample)	
	Positive samples		Positive samples	
	No.	%	No.	%
1- AFB1	9.00	45.00	00.00	00.00
2- AFB2	4.00	20.00	2.00	10.00
3- AFG1	00.00	00.00	00.00	00.00
4- AFG2	3.00	15.00	00.00	00.00
total	16.00	80.00	2.00	10.00

Table 2, showed that the incidence of AFs (B1, B2 and G2) residues in examined samples of table eggs “brown” were 9 (45%); 4 (20%) and 3 (15%) respectively. Although, our findings failed to detect AFG1 in the same samples. While, two out of 20 (10%) table eggs “balady” samples were contaminated only with AFB2. Indeed, these toxins were found in the food in spite of disappearing fungal organisms Mislivec (1981); Fraizer (1983) and Macdonald & Castle (1996). As regard to AFs residues Stoloff & Trucksess (1978) recorded lower result (0.89%) contaminated with AFB1. Whereas, different results for AFB1 residues in chicken eggs was recorded by Oliveira et al. (2000 & 2003) and Pandey & Chauhan (2007). Moreover, higher result was found by Hassan (1995) that 1 (4%) of table eggs “balady” samples was contaminated with AFG2. On contrary, our findings failed to detect neither AFB1 nor, AFG1 nor, AFG2 in table eggs “balady” samples.

From the above achieved findings, it could be noticed that table eggs “balady” have a best quality to some extent in comparison to table eggs “brown”. But, it needs more care during producing, handling and storing to minimize mould contamination to safeguard human from being infected. Unfortunately, the highest aflatoxins contamination in table eggs “brown” gave indication for poor conditions during producing, handling and storing, exhibit a wide array of hazardous impacts on animal and human health.

Regarding the public health significance, mycotoxins attract worldwide attention because of the significant associated with their impact on human health, animal productivity and trade CTA (1997). Exposure to mycotoxins can produce both acute and chronic toxicities ranging from death to deleterious effects on the central nervous, cardiovascular, pulmonary and digestive systems. Mycotoxins may also be carcinogenic, mutagenic, teratogenic and immunosuppressive. The ability of some mycotoxins to compromise the immune response and consequently, to reduce resistance

to infectious diseases is now widely considered to be the most important effect of mycotoxins particularly in developing countries **FAO (2001)**.

Aflatoxins, were considered as hepatotoxin and hepatocarcinogens in man and different animal species, **Krishnamachari et al. (1975)**; **Mirocha (1983)**; **Varman & Evans (1991)**; **Raisuddin et al. (1993)** and **Berera (1998)**. Those toxins could produce degenerative changes in liver due to the ability to bind with DNA and impair protein synthesis **Pler & Heddleston (1970)**; **Cheford & Pees (1976)**; **Ngindu et al. (1982)** **Smith (1982)**; **Van Rensburg et al. (1985)** and **Carnaghan et al. (1986)**. AFB1 has been shown to bind covalently to liver mitochondria especially to nuclear DNA **Niranjan et al. (1982)**; **Jonsyn (1999)** and **Smela et al. (2001)**. Epidemiological studies have shown a strong correlation between exposure to aflatoxins and primary liver cancer **Alpert & Davidson (1979)**; **Haggag et al. (2001)**; **Bhat & Vasanthi (2003)** and **Sherif (2003)** which add, that the liver is the main target of aflatoxin toxicity and carcinogenicity. All AFs, are chronically toxic to varying degrees. Otherwise, AFB1 is considered to be the most potent of mycotoxins and has been linked epidemiologically with cases of human liver cancer in a number of developing countries **Aikins & Norman (1998)**. It is noteworthy to mention that primary liver cancer is not a common disease in most areas of the world. There are particular geographic areas, however, where the annual liver cancer rate is reported to be well above the level (2 cases /100,000 people). Certain populations in Africa, southern India, Japan, and Southeast Asia have unusually high incidences of liver cancer. In addition AFs, have a nephrotoxic effect through the degeneration of kidney tubules **Raj & Venkitasubramanian (1978)**; **Cortina & San Gabriel (1982)**; **Orriss (1997)** and **Haggag et al. (2001)**.

AFs, also have carcinogenic, mutagenic, teratogenic and immunosuppressive properties **Hiroshi (1978)**; **Ueno & Ueno (1978)**; **Hayes (1980)**; **Niranjan et al. (1982)**; **Mirocha (1983)**; **Olufemi et al. (1983)**; **Hayes et al. (1984)**; **Anonymous (1985)**; **Beuchat (1987)**; **El-Tahan (1992)**; **Krough (1992)**; **Gourama & Bullerman (1995)**; **Harvey et al. (1995-a & 1995-b)**; **Mon et al. (1998)**; **Sabbioni et al. (1998)**; **Jonsyn (1999)**; **Peraico et al. (1999)**; **Pitt (2000)**; **Abarca et al. (2001)**; **FAO (2001)**; **Yu (2002)**; **Trucksess et al. (2006)** and **Sapkota et al. (2007)**.

AFs, can produce deleterious effects up on several systems, as central nervous **FAO (2001)** and **Trucksess et al. (2006)**. Also, cardiovascular **FAO (2001)**. However, the gastrointestinal tract could show esophageal cancer **Sydenham et al. (1990)**, stomach and colon cancer **Deger (1976)** and **Cortina & San Gabriel (1982)**. Moreover, an acute aflatoxicosis was recently reported in Western India showed clinical pattern of jaundice, vomiting and anorexia **Bullerman et al. (1984)** and **Sayed et al. (2000)**.

AFs, in relation to reproductive system, AFB1 could induced testicular degeneration in laboratory animals **Sahay (1993)** and developmental toxicity **Trucksess et al. (2006)** and pregnancy failed to complete **Badawy (1997)**. Also, act as transplacental carcinogens transfer from mother to her child, can cause genetic defects at foetal stages itself **Tomatis (1974)** and **Maxwell et al. (1989)**. In addition to transfer of AFs in breast milk of mothers who ingest aflatoxin-contaminated food, expose the fetus and young infant to aflatoxins unpleasant health effects **Sherif (2003)**.

Concerning hematological parameters **Harvey et al. (1995-a)**; **Abdelhamid et al. (2002)** and **Allam et al. (2002)** who recorded decrease in most hematological parameters as a result to aflatoxicosis. AFs, also show deficiencies or coagulation

disorders with sever hemorrhages in many parts of human body **Lopez & Crawford (1983)**; **Beuchat (1987)**; **Wilkinson et al. (1988)** and **Ruston (1997)**.

From child health point of view, children exposed to AFs may become stunted, underweight, child neurological impairment and more susceptible to infectious diseases in childhood and later life **Bhat & Vasanthi (2003)** and **Gong et al. (2003)**. Meanwhile, epidemiological evidences implicate aflatoxins in the causation of Kwashiorkor, unexplained neonatal jaundice, hepatitis cirrhosis and Reye's syndrome and decreased immunity **Sherif (2003)**.

Synergistic effects may observe as a result from interaction of mycotoxin with ochratoxin A and citrinin in both animals and in vitro systems **Sansing et al. (1976)** and **Tapia & Seawright (1985)** and as a result from interaction with nontoxic substances as dimethylnitrosamine on liver tumor induced by the most hepatocarcinogenic mycotoxin AFB1 **Angsubhakorn et al. (1981)**. Also, AFs and alcohol when consumed concurrently can act synergistically in developing of primary liver cancer in man **Bulato-Jayme et al. (1982)**. It also has a synergistic effect with hepatitis B virus in the etiology of liver cancer and could interact with HIV/AIDS **Montesano et al. (1997)** and **FAO (1997 & 2001)**.

As a final point, fatal outbreaks of aflatoxicosis have been reported in different localities all over the world resulted from widespread aflatoxin contamination **Pitt (2000)**; **Kovacs (2004)** and **MMWR (2004)**.

CONCLUSION AND RECOMMENDATION

Effective integrated mycotoxins management programs not only cover prevention of mycotoxins formation in agricultural products or their detoxification/decontamination, but also involve routine surveillance, regulatory measures to control the flow of mycotoxins-contaminated material in national and international trade and information, education and communication activities. Food surveillance and the enforcement of proper safety legalization provide the basis for a control strategy.

The simultaneous detection of toxins in chicken eggs samples, randomly collected is not unexpected because poultry feeds and its related ingredients (corn, wheat, soybean and concentrated protein) have been found to be contaminated with toxins in Egypt and also, all over the world.

Methods for reducing levels of aflatoxins in foods include physical, chemical and biological treatments. Good agricultural practices during pre-harvest and post-harvest minimizes the problem of contamination by mycotoxins. These include appropriate drying techniques, maintaining proper storage and taking care not to expose grains or oil seeds to moisture during transport and marketing.

The development of physical, chemical and biotechnological tools to improve seed production, cultivation, harvest and storage of forages and cereals is essential to reduce the level of contamination of food and feeds. However, the total elimination of moulds and their toxins must be considered as impossible.

If contaminated eggs are consumed raw or semi-raw may be responsible for sporadic or epidemic diseases. Moreover, some species of fungi encountered are known to be mycotoxin-producers which threaten human health. Therefore, to safeguard human from

being infected, the hygienic measures adopted in the farm during handling and storage are necessary for obtaining good quality eggs and fit for consumption. These problems were eliminated by providing the birds with good quality feed ingredients.

The need for setting maximum levels of aflatoxins in foods and feeds is generally recognized. Several countries, particularly some industrialized ones, have already set specific regulations. Limits for aflatoxin B1 in foodstuffs of 0 to 30 µg/kg, while those for total aflatoxins range from 0 to 50 µg/kg. In Africa, fifteen countries, accounting approximately 59 percent of the continent's population, were known to have specific mycotoxin regulations.

For this reason, there is a need for additional current preventive measures with the use of agents that are able to bind toxins, consequently limiting their bioavailability in animals and perhaps also in human as well as, the national or international maximum tolerated levels should be fulfilled during analysis of feed and foodstuffs.

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تواجد سموم الأفلاتوكسين في بيض المائدة المباع في مدينة دمياط مع بيان الأهمية الصحية

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المُلخَص العربي

أجريت هذه الدراسة لبيان مدى تواجد سموم الأفلاتوكسين (AFs) في بيض دجاج المائدة، حيث تم إجراء الدراسة على عدد ٤٠ عينة عشوائية (٢٠ عينة من كل من بيض دجاج المائدة "البلدي" والمعروف عند المستهلك ببيض المزارع الأحمر وبيض دجاج المائدة "البلدي")، تم تجميع العينات من محلات البقالة والسوبر ماركت بمدينة دمياط، وتم إجراء عزل فطري للعينات وتحديد أنواعها وفصل سموم الأفلاتوكسين من العينات وتحديد أنواعها المفصولة.

دلت النتائج على عزل ثلاث أجناس مختلفة من الفطريات، حيث تواجدت بنسبة ١٥% في عينات بيض دجاج المائدة "البلدي" و ١٠٠% في عينات بيض دجاج المائدة "البلدي". حيث كان جنس *Aspergillus* الأспيراجيلس الأقل تواجداً في جميع عينات الفحص وكان بنسبة ٥%، بينما جنس *Trichophyton* الترايكوفيتون تواجد بنسبة ١٠% في عينات بيض دجاج المائدة "البلدي". أما جنس *Cladosporium* الكلاوسبوريم كان الأكثر تواجداً في جميع عينات الفحص وبنسبة ٨٠%، وكان جنس *Aspergillus* الأспيراجيلس متواجداً بنسبة ٢٠% في عينات بيض دجاج المائدة "البلدي".

من ناحية أخرى تم فصل أنواع مختلفة من سموم الأفلاتوكسين وهي *AFB1* و *AFB2* و *AFG2* وبنسب مختلفة وهي ٤٥% و ٢٠% و ١٥% على التوالي، بينما لم تتواجد سموم الأفلاتوكسين من نوع *AFG1* في عينات بيض دجاج المائدة "البلدي".

بينما تم فصل سموم الأفلاتوكسين من نوع *AFB2* بنسبة أقل كانت ١٠%، ولم يستدل على وجود سموم الأفلاتوكسين من نوع *AFB1* و *AFG1* و *AFG2* في عينات بيض دجاج المائدة "البلدي". هذا وقد تم مناقشة النتائج والأهمية الصحية العامة ومدى خطورة وتأثير السموم الفطرية على صحة الإنسان وما يجب أن يتبع لإنتاج بيض مائدة أقل تلوثاً بالسموم الفطرية.