Assiut Vet. Med. J. Vol. 36 No. 72, January 1997

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MYCOLOGICAL QUALITY OF COMMERCIAL HEN'S EGGS

(With 3 Tables)

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(Received at 13/12/1996)

التقييم الميكولوجى لبيض الفراخ التجارى

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يعتبر البيض من أهم مصادر الغذاء للانسان ، إلا أنه قد يكون ملوثا بالفطريّات التى تعد من أخطر الميكروبات التى يتعرض لها الانسان والحيوان على حد سواء . لذلك تم إجراء هذه الدراسة لتقييم بيض الفراخ من الناحية الميكولوجية وذلك بفحص ٢٢٥ عينة جمعت عشوائيا من مدينة أسيوط لعد وعزل وتصنيف الفطريات الملوثة لكل من قشرة ومحتويات البيض. وقد تبين من الفحص أن متوسط العدد الكلى للفطريات هو ٣ × ١٠^{٥} لكل قشرة ، ٤ × ١٠^{٢} لكل مللى فى كل من قشرة ومحتويات البيض على التوالى ، وهذا يوضح تزايد عدد الفطريات المعزولة من قشرة البيض عنها فى محتوياتها . وأظهرت النتائج أيضا أن اكثر هذه الفطريات تواجدا هو فطر *Aspergillus* بنسبة ٦٣,٦٨٪ فى قشرة البيض و ٥١,٧٥٪ فى محتويات البيض ، ممثلا أساسا فى *A. flavus, A. niger* . بينما أمكن التوصل الى عزل وتصنيف ١٤ نوع تعزو الى ٩ أجناس من الفطريات ، ١٣ نوع تعزو الى ٨ أجناس من كل من قشرة ومحتويات البيض ، على التوالى . من هذه الأنواع *Penicillium, Cladosporium, Alternaria, Aphanoascus, Cochliobolus, Fusarium, Epicoccum, Rhizopus Vlocladium, Drechslera, Mucor* ولكنها بنسب قليلة. وقد تم عزل *Acremonium fusidioides* من محتويات البيض وذلك لأول مرة فى محافظة أسيوط. هذا وقد تم مناقشة تأثير الفطريات التى تم عزلها على صحة المستهلك وكذلك الخسارة الاقتصادية الناتجة من فساد البيض .

SUMMARY

A total of 225 samples of commercial hen's eggs (45 group) were collected randomly from different groceries, and farmers houses in Assiut City supermarkets, and examined to evaluate the mycological quality of both shells and egg contents. The mycological analysis, reveals that 38 (84.44%) and 22 (48.88%) of egg shell and content samples were positive for molds, respectively, with an average counts of 3 x 10^5/shell and 4 x 10^2/ml. The identification of molds reveals that, 66 species belonging to 10 genera were isolated from egg shells, while in the egg contents 58 species belonging to 9 genera were identified. *Aspergillus* was found to be the most prevalent genus recovered from shells and contents in percentages of 63.68% and 51.75%, respectively. Both *Cladosporium* and *Penicillium* were constituting 9.09% of egg shell samples, however, *Penicillium* recovered from egg contents in a percentage of 3.44% while *Cladosporium* was existed in 15.53%. The infrequent genera were *Alternaria, Aphanoascus, Cochliobolus, Fusarium, Epicoccum, Rhizopus* and *Vlocladium* which were isolated from shells, while those recovered from content were *Acremonium* (*A. fusidioides* was a new record in Assiut), *Alternaria, Drechslera, Fusarium, Mucor* and *Rhizopus*. The public health implications and suggestive measures for improving the hen's eggs quality are discussed.

Key words: *Hen's eggs-Quality-Mycology.*

INTRODUCTION

It is generally accepted that milk and eggs are nature's most perfect food. The contents of the eggs are as rapidly perishable as milk, yet the fragile shell, if dry undamaged will usually keep the egg edible for long time. Although hen's eggs are remarkable natural packages, their deterioration may occur due to the penetration of different microorganisms to egg contents as a result of bad handling and storage.

Molds as undesirable microorganisms are widely distributed in nature as environmental contaminants of air, water, soil and dust. They can grow on a variety of substrates and under a diversity of conditions of moisture, pH and temperature. However, only a relatively small proportion is responsible for spoilage of food and feed materials, they cause a serious economic losses. Furthermore, certain molds are capable of producing toxic and carcinogenic metabolites (Chapman et al., 1983) or mycotoxins (Gourama & Bullerman, 1995a) which could be regarded as potential health hazard. Also, they are responsible for many serious diseases of liver, kidney, blood circulation system and blood forming organs (Cole & Cox, 1981) as well as carcinogenic effect (Mossel, 1982).

Unfortunately, eggs are susceptible to fungal contamination at different stages till consumption (Fajardo et al., 1995). Various species of molds that causing moldiness of eggs could be isolated (Frazier, 1967). Also, different types of molds were recovered from deteriorated eggs examined by Ahmed et al. (1974) and Moursy *et al.* (1982). Moreover, Ahmed et al. (1987) recorded that the mean value of yeast and mold count/ml of egg contents was 1×10^2. While El-Prince (1988) found that the average counts of these microorganisms recovered from egg shells and contents collected in summer were 7.4×10^4/shell and 9.8 x 10/ml, respectively.

This work was planned to evaluate the mycological quality of commercial hen's eggs sold in Assiut City.

MATERIAL and METHODS

Collection of samples:

A total of 225 samples commercial hen's eggs (45 group) were collected randomly from different groceries, and farmers houses in Assiut City supermarkets. Every 5 eggs (one group) were placed in a sterile plastic bag and dispatched to the laboratory with a minimum of delay, where they were prepared and examined for the presence of different types of molds.

Preparation of samples:

I - Egg shells:

Egg shells were tested by a surface rinse method as described by Moats (1979).

II- Egg contents:

The egg contents were prepared according to Speck (1976).

Dilutions:

Ten-fold serial dilutions up to 10^{-6} were aseptically prepared from the rinse solution, as well as from the homogenized egg contents using sterile 0.1% peptone water. Molds count was determined by using Malt extract agar according to Harrigan and McCance (1976).

Identification of the isolated strains:

Suspected mold strains were identified on glucose-Czapek's agar medium according to Raper and Fennell (1965), Ellis (1976), Booth (1977), Pitt (1979), Domsch et al. (1980), Sivanesan (1984) and Moubasher (1993).

RESULTS

The obtained results were summarized in Tables 1 - 3.

Table 1. Statistical analytical results of molds recovered from commercial hen's eggs.

Samples	No. of examined samples	Positive samples		Count / ml		
		No.	%	Min.	Max.	Average
Egg shell	45	38	84.44	$1x10^2$	$1x10^7$	$3x10^5$
Egg content	45	22	48.88	5x10	$3x10^3$	$4x10^2$

Table 2. Incidence of isolated molds recovered from egg shell.

Mold genera and species	Number of cases	%
Alternaria species		
Alternaria alternata (Fr.) Keissler	3	4.55
Aspergillus species		
A. flavus Link	12	18.20
A. flavus var columnaris Raper & Fennell	4	6.06
A. fumigatus Fresenius	2	3.03
A. japonicus Saito	4	6.06
A. niger Van Tieghem	18	27.30
A. versicolor (Vuill) Tiraboschi	2	3.03
Aphanoascus species		
A. fulvescens (cock) Apinis	1	1.51
Cladosporium species		
C. cladosporioides (Fries) de Vires	2	3.03
C. sphaerospermum Penz	4	6.06
Cochliobolus species		
C. spicifer Nelson	1	1.51
Fusarium species		
F. moniliforme Sheldon	2	3.03
F. oxysporum Schlecht. Fr.	1	1.51
Epicoccum species		
E. purpurascens Ehrenb. Fr.	1	1.51
Penicillium species		
P. chrysogenum Thom	1	1.51
P. corylophilum Dierckx	3	4.54
P. jenseni Zaleski	1	1.51
P. simplicissimum (Oud.) Thom	1	1.51
Rhizopus species		
Rh. stolonifer (Ehrenb.) Lindt	2	3.03
Vlocladium species		
V. alternariae (Cke.) Simmons	1	1.51
Total	**66**	**100.00**

Table 3. Incidence of isolated molds recovered from egg content.

Mold genera and species	Number of cases	%
Acremonium species		
* *A. fusidioides* (Nicot) W. Gams	1	1.72
A. strictum W. Gams	6	10.34
Alternaria species		
A. alternata	2	3.45
A. tenuissima (Kunze : Pers) Wiltschire	1	1.72
Aspergillus species		
A. flavus Link	7	12.07
A. flavus var columnaris Raper & Fennell	5	8.63
A fumigatus Fresenius	3	5.17
A japonicus Saito	2	3.45
A. niger Van Tieghem	9	15.53
A. parasiticus	2	3.45
A. sydowii (Bain. & Sart.) Thom & Church	2	3.45
Cladosporium species		
C. cladosporioides	5	8.63
C. sphaerospermum Penz	4	6.90
Drechslera species		
D. halodes (Drechsler) Subramanin & Jain	3	5.17
Fusarium species		
F. equseti Miller, Giddens & Foster	1	1.72
F. moniliforme Sheldon	1	1.72
Mucor species		
M. hiemalis Wchmer	1	1.72
Penicillium species		
P. corylophilum	1	1.72
P. jenseni	1	1.72
Rhizopus species		
Rh. stolonifer	1	1.72
Total	**58**	**100.00**

* New Record in Assiut.

DISCUSSION

The results recorded in Table 1, reveal that mold counts of the examined egg shell and content samples ranged from 1 x 10^2 to 1 x 10^7/shell and 5 x 10 to 3 x 10^3/ml with an average count of 3 x 10^5/shell and 4 x 10^2/ml, respectively. Nearly similar results were obtained by Ahmed et al. (1987) and El-Prince (1988).

The incidence of molds given in Table 2 and 3 reveal that, *Aspergillus* was the most prevalent genus encountered in egg shell and content samples comprising 63.68 and 51.75% of total molds, respectively. This genus is represented mainly by *A. niger* and *A. flavus* 27.30 & 15.53 and 18.20 & 12.07% of total molds which recovered from egg shells and contents, respectively. While, *A. flavus var columnaris*, *A. fumigatus*, *A. japonicus*, *A. versicolor*, *A. parasiticus* and *A. sydowii* were infrequently recovered from the examined samples.

Species of *Aspergillus* were known to be common contaminants of human food and animal feeds (Gourama & Bullerman, 1995 b). In animals, the pathogenic *Aspergilli* induce various clinical forms of avian aspergillosis and bovine mycotic abortion (Ainsworth & Austwick, 1973). Moreover, the presence of *Aspergilli* is not only of economic importance but also represents a real hazard to human health. They can be of allergic, toxigenic and pathogenic effect through production of mycotoxins which cause mycotoxicosis in animals (Bullerman, 1979) and implicated in human cases of food poisoning and neoplastic diseases, such as leukemia and other cancers (Deger, 1976; Stoloff, 1976 and Bullerman, 1980). Furthermore, *A. niger* was encountered in otomycosis (Rippon, 1982). Also, these toxigenic *Aspergilli* can grow on food during storage at a suitable temperature, and produce aflatoxins (Applebaum et al., 1982).

Both *Cladosporium* and *Penicillium* constituted 9.09% of the examined egg shell samples (Table 2). *Penicillium* was represented by four species, *P. corylophilum* (4.54%) and *P. chrysogenum*, *P. jenseni* and *P. simplicissimum* (1.51%), while *Cladosporium* included two species *C. cladosporioides* (3.03%) and *C. sphaerospermum* (6.06%). However, *Penicillium* species recovered from egg contents in a lower percentage (3.44%) while *Cladosporium* was detected in a higher percentage (15.53%) as compared with those recovered from

egg shells (Table 3). It has been stated that some species of *Penicillium* were found to be associated with pulmonary and urinary tract infections as well as "yellow rice disease" causing several deaths in man (Banwart, 1980).

The examination of egg shells reveals that, *Alternaria alternata*, *Aphanoascus fulvescens*, *Cochliobolus spicifer*, *Fusarium moniliforme*, *Fusarium oxysporum*, *Epicoccum purpurascens*, *Rhizopus stolonifer* and *Vlocladium alternariae* were infrequently encountered (Table 2). Contamination of egg shells with yeasts and molds had been detected previously by Romanoff and Romanoff (1949) and Garibaldi and Stokes (1958).

As recorded in Table 3, another 9 species belonging to 6 genera were existed in percentages of 12.06% for *Acremonium* and 17.22% for the other genera recovered from egg contents. *Acremonium* was represented by *A. strictum* (10.34%) and *A. fusidioides* (1.72%) which is thought to be a new record in Assiut.

Molds causing moldiness of eggs include various species such as *Penicillium*, *Cladosporium*, *Mucor*, *Sporotricum*, *Thamnidium*, *Botrytis*, *Alternaria* and other genera (Frazier, 1967). In addition, yeasts and molds were established as a cause of spoilage and could be isolated from rotten eggs (Ahmed et al., 1974; Frazier & Westhoff, 1978 and Moursy *et al.*, 1982).

The assesment of the obtained resutls allow to conclude that some of the encountered molds are known to be mycotoxin producers, which can threaten human health. The contamination of eggs with these microorganisms is an indicative of bad sanitary measures adopted in poultry farms. Therefore, to safeguard consumers from being infected and to safe eggs from being deteriorated, efforts should be made to prevent mold growth. This could be accomplished by proper sanitation and quality control of eggs.

REFERENCES

Ahmed, A.A.; Abou-Gabal, M.; Enab, S.M. and Moustafa, T.H. (1974): Investigation of microbial spoilage of hen eggs. Assiut Vet. Med. J., 1(1&2): 77-84.

Ahmed, A.A-H.; Saad, Nagah M. and Moustafa, M.K. (1987): Microbial contamination of market hen eggs. Assiut Vet. Med. J., 18 (36): 125-131.

Ainsworth, G.C. and Austwick, P.K.C. (1973): Fungal diseases of animals, 2nd ed. Common Wealth Agricultural Bureau, Slough, U.K.

Applebaum, R.S.; Brakett, R.E.; Wieserman, D.W. and Marth, E.H. (1982): Responses of dairy cows to dietary aflatoxin feed intake and yield, toxic content, and quality of milk of cows treated with pure and impure aflatoxin. J. Dairy Sci., 65: 1503-1508.

Banwart, G.J. (1980): Basic food microbiology. A bridged text book Ed. Avipublishing Co., INC., Westport, Connecticut, USA.

Booth, C. (1977): Fusarium: Laboratory guide to identification of the major species. Common Wealth Mycological Institute, Kew, Surrey, England, 1-58.

Bullerman, L.B. (1979): Significance of mycotoxins to food safety and human health. J. Food Prot., (42) 1: 65-86.

Bullerman, L.B. (1980): Incidence of mycotoxic molds in domestic and imported cheeses, J. Food Safety, 2 (1): 47-58.

Chapman, W.B.; Sara, J. Cooper; Norton, D.M.; Williams, A.R. and Jarvis, B. (1983): Mycotoxins in molded cheeses. Proc. Int. Symp. Mycotoxins. 363-373.

Cole, R.J. and Cox, R.H. (1981): Handbook of toxic fungal metabolites. New York, Academic Press.

Deger, G.E. (1976): Aflatoxin-human colon carcinogenosis. Ann. Infern. Med., 774.

Domsch, K.H.; Gams, W. and Anderson, T.H. (1980): Compendium of soil fungi. Academic Press, London, 1-859.

Ellis, M.B. (1976): More *Dematiaceous Hyphomycetes*. Common Wealth Mycological Institute, Kew, Surrey, England, 1-507.

El-Prince, Enas (1988): Microbiological quality of hen's eggs. M.V.Sc. Thesis. Fac. Vet. Med., Assiut Univ., Egypt.

Fajardo, T.A.; Auantheswaran, R.C.; Puri, V.M. and Kanbel, S.J. (1995): Penetration of *Salmonella enteritidis* into eggs subjected to rapid cooling. J. Food Prot., 58 (5): 473-477.

Frazier, W.C. (1967): Food Microbiology, 2nd ed., TATA, McGraw Hill Publishing Co. Ltd. New Delhi.

Frazier, W.C. and Westhoff, D.C. (1978): Food Microbiology. 3rd ed., TATA. McGraw Hill INC. New York. Contamination, preservation and spoilage of eggs. P. 256-269.

Garibaldi, J.A. and Stokes, J.L. (1958): Protective role of shell membranes in bacterial spoilage of eggs. Food Res., 23: 283-290.

Gourama, H. and Bullerman, L.B. (1995 a): Detection of molds in foods and feeds: Potential rapid and selective methods. J. of Food Prot. 58 (12): 1389-1394.

Gourama, H. and Bullerman, L.B. (1995 b): Aspergillus flavus and *Aspergillus parasiticus*: Aflatoxigenic fungi of concern in foods and feeds: A review. J. of Food Prot., 58 (12): 1395-1404.

Harrigan, W.F. and McCance, M.E. (1976): Laboratory methods in food and dairy microbiology. Academic Press, London, New York, San Francisco.

Moats, W.A. (1979): The effect of washing eggs under commercial conditions on bacterial loads on egg shells. Poult. Sci., 58: 1228-1233.

Mossel, D.A.A. (1982): Microbiology of foods, 3rd ed. The Univ. of Uterecht., The Netherlands. ISPN.

Moubasher, A.H. (1993): Soil fungi in Qatar and other arab countries. The Scientific and Applied Research Centre, University of Qatar, Doha, Qatar, 1-566.

Moursy, A.W.; Al-Ashmawy, A.M. and Moursy, E.A. (1982): Microbiological studies on deteriorated hen eggs. Assiut Vet. Med. J., 9: 91-96.

Pitt, J.I. (1979): The genus *Penicillium* and its teleomorphic states *Eupenicillium* and *Talaromyces*. Academic Press, London, 1-398.

Raper, K.B. and Fennell, D.J. (1965): The genus *Aspergillus*. Williams and Wilkins, Baltimore, USA, 1-878.

Rippon, J.W. (1982): Medical mycology. The pathogenic fungi and pathogenic *Actinomycetes*. W.B. Saunders Co., Philadelphia.

Romanoff, A.L. and Romanoff, A.J. (1949): Avian egg, June Wiley & Sons, Inc. New York, Chapman, Limited, London.

Sivanesan, A. (1984): The bitunicate *Ascomycetes* and their anamorphs. Strauss and Cramer GmbH, Germany, 1-701.

Speck, M.L. (ed) (1976): Compendium of methods for the microbiological examination of foods. American Public Health Association, Washington, DC.

Stoloff, L. (1976): Occurrence of mycotoxins in foods and feeds, p. 23-50. In: J.V. Rodricks (ed), Mycotoxins and other fungal related food problems. Adv. Chem. Ser. 149. American Chemical Society, Washington, D.C.