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MYCOLOGICAL STUDY ON SOME PROCESSED MEAT PRODUCTS EXPOSED FOR SALE IN MARKETS

(With 6 Tables)

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دراسة فطرية على بعض منتجات اللحوم المصنعة المعروضة للبيع فى الأسواق

عزة على حسين التابعى

تعتبر منتجات اللحوم من أهم وأرخص مصادر البروتين الحيوانى ولما كانت منتجات اللحوم وسط مناسب لنمو وإنتشار الفطريات حيث أنها تقاوم الظروف البيئية الغير مناسبة فإن لها تأثير كبير فيما تسببه من مشاكل فى منتجات اللحوم من حيث فساد المنتج أو تكون نكهات غير مرغوبة أو الإضرار بصحة المستهلك ، لذا أجريت هذ الدراسة لمعرفة مدى تواجد الفطريات وسمومها فى بعض منتجات اللحوم المعدة للبيع بمدينة بورسعيد ، حيث تم جمع ٣٠ عينة من منتجات اللحوم بواقع ١٠ عينات من كل من اللانشون والسجق المجمد واللحم المفروم. وقد أسفرت النتائج على أن متوسط العد الكلى الفطريات كان $٢,٤ \times ١٠^{٣}$ ، $٢,٩ \times ١٠^{٢}$ و $٤,٩ \times ١٠^{٢}$ ومتوسط العد الكلى للخمائر $٤,٤ \times ١٠^{٣}$ ، $١,٨ \times ١٠^{٢}$ و $١,٤ \times ١٠^{٣}$ فى كل من اللانشون والسجق المجمد واللحم المفروم على التوالى. وقد تم تصنيف الفطريات والخمائر المعزولة من منتجات اللحوم. وقد أمكن إنتاج الأفلاتوكسين من مجموعة الأسبرجلس فلافس والباراسيتكس. وقد دلت النتائج على تواجد السموم الفطرية بنسب ومقادير مختلفة ، مما يسبب خطورة على صحة المستهلك وخصوصا عند إستهلاكها لفترات طويلة، حيث أنها تسبب أمراض خطيرة من أهمها مرض السرطان عامة وسرطان الكبد خاصة. وقد نوقشت الأهمية الصحية لتواجد الفطريات وسمومها والإحتياطات اللازمة لمنع التلوث بها وعلاقة ذلك بصحة المستهلك.

SUMMARY

A total of thirty samples of luncheon, frozen sausage and minced meat (10 of each) were collected from different groceries of different sanitation levels at Port-Said city for estimation of their mycological profiles. The mean values of total mould count were 2.4×10^3,

2.9 X 10^2 and 4.9 X 10^2 CFU/g and the mean values of yeast count were 4.4 X 10^3, 1.8 X 10^2 and 1.4 X 10^3 CFU/g for luncheon, frozen sausage and minced meat respectively. The isolated mould genera from luncheon, frozen sausage and minced meat were belonged to genera *Aspergillus, Penicillium, Alternaria, Fusarium, Cladosporium* and *Mucor*, while that of yeasts were *Saccharomyces, Candida* and *Torulopsis*. The isolated *A. flavus, A.parasiticus* were capable to produce aflatoxins. The presence of aflatoxins residues in the examined samples were also investigated. The public health significance of mycotoxins and measures to prevent contamination of meat products with mycotoxins were discussed.

Key words: *Mycology, meat products, luncheon, sausage, minced meat.*

INTRODUCTION

Meat products are the most palatable and fast food meal. They are considered the best alternative for the fresh meat due to their low price, easily preparation and palatability. On the other hand, meat products are an ideal medium for growth of different microorganisms, as they are rich in nutritious elements and moisture. Mould and yeast comprise a large group of microorganisms which are widely distributed in nature, they are responsible for a major portion of food deterioration in developing countries due to the contamination and suitable environmental condition predisposing their growth (Frazier and Westhoff 1988).

Fungal contamination is considered as one of the important spoilage agent of meat and meat products, and may occur during slaughtering of animals, transportation or processing of meat products through the use of contaminated equipments or other additives and spices as they considered the most important sources of mould contamination in meat products (Scott and Kennedy, 1973, Flannigan and Hui, 1976, Misra 1983 and Abdel-Rahman, 1987).

Mycotoxins occurrence and related toxic effects in humans and animals represent a major part of food safety (FAO, 1999). Aflatoxins are toxic and thermostable chemical compounds produced by toxigenic strains of *Aspergillus flavus* and *A.parasiticus*.

The present work aimed to study fungal contamination and aflatoxin residues of some meat products marketable in Port-Said groceries, with reference to their significance for human health.

MATERIALS and METHODS

A total of 30 meat products samples (luncheon, frozen sausage, and minced meat, 10 of each) were collected randomly from different groceries distributed in Port-Said city. The collected samples were transferred immediately to the laboratory and subjected to mycological investigations.

Mycological analysis:

1- All samples were analyzed mycologically for enumeration of fungi on dicloran rosebengal medium as prepared by King *et al.*, (1979).

2- Identification of the isolated fungi was done according to Samson *et al.*,(1981).

3- Screening of the isolated *Aspergillus* for production of aflatoxins was done according to Davis *et al.*, (1966).

4- Detection and determination of aflatoxins residues: Aflatoxins standards were obtained from Sigma chemical company. The standards were prepared and calculated to give a concentration of 0.2 µg/ul. in meat products using thin layer chromatography method according to the technique recommended by AOAC (1990).

RESULTS

Table 1: Statistical analytical results of total mould count (CFU/g.) of the examined meat products samples (n= 10 of each)

Meat products samples	Positive samples		Mould CFU/g			
	No.	%	Min.	Max.	Mean	± SE
Luncheon	8	80	$2X10^2$	$9X10^3$	$2.4\ X10^3$	$5.7\ X10^2$
Frozen sausage	5	50	4	$9X10^2$	$2.9\ X10^2$	$6.2\ X10^1$
Minced meat	5	50	$3X10^1$	$2X10^3$	$4.9\ X10^2$	$9.6\ X10^1$

Table 2: Statistical analytical results of total yeast count (CFU/g.) of the examined meat products samples (n=10 of each)

Meat products samples	Positive samples		Yeast CFU/g			
	+ve	%	Min.	Max.	Mean	±SE
Luncheon	7	70	$5X10^2$	$2X10^4$	$4.4X10^3$	$2.5X10^2$
Frozen sausage	3	30	$4X10^1$	$5X10^2$	$1.8X10^2$	$2.9X10^1$
Minced meat	6	60	$3X10^2$	$4X10^3$	$1.4X10^3$	$1.1X10^3$

Table 3: Frequency distribution of mould and yeast species recovered from the examined meat products samples.

Isolated moulds and yeasts	Luncheon		Frozen sausage		Minced meat	
	+ve	%*	+ve	%	+ve	%
Aspergillus flavus	3	21.4	2	28.6	1	12.5
Aspergillus parasiticus	1	7.1	2	28.6	1	12.5
Aspergillus niger	2	14.3	1	14.3	2	25
Penicillium spp.	4	28.6	-	-	1	12.5
Alternaria spp.	-	-	-	-	2	25
Fusarium spp.	1	7.1	-	-	1	12.5
Cladosporium spp.	2	14.3	1	14.3	-	-
Mucor spp.	1	7.1	1	14.3	-	-
Total mould isolates	14	100	7	100	8	100
Saccharomyces	5	62.5	3	75	7	70
Candida spp.	2	25	1	25	1	10
Torulopsis spp.	1	12.5	-	-	2	20
Total yeast isolates	8	100	4	100	10	100

* The percentage related to No. of total isolates of moulds or yeasts in each product.

Table 4: Amounts of aflatoxins (µg/L media) obtained from *Aspergillus* group isolated from meat products.

Aspergillus genera	No. of isolates	Toxigenic strains	Average			
			B_1	B_2	G_1	G_2
A.flavus	6	3	60.0	31.0	33.24	11.1
A.parasiticus	4	2	37.87	20.0	15.65	12.89

Table 5: Incidence of aflatoxins in the examined meat products samples (n=30).

samples	B_1		B_2		G_1		G_2	
	+Ve	%	+Ve	%	+Ve	%	+Ve	%
Luncheon	2	25	1	16.7	3	42.9	-	-
Frozen sausage	6	75	3	50	3	42.9	4	100
Minced meat	-	-	2	33.3	1	14.3	-	-
Total (30)	8	26.7	6	20	7	23.3	4	13.3

Table 6: Statistical analytical results of aflatoxins residues (µg kg) in the examined meat products samples.

Samples	B_1	B_2	G_1	G_2
Luncheon				
Min	0	0	0	-
Max	4.3	3.5	1.8	-
Mean	2.88	0.4	1.6	-
±SE	±0.48	±0.20	±0.06	-
Frozen sausage				
Min	0	0	0	0
Max	12.9	7.5	9.3	2.4
Mean	6.84	3.79	4.97	2.20
±SE	±1.20	±0.54	±1.47	±0.07
Minced meat				
Min	-	0	0	-
Max	-	5.17	6.1	-
Mean	-	2.89	1.54	-
±SE	-	±2.21	±0.40	-

*µg/Kg =ppb

DISCUSSION

Mould affections are considered the most important problems affecting meat and meat products as mould spores are ubiquitous and can grow at various conditions. The present results in Tables (1&2) revealed that luncheon was commonly contaminated by moulds (80%) and yeasts (70%) but frozen sausage had lower incidence (50% and 30%) whereas minced meat had 50% and 60% for moulds and yeasts respectively. The results obtained from luncheon were nearly similar to that recorded by Abdel-Rahman *et. al.*, (1984), Aziz and Youssef (1991) and Sayed *et. al.*, (2000) and it was higher than that obtained by Aiedia (1995), El-Gazzar (1995) and Nouman *et. al.*, (2001a). The results of minced meat were in agreement with that obtained by Leistner and Ayers (1976), Jay (1978), and slightly lower than that obtained by Abdel-Rahman and EL-Khateib (1989) and Abdel-Rahman *et. al.*, (1984). The higher incidence of moulds in luncheon samples could be attributed to the use of different untreated food additives and spices which may be the main source of mould contamination in meat products (El-Khateib *et. al.*, 1987 and Hadlok 1969). Meanwhile, the results in frozen sausage were lower than that obtained by Nouman *et. al.*, (2001 b) and Shaltout (1996).

It is obvious from the results recorded in Tables (1&2) that the mean values of total mould count/g were 2.4 X 10^3, 2.9 X 10^2 and 4.9 X 10^2 while the averages total yeast count/g were 4.4 X 10^3, 1.8 X 10^2 and 1.4 X 10^3 for luncheon, frozen sausage and minced meat respectively. The variation in quantitative estimation of mould counts might be attributed to improper sanitation during slaughter, preparation, manufacturing, additives specially using spices of low quality or during transportation, storage and marketing of the products (Abobaker, 1986, Refai *et al.* 1990, Roushdy *et al.* 1996). Although the total mould count of any food article is not indicative of its safety for consumption yet it is of supreme importance in judging the hygienic condition under which it has been produced ,handled and stored (Martin and Lowery, 1992).

Table (3) illustrated the frequency distribution of mould and yeast genera isolated from the examined meat products. The predominant mould species recovered from luncheon were *Aspergillus flavus, A. niger*, and *A. parasiticus* with percentage of 21.4, 14.3 and 7.1% respectively followed by *Penicillium* species (28.7%), *Cladosporium* (14.3%), *Fusarium* and *Mucor* (7.1% for each). While the most common species isolated from sausage were *A. flavus* and *A.parasiticus* (28.6% for each). *A. niger* and *Alternaria* were the most isolated species from minced meat samples. The results of mould identification declared that the most predominant mould genera in meat products samples were *Aspergillus* and *Penicillium*, nearly similar results were obtained by Wu *et al.*, (1974), Abdel-Rahman *et al.*, (1984), Beuchat (1987), Lotfi *et al.*, (1987) and Aideia (2005).

Regarding the frequency distribution of yeast genera, the results tabulated in Table (3) revealed that *Saccharomyces* were recovered at rate of 62.5, 75 and 70% from luncheon, frozen sausage and minced meat respectively. *Torulopsis* was recovered from luncheon and minced meat at rate of 12.5 and 20%, respectively. Hessel-Schmal-Fuss, (1976), Jay, (1978) and Abdel-Rahman *et. al.*, (1984) isolated the aforesaid yeast genera from minced meat and luncheon.

Table (4) revealed that *A. flavus* and *A. parasiticus* isolated from the examined samples were capable of production of aflatoxins in high levels. Nearly similar results were reported by Biomy (1993) and Shabana (1995). Therefore, the presence of mycotoxins in this samples were expected.

The results recorded in Tables (5&6) showed the incidence and the mean values of aflatoxins residues among 30 meat product samples. Aflatoxin B_1 (AFB_1) was the major contaminant with an overall

incidence of 26.7% (8 out of 30 samples). The mean values of AFB_1 were 2.88±o.48 and 6.84±1.20 µg/kg for luncheon and frozen sausage respectively. The incidence and mean value of AFB_2 were lower than AFB_1, where 6 samples (20%) had AFB_2 with mean values of 0.4±0.20, 3.79±0.54 and 2.89 ±2.21. AFG_1 was found in 7 samples (23.3%) with mean values of 1.6 ±0.06, 4.97±1.47 and 1.54±0.40 µg/kg luncheon, frozen sausage and minced meat respectively while AFG_2 was found in frozen sausage only with a mean value of 2.20±0.07 µg/kg. The variations in results may be due to the fact that the level of contamination with aflatoxins in feed stuffs is an important factor influencing tissue aflatoxin level (Cespedes and Diaz, 1997, Bintvihok *et al.*, 2002). In toxicity and carcinogenicity aflatoxin B_1 demonstrate greater activity than G_1, aflatoxins B_2 and G_2 are many times less toxic (Van rensburg, *et al*, 1985). The low levels of aflatoxins in luncheon may be attributed to the addition of curing ingredients specially sodium nitrite which tend to reduce aflatoxin production (Bullerman *et. al.*, 1969) while Martin and Lowery (1982) recorded that the casing of meat products prevent aflatoxins production. The higher incidence of toxin residues in sausage was attributed to the use of garlic which was found to stimulate toxin production (Bullarman *et. al.*, 1969).

The present results showed that the aflatoxins extracted from luncheon, frozen sausage and minced meat were lower than the permissible limits (20 µg/kg) ,which was recommended by FDA (2002) for aflatoxins in human foods. In spite of these results the aflatoxins residues is still very dangerous due to the repeated exposure of consumers to low concentration of the toxins results in over toxicosis (Stephan and Charles 1985). Exposure to mycotoxins can produce both acute and chronic toxicities ranging from death to deleterious effects upon the central nervous, cardiovascular and pulmonary systems and upon the alimentary tract. Mycotoxins may also be carcinogenic, mutagenic, teratogenic and immunosuppressive. Also mycotoxins have the ability to reduce the resistance to infectious disease, is now widely considered to be the most important effect of mycotoxins particularly in developing countries (FAO 2001).

The occurrence of moulds and mycotoxins can be alleviated by the variety of prevention measures along the entire food chain would be to prevent its formation in crop production and/or during storage of feed stuffs (Meier *et. al.*, 2000, Obst *et al.*, 2000). The animal feed stuffs must be checked periodically for the presence of mycotoxins. Also meat

and meat products should be stored in suitable freezing chambers at -18°C.

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