OCCURRENCE OF ENTEROTOXIGENIC STAPHYLOCOCCUS AUREUS IN SOME CHEESE VARIETIES IN ASWAN CITY – UPPER EGYPT

LAILA M. EL MALT
Department of Food Hygiene, Faculty of Veterinary Medicine, South Valley University, Qena Upper Egypt.
Email: lailael.malt@ymail.com

Received at: 21/9/2013

Accepted: 24/10/2013

ABSTRACT

A total of 120 random samples of some cheeses were collected from various dairy shops, street vendors and supermarkets located in Aswan city (upper Egypt) and The samples were examined microbiologically for enumeration, *isolation and identification of Staph. aureus* and *detection of enterotoxigencity of the isolated Staph. aureus strains* These samples included fresh Kareish, pickled Kareish, Domiati and Processed cheese (30 samples each).' Of the examined cheese samples 28(93%), 25(83%), 21(70%) and 6(20%), respectively were contaminated with Staph.aureus. With mean counts of 3.08 X 10^6 ± 1.79 $x10^6$, 3.40 x 10^5 ± 1.59$x10^5$, 7.24 $x10^5$± 4.10 $x10^5$ and 1.15 $x10^2$±8.98, respectively. Incidence of coagulase positive and coagulase negative strains of S. aureus in the examined cheese samples were (15 (54%) and (13 (46 %), (7 (28%) and (18(72%), (13(62%) and (8 (38%) and (0% and 6(100%), respectively. Methicillin resistant *S. aureus* (MRSA) was isolated in an incidences of 8(26%), 7(23%), 9(30%). and 0(0%) from examined cheese samples respectively while vancomycin resistant Staph aureus (VRSA) was isolated in an incidences of 3(10%), 5(17%), 8(27%) and 0(0%). Three out of twelve strains of MRSA and VRSA were isolated from fresh Kareish cheese 4 strains from Pickled Kareish cheese and 5 strains of Domiati cheese. Two strains produced enterotoxins A were isolated from fresh Kareish cheese which can also synthesis entertoxins B and another 2 strains isolated from Domiati cheese produce entertoxins A and also one strain synthesiss entertoxins B, entertoxins C, and enterotoxins D. The selected 12 strains were strongly produce coagulase only 4 strains were enterotoxigenic.

Key words: *Staph.aureus, Enterotoxigenic, cheese.*

INTRODUCTION

Cheeses are ready to eat food products that do not undergo any further treatment to ensure their safety before consumption Although cheeses have been characterized as one of the safest food products by some authors (Little *et al.*, 2008). In 2006 the consumption of contaminated cheese accounted for the 0.4% of the total food borne outbreaks in Europe (European Food Safety Authority "EFSA", 2008), furthermore, the scientific literature has reported severe food poisoning out breaks associated with various types of cheese. (Kongo *et al.*, 2008).

Staphylococcus aureus is a leading cause of gastroenteritis resulting from the consumption of contaminated food. Staphylococcal food poisoning is due to the absorption of Staphylococcal enterotoxins preformed in the food (Loir *et al.*, 2003)

Many contaminants find their way to raw milk, from which they gain access to dairy products (Al–khatib and Al-Mitwalli, 2009). Chapaval *et al.*, (2010) found *staphylococcal* enterotoxins in milk when milk was stored at temperatures of 37 °C to 42 °C or when exposed to variations in temperature.

On heating at normal cooking temperature, the bacteria may be killed but the toxins remain active (Presscott *et al.*, 2002). Staphylococcal enterotoxins are highly heat resistant and are thought to be more heat resistant in foodstuffs than in a laboratory culture medium (Bergdoll 1983).

Besides enterotoxins producing S. aureus are most dangerous and harmful for the human health about 50 % of this organism are able to produce enterotoxins associated with food poisoning (Payne, 1974). Illness through S. aureus ranges from minor skin infection such as pimples, boils, cellulites, toxic shock syndrome impetigo, and abscesses to life threatening

disease such as pneumonia, meningitis, endocarditis, and septicemia (Soomro *et al.*, 2003).

SEA and SED are the most common Ses involved in food poisoning outbreaks (Genigeorgis, 1989). Growth of enteroxigenic strains of *Staph. aureus* to a population of 10^6 or more cells/g of food is generally considered necessary for production of a sufficient amount of enterotoxins to cause intoxication if the food consumed (Newsome, 1988). The amount of SEA necessary to cause symptoms in humans is about 100ng (Balaban and Rasooly, 2001). The Ses functions as potent gastrointestinal toxins. After ingestion of contaminated food, the toxins are resorbed into the blood in the gastrointestinal tract, activate an emetic reflex and cause nausea, emesis, abdominal cramps an diarrhea (Tortora, 1995) after 2-6 hours of eating contaminated food and the recovery takes 1-3 days, while deaths occur rarely, specifically in very young or old age (Martin and Iandolo, 2000) Time of onset and severity of symptoms depend on the quantity of toxins consumed and the individuals susceptibility (Baird and Lee, 1995).

To detect the level of enterotoxin (100ng) several sensitive detection methods for SE detection have been developed in particular the rapid methods based on specific antibodies such as a radioimmunoassay (RIA) an enzyme linked immunsorbent assay (ELISA) or enzyme immunoassay (EIA) and reversed passive latex agglutination assay (RPLA) are commonly used in staphylococcal intoxication the use of polymerase chain reaction (PCR) and DNA probes is limited in detection only the presence of *Staph aureus* genes, but not in detection the presence of preformed toxins (Di Pinto *et al.*, 2004).

Antimicrobial resistance is a major public health problem in many countries due to the persistent circulation of resistant strains of bacteria in the environment and the possible contamination of bacteria in the environment and the possible contamination of water and food. The development of resistance both in human and animal bacterial pathogens has been associated with the extensive therapeutic use of antimicrobials or with their administration as growth promoters in food animal production (Barber *et al.*, 2003)

The present study was planned to deal with the following:

1-Enumeration, isolation and identification of *staph. aureus* in some cheese varieties sold in Aswan city.

2- Identification of antibiotics resistant strains of *staph. aureus.*

3- Detection of enterotoxigencity of the isolated *staph. aureus* organisms

MATERIALS and METHODS

A) Collection of samples:
A total of 120 random samples of some cheese varieties were collected from various dairy shops, street vendors and supermarkets located at Aswan city. These samples included Fresh Kareish, pickled Kareish, Domiati and Processed cheese (30 samples each). Collected samples were transferred in an ice box directly to the laboratory with a minimum of delay to be examined.

B) Preparation of serial dilutions **(ISO 8261. 2001)**:
C) Microbiological examination:

1- Enumeration and isolation of *Staph. aureus (A.*O.A.C., 2000):
Over duplicated plates of a dry surface of Baird Parker (B-P) agar, 0.1 ml from each prepared dilutions of examined samples were transferred and evenly spread using surface plating technique (Thatcher and Clark, 1988).

- The inoculated B-P agar plates were incubated at 37°C for 24-48 hrs. Suspected colonies are circular, smooth, convex, moist, 2-3 mm in diameter, gray to jet black, shiny, with light color (off-white) narrow margin, surrounded by hallow zones and had buttery to gummy consistency when touched with inoculating needle were counted. The plates were then reincubated for additional 18-24 hrs before being counted for further growth, *Staph.* aureus count/ g were recorded.

- Furthermore, an appropriate amount from each prepared sample was inoculated into sterile Nacl 10% broth. Inoculated Nacl 10% broths were inoculated at 37 °C for 24 hrs. A loopful of the incubated broth was streaked onto sterile plates of Mannitol Salt agar. (A.O.A.C., 2000)

Identification of *Staph. aureus* recovered from the examined samples:

A- Morphological characters for all isolates:

1- Staining reaction (A.P.H.A., 2004):-

B- Biochemical reactions:- catalase activity (Koneman *et al.*, 2005), anaerobic mannitol fermentation (Baird-Parker, 1962), coagulase test. according to (Cruickshank *et al.*, 1973).

2- Isolation of antibiotics resistant strain of *staph. aureus.*

a- Identification of methicillin resistant *Staph aureus* (MRSA): (Simor *et al.*, 2001).

The cultures of *Staph aureus were* subcultured on Oxacillin Resistance Screen Agar Base (ORSAB) (Oxoid Limited, Basingstoke, England) containing ORSAB Selective Supplement contained two antibiotics-oxacillin at 2mg/L and polymyxin B

50.000 IU/l. The plate was incubated at 37 ^{O}C for 24-48 h and examined for the presence of MRSA colonies, which were blue on ORSAB.

b- Identification of vancomycin resistant *Staph aureus* (VRSA) ⊗Tiwari and Sen, 2006).

3- Detection of the enterotoxigencity of Staph. aureus (Park *et al.*, 1994)

Using RIDASCREE®SET A, B, C, D, E (Art. No.:R4101) manufactured by R- Biopharm AG, Germany.

Selected strains of the isolated MRSA and VRSA from the examined samples were tested for –ability to produce entertoxins.

RESULTS

Table 1: Statistical analytical results of *Staph.aureus* count in the examined cheese samples.

Examined samples	No. of examined samples	Positive samples		Count /g				No. of samples above E.S.*	
		No.	%	Min.	Max.	Mean	S.E.	No.	%
Fresh Kareish cheese	30	28	93	$1x10^3$	$5.25x10^7$	$3.08x10^6$	$1.79x10^6$	28	93
Pickled Kareish cheese	30	25	83	$4.17x10^2$	$4.70x10^6$	$3.40x10^5$	$1.59x10^5$	25	83
Domiati cheese	30	21	70	1x10	$1.27x10^7$	$7.24x10^3$	$4.10x10^5$	21	70
Processed cheese	30	6	20	1x10	$1.90x10^2$	$1.15x10^2$	8.98	6	20

*E.S.: Egyptian Standard (2005).

Table 2: Frequency distribution of positive cheese samples based on their S.aureus count.

Count /g	Fresh Kareish cheese		Pichled Kareish cheese		Domiati cheese		Processed cheese	
	No./28	%	No./25	%	No./21	%	No./6	%
	-	-	-	-	-	-	-	-
10^1>	-	-	-	-	-	-	-	-
10^2>	-	-	2	8	9	43	6	100
10^3>	8	28.5	11	44	7	33	-	-
10^4>	6	21	2	8	8	14	-	-
10^5>	5	18	9	36	-	-	-	-
10^{6}>	8	28.5	1	-	1	5	-	-
10^7>	1	4	-	4	1	5	-	-

Table 3: Incidence of coagulase positive and coagulase negative strains of *S. aureus* in the examined cheese.

Examined samples	No. of examined samples	No. of isolated S.aureus	coagulase positive		coagulase negative	
			No.	%	No.	%
Fresh Kareish cheese	30	28	15	54	13	46
Pickled Kareish cheese	30	25	7	28	18	72
Domiati cheese	30	21	13	62	8	38
Processed cheese	30	6	-	-	6	100

Table 4: Frequency distribution of MRSA and VRSA and recovered from the examined cheese samples.

Examined samples	No. of isolated S.aureus	MRSA		VRSA		Both	
		No.	%	No.	%	No	%
Fresh Kareish cheese	28	8	29	3	11	3	11
Pickled Kareish cheese	25	7	28	5	20	4	16
Domiati cheese	21	9	43	8	38	3	14
Processed cheese	6	-	-	-	-	-	-

Table 5: Incidence of MRSA and VRSA strains isolated from the examined cheese

Examined samples	No. of examined samples	MRSA		VRSA	
		No.	%	No.	%
Fresh Kareish cheese	30	8	26	3	10
Pickled Kareish cheese	30	7	23	5	17
Domiati cheese	30	9	30	8	27

Table 6: Enterotoxins produced by some strains of *Staph. aureus* isolated from the examined cheese samples.

Examined samples	No. of strains tested	No. of strains producing enterotoxins	Types of produced enterotoxins				
			A	B	C	D	E
Fresh Kareish cheese	3	2	2	1	-	-	-
Pickled Kareish cheese	4	-	-	-	-	-	-
Domiati cheese	5	2	2	1	1	1	-

Table 7: Entertoxins production in relation to coagulase producing *Staph. aureus* strains.

No. of strains tested	Coagulase producing strains.		Enterotoxigenic Staph. aureus strains	
	No.	%	No.	%
12	12	100	4	33.3

DISCUSSION

The results recorded in Table 1showed that 28(93%), 25(83%), 21(70%) and 6(20%) of the examined fresh Kareish, Pickled Kareish, Domiati and processed cheese samples were contaminated with *Staph.aureus*, respectively.

The *S.aureus* count ranged from 1 $x10^3$ to 5.25 $x10^7$ with a mean count of 3.08 X $10^6 \pm 1.79 x10^6$ in fresh Kareish cheese samples, from 4.17 $x10^2$ to 4.70 $x10^6$ with a mean Count of 3.40 x $10^5 \pm 1.59x10^5$ in Pickled Kareish cheese samples, from 1 x10 to 1.27 $x10^7$ with a mean count of 7.24 $x10^5 \pm$ 4.10 $x10^5$ and for Processed cheese the counts ranged from 1 x10 to 1.90 $x10^2$ with a mean count of 1.15 $x10^2 \pm 8.98$ cfu/g.

Egyptian standards (2005) pointed out that the cheese must be free from Staph. aureus and its toxins It was evident that 28(93%), 25(83%), 21(70%) and 6(20%) of the examined fresh Kareish, Pickled Kareish Domiati and processed cheese samples failed to comply with the standerd, respectively.

The results in Table 2, revealed that the highest frequency distribution of fresh Kareish cheese samples was 8(28.5%) lied in the range of 10^3 <10^4 and 10^6 <10^7 cfu/g, while the rest of the positive samples were distributed as 21,18 and 4% lied in between 10^4 <10^5, 10^5 <10^6 and 10^7 <10^8 cfu/g, respectively. In case of Pickled Kareish cheese samples the highest frequency distribution was (44%) lied in the range of 10^3 <10^4 cfu/g, while the rest of the positive samples were distributed as 36,8,8 and 4% lied in between 10^5 <10^6, 10^2 <10^3, 10^4 - <10^5 and 10^7 <10^8 cfu/g, respectively.

For Domiati cheese samples the highest frequency distribution was (43%) lied in the range of 10^2 <10^3 cfu/g, while the rest of the positive samples were distributed as 33, 14 and 10 % lied between 10^3 <10^4, 10^4 <105 and 10^6 <10^7, respectively. While the Processed cheese positive samples all of them lie in range10^2 <10^3 fresh Kareish cheese samples ,comparatively higher counts were obtained by Tawfek *et al.* (1988) Relatively lower counts were detected by Shelaih (1979), Ahmed (1980). Ewida (2009), De Reu *et al.* (2002) and In case of the examined Pickled Kareish cheese samples comparatively lower counts were obtained by Kaldes (1997). While for Domiati cheese samples results, higher counts obtained by Sallam *et al.* (1985). Relatively lower counts obtained by El-Malt (1993) and Hassan (2009). Lower incidence of S.aureus in curd (6.66 %) was reported by Kumar and Prasad (2010) and (3.33 %) Thaker *et al.* (2013). The difference in the prevalence of S.aureus between different types of cheese may originate from the method of manufacture, storage and handling. Turkoglu *et al.* (2001) and Soncini *et al.* (2002) failed to detect the S. aureus in the examined Processed cheese samples.

Fresh Kareish cheese is a popular Egyptian food. It is made at farmers houses from raw skim milk and the fresh product is either consumed fresh or consumed after pickling in its salted fresh whey with added spices and medical plants (mish cheese) so the source of *Staph.aureus* in Kareish cheese are the raw milk, during processing and distribution in village markets these sources lead to the expected high count of *Staph. aureus*. Domiati cheese is considered to be the main national popular pickled soft cheese produced in Egypt. It is traditionally prepared from raw milk without addition of lactic acid culture starters but by addition of appreciable amount of salt to milk before renneting. It can be consumed fresh, but usually it is consumed after pickling in its salted fresh whey for a period of not less than 8 weeks, the product may be soiled with pathogens which enter through different environmental sources that render it a source of infection by many diseases. Genigeorgis (1989) demonstrated that the higher concentration of competing microorganisms in milk, the lower the rate of *S. aureus* growth and SE production. Competition with lactic acid bacteria has been reported in other research on cheese (Otero *et al.*, 1988).

Table 3 showed that 15(54%), 7(28%) and 13 (62 %) of the examined staphylococci strains recovered from fresh Kareish cheese, Pickled Kareish cheese and Domiati cheese, respectively. were coagulases positive. While, 13(46%), 18(72%), 8(38%) and 6(100%) of the isolated strains of staphylococci of the examined cheese, respectively. were coagulase negative. Concerning the result of coagulase positive *S.aureus* in the examined fresh Kareish cheese samples, higher results were obtained by Ahmed (1980), Al-Hawary *et al.* (2009) and Helmy *et al.* (2009). Lower results were detected by Aman (1994); Kaldes (1997) and Kolluman *et al.* (2011).

In case of coagulase positive *S.aureus* in the examined Domiati cheese samples, lower results were detected by Coveney *et al.* (1994), Kaldes (1997); Normanno *et al.* (2005) and Kolluman *et al.* (2011).

Various examples of staphylococcal food poisoning are described in the literature. In one case, cheese was involved in an outbreak because it had been made from unheat treated milk, milk contaminated after pasteurization and before inoculation with lactic starter culture or did not use culture starter. In this particular case, the starter culture did not grow properly, resulting in a fermentation accident that allowed the S. aureus strain to develop and produce SE (Vasavada, 1988) Although milk and milk products are frequently contaminated with S. aureus, dairy products are rarely involved in staphylococcal

food poisoning because the critical cell density of >105 cfu/g-1 is usually not reached (Altekruse *et al.*, 1994).

The result in Table 4 showed that 8(29%), 3(11%) and 3(11%) showed *staph aureus* strains isolated from fresh Kareish cheese were MRSA, VRSA, respectively. While 7(28%), 5(20%) and 4(16%) *Staph aureus* strains isolated from Pickled Kareish cheese were MRSA, VRSA, respectively. The frequency distribution of MRSA and VRSA in the Domiati cheese samples were 9(43%) and 8(38%) while 3(14%) can be resistant for both drugs. In Processed cheese there is no of isolated strains of MARSA, VRSA or resistant for both drugs.

In Table 5 the incidences of MRSA and VRSA from the examined samples were 26%, 3(10%), 7(23%), 5(17%), 9(30%) and 8(27%) of fresh Kareish cheese, Pickled Kareish cheese and Domiati cheese respectively.

Inspection of Table 6 revealed that out of 12 methicillin and vancomycin resistance *Staph aureus* strains tested for enterotoxins production 3 strains of fresh Kareish cheese, 4 strains of Pickled Kareish cheese and 5 strains of Domiati cheese. From fresh Kareish cheese 2 strains were synthesized enterotoxins A and SEB (one strains) in Pickled Kareish cheese there is no strains producing interotoxins. Domiati cheese 2 strains were isolated and produce enterotoxins A(2 strains), SEB (one strain), SEC (one strain) and SED (one strain).

This finding was in contrast to other studies from Spain, Kenya, Switzerland, Brazil, South Korea, the USA, Slovakia and Palestine, where most of the enterotoxigenic *Staph aureus* isolated usually synthesized the toxins SEA, SEC, or SED (Scherrer *et al.*, 2004).

The higher percentages of SEA among Staph aureus strains isolated from milk and milk products may be due to the fact that enterotoxins A are less common among the strains of animals origin than from human origin (Hajek and Marsalek, 1973). These strains of human origin contaminate milk and milk producte during different stages of production or at consumer outlet. On the other side the presence of SEC and SED can be attributed to the increased incidence of *Sraphylococcal* mastitis as enterotoxins C and D were found to be produced by Staph aureus strains isolated from bovine mastitis and were designated as "animal strains" (Olson *et al.*, 1970).

Although the selected 12 isolates were strongly producing coagulase, only 4 were enterotoxigenic which confirm what was stated by A.P.H.A. (1992) and Ryser (2001) Table (7) that attempts to associate enterotoxin production by *Staph aureus* with specific biochemical properties were generally failed. Consequently, confirmation of the toxin by serological or other means provide the only proof that the particular strain is enterotoxigenic.

Park *et al.* (1994) evaluated the RIDASCREEN SET kit for its efficiency. They concluded the major advantages of the kits were a high degree of specify (neither false –positive results due to the growth of nonstaphyloccal microorganisms nor cross –reaction among reagents of the kits was reported), excellent sensitivity, simplicity rapidity (results can obtained in 3h) and semi quantitative results.

CONCLUSION

The obtained results allow concluding that fresh Kareish cheese, Pickled Kareish cheese, Domiati cheese and Processed cheese samples sold in Aswan city markets were produced, handled, packed and distributed under neglected hygienic measures. The information given by the achieved results proved that most of the examined fresh Kareish cheese, Pickled Kareish cheese, Domiati cheese and Processed cheese samples sold in Aswan city market were highly contaminated with high number of Staph aureus which may lead to undesirable changes of these products that render them unfit for consumption and indicate unpersonal hygiene and un- sanitary conditions during processing and handling of such product in compared with examined Processed cheese which show lower incidences or counts.

In the resent years, the extensive therapeutic use of antimicrobials or with their administration as growth promoters in food animal production lead to development of resistance both in human and animal's pathogens as occurred with *Staph aureus*. Nowdays, *Staph aureus* can resist many antibiotics as methicillin and vancomycin which lead to difficulty in their susceptibility of *Staph aureus to these antibiotics and other group of antibiotics. In this study, methicillin resistance* Staph aureus (MRSA) and vancomycin *resistance* Staph aureus (VRSA) could be isolated from examined samples in different percentages.

REFERANCE

A.O.A.C. (2000): Official Methods of Analysis, Food Composition, additives and contamination 7th Ed., Vol. 3 Public Association of Official Analytical Chemists, North Frederick Avenue, Gaitherdburg, Maryland, USA.

A.P.H.A. American Public Health Association"(2004): Standards Methods for Examination of Dairy Products. 17th Ed., American Public Health Association, Washington DC, USA.

A.P.H.A. (1992): Standard Method for the Examination of Dairy Products 16th Ed Vol. 2.

American Public Health Association New York.

Ahmed, A.A.-H. (1980): Studies on the enterotoxigenicity of Staph. aureus isolated from milk and milk products. Ph, D. Thesis, Fac. Vet. Med., Assiut Univ., Egypt.

Al-Hawary, I.I.; Ahmed, H.F.; Ewina, M.A. and El-Sebaey, E.F. (2009): Quality evaluation of some dairy products. Kafr El-Sheikh. Vet. Med. J., 3rd Sci. Congress, 234-252.

Al-Khatib, I.A. and Al-Mitwalli, S.M. (2009): Microbiological quality and sample collection policy for dairy products in Ramallah and Al-Bireh district, Palestine. East Mediterranean Heal., 15: 709-716.

Altekruse, S.; Hyman, F.; Klontz, K.; Timbo, B. and Tollefson, L. (1994): Food borne bacterial infections in individuals with the human immunodeficiency virus. South Med. J., 87: 69-73.

Aman, I.M. (1994): Microbiological quality of Kareish cheese in Kafr El-Sheikh city. Assiut Vet. Med. J., 31: 182.

Baird, R.M. and Lee, W.H. (1995): Media used in the detection and enumeration of Staph. aureus Food Microbiol., 26: 15-24.

Baird-Parker, A.C. (1962): An improved diagnostic and selective medium for isolation of coagulase positive *Staphylococci*. J. Appl. Bacteriol., 25: 12-19.

Balaban, N. and Rasooly, A. (2001): Analytical chromatography for recovery of small amounts of staphylococcal entertoxins from food. Int. J. Food Microbiol., 64: 33-40.

Balaban, N. and Rasooly, A. (2000): Staphylococcal enteroyoxins. Int. J. Food Microbiol., 61: 1-10.

Barber, D.A.; Miller, G.Y. and McNamara, P.E. (2003): Models of antimicrobial resistance and foodborne illness examining assumptions and practical applications. J., Food Prot., 66: 700-709.

Barber, D.A.; Miller, G.Y. and McNamara, P.E. (2003): Models of antimicrobial resistance and foodborne illness examining assumptions and practical applications. J., Food Prot., 66: 700-709.

Bergdoll, M.S. (1983): Enterotoxins. *Staphylococci* and *Staphylococcal* Infections (Easman, C.S.F. and Adlam, C., eds.). *Academic Press, London, UK.*: 559-598.

Chapaval, L.; Moon, D.H.; Gomes, J.E.; Duarte, F.R. and Tsai, S.M. (2010): Efeito da temperatura sobre a producao de enterotoxina estafilococica en leit. Revista Higiene Alimentar., 24: 180-181.

Coveney, H.M.; Fitzgerald, G.F. and Daly, C. (1994): A study of the Microbiological status of Kareish farm house cheese with emphasis on selected pathogenic and spoilage microorganisms. J. Appl. Bacteriol., 77: 621-630.

Cruickshank, R.; Duguid, J.P.; Marmion, B.P.; Mackie, T.J. and Swain, R.H.A. (1973): Medical microbiology: a guide to the laboratory diagnosis and control of infection. 12th Ed., E. and S. Churchill Livingstone Ltd., Edinburgh and London.

De Reu, K.; Debeuckelaere, W.; Botteldoorn, N.; De Block, J. and Herman, L. (2002): Hygienic parameters, toxins and pathogen occurrence in raw milk cheese. J. Food Safety, 22:183.

Di Pinto, A.; Forte, V.T.; Ciccarese, G.; Conversano, M.C. and Tantillo, G.M. (2004): Comparison of reverse passive aggulatination test and immunoblotting for detection of staphylococcal enterotoxins A and B.J. Food Safety, 24(4): 231-238.

European Food Safety Authority (EFSA). (2008): Zoonoses data collection reports. _http://www.efsa.europa.eu/en/science/monitoring zoonoses/reports.html Retrieved 25. 09. 08.

El-Malt, Laila, M. (1993): Search for microorganisms of public health hazard in locally manufactured cheese. M.V.Sc. Thesis, Fac. Vet. Med., Assiut Univ.

EOSQC (2005): Egyptian Organization for Standardization and Quality Control. Egyptian Standards (E.S.): "Domiati cheese 1008-3/ Kareish cheese1008-4/ Ice cream 1185-1/ Yoghurt 1633".

Ewida Rania, M.M.A. (2009): Some studies on staphylococcus aureus in milk and some milk product sold in Assiut city with special reference to antibiotics resistance staphylococcus aureus. Ph. D. Thesis, Fac. Vet. Med., Assiut Univ., Egypt.

Genigeorgis, C.A. (1989): Present state of knowledge on staphylococcal intoxication. Int. J. Food Microbiol., 9: 327-360.

Hajek, V. and Marsalek, E. (1973): The occurrence of enterotoxigenic. Staphylococcus aureus in hosts of different animal species. Zentralblatt f ür Bakteriologie, Parasittenk. Und,Infections Krankheiten und Hygiene. I. Abt: Orig, 223,63 (Cited after Ahmed, 1980 a).

Hassan, Walaa, Sh. (2009): Microbiological evaluation of some white soft cheese locally manufactured and sold in Assiut city. M.V.Sc. Thesis, Fac. Vet. Med., Assiut Univ.

Helmy, A.T.; Emad, M.R. and Sara, M.R. (2009): Prevalence of *Staph. aureus* and its toxins in street marketing milk and some milk products. Alex. J. Vet. Science, 28(1): 93-101.

ISO "International Organization for Standardization" 8261. (2001): General guidance for preparation of test samples, initial suspentions and decimal dilutions for microbiological examination of milk and milk products.

Kaldes, Y.Th. (1997): Microbiological examination of soft cheeses manufactured in Minia city. Assiut Vet. Med. J., 38: 40.

Kolluman, Ahmet; Unlu, Teoman; Dikici, Abdullah; Tezel, Atanur; Nazan Akcelik, E. and Burkan, Zeynep (2011): Presence of Staphylococcus aureus and Staphylococcal enterotoxins in different foods Kafkas Univ. Vet. Fak Derg 17 (Suppl A): S55-S60.

Koneman, E.; Washington Winn, Jr.; Allen, S.; Janda, W.; Procop, G.; Paul, S. and Gail, W. (2005): Color atlas and textbook of diagnostic microbiology. 6th Ed., Lippincott Williams & Wilkins Company, Philadelphia.

Kongo, J.M.; Gomes, A.P. and Xavier, F. (2008): Monitoring and identification of bacteria associated with safety concerns in the manufacture of S¸o Jorge, a Portuguese traditional cheese from Raw cow's milk. Journal of Food Protection, 71, 986–992.

Kumar, R. and Prasad, A. (2010): Detection of *E.coli* and *Staphylococcus* in Milk and Milk Products in and around Pantnagar. Vet. World., 3(11): 495-496.

Little, C.L.; Rhoades, J.R.; Sagoo, S.K.; Harris, J.; Greenwood, M. and Mithani, V. et al. (2008): Microbial quality of retail cheese made from raw, thermised or pasteurized milk in UK. Food Microbiology, 25, 304–312.

Loir, Y.L.; Baron, F. and Gautier, M. (2003): Staphylococcus aureus and food poisoning. Gen. & Mol. Res., 2 (1): 63-76.

Martin, S.E. and Landolo, J.J. (2000): Staphylococcus. In: Encylopedia of Food Microbiology. Robinson, R.K.; Batt, C.A. and Patel, P.D.(eds.). Academic Press, pp.: 2062-2065.

Newsome, R.L. (1988): Staph. aureus. Food Technol., 42:194-185.

Normanno, G.; Firinu, A.; Virgilio, S.; Mula, G.; Dambrosio, A.; Poggiu, A.; Decastelli, L.; Scuota, S.; Bolzoni, G.; Giannatalè, Di.; Salinett, A.P.; La Salandra, G.; Bartoli, M.; Zuccon, F. and Celano, G.V. (2005): Coagulase positive *Staphylococci* and *Staph. aureus* in food products marketed in Italy. International Journal of Food Microbiology, 98: 73-79.

Olson, J.C.; Casman, E.P.; Baer, E.P. and Stone, J.E. (1970): Staphylococcus aureus. In, Doyle, MP (Ed): Foodborne Bacterial Pathogens. pp. 463-523. Marcel Dekker, Inc., New York, NY, USA, 1989.

Otero, A.; Garcia, M.C.; Garcia, M.L. and Moreno, B. (1988): Effect of a cpmmercial starter culture on growth of Staphylococcus aureus and thermonuclease and entertoxins (C1 and C2) production in both cultures. Int. J. Food Microbiol, 6, 107-114.

Park, C.E.; Akhtar, M. and Rayman, M.K. (1994): Evaluation of a commercial enzyme immunoassay kit (RIDASCREEN) for detection of Staphylococcal entertoxins A,B,C,D, and E in foods Appl. Environ. Microbiol., 60(2): 677-681.

Payne, D.N. and Wood, J.M. (1974): The incidence of enterotoxin production in strains of *Staphylococcus aureus* isolated from food. *J. Appl. Bacteriol.*, 37(3): 319-325.

Presscott, L.M.; Harley, J.P. and Klein, D.A. (2002): Text book of Microbiology. *Brown Publishers.* 5th ed.: 441-442.

Ryser, E.T. (2001): Public health concerns In: Applied Dairy Microbiology, Marth, E.H. and Steele, J,I, (eds.).Marcel Dekker, Inc.New York, pp.: 478- 486.

Sallam, S.S.; Hafez, Ragaa, S. and Moursy, A.W. (1985): Staph. *aureus* in some varieties of Egyptian cheese. Vet. Med. J., 33:191.

Scherrer, D.; Corti, S. and Muchlherr, J.E. (2004): Phenotypic and genotypic characteristics of *Staph. aureus* isolates from raw bulk-tank milk samples of goats and sheeo. Vet. Microbial., 101: 101-107.

Shelaih, M.A. (1979): Microbiological studies on Egyptian soft cheese. Ph.D. Thesis, Fac. Vet. Med., Cairo Univ.

Simor, A.E.; Goodffellow, J.; Louie, L. and Louie, M. (2001): Evaluation of a new medium, Oxacillin Resistance Screening Agar Base, for the detection of methicillin resistant Staph. aureus from clinical specimens. J. Clin. Microbio., 39(9): 3422.

Soncini, G.; Valnegri, L. and Marchisio, E. (2002): Research on the quality of industrial milk and cheese products.Industrie alimentary. 2002. 41: 417, 933-936.

Soomro, A.H.; Arain, M.A.; Khaskheli, M. and Bhutto, B. (2003): Isolation of *Staphylococcus aureus* from milk products sold at sweet meat shops of Hyderabad. Online J. Biol. Sci., 3(1): 91-94.

Tawfek, N.F.; Sharaf, O.M. and Hewedy, M.M. (1988): Incidence of pathogens and Staphylococcal enterotoxins in Kareish cheese. Egyptian J. Dairy Sci., 16: 295.

Thaker, H.C.; Brahmbhatt, M.N. and Naya, J.B. (2013): Isolation and identification of Staphylococcus aureus from milk and milk products and their drug resistance patterns in Anand, Gujarat. doi:10.5455/vetworld.10-13.

Thatcher, F.S. and Clark, D.S. (1988): Microorganisms in Foods 1: Their significance and methods of enumeration. Revised 2nd Ed., University of Toronto Press, Toronto, Ontario, Canada.

Tiwari, H.K. and Sen, M.R. (2006): Emergence of vancomycin resistance Staph. aureus (VRSA) from a tertiary care hospital from northern part of India. BMC Infectious Dis., 6:156.

Tortora, G.J. (1995): Staphylococcal food poisoning (Staphylococcal enterotoxicosis) In: Microbiology, Tortora, G.J.; Funke, B.R. and Case, C.L. (eds) Benjamin/ Cummings Publishing Publishing Company, Inc, Redwood City, CA, pp.: 616-618

Turkoglu, H.; Ceylan, Z.G. and Caglar, A. (2001): Some microbiological properties of processed cheese made with vegetable oils at different levels. Ziraat fakultesi dergisi, Ataturk university. 2001.32: 4,447-483.

Vasavada, P.C. (1988). Pathogenic bacteria in milk-A review. J. Dairy Sci., 71: 2809-2816.

تواجد المكور العنقودى الذهبى المفرز للسموم المعوية فى بعض الجبنة المباعة بمحافظة أسوان مصر العليا

ليلى مصطفى كامل الملط
Email: lailael.malt@ymail.com

يعتبر الميكروب المكور العنقودى الذهبى من أهم الميكروبات التى تساهم فى احداث حالات التسمم الغذائى التى اهم اعراضها الاسهال وما زال يسبب الكثير من المشاكل الصحية الاخرى على مستوى العالم مثل تسمم الدم البكتيرى والتهاب عضلة القلب والتهاب العظام. هذا وتعتبر الالبان ومنتجاتها من اهم الاغذية التى يمكن ان يصل اليها الميكروب حيث انه يعيش فى الامعاء والجهاز التنفسى وعلى جلود الانسان والماشية وكذلك أيضاً يوجد فى ضرع الماشية ويسبب الالتهاب الضرعى ولذلك يكون من السهولة تلوث اللبن به. أجريت الدراسة على عدد مائة وعشرين عينة من الجبن المصنع محليا ثلاثون عينة من كل من (الجبن القريش الطازج ، الجبن القريش المملح ، الجبن الدمياطى)، والجبن المطبوخ المصنع فى شركات الالبان وقد جمعت العينات من الباعة الجائلين وأسواق مدينة أسوان بطريقة عشوائية لفحصها وقد تبين بالفحص البكتريولجى أن نسبة تواجد الميكروب المكور العنقودى الذهبى ٢٨(٩٣%) ، ٢٥(٨٣%)، ٢١(٧٠%) و٦(٢٠%) لكل من الجبن القريش الطازج ، الجبن القريش المملح ، الجبن الدمياطى والجبن المطبوخ على التوالى. وكان متوسط العدد الكلى للمكور العنقودى الذهبى لهذه المنتجات على التوالى $٣.٠٨ \times ١٠^{٦} \pm ١.٧٩ \times ١٠^{٦}$ ، $٣.٤٠ \times ١٠^{٥} \pm ١.٥٩ \times ١٠^{٥}$ ، $٧.٢٤ \times ١٠^{٥} \pm ٤.١٠ \times ١٠^{٥}$ و $١.١٥ \times ١٠^{٢} \pm ٨.٩٨$ جم. وقد امكن عزل الميكروب المكور العنقودى الذهبى المقاوم للمثيسيللين من العينات المفحوصة بنسبة ٨(٢٦%) ، ٧(٢٣%) ، ٩(٣٠%) و ٠(٠%) على الترتيب. بينما تم عزل الميكروب المكور العنقودى الذهبى المقاوم للفانكوميسين بنسبة ٣(١٠%)، ، ٥(١٧%)، ٨(٢٧%) و٠(٠%) من العينات المفحوصة على التوالى. وباستخدام اختبار ELISA تم تصنيف السموم المفروزة من اثنى عشرة عترة من الميكروب المكور العنقودى الذهبى المقاوم للمثيسيللين وللفانكوميسين (ثلاثة عترات من جبن القريش ، واربعة عترات من الجبن القريش المملح وخمس عترات من الجبن الدمياطى) وقد وجد ان اربعة عترات من (عترتين تم عزلهم من جبن القريش وعترتين تم عزلهم من الجبن الدمياطى) لهم قدرة على افراز سم A وتم عزل عترة واحدة من جبن القريش لها القدرة على افراز سم B وكذلك عترة من الجبن الدمياطى تفرز ثلاث انواع من السموم B, C&D