

STUDIES ON ENTEROTOXIGENIC STAPHYLOCOCCUS AUREUS IN MILK AND SOME DAIRY PRODUCTS

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ABSTRACT

Staphylococcus aureus is one of the commonest aetiological agents of bacterial diseases. Food intoxication by *S. aureus* was listed as the third etiology of food intoxication all over the world. The current study was designed to record the prevalence of *S. aureus* in milk and some dairy products. A total of 200 samples consisting of (25 raw cow's milk, 25 raw buffalo's milk, 50 kareish cheese, 25 small scale ice-cream, 25 large scale ice-cream, 25 small scale yoghurt and 25 large scale yoghurt samples) were collected from dairy shops and street vendors in Beni-Suef Governorate, Egypt. The prevalence rate of *S. aureus* was 13 (52%), 16 (64%), 34 (68%), 20 (80%), 6 (24%), 22 (88%) and 9 (36%) with an average count of $1.62 \times 10^8 \pm 9.5 \times 10^7$, $7.88 \times 10^7 \pm 5.19 \times 10^7$ CFU/ml, $8.68 \times 10^7 \pm 2.61 \times 10^7$, $6.64 \times 10^7 \pm 3.29 \times 10^7$, $6.52 \times 10^5 \pm 4.41 \times 10^5$, $3.67 \times 10^6 \pm 1.68 \times 10^6$ and $5.27 \times 10^5 \pm 3.45 \times 10^5$ CFU/g in the concerning samples, respectively. The molecular results confirmed that 30% of the examined *S. aureus* strains were enterotoxigenic by PCR technique as carried one or two SE-genes. High *S. aureus* counts in milk and milk products constitute a public health hazard to the consumers and emphasizes the need for improved hygienic standards.

Key words: *S. aureus*, Milk, Kareish cheese, Ice-cream, Yoghurt, Enterotoxins, PCR.

INTRODUCTION

Milk is a highly nutritious food that serves as an excellent growth medium for a wide range of food borne pathogens (Richter *et al.*, 1992). In Egypt, direct consumption of raw milk is much frequent and more popular than consumption of pasteurized one because it's believed, especially in rural areas, that raw milk and its byproducts have nutritional advantages over the pasteurized one. Furthermore, milk is produced mainly by individual in small farms that lack proper sanitary measures and may be either consumed fresh, manufactured into dairy products or sell in retail markets that alarming as a major source of Staphylococcal enterotoxin poisoning (El-Sayed *et al.*, 2011) and represent a serious human health problem.

Kareish cheese is considered one of the most food products rich in calcium and phosphorus. These elements are essential for bones and teeth formation, it is also rich in sodium and potassium, which play an important role in the formation of body liquids and muscles (Mahmoud *et al.*, 2013). Ice-cream is a

nutritionally enriched congealed dairy product consumed by all age groups particularly children during summer (El-Sharef *et al.*, 2006). Yoghurt is the best known of all cultured milk products and the most popular almost all over the world. Consumption of fermented milk products are associated with several types of human health benefits, including it's potential effect against development of colon tumors (Wollowski *et al.*, 2001), reducing risk of hypertension, improving lactose digestion, preventing diarrhea, reducing serum cholesterol (McDonagh *et al.*, 1997).

Dairy products are liable to be contaminated with *S. aureus* from different sources during their production, processing and handling that make them unfit for human consumption or even a dangerous source of infection among consumers constituting a potential health hazard caused by enterotoxin production. This can be occurred under certain conditions during production as well as when they are cut and packaged for consumption (Wauschkuhm, 1970).

S. aureus is a Gram-positive microorganism that colonizes the nasal passages and skin of approximately 50% of healthy individuals. *S. aureus* grows in a wide range of temperatures and pH, from 7 °C to 48.5 °C, and 4.2 to 9.3, respectively. *S.*

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aureus can adapt to grow in various foods and causes food poisoning by secreting enterotoxins (Balaban and Rasooly, 2000).

Staphylococcal Food Poisoning is one of the most commonly occurring Food Borne Disease (FBD) worldwide with high occurrence second to salmonellosis (Aycicek *et al.*, 2005). The significance of finding *S. aureus* in foods suspected of causing staphylococcal poisoning should be interpreted with caution (Robinson and Tamime, 2002). Up to 50% of humans may carry this organism in their nasal passages, throats and on their hair and skin. So it is good indicator of the personal hygiene of the workers with a respiratory infections (Kamat *et al.*, 1991).

The staphylococcal enterotoxins (SEs) are recognized agents of the staphylococcal food poisoning syndrome and may be involved in other types of infections with sequelae of shock in humans and animals (Bergdoll, 1983). Nine major antigenic types of SEs have been recognized and designated SEA, SEB, SEC, SED, SEE, SEG, SEH, SEI, and SEJ (Betley and Mekalanos, 1987). The most common symptoms resulting from the ingestion of food contaminated by SEs are nausea, vomiting, diarrhea and abdominal cramps, which occur within 2–6 hrs of eating SE-contaminated food (Tranter, 1990). Further SE toxins have been identified (SEK, SEL, SEM, SEN, SEO and SEU) (Orwin *et al.*, 2001). It is known that about 95% of staphylococcal food poisoning outbreaks are caused by SE types SEA to SEE (Bergdoll, 1983).

Enterotoxins have been detected by various methods, but the PCR technique has the ability of detecting specific gene sequences by DNA amplification (Omoe *et al.*, 2002). PCR is much faster and can be applied to detect SEs in most kinds of food such as milk and cheese products rather than available conventional techniques (compared with animal tests) (Shijia *et al.*, 2016).

The presence of *S. aureus* in milk and dairy product are not only of economic importance, but also constitute a public health hazard. Therefore, the present work was planned to investigate the incidence of *S. aureus* in milk and some dairy products and to throw the light upon the public health significance, preventive and control measures of *S. aureus*.

MATERIALS AND METHODS

Part I: Isolation and identification of *S. aureus*:

I-1- Collection and handling of the samples:

A total of 200 samples consisting of 25 raw cow's milk, 25 raw buffalo's milk, 50 kareish cheese, 25 small scale ice-cream, 25 large scale ice-cream, 25 small scale yoghurt and 25 large scale yoghurt

samples were collected from dairy shops and street vendors in Beni-Suef Governorate, Egypt. The collected samples were delivered as soon as possible to the laboratory in an insulated ice box and examined in the same day. Each milk sample was thoroughly mixed before divided into two parts. The first part was used for Storch's test (Lampert., 1987) to exclude the heat treated milk, while the second one was used for bacteriological examination.

I-2- Preparation, Enumeration and identification of *S. aureus* according to (Quinn *et al.*, 1994):

Raw milk and yoghurt samples were thoroughly mixed before preparation of serial dilutions, while ice-cream samples were melted in thermostatically controlled water bath at a temperature of up to 40°C for not more than 15 min and mixed well. Eleven ml/grams of samples were diluted in 99 ml of sterile 0.1% peptone water for initial dilution, subsequent decimal dilutions were prepared with the same diluent. Eleven grams of kareish cheese samples were transferred into a sterile blender and thoroughly homogenized with 99 ml of sterile 2% sodium citrate solution to obtain a dilution of 1:10, and then serial dilutions were carried out.

From each dilution 0.1 ml was spread onto a dry surface of double sets of Baird-Parker agar plate supplemented with egg yolk tellurite emulsion. The streaked plates were incubated at 37°C for 24-48 h. Typical colonies of *S. aureus* (black shining convex colonies, 1-1.5 mm in diameter with narrow white margin and surrounded by a clear zone extending into opaque medium) were selected and isolated. The isolates were subjected to further microscopic and biochemical identification (Catalase test, Coagulase test, Mannitol salt agar medium) as described by (Quinn *et al.*, 1994).

Part II: Detection of enterotoxins genes of *S. aureus* by PCR:

1- Extraction of DNA according to QIAamp DNA mini kit instructions (Mehrotra *et al.*, 2000):

20 µl QIAGEN protease were pipetted into the bottom of a 1.5 ml microcentrifuge tube, then add 200 µl of the sample and 200 µl buffer AL, mixed by pulse vortexing for 15 seconds. After that the mixture was incubated at 56°C for 10 min and centrifugated to remove drops from the inside of the lid. 200 µl ethanol (96%) were added to the sample, and mixed again by pulse vortexing for 15 seconds. After mixing, make centrifugation to remove drops from the inside of the lid. The solution was carefully transferred to the QIAamp mini spin column (in a 2ml collecting tube) without wetting the rim. the cap was closed, and centrifugated at 8000 rpm for 1 min. The QIAamp mini spin column contents were transferred into a clean 2 ml collection tube, and the tube containing the filtrate was discarded, after

centrifugation. Then add 500 µl buffer AW1 was added without wetting the rim. The cap was closed, centrifugated at 8000 rpm for 1 min and 500 µl buffer AW2 were added. The cap was closed, and centrifugated at full speed for 3 min. The old collection tube was discarded with the filtrate. Centrifugation at full speed for 1 min was done. After that the QIAamp mini spin column was transferred to a clean 1.5 ml microcentrifuge tube, and the collection tube containing the filtrate was

discarded. It was carefully opened and 100 µl buffer AE were added and incubated at room temperature (15-25°C) for 1 min and then centrifugated at 8000 rpm for 1 min.

2- Cycling conditions of the primers during cPCR:

Temperature and time conditions of the primers during PCR are shown in Table:

Primary denaturation	Secondary denaturation	Annealing	Extension	No. of cycles	Final extension
94°C 5 min.	94°C 30 sec.	57°C 30 sec.	72°C 30 sec.	35	72°C 7 min.

3- Agarose gel electrophoreses with modification (Sambrook *et al.*, 1989):

The gel was photographed by a gel documentation system and the data was analyzed through computer software.

RESULTS

Table 1: Statistical analytical results of coagulase- positive *S. aureus* in the examined raw milk samples (cfu/ml).

Samples	No. of examined samples	No. of positive samples	%	Minimum	Maximum	Mean	±SEM
Raw cow's milk	25	13	52	<10	9.8×10^8	1.62×10^8	9.5×10^7
Raw buffalo's milk	25	16	64	<10	8.5×10^8	7.88×10^7	5.19×10^7

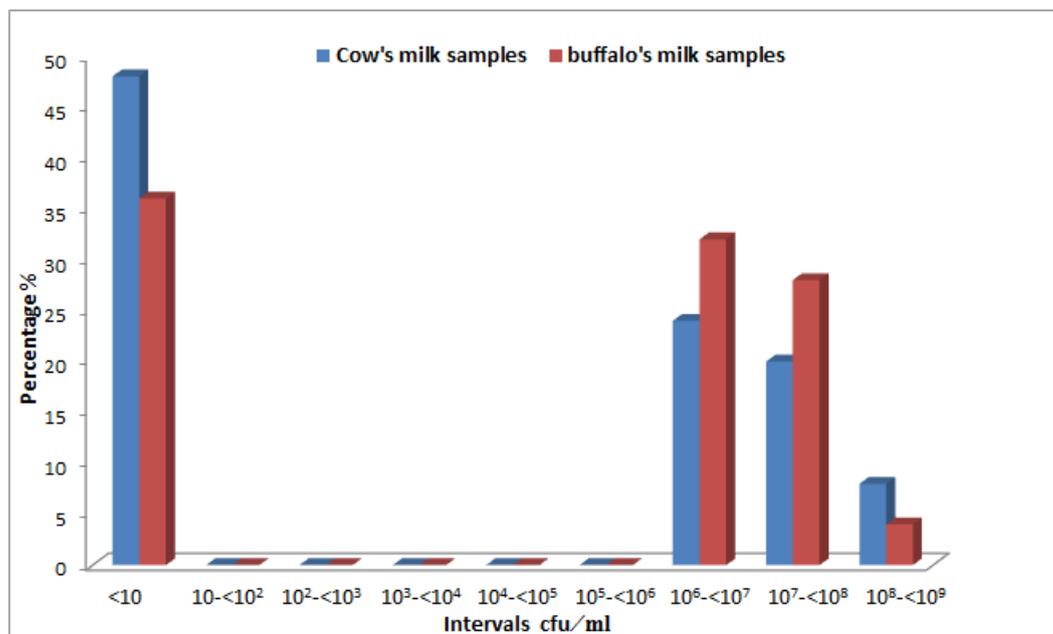


Figure (1): Frequency distribution of *S. aureus* counts/ml in the examined raw cow's and buffalo's milk samples.

Table 2: Statistical analytical results of coagulase- positive *S. aureus* counts in the examined dairy products samples (cfu/g).

Samples	No. of examined samples	No. of positive samples		Minimum	Maximum	Mean	±SEM
		No.	%				
Kareish cheese	50	34	68	<10 ²	7.3×10 ⁸	8.68×10 ⁷	2.61×10 ⁷
Small scale ice- cream	25	20	80	<10 ²	7.8×10 ⁸	6.64×10 ⁷	3.29×10 ⁷
Large scale ice- cream	25	6	24	<10 ²	1.1× 10 ⁷	6.52×10 ⁵	4.41×10 ⁵
Small scale yoghurt	25	22	88	<10 ²	3.2× 10 ⁷	3.67× 10 ⁶	1.68× 10 ⁶
Large scale yoghurt	25	9	36	<10 ²	8.5× 10 ⁶	5.27× 10 ⁵	3.45× 10 ⁵

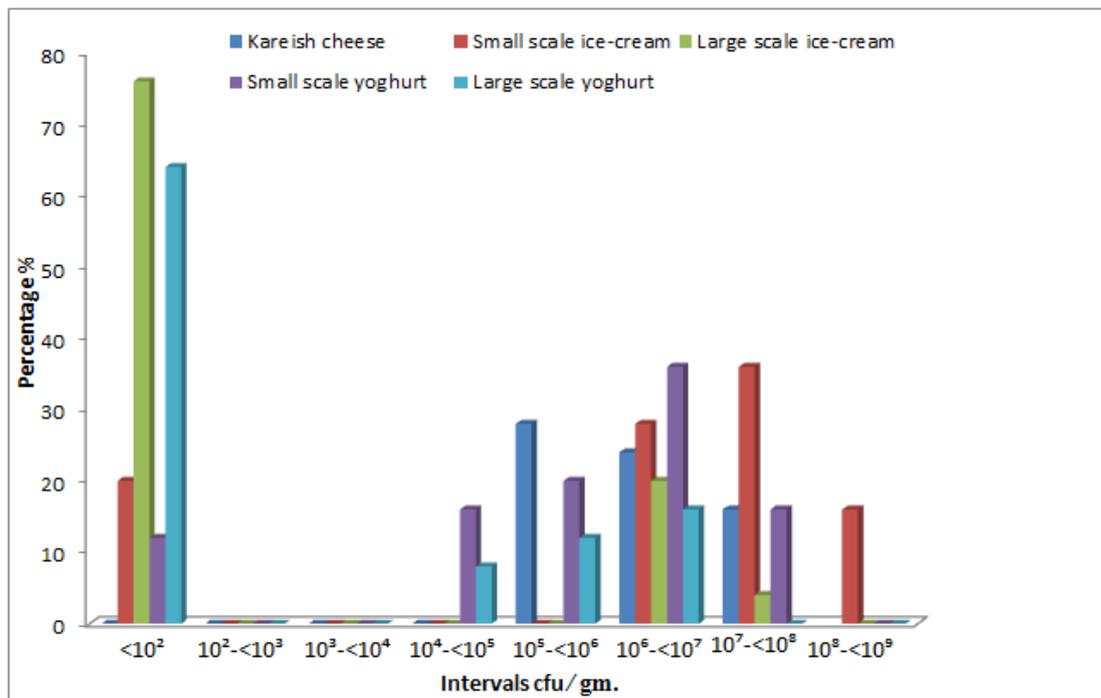


Figure (2): Frequency distribution of coagulase- positive *S. aureus* counts in the examined dairy products samples.

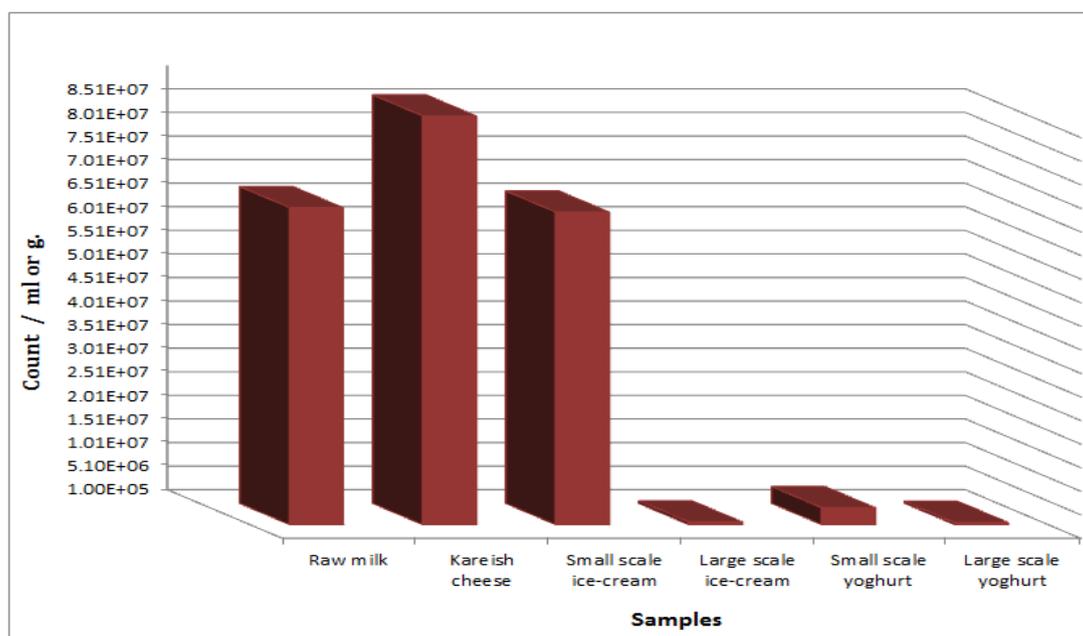
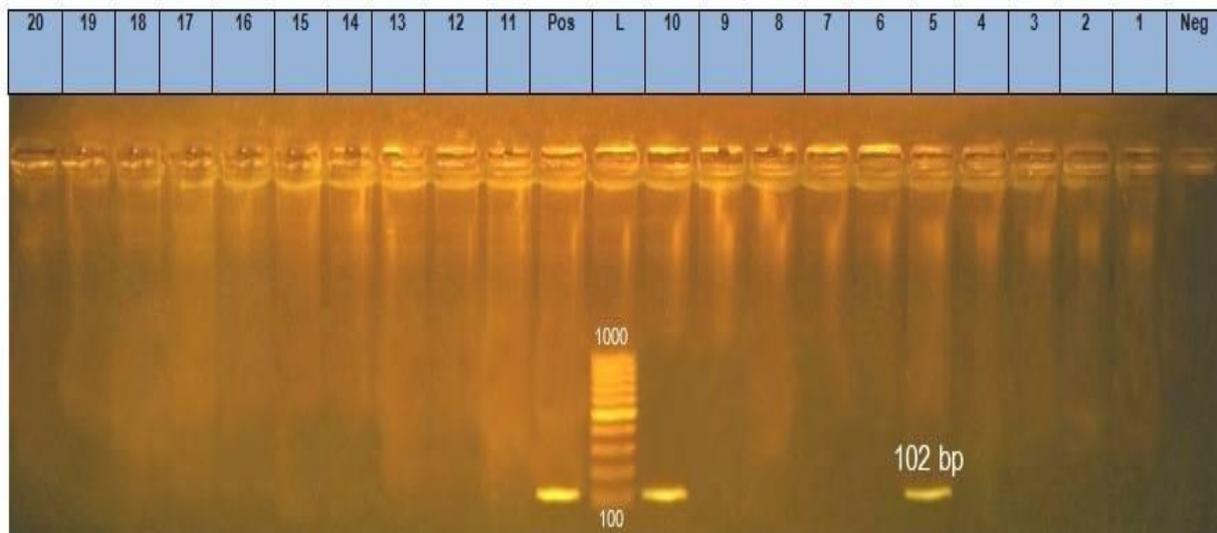


Figure (3): Coagulase- positive *S. aureus* counts (/ml or g.) in the examined samples.

Table 3: PCR results for detection entrotoxicogenic *S. aureus* of Sea and Seb toxins.

No.of samples	origin	Results	
		<i>Sea</i>	<i>Seb</i>
1	Yoghurt	-	-
2	Yoghurt	-	+
3	Yoghurt	-	-
4	Yoghurt	-	-
5	Yoghurt	+	+
6	Milk	-	+
7	Milk	-	-
8	Milk	-	-
9	Milk	-	-
10	Milk	+	+
11	Ice-cream	-	-
12	Ice-cream	-	-
13	Ice-cream	-	+
14	Ice-cream	-	-
15	Ice-cream	-	-
16	Kareish cheese	-	-
17	Kareish cheese	-	-
18	Kareish cheese	-	-
19	Kareish cheese	-	+
20	Kareish cheese	-	-

**Figure (4):** PCR results for *S. aureus* Sea gene

L: 100-1000 bp DNA ladder.

Lane 5, 10: positive samples for sea gene.

Lane 1-4,6-9,11-20: negative samples.

Pos.: Positive control.

Neg.: Negative control.

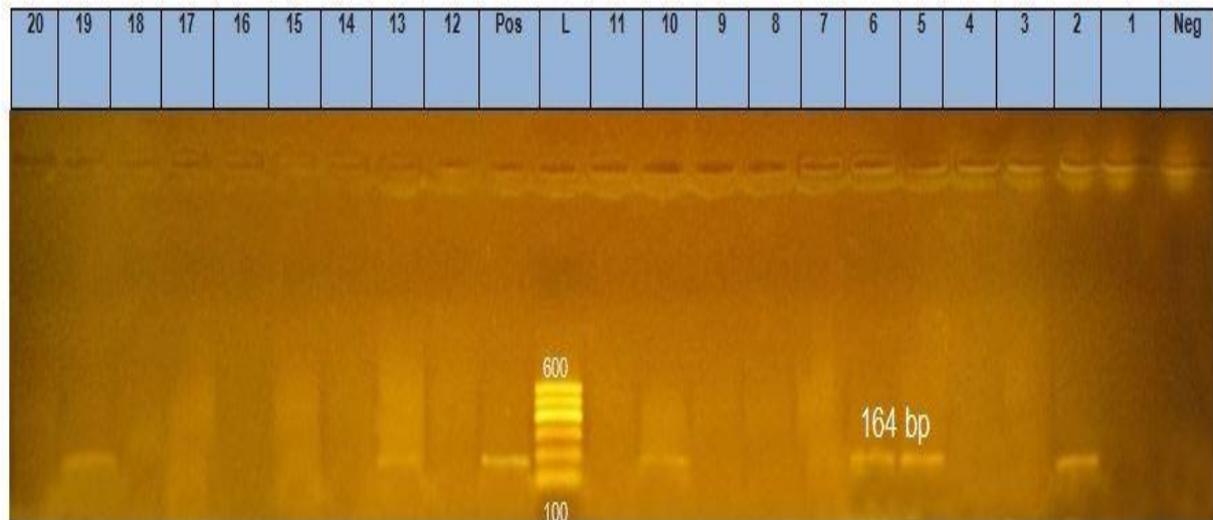


Figure (5): PCR results for *S. aureus* Seb gene.

L: 100-600 bp DNA ladder

Lane 2,5,6,10, 13 and 19: positive samples For seb gene.

Lane 1, 3, 4, 7-9, 11,12, 14-18,20: negative samples

Pos. : Positive control

Neg. : Negative control

DISCUSSION

In the present study, Table (1) and Figures (1&3) showed that 13 (52%) and 16 (64%) of raw cow's and buffalo's milk samples were contaminated by *S. aureus* respectively. Also revealed that the count of *S. aureus* in raw cow's milk ranged from <10 to 9.8×10^8 with a mean value of $1.62 \times 10^8 \pm 9.5 \times 10^7$ CFU/ml and the highest frequency distribution (48%) was in a range of <10 . The count of *S. aureus* in raw buffalo's milk ranged from <10 to 8.5×10^8 with a mean value of $7.88 \times 10^7 \pm 5.19 \times 10^7$ CFU/ml and the highest frequency distribution (36%) was in a range of <10 .

To some extent nearly similar results for the incidence of *S. aureus* were postulated by Rall *et al.* (2008), De Oliveira *et al.* (2011), Charaya *et al.* (2014) and Pourtaghi *et al.* (2015). While higher results were reported by Gitau *et al.* (2010), Ayele *et al.* (2017) at farm level, and Kandil *et al.* (2018). On the other side, lower results were demonstrated by Abdou *et al.* (2016) and Matallah *et al.* (2019).

S. aureus is frequently found in raw milk and the infections of mammary gland (mastitis) represent a significant reservoir of toxigenic strains in raw milk. Storage of raw milk under high environmental temperature permitting growth of *S. aureus* can stimulate the production of their enterotoxin. This results highlights the unhygienic handling and inadequate personal hygiene as postulated by Meshref (2013).

The obtained results in Table (2) and Figures (2&3) revealed that the count of *S. aureus* in kareish cheese ranged from $<10^2$ to 7.3×10^8 with a mean value of $8.68 \times 10^7 \pm 2.61 \times 10^7$ CFU/g and an incidence of 68%. The highest frequency distribution (32%) was in a range of $<10^2$.

To some extent nearly similar results for the count were indicated by Ewida (2009) and Salama *et al.* (2015), with respect to the *S. aureus* incidence higher results were recorded by El-Kholy *et al.* (1995) and Kandil *et al.* (2018), but lower results were reported by Mousa (2017) and Abo El-Makarem and Amer (2018). While lower results for the count were recorded by Moawad *et al.* (2002), Al-Tahiri (2005), Bahout and Moustafa (2006), El-Bessary (2006), Aly *et al.* (2007), Sadik (2009) and Armanios (2013).

S. aureus is frequently found in milk, however their presence in kareish cheese is usually due to primary contamination from raw milk, unhygienic handling of the cheese and/or on extensive contamination by personnel possibly involved in cheese making and marketing; as humans are the common carriers.

The high incidence of *S. aureus* in the examined kareish cheese was due to that the kareish cheese is made by farmers from raw milk that isn't subjected to heat treatment. Street vendors put kareish cheese in pans exposed to dust and flies (Zakary *et al.*, 2011). So, kareish cheese are involved in food poisoning outbreak from public health point of view,

S. aureus had been implicated in many cases of food poisoning and gastroenteritis among consumers (Eley, 1996).

The results in Table (2) and figures (2&3) showed that the count of *S. aureus* in the examined small scale ice-cream ranged from $<10^2$ to 7.8×10^8 with a mean value of $6.64 \times 10^7 \pm 3.29 \times 10^7$ CFU/g and an incidence of 80%. The highest frequency distribution (36%) was in a range of 10^7 - $<10^8$. To some extent nearly similar result for the count was pointed by Nawar (2001), but for the incidence was recorded by Ahmed *et al.* (2019). While higher result for incidence was reported by Kandil *et al.* (2018) and lower result for the count was detected by Ewida (2009).

It's noticed from the results in Table (2) and figures (2&3) that the count of *S. aureus* in the examined large scale ice-cream ranged from $<10^2$ to 1.1×10^7 with a mean value of $6.52 \times 10^5 \pm 4.41 \times 10^5$ CFU/g and an incidence of 24%. The highest frequency distribution (76%) was in a range of $<10^2$. Nearly similar result for the count of *S. aureus* was reported by Hassan (2015), while nearly result for the incidence of *S. aureus* was recorded by Abd El-Fattah (2013). In addition to, lower result for the incidence was reported by El-Ansary (2015). From another side higher results for the incidence were detected by Zakary *et al.* (2011) and Kandil *et al.* (2018).

Ice-cream has been incriminated as a vehicle of staphylococcal enterotoxin in several food poisoning outbreaks as ice-cream mix was usually contaminated at the time of preparation and during the long period of cooling as well as before the mix freezing (APHA, 1992). The high contamination rates of examined ice-cream samples are mainly due to use poor quality of milk and materials used in ice-cream manufacture, environmental contamination as unclean hands of worker either suffering from diseased or apparent healthy carriers, unsanitary production and marketing practices (Araujo *et al.*, 2002).

The results in Table (2) and figures (2&3) revealed that the count of *S. aureus* in the examined small scale yoghurt ranged from $<10^2$ to 3.2×10^7 with a mean value of $3.67 \times 10^6 \pm 1.68 \times 10^6$ CFU/g and an incidence of 88%. The highest frequency distribution (36%) was in a range of 10^6 - $<10^7$. Nearly similar result for the incidence was recorded by Abou El-Makarem (2013), while lower results for count and incidence were indicated by Hafez (2010) and Kandil *et al.* (2018).

In addition, the occurrence of *S. aureus* in the examined large scale yoghurt samples were present in 9 (36%) and ranged from $<10^2$ to 8.50×10^6 with a mean value of $5.27 \times 10^5 \pm 3.45 \times 10^5$ CFU/g. The

majority of the samples 16 (64%) contained $<10^2$ CFU/g. To somewhat extent similar results were reported by Haggag (2006), Sadik (2009), El-Kholy *et al.* (2014) and Shahin (2015), while comparatively higher result for the incidence was noted by Ahmed *et al.* (2014), but lower result was obtained by Osman (2015). In contrast, no *S. aureus* were detected in any of the examined samples by El-Bessery (2001), El-Kholy *et al.* (2016).

The variation between these results and the others was attributed to the difference in localities where samples were collected; also, there are different sources for contamination by *S. aureus* as from respiratory infections, skin or mouth of the workers handling the dairy products. So it's good indicator of the personal hygiene of the workers (Kamat *et al.*, 1991). Finally, we should show that the high incidence of *S. aureus* is indicative of poor hygienic measures during production, handling and distribution (Joshi *et al.*, 2004). The difference in prevalence rates of *S. aureus* between the examined products may originate from the method of manufacture, storage and handling. The lowest prevalence rate (24 and 36%) of *S. aureus* which were recorded in large scale ice-cream and large scale yoghurt, respectively. This might be attributed to the effect of heating then freezing during their manufacturing which inhibit the multiplication of the microorganism and kill the microorganism (Zakary *et al.*, 2011).

It's worth in mention that the presence of *S. aureus* in milk and dairy products even in low numbers must be regarded a public health hazard; because it has been established that the *S. aureus* may lose its viability in food, but enterotoxins still exist. The viability of *S. aureus* during the manufacturing of dairy products as cheese, yoghurt and ice-cream, depend on the addition of salt concentration, starter culture and storage time (Erkmen, 1995). The higher incidence in our results are due to the poor hygienic conditions during milking and cheese making and inappropriate conditions of their storage negatively influenced the cheese quality.

The results in Table (3) and Figures (4&5) regarding that twenty *S. aureus* strains (5 out of milk samples and 5 of each product of kareish cheese, ice-cream and yoghurt samples) were examined for enterotoxin genes detection and 30% of the examined *S. aureus* strains were enterotoxigenic by PCR technique as carried one or two SE-genes. Sea gene is detected in yoghurt and milk for Sea and Seb genes by PCR samples with a percentage 20% of each while Sea was not detected in any ice-cream and kareish cheese samples. While Seb gene was found in 6 (30%) of the strains isolated dairy products in this study. It was interesting that most of the *S. aureus* isolated from yoghurt and milk in our study harbored the Seb (40%) gene, and the strains

isolated from ice-cream and kareish cheese samples contained the Seb gene at level of 20%. These results are similar to (Lee *et al.*, 2001 and Peles *et al.*, 2007).

While comparatively higher results for Sea gene than our obtained result in this study was recorded by (Mathenge *et al.*, 2015), but lower results for Sea gene were reported by (Ghaleb *et al.*, 2005 and Naffa *et al.*, 2006). On the other hands, the higher result for Seb gene was recorded by (Ghaleb *et al.*, 2005), but lower results for Seb gene were reported by (Mathenge *et al.*, 2015 and Zouharova and Rysanek, 2008).

Although pasteurization destroy *S. aureus* pathogen, but thermostable SEs can resist heat treatment and still are potent even in very small amount ranging from 20 ng to <1 µg can produce symptoms to human beings (Bergdoll, 2012). Enterotoxigenic strains of *S. aureus* have been reported to cause a number of diseases or food poisoning outbreaks due to ingestion of contaminated dairy products (Oliver *et al.*, 2005). SEs have ability to withstand the environmental conditions such as drying, heat and freezing (Le Loir *et al.*, 2003). In this study, we found enterotoxigenic *S. aureus* in yoghurt samples which has low acidity due to that the enterotoxins of *S. aureus* can survive at lowered pH and proteolytic enzymes such as pepsin or trypsin that destroy them completely functional in the gastrointestinal tract after consumed. They also are one of the superantigens family, which demolishes the immune system of the host by targeting the innate and adaptive system responses (Argudin *et al.*, 2010).

Neither the absence of *S. aureus* nor the presence of small numbers of organism can provide complete assurance that the milk and dairy products are safe, since conditions inimical to the survival of *S. aureus* may result in a diminished population or death of viable microbial cells, while sufficient toxins remain to elicit symptoms of staphylococcal food poisoning (Bennett and Monday, 2003). Although foods must contain at least 10⁶ enterotoxigenic *S. aureus* CFU/g to induce illness, small numbers of *S. aureus* present in thermally processed foods may represent the survivors of very large populations (Robinson and Tamime, 2002).

CONCLUSION

Results of this study clearly indicate that milk and milk products in the examined areas have high prevalence of *S. aureus* and found some secreted enterotoxins in this samples which lead to presence food poisoning symptoms, where we found that the small scale yoghurt and small scale ice-cream samples have higher contamination percentage with *S. aureus* than kareish cheese and raw milk samples than large scale yoghurt and large scale ice-cream

samples have lower contamination level, and this due to poor hygienic handling, inadequate personnel hygiene and insufficient pasteurization. So, It's required improving hygiene practices during milking routine and careful handling of animal during milking should be followed to limit the spread of *S. aureus* to humans and less than 100 cell/ml milk of bacterial counts can be achieved if some better hygienic practices implemented. Introduction of cooling system for the milk during production, transportation and during distribution process particularly during the summer season to not allow *S. aureus* to form SEs. Milk must be produced, distributed, handled and marketed under the control of milk commission and the commission must have a sanitary inspector and veterinarian to enforce its methods and standards.

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دراسات على ميكروب المكور العنقودي الذهبي المفرز للسموم المعوية في اللبن وبعض منتجاته

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الجزء الاول من هذه الدراسة تم تجميع ٢٠٠ عينة (٢٥ عينة لبن بقرى خام و ٢٥ عينة من اللبن الجاموسي الخام و ٥٠ عينة من الجبن القريش و ٢٥ عينة من الايس كريم من صغار المنتجين و ٢٥ عينة من الايس كريم المنتج من المصانع الكبيرة و ٢٥ عينة من الزبادي المنتج من صغار المنتجين و ٢٥ عينة من الزبادي المنتج من المصانع الكبيرة) من مدينة بني سويف وقد تم فحص هذه العينات عن مدى تواجد ميكروب المكور العنقودي الذهبي في اللبن وبعض منتجاته وأسفرت الدراسة عن النتائج الآتية: ١٣ (٥٢%) و ١٦ (٦٤%) بمتوسط $1.62 \times 10^4 \pm 9.5 \times 10^7$ و $7.88 \times 10^7 \pm 0.19 \times 10^7$ و 34 (٦٨%) بمتوسط $8.68 \times 10^7 \pm 2.71 \times 10^7$ و 20 (٨٠%) بمتوسط $6.64 \times 10^7 \pm 3.29 \times 10^7$ و 6 (٢٤%) بمتوسط $6.02 \times 10^6 \pm 4.41 \times 10^6$ و 22 (٨٨%) بمتوسط $3.67 \times 10^6 \pm 1.68 \times 10^6$ و 9 (٣٦%) بمتوسط $5.27 \times 10^6 \pm 3.45 \times 10^6$ لكل مللي او جرام على الترتيب. وبالنسبة للجزء الثاني، أشارت النتائج بأن ٣٠% من سلالات العنقودية الذهبية التي تم فحصها عن طريق تفاعل البلمرة المتسلسل حيث حملت واحد او اثنين من الجينات المفرزة للسموم المعوية. وقد تمت مناقشة الأهمية الصحية بالإضافة الى الأهمية الاقتصادية للميكروب العنقودي الذهبي لتحسين جودة اللبن وبعض المنتجات اللبنية للحفاظ على صحة المستهلك.