

## DETECTION OF STAPHYLOCOCCUS AUREUS IN EGYPTIAN SOFT CHEESES

**Maha A. M. Al-Ashmawy**

Food Hygiene and Control Department,  
Faculty of Veterinary Medicine, Mansoura University

### SUMMARY

Seventy five random samples of Egyptian soft cheeses (Kareish cheese, Damitta fresh cheese and Damitta ripened cheese "25 samples of each") were collected from retail outlets and supermarkets in Mansoura city, Egypt. *Staphylococcus aureus* was detected in 84%, 64% and 68% of examined Kareish cheese; Damitta fresh cheese and Damitta ripened cheese samples. The mean counts of *Staphylococcus aureus* in Kareish cheese; Damitta fresh cheese and Damitta ripened cheese were  $3.3 \times 10^3$ ;  $3.0 \times 10^1$  and  $4.1 \times 10^2$  respectively. 68% of Kareish cheese samples; 52% samples of Damitta fresh cheese and 44% samples of Damitta ripened cheese had *Staphylococcus aureus* counts marginally exceed the desire maximum count of *S. aureus* reported by EC Food Legislation and UK Microbiological Guidelines for good hygiene practices in manufacture of soft and fresh cheeses. Preventive measures for decreasing contamination of soft cheeses were discussed.

### INTRODUCTION

Milk and dairy products have been implicated in Staphylococcal food-borne outbreaks. In general, the bacterium grows in dairy products and produces toxins without adversely affecting its acceptance quality. Cheeses specially soft cheeses have been more frequently implicated (Ray, 1996).

*Staphylococcus aureus* may be contaminating dairy products from the udder of dairy animal, from human beings and from inadequately cleaned equipment (Takeuchi et al., 2001). Enterotoxin-producing *Staphylococcus aureus* strains have generally been associated with Staphylococcal food intoxication. Although strains of several *Staphylococcus* species are known to be enterotoxin producers, their involvement in food poisoning is not fully known (Ray, 1996).

The enumeration of Staphylococci was routinely conducted on dairy products specially cheeses as a good indicator of quality of sanitation during its production and distribution and to es-

tablish the occurrence of post-processing contamination (ICMSF, 1996). Therefore, the present investigation was planned to throw light on the incidence and count of coagulase positive *Staphylococcus aureus* and study the physiological character of the isolates.

## MATERIAL AND METHODS

Seventy five random samples of soft cheeses (Kareish cheese; Damitta fresh cheese and Damitta ripened cheese "25 samples of each"), collected from retail outlets and supermarkets in Mansoura city, Egypt, were examined for detection of *Staphylococcus aureus* organisms.

### Collection and preparation of samples:

Random samples of each type of cheeses were collected from different groceries, supermarkets, dairy shops and small dairies in sterile air tight sampling jars and transferred to the laboratory without delay to be prepared according to APHA. (1992).

### Enumeration and isolation of *Staphylococcus aureus*:

The numbers of *S. aureus* were determined according to Roberts et al., (1995) by using surface plate technique onto *Staphylococcus* Medium No.110 and incubated at 35°C/48hs. (Oxoid, 1998). The suspected colonies (deep orange colonies) were picked up onto slope agar, purified and identified according to Cowan and Steel, (1974).

*Staphylococcus aureus* isolates which were previously identified were confirmed by using Latex slide agglutination test for detection of *S. aureus* "Dry Spot Staphytest" (Oxoid, 2004).

## RESULTS AND DISCUSSION

### \*Incidence and count of coagulase positive *Staphylococcus aureus* in soft cheese samples:

The statistical analytical results reported in Table (1) indicated that *Staphylococcus aureus* was detected in 21 (84%); 16 (64%) and 17 (68%) samples of examined Kareish cheese; Damitta fresh cheese and Damitta ripened cheese respectively. The count of *Staphylococcus aureus* in Kareish cheese samples/g ranged from  $10 - 3.3 \times 10^4$  with a mean value of  $3.3 \times 10^3 \pm 1.7 \times 10^3$ ; in Damitta fresh cheese  $10 - 6.7 \times 10^4$  with a mean value of  $3.0 \times 10^4 \pm 0.30 \times 10^4$ ; while in Damitta ripened cheese the count/g varied from  $10 - 3.4 \times 10^3$  and the mean value  $4.1 \times 10^2 \pm$

$2.0 \times 10^2$  (Table 1). These findings substantiate the results reported by **Nour et al., (1991)**. Also, the presence of *Staphylococcus aureus* in soft cheeses was reported by several investigators (**Ashenafi, 1990; Pereira et al., 1991; Dardir, 1999 and Aranlo et al., 2002**).

The frequency distribution of examined soft cheese samples based on coagulase positive *Staphylococcus aureus* count (Table 2) indicated that 17 (68%) samples of karcish cheese have *S. aureus* count ranged from  $10^2 - 10^5$ /g, while 13 (52%) samples of Damitta fresh cheese have *S. aureus* count/g ranged from  $10^2 - 10^5$ /g and 11 (44%) samples of Damitta ripened cheese have *S. aureus* count varied from  $10^2 - 10^4$ /g.

The enumeration of *Staphylococcus aureus* was routinely conducted for dairy products as a good indicator of quality of sanitation during its production, handling and distribution and to establish post-processing contamination. The results obtained in this work marginally exceed the desire maximum counts of *Staphylococcus aureus* reported by European Economic Communities Food Legislation and United Kingdom Microbiological Guidelines for good hygiene practices in manufacture of soft and fresh cheeses (**Roberts et al., 1995**). Moreover, the Egyptian Standards (2000) concerning that soft cheeses must be free from pathogenic organisms and their toxins.

Growth of *Staphylococci* can occur either before or during cheese making. Cheeses have been implicated in several food poisoning outbreaks in recent years (**Bowen and Henning, 1994**). High populations of *Staphylococci* in cheese are ground for suspicious, but should not be taken as presumptive evidence for presence of enterotoxins.

Physiological characteristics of isolated *Staphylococcus aureus*:

Regarding the summarized results of coagulase positive *Staphylococcus aureus* reported in Table (3) showed the haemolysis, mannitol fermentation, gelatin liquefaction and DNase of the isolated *Staphylococcus aureus* strains from soft cheeses.

These findings indicated that no single physiological characteristics or combination of characteristics possessed by *Staphylococcus aureus* are found to be an absolutely reliable indicator of enterotoxigenicity (**Minor and Marth, 1971**).

**Koenig and Marth, (1982)** described that virtually cheeses were positive for enterotoxin, with the highest toxin levels being recorded in high-salt cheeses ripened at  $10^\circ\text{C}$ .

As *Staphylococcus aureus* is ubiquitous within the farm environment and carried by human population, soft cheeses contain low levels of enterotoxigenic *Staphylococcus* however, growth and enterotoxin production are easily prevented by proper refrigeration.

Therefore, increased recognition of *Staphylococcal* mastitis in dairy animals coupled with im-

provements in milk handling, processing and cooling practices have made dairy-related outbreaks of Staphylococcal food poisoning an uncommon occurrence in the countries.

*Table (1): Statistical analytical results of coagulase positive Staphylococcus aureus count/g soft cheeses*

| Soft cheeses           | Positive samples |    | Min. | Max.              | Mean              | SEM                   |
|------------------------|------------------|----|------|-------------------|-------------------|-----------------------|
|                        | No.              | %  |      |                   |                   |                       |
| Kareish cheese         | 21               | 84 | 10.0 | $3.3 \times 10^4$ | $3.3 \times 10^3$ | $\pm 1.3 \times 10^3$ |
| Damitta fresh cheese   | 16               | 64 | 10.0 | $6.7 \times 10^4$ | $3.0 \times 10^4$ | $\pm 0.3 \times 10^4$ |
| Damitta ripened cheese | 17               | 68 | 10.0 | $3.4 \times 10^3$ | $4.1 \times 10^2$ | $\pm 2.0 \times 10^2$ |

*Table (2) Frequency distribution of examined soft cheeses samples based on their coagulase positive Staphylococcus aureus*

| Intervals     | Kareish cheese |      | Damitta fresh cheese |      | Damitta ripened cheese |      |
|---------------|----------------|------|----------------------|------|------------------------|------|
|               | No.            | %    | No.                  | %    | No.                    | %    |
| $0 - < 10^1$  | 4              | 16.0 | 9                    | 36.0 | 8                      | 32.0 |
| $10^1 < 10^2$ | 4              | 16.0 | 3                    | 12.0 | 6                      | 24.0 |
| $10^2 < 10^3$ | 7              | 28.0 | 4                    | 16.0 | 9                      | 36.0 |
| $10^3 < 10^4$ | 8              | 32.0 | 4                    | 16.0 | 2                      | 8.0  |
| $10^4 < 10^5$ | 2              | 8.0  | 5                    | 20.0 | -                      | 0    |

Table (3): Physiological characteristics of confirmed coagulase positive\* *Staphylococcus aureus* strains isolated from soft cheeses

| Type of cheese         | No. of coagulase +Ve isolates | Haemolysis |      |     |      | Mannitol fermentation |      |     |      | Gelatin Liquefaction (stone reaction) |      |     |      | DNase |      |     |      |
|------------------------|-------------------------------|------------|------|-----|------|-----------------------|------|-----|------|---------------------------------------|------|-----|------|-------|------|-----|------|
|                        |                               | +Ve        |      | -Ve |      | +Ve                   |      | -Ve |      | +Ve                                   |      | -Ve |      | +Ve   |      | -Ve |      |
|                        |                               | No.        | %    | No. | %    | No.                   | %    | No. | %    | No.                                   | %    | No. | %    | No.   | %    | No. | %    |
| Karcish cheese         | 54                            | 41         | 75.9 | 13  | 24.1 | 46                    | 85.2 | 8   | 14.8 | 11                                    | 20.4 | 43  | 79.6 | 42    | 77.7 | 12  | 22.2 |
| Damitta fresh cheese   | 36                            | 30         | 83.3 | 6   | 16.7 | 23                    | 63.9 | 13  | 36.0 | 15                                    | 41.7 | 21  | 58.3 | 25    | 69.4 | 11  | 30.6 |
| Damitta ripened cheese | 34                            | 29         | 85.3 | 5   | 14.7 | 28                    | 82.4 | 6   | 17.6 | 8                                     | 23.5 | 26  | 76.5 | 20    | 58.8 | 14  | 41.2 |

\*By Latet slide agglutination test (Dry-spot staphy tests)

### REFERENCES

- American Public Health Association "APHA" (1992)** : Compendium of methods for the microbiological examination of foods. 2nd Ed., APHA, Washington D.C., USA.
- Aranio, V. S.; Pagliares, V. A.; Quelroz, M. L. and Freitas-Almeida, A. C. (2002)** : Occurrence of Staphylococcus and enteropathogens in soft cheese commercialized in the city of Rio de Janeiro, Brazil. *J. Appl. Microbiol.*, 92 (6): 1172-7.
- Ashenafi, M. (1990)** : Microbiological quality of Ayib, a traditional Ethiopian Cottage cheese. *Int. J. of Food Microbiol.*, 10 (3-4): 263-268.
- Bowen, D. A. and Henning, D. R. (1994)** : Coliform bacteria and Staphylococcus aureus in retail natural cheeses. *J. Food Prot.* 57: 253.
- Cowan, S. T. and Steel, K. J. (1974)** : Manual for the identification of medical bacteria, 2nd Cambridge Univ., Press, England.
- Dardir, H. A. (1999)** : Novel trends for rapid detection of some food-borne pathogens in milk and some dairy products. Ph.D. Thesis (Milk Hygiene). Fac. of Vet. Med., Cairo Univ.
- Egyptian Standards "E. S" (2000)** : 1008, 2000 International Committee on Microbiological Specifications for Foods "ICMSF" (1996): Microorganisms in food, their significance and methods of enumeration. 2nd Ed., Univ. of Toronto Press, Toronto, Buffalo and London.
- Koenig, S. and Marth, T. H. (1982)** : Behavior of *S. aureus* in Cheddar cheese made with sodium chloride or a mixture of sodium chloride and potassium chloride. *J. Food Prot.* 45: 996.
- Minor, T. E. and Marth, E. H. (1971)** : Staph. aureus and Staphylococcal food intoxication. A review. I. The Staphylococci: Characteristics, isolation and behaviour in artificial media. *J. Milk Food Technol.* In press.
- Nour, M.A.; Naguib, M.M. and Tohamy, M.M. (1991)**: Contamination level and enterotoxigenicity of Staphylococcus aureus isolated from market Domiatii cheese. *Egyptian J. of Dairy Sci.*, 19: 1, 93-98.
- Oxoid, (1998)**: The Oxoid Manual. 8th Ed. Unipath. Ltd., United Kingdom.
- Oxoid, (2004)**: Oxoid Product List. Oxoid Ltd., England.
- Pereira, M.L.; Lara, M.A.; Dias, R.S. and Carma, L.S. (1991)**: Staphylococcal food poisoning by cheese "Tipo Minas". *Revista-de-Microbiol.*, 22 (4): 349-350.
- Ray, B. (1996)**: Fundamental Food Microbiology, CRC Press, Inc.

**Roberts, D.; Hooper, W. and Greenwood, M. (1995):** Practical food microbiology, Public Health Laboratory Service, London, 2nd Edition.

**Takeuchi, S.; Maeda, T.; Hashimoto, N.; Imazumi, K.; Kaidoh, T. and Hayaka, Y. (2001):** Variation of the agr Locus in *Staphylococcus aureus* isolates from cows with mastitis. *Vet. Microbiol. Apr. 2; 79 (3): 267-74.*

## الملخص العربي

## الكشف عن المكور العنقودي الذهبي في الجبن الطري المصري

مها عبده محمد عبده العشماوى

قسم الرقابة الصحية على الأغذية - كلية الطب البيطرى - جامعة المنصورة

تم فحص ٧٥ عينة من الجبن الطري المصري (٢٥ عينة من كل من الجبن القريش ، الجبن الدمياطي الطازج ، الجبن الدمياطي المعتق) جمعت من مراكز البيع و الأسواق من مدينة المنصورة بمصر. تم الكشف عن الميكروب العنقودي الذهبي في ٨٤٪ ، ٦٤٪ ، ٦٨٪ من عينات الجبن القريش والجبن الدمياطي الطازج والجبن الدمياطي المعتق علي التوالي. المتوسط العددي للمكور العنقودي الذهبي في الجبن القريش ٣٣ × ٣١٠ والجبن الدمياطي الطازج ٣ × ٤١٠ والجبن الدمياطي المعتق ٤١ × ٢١٠.

وبمقارنة النتائج بالحد الأدنى لعدد المكور العنقودي الذهبي للمواصفات القياسية للدول الأوروبية والخطوط الميكروبيولوجية المرشدة لتصنيع الجبن الطري والطازج بالمملكة المتحدة تبين أن ٦٨٪ من الجبن القريش ، ٥٢٪ من الجبن الدمياطي الطازج ، ٤٤٪ من الجبن الدمياطي المعتق يزيد عن الحد الأدنى المرغوب فيه كحد أقصى.

وقد تم مناقشة الإجراءات الوقائية لتقليل من مدى تلوث الجبن الطري الأبيض أثناء الإنتاج والتداول والتخزين.