



Beni-Suef University

Faculty of Veterinary Medicine

Food Hygiene and Control Department

Studies on enterotoxigenic Staphylococcus aureus in milk and some dairy products.

Thesis presented by

Walaa Ahmed Ashour Ahmed

(B.V.Sc.Fac.Vet. Med., Beni-Suef University, 2011,

M.V.Sc. Fac.Vet. Med., Beni-Suef University, 2016).

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Under the supervision of

Prof. Dr. Arafa M. Soliman

Vice Dean for Education and Student Affairs
Professor and Head of Food Hygiene Department
Faculty of Veterinary Medicine
Beni-suef University

Prof. Dr. Gamal M. Hassan

Professor of Milk Hygiene
Faculty of Veterinary Medicine
Beni-suef University

Prof. Dr. Emad M. Riad

Chief researcher of Bacteriology Department
Animal Health Research Institute Dokki, Giza

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Abstract

A total of 200 samples consisting of (25 of each raw cow's and buffalo's milk samples, 50 kareish cheese samples, 25 of each small scale and large scale ice-cream samples, 25 of each small scale and large scale yoghurt samples) were collected from different localities as dairy shops and street vendors in Beni-Suef Governorate, Egypt for the determination of prevalence rate of *S. aureus* by using conventional method and molecular technique. The obtained results revealed that 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, $8.68 \times 10^7 \pm 2.61 \times 10^7$, $6.64 \times 10^7 \pm 3.29 \times 10^7$, $6.52 \times 10^5 \pm 4.41 \times 10^5$, $3.67 \times 10^6 \pm 1.68 \times 10^6$, $5.27 \times 10^5 \pm 3.45 \times 10^5$ 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.

The present work evaluated the influence of nisin on viability of enterotoxigenic *S. aureus* in manufactured kareish cheese using two different concentrations of nisin (10&12.5 ppm) and the obtained results showed the gradual reduction of *S. aureus* count during storage period in refrigerator for 30 days.

Keywords: *S. aureus*, Milk, Kareish cheese, Ice-cream, Yoghurt, Enterotoxins, PCR, nisin.

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Summary

In the present study, we focused on three important points to improve the quality of milk and some dairy products.

The first part was the estimation of the incidence of *S. aureus* in milk and some dairy products samples (kareish cheese, small scale ice-cream, large scale ice-cream, small scale yoghurt and large scale yoghurt samples).

A total of two hundred random samples of milk and dairy products including (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 different localities as dairy shoppes, markets and street vendors in Beni-Suef Governorate, Egypt. All the samples were examined bacteriologically for detection of *S. aureus*. The isolated strains were identified using conventional method and then the identified *S. aureus* strains were evaluated by PCR. Last but not least, nisin was applied to examine its inhibitory effect on the growth of the enterotoxigenic *S. aureus* in manufactured kareish cheese.

The obtained results revealed that the prevalence rate of *S. aureus* was 13 (52%), 16 (64%), 34 (68%), 20 (80%), 6 (24%), 22 (88%) and 9 (36%) in raw cow's milk, raw buffalo's milk, kareish cheese, small scale ice-cream, large scale ice-cream, small scale yoghurt and large scale yoghurt samples were contaminated with *S. aureus*, respectively using conventional method, 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, $8.68 \times 10^7 \pm 2.61 \times 10^7$, $6.64 \times 10^7 \pm 3.29 \times 10^7$, $6.52 \times 10^5 \pm 4.41 \times 10^5$, $3.67 \times 10^6 \pm 1.68 \times 10^6$, $5.27 \times 10^5 \pm 3.45 \times 10^5$ CFU/g. respectively.

The public health in addition to economic importance of isolates was discussed to improve the hygienic quality of milk and milk products.

The second part was detection of enterotoxins genes of *S. aureus* by PCR assay.

The results in 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 isolates for Sea and Seb genes by PCR with a percentage 20% of each while Sea was not detected in any ice-cream and kareish cheese isolates. While Seb gene was found in 6 (30%) of the strains isolated from 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, while the strains isolated from ice-cream and kareish cheese samples contained the Seb gene at level of 20% .

The obtained results proved that PCR technique is more sensitive and specific in detection of these genes than other conventional techniques and help to understanding the role of these genes in pathogenicity pathway in different disease conditions and construction of effective control measure.

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.

The third part was the evaluation the effect of nisin on the survival of *S. aureus* in manufactured kareish cheese.

1- Effect of 10 ppm nisin on *S. aureus* count / g. in kareish cheese samples during the refrigerator storage:-

The obtained results noted that the count of *S. aureus* in manufactured kareish cheese decreased gradually from 4×10^8 to 1.1×10^3 CFU/gm., while in control ones decreased to 4×10^4 CFU/gm. at the 30th day through the storage period in refrigerator at 4°C

The statistical analysis of results approved that there were significant differences between count of *S. aureus* in control and cheese with nisin which indicated that the addition of 10 ppm of nisin concentration to cheese inhibits the growth of *S. aureus*.

2- Effect of 12.5 ppm nisin on *S. aureus* count / g. in kareish cheese samples during the refrigerator storage:-

The results revealed that the count of *S. aureus* in manufactured kareish decreased gradually from 4×10^8 to 1×10^2 CFU/gm. during storage in refrigerator for 30 days. the results indicated that nisin of 12.5 ppm concentration had higher inhibitory and preservative effect than nisin of 10 ppm concentration against *S. aureus* growth in cheese.

The results suggested that cheese with nisin (10 and 12.5 ppm concentrations) can be used for preservation of cheese; due to its inhibitory effect against food borne pathogens.



جامعة بني سويف
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قسم الرقابة الصحية على الأغذية

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دراسة مقدمة من

طب / ولاء أحمد عاشور أحمد

(بكالوريوس العلوم الطبية البيطرية - جامعة بني سويف - 2011 ماجيستر العلوم

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كلية الطب البيطري- جامعة بني سويف

تحت إشراف

أ.د/ عرفة مشرف سليمان

وكيل الكلية لشئون التعليم والطلاب

أستاذ ورئيس قسم الرقابة الصحية على الأغذية

كلية الطب البيطري - جامعة بني سويف

أ.د/ جمال محمد حسن

أستاذ الرقابة الصحية على الألبان ومنتجاتها

كلية الطب البيطري - جامعة بني سويف

أ.د/ عماد مختار رياض

رئيس بحوث قسم البكتيريولوجي

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