



Investigation of *S. Aureus* in Butter and Ghee in Some Markets in Nyala Locality, South Dafour State, Sudan

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ABSTRACT:

This study was aiming at investigating *S. aureus* in butter and ghee in Nyala city in South Darfour State during the year 2021. A total of 80 milk products' samples (40 butter and 40 ghee) were collected from 4 markets (Nyala, El Ban gadeed, Manawashy and El Laloba) in Nyala Locality. Samples were examined bacteriologically using conventional and rapid API methods. All butter samples (100%) collected from El Laloba and Manawashy markets were positive for *S. aureus*. Eighty percent of the samples collected from Nyala market were positive for *S. aureus*. Sixty percent of the samples collected from El Ban gadeed market were positive for *S. aureus*. All ghee samples (100%) collected from El Laloba and El Bangadeed markets were negative for *S. aureus*. Sixty percent of the samples collected from Nyala and Manawashy markets were positive for *S. aureus*. The prevalence of *S. aureus* in butter samples in Nyala Locality was 85%. The prevalence of *S. aureus* in butter samples in Nyala Locality was 30%. Butter in Nyala Locality is highly contaminated and ghee is less contaminated with *S. aureus*. Pasteurization and heat treatment are highly recommended before processing of milk.

KEY WORDS: Aerobic bacteria, Butter, Ghee, Nyala Locality, Sudan.

I. INTRODUCTION

Butter is a traditional natural food used world-wide and is essential for human nutrition Due to its high fat content. It is an important source of energy and contains many other nutritionally important components, such as minerals and fat soluble vitamins. Butter oil (ghee, samin) is a rich source of energy, fat soluble vitamins, essential fatty acids and other growth-promoting Factors. Butter oil is the anhydrous form of milk fat defined as the product exclusively obtained from milk, cream or butter from various animal species [1]. The genus Staphylococci includes over 30 species; 18 of these species and subspecies are of potential hazard in food poisoning as they produce coagulate, heat stable nuclease or enterotoxins. The coagulase positive species are *S. aureus* subsp. *aureus* which is the most common enterotoxigenic species [2] [3]. *S. aureus* is a common pathogen associated with serious community and hospital acquired diseases and has for long been considered as a major problem of public health [4]. The pathogenicity of *S. aureus* and its ability to cause diseases is attributed to a number of virulence factors such as the heat stable enterotoxins [5]. Staphylococcal food poisoning is one of the most common foodborne diseases in both humans and animals globally, resulting from the ingestion of staphylococcal enterotoxins preformed in food by enterotoxigenic strains of coagulase-positive staphylococci, mainly *S. aureus*. Foodborne diseases are of major concern worldwide. There have been around 250 different foodborne diseases described, and bacteria are the causative agent of two third of foodborne diseases outbreaks [6]. Contamination of food products with *S. aureus* pathogens may result primarily from their presence in the basic raw material, milk. In such cases, the source of pathogen is the dairy cow or sheep [7]. Milk is normally sterile in the udder of the cow and buffalo, if they do not suffer from mastitis (udder infection). If they have mastitis, a large number Streptococcus spp. and Staphylococcus spp. may be present in milk. *S. aureus* may be in milk when it leaves the udder [8]. [9] reported Staphylococci represented the predominant bacteria (27.8%) isolated from mastitic milk samples in Shandi and Bahri Localities. [10] identified *S. aureus* (18.3%), *S. epidermidis* (8.4%), *S. chromogenes* (4.8%) from Cases of Bovine Subclinical Mastitis in White Nile State.

The objective of this study is to investigate *S. aureus* in butter and ghee in some markets in Nyala Locality, South Darfour State, Sudan.

II. MATERIALS AND METHODS

Area of the study

A total of 80 milk products' samples (40 butter and 40 ghee) were collected from 4 markets (Nyala, El Ban gadeed, Manawashy and El Laloba) in Nyala Locality, South Darfour State (table 1).

Sampling procedure

Samples were collected and put in sterile containers. The collected samples were put in ice box containing ice, transported to the laboratory of faculty of Veterinary Medicine University of Bahri and examined.

Isolation, identification and characterization of bacterial isolates

Isolation, identification and characterization of bacterial isolates was done according to [11].

Biological and biochemical identification

The purified isolates were identified as previously described [12] and [11].

Identification of Staphylococcal Isolates Using API staph (BIOMERIEUX, France) Identification System

API staph (Analytical Profile Index for identification of the Genus Staphylococcus) is a standardized system for the identification of the Genera: Staphylococcus, Micrococcus and Kocuria, which uses miniaturized biochemical tests and specially adapted database. It was used according to [13] to identify gram positive bacteria.

Statistical analysis

Statistical analysis was done through Microsoft office Excel 2007.

Table (1): Butter and ghee samples collected from markets of Nyala Locality

Market	No. of butter samples	No. of ghee samples
Nyala	10	10
El Ban gadeed	10	10
Manawashy	10	10
El Laloba	10	10
Total	40	40

3. RESULTS

S. aureus in butter samples:

All butter samples (100%) collected from El Laloba and Manawashy markets were positive for *S. aureus*. Eighty percent of the samples collected from Nyala market were positive for *S. aureus*. Sixty percent of the samples collected from El Ban gadeed market were positive for *S. aureus* (Table 2 and Figure 1).

S. aureus in ghee samples:

All ghee samples (100%) collected from El Laloba and El Bangadeed markets were negative for *S. aureus*. Sixty percent of the samples collected from Nyala and Manawashy markets were positive for *S. aureus* (Table 3 and Figure 2).

Table (2): *S. aureus* in butter samples

Market	No. of butter samples	No./% of samples positive for Staphylococci
Nyala	10	8 (80%)
El Ban gadeed	10	6 (60%)
Manawashy	10	10 (100%)
El Laloba	10	10 (100%)
Total	40	34(85%)

Table (3): *S. aureus* in ghee samples

Market	No. of ghee samples	No./% of samples positive for Staphylococci
Nyala	10	6 (60%)
El Ban gadeed	10	0 (0%)
Manawashy	10	6 (60%)
El Laloba	10	0 (0%)
Total	40	12(30%)

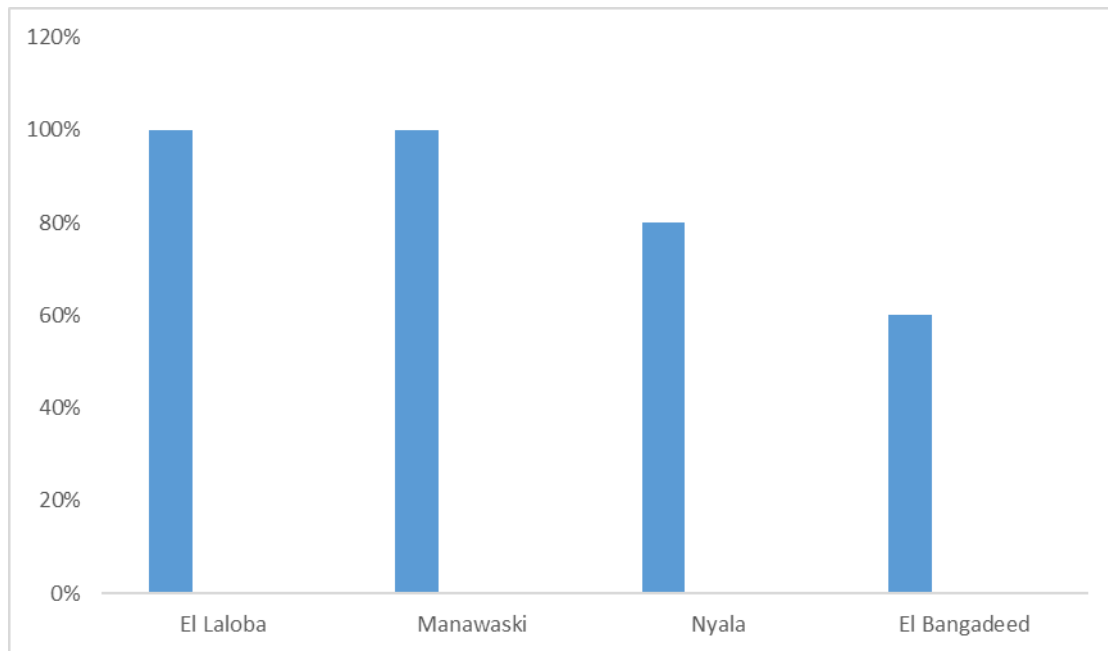


Fig. (1): *S. aureus* in butter samples

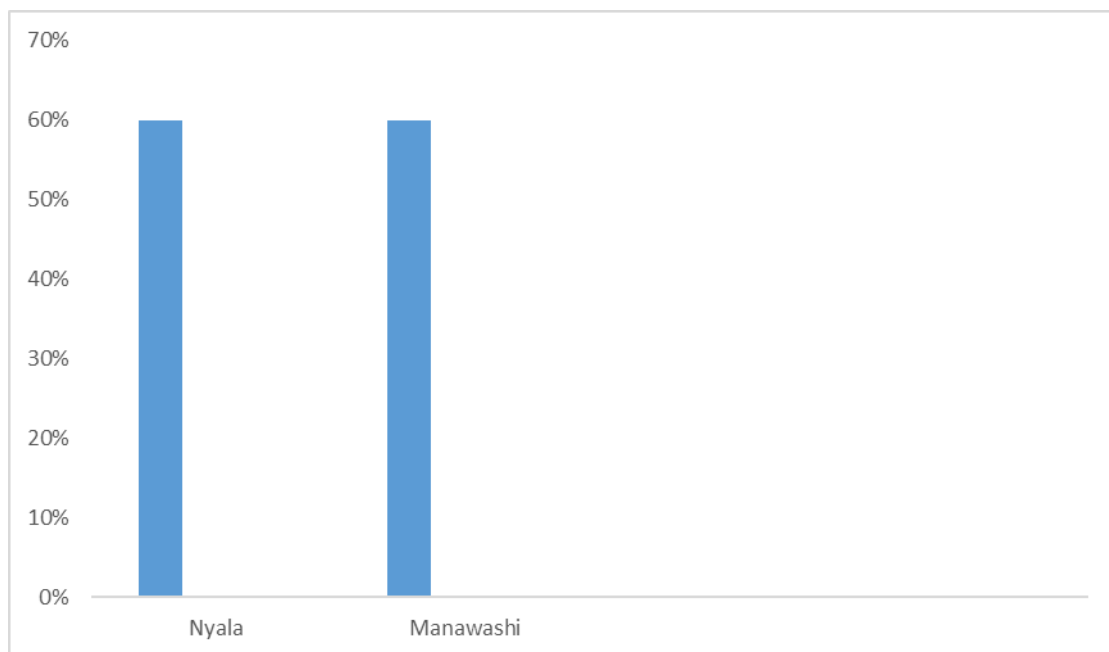


Fig. (2): *S. aureus* in ghee samples

Prevalence of S. aureus in butter samples in Nyala Locality

Out of 40 butter samples collected from markets of Nyala Locality, 34 samples were positive for *S. aureus* (Table, 2). Prevalence of *S. aureus* in butter samples in Nyala Locality was 85%.

Prevalence of S. aureus in ghee samples in Nyala Locality

Out of 40 ghee samples collected from markets of Nyala Locality, 12 samples were positive for *S. aureus* (Table, 3). Prevalence of *S. aureus* in butter samples in Nyala Locality was 30%.

Identification and characterization of *S. aureus*

Primary and secondary tests

Table (4) and Figures (3 and 4) illustrate the results of primary and secondary tests for identification of *S. aureus*.

Table (4): Cultural characteristics, bacterial morphology and biochemical tests of the isolated *S. aureus*.

Test	Result	Test	Result
Aerobic growth	+	Oxidase	-
Colonies on MacConkey	Pink	Indole	-
Haemolysis on blood agar	+	Methyl red	+
Gram reaction	+	VP	-
Shape	Cocci	Citrate	-
Spore	-	H ₂ S	-
Motility	-	O/F	+
Catalase	+	Glucose	+

Api Staph Identification system

Table (5) and Figure (5) illustrate the results of Api Staph Identification system for identification of *S. aureus*.

Table (5): Api Staph Identification system.

Test	Result	Test	Result
(0)	-	Potassium Nitrate (NIT)	+
D-glucose (GLU)	+	B-Naphthyl phosphate (PAL)	+
D-fructose (FRU)	+	Sodium pyruvate (VP)	+
D-mannose (MNE)	+	D-raffinose (RAF)	-
D-maltose (MAL)	+	D-xylose (XYL)	-
D-lactose (LAC)	+	D-saccharose (SAC)	-
D-trehalose (TRE)	-	Methyl- α D-Glucopyranoside (MDG)	+
D-mannitol (MAN)	+	N-acetyl-glucoseamine (NAG)	+
Xylitol (XLT)	-	L-arginine (ADH)	-
D-melibiose (MEL)	-	Urease (URE)	+

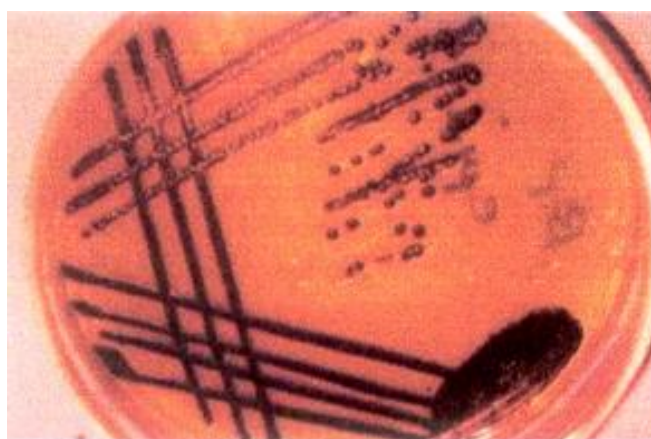


Fig. (3): Black colonies of *S. aureus* on Baird-Parker medium.



Fig. (4): β -haemolysis of *S. aureus* on Blood Agar.



Fig. (5): API staph strip positive for *S. aureus*.

4. DISCUSSION

Milk is normally sterile in the udder of the cow and buffalo, if they do not suffer from mastitis (udder infection). If they have mastitis, a large number Streptococcus spp. and Staphylococcus spp. may be present in milk. *S. aureus* may be in milk when it leaves the udder [14]. In this study all butter samples collected from El Laloba and Manawashy markets, 80% of the samples collected from Nyala market and 60% of the samples collected from El Ban gadeed market were positive for *S. aureus*. Sixty percent of ghee samples collected from Nyala and Manawashy markets were positive for *S. aureus*. Also [15] isolated a total of 133 staphylococci from 100 bovine mastitic samples in River Nile State, Sudan. [16] reported that Staphylococci isolated from ewes' mastitic milk samples collected from in El Nuhud city in West Kordofan State in Sudan, represented 60.0% of the total bacterial isolates. He also reported that Staphylococci isolated from goats' mastitic milk samples represented 80.0% of the total bacterial isolates. [17] isolated *S. aureus* (25.0%) from white cheese in El Obied city in North Kordofan State Sudan. [18] reported that Staphylococci represented 44.5% of the microorganisms isolated from mastitic milk samples in Khartoum area. Low percentages of isolation of Staphylococci from milk and milk products were reported by some authors. In Sudan [19] [20] isolated Staphylococci (21.1%) from cases of Caprine Mastitis in River Nile State. In the same State [21] isolated Staphylococci (21.0%) from ewes' milk samples. In this study the prevalence of *S. aureus* in butter samples in Nyala Locality was 85% and 30% in butter samples. In Iran [22] reported that the prevalence of *S. aureus* was found in traditional cheese (11.1%), followed by traditional ice-cream (5.9%), cream (5.6%), and butter (5.3%). In India [23] isolated *S. aureus* (6.25 %) from milk and milk products. [24] reported the prevalence of 29.5% of *S. aureus* in milk products in Ethiopia. [25] reported that Coagulase positive Saphylococci (CPS) represented 50.0% of the total Staphlococci isolates from ewes' mastitic milk samples collected from in El Nuhud city in West Kordofan State in Sudan. He also reported that Coagulase Positive saphylococci (CPS) represented 81.5% of the total Staphylococci isolated from goats' mastitic milk samples.

5. CONCLUSION AND RECOMMENDATIONS

From this study we conclude the high contamination of butter and low contamination of ghee in Nyala Locality with *S. aureus*. The prevalence of *S. aureus* in buttein Nyala Locality was 85% and 30% for ghee.

We recommend further studies in different Localities of the State and an extensive study of the significance of *S. aureus* in milk products. Pasteurization and heat treatment are highly recommended before processing of milk.

6. ACKNOWLEDGEMENT

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