A Safety Evaluation Study Of Locally Manufactured Dairy Products

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Staphylococcus aureus, Bacillus species, lactobacillus, and Enterobacter species

Summary
Introduction: Locally produced dairy products safety issue, is of a pressing public health concern, as they can pose health risks to consumers due to potential contamination and inadequate safety controls.
Objective:This study sought to assess the safety levels of locally produced dairy products. Samples of locally produced cow milk dairy product were collected randomly from Fulani hawkers busy Hausa settlements in Edo/Delta state (Benin, Abraka, Obiaruko).
Methods: Microbial and biochemical processes were carried out following appropriate microbiological and biochemical techniques. The bacterial species isolated include: *Staphylococcus aureus, Bacillus species, lactobacillus, and Enterobacter species.*
***Results**:* The organism with the highest percentage occurrence was *Enterobacter species* (45.4%) followed by *Staphylococcus aureus* (27.3%), then *Bacillus species* (18.2%) and *lactobacillus* (9.1%). Sample A which was obtained from Abraka had a Total bacterial count which was Too numerous to count (TNTC), and Sample B which was obtained form Benin city had a Total bacterial count of 14.5 and a colony forming unit of 1.45×10^8, and Sample C which was obtained from Obiaruko had a Total bacterial count of 14.0 and a colony forming unit of 1.40×10^8.
Conclusion: According to the results, the bacteria present in the sample exceeds the recommended limit. The study highlights the need for improved sanitary practices and quality control measures to ensure consumer safety and reduce the risk of food borne illnesses.

INTRODUCTION

Dairy products are various products obtained from the milk of cows or otherfemale mammals such as goats, sheep, yaks, horses, and camels. They includemilk and any of the food made from milk, including butter, cheese, ice cream,yoghurt, and condensed and dried milk [1]

The importance of dairy products in human nutrition and economic developmentis well-documented, making them a crucial component of dietary regimens across the globe. Dairy products contribute significantly to the nutritional needs by providing essential proteins, vitamins, and minerals necessary for human health [2]. In many regions, especially in developing countries, locally produced dairy products not only satisfy these nutritional needs but also support livelihoods and local economies [3].

Despite their benefits, the safety of locally produced dairy products often comes into question due to varying standards of production practices. Issues such as inadequate sanitary conditions, lack of proper storage facilities, and minimal regulatory oversight can lead to contamination risks, affecting the safety and quality of the products [4]. Common contaminants in dairy include pathogens like Salmonella, Listeria, and E. coli, which are associated with serious health risks [5].

Moreover, the use of antibiotics in dairy cattle, a common practice to prevent diseases, poses additional risks as it can lead to antibiotic resistance. The World Health Organization has tagged antibiotic resistance as one of the top ten global public health threats facing humanity, which underscores the need for careful management and regulation of antibiotic use in livestock [6].

Furthermore, economic pressures and the informal nature of local dairy sectors can exacerbate these safety challenges. Small-scale producers may lack the resources to implement necessary safety measures, leading to inconsistencies in product quality and safety standards [7]. This not only puts public health at risk but also impacts the marketability of these products both locally and in broader markets.

Evaluating the safety of locally produced dairy products is, therefore, crucial to ensure public health, enhance consumer confidence, and improve market access for these products. Such evaluations can help identify critical control points in the production and distribution processes and foster the development of targeted interventions to mitigate health risks associated with dairy consumption [8]. Moreover, strengthening the local dairy sector's

compliance with national and international safety standards can play a pivotal role in boosting economic opportunities for local communities [9].

Through this study, we aim to provide a comprehensive evaluation of the safety practices associated with locally produced dairy products, identify prevalent risks, and propose actionable strategies to enhance safety protocols. This research not only contributes to the academic discourse on food safety but also serves as a guide for policy makers, stakeholders, and producers in the dairy industry [10].

MATERIALS AND METHODS

Study Area

The study areas are Ethiope East Local Government Area of Delta state (Sample) A), Ukwuani Local Government Area of Delta State (Sample C) and Ikpoba Okha Local Government Area of Edo State (Sample B). The population for this study was selected from random Fulani dairy product hawkers in the study areas

Sample Collection

Samples of locally produced dairy products were randomly collected from various local farms and markets within the study area. Each sample was aseptically collected in sterile containers and transported to the laboratory in ice-packed coolers within two hours of collection.

Sterilization

The working surfaces were disinfected before and after working by cleaning with cotton wool soaked with 70% ethanol. The wire loops were sterilized by flaming to red hot before use. The L-shaped glass rod was sterilized by soaking in ethanol and then flaming, all glass wares and media were sterilized in the autoclave at 121℃ for 15 minutes at 15psi

Preparation of culture media

Nutrient agar, MacConkey Agar, Mannitol Agar, Cetrimide were prepared following manufacturer's specification.

Sample Preparation

Ten-fold serial dilution were done for each samples using normal saline as blank. 1ml of sample was dispense into 9 ml of blank in a sterile test tube using a sterile Pasteur pipette and mixed properly. A volume of one milliliter of the mixture was then transferred into the second test tube already containing 9 ml of the blank and mixed also to obtain a 1:10 dilution. This was repeated further to obtain dilution of 1:100 and 1:10,000 repeatedly until a 1:1,000,000 dilution was obtained.

Inoculation and Incubation

The media were prepared following the manufacturer’s guidelines and sterilized by autoclaving at 121℃ for 15 minutes. The media were allowed to cool to 45℃ and then transferred into sterile Petri dishes under aseptic conditions and then allowed to solidify. A volume of 0.1ml of the serially diluted sample was asepticallytransferred from the sixth dilution into the solidified agar plates using a sterile Pasteur pipette, a L-shaped glass rod was sterilized and used to spread the sample across the plates. The plates were incubated in an inverted position in an incubator for 24 hours at 37ºC.

Colony Count

Colony count was performed on the various culture media used. Discrete colonies appearing on the plate after appropriate incubation were counted and recorded. The total bacterial count was obtained by counting discrete colonies on the Nutrient Agar media. The numbers of colonies counted were multiplied by the reciprocal of the dilution factor plated and divided by the volume of the inoculums used, to obtain the colony forming unit per millimeter (cfu/ml) of each sample.

CFU/ml = Number of distinct colonies
numbers of aliquot × dilution factor
Key: CFU = colony forming unit

Identification of Isolates

Isolates from culture after incubation were identified using cultural and biochemical test.

Prior to the chemical test, Gram staining was carried out to differentiate the isolates into Gram-positive and Gram-negative organisms. Cultural features of the bacterial such as morphology and color were also reported. Gram staining and other Biochemical Identifications such as Catalase Test, Indole Test, Citrate Test, Coagulase Test, Motility Test, Fermentation and Gas Test, Methyl Red Test, Oxidase Test and Urease Test were carried out following standard procedures as described [12].

RESULTS AND DISCUSSION

Three samples of locally produced cow milk dairy product collected randomly from Fulani hawkers in the study areas were assessed. The result of the Total Bacteria Count (TBC) and the Colony Forming Unit (CFU) of the different samples are shown in Table 1. The result of cultural characteristics, microscopic and biochemical test are shown in Table 2 and the result for the percentage occurrence of the bacteria isolates obtained from the samples are shown in Table 3 below.

Table 1 Total bacterial count of the three samples.

S/N	SAMPLE	NUMBER OF COLONIES	CFU= No. of colonies/vol. of aliquot × dilution factor (CFU/ml)
1.	A	TNTC	TNTC
2.	B	14.5	1.45×10^8
3.	C	14.0	1.4×10^8

KEY: TNTC= Too numerous to count.

From the study, the microbial load analysis of the dairy products revealed significant variations in the total bacterial count (TBC) among different producers. The TBC values indicated a range of contamination levels, with some samples exhibiting higher bacterial loads than others. For instance, milk samples from sample A had a notably higher TBC which was Too numerous to count(TNTC) compared to those from sample B (1.45×10^8) and sample C (1.4×10^8). This discrepancy suggests differences in hygiene practices and production processes among producers.

Table 2: Identification of the bacterial isolates (biochemical tests)

S/N	Sample isolates	CAT	CIT	MTY	UREA	IND	COA	H2S	OX	G	L	S	MR	Gram stain	Cell morph-ology	INFERENCE
1	A N/A White	+	-	-	+	-	-	-	-	A	A	A	-	+ve	Rod-like	*Lactobacillus spp*
2	B MAC cream	+	-	+	+	-	-	-	-	AG	AG	AG	-	-ve	Rod-like	*Enterobacter spp*
3	B MSA	+	-	-	-	-	+	-	-	AG	A	A	-	+ve	cocci	*Staphylococcus aureus*
4	B MAC pink	+	-	+	-	-	-	-	-	AG	AG	AG	-	-ve	Rod-like	E*nterobacter spp*
5	C MSA Yellow	+	-	-	-	-	+	-	-	A	A	A	-	+ve	cocci	*Staphylococcus aureus*
6	A MSA	+	-	-	+	-	+	-	-	A	A	A	-	+ve	cocci	*Staphylococcus aureus*
7	A MAC Cream	+	-	+	+	-	-	-	-	AG	AG	AG	-	-ve	Rod-like	E*nterobacter spp*

8	A MAC Pink	+	-	+	-	-	-	-	-	A G	A G	A G	-	-ve	Rod-like	E*nterobacter spp*
9	B N/A White	+	+	+	-	-	-	-	-	A	A	A	-	+ve	Rod-like	*Bacillus spp*
10	C N/A White	+	+	+	+	-	-	-	-	A G	A G	A G	-	+ve	Rod-like	*Bacillus spp*
11	C MAC Pink	+	-	+	+	-	-	-	-	A G	A G	A G	-	-ve	Rod-like	*Enterobacter spp*

KEY: CAT= catalase test, CIT= citrate test, MTY= motility test, UREA= urease test, H2S= hydrogen sulphide test, IND= indole test, COA= coagulase test, OX= oxidase test, G= glucose, L= lactose, S= sucrose, MR= methyl red test, += positive reaction - = negative reaction, spp= species, A= acid production, G= gas production, CET= cetrimide agar, N/A= nutrient agar, MSA= mannitol salt agar, MAC= MacConkey agar, +ve= Gram positive organism, -ve= Gran negative organism.

The identification and characterization of bacteria through Gram staining and biochemical tests provided detailed insights into the microbial diversity present inthe dairy products. Gram staining results showed a predominance of Gram-positive bacteria. These included common lactic acid bacteria such as the *Lactobacillus species,* which are beneficial for fermentation but also highlighted the presence of pathogens like *Staphylococcus aureus.*

Table 3 Percentage frequency of bacterial isolates based on biochemical tests.

S/N	ISOLATES	NUMBER OF ISOLATES	PERCENTAGE (%)
1	*Lactobacillus spp*	1	9.1
2	*Enterobacter spp*	5	45.4
3	S*taphylococcus aureus*	3	27.3
4	*Bacillus spp*	2	18.2
	TOTAL	**11**	**100**

Biochemical tests confirmed the presence of various bacterial isolates such as *Staphylococcus aureus, Lactobacillus species, Enterobacter species, and Bacillus species* and their percentage frequency of occurrence include 27.3%, 9.1%, 45.4%,and 18.2% respectively.

CONCLUSION

In conclusion, the microbiological analysis of locally produced dairy products inthe study area reveals significant variations in microbial load and the presence of various bacterial species, including potential pathogens. The total bacterial count found in several samples exceeded acceptable safety standards, indicating sub optimal hygiene practices and the need for stringent quality control measures.

The identification of pathogenic bacteria such as *Enterobacter species, Staphylococcus aureus, and Bacillus species* underscores the potential health risks associated with these products and highlights the importance of regular monitoring and adherence to sanitary protocols.[13]

Overall, this study emphasizes the critical need for improved sanitary practices and stringent quality control measures in the production and handling of dairy products to ensure their safety for consumers. By addressing the identified microbial contamination issues and implementing robust hygiene standards, producers can significantly reduce the risk of food borne illnesses and enhance the overall quality and safety of locally produced dairy products

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