Microbial Quality and Safety of Traditional Fermented Camel Milk Product Suusac Sampled from Different Regions in North Eastern, Kenya

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Authors’ contributions

This work was carried out in collaboration among all authors. Author IMM designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors DWMK, JW and SM managed the analyses of the study. Author IMM managed the literature searches. All authors read and approved the final manuscript.

ABSTRACT

Aim: The study was carried out to assess the microbial quality and safety of fermented camel milk product (Suusac) from North Eastern Kenya.

Methodology: Twenty-eight samples (n=28) of Suusac from different areas of the region sold in informal markets at Eastleigh in Nairobi were aseptically collected at the sales points. The quality and safety of the Suusac with reference to selected pathogens namely E. coli, S. aureus, Shigella, and Klebsiella spp was evaluated using the standard analytical methods.

Results: Escherichia coli were detected in all the samples while Staphylococcus aureus was detected in 63.09% of the samples analyzed. Shigella spp was detected in 88.1% of the samples analyzed and Klebsiella spp was detected in 77.4% of the samples. The mean log_{10} counts for E. coli, S. aureus, Shigella, and Klebsiella spp were 3.135, 2.576, 2.784 and 3.138, CFU/mL, respectively. There is a potential public health concern posed by Suusac which is sold for direct consumption due to the presence of the life-threatening bacterial pathogens.

Conclusion: The Suusac being sold at Eastleigh market in Nairobi from North Eastern Kenya may...
be responsible for transmission of the pathogens to the consumers. Training on food hygiene, improving production technology, hygienic conditions and implementing the food legislations along the value chain can minimize the risk.

Keywords: Milk; fermentation; bacterial pathogens.

1. INTRODUCTION

Suusac is fermented camel milk that is consumed by the communities that have inhabited the arid and semi-arid areas of Kenya. Preparation of Suusac is through spontaneous fermentation of camel milk which is carried out in gourds treated with smoke [1]. The camels are milked directly into a gourd that has been cleaned, smoothed and treated with smoke. It was found out that the smoke improved color, taste and improves the shelf-life by up to 20 days. The milk used is usually raw without any kind of heating. Suusac fermentation is carried out for a period of one to two days and this takes place at room temperature of between 26-29°C. There is rising public health concern associated with microbial food safety with reports implicating unpasteurized and raw camel milk products as major contributing factors to illnesses caused by foodborne pathogens [1,2]. Traditional milk products in Kenya are fermented spontaneously in gourds while modern techniques of milk fermentation involve the use of starter cultures to produce consistent and safe products with improved shelf-life [3]. In some communities, fermentation is carried out by the use of raw milk and this may lead to safety concerns while in other products like mursik, the milk is boiled before fermentation [4].

Concerns regarding the safety of products of spontaneous fermentation due to the presence of pathogenic bacteria and chemical toxins produced by the bacteria have been raised. These concerns have been demonstrated by reports on the possible causes of diseases due to mursik consumption in Kenya [5,6]. A study carried out by Kaindi et al. [7] showed that, 25% of the milk at Isiolo market and 75% of the milk at the final market in Nairobi, was not acceptable. Suusac production is also associated with unknown factors such as poor udder health, poor milking personnel practices like tying the quarters to prevent suckling by the calf, dusty milking environment, and lack of water may act as points of contamination. Various microorganisms have been reported in traditional fermented milk products in Kenya [8,9,10] and at the same time there exists no information on the microbial quality and safety of suusac. Therefore, the objective of this study was to determine the quality and safety of traditionally fermented camel milk product (Suusac) from North Eastern Kenya.

2. MATERIALS AND METHODS

2.1 Study Site

The study was carried out in various regions in North Eastern part of the country which is camel milk producing zone. The area has an average temperature ranging between 12 and 28°C and receives low rainfall ranging between 300 and 500 mm per year. However, milk samples from those areas were collected in Eastleigh, Nairobi County which is a major urban consumption center for camel milk. North Eastern Kenya has approximately about 40,300 camels, mostly owned by Borana and Somali communities and produces about 50,000L of milk daily. It is estimated that 87.5% of the produced camel milk is for home consumption or sold to locals in nearby trading centers while 12.5% is supplied to Eastleigh, the main market in Nairobi [10].

2.2 Milk Sampling

A total of 28 milk samples were collected from selected places in the seven Counties of North Eastern, Kenya: Isiolo (14), Tana River (5), Marsabit (1), Namanga (3), Garissa (3), Moyale (1), Madera (1) were collected for this study. Approximately 50mls of each sample was obtained from bulking containers in selected trader shops in Eastleigh. At the shops, 30 traders were selected. Each sample was coded numerically to show the Sub County from where it was sourced from in each County.

2.3 Determination of pH of Milk Samples

pH determination was done using an electronic digital pH meter (Orion Research Inc., Cambridge, MA, USA) which was calibrated using a Buffer solution of pH 4 and 7 following the ISO 26323:2009(en) method. Samples of the camel milk were taken and analyzed for pH. Readings were taken by immersing the pH meter electrodes into the samples and steady values displayed.
2.4 Microbial Analyses

2.4.1 Staphylococcus aureus

The ISO 6888-1:1999 method was used for enumeration of \textit{Staphylococcus aureus}. Appropriate dilutions of homogenate samples were pipetted on the surface of previously dried Baird-Parker agar plates and spread with a sterile bent glass rod in duplicate. The plates were incubated at 37°C for 24 hours. The enumeration was done using colony counter for colony forming units and expressed per mL of the sample (CFU mL\(^{-1}\)). The colonies were identified based on colour which was black and shiny, with narrow white margins, surrounded by clear zones extending into the opaque medium. The colonies were confirmed by conducting catalase, lipase test and glucose fermentation.

2.4.2 Escherichia coli

ISO 16649-2:2001 method was used for enumeration of \textit{Escherichia coli}. Appropriate dilutions of homogenate samples were pipetted on the surface of dried Hi-Crome agar plates in duplicate and spread with a sterile bent glass rod. The plates were incubated at 30°C for 4 hours and then at 44°C for 18 hours. Enumeration was then done using colony counter for colony forming units on colonies which had bluish-green coloration and expressed per mL of the sample (CFU mL\(^{-1}\)). The colonies were confirmed by conducting Methyl Red Voges Proskauer test.

2.4.3 Shigella spp

The ISO 21567:2004 method was used for enumeration of \textit{Shigella spp}. Appropriate dilutions of homogenate samples were pipetted on the surface of dried plates of XLD agar in triplicates and spread with a sterile bent glass rod. The plates were incubated at 37°C for 24 hours. Enumeration was then done using colony counter for colony forming units and expressed per mL of the sample (CFU mL\(^{-1}\)). Counting was done on presumptive Shigella colonies which appeared uniformly red. The colonies were confirmed by conducting oxidase, urea agar test, Methyl Red Voges Proskauer test.

2.4.4 Klebsiella spp

The ISO 21528-2:2004 method was used for enumeration of \textit{Klebsiella spp}. Appropriate dilutions of homogenate samples were pipetted on the surface of dried plates of XLD agar in triplicates and spread with a sterile bent glass rod. The plates were incubated at 37°C for 24 hours. Enumeration was then done using colony counter for colony forming units and expressed per mL of the sample (CFU mL\(^{-1}\)). Methyl Red Voges Proskauer test was done for confirmation.

2.5 Statistical Analysis

Statistical analysis of microbial cell counts was performed using Genstat software version 15 for windows. Data obtained from the Laboratory analysis of the samples was evaluated statistically using analysis of variance (ANOVA). Mean rating and Fischer’s Least Significant Difference was calculated.

3. RESULTS

3.1 pH and Microbial Counts Isolated from the Milk Samples

The pH and presence of selected microbial pathogens found in Suusac samples (n=28) from different areas of North Eastern region of Kenya are summarized in Table 1. The pH values for the samples range from 4.17 to 4.95. Samples coded 8 from Tana River, 10 from Mandera and 15 from Isiolo had the highest pH values averaging 4.93 while sample number coded 6 from Tana River, 24 and 27 from Isiolo had the lowest pH values. However, in terms of sites, all samples from Mandera had the highest pH values while samples from Tana River had the least pH values.

Sample coded number 25 from Tana River had the lowest \textit{E. coli} population count of 2.39 log\textsubscript{10} CFU mL\(^{-1}\), and the highest was sample number 27 from Isiolo which had a count of 3.41 log\textsubscript{10} CFU mL\(^{-1}\). \textit{Klebsiella spp}. was not detected in five samples coded 9, 10, 13, 21 and 23 from different parts of North Eastern Kenya but was present in the other samples. The highest count of 4.24 log\textsubscript{10} CFU mL\(^{-1}\), was in sample coded 20 from Isiolo. The highest number of \textit{Shigella spp} was in sample number 27 from Tana River, which had an average count of 3.28 log\textsubscript{10} CFU mL\(^{-1}\), and was absent in five samples number 8, 13, 14, 23 and 25 from different areas of North Eastern Kenya. The highest number of \textit{Staphylococcus aureus} was in sample number 27, and had a count of 2.86 log\textsubscript{10} CFU mL\(^{-1}\) and was absent in samples 3, 5, 6, 8, 11, 15, 18, 19, 21 and 25.
Table 1. Bacterial contamination and pH of suusac milk samples from different regions in North Eastern Kenya

<table>
<thead>
<tr>
<th>Samples (n = 28)</th>
<th>Region</th>
<th>pH</th>
<th>E. coli</th>
<th>Klebsiella spp</th>
<th>S. aureus</th>
<th>Shigella spp</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Isiolo</td>
<td>4.45</td>
<td>3.32±0.07&lt;sup&gt;abc&lt;/sup&gt;</td>
<td>3.42±0.09&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>2.6±0.3&lt;sup&gt;abc&lt;/sup&gt;</td>
<td>3.02±0.11&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
<tr>
<td>2</td>
<td>Isiolo</td>
<td>4.44</td>
<td>3.39±0.08&lt;sup&gt;ab&lt;/sup&gt;</td>
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<td>2.1±0.17&lt;sup&gt;cd&lt;/sup&gt;</td>
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</tr>
<tr>
<td>3</td>
<td>Namanga</td>
<td>4.58</td>
<td>3.5±0.07&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.16±0.06&lt;sup&gt;b&lt;/sup&gt;</td>
<td>ND</td>
<td>2.54±0.28&lt;sup&gt;abcd&lt;/sup&gt;</td>
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<td>4</td>
<td>Garissa</td>
<td>4.48</td>
<td>3.13±0.16&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>2.67±0.62&lt;sup&gt;d&lt;/sup&gt;</td>
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<td>2.83±0.16&lt;sup&gt;abcd&lt;/sup&gt;</td>
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<td>8</td>
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<td>ND</td>
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<td>10</td>
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<td>2.48±0.44&lt;sup&gt;abcd&lt;/sup&gt;</td>
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</tr>
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<td>3.14±0.00&lt;sup&gt;cd&lt;/sup&gt;</td>
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<td>20</td>
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<td>2.99±0.11</td>
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<td>21</td>
<td>Marsabit</td>
<td>4.48</td>
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<td>ND</td>
<td>ND</td>
<td>2.59±0.11&lt;sup&gt;abcd&lt;/sup&gt;</td>
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<td>ND</td>
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<td>3.17±0.12&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

LSD  
4.48  
0.25  
0.36  
0.43  
0.44

*Value along a column whose superscripts are different letters are significantly different at P<0.05. Units of bacterial counts are in log<sub>10</sub> CFU/mL. Each value is mean ± standard deviation for triplicate experiments. 
ND* = Not detected

Fig. 1. Percentage of samples contaminated by various bacterial pathogens
Camel milk is believed to have therapeutic ability but contamination may be common from raw camel milk and is consumed directly detecting Klebsiella spp with assessed by enumerating bacterial pathogens (present microorganisms samples could be due to production lactic acid by lowest pH values 6 from Tana River, 24 and 27 from Isiolo had the maximum of 3 Shigella spp to 4 from Mandera and 15 from Isiolo had the minimum of 3. Shigella spp had a mean value of 2. S. aureus was not detected in 22% of the samples. Escherichia coli in the samples collected had a mean value of 3.135 log10 CFU/mL, with a maximum of 3.591 log10 CFU/mL and a standard deviation of ±0.325. E. coli was detected in all the samples analyzed. Shigella spp in the samples collected had a mean value of 2.784 log10 CFU/mL, with a maximum of 3.322 log10 CFU/mL, and a standard deviation of ±0.350. It was not detected in 11.9% of the samples analyzed. Staphylococcus aureus in the samples collected had a mean value of 2.576 log10 CFU/mL, with a maximum of 3.041 log10 CFU/mL, and a standard deviation of ±0.300. It was not detected in 36.91% of the samples analyzed (Fig. 1).

### 3.2 Summary of Selected Pathogens Isolated from the Suusac Samples

Summary of each pathogen from the samples is shown in Table 2. *Klebsiella spp* in the samples collected had a mean value of 3.138 log10 CFU/mL, with a maximum of 4.003 log10 CFU/mL, and a standard deviation of ±0.4738. Klebsiella was not detected in 22.6% of the samples. *Escherichia coli* in the samples collected had a mean value of 3.135 log10 CFU/mL, with a maximum of 3.591 log10 CFU/mL and a standard deviation of ±0.325. *E. coli* was detected in all the samples analyzed. *Shigella spp* in the samples collected had a mean value of 2.784 log10 CFU/mL, with a maximum of 3.322 log10 CFU/mL, and a standard deviation of ±0.350. It was not detected in 11.9% of the samples analyzed.

### 4. DISCUSSION

The pH values for the samples range from 4.17 to 4.95. Samples number 8 from Tana River, 10 from Mandera and 15 from Isiolo had the highest pH values averaging 4.93 while sample number 6 from Tana River, 24 and 27 from Isiolo had the lowest pH values. The acidic nature of the milk samples could be due to production lactic acid by microorganisms. The microbiological hazards present in traditionally fermented camel milk (*Suusac*) in the Kenyan main market was assessed by enumerating bacterial pathogens. The results show high contamination of *Suusac* with *E. coli, S. aureus, Shigella spp,* and *Klebsiella spp* which the study aimed at detecting. Being that *Suusac* is traditionally made from raw camel milk and is consumed directly without undergoing any processing food contamination may be common.

Camel milk is believed to have therapeutic ability against many bacterial spp. due to the lytic action of lactoferrin and lysozyme present [11,12,13], but it is still a significant source of human infections [14,15,16]. The results indicated high level of microbial hazards in the products. *Klebsiella spp* was detected in 77.4% of the samples analyzed with an average of 3.138 log10 CFU/mL. The occurrence of this pathogen may be as a result of infection of the udder, poor hygiene of the handlers, cleaning, and disinfection of the *Suusac* containers [17]. The pathogen is associated with pneumonia, intraabdominal infections, urinary tract and bloodstream infections to humans and animals [18].

*Escherichia coli* was detected in many of the samples but according to KEBS standards (KS 941:2018), *Escherichia coli* should be totally absent in fermented milk. Various studies have shown *E. coli* O157:H7 is resistant to acid [19] and it can survive for long periods of time in fermented milk products [20,21,22]. There was a 36% prevalence of Shiga toxin-producing *Escherichia coli* (STEC) in camel milk with half of the isolates being from *Suusac* [23]. In Zimbabwe, 100% of all naturally fermented milk had *E. coli* [2]. The results indicate that *Suusac* could be an important medium for the transmission of pathogens to humans for instance strains of *E. coli* like O555, O111, O127 cause infantile diarrhea, while others like 06:H16, O5:H11, and O25:H42 produce potent enterotoxins capable of producing acute diarrhea.

The population of *Shigella spp* averaged 2.784 log10 CFU/mL and was detected in 88.1% of the samples analyzed. When ingested, *Shigella spp* grows in the intestine, then lyses and release endotoxins causing an infection called shigellosis. *Shigella spp* existed in raw camel milk samples but not detected in any tested samples of fermented camel milk in Iran [24]. Contamination of raw milk is usually from external sources [25]. Therefore, the results from this study clearly indicate there was contamination of the *Suusac* during production, storage or at the sales point.

### Table 2. Microbial analysis summary of selected pathogens from the Suusac samples

<table>
<thead>
<tr>
<th>Spp</th>
<th>N</th>
<th>Mean</th>
<th>Standard Deviation</th>
<th>Minimum</th>
<th>Maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Klebsiella spp</em></td>
<td>65</td>
<td>3.138</td>
<td>0.4738</td>
<td>2.000</td>
<td>4.303</td>
</tr>
<tr>
<td><em>E. coli</em></td>
<td>84</td>
<td>3.135</td>
<td>0.325</td>
<td>2.000</td>
<td>3.591</td>
</tr>
<tr>
<td><em>Shigella spp</em></td>
<td>70</td>
<td>2.784</td>
<td>0.350</td>
<td>2.000</td>
<td>3.322</td>
</tr>
<tr>
<td><em>S. aureus</em></td>
<td>53</td>
<td>2.576</td>
<td>0.3000</td>
<td>2.000</td>
<td>3.041</td>
</tr>
</tbody>
</table>

Data are mean values of triplicate samples. Units of bacterial counts are log10 CFU/mL.
Staphylococcus aureus in the samples collected had a mean value of 2.576 log_{10} CFU mL^{-1} and was detected in 63.1% of the samples analyzed. This showed the samples were highly contaminated with S. aureus which is an enterotoxins producer that causes gastroenteritis after consumption of contaminated food [26]. These results concur with those reported in Morocco where S. aureus was present in 30% of the samples with an average count of 2.32 CFU mL^{-1} [27]. Similarly, work done on Roub, a Sudanese traditionally fermented dairy product found that S. aureus was present in 60% of the samples analyzed with a bacterial count of 6.18 CFU mL^{-1} [28]. Another study found that this microbe is the most commonly isolated from udder infections in camels and causes diseases to both humans and animals [29]. Nosocomial and community-acquired staphylococcal infections are the most common cases reported in humans [30]. Coagulase positive and negative Staphylococci are pathogens which cause mastitis in animals [31,32].

Many factors along the informal Suusac production and market chain contribute to its quality and safety. The slow fermentation process of Suusac by traditional methods of production which is usually as a result of weak starter culture leads to contamination with pathogenic and toxigenic bacteria, molds, and other unwanted changes in the milk [33]. These pathogens have been found to grow faster than lactic acid bacteria [34]. The environment is a contributing factor to cross-contamination of Suusac. Milking area is usually open and dusty hence possibility of contaminating the milk and milk containers with microorganisms from the soil, milking personnel or camel coat during milking [35,36]. The study was done in the months of July and August when it was extremely dry and dusty hence high levels of contamination from the dust.

Suusac is traditionally prepared from unpasteurized milk [37]. Traditional preparation methods can mitigate foodborne diseases. Suusac flows through a long informal value chain in order to meet the increased demand from the urban areas, resulting in increased risk. There is increased handling of the product and also the informal end markets are in poor hygienic conditions. These informal markets are highly preferred by the poor and middle-class people because they are cheap, have trusted vendors who can give credit facilities but there is a high risk of product contamination from the dirty open drainages and dusty surroundings. There is no strict implementation of the food safety legislation hence this is a public health concern. Good quality water and proper sanitation are important if milk contamination is to be avoided [38]. Containers should be cleaned with clean potable water to avoid contamination [39] but another study reported that water in the ASALs is highly contaminated and scarce, thus difficult to improve the hygiene standards at the milking level [40]. This could have been a contributing factor to the microbial contamination.

In this study, it was also found that plastic containers of five, ten and twenty liters which are opaque with narrow openings are used for handling, storage, and transportation of Suusac. Therefore, this creates a problem in cleaning [41,42,43,44] and therefore a contributing factor to the pathogenic contamination.

Lactating camels with mastitis also contribute to foodborne pathogens and therefore, they can also be linked as a source of the pathogens [45,46,47]. The poor microbial quality of Suusac was contributed by the many interactive factors discussed above. Therefore, production of Suusac with unpasteurized milk with the poor hygienic conditions along the value chain as it is currently, poses potential public health risk as was reported in other studies [48,49].

5. CONCLUSION
The study concludes that the microbiological quality determined for Suusac was poor and of public health concern due to the presence of pathogens in the samples which according to KEBs standards (KS 941:2018) should be absent. This may be due to production processes, handling practices, storage vessels, selling method, and or sales environment.

The presence of life-threatening pathogens is a potential health risk which makes the product unfit for consumption. The Suusac being sold at Eastleigh market in Nairobi may be responsible for transmission of the pathogens to the consumers; therefore, there is a need for effective diagnosis.

COMPETING INTERESTS
Authors have declared that no competing interests exist.
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