



## BACTERIOLOGICAL EVALUATION OF SANITARY QUALITY OF MILK AND MILK PRODUCTS SOLD IN DUTSE METROPOLIS

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**ABSTRACT:** Milk is a rich source of protein and other nutrients in diet, but its perishability and high nutritional value makes it an ideal medium for the development of various microorganisms provided the condition is favourable. This characteristic makes it harbor pathogens of epidemiological importance. It has been previously established that milk and milk products harbor bacteria, predominantly *Escherichia coli*, *Staphylococcus aureus* and *Listeria spp.* Other species not commonly associated with milk include *Bacillus cereus*, *Enterococcus faecalis*, *Klebsiella spp.*, *Proteus spp.*, *Pseudomonas spp.*, and *Staphylococcus spp.* The poor quality of milk, unhygienic practices, poor animal husbandry practices, organization of milk supply chains and dysfunction of the regulatory authorities predispose the public to the risk of contracting milk-borne infections. However, the precise bacteriological sanitary quality of milk and milk products sold in Ranch (Riga), Yan tifa, Hakimi, Dutse central market and Love Garden (Federal University of Dutse) Dutse Local Government Area of Jigawa State from microbiological point of view remained largely unknown. The current study establishes for the first time the bacteriological profile of milk and milk products, estimated the microbial load and isolated selected bacterial pathogens along the milk value chain of the study area. The results revealed poor practices and lack of formal training on milk hygiene among most of the actors. More than 90% of all handled milk samples had Total Bacterial Count (TBC) above the European Union and the United States and National Agency for Food and Drug Administration and Control (NAFDAC) standards. The bacteria isolated in this study were *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella spp.* and *Shigella spp.* Educating the public, especially milk handlers in rural areas and also vendors, is a necessary action to ensure quality along milk value chain. To ensure this, policies by government, support services and training, and programmes through both government and nongovernment organizations should be created. Focus on research into health risks associated with milk contamination should also be prioritized.

**KEYWORDS:** Milk, Milk Hygiene, Bacteria, Contamination, Animal Husbandry Practices, Milk Pathogens.



## INTRODUCTION

Milk is an excellent source of protein and nutrients that are needed for normal growth and development. Milk and milk products are consumed across the globe as sources of energy, forming a major component of breakfast for adults and also being fed to infants as a substitute to breast milk. Milk is primarily obtained from bovines and caprines, especially cows and goats, and it is processed either locally or in factories to produce milk products, including but not limited to powdered milk, yoghurt, and cheese. The nutritional content of raw milk is excellent and is thus favourable for bacterial growth (Agustina Tan *et al.*, 2023; Azeez *et al.*, 2022; Machado *et al.*, 2017).

The nutrient present is also adequate for the growth of various pathogens. Results of extensive investigation over many years have proven that both human and animal diseases are often spread by milk and milk products. The bacterial species most commonly isolated from contaminated milk include *Campylobacter spp.*, *Salmonella spp.*, *Escherichia coli*, *Listeria monocytogenes* and *Cryptosporidium parvum* (Gebeyehu *et al.*, 2022; Hoffmann *et al.*, 2022). Other organisms commonly isolated from milk includes *staphylococci*, *streptococci*, *micrococci*, *Corynebacterium bovis*, *Mycobacterium spp.*, *Coxiella burnetii*, and coliform bacteria (Sipahi *et al.*, 2023; Hamouda & Abdul-Hafeez, 2023). The milk secreted into the udder of healthy bovines and caprines can be said to be sterile as no bacterial flora has been established to be resident in milk. Faecal contamination is the most likely route responsible for the presence of microbial pathogens in raw milk. Poor handling and processing practices could be another route of pathogen introduction into milk and milk products (Ghali-Mohammed *et al.*, 2022).

Domestication of animals or livestock has played a key role in the development of human civilisations. The cow has now become the main dairy animal associated with milk, with the term "milk" being almost synonymous with cow milk in most people's minds. However, milk from a range of other sources are also consumed and will therefore be covered in this chapter. The demand for milk in developing countries is expected to increase by 25% by 2025 (FOA, 2008).

All factors put together, the general condition of milk handling and marketing is likely to cause health complications to the public. These risks are associated with milk contamination leading to the growth and survival of harmful microorganisms and creating an avenue for the multiplication of other microorganisms as storage time and conditions, such as temperature and humidity, become more conducive (Hyera, 2015).

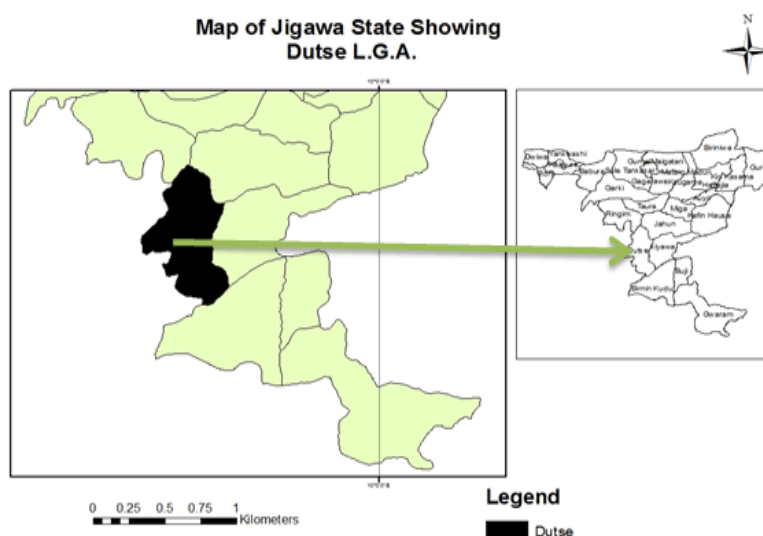
The aim of this study was to investigate the bacteriological sanitary quality of milk and milk products in Dutse Metropolitan. This study suggests that there is high bacterial load and low sanitary quality in milk and milk products sold in the study area.

## MATERIALS AND METHODS

### Study Area

This study was carried out in different districts of Dutse Metropolis (latitude 11° 45' N, 9° 20' E at 444 meters above sea level) in Jigawa State, Nigeria.

**Figure 1: Map of Jigawa State showing Dutse LGA. Source: GIS Lab., Federal University Dutse, Nigeria**



### Collection and Handling of Milk Samples

Eighteen (18) milk samples were collected from different locations in the study areas. Out of these samples, eight (8) were collected directly from the udders of milking cows and ten (10) were taken from other vendors/traders of milk and milk products and collection centres. The collection of samples was completely aseptic using a sterile container of 50 ml for each sample. The samples were instantly transferred to the central laboratory of Federal University, Dutse preserved in the refrigerator at  $-20^{\circ}\text{C}$  for storage until further analysis.

### Analyses of Milk Samples

Sample analyses were carried out in the Microbiology and Biotechnology Laboratory in the Faculty of Science at Federal University, Dutse using conventional microbiological methods. Media preparation was followed by suspension of samples and dilution. Microorganisms associated with sanitary quality of milk were isolated and identity was confirmed by gram staining and biochemical tests.

### Media Preparation

The culture media used included Peptone water, Nutrient agar, Eosin methylene blue, Mannitol salt agar, Cetrimide Agar and Salmonella-Shigella agar. The media were prepared according to the manufacturer's specification. These media were sterilized in an autoclave at  $121^{\circ}\text{C}$  for 15 minutes.



## Enumeration and Isolation of Pathogens

To evaluate the bacteriological sanitary quality of milk and milk products sold in Dutse metropolis, total bacterial count (TBC) and the presence of certain pathogenic bacteria in the milk samples were detected and compared to limits set by regulating agencies such as EU/US and NAFDAC. In this research, we focused on a few selected pathogens including *E. coli*, *S. aureus*, *Salmonella spp.*, *Shigella spp.* and *Pseudomonas aeruginosa*.

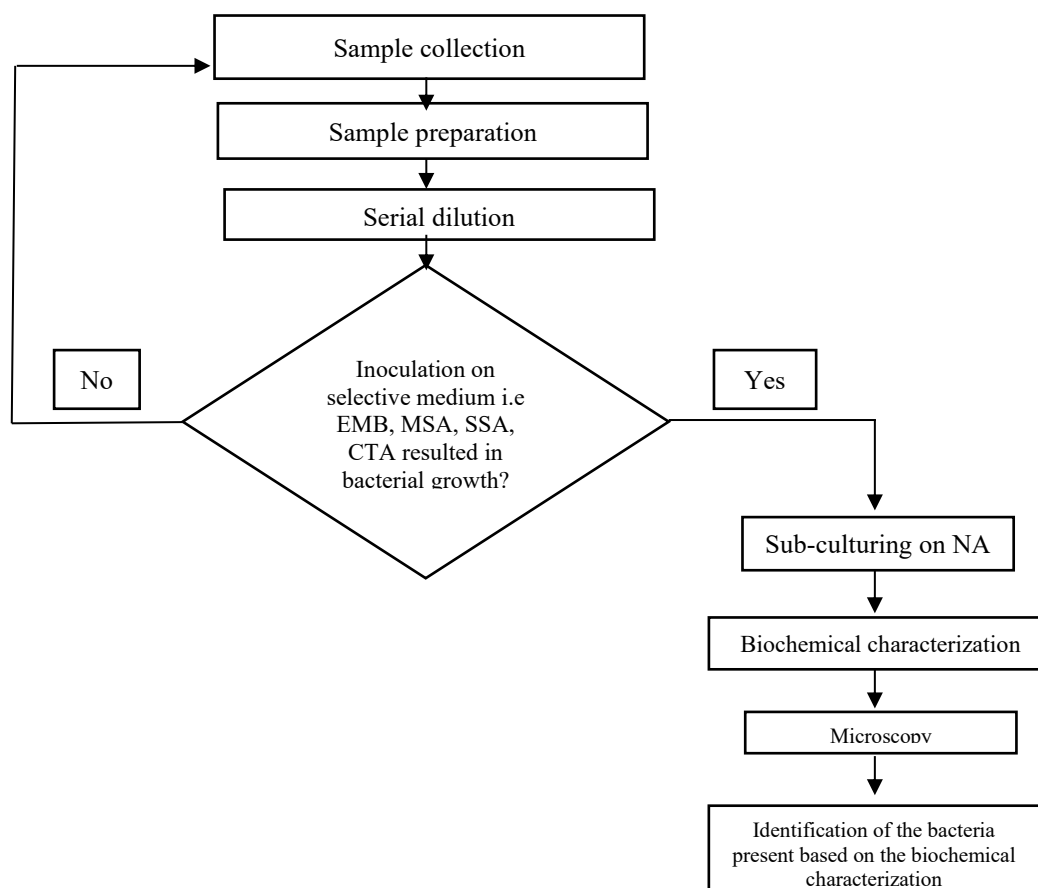
## Staining and Biochemical Tests

Gram stain and biochemical tests were carried out on the isolates. The biochemical tests were: Catalase test, Indole test, Methyl red test, Voges Proskauer test and Citrate test.

## Consent and Ethical Consideration

Data collected and results of laboratory microbial analysis of milk samples were under the protective care of researchers as confidential.

### Experimental Design





## RESULTS

### Results

#### Bacteriological Analysis

Among all the eighteen (18) samples, fifteen (15) samples (75%) were positive for bacteriological growth. Highest numbers of positive samples were noted in raw milk samples, where 5 out of 5 samples were positive for bacteriological growth, followed by milk products, where 8 out of 10 samples were noted positive, while in fresh milk, 2 out of 3 samples were noted positive.

*Salmonella* was noted with the highest percentage with 33.33%, *Staphylococcus aureus* with 30.56%, *E. coli* with 16.67%, and *Shigella* with 13.89%. *Pseudomonas aeruginosa* was detected in 5.56% samples.

#### *Escherichia coli*

*Escherichia coli* was detected in 6 (16.67%) samples. In Riga (5) and Love garden (1), the samples were observed positive for *E. coli*.

#### *Salmonella spp.*

*Salmonella* was detected in 12 (33.33%) samples. In Riga (5), Hakimi (1), Love garden (1), Yan tifa (2) and Dutse central market (3), one sample was observed positive for *Salmonella*.

#### *Shigella spp.*

*Shigella* was detected in 5 (13.89%) samples. All these (5) samples were observed to be positive for *Shigella* were in the milk samples collected from Riga.

#### *Staphylococcus aureus*

*Staphylococcus aureus* were detected in a total of 11 (30.56%) samples—in samples collected from Riga (5), Hakimi (2), Love garden (2), Dutse central market (1), and Yan tifa (1).

#### *Pseudomonas aeruginosa*

*Pseudomonas aeruginosa* was detected in a total of 2 (5.56%) samples. These two samples observed to be positive for *Pseudomonas aeruginosa* were in the milk samples collected from Riga.



**Table 1: Prevalence of gram positive and gram negative bacteria in milk and milk products**

Parameters	<i>E.coli</i> (-)		<i>S. aureus</i> (+)		<i>Salmonella spp</i> (-)		<i>Shigella spp</i> (-)		<i>P.aeruginosa</i> (-)	
	No.	%	No.	%	No.	%	No.	%	No.	%
Raw milk	5	100	5	100	5	100	5	100	2	40
Fresh milk	0	0	1	33.33	2	66.66	0	0	0	0
Milk products	1	10	5	50	5	50	0	0	0	0

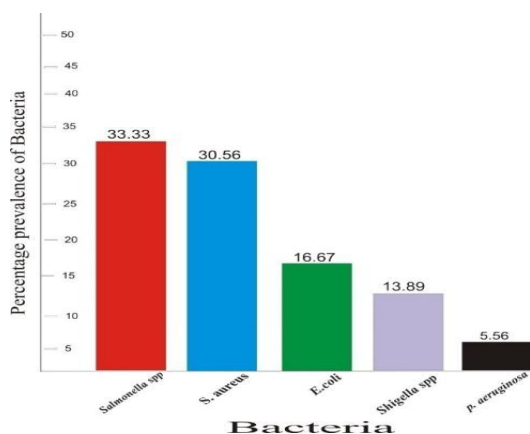
Table 1 shows the prevalence of some gram positive and gram negative bacteria in milk and milk products along the milk value chain.

**Table 2: Area wise microbial load in milk samples**

Sampling Samples	area	TBC(CFU/ml)	EU/US	Microorganisms			<i>Nafdac</i>		
		Mean count	Limits	<i>E.coli</i>	<i>S. aureus</i>	<i>Shigella</i>	<i>P.aeruginosa</i>		
		<i>Salmonella spp</i>				Limits			
Riga	5	$7.02 \times 10^4$	$<10^4$	5	5	5	5	2	Nil
Hakimi	2	$0.1 \times 10^4$	$<10^4$	0	2	0	0	0	Nil
Love garden	4	$0.96 \times 10^4$	$<10^3$	0	2	0	0	0	Nil
Yan tifa	3	$1.7 \times 10^4$	$<10^4$	0	1	1	0	0	Nil
Dutse Central market	4	$1.11 \times 10^4$	$<10^3$	1	1	1	0	0	Nil

Table 2 shows the area wise microbial load in milk and milk product samples along the milk value chain.

**Figure 3: Bacterial isolates' prevalence in milk and milk products.**





## DISCUSSION

In the present study, eighteen (18) milk samples were randomly collected from five (5) different locations of Dutse, including Riga, Yan tifa, Hakimi, Dutse central market and Love garden (FUD). Milk samples collected from each location were subjected to various tests for the detection of bacteria. Being an important food for humans, milk also serves as a good source of growth for various microorganisms.

Pathogens that have been found to be involved in food-borne outbreaks associated with the consumption of milk include *Escherichia coli*, *Salmonella spp.*, *Shigella spp.*, and *Staphylococcus aureus*. The presence of such types of pathogenic bacteria in milk has caused major public health problems, especially for those individuals who use such contaminated milk. *E. coli* was detected in 6 (33.33%) samples. These results are in agreement with those of Sarba *et al.* (2023) who reported that 33.8% samples were contaminated with *E. coli*. Ariyanti *et al.* (2022) reported 23.85% out of 109 fresh milk samples contaminated with *E. coli*, of which 17 (65.38%) out of the 26 isolates were confirmed as *E. coli* 0157:H7.

Milk and milk products can easily be contaminated with *Escherichia coli* and coliform bacteria and their presence in the milk is a sign of contamination of milk products. The presence of *E. coli* points to fecal contamination of the sample. *E. coli* 0157:H7, which is among the main leading and important bacteria indicating potential health risk, has posed a serious danger to the dairy industry for several years, causing outbreaks of many diseases in several countries (Mesele *et al.*, 2023).

Salmonella was detected in 12 (66.67%) samples. The presence of Salmonella and such other types of pathogenic bacteria in milk poses a great health risk and calls for alarm especially for individuals who consume Salmonella contaminated milk. Research by Shimelis (2023) in Bishoftu town showed a prevalence of 21.3% Salmonella in dairy cattle products with high resistance to multiple antibiotics. A study by Bedassa *et al.* (2023) in Areka town showed a 9.3% prevalence of Salmonella in raw milk samples. High contamination levels of Salmonella in raw milk samples at production and collection stages emphasize the need for targeted intervention strategies to reduce contamination risks (Bedassa *et al.*, 2023).

The incidence of *Staphylococcus aureus* isolates was 30.56% in this research, which is a little bit higher compared with that reported by Zhang *et al.* (2023), who obtained a prevalence of 20.8% in raw milk from 100 dairy farms in China. Thus, the maintenance of hygienic conditions in handling milk and processing milk products is important to prevent microbial contamination and ensure quality and safety (Fekata *et al.*, 2023).

Based on colony morphology and gram stain, the bacteria identified in the present study include *E. coli.*, *Staphylococcus spp.*, *Salmonella spp.*, *Shigella spp.* and *Pseudomonas spp.* The possible causes of these bacterial presence in milk and milk products could be due to either unhygienic handling practices or mastitis cow milk. The high proportion of *Staphylococcus spp.* isolated in the present research can be attributed to them being part of the normal teat skin flora and mucosa of humans and animals and, also, species are found free living in the environment.



## CONCLUSION AND RECOMMENDATIONS

### Conclusion

In the present study, eighteen (18) samples of milk were randomly collected from five (5) different locations of Dutse, including Riga, Yan tifa, Hakimi, Dutse central market and Love garden (FUD).

The present study revealed that raw cow milk and milk products sold in the market are unhygienic due to microbial contamination, and they have significantly high numbers compared to the NAFDAC, European Union and United states standards. An important source of microbial contamination of the milk is faecal pollution probably from cow dung and poor handling practices. Programs like ‘good hygiene practices’ and ‘good farm practices’ should be adopted at every step in milk handling and processing. Moreover, raw milk should not be used without processing (at least boiling) and, also, a strict check and balance system should be developed to control chemical adulteration.

The bacteriological profile of sanitary quality of milk and milk products along the milk value chain in the present research is challenging and needs the attention of all actors in the chain.

### Recommendations

In view of the conclusions above, it is therefore recommended that:

- i. There is a need to improve practices with respect to animal husbandry, milk processing and handling so as to minimize contact between microorganisms and lactating animals, environment and equipment by adhering to general hygiene practices and environmental cleanliness.
- ii. Sensitization of stakeholders along the milk value chain should be prioritized to improve their understanding of the importance of safe handling and management of animals, milk processing, milk hygiene and modern commercialization of milk.
- iii. Limited awareness on health risks associated with consumption of raw and/or raw fermented milk needs to be made by responsible authorities such as National Agency for Food and Drug Administration and Control, and the health sector, and the existing regulations must be instituted in order to safeguard community health.
- iv. Sector policies, organizational structures and support services for farmers and other actors must be properly concentrated in order to stimulate dairy sector development, particularly by strengthening the dominant informal sector.
- v. Further studies should be conducted to investigate the presence of other Verocytotoxigenic *E. coli* serotypes which are pathogenic for both humans and animals, to enable necessary action by concerned authorities.
- vi. The research into public health risks in milk should keep going and, in particular, special consideration should be given to cultural and milk handling practices which are the likely factors predisposing humans to infectious diseases, through the consumption of contaminated milk and milk products which are so common in various parts of the country.



## Authors' Contributions

**Corresponding Author:** Led the construction of the initial manuscript, conducted the laboratory analysis and rigorously monitored the editing process, ensuring consistency and clarity throughout. Conducted a thorough analysis and detailed comparison investigation, which increased the depth and breadth of the article's content

**Co-Author 1:** Provided crucial supervision throughout the process, offering advice and expertise that greatly improved the article's quality and trustworthiness.

**Co-Author 2:** Played an important role in the editing process, methodically evaluating and revising the content.

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## Competing Interest

The authors declare that they have no known competing interests or personal relationships that could have appeared to influence the review of this paper.

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