Assessment of the microbiological quality of beef marketed in commune I of Bamako district

ABSTRACT

Aims:
To assess the microbiological quality of beef marketed in commune I of Bamako district to determine its level of contamination and identify the main pathogens present.

Study design
A descriptive and analytical study was conducted between [from July to November 2023]. It involved meat samples collected from different points of sale in commune I.

Methodology:

Beef samples were collected randomly from markets, butcher shops and street stalls. Microbiological analyses were performed according to standardized protocols to enumerate total mesophilic flora, total coliforms, fecal coliforms, as well as to detect specific pathogens such as *Salmonella* spp., Escherichia coli and *Staphylococcus aureus*.

Results:
The results showed high microbiological contamination in [65,68%] of the samples analyzed, exceeding the thresholds set by food safety standards. Total and faecal coliforms were present in [23,35%]. Pathogens such as *Escherichia coli*, *Salmonella* spp., and *Staphylococcus aureus* were isolated in [26,39%]. Contamination rates were higher in informal outlets compared to licensed butchers.

Conclusion:
Beef marketed in Commune I has a worrying microbiological quality, constituting a potential risk to public health. There is an urgent need to strengthen hygiene measures throughout the production and sales chain, as well as to increase awareness of food safety among stakeholders.

Keywords: Microbiological quality, Fresh meat, Bovine, Market

Introduction

Mali has the largest cattle herd in West Africa, with an estimated national population of 12,111,128 heads (Teno, 2022; FAO, 2013). This potential place the country as the second largest livestock-producing nation in ECOWAS (Economic Community of West African States), after Nigeria, and first in UEMOA (West African Economic and Monetary Union) (Gning, 2021). Livestock farming plays a major role in the Malian economy, accounting for 30% of the primary sector's contribution to Gross Domestic Product (GDP) and 9% of national GDP (Samaké et al., 2008).
In Bamako, the capital of Mali, two large refrigerated slaughterhouses and seven other regional slaughterhouses serve the city and its surroundings with a total annual production capacity of around 12,000 tons (Samaké et al., 2008; Santara et al., 2019). Beef, particularly

in its fresh and unprocessed form, is an essential component of the local diet due to its richness in proteins and essential amino acids such as lysine and histidine. It plays a key role in human development, both physically and cognitively (Oumokhtar et al., 1998).
However, meat is also a favorable substrate for microbial proliferation due to its nutrient composition. Failures to comply with good hygiene practices at different stages of the supply chain (slaughter, transport, storage, and marketing) constitute a potential source of contamination and food poisoning. These gaps may result from poor operator training, inadequate hand washing, the use of non-sterile equipment, or the conditions in which meat is displayed on shelves (Boubaker Fattoum, 2021).
The microbiological quality of meat is an essential criterion for meeting consumer expectations in terms of food safety, nutritional quality, and preservation. Pathogenic microorganisms responsible for meat spoilage include bacteria (e.g., *Salmonella*, *Escherichia coli*, *Staphylococcus aureus*), yeasts, molds, and parasites (Daube, 2007; Barro et al., 2005). The preservation of fresh meat is particularly critical due to its short shelf life and requires appropriate techniques to prevent the multiplication of pathogens (Hamaidia, 2019; Ellies-Oury, 2016; Bellés et al., 2017).
Faced with these challenges, this study aims to count total mesophilic aerobic flora (TMAF), total and fecal coliforms, *Escherichia coli*, *Salmonella*/*Shigella*, *Staphylococcus aureus*, as well as yeasts and molds in marketed beef, to determine the microbial concentrations expressed in Colony Forming Units (CFU) per gram for each parameter and to evaluate the microbiological quality of meat sold in the markets of commune I of Bamako.

II. MATERIAL AND METHODS

2.1. Materials

2.1.1 Sampling Sites

This study was conducted in three different markets located in Commune 1 on the Right Bank. These markets, designated as Market 1 (M1), Market 2 (M2) and Market 3 (M3), were selected to represent beef sales areas within the commune.

2.1.2 Meat Sample Collection

Samples were collected in the morning, around 8 am, after post-mortem inspection of bovine carcasses. Thirty (30) samples were collected per market according to ISO 17604 (2003), for a total of ninety (90) samples collected randomly. Samples were collected three times a week over a period of four months.
Each sample was packaged in sterile bags and hermetically sealed, then labeled with information regarding the sample code, date, time and place of collection. The bags were transported in a cooler to the laboratory and stored at 4 °C before analysis.

2.2 Methods

2.2.1 Preparation of stock suspensions and decimal dilutions

Twenty-five grams (25 g) of each sample were homogenized in 225 mL of sterile physiological water to obtain a stock suspension. Decimal dilutions were then performed: 1 mL of the stock suspension was added to 9 mL of physiological water to obtain a 10^{-1} dilution, then repeated to achieve dilutions up to 10^{-}□.

2.2.2 Inoculation and counting of germs

2.2.2.1 Total mesophilic aerobic flora (FMAT) (Standard NF EN ISO 4833-1:2013)

One milliliter (1 mL) of each dilution was inoculated en masse in Petri dishes containing an appropriate agar medium. The dishes were incubated at 37 ± 1 °C for 72 ± 3 hours. The colonies developed were counted and expressed in colony-forming units per gram (CFU/g), retaining only the dishes containing between 30 and 300 colonies.

2.2.2.2 Total and faecal coliforms (NF V08-050:2009 Standard)

The samples were inoculated on Deoxycholate Agar and incubated at 37 °C for total coliforms (24 h) and at 44 °C for faecal coliforms (48 h). The red colonies observed were counted and expressed in CFU/g

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2.2.2.3 *Staphylococcus aureus* (NF EN ISO 6888-2:1999 Standard)

The samples were inoculated on Chapman agar and incubated at 37 °C for 48 hours. The bright yellow colonies were counted and expressed in CFU/g.

2.2.2.3 *Salmonella* sp. (Standard NF EN ISO 6579/A1:2012)

The Salmonella search included four steps:

1. Pre-enrichment: 25 g of meat in 225 mL of buffered peptone water, incubated at 37 °C for 8 to 24 hours.

2. Enrichment: 0.1 mL of pre-enrichment in 10 mL of Rappaport-Vassiliadis broth, incubated at 42 °C for 18 to 24 hours.

3. Isolation: Inoculation on Hektoen agar, incubated at 37 °C for 24 hours. Blue-green colonies with or without black center were presumed positive.

4. Identification: Biochemical test with TSI medium. After incubation incubated at +36.0 °C ± 1.0 °C, for 24 hours; Table 1 was used to identify Salmonella.

Tableau 1. Lecture des tests sur gélose triple sucre-fer (TSI)

	Lactose	Glucose	Saccharose	H_2S	Gaz
S.Typhi	-	+	+	+	-
***S.Paratyphi*A**	-	+	+	-	+
Other *Salmonella*	-	+	+	+	+

2.2.2.4 *Escherichia coli* (AFNOR SDP 07/1-07/93 method)

The samples were inoculated on TBX agar, incubated at 44 °C for 48 hours. The characteristic colonies of E. coli were counted directly in CFU/g.

2.2.2.5 Yeasts and molds (NF ISO 21527-2:2008 standard)

The samples were inoculated on Sabouraud Dextrose Agar and incubated at 37 °C for 48 hours. The colonies were counted and expressed in CFU/g.

Data analysis

The results were expressed in CFU/g and compared to international microbiological criteria in accordance with Regulation 2073/2005/EC. Data were analyzed using Excel pivot tables to calculate means, standard deviations, and covariances of the studied parameters.

3. RESULTS

3.1 Total Mesophilic Aerobic Flora (TMAF), Total and Fecal Coliforms, *Escherichia coli*, Salmonella/Shigella, *Staphylococcus aureus,* and Yeasts and Molds in Marketed Beef

Meats from the markets studied were highly contaminated by microbial pathogens, with significant variability between markets (Table 2). However, with the exception of coliforms, contamination did not vary significantly between repeat samples collected from each market.

Table 2. Analysis of variance for Total Aerobic Mesophilic Flora (FAMT), total and fecal coliforms, *Escherichia coli*, *Salmonella/Shigella*, *Staphylococcus aureus*, yeasts and molds in commercial beef

Sources of variation	Pathogenic microorganisms						
	DF	FAMT	*E. coli*	CF	CT	Staph	Moisissures
Markets	2	1213,58***	156,59***	28,69**	6353419***	648,26***	35979.1***
Repetitions	2	1,12NS	1,02NS	1,03NS	33,14**	1,35NS	1,32NS

****. ***,** significant at p<0.01 and p<0.001 respectively, NS: not significant, DOF; degree of freedom.

Fisher's test shows that meat from market 3 is the most contaminated by total coliforms, fecal coliforms and molds. On the other hand, meat from market 2 has higher concentrations of FMAT, *Escherichia coli* and *Staphylococcus aureus*. Market 1 is distinguished by a particularly high contamination by fecal coliforms (Table 3).

Table 3. Comparison of the means of total mesophilic aerobic flora (TMAF), total and fecal coliforms, *Escherichia coli*, *Salmonella/Shigella*, *Staphylococcus aureus*, as well as yeasts and molds in beef marketed in the market

Markets	**FMAT**	***E. coli***	**Fecal coliforms**	**Total coliforms**	**Staph**	**Yeasts and moulds**
Market 1	10982.53b	10285b	8134.3a	14939.3c	5342.33b	14939.3c
Market 2	8841.33a	18847a	6045.0b	15668.0b	9644a	15668b
Market 3	11218.67c	7919c	8734.0a	18771.3a	5010c	18771.3a

Apart from total coliforms, contamination by other microorganisms is not influenced by the repetition of sampling (Table 4). Contaminants appear to depend mainly on the specific conditions of each market (sources of supply, transport and hygiene at sale).

Table 4. Comparison of the means of total mesophilic aerobic flora (TMAF), total and faecal coliforms, *Escherichia coli*, *Salmonella/Shigella*, *Staphylococcus aureus*, as well as yeasts and moulds in beef marketed by repetitions

Repetitions	**FMAT**	***E. coli***	**Fecal coliforms**	**Total coliforms**	**Staph**	**Yeasts and moulds**
R1	10301.67a	11814.3a	7328.3a	12147b	16508.3a	6647.67a
R2	10371.93a	12619.0a	7791.3a	12159.33a	16515.0a	6671.33a
R3	10368.93a	12617.7a	7793.7a	12162.67a	16355.3a	6677.33a

3.2 Microbiological quality of meat marketed in the markets studied

The average microbial contents show that except for FMAT and faecal coliforms, the microbial loads of *Escherichia coli*, total coliforms, staphylococci and yeasts/moulds are higher than the standards (Table 5).

Table 5. Microbiological quality of meat marketed in the markets studied

Microorganismes recherchés et quantifies (UFC/g)						
Markets	**FMAT ($x10^4$)**	**E. coli ($x10^3$)**	**Fecal coliforms ($x10^4$)**	**Total Coliforms ($x10^4$)**	**Staph ($x10^3$)**	**Yeasts and moulds($x10^3$)**
Market 1	1.10	10.30	0.81	1.50	5.3	14.9
Market 2	0.88	18.90	0.61	1.57	9.6	15.7
Market 3	1.12	7.90	0.87	1.88	5.0	18.8
AFNORNorms	5.10^6 UFC/g	10^3 UFC/g	10^4 UFC/g	10^4 UFC/g	10^3 UFC/g	10^3 UFC/g

DISCUSSION

Microbiological contamination

Of the 90 samples analyzed (30 per market), FMAT concentrations were below the standards of 5×10 CFU/g, reflecting a good general state of preservation. These results are in agreement with those of Boukhenfar et al. (2019) and Hamaidia and Rouachdia (2019), who reported similar microbial loads on meats sampled in Algeria.
The presence of total and fecal coliforms, although fecal coliforms were below the standard, indicates improvable hygiene conditions. Djabou and Rafai (2021) also reported similar levels, although compliant with AFNOR/CODINORM and FCD (2015) standards.

Major pathogens

Escherichia coli concentrations were above the standards in all markets, with a peak at M2 (18500 CFU/g). These results differ from those reported by Boukhenfar et al. (2019), who found compliant loads under similar conditions.

For *Staphylococcus aureus*, our results corroborate the observations of Chadli and Farricha (2017) on meat products in Morocco, highlighting the risks associated with improper handling of meat.

Fungal contamination

Yeast and mold load largely exceed standards in all markets. These results are consistent with those of Boudjehem and Mazouni (2014), who reported high loads in similar products.

Presence of *Salmonella* spp.

The overall compliance with *Salmonella* spp. (92%) is satisfactory, but the 8% of non-compliant samples require increased monitoring. These results are consistent with those of Boukhenfar et al. (2019), who reported similar proportions.

CONCLUSION

The results highlight significant microbiological contamination in the three markets, mainly due to poor hygiene conditions and improper handling. Corrective measures, including safe handling practices and increased awareness among vendors, are essential to improve the sanitary quality of meat**.**

DISCLAIMER

The authors hereby declare that no generative AI technologies such as large language models (ChatGPT, COPILOT, etc.) and text-to-image generators were used in the writing or editing of this manuscript.

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