

Original Research Article

Hygiene practices and microbial contamination of liquid *Gapal* sold in Ouagadougou, Burkina Faso

ABSTRACT

Aims: Liquid gapal is a fermented food prepared mainly from millet paste and milk, and is increasingly consumed in Burkina Faso's major cities. This study aims to contribute to knowledge of the microbiological quality of liquid gapal marketed in Ouagadougou.

Methodology: To achieve this objective, we surveyed eleven (11) gapal producers located in ten (10) different districts of the city. Thirty-three (33) samples of liquid gapal randomly collected at a rate of three (3) samples per producer were then analyzed using standard microbiological analyzes methods.

Results: The results showed that some producers had poor control over good hygiene and production practices. They used of public mills to grind millet and lack of backup power systems that can contribute to promote microbial contamination. Microbiological analyzes showed that the level of microbiological contamination varied from one gapal producer to another. Microbial loads varied from 4.9 to 8.1 log₁₀ CFU/mL for total aerobic mesophilic microorganisms, 4.3 to 5.7 log₁₀ CFU/mL for yeasts and moulds, 4.6 to 8.8 log₁₀ CFU/mL for lactic acid bacteria, 1.4 to 4.6 log₁₀ CFU/mL for enterobacteria, 0.8 to 4.6 log₁₀ CFU/mL for thermotolerant coliforms, <1.0 UFC/mL to 3.7 log₁₀ CFU/mL for *Staphylococcus aureus* and 0.6 to 3.0 log₁₀ CFU/mL for *Bacillus cereus*.

Conclusion: These results underline the need to strengthen the capacities of these gapal producers in order to improve the quality of the products they marketed.

Keywords: gapal, Microbiological quality, *Staphylococcus aureus*, *Bacillus cereus*, Enterobacteriaceae.

1. INTRODUCTION

Milk and dairy products are widely consumed around the world. They are highly nutritious for humans, but also provide a good growth medium for micro-organisms (Ntuli et al., 2023). In general, contamination of foodstuffs by microbial agents can occur at any stage in food chain, from farm. There is therefore a need for rigorous monitoring of good hygiene and manufacturing practices throughout food chain to prevent microbiological contamination. Which contamination can result a high incidence of morbidity and mortality among consumers (Tropea, 2022). However, in many countries, mainly in developing countries, many foods are produced and marketed without quality control of either raw materials or final products. As a result, some foodstuffs on market are sources of food poisoning or food poisoning-infections.

In the case of dairy products, all stages from production to processing constitute risk situations for microbial contamination, as they can be influenced by animal health, milking, transport, storage and processing conditions (Owusu-Kwarteng et al., 2020). Dairy products (milk, yoghurt or fermented milk) are sometime added to other foods to make compound foodstuffs. This is the case of gapal or gappal in Burkina Faso, which is made from a mixture of millet paste and fresh milk, curdled milk or yoghurt. This traditional food, once popular with livestock farmers, is increasingly consumed in Ouagadougou, the capital of Burkina Faso (Tankoano et al., 2017). The special feature of this food is that it undergoes no heat treatment, unlike other foods made from millet and milk, such as dèguè, for which millet lumps undergo steam cooking (Hama et al., 2009). The addition of millet without any prior treatment other than washing with water, hulling (which is not always applied) and grinding could therefore constitute another source of contamination of the finished product (Bayili et al., 2024). In fact, post-harvest cereal practices, particularly during millet threshing, which is often carried out on the ground, could constitute a risk factor of microbial contamination that could lead to a rapid deterioration in the organoleptic and sanitary quality of non-heat-treated cereal-based foods (Albaridi, 2022). Millet can therefore be a source of telluric germs such as *Bacillus*, but it could also be a source of enterobacteria and other pathogenic microorganisms. Also, poor hygiene practices during gapal production could be a source of many other pathogenic microorganisms.

Previous studies have shown pathogenic microorganisms in foods made from milk and millet. This is the case of Ouattara et al. (2015), which showed the presence of *Klebsiella pneumoniae*, *Enterobacter agglomerans*, *Yersinia pestis*, *Serratia marcescens* and *Proteus mirabilis* in dèguè marketed in Burkina Faso (Ouattara et al., 2015). These results were recently confirmed by a study which showed a high load of thermotolerant coliforms, yeasts and moulds in samples of degué, gapal, wagashi and yoghurt from Bobo-Dioulasso (Bayili et al., 2024). In addition, other works have shown the contamination of Zoom-Koom, a local drink in Burkina Faso made from millet, as well as millet porridges by high-burden enterobacteria, yeasts and molds (Bsadjo-Tchamba et al., 2014; Kagambèga et al., 2019; Soma et al., 2020).

The present work therefore aims to investigate the microbiological quality of gapal marketed in the city of Ouagadougou. It will enable to update data on this foodstuff and contribute to improving the national food standardization system.

2. MATERIAL AND METHODS

2.1 Liquid gapal sampling

Sampling consisted in purchasing 500 mL of liquid gapal from eleven women producers not included in previous study of (Tankoano et al., 2017) and located in different arrondissements in Ouagadougou, the capital of Burkina Faso. Three productions were sampled, each spaced one week. A total of 33 samples were randomly collected. Samples were transported to the laboratory in a cooler containing melting ice to maintain the transport temperature at around 4°C. Samples were stored at 4°C and analyzed within 24 hours for Total Aerobic Mesophilic microorganisms, Enterobacteriaceae, Yeasts and Molds, Lactic Acid Bacteria, *Staphylococcus aureus* and *Bacillus cereus* determination.

2.2 Survey on compliance with good hygiene practices and good production practices for liquid gapal in Ouagadougou

The survey was carried out in two phases. The first phase involved identifying the main sellers of gapal in Ouagadougou. Once the production/sales sites had been identified, a questionnaire was drawn up and submitted to producers. Parallel information relating mainly to hygiene practices was collected directly on site during gapal production monitoring.

2.3 Assessment of the microbiological quality of liquid gapal

Ten grams of sample was mixed with 90 mL sterile peptone (1%) and NaCl solution (0.85%, w/v) into a homogenous suspension. Thereafter tenfold serial dilutions (10^{-1} - 10^{-7}) in peptone and NaCl solution were carried out. Total aerobic mesophilic microorganisms were determined according to (ISO 4833, 2003), lactic acid bacteria according to (ISO 15214, 1998), yeasts and molds according to (21527-1, 2008) enterobacteria according to NF (NF ISO 21528-2, 2017), thermotolerant coliforms according to (ISO 4832, 2006) *S. aureus* and *B. cereus* were counted according to (ISO 6888, 2003) and (ISO 7932, 2020) respectively.

2.4 Statistical processing of data

Data were analyzed using R version 3.6 and XLSTAT Pro version 2014. Analysis of variance (ANOVA) at the 5% significance level was performed using Tukey test. Principal component analysis (PCA) was performed using R software version 3.6.

3. RESULTS

3.1 Application of good hygiene and production practices

The results of the survey show that 90% of gapal producers included in this study do not wear appropriate clothing, and none of them wear bibs (Figure 1). However, work equipment and production area are cleaned before and after each production run, and hair is covered with scarves. Production monitoring showed that liquid gappal production processes vary according to producer. Differences were noted in terms of ingredients and sieving stage, which is not applied everywhere. As a result, two types of gapal were marketed, one whitish and the other grey. In addition, all producers use public mills for flour production. None of producers surveyed has of backup power systems, such as generators, solar system or uninterruptible power supplies (UPS). Only 18% of producers applied a recommended pasteurization schedule.

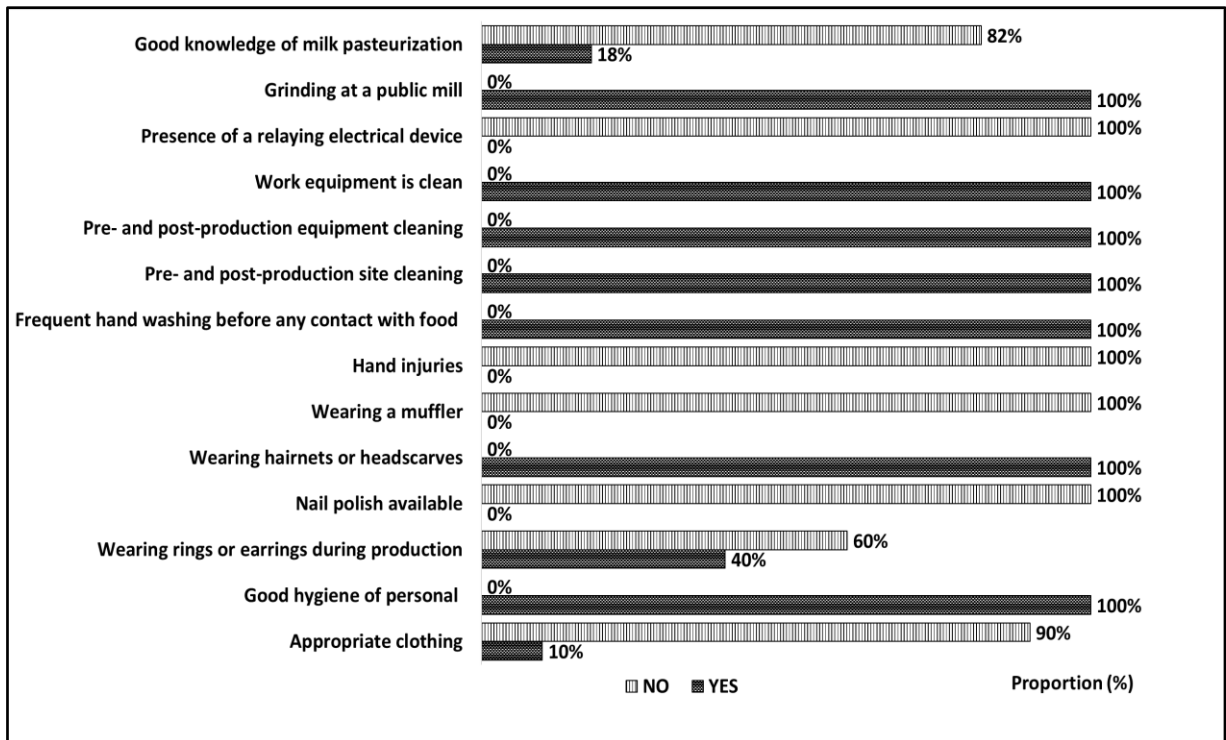


Fig. 1. Application of good hygiene during gupal production practices

Milk powder was used by 90% of producers and cow's milk in only 10% (Figure 2). As for ferment, 90% use yoghurt produced by another dairy, while the remainder keep yoghurt produced the previous day or curdled milk from the day before. About used millet 30% of use any variety available on the market, while 70% prefer a variety with large black grains that seems more suitable. Milk and gupal conservation are the main constraint encountered by producers. In fact, 80% of producers have difficulty in storing liquid gappal because of frequent power cuts.

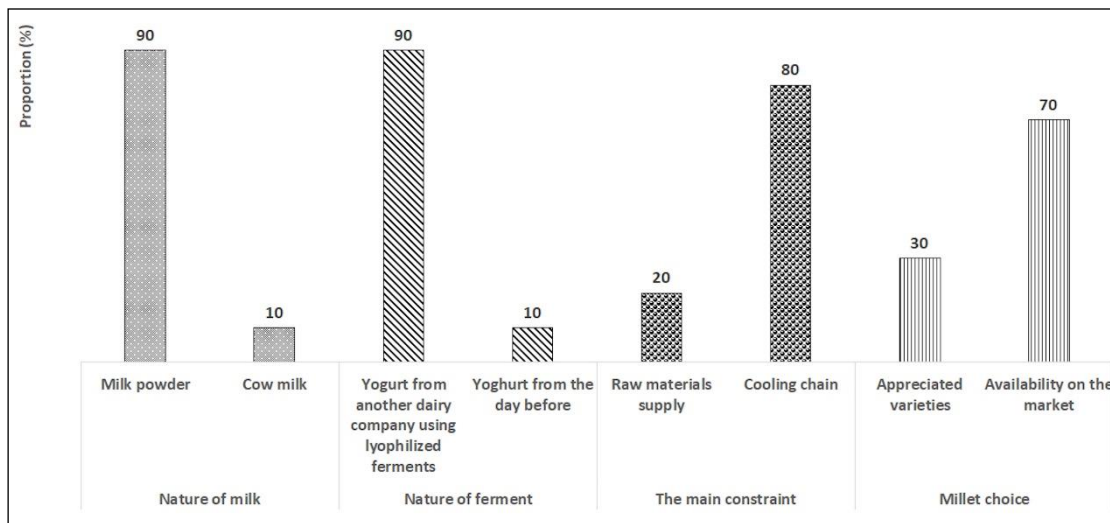


Fig. 2. Nature of main production raw materials and main constraints.

3.2 Microbiological quality of liquid gapal produced in Ouagadougou

The results of microbiological analyses are shown in Figure 3 and Figure 4. The average microbiological load was $7.8 \log_{10}$ CFU/mL for FMAT, $5.4 \log_{10}$ CFU/mL for yeasts and molds, and $8.0 \log_{10}$ CFU/mL for lactic acid bacteria. As for pathogenic microorganisms, we noted $3.8 \log_{10}$ UFC/mL for enterobacteria, $3.7 \log_{10}$ UFC/mL for thermotolerant coliforms, $3.0 \log_{10}$ UFC/mL for *S. aureus* and $2.4 \log_{10}$ UFC/mL for *B. cereus*. Analysis of variance shows that loads of total microorganisms, yeasts and molds, enterobacteria and lactic acid bacteria vary from one producer to another while loads of thermotolerant coliforms, *S. aureus* and *B. cereus* do not vary according to producer (Figure 5).

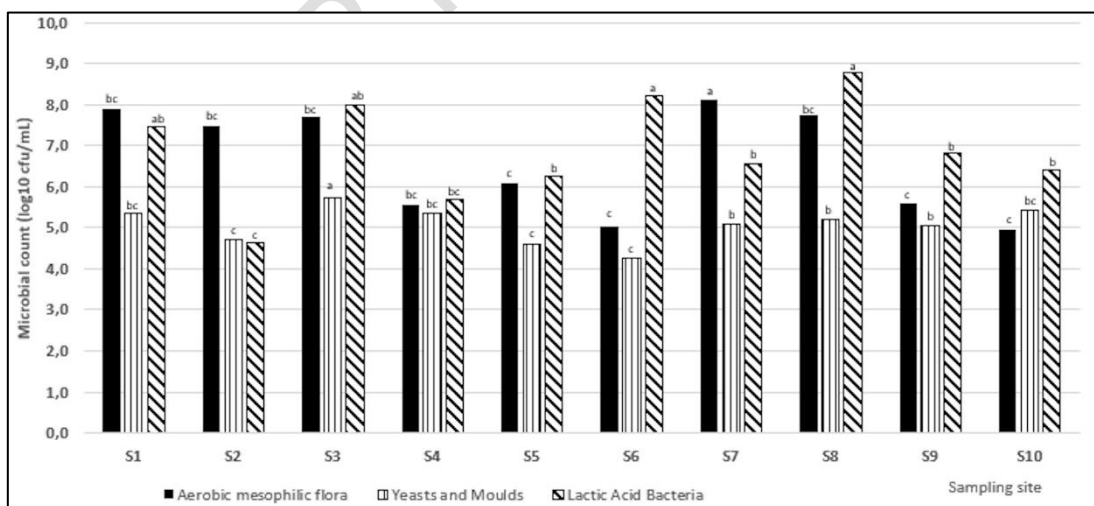


Fig. 3. Fermentative and alternative microorganisms

Interpretation of ANOVA is done by parameter according to sampling sites. When the letters are different for the same parameter from one sampling site to another, there is a significant difference ($p < 0.05$).

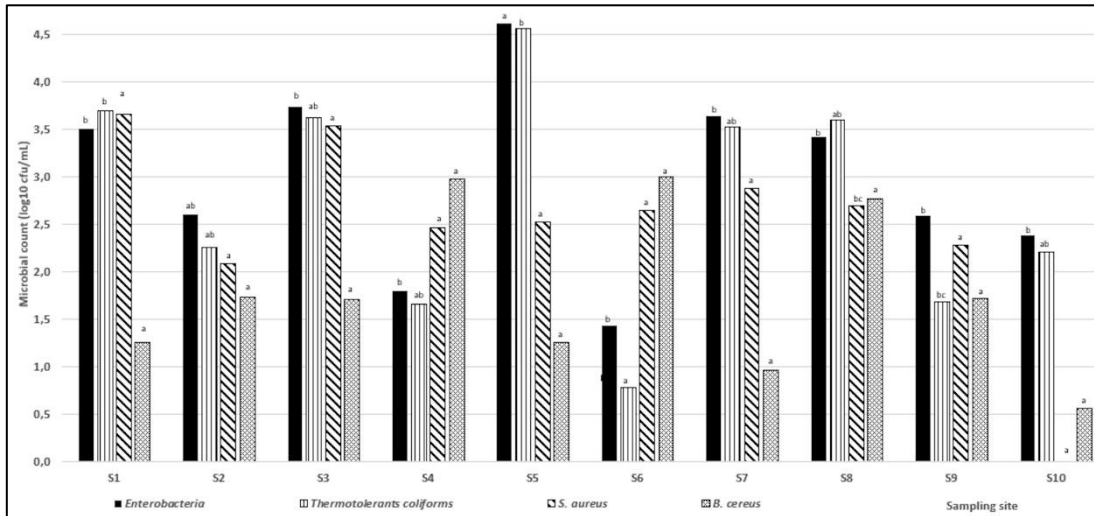


Fig. 4. Pathogenic microorganisms

Interpretation of ANOVA is done by parameter according to sampling sites. When the letters are different for the same parameter from one sampling site to another, there is a significant difference ($p < 0.05$).

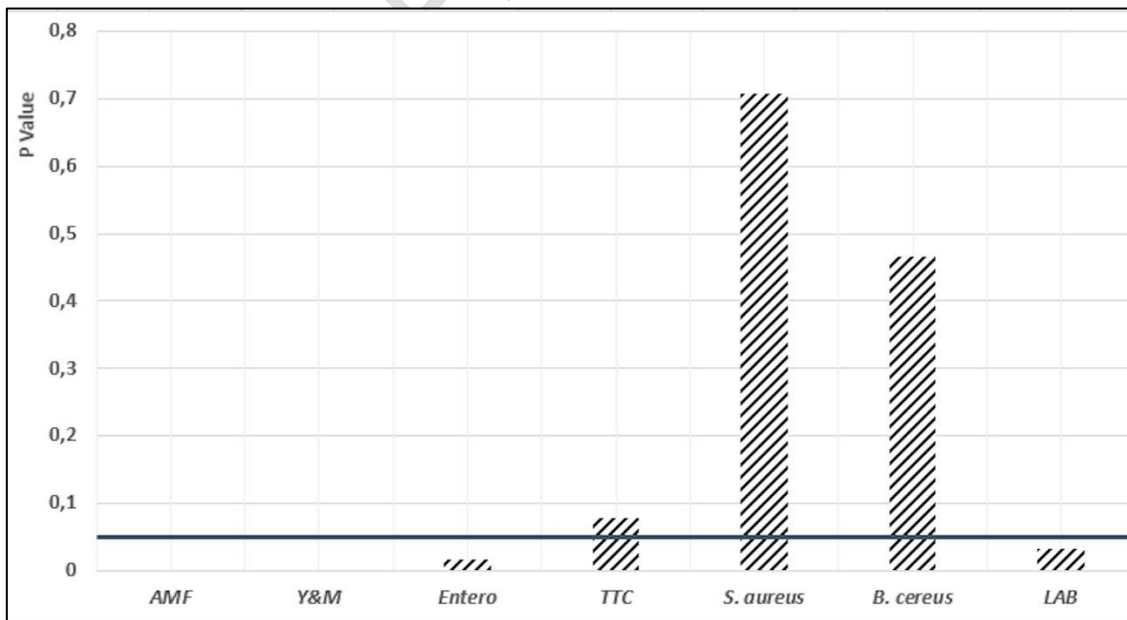


Fig. 5. Overall variability of microbial loads according to sampling sites

Legend:

AMF: Aerobic Mesophilic Microorganisms, Y&M: Yeast & Moulds, Entero: enterobacteria, LAB: Lactic Acid Bacteria

Table 1 shows the correlations between studied variables. It emerges that the increase in total mesophilic aerobic microorganisms in liquid gapal depend on the increase in yeast and mold load. Similarly, enterobacteria are strongly dominated by thermotolerant coliforms.

Table 1. Correlation between production conditions and microbiological quality

Variables	AMF	Y&M	Entero	TTC	<i>S. aureus</i>	<i>B. cereus</i>	LAB
AMF	1	0,88	-0,07	-0,05	0,07	0,06	0,2
Y&M	0,88	1	-0,09	-0,08	-0,01	0,03	0,11
Entero	-0,07	-0,1	1	0,97	-0,01	-0,13	-0,06
TTC	-0,05	-0,1	0,97	1	0,07	-0,13	-0,01
<i>S. aureus</i>	0,07	0	-0,01	0,07	1	-0,08	-0,02
<i>B. cereus</i>	0,06	0,03	-0,13	-0,13	-0,08	1	0
LAB	0,2	0,11	-0,06	-0,01	-0,02	0	1

Legend:

AMF: Aerobic mesophilic microorganisms, Y&M: Yeasts & Moulds, Entero: enterobacteria, TTC: Thermotolerant coliforms, LAB: Lactic Acid Bacteria

4. DISCUSSION

The results of the survey showed that some producers do not properly apply good hygiene and gapal manufacturing practices. The use of hands-on garments during gapal production could increase the risk of microbial contamination. This observation had been made previously in the milk sector in Burkina Faso but also in Senegal (Leone et al., 2022; Millogo-dah et al., 2019). All producers pasteurize milk (liquid or powder) but the survey revealed that time/temperature schedules are not perfectly mastered. Inadequate pasteurization temperatures may not eliminate pathogens if they were already in milk, and may even encourage faster pathogen growth (Owusu-Kwarteng et al., 2020). Using yoghurt produced the day before as ferment, or even yoghurt produced in other companies, does not guarantee quality of the ferment and therefore of gapal as well. This practice increases the risk of contamination of several products. Poor sanitation of processing environments and inadequate personal hygiene can lead to direct contamination of processed dairy products by pathogenic micro-organisms. In addition, the use of public mills to hull and grind millet is another factor in the contamination of millet flour used to produce gapal. In fact, the products processed in these public mills come from various horizons and actors. Quality control in these milling conditions is complicated by the diversity of the products to be ground and the miller's need to make a profit, which prevents him from cleaning the mill properly. These milling conditions may justify the high total aerobic mesophilic microorganisms load of 7.8 log₁₀ CFU/mL. The average value is higher than that obtained in gapal made from powdered milk sold in Bobo-Dioulasso the second city of the country, which was 6.96 log₁₀ CFU/mL. It is higher than the values obtained in 2017 from gapal sellers in the same town, which was 8.5.10⁸ CFU/mL or 8.9 log₁₀ CFU/mL (Tankoano et al., 2017). On the other hand, microbiological load is close to the value obtained on gapal made from cow's milk sold in the same town of Bobo-

Dioullasso, which was 7.74 log₁₀ UF/mL (Bayili et al., 2024; Tankoano et al., 2017). This total microorganism load could lead to rapid a food spoilage in case breaking in the cold chain. Indeed, the lack of backup power systems, such as generators or uninterruptible power supplies (UPS), can contribute to temperature fluctuations, which promote microbial growth.(Kussaga et al., 2014). The variation in total aerobic mesophilic microorganisms from one producer to another, which was also reported by (Tohoyessou et al., 2020) on dèguè sold in Cotonou, Benin, bears witness to the difference in the level of mastery of good hygiene and gapal manufacturing practices by female producers. It could also be linked to the difference observed in the production technology, mainly the absence of millet hulling among some female producers observed during the survey.

Yeast and mold load varied depending on the sampling sites (p=0.004) with an average of 5.4 log₁₀ CFU/mL. Contamination of gapal by yeasts and molds has been reported in other previous studies (Bayili et al., 2024; Tankoano et al., 2017). Yeasts and molds have been reported in other foods made from milk and millet such as dèguè has in Burkina Faso and Benin (Hama et al., 2009; Tchekessi et al., 2014). The load of yeasts and molds obtained in this study is lower than that found in millet dèguè sold in Cotonou, Benin which was 6.03.10⁷ CFU/mL or 7.8 log₁₀ CFU/mL. Table 1 shows that the total mesophilic aerobic microorganisms is strongly correlated with yeasts and molds. Yeasts and molds could therefore be the major aerobic mesophilic contaminants of liquid gapal. Yeasts and molds are common food contaminants. They can be carried by the environment and found in milk and dairy products ((Cissé et al., 2019; Tankoano et al., 2016). In the case of gapal, millet could be a source of contamination when it is not sufficiently washed before processing.

Lactic acid bacteria load in liquid gapal also varies depending on sampling sites (p= 0,034). Lactic acid bacteria are mainly provided by yogurts introduced as ferments. Lactic acid bacteria load obtained in this study is lower than that obtained on millet dèguè sold in Abomey Calavi in Benin which was 1.14×10⁹ CFU/ml or 9.1 log₁₀ CFU/mL (Tohoyessou et al., 2020). This difference in load could be explained by the quality of the ferments used as well as the difference in the proportions of millet flour or millet lumps used in the two cases. However, the use of lactic ferments makes it possible, among other things, to increase shelf life, improve health quality, improve nutritional quality, and produce flavors and aromas in foods. They can then significantly contribute to slowing down unwanted germs in the gapal (Huang et al., 2024; Xu et al., 2023; Yu et al., 2011).

The enterobacteria load varies depending on the gapal production sites with average values of 3.7 log₁₀ CFU/mL while that of and thermotolerant coliforms is identical with an average of 3.6 log₁₀ CFU/mL. The thermotolerant coliform load obtained in this study is higher than that found by Bayili *et al.* (2024) in the gapal made from powdered milk and sold in bobo which is characterized by an absence of thermotolerant coliforms. It is also higher than the values found on gapal made from cow's milk sold in the same city of Bobo-Dioullasso where the load was 1.55 log₁₀ CFU/g. On the other hand, it is in the range of the values found on thiakry sold at the large market of Bamako in Mali which varied between 1.9 log₁₀ CFU/g and 3.9 log₁₀ CFU/g (Sissoko et al., 2023). The presence of enterobacteria in liquid gapal, particularly thermotolerant coliforms, is an indicator of poor hygiene practices. This group of microorganisms, whose characteristic species is *Escherichia coli*, also harbors other strains of coliforms, such as *Citrobacter* spp, *Enterobacter* spp and *Klebsiella* spp responsible for toxic infections. Thermotolerant coliforms are the main enterobacteria in gapal (Table 1). The presence of thermotolerant coliforms is particularly significant, as it could be linked to recent faecal contamination in the food, potentially suggesting the presence of other enteric pathogens.

The *S. aureus* load which does not vary depending on the sampling sites is higher than the result found in liquid gapal sold in Ouagadougou in 2017 which was 1.5 log₁₀ CFU/mL and lower than the values obtained on thiakry sold on the large market of Bamako where the average load was 4.3 log₁₀ CFU/mL (Sissoko et al., 2023). *S. aureus* is a major foodborne pathogen and one of the main causative agents of mastitis in dairy ruminants. Its presence in gapal can come from operators but also from fresh cow's milk used in certain gapal production units when the pasteurization scale is not controlled. *Staphylococcus aureus* in dairy products in general has been documented previously because dairy products constitute good substrates for its multiplication (Tankoano et al., 2016; Tohoyessou et al., 2020). This pathogen can produce a variety of extracellular toxins, including shock syndrome toxin 1 (TSST-1), exfoliative toxins, staphylococcal enterotoxins (SE), hemolysins, and leukocidins (Abril et al., 2020). All makes it a microorganism to be feared in foodstuffs.

Bacillus cereus is a germ strongly associated with soil. In general, cereals which is an high starch content food are common vectors for *Bacillus cereus*, which is a common cause of food poisoning. It is a common environment bacterium which can grow in food (Albaridi, 2022). The sporulated nature of this bacterium makes it a difficult contaminant to eliminate. Although cooking at more than 55°C can kill the vegetative cells of *B. cereus*, but spores can survive cooking and then germinate when conditions are favorable (Albaridi, 2022). This could explain the load of *B. cereus* in gapal which is produced without any heat treatment. The process then increases the risk of proliferation of both vegetative form of and spores *B. cereus* in millet. *B. cereus* load in this study is significantly lower than that found in the previous study in 2017, which reported 7.2 log₁₀ CFU/mL. This decrease in *B. cereus* load could be linked to an improvement in millet washing processes that significantly contribute to reduce vegetative forms when it made in appropriate way. However, the current loads are high enough to be a health risk for consumers base on it ability to produce toxins. Over the past decade, food-borne epidemics and individual cases caused by bacterial toxins have been on an increasing trend. And the main culprits are enterotoxins and cereulide produced by *B.cereus*, which can cause diarrheic and emetic form of the disease (Jovanovic et al., 2022).

5. CONCLUSION

The study allowed an observation of hygienic conditions in which liquid gapal is produced in Ouagadougou and to evaluate its microbiological quality. Based on criteria applicable to foods close to the liquid gapal marketed, it appears that its microbiological quality needs improvement. There is a significant presence of mesophilic aerobic microorganisms, yeasts and moulds that can increase the rapid deterioration of gapal and the presence of pathogenic microorganisms including thermotolerant coliforms, *S. aureus* and *Bacillus cereus*. It is therefore important that awareness programs be settle to improve product quality and protect consumer health.

CONSENT

Not applicable

ETHICAL APPROVAL

Not applicable

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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