



A BRIEF DISCUSSION ON CURDLING TECHNOLOGY, MICROBIOLOGICAL RISKS AND CONTAMINANTS OF FERMENTED MILK NAMED *BÛSIM MISSIGA*

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ABSTRACT

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The objective of this study was to evaluate endogenous curdling knowledge, microbiological hazards and other contaminants of *Bisim missiga*. Thus, a survey was conducted among local producers to collect information on curdling technologies in five cities to Burkina Faso. The quality of *Bisim missiga* samples collected was assessed according to standard microbiological criteria. Contaminants and their sources have been identified according to literature review. The results of this survey revealed that fresh and powdered milk are the most used raw materials to production of *Bisim missiga*. Curdling technologies are similar in cities surveyed. Microbiological quality of *Bisim missiga* analysed was unsatisfactory according to criteria used. The presence of pathogenic potential was detected during microbiological analyses. Three types of contaminations have been identified according to literature. In sum, this study revealed that the *Bisim missiga* analysed do not comply with the criteria in force. Corrective measures must be taken to improve the quality of *Bisim missiga* sold in Burkina Faso.

Contribution/Originality: This study contributes to the existing literature knowledge of endogenous milk curdling technologies in Burkina Faso. In our study we are focused as well on the diversity of biological, physical and chemical contaminants of curd milk in generally. This study documents on the sanitary risk of the dairy products chain.

1. INTRODUCTION

Sahelian country, Burkina Faso is a country with agro-pastoral vocation, where livestock represents respectively the third export revenue of country after gold and cotton. Livestock contributes nearly 20% of gross domestic product to Burkina Faso (Duteurtre and Vidal, 2018). In West Africa, livestock breeding is one of driving forces for dynamisation to markets of agricultural and food products. In addition, this link in the economy is also of socio-economic importance since it occupies 30% of *Burkinabé*. This sector contributes to strengthening both food

and nutrition security and poverty reduction (CSAO-OCDE/CEDEAO, 2008). Strategies to improve food security and fight against poverty in households are put in place by the political authorities for the supporting livestock sector (Duteurtre and Vidal, 2018).

The production and sale of *Bïsim missiga* is an important strategy implemented in the country. Indeed, milk is a highly valued food whose nutritional and economic contribution within producing households is no longer demonstrated (Boko *et al.*, 2016; Cissé *et al.*, 2018). Several products are derived from milk processing, the best known in Burkina Faso are pasteurized milk, *Bïsim missiga*, butter (solid or liquid) and cheese (Hamadou and Sanon, 2005; Cissé *et al.*, 2019). The development of standard living has changed the technological processes and eating habits of populations.

Current trends show that demand of dairy products is growing in Burkina Faso. It should also be noted that from consumer trafficking through processing and sale, milk follows a short marketing cycle, that is from producer to consumer, but the intervention of milk collectors and resellers gives rise to a long circuit therefore the conditions of collection, storage and sales are very precarious (Kouamé-Sina *et al.*, 2010; Tankoano *et al.*, 2016; Cissé *et al.*, 2018). During this phase, milk is exposed to microbiological, physical and chemical contaminants. Its contaminants pose a significant health risk to consumers (Cissé *et al.*, 2018). The contamination of milk thus becomes a major public health problem, especially with the presence of microorganisms responsible for food poisoning. The consumption of defective hygienic milk causes digestive disorders that are often serious for consumers (Millogo *et al.*, 2018; Millogo-Dah *et al.*, 2019).

Thus, several risk factors for contamination of milk at different stages of production come into play. It is therefore important to control the quality of dairy products sold (Lompo *et al.*, 2006; Ogunshe *et al.*, 2015). To overcome this problem, standards are used in Burkina Faso to enforcement of laws and regulations related to food control. They impose on industrialists a harsh constraint but constitute the guarantee of hygienic and commercial quality assurance of products (Lompo *et al.*, 2006). This will allow the development of milk sector and reduction health risks, hence the interest of this study on the knowledge of traditional processes to production of *Bïsim missiga*, microbiological risks and contaminants'.

2. MATERIALS AND METHODS

2.1. Survey on Curdling Technology

A survey was carried out among producers-sellers, households and small milk processing units in the cities of Bobo-Dioulasso, Djibo, Dori, Gorom-Gorom and Sebba, whose objective was to know endogenous milk curdling technology for production of *Bïsim missiga*.

2.2. Microbiological Risk Assessment

Microbiological risks were assessed by applying the criteria for assessing the microbiological quality of *Bïsim missiga* by combining several standards. Data obtained in our previous studies on *Bïsim missiga* were interpreted according to criteria listed in Table 1 (Cissé *et al.*, 2019). In this study, only pathogenic potential microorganisms were considered. Thus, total coliforms and thermotolerant coliforms were counted on eosin methylene blue agar at 37°C and 44°C for 24-48 h. *Staphylococcus aureus* were counted on Baird-Parker agar supplemented with tellurium egg yolk and incubated at 37°C for 24-48 h, the black brilliant or dark gray colonies surrounded a clear halo were selected and confirmed by Gram, catalase, and coagulase tests. Three-class plan was used to interpretation of load in total coliform, thermotolerant coliform and *Staphylococcus aureus*. For thermotolerant coliforms, we used the standard according to National Public Health Laboratory (Burkina Faso), which is based on FASONORM standards based on French standards (Lompo *et al.*, 2006).

Table-1. Microbiological criteria used.

Appreciation	Total coliforms	Thermotolerant coliforms	<i>Staph. aureus</i>
Satisfactory	$N < 10$	$N < 1$	$N < 10$
Acceptable	$10 \leq N \leq 100$	$1 \leq N \leq 10$	$10 \leq N \leq 100$
Unsatisfactory	$N > 100$	$N > 10$	$N > 100$

Source: (Australia New Zealand Food Authority (ANZFA), 2001; Lompo *et al.*, 2006; Federation of Commerce and Distribution Companies (FCD), 2016; RCM, 2018) N: Number of microorganisms in colony forming units per millilitre.

2.3. Milk Contaminants

Milk contaminants and their effects on consumer health were searched in many databases as CrossRef, Google Scholar, Mendeley, PubMed, Science Direct and Scopus with keywords: Milk and Contaminants, Milk and Chemical Contaminants, Milk and Physical Contaminants, Milk and Organic Contaminants, Pathogens and Milk, Milk and Risks. Figure 1 shows the procedure for selecting articles and information used in this study.

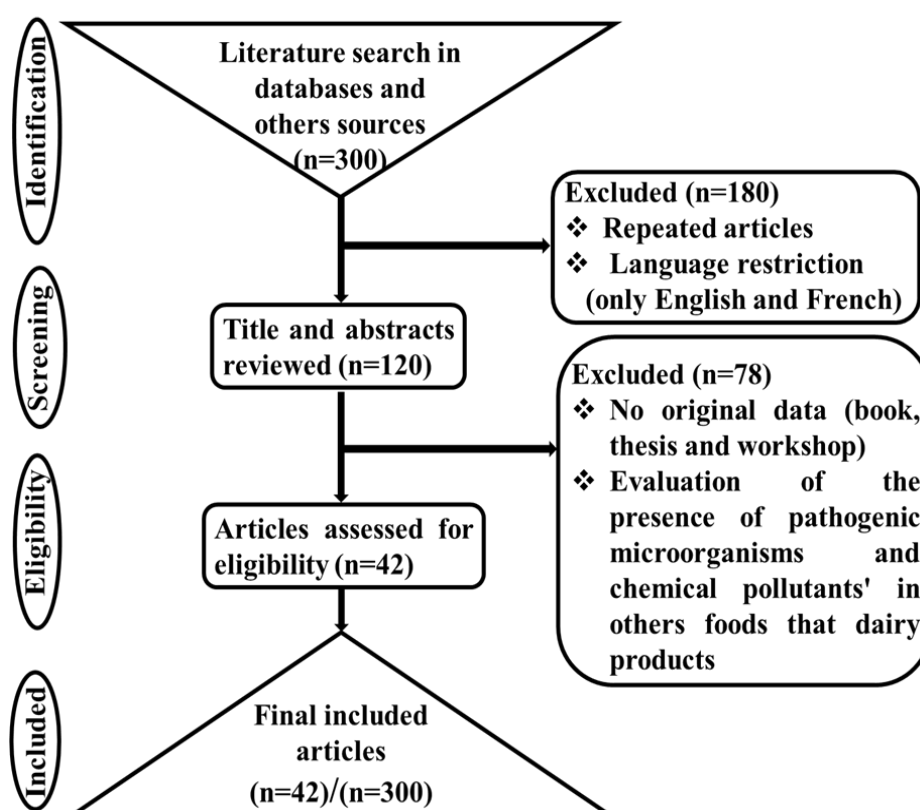


Figure-1. Procedure for selecting articles and information used.

2.4. Statistical Analyses

The data were processed by the XLSTAT 2016 software. Tukey HSD test was used for variance analysis (ANOVA) between modalities with a 95% confidence interval.

3. RESULTS

3.1. Endogenous Curdling Technologies in Burkina Faso

Figure 2 shows the traditional technology of curdling milk from milking in households in some cities of Burkina Faso. Figure 3 presents the technology for producing *B̄isim missiga* and *B̄is-ḡoem* from milk powder in households and small milk processing units in these cities. Figure 4 shows the conditions of milking, material used for curdling and exposure of *B̄isim missiga* to contamination during sales of markets this study.

Thus, according to information collected in this study near households and small milk processing units in Burkina Faso, curdling or fermentation technology is little variant. It is done by physical processes and biochemical

reactions without any chemical treatment in urban and rural areas. The main processes are heating, filtration, decantation, homogenization, fermentation and skimming. The heating ensures the sanitary quality of milk and its shelf life. It helps to guarantee certain technological characteristics, textural and enzymatic coagulation. Filtration consists of separating milk from physical contaminants (hair, sand, etc.). Settling allows a separation of contaminants according to their density. Homogenization is an operation to prevent the separation of milk constituents in different phases during the production of fluid milk and *Bîs-gôem*. The variants are at the level of the treatments of milk after milking, the raw material and the type fermentation used (spontaneous or use of ferments). These technologies are transmitted from generation to generation in certain ethnic groups' nomadic peoples such as *Fulani*, *Bella* and *Touareg*.

Nevertheless, we can note that in Burkina Faso, milk processing has undergone an innovative technological development in recent years. It has gone from ancestral practices to use of well-adapted industrial ferments and enrichments of dairy products with ingredients as fruits, Morinaga and other flavouring agents. These practices have been observed in some cities such as *Bobo Dioulasso*, *Djibo* and *Dori* with women's groups, which market dairy products with high added value in nutrients. According to producers, these products have some very interesting functional and nutritional properties.

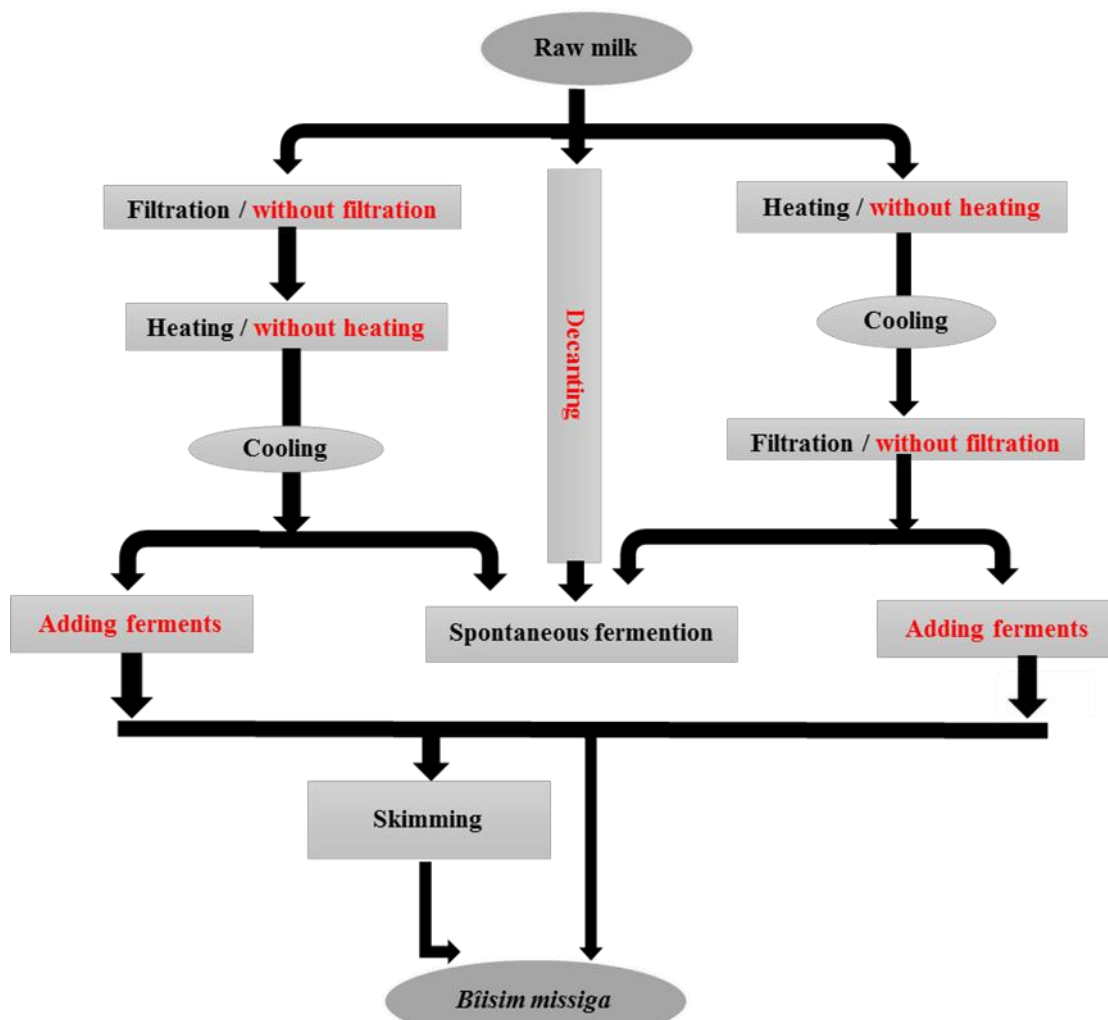


Figure-2. Technology for producing *Bîsim missiga* from milk after milking.

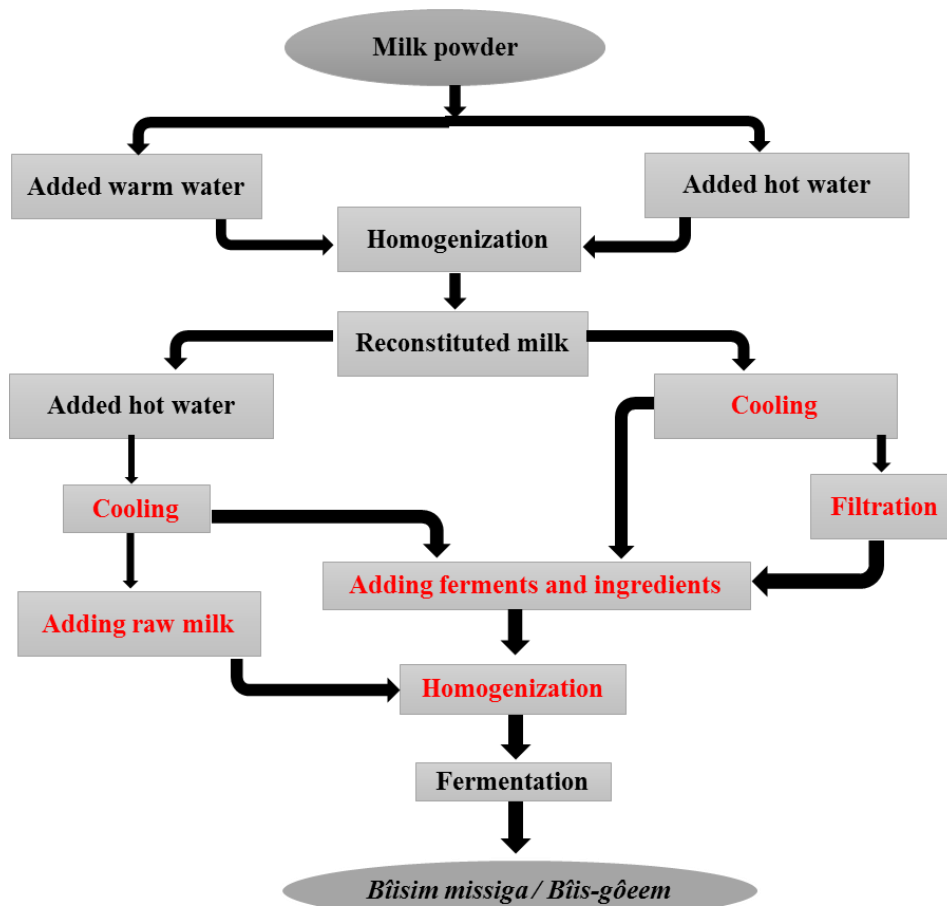


Figure-3. Technology for producing *B̄isim missiga* from milk powder.

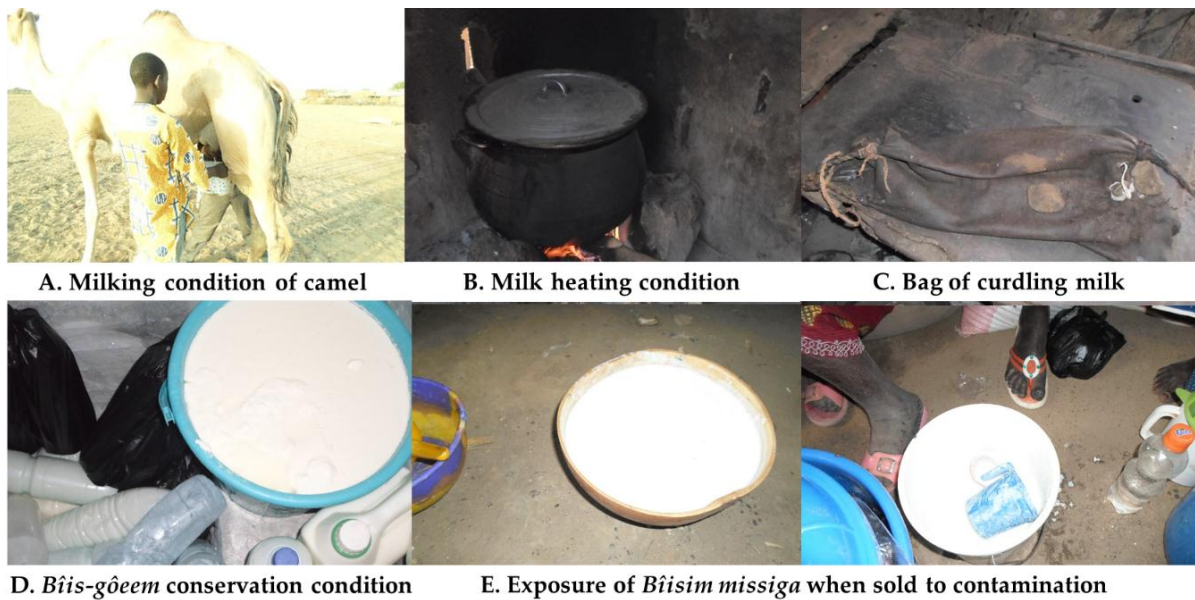


Figure-4. Practical aspects of milking to produce the *B̄isim missiga*.

3.2. Microbiological Risk

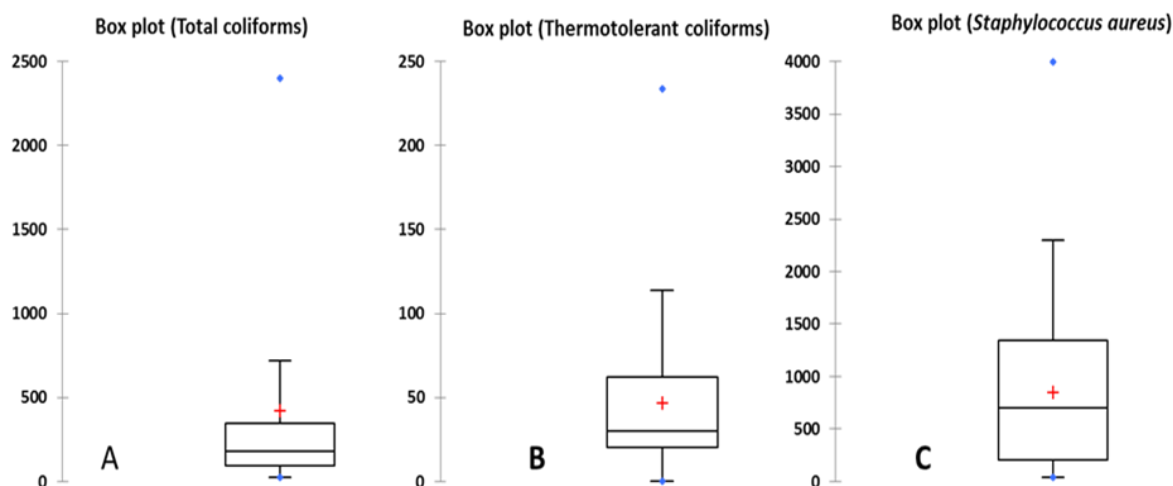
The results of microbiological analyses of *B̄isim missiga* are given in Table 2. The average loads of total coliforms, thermotolerant coliforms and *Staphylococcus aureus* were respectively 0.17×10^3 to 0.70×10^3 CFU/mL; 0.03×10^3 to 0.08×10^3 CFU/mL and 0.58×10^3 to 1.27×10^3 CFU/mL. The high loads in the latter come from *Gorom-Gorom* (total coliforms) and *Sebba* (thermotolerant coliforms and *Staphylococcus aureus*). The statistical analysis reveals that there is no significant difference ($P > 0.05$).

Table-2. Microorganism load of *B. missiga* analysed in CFU/mL.

Locality	Load	CT	CTT	<i>Staph. aureus</i>
<i>Bobo-Dioulasso</i> (n=10)	Mean	0.17x10 ^{3a}	0.03x10 ^{3a}	0.62x10 ^{3a}
	Min-Max	0.05-0.37x10 ³	<1-0.07x10 ³	<10-0.11x10 ³
<i>Djibo</i> (n=24)	Mean	0.22x10 ^{3a}	0.05x10 ^{3a}	1.06x10 ^{3a}
	Min-Max	<10-1.01x10 ³	<1- 0.23x10 ³	0.12-2.30x10 ³
<i>Dori</i> (n=26)	Mean	0.17x10 ^{3a}	0.03x10 ^{3a}	0.58x10 ^{3a}
	Min-Max	0.03-0.33x10 ³	<1-0.08x10 ³	0.04-2.20x10 ³
<i>Gorom-Gorom</i> (n=30)	Mean	0.70x10 ^{3a}	0.04x10 ^{3a}	0.77x10 ^{3a}
	Min-Max	<10-2.40x10 ³	<1-0.09x10 ³	0.14-2.12x10 ³
<i>Sebba</i> (n=24)	Mean	0.68x10 ^{3a}	0.08x10 ^{3a}	1.27x10 ^{3a}
	Min-Max	0.11-1.57x10 ³	0.02-0.17x10 ³	0.05-4.00x10 ³
P-value	-	0,067	0,207	0,257

Values in the same column with identical lowercase letters are not significantly different ($p > 0.05$); Mean: Average, Min: Minimum, Max: Maximum, CT: Total coliforms, CTT: Thermotolerant coliforms; *Staph. aureus*: *Staphylococcus aureus*.

Figure 5 shows that there is a great disparity of microorganisms from one sample to another ($p > 0.05$). Some samples are free of microorganisms and others contain them in abundance with only one extreme value.

**Figure-5.** Box plot of variation the number of microorganisms in *B. missiga*.

The results of correlation tests reported in Table 3 indicate a weakly positive correlation between all microbiological parameters of *B. missiga* analysed. However, these parameters are negatively correlated with origin of *B. missiga* except for Sebba, which has a weak positive correlation with them. These results show that the presence of thermotolerant coliforms in *B. missiga* involves total coliforms. *Staphylococcus aureus* and total coliforms are weakly correlated to origin of *B. missiga*.

Table-3. Correlation matrix between microbiological parameters and sampling localities.

<i>B. missiga</i>	Bobo	Djibo	Dori	Gorom	Sebba	<i>Staph</i>	CT	CTT
Bobo	1	-0,127	-0,161	-0,161	-0,127	-0,077	-0,112	-0,089
Djibo	-0,127	1	-0,301	-0,301	-0,237	0,127	-0,165	0,068
Dori	-0,161	-0,301	1	-0,382	-0,301	-0,234	-0,271	-0,134
Gorom	-0,161	-0,301	-0,382	1	-0,301	-0,066	0,290	-0,162
Sebba	-0,127	-0,237	-0,301	-0,301	1	0,261	0,214	0,324
<i>Staph</i>	-0,077	0,127	-0,234	-0,066	0,261	1	0,295	0,078
CT	-0,112	-0,165	-0,271	0,290	0,214	0,295	1	0,353
CTT	-0,089	0,068	-0,134	-0,162	0,324	0,078	0,353	1

Table 4 presents the interpretation of the quality of *B. missiga* analysed by applying the assessment criteria given in Table 1. This interpretation reveals that none of *B. missiga* analysed complies with the

recommendations of standards. However, when looking at microorganisms in turn, we note that on to 114 of *Bisim missiga* analysed, only 1 was satisfactory in *Staphylococcus aureus*, 14 were satisfactory in thermotolerant coliforms and 6 satisfactory in total coliforms. This result shows that there is a high microbiological risk on health related to consumption of these *Bisim missiga* analysed during this study. Thus, the quality of food is based on the absence of pathogenic and unwanted germs causing infections or organoleptic changes. The non-compliance of certain quality criteria according to international standards does not immediately render the food unsafe for human consumption.

Table-4. Overall assessment the results of microbiological analyses.

Locality	Appreciation	CT	CTT	<i>Staph. aureus</i>	Global app
<i>Bobo</i>	Satisfactory	0/10(0%)	2/10(10%)	1/10(10%)	0/10(0%)
<i>Dioulasso</i> (n=10)	Acceptable	4/10(40%)	3/10(30%)	3/10(30%)	1/10(10%)
	Unsatisfactory	6/10(60%)	5/10(50%)	6/10(60%)	9/10(90%)
<i>Djibo</i> (n=24)	Satisfactory	2/24(8.33%)	2/24(8.33%)	0/24(0%)	0/24(0%)
	Acceptable	7/24(29.17%)	3/24(12.5%)	10/24(41.67%)	3/24(12.5%)
	Unsatisfactory	15/24(62.50%)	19/24(79.17%)	14/24(58.33%)	21/24(87.5%)
<i>Dori</i> (n=26)	Satisfactory	0/26(0%)	4/26(15.38%)	0/26(0%)	0/26(0%)
	Acceptable	16/26(61.54%)	8/26(30.77%)	15/26(57.69%)	2/26(7.69%)
	Unsatisfactory	10/26(38.46%)	14/26(53.85%)	11/26(42.31%)	24/26(92.31%)
<i>Gorom-Gorom</i> (n=30)	Satisfactory	4/30(13.33%)	06/30(20%)	00/30(0%)	0/30(0%)
	Acceptable	9/30(30%)	4/30(13.33%)	7/30(23.33%)	0/30(0%)
	Unsatisfactory	17/30(56.67%)	20/30(66.67%)	23/30(76.67%)	30/30(100%)
<i>Sebba</i> (n=24)	Satisfactory	0/24(0%)	0/24(0%)	0/24(0%)	0/24(0%)
	Acceptable	8/24(33.33%)	6/24(25%)	8/24(33.33%)	0/24(0%)
	Unsatisfactory	16/24(66.67%)	18(75%)	16/24(66.67%)	24/24(100%)

CT: total coliforms, CTT: thermotolerant coliforms, *Staph. aureus*: *Staphylococcus aureus*, Global app: Global appreciation, Values in brackets refer the ratio of number of samples according to criteria standards of assessment to the total number of samples analysed.

3.3. Overview of Milk Contaminants

The contaminants founded in milk and others dairy products are listed in Table 5. These contaminants can be defined as any form of pollution or contamination that makes a food unfit for consumption. Thus, according to *Codex Alimentarius*, a food hazard is any agent present in a food causing health damage after his ingestion. These contaminants may be of a chemical nature (pesticides, antibiotics, mycotoxins, trace elements metal (TEMs)), biological (microorganisms, enzymes) or physical (grains of sand, hair). Their origin being either endogenous or exogenous, makes the sanitary quality of local fermented foods unpredictable. Thus, there is a health risk with negative impact. This impact can be immediate in case of poisoning and allergies or delayed in case of cancers and degeneration of certain organs and physiological functions.

Table-5. Sources of hazards in fermented dairy products.

Danger	Examples of contaminants
Microbiological	Pathogenic microorganisms (<i>Pseudomonas</i> , <i>Aeromonas</i> , <i>Serratia</i> , <i>Acinetobacter</i> , <i>Alcaligenes</i> , <i>Achromobacter</i> , <i>Enterobacter</i> , <i>Chryseobacterium</i> , <i>Flavobacterium</i> , <i>Bacillus</i> , <i>Clostridium</i> , <i>Corynebacterium</i> , <i>Microbacterium</i> , <i>Micrococcus</i> , <i>Staphylococcus</i> , <i>Salmonella</i> , <i>Listeria</i> , <i>Shigella</i> , <i>E. coli</i>), toxigenic (<i>Aspergillus</i> , <i>Fusarium</i> , <i>Penicillium</i>), parasites (<i>Toxoplasma gondii</i> , <i>Entamoeba histolytica</i>), virus (<i>Vibrio cholera</i> , Hepatitis A virus, <i>Norovirus</i>) and prions (BSE, GMO)
Chemical	Biogenic amines (putrescine, histamine, tyramine, cadaverine, sperimine, tryptamine) mycotoxins (aflatoxin, ochratoxin A, patulin, citrinin, paxillin, fumonisins, zearalenone, trichothecenes), ethyl carbamate, food additives, pesticide / antibiotic residues, chemical contaminants (rusty dishes and mines), allergens, phycotoxins (algal toxin) TEMs (Ni, Cr, Cu, Zn, Pb, As) and radioactive elements
Physical	Debris of metals, plastics, plants, glasses, grains of sand, animal hair, human hair and bone fragments

Source: This table is the results of 34 articles dealing with contaminants in curdled milk.

4. DISCUSSION

Milk, both a food and a drink, is of great nutritional value for the consumer (Manzi *et al.*, 2013). During his processing, milk from milking can be heated or unheated and then seeded with *Bifidobacterium missiga* from old or unseeded (Figure 1 and 2). This transformation is often carried out under very precarious hygienic conditions according to our observations. Thus, the poor hygienic quality of equipment, ingredients used and the non-respect or ignorance the rules of hygiene expose the product to contamination (Moullec, 2002; Millogo *et al.*, 2018; Ayaz *et al.*, 2018). Thus, from milking to consumption through processing, milk can undergo significant change due to action of microorganisms (Bonfoh *et al.*, 2006; Kouamé-Sina, 2013). The deterioration of organoleptic qualities (taste, aroma and texture) and reduction of shelf life the milk are to do the action of alteration flora. This flora is composed of sporulating bacteria (*Bacillus* and *Clostridium*), certain yeasts and moulds, *Escherichia coli* and *Enterobacter*. Harmful, it often becomes pathogenic for consumers. However, contaminated, *Bifidobacterium missiga* can be a vector for transmission of zoonoses (viral, bacterial and parasitic), prions and other toxic elements to humans (Lompo *et al.*, 2006; Early, 2017; Asante *et al.*, 2019). These pathogenic are responsible the damage on health of consumers and comes mainly from human, animals and environment (Edalati *et al.*, 2019). Thus, *Bacillus cereus*, *Campylobacter*, *Clostridium*, *Escherichia coli*, *Listeria monocytogenes*, *Salmonella* and *Staphylococcus* are the pathogens frequently found in milk (Yobouet *et al.*, 2014; Amenu *et al.*, 2019). These microorganisms are the origin of food poisoning and digestive disorders. The level of this contamination is closely related to hygienic conditions of milking, state cleanliness of animal, material used, environment, knowledge and to application the good practices of rules hygiene by the actors (Maiwore *et al.*, 2018; Millogo *et al.*, 2018; Cissé *et al.*, 2019). Some microorganisms pass transiently into their food producing toxins or spores, which after ingestion are very fatal for consumers (Murphy *et al.*, 2019; Xiong *et al.*, 2019). The presence of certain contaminants does not render this food completely unfit for human consumption, but depends the limited dose. This is the case of biogenic amines whose presence constitutes a significant health risk. But their low-grade consumption can't be a serious risk. Because they can be degraded by amine oxidases in intestinal lumen (Pessione and Cirrincione, 2016; Gu *et al.*, 2018).

When milk is kept in poor conditions, it is prone to contamination by toxigenic moulds and bacteriophages (Fernández *et al.*, 2017; Nishimwe *et al.*, 2019). The presence of these moulds in dairy products as *Aspergillus*, *Fusarium*, *Stachybotrys* and *Penicillium* potential mycotoxin producers represents a danger for human and animal health (Abo El-Makarem *et al.*, 2019; Alnaemi, 2019). Toxins produced by these moulds must be taken into account when monitoring the sanitary quality of fermented dairy products intended for consumption (Melini *et al.*, 2017). These toxins can cause several disorders at the consumer manifested by many syndromes such as hepatotoxic, neurotoxic, mutagenic, teratogenic, carcinogenic and immunosuppressive effects (Alshannaq *et al.*, 2018; Ezekiel *et al.*, 2018). These effects can be manifested by gastroenteritis, haemorrhage, convulsions, paralysis, oestrogen, kidney and liver damage leading to cancer (Okpala and Ifeoma, 2019; Gauthier, 2016). Some practices, as feeding animals with altered foods, provide a route for transfer of these toxins to humans. These practices are real public health problems in rural areas where feed is mixed with food damaged by mould. Many studies were found that animals were alimented by contaminated feed and waiting time for milking after administration of antibiotics was not respected (Wanniatie *et al.*, 2019). Similarly, we have also noticed the dirty environments of sales and transformations, the washing of materials used in traditional gold panning sites at watering points of animals (Cissé *et al.*, 2018).

Others TEMs toxic as Aluminum and Barium can be found in many dairy and agricultural products (Bakircioglu *et al.*, 2018; Tanoi *et al.*, 2019). However, the presence of pesticide, antibiotic and TEMs in food can be fatal to consumers (Bakircioglu *et al.*, 2018; Ziarati *et al.*, 2018; Lapierre *et al.*, 2019). Without doubt, the presence of all its contaminants is a logical result of lack of awareness and poor leadership on the concept of "One Health", a key factor in food safety (Roesel and Grace, 2016; Bell *et al.*, 2018). Thus, hazard identification is the most important steps in risk assessment for locally sold foods to ensure everyone's health (Tourette, 2002). If all parameters of this

factor are taken, there will be a reduction risk leading to positive impact on public health, ecology and economy according to Figure 6 (Bell *et al.*, 2018).

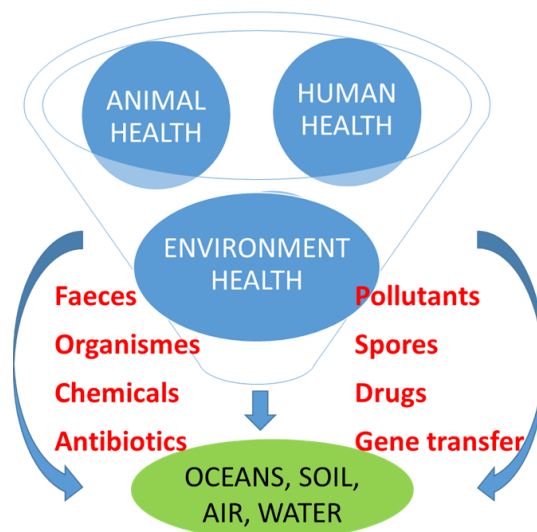


Figure-6. General concept of the one health.

5. CONCLUSION

This study shows that there is a high microbiological risk in to *Bisim missiga* production and processing chain in Burkina Faso. Monitoring and evaluation the health quality of milk for processing and consumption is necessary to risk reduction. To overcome this problem, establishment of training, awareness cleanliness, animal, environmental health, hygiene and storage conditions to benefit for farmers, processors and sellers are necessary.

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