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Assessment of the risk of *Staphylococcus aureus* infection associated with the consumption of braised beef meat sold on the streets in Côte d'Ivoire

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Abstract

Braised beef meat sold on the streets, a meat product that poses a potential risk of food poisoning to humans. This study was to assess the risk of foodborne infection by *S. aureus* associated with the consumption of braised beef in Côte d'Ivoire, to improve food safety in this country. A preliminary survey was first carried out in six (06) separate towns in Côte d'Ivoire, among consumers (n = 900) and sellers (n = 300) of braised beef meat with a view to characterizing the behavior of these actors. A microbiological analysis was then carried out in accordance with the ISO 6888-1/-3 standard (1999) on 189 samples of this braised beef meat taken from various sales outlets in these towns. A risk model was developed. The risk of infection linked to the consumption of this braised beef was estimated using the Monte Carlo simulation procedure. The results of the consumer survey showed that the percentage of the population consuming braised beef meat (BBM) was 74.4%, with an average consumption of 114.3 ± 0.5 g/person/day. Microbiological analysis revealed the isolation of 92% *S. aureus* with a mean load of 6.0 ± 0.19 log10·cfu/g greater than 10^5 CFU/g. The probability of ingesting a dose greater than 10^8 *S. aureus* bacteria ranged from 4.2% to 4.3%. Braised beef meat sold in the streets of Côte d'Ivoire's towns and cities poses a real risk of infection. *S. aureus* is one of the causes. *S. aureus*-associated gastroenteritis is caused by failure to observe simple hygiene rules. The risk of infection should be mitigated by cleaning up the places where this product is sold and promoting good hygiene practices in the informal sector.

Keywords: Braised beef meat; Inadequate handling; *Staphylococcus aureus*; Risk of infection; Simulation of monte carlo

1. Introduction

In all countries of the world, as well as in the Third World, the consumption of traditionally produced meat products has been gaining increasing interest in recent years, due to their potential health benefits such as sensory quality, nutritional value and natural composition (micronutrients and macronutrients) [1] [2] [3]. Moreover, in developing countries, pathogenic microorganisms are a hazard in food. Food poisoning is common in these countries, where the sale of street food is commonplace, constituting a permanent risk [4]. In West Africa, as in Côte d'Ivoire, the most dreaded bacteria frequently identified in cases of foodborne infection are *Staphylococcus, Salmonella, Coliforms* and *Clostridium* [5] [6]. For example, the prevalence of *salmonella* in cooked meat gizzards sold in the Abidjan district is 3.3% [7]. Braised beef meat, known as 'barbecue' in the West and "*choukouya"* in Côte d'Ivoire. This meat-based food product, most of which comes from farmed beef, is a dish made up of different meats cooked in the open-air using embers or wood [8]. This meat is particularly appreciated for its taste, availability and relatively low cost. However, this

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meat is mainly prepared and sold on the streets, in inadequate informal environmental conditions, such as handling practices, sales, storage in the open air and at inappropriate temperatures, and combinations in the absence of strict food safety regulations. This situation exposes them to contamination of all kinds, and the consumer to a very high risk of microbial food poisoning, due to the unsanitary environment, which is conducive to the development of flies and mosquitoes in the water jars [9]. *S. aureus* is one of the most dreaded pathogenic micro-organisms contaminating this product. This bacterium, often identified in many cases of toxic infections reported in developing countries, produces staphylococcal toxins. Ingestion of these toxins can cause serious food-borne infections in consumers [10]. Similarly, *S. aureus* is an indicator of handling hygiene contamination, as the presence of resistant strains of *S. aureus* in food constitutes a major health risk [11]. In addition, meat dishes are highly vulnerable to bacterial contaminants [12] [13] because meat is a substrate for the proliferation of microbial flora when handled or stored in poor hygienic conditions [14] [15]. Moreover, waste water gutters and puddles with a pervasive unpleasant odor can be found near these sales outlets [16], making the environment even more unhealthy. Most studies in Côte d'Ivoire have focused on determining the level of contamination of certain food products and the probability of consuming food contaminated by a pathogen [17] [18]. It has been noted that very little interest has been shown in the biological or chemical risk assessment studies carried out [19] in Côte d'Ivoire. In some countries, quantitative assessments of the microbiological risks associated with various widely consumed food products have been carried out [20]. This situation poses a major challenge for public health, where cases of food-borne infections linked to the consumption of street food are potentially frequent. Braised beef meat sold in the street could be contaminated by toxin-producing *S. aureus* because of its handling and exposure to the open air on the public highway during marketing. This source of microbial contamination poses a health risk to consumers. It is therefore necessary to know the level of risk associated with the infection linked to the presence of *S. aureus* in this braised beef meat dish for the exposed population. This study was initiated to assess the risk of infection associated with the consumption of braised beef meat contaminated with *S. aureus*. This assessment involves obtaining a numerical value for the risk faced by the population [21]. The study will aim to guarantee food safety in Côte d'Ivoire by guiding policies in the implementation of sanitation measures, particularly in areas where the consumption of street food is common.

2. Material and methods

2.1. Study site

This study was conducted from July 2013 to June 2015, in six major cities of Côte d'Ivoire namely Korhogo (North), Daloa (West), Bouaké (North-Central),Yamoussoukro (South-central), Abengourou (East), Abidjan (South and North). The sites were selected purposively, based on the importance of markets for pro- visioning of foodstuff to the populations and the helpfulness of actors (Vendors, consumers) of the bovine sector to participate in the study

2.2. Questionnaire survey

The questionnaire survey was conducted among consumers and vendors of Braised beef meat on the streets in the six (06) selected cities for the study. The questions includedthe quantity consumed, the frequency of consumption of braised beef meat and the sales practices and conditions used. The interest of the survey was to gather information to assess the impact of consumption of braised beef meat onconsumer health. In total, 1200 people comprising nine hundred (900) consumers and three hundred (300) vendors were interviewed.

2.3. Sampling of Braised Beef Meat

Braised beef meat samples were collected from 27 vendors of braised beef meat on all sites. Three campaigns were carried out per site and a total of one hundred eighty-nine (189) samples were collected from the whole sites. For each campaign nine (9) samples of approximately 100 g per sample of braised beef meat were purchased from each vendor of braised beef meat It shouldbe underline that each sample bought and labeled (site, date and time of collection) is placed in a sterile bag "Stomacher". The ideal conditions to ensure the preservation of samples were a quick transport into a cooler containing cold packs, so as keeping the temperature under 5°C. The maximum storage periodof samples from point of sale to the laboratory where they undergo microbiological analyzes was 16 hours.

2.4. Laboratory procedures

Once at the laboratory, 10 g of each sample were homogenized in 90 mL of sterile bacteriological peptone (Oxoid, Hampshire, England). Decimal dilutions (1:10) of all samples were made in buffered peptone water (BioRad) was carried according to the method [22]. Enumeration of *S. aureus* was carried out according to [23]. Potassium tellurite egg yolk was added at 0.1 ml of the appropriate dilution (10-1 to 10-4) to a sterile Petri dish containing 15 ml of solid Baird Parker (Oxoid) agar and incubated at 37°C for 24 hours. The characteristic colonies of presumptive staphylococci were counted for each dish. *S. aureus* give black colonies (reduction of tellurite to telluride) with a clear halo due to proteolysis of egg yolk proteins and a white-opaque border linked to the precipitation of fatty acids resulting from the action of lecithinase (hydrolysis of the lecithin in the egg yolk). Only Petri dishes with a colony count of between 15 and 150 were accepted. The coagulase test was performed to identify and confirm suspected *S. aureus* colonies. The inoculum from each colony with a characterized and well-isolated morphology was transferred to brain heart broth (BCC) and incubated at 37°C for 18-24 hours. A volume of 0.2 mL of each BCC broth culture was transferred to sterile hemolysis tubes containing 0.5 mL of rabbit plasma. The mixture was incubated at 37°C and examined after 1 h and 4 h. The tubes must not be shaken during incubation. The presence of a firm clot which does not move even when the test tube is tilted confirmed a positive result for the presence of a *S. aureus* colony (coagulase + reaction).

The unacceptable microbiological limits for which the braised beef meat samples were considered potentially hazardous in this study were based on the standards for ready-to-eat foods [24].

2.5. Quantitative Risk Assessment for of *Staphylococcus aureus* pathogenic species

The use of reference pathogens is an accepted practice in the field of Quantitative Microbial Risk Assessment [25] [26] to represent the potential adverse health effects of each broader microbial group. Risk assessment of infection linked to the consumption of braised beef meat was carried out according to the *Codex Alimentarius* approach [27]. It is a scientific approach carried out in four steps: hazard identification, hazard characterization, exposure assessment and risk characterization.

2.5.1. Hazard Identification (Pathogen)

The step of hazard identification is a literature search on the hazard that may cause adverse health effects, and which may be present in a food [21] [28]. Braised beef meat, classified as street food, has been linked to a number of serious illnesses [29] [30]. Braised beef meat can be contaminated with bacteria that pose a risk to human health [5] [31]. In this study, *S. aureus* was selected based on its probable presence in beef meat contamination [32] and considered as an indicator of handling hygiene contamination and the more a food is handled, the higher the risk of recontamination. This bacterium is present on human skin and mucous membranes [33]. It can produce preformed enterotoxins in braised beef, which are responsible for its pathogenicity [34]. This meat-based dish is characterized by a relatively high protein content, a factor that encourages rapid germ growth. *S. aureus* is stored at temperatures between 6°C and 48°C with an optimal pH between 6.0 and 7.0. The optimal temperature to produce its enterotoxins is between 34 - 40°C and its basic pH is 7.0 to 8.0 [32]. These parameters factor the multiplication of this germ, particularly when factors such as an unhealthy environment, inappropriate preparation conditions and the type of preparation requiring intensive handling of the food are combined. These toxin-producing bacteria are identified as dangerous because they cause food-borne infections around the world, [35] [36] [37] particularly in Côte d'Ivoire. The main symptoms of the disease are fever, vomiting and diarrhea, but fatalities have been identified in the elderly and young children [38]. In this study, risk describes the probability of infection following consumption of contaminated braised beef meat.

2.5.2. Hazard characterization

The characterization of the hazard allows to assess qualitatively and quantitatively the nature of the adverse health effects associated with the pathogen present in the food. This step provides a description of the severity of the effectsresulting from the ingestion of the hazard in the food. Establishment of the dose-response relationship or the relationship between the level of the hazard exposure and the level of a toxic effect should allow knowing the infective doseor disease [39]. The maximum concentration of the bacteria in food or culinary preparations is 10^6 cfu/g, a concentration above which there is a probability of multiplication of the bacteria and production of enterotoxins preformed in the food. According to the literature, the infectious dose of *S. aureus*, which is the dose-effect, is 10^8 bacteria per gram [32]. Thus, the ingestion of food containing 10^8 or more *S. aureus* bacterial cells can lead to staphylococcal food poisoning, a type of gastroenteritis causing inflammation of the mucosa of the stomach, small intestine and colon.

2.5.3. Exposure Assessment

A stochastic methodology was used to estimate infection from pathogenic microorganisms through ingestion of product [40]. Individuals are assumed to ingest amount of product meat beef.

Diagram of the evaluation model

The step of exposure assessment is the quantitative or qualitative estimation of the likely ingestion of biological or chemical hazards through food aliments [27]. The exposure estimation is based on consumption scenarios of contaminated food (Figure 1). The realization of these scenarios considers various identified parameters identified to the risk to be evaluated. After cooking, which is usually done at high temperatures between 100°C and 120°C. Braised beef meat is exposed ambient temperature (27°C to 32°C). In most cases, it is consumed immediately after cooking or after a period of exposure to the often-unsanitary environment and may be subject to human handling, which can lead to cross-contamination and recontamination. All these reasons allow to suppose that the contamination of the braised meat occurs atthe time of its sale [41].

The exposure assessment consists of estimating the level of danger to which the consumer is exposed at the time of consumption of the food. Thus, the concentration *(C)* of *S. aureus* in the braised meat could be supposed to be equal to its initial concentration (C_i) at the time of its sale. Under these conditions, exposure was assessed by estimating the dose (I) of the toxin-producing bacteria and its toxins ingested by the consumer each time the cooked meat was eaten.

Mathematical model of ingested dose

Stochastic modeling requiring probability distributions was used for the exposure assessment. Thus, a diagram showing all the events that can lead to infection in the consumer has been established. In our study, exposure or Ingested dose (D_i) of *S. aureus* in braised beef meat was estimated using (Figure 1).

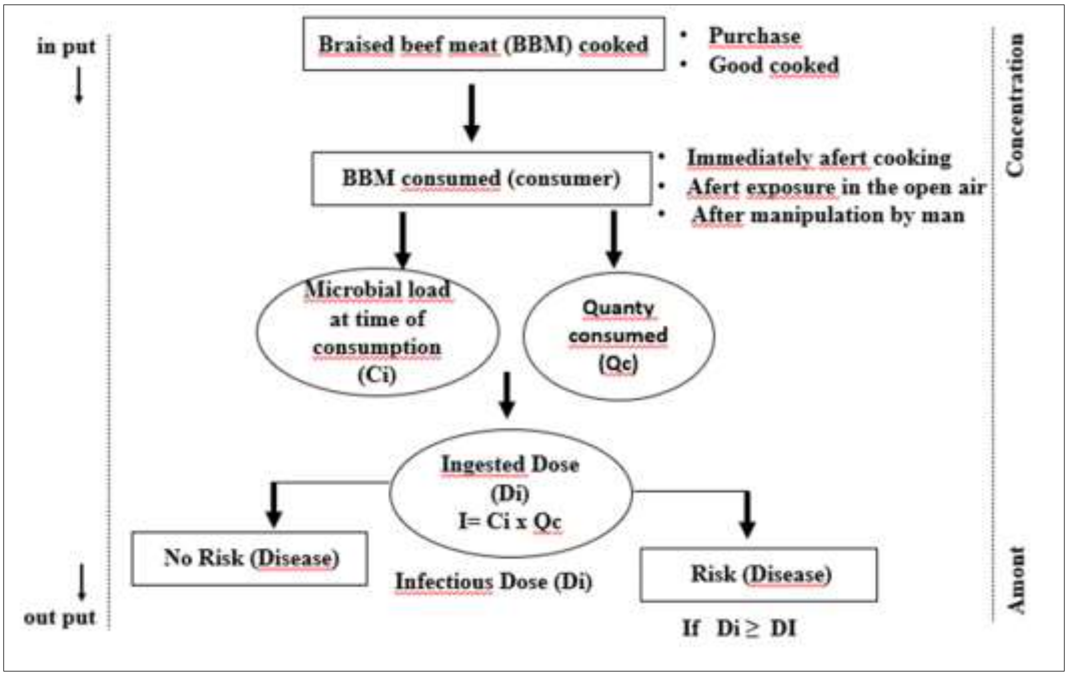

Figure 1 Fault tree on the infection risk with braised beef meat (BBM)

The parameters of pathogen concentration in braised beef meat (C_i), the quantity of braised beef meat consumed (Q_c). The number of the germs ingested (I) by the consumers expressed as Colonies Forming Unit (cfu) was calculated as follow:

I= Ci X Qc

- *I*: Ingested dose of the pathogen in braised beef meat
- C_i: Concentration of the pathogen in braised beef meat in (cfu/g)
- Q_C: Amount consumed from braised beef meat (g/person/intake)

The use of this equation requires data on the concentration of *Staphylococcus. aureus* (expressed in cfu/g) in the braised beef meat sold in the popular streets of the selected cities for the study and survey data consumption of a consumer population. The data collected on these two parameters each gave a distribution reflecting their uncertainties. The uncertainty on the parameters of the model was quantified by the bootstrap method **[40].** Parameter values were estimated based on laboratory and survey results. The distribution of the ingestion of the pathogen by the consumers is obtained after the calculations. Monte Carlo simulation was performed for 1500 iterations using the MATLAB R 2015a software. Following the simulations, cumulative probabilities of realizations or probabilities are obtained.

2.5.4. Risk characterization

The characterization incorporates the results of hazard identification, hazard characterization and exposure assessment. Based on these results, the risk characterization must quantitatively estimate the severity (*i.e.* probability) of adverse effects caused by *S. aureus* on health of consumers. Thus, based on the distribution of contaminant ingestion obtained using the Monte-Carlo method, the value of the infectious dose greater than 108 bacteria per gram is reported for the estimation of the probability or risk of developing an infection per 100,000 exposed inhabitants [42].

2.6. Statistical Data Analysis

The sociological survey data were entered on Excel and analyzed with the Statistica software 7.1. Data collected for the microbiological quality of braised beef meat were analyzed using analysis of variance (ANOVA) with two factors (sampling site and campaign). The difference in Factor is statistically significant at 5% level (two-tailed test) if the probability (or p-value) is less than 0.05. The comparison of means was done using LSD's post hoc test, to determine the source of the significant variations. Concerning the risk assessment, the infection risk associated with the consumption of braised beef meat contaminated with *S. aureus* was carried out by the stochastic method. The Bootstrap method was used to resample the input data of the exposure assessment model (i.e. the data of the concentration of *S. aureus* (C_i) in braised beef meat and the data of consumption from the survey of household). Then the Monte Carlo simulations were performed to obtain thedistribution of the exposure assessment model which is also the ingested dose of *S. aureus*. Fifteen hundred iterations were performed for each simulation creating two thousand cumulative frequency distribution curves for the ingested dose [43]. Bootstrap and Monte Carlo type simulations were performed by the software MATLAB R 2015a.

3. Results

3.1. Risk factors for Contamination of Braised Beef Meat

Braised beef meat consumed at the various sites is packaged. The survey identified the types of packaging and devices used. Most of the braised beef meat is packaged in cement (49.3%), compared with 18.2% in aluminum foil (Table 1). On the one hand, some customers eat this dish on the spot and are often served either on plastic plates (21.3%) exposed to the air, as opposed to steel plates (11.2%). On the other hand, it was observed that the various sales outlets are in poorly maintained environments, with the majority of the product exposed to the open air (82.5%). The other points of sale were protected in display cases (17.5%). Most of the sales assistants (76.7%) did not wear a smock.

Table 1 Contamination factor of braised beef meat sold in the streets of cities in Côte d'Ivoire

braised beef meat (BBM) n = 390		
Type of utensil and packaging used		
Plastic plate	83 (21.3%)	
Steel plates	44 (11.2%)	
Cement packaging	192 (49.3%)	
Aluminum foil	70 (18.2%)	
Point-of-sale devices		
Exposure to the open air	322(82.5%)	
Protection in show case	68(17.5%)	
Wearing the blouse (Body hygiene	Yes	No
Seller	91 (23.3%)	299 (76.7%)

3.2. Average load of *Staphylococcus aureus* in braised beef at the sites

The results of the microbiological analysis of *S. aureus* are shown in (F**igure 2**). *S. aureus* counts vary from one site to another and from one campaign to another**.** The average microbial loads of the bacteria are of 6.0 ± 0.66 log10·cfu/g for Korhogo 6.0 ± 0.58 log10·cfu/g for Abengourou, 5.0 ± 0.27 log10·cfu/g for Daloa, 6.0 ± 0.10 log10·cfu/g for

Yamoussoukro, 6.0 ± 0.79 log10·cfu/g for Bouaké, 7.0 ± 1.47 log10·cfu /g for Abidjan-Sud, 7.0 ± 0.73 log10·cfu/g for Abidjan- Nord. Analysis of the average microbial loads for the three campaigns shows that *S. aureus* is above the unacceptable microbiological limit (M = 10^4 cfu/g) at all the sites studied. The highest average loads were found in Abidjan-Sud and Abidjan-Nord, while the Daloa site recorded the lowest average load. Statistical analysis revealed a significant difference ($p < 0.05$) between the Abidjan-North and Daloa sites. No significant difference was observed between the Abidjan-Sud, Korhogo, Abengourou, Yamoussoukro and Bouaké sites, which had statistically similar microbial loads.

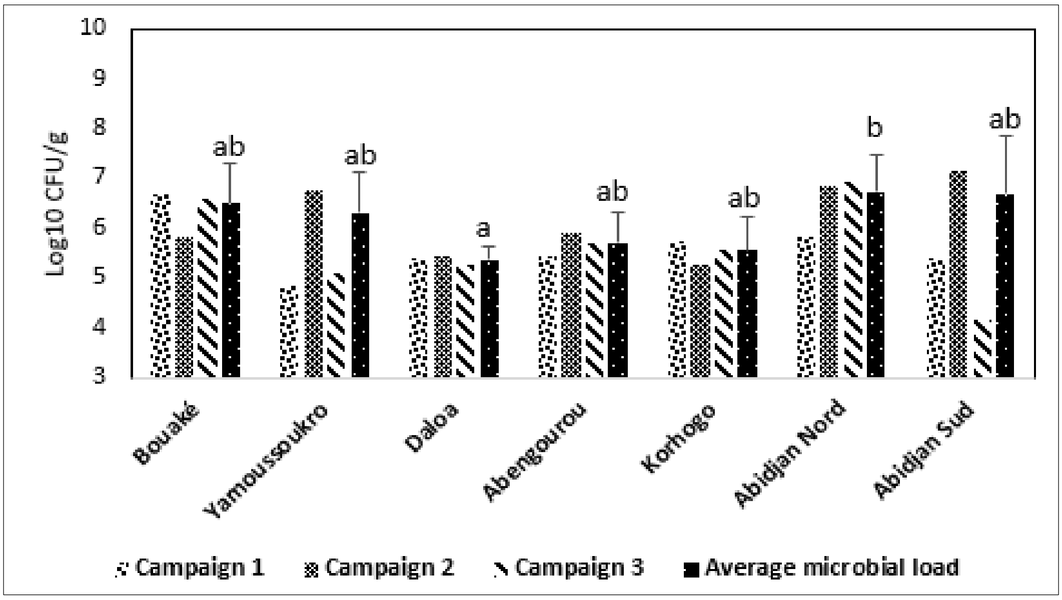

Figure 2 Average load of *Staphylococcus aureus* according to the origin of the braised beef meat. (Different letters in the same figure indicate significant differences ($p < 0.05$) between the average microbial loads of the sites. The same letters indicate similar averages)

Table 2 Tests of significance giving the probability (p-value) to accept means equality

Sources of variation	*Staphylococcus aureus*
Sites	0.416
Campaigns	0.036
Sites * Campaigns	0.215

Table 2 shows the p-values associated with the mean bacteria loads for each source of variation considered. At the sampling sites, for braised beef, the p-value was less than 0.05. There was therefore a significant difference between the average microbial loads of *S.aureus* from one sampling site to another. No significant difference ($p > 0.05$) in the average load of *S. aureus* was observed between the different campaigns. Similarly, in terms of interactions, the p-value analysis shows that there is no interaction effect between the sampling site and *S. aureus* enumeration campaigns. *S. aureus* microbial load results observed at the sites therefore do not depend on the campaign period.

3.3. Beef meat consumption patterns

The survey revealed an average of 114.3 g of beef meat consumed per person per meal (Table 3). The most popular way of eating beef meat was braised (74.3%), as opposed to cooked in a sauce (25.6%). Among consumers of braised beef meat, 12.8% eat it more than three times a week. The largest proportion of consumers (47.5%) were young people (aged 20-30). However, braised beef meat is consumed by all age groups.

Table 3 Consumption patterns of braised beef meat

Variable	Characteristic	Frequency (%)
Consumption mode	Meat cooked in sauce	217 (25.6)
	Meat cooked in braised	630 (74.3)
Frequency of consumption of braised meat	Occasionally (one time/month)	265 (42)
	Every 2 weeks	146 (23.1)
	Weekly	142 (22.5)
	2 or 3 to 4 times/week	81 (12.8)
People who consume purchased meat	youngest (>20 years)	41 (6.5)
	Young people (20 - 30 years)	299 (47.5)
	Adults (30 - 40 years)	271 (43)
	Elderly (50 years and over)	21 (3.3)
Average quantity of braised meat consumed per person		
per part (g)	114.3 g/person/intake	

3.4. Risk of infection Linked to the Consumption of Braised beef meat Sold on Public Roads According to the Stochastic Method.

3.4.1. Proportion of Staphylococcus aureus

S. aureus was isolated from braised beef meat. The samples contained a *S. aureus* contamination rate of 92% with mean loads of 6.0 ± 0.19 log10·cfu/g (**Table 4**). These contamination loads are above 10^5 cfu/g, the critical threshold for ready-to-eat foods.

Table 4 Proportion of braised beef meat contaminated with *Staphylococcus aureus*

	Staphylococcus aureus
Number and percentage (%) of contaminated samples	174/189
Contamination rate	92 %
Average load (log10·cfu/g)	6.0 ± 0.19

3.4.2. Levels of contamination of braised beef meat by Staphylococcus aureus

The results of the distribution of the different concentration levels of *S. aureus* in the braised beef meat samples are shown in Figure 3. In general, most of these values exceed the critical limit for the pathogen *S. aureus*, which is 10^5 CFU/g in a ready-to-eat food. Analysis of Figure 2 shows that 73.5% of the 189 samples had contamination levels above 10^5 cfu/g, which is the maximum concentration in ready-to-eat foods. After analysis, approximately 74 portions of braised beef meat contained concentrations of *S. aureus* that could lead to the multiplication and production of preformed enterotoxins in the food.

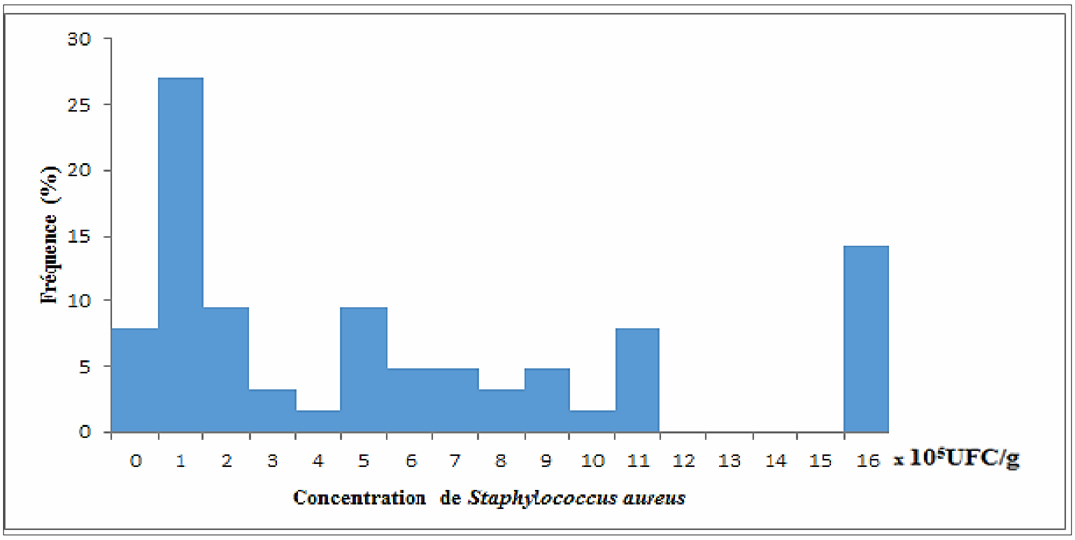

Figure 3 Distribution of *S. aureus* contamination levels in braised beef meat samples

3.4.3. Microbiological quality of braised beef meat

The microbiological quality of the braised beef meat was determined on the basis of the average *S. aureus* load in accordance with European standards for the interpretation of analytical results in food microbiology **[44].** With regard to the results presented in the table, 4.8% are of satisfactory microbiological quality with no risk of toxic infection for the consumer, while 27% have loads that exceed acceptable limits. Of these, 27% of the samples were of unsatisfactory microbiological quality with no risk to the consumer, 47.6% were of unsatisfactory microbiological quality with a risk of toxi-infection, and 20.6% were of unsatisfactory microbiological quality with a high risk to health by having a *S. aureus* microbial load of more than 10^5 CFU/g. Analysis of the average microbial load for the three campaigns shows that *S. aureus* is above the unacceptable microbiological limit (M = 10^4 cfu/g) at all the sites studied.

Table 5 Microbiological quality of braised beef meat

Microbiological quality (BBM)	Number of samples (n = 189)	Percentage (%)
Satisfactory	9	4.8
Unsatisfactory	51	27
Unsatisfactory with risk of toxi-infection (Poor product)	90	47.6
Unsatisfactory with high risk of toxic infection (corompound)	39	20.6

3.4.4. Gastroenteritis symptoms observed in consumers of braised beef meat

Table 6 shows the percentage of consumers who had gastroenteritis symptoms that could be linked to the consumption of braised beef 'street food'. Potential previous symptoms associated with food poisoning related to the consumption of this delicacy were reported by 25.7% of consumers of braised beef as 'street food'.

The three main symptoms reported were diarrhea, vomiting and fever. In this case of consumption, diarrhea was the symptom most frequently mentioned, with prevalence rates of 46.4% for BBM as a 'street food'.

Table 6 Symptoms of gastroenteritis developed after eating braised beef meat defined as 'street food'.

Parameters	**Number of consumers**	**Prevalence**
	n= 630	**(%)**
No ill	468	74.3
Ill	162	25.7
Symptoms reported	*n= 162*	
Diarrhea	124	76.5
Fever	21	12.9
Vomiting	10	6.3
Fever + Vomiting	5	3.1
Diarrhea + Vomiting + Fever	2	1.2

3.4.5. Consumption of serving of Braised beef meat

Nine hundred (900) individuals were interviewed during the consumer survey. The results of the evolution of the distribution of the consumption of braised beef meat by the population of the different sites were treated and then expressed in frequency (%) for the different classes of serving during consumption per intake (Figure 4). The minimum serving size ranges from 50 to 100 g. The maximum serving weight ranges from 1000 g to 1050 g. Thus, the size of the most consumed portion which is the mode is between 100 and 150 grams per personper intake and corresponding to a frequency of 20.8%.

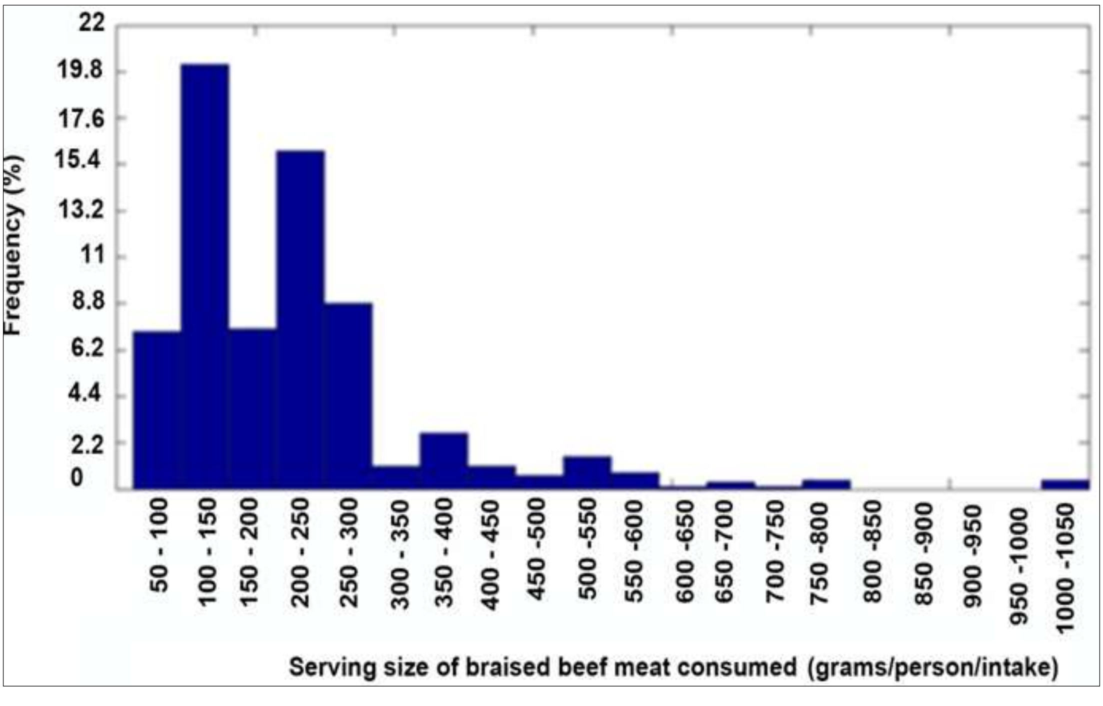

Figure 4 Distribution of consumption of braised beef meat

3.4.6. Exposure Assessment

The result of the Monte Carlo simulation was the distribution of the exposure assessment model represented by a cumulative frequency distribution curve for ingested doses. One thousand five hundred (1500) simulations were carried out, representing one thousand five hundred (1500) ingestion dose curves (Figure 5). This figure gives the probability Y, within an uncertainty interval, with an ingested dose of *S. aureus* (expressed in cfu/g) below a limit value X. The infectious dose of *S. aureus* that can cause food poisoning by ingesting a quantity of braised beef is estimated to be more than 10^8 cfu/g. Assuming that the infectious dose is 10^8 bacteria, the probability of developing food poisoning due to *S. aureus* by consuming a portion of braised beef meat sold in the streets of Côte d'Ivoire cities is at least 4.2% and at most 4.3%.

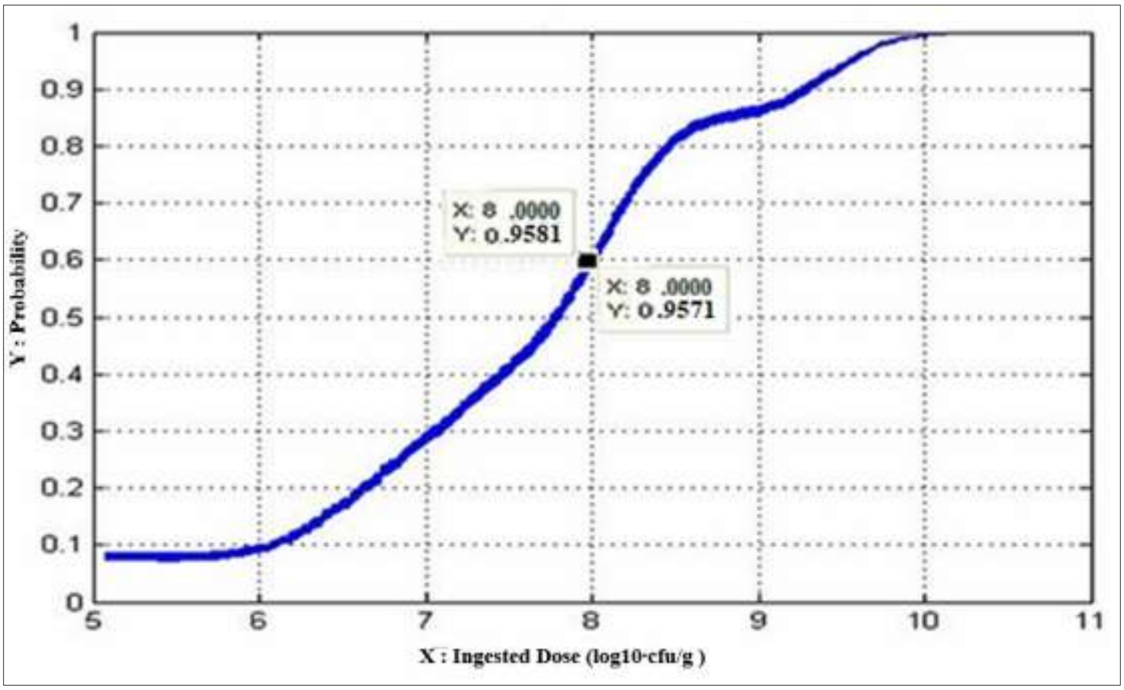

Figure 5 Cumulative probability of ingested doses of *Staphylococcus aureus*

3.4.7. Risk characterization

The infectious dose of *S. aureus* causing foodborne problems by eating one portion of braised beef per day is greater than 10^8 bacteria/g. Thus, on the basis of the exposure assessment model, the probability of injecting a dose greater than 10^8 bacteria could be between 42% and 43%. The risk of developing food poisoning due to *S. aureus* when consuming a portion of braised beef sold on public highways is a minimum of 4.2% and a maximum of 4.3%. Based on this risk, we can deduce between 4,200 and 4,300 cases of food poisoning due to *S. aureus* per 100,000 inhabitants consuming contaminated braised beef meat. This risk estimate represents a quantified estimate of the probability and severity of adverse reactions following the consumption of braised beef meat.

4. Discussion

The risks associated with the consumption of braised beef meat sold in the streets of Côte d'Ivoire were assessed by analyzing the contamination of braised beef meat by *S. aureus* and the behavior of food consumers. Several factors relating to packaging, utensils used, the environment at the point of sale and personal safety practices can influence the safety of braised beef meat sold on the streets of towns in Côte d'Ivoire. Unsuitable packaging, such as cement packaging, is used by 49.3% of vendors. This type of packaging encourages recontamination of the product by germs such as *S. aureus* after cooking. Most braised beef sellers (76.7%) do not comply with the gowning rule. This means that cross-contamination between packaging, utensils, the vendor's hands and the product is inevitable [45]. The practices of sales staff at the point of sale are the source of contamination of the product by certain bacteria. A study carried out on an episode of toxi-infection that occurred in Florida in September 1997 demonstrated through an investigation that a contaminated slicer was the source of the transmission of the *S. aureus* germ to the food [46]. These bacteria are capable of growing and spoiling the product [47] [48]. However, the hygiene provisions observed in this study and the methods used seem to be conducive to contamination due to the direct contact of the sellers with the product [49]. The level of *S. aureus* contamination in braised beef sold on the streets of Côte d'Ivoire varied according to the towns selected for the study. This variability reflects poor practices in these areas and could show that the sampling sites have different unsanitary environments that favors contamination. All the sampling sites showed average loads of *S. aureus* above the unacceptable microbiological limits: 10^4 cfu/g. In addition, microbiological analyses revealed the presence of microorganisms such as *S. aureus* in beef skewers [50]. In the work of [51], a number of lettuce sales outlets located near sources of contamination such as rubbish bins and public toilets were identified in various communes of Abidjan.

These unhealthy environments near sales outlets have an impact on the health quality of products sold nearby [52], leading to high levels of contamination. This contamination is strongly correlated with the lack of personal hygiene among vendors and the unsanitary environment of sales and consumption sites. The same observation was made in Dakar, Senegal, during a similar study of beef [53]. The natural habitat of *S. aureus* is the mucous membranes of animal species and humans [33]. However, these are germs which, at a certain level (10^6 CFU/g) in food, are pathogenic and can multiply, posing a risk to public health. This species is used as a personal hygiene indicator and is widely known to produce enterotoxins that are harmful to the body [54].

As a result, contamination and proliferation of this bacterium may be unavoidable because of handling. Packaging and reheating are ways of preserving the microbiological quality of food after cooking. However, they are also risk factors for contamination. Reliable food preservation temperatures are difficult to maintain. The growth temperature of *S. aureus* and its toxins can be as high as 48°C [32]. At this temperature, cuts in the still-warm food in the container or packaging can encourage contamination. In these ideal conditions, the *S. aureus* germ develops on all food surfaces [46]. These strong growths of *S. aureus* lead to the production of enterotoxins. *S. aureus* was present in 73.5% of the samples analyzed. These results are very high compared with the work of [55] on samples of beef meat, chicken, salad and sauce collected from street vendors in South Africa. In their studies, these authors found 3% *S. aureus* in samples of beef, chicken, salad and sauce. Braised beef meat, identified as street food, is subject to a great deal of handling during preparation and sale. This food is not effectively protected, which encourages recontamination. Wastewater and rubbish are dumped in the streets, attracting flies that could harbor food-borne pathogens [56] [57].

The preliminary survey revealed that 74.3% of people prefer to eat beef meat in braised form. In this form, beef meat is eaten three to four times a week by 12.8% of people, but much more so by young people (47.5%) followed by adults (43%). In fact, this dish is highly appreciated by the Ivorian population, with an average consumption of 114.3 ± 0.5 g per person per serving. These works are aligned with those of [50], which showed that the number of male consumers was equal to that of female consumers (50%). This considerable appreciation of braised beef meat by the population is justified by the fact that food obtained in the traditional way has excellent potential health characteristics such as sensory, natural and nutritional quality [1] [2]. The interest in this food is also justified by its availability as a source of nutritious, inexpensive meals [58]. It is widely consumed in many entertainment activities, such as outings for relaxation, birthday and wedding ceremonies, in refreshment bars and other places of relaxation in the country's main cities. These activities are supported by the strong demographic growth observed in these towns [59]. Note that the risk of hand-carried contamination is very high. According to [51], a single handful of portions contaminated with this bacterium can act as a reservoir for contaminating an entire family. What's more, the germs transmitted from one person to another can circulate as far as the sixth person. Epidemics associated with meat-based meals contaminated with *S. aureus* have already been reported [60] [61].

The epidemiological nature of *S. aureus* at high doses in food could be linked to the virulence mechanism expressed by the excretion of a heat-labile toxin preformed in the food (*S. aureus*). It is important to know that *S. aureus* causes emetic poisoning with symptoms of nausea, vomiting and fever [60]. In view of the above, a quantitative assessment of the microbiological risks was carried out in accordance with the approach of the *Codex Alimentarius* method [62]. Thus, *S. aureus* was chosen as a hazard because of its ability to produce staphylococcal enterotoxins and its virulence. The quantitative risk assessment carried out in this study was based on a probabilistic approach to assessing exposure, which was also the ingested dose of *S. aureus* from the consumption of braised beef meat. This probabilistic method requires probability distributions to represent the variability or uncertainty of the parameters and leads to a probability distribution of the risk and refines the interpretation of the model results [21] [28]. Using Monte-Carlo simulations, it is possible to carry out probability distributions of variables when the model for each variable is determined [39] [63]. The evaluation model was defined on the basis of real data collected on 2 parameters: the distribution of *S. aureus* concentrations (Ci) and the distribution of the portion size (QC) of braised beef meat consumed at each meal per day and per individual. This method was described by [41] in their study on risk assessment of attieke consumption. *S. aureus* concentration levels ranged from 10^2 cfu/g to 10^8 cfu/g with 75.8% of braised beef meat portions above 10^5 cfu/g which is the maximum concentration of *S. aureus* in ready-to-eat foods. At high levels in food, *S. aureus* produces staphylococcal enterotoxins. Thus, ingested doses of *S. aureus* can be considerable, with an average of 10^3 to 10^5 CFU. However, this dose can increase to 10^6 or 10^8 CFU in the event of infection [64]. Although this germ can be easily destroyed at high temperatures, their toxins are able to resist cooking because they are thermostable. Given that the level of *S. aureus* in the suspected food is greater than 10^5 colony-forming units (cfu)/g [65], the diagnosis of staphylococcal food poisoning could be confirmed, and the consumer could therefore be exposed to a food-borne infection [33]. Thus, for 10^8 bacteria ingested, the individual risk of food poisoning due to *S. aureus* for the consumption of portions of braised beef meat per intake is obtained. After Monte Carlo simulations, the results of the assessment of the risk of infection associated with the consumption of braised beef meat contaminated with *S. aureus* revealed that the risk varies between 4.2% and 4.3%, or 4,200 to 4,300 cases of *S. aureus* disease for a total of 100,000 consumers.

These results are higher than those obtained by [41] in attieke sold on the open market. This risk appears relatively real and high compared to the risk assessment for certain germs present in other food matrices in developed countries. Several cases of disease have been linked to *S. aureus* [66] [61] [16]. According to data extracted from reports by the European Food Safety Authority (EFSA), 164 outbreaks of toxi-infections due to staphylococcal enterotoxins were responsible for 1692 cases of illness, 21.5% of which required hospitalization, with meat products being the most incriminating [67]. In their study of a *S. aureus* family food poisoning outbreak, [68] demonstrated the presence of toxin by radioimmunology in meat containing 7.5 x 10^9 CFU/g of *S. aureus*. Contrary to the works of [69], where the microbiological risk assessment mentioned *Escherichia Coli* doses lower than the infectious dose of 10^8 CFU in drinking water. In one case of food poisoning, a strain of *Staphylococcus aureus* was implicated in the United States [70]. In some cases, there are more than 26,000 cases of the disease per 100,000. The risk posed by eating braised beef sold on the streets of Côte d'Ivoire is high. This high probability of developing a disease can be explained by the high contamination of samples, which exceeds the acceptable limits set by quality standards, because in sufficient quantities in the food, staphylococcal enterotoxins can cause symptoms such as nausea followed by incoercible vomiting, diarrhea and abdominal pain [32]. The high bacterial load of braised beef sold on the streets, the quantity of braised beef consumed, and the frequency of consumption are potential risk factors [18]. The different processes involved in the scenarios could explain this phenomenon. The cooking stage eliminates or reduces the risk of proliferation of micro-organisms, hence the probable presence of pathogenic germs in the food. The food only becomes toxic if conditions favorable to significant bacterial multiplication and toxinogenesis are present. Once produced in the food matrix, enterotoxins stabilize as a function of environmental conditions such as temperature and pH. Compliance with the time/temperature pairing is sufficient to destroy most microorganisms [71]. However, in our case, this observation is justified by the high-risk preparation conditions and practices applied and the ‘eating behavior’ of consumers. It should be borne in mind that while microbial contamination of ready-to-eat foods may be reduced or controlled in the home environment, it is less so in foods prepared away from home, which are channels for the transmission of food-borne infectious diseases [51]. Diarrhea was the symptom most frequently mentioned when consuming braised beef meat (76.5%). This result is higher than that of studies by [9] which showed that diarrhea predominated with a frequency of 16% compared with 10% for vomiting and 2% for fever. Studies carried out on cooked beef sold on the streets of Abidjan in Côte d'Ivoire showed that cooked kebabs were more likely to cause diarrhea in 55.25% of cases. The works of [50], also recorded diarrhea (18%), abdominal pain (11%) and vomiting (6%) among consumers of beef skewers sold in the streets of the town of Adjame in Côte d’Ivoire. It should be noted that in Côte d'Ivoire, consumers rarely report their illness to health centers, or they find it difficult to make the link between their illness and the food they have eaten. However, bacterial infections are common in developing countries [72]. According to [63] food poisoning caused by *S. aureus* is like the syndromes associated with diarrheal and emetic disorders leading to food-borne illnesses like food-borne infections caused by *S. aureus.* The toxinogenic nature of this pathogen means that the food studied under the conditions described in this study could be incriminated. Consequently, the consumption of braised beef meat as a street food could lead to health consequences for consumers. Furthermore, studies by [73] have shown that there is a relationship between the food consumed and the occurrence of diarrhea. These studies showed a diarrhea stool rate of 12.7% was observed in patients depending on starch-protein associations. [74] showed cases of salmonellosis are associated with beef consumption in a report of *Salmonella* infection positivity rate of [75]. The association between braised beef considered a street food and foodborne infections has been reported by several authors [29] [30] [76] [77] [78]. Those involved in the informal sector need to be aware of the threat posed by poorly processed meat dishes and non-compliance with good hygiene practices at the point of sale. For this reason, inspections by the public health authorities must be carried out to improve sales conditions and the quality of products from this sector.

5. Conclusion

The study highlighted the poor hygienic quality of braised beef meat sold in the popular streets of several towns in Côte d'Ivoire. The average microbial load of *S. aureus* in braised beef differed significantly from one site to another. This dish, which is highly prized by the Ivorian population, represents a health risk for consumers due to the high concentration of *S. aureus* found in the samples analyzed. The probability of ingesting a dose greater than 10^8 CFU is between 4.2% and 4.3% for *S. aureus*. However, the risk of *S. aureus* infection is real and significant for consumer health in Côte d'Ivoire. The study assessing the risk of *S. aureus* infection linked to the consumption of braised beef thus appears real and high. This risk could result in between 4,200 and 4,300 cases of illness per 100,000 inhabitants consuming braised beef meat, defined as ‘street food’. These results should draw the authorities' attention to the need to raise awareness of good hygiene and product manufacturing practices.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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