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Bacteriological Profile of Contamination of Artisanal Peanut Pastes Sold in the City of Bangui in the Central African Republic

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Abstract

Peanut pastes are the most popular ingredients in Central African cuisine. The processing, preservation, transportation and sales locations can promote the development of bacteria. The objective of this study was to count the germs that are indicators of hygiene and fecal contamination, isolate and identify them in peanut pastes. This is a cross-sectional study with an analytical aim. A total of 320 samples of peanut pastes sold in the city of Bangui were collected by random methods and analyzed in the food microbiology unit of the National Laboratory of Clinical Biology and Public Health. The indicator germs of hygiene and fecal contamination were counted and the other enterobacteria were isolated and identified. The enumeration of mesophilic flora was carried out according to the references and the standard in force NF V 08-051, total coliforms by the standards NF V 08-050, 2010 and NF V 08-060, 2010, fecal coliforms by the standard NF ISO 16649-2, 2001 and enterococci by the horizontal enumeration method according to the standard NFT 90-416. The data were collected on the ODK collection software. The Epi info version 6 software was used for data analysis and processing. The results obtained from this study showed that 96.3% of the peanut pastes were marketed by women. Among the sellers, 6.3% were illiterate; 65.6% had dropped out of primary school; 28.2% had dropped out of high school. The search for critical points using the HACCP method revealed that 3% of the sellers had knowledge of food hygiene, 27% could store peanut pastes well and 1% had an idea of the contamination of peanut pastes by germs. No seller had any knowledge of toxiinfection and food poisoning. The personal hygiene of the vendors showed that 14% had dirty clothes, and 11.88% had lesions on their skin. Note that 61.25% of peanut legs were preserved by humidity, and 1.88% by exposure to the sun.

The microbial load of germs indicating hygiene and fecal contamination varied from 56×10^6 CFU/g to 219×10^6 CFU/g for mesophilic aerobic germs; 69×10^6 CFU/g to 71×10^6 CFU/g for total coliforms; 43×10^6 CFU/g to 88×10^6 CFU/g for fecal coliforms and enterococci from 3.65×10^6 CFU/g to 111.8×10^6 CFU/g depending on the place of sale (market). The overall prevalence of contamination was 54% by enterobacteria. Strains *of Klebsiella pneumonae* were 3.4%, *Citrobactere koserii* 4.6%, those of *Enterobacter sakazakii*, *Flaviomonas horzitiabita*, *Klebsiella oxytoca* and *Pseudomonas aeruginosa* were in the order of 5.7%. Strains of Proteus mirabilis, *Enterobacter cloacae* and *Klebsiella spp* were in the order of 10.3% and those of *Escherichia coli* were 29.9%. The results of this study revealed the presence of hygiene indicator germs, fecal contamination that could be the cause of food-borne infectious diseases. The unsanitary conditions of sales locations and the lack of knowledge of the hygiene rules applicable to foodstuffs are a risk to consumer health.

Keywords

Sale, Peanut Paste, Contamination, Bacteria, Bangui

1. Introduction

Peanut is a plant of the legume family in which the central stem produces peanut flowers [1] [2]. The fruit is a pod containing one to three seeds that develop in the branches and are pushed underground [3] [4]. Peanut, were introduced during the colonial era in Africa, during the cotton crises as one of the ways to ensure additional income to that of cotton. This has been a source of income to boost the economy of African countries located in savannah areas [5]. Peanut has a high nutritional value due to the presence of proteins, fatty acids, carbohydrates, fiber, vitamins, calcium and phosphorus [4]. Studies have highlighted the medical interest of consuming peanuts to reduce the risk of cardiovascular diseases and type 2 diabetes [5]. In the savannah areas of northern Cameroon, southern Chad and the Central African Republic, peanuts have emerged as one of the ways for rural people as well as for traders and new deflators from public administrations and private organizations. Peanuts have emerged as a truly regional product, very famous in African cuisine for their multiple uses as condiments [6]. Traditional techniques for processing peanuts into paste or butter, preservation, storage and transportation are still used. These practices are optimal conditions for the development of pathogenic microorganisms, which constitutes a serious problem for human health [1] [7]. This could lead to the growth of bacteria such as *Escherichia coli, Proteus sp,* and *Salmonella sp* in peanut pastes as signs of microbial proliferation presenting a potential risk related to the pathogenicity of these germs on human health [8]. Apart from contamination by toxins, in particular aflatoxins resulting from the growth of fungi in their seeds during storage, handling and transport, the bacterial risk remains higher [9]. In the Central African Republic as in other countries in the sub-region, peanuts have often been considered mainly

as a secondary crop grown around huts. Its cultivation was only practiced by women and children. The share of area reserved for this crop is increasing significantly and is respectively 17% for an average farm in the CAR, *i.e.* an area of 100,000 ha [10]. Peanut seeds are used to make oils often used for preparing food in kitchens. Peanut seeds are also eaten raw or roasted [11]. Transformed into pastes, it is one of the most popular ingredients in Central African cuisine, either consumed directly in the form of "GBAKPA" butter. Thus transformed, artisanal processing, means of conservation, transport and places of sale can promote the development of bacteria and mold [1] [12]. Peanut pastes from artisanal processing are sold throughout the territory of the Central African Republic (CAR) in precarious hygiene conditions and at the risk of proliferation of various microorganisms that could raise the problem of poisoning and toxiinfection among consumers. Because no study has been carried out in this area to my knowledge in the CAR, it is in this context that this study was conducted, the objective of which was to determine the profile of germs contaminating peanut pastes for human consumption sold in the city of Bangui.

2. Material and Methods

2.1. Study Framework

The study was conducted at the National Laboratory of Clinical Biology and Public Health in the food microbiology unit.

2.2. Type and Duration of Study

This is a cross-sectional study with an analytical aim lasting six months conducted in the main markets of the city of Bangui.

2.3. Biological Material

The biological material consisted of peanut paste which is used as a condiment in the preparation of sauces popular with the Central African population.

Table 1 gives the nutritional composition for 100 g of peanut seed.

Table 1. Composition of 100 g of peanut seed (FAO 2017).

Constituents	Raw with skin (%)	Raw without skin (%)
Water	5.66	5.4
Proteins	26	26.3
Fats	47.5	48.4
Carbohydrates	18.6	17.6
Fibers	2.4	1.9
Ashes	2.3	2.3
Minerals	1.15	1.15
Others	0.5	0.5
Energy (J)	2.361	2.378

3. Method

3.1. Sampling

A total of 320 samples of peanut paste sold in the eight (8) main markets of the districts of the city of Bangui were collected for analysis. The samples were taken by random method from the sellers during the period from October to November 2023 under aseptic conditions.

3.2. Field Survey

Interview-type questionnaires were sent to the sellers in order to collect information on the production technology, conservation and the assessment of the level of knowledge of the sellers on the concepts of food safety. These data were recorded directly by the ODK collection application v2023.3.1.

3.3. Bacteriological Analysis

The bacteriological analyses were based on conventional food analysis methods and the standard (NF EN ISO 4833, 2006) [13]. 25 g of each weighed sample was suspended in 225 mL of buffered peptone water to give the mother solution which would be revived for 24 hours in an incubator at 37˚C. Decimal dilutions were made from the mother solution, *i.e.* 1 mL of the mother solution was added to 9 mL of peptone water. These dilution series were carried out in 6 tubes. The count of germs was carried out according to the technique of counting on agar medium by making a inoculation by incorporation according to the standards. The inoculated media were placed on the bench for solidification, then incubated in an incubator at 37 and 44˚C for 24 hours. The mesophilic flora was counted according to the standards in force on the Plat Count Agar (PCA) medium compared to the NF V 08-051 standard [1] [14]-[18]. Total Coliforms were counted according to the standards (NF V 08-050, 2010 and NF V 08-060, 2010), fecal coliforms (FC) by the NF ISO 16649-2, 2001 standard [19].

The enterococci were counted by the horizontal counting method according to the NFT 90-416 standard [20]-[22]. The search for other bacteria of medical interest was done by the conventional bacteriology technique. The mother solution previously revived for 24 hours in the incubator was inoculated in the Salmonella and Shigella, Mac cockney, Hektoen, Bile esculin agar media and incubated in the incubator for 24 hours at 37˚C [23] [24]. The viable colonies on the surface of the agar were collected and identified in the identification medium, the Api20E gallery. A single pure colony from 24 hours old was collected, added into 5 ml of sterile distilled water for the preparation of the bacterial suspension. The preparation of the gallery was done by distributing a little water in the cells at the bottom to create a humid atmosphere and place it neatly in the box. Using a sterile Pasteur pipette, the bacterial suspension was taken and the tip of the pipette was placed on one side of the well allowing the suspension to flow gently. For the framed characters (CIT, VP, GEL), the cups were filled completely and the underlined characters (ADH, LDC, ODC, H2S, URE) were filled just at the level of the me-

niscus leaving the orifices of the cup which were filled with sterile vaseline oil. The box was closed and placed in the oven at 37˚C for 18 to 24 hours. After 24 hours of incubation, the gallery was read after checking the purity of the strain and the reactions. The interpretation was done using the Api Version system [1] [25]. These manipulations were carried out aseptically under a laminar flow hood.

3.4. Statistical Analysis of Data

The data were collected on the ODK collection software. The Epi info version 6 software was used for data analysis and processing. Descriptive statistical tests were used to calculate central tendencies. With $p < 0.05$ in bilateral, the test was considered statistically significant with a confidence interval of 95%. Some results were expressed in frequency, in percentage and the SPS software was used for some trend analyses.

4. Results

4.1. Manufacturing Flow Chart

The production of peanut paste involves different steps such as shelling, roasting, whitening of seeds and grinding. The red ochre skin was removed before grinding, possibly by hand.

Figure 1 shows the peanut paste manufacturing flow chart.

Figure 1. Peanut paste manufacturing diagram.

4.2. Socio-Demographic Characteristics of the Vendors

The age of the vendors ranged from 19 to 52 years with an average of 22 years. The sex ratio male/female was 0.04.

Figure 2 shows the distribution by level of education of the vendors.

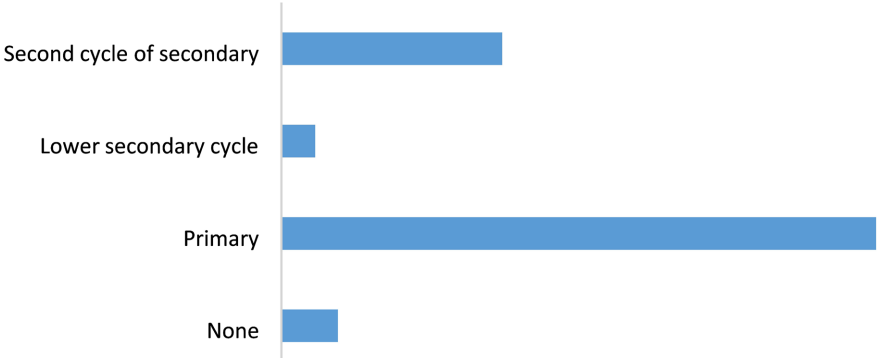

Figure 2. Distribution by level of education of sellers.

The illiteracy rate among sellers is around 6.3%.

4.3. Risk Assessment According to the HACCP Method

Figure 3 shows the assessment of sellers' knowledge of food poisoning and toxi-infection from production to marketing.

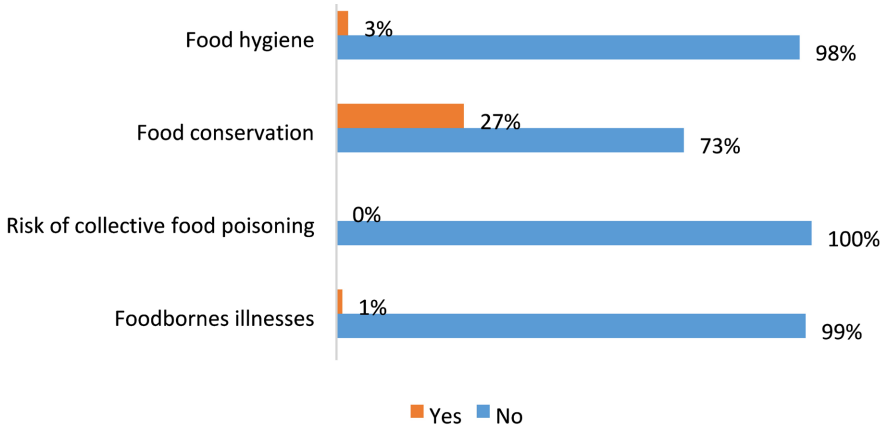

Figure 3. Level of knowledge of sellers and vendors on Hygiene, Conservation and food poisoning.

99% to 100% of sellers have no knowledge about food poisoning and foodborne illnesses, 73% have no knowledge about food preservation technology.

Table 2 presents the personal hygiene of peanut paste sellers.

Table 2. Evaluation of the peanut paste sales environment by sellers.

State of play	No	Yes
Own	26.25%	73.75%
Presence of flies	17%	83%
Presence of puddles	62%	38%
Dusty space	47.5%	52.5%

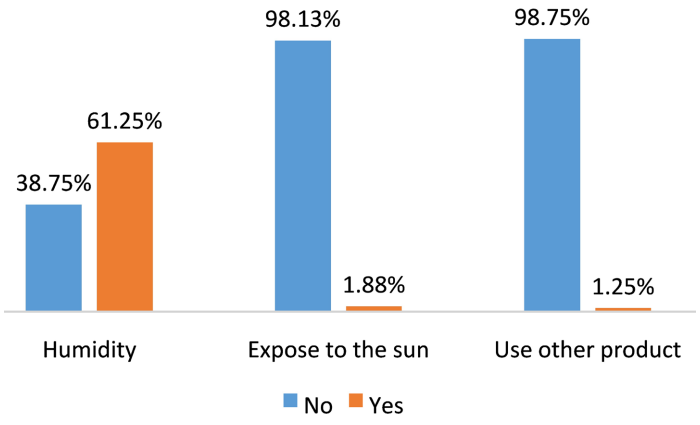

Figure 4. Different methods of preserving peanut pastes.

26.25% of the vendors had dirty clothes and 52.5% sold peanut paste in dusty spaces.

Methods of preservation and storage of peanut pastes.

Figure 4 presents the different techniques of preservation of peanut pastes.

61.25% of the sellers preserved their peanut pastes in humid places and 1.25% used additives.

Figure 5 presents the hygienic characteristics of the sellers.

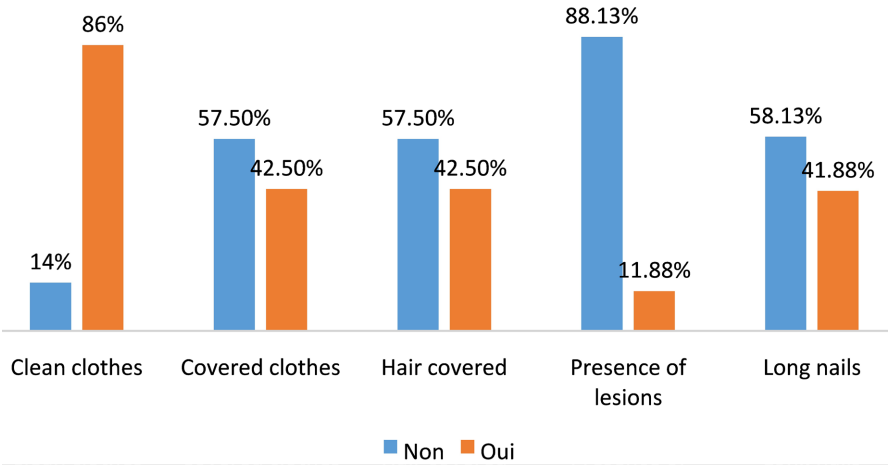

Figure 5. Assessment of the property status of sellers.

According to Figure 5, 41.8% of sellers had long nails, 14% had dirty clothes and 11.8% had injuries to the upper limbs.

Table 3 presents the microbial load by market and by type of microorganisms found in peanut pastes.

Table 3. Enumeration of germs indicating hygiene and fecal contamination.

Marcket	Mesophiles Flora $\times 10^6$ CFU/g	Coliforme Fécaux $\times 10^6$ CFU/g	Total Coliform $\times 10^6$ CFU/g	Enterococci $\times 10^6$ CFU/g	Sulfite-reducing anaerobic $\times 10^6$ CFU/g
Boy-rabe	219	65.5	69	24.4	57.55
Centrale	15.25	51.45	71.1	72	39.25
Combattant	78	88	67.5	92	56
Mamadou Mbaïki	77.4	60.4	71	3.65	26.7
Miskine	148.65	76.65	71.2	82.9	59.8
Ouango	81.2	43.8	42	60.2	56.5
Pétévo	160.4	71.2	43.25	111.85	35.5
Sango	56.5	62.3	71.15	49.9	70.8

The number of mesophilic aerobic germs in peanut pastes was very high at the Boy-rabe market, total coliforms at the Miskine market and fecal coliforms at the Centrale market, Miskine market and Sango market. Enterococci were much more

observed in peanut pastes at the Petevo market and Sulfito-reducing anaerobes and enterococci contaminated peanut pastes at the Sango and Ouango markets much more.

Figure 6 shows us the different colonies isolated and counted in peanut pastes.

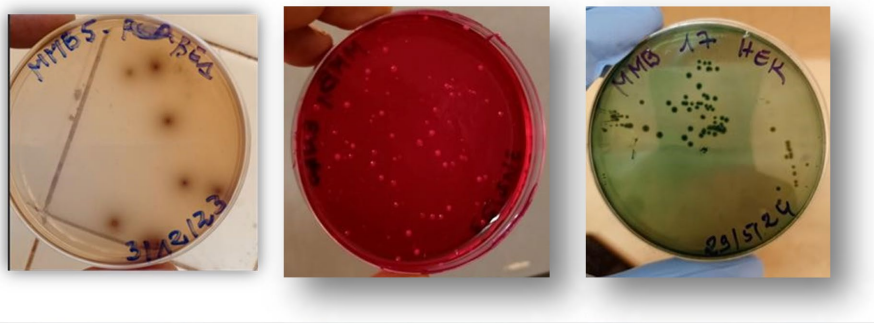

Figure 6. Photo of the different colonies of what and what that developed in which environments?

Figure 7 shows the prevalence of contamination of peanut pastes in the different markets of the city of Bangui.

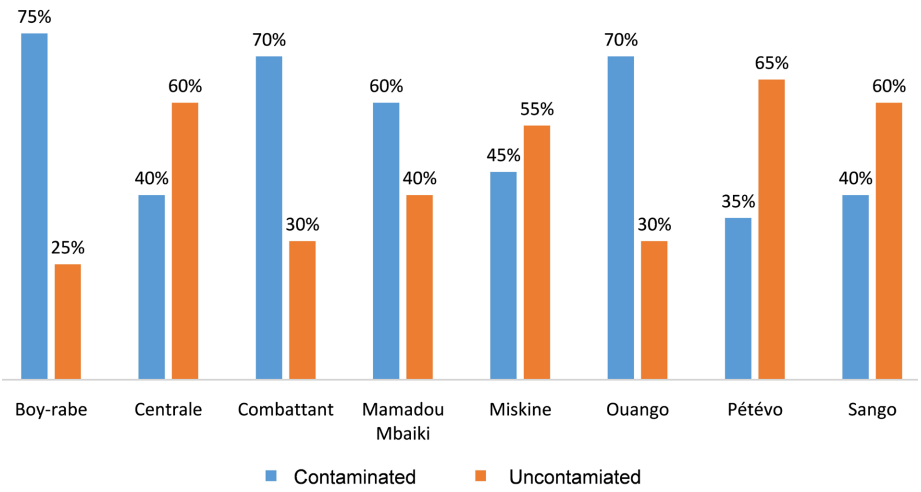

Figure 7. Prevalence of contamination of peanut pastes in the different markets of the city of Bangui.

The prevalence of contamination of peanut pastes at the Boy-rabe market by enterobacteria was 75% that of the Combattant and Ouango markets was 70% each and that of the Mamadou Mbaïki market was 60%.

Table 4. Prevalence of contamination of peanut pastes according to storage duration.

	Contaminated	Uncontaminated
3 days and more	76.23%	23.75%
Less than 3 days	26.25%	73.25%

Peanut pastes stored for more than 3 days were the most contaminated (76.23%) than those stored for less than 3 days (26.25%) (Table 4).

Figure 8 shows the different bacterial species isolated in peanut pastes.

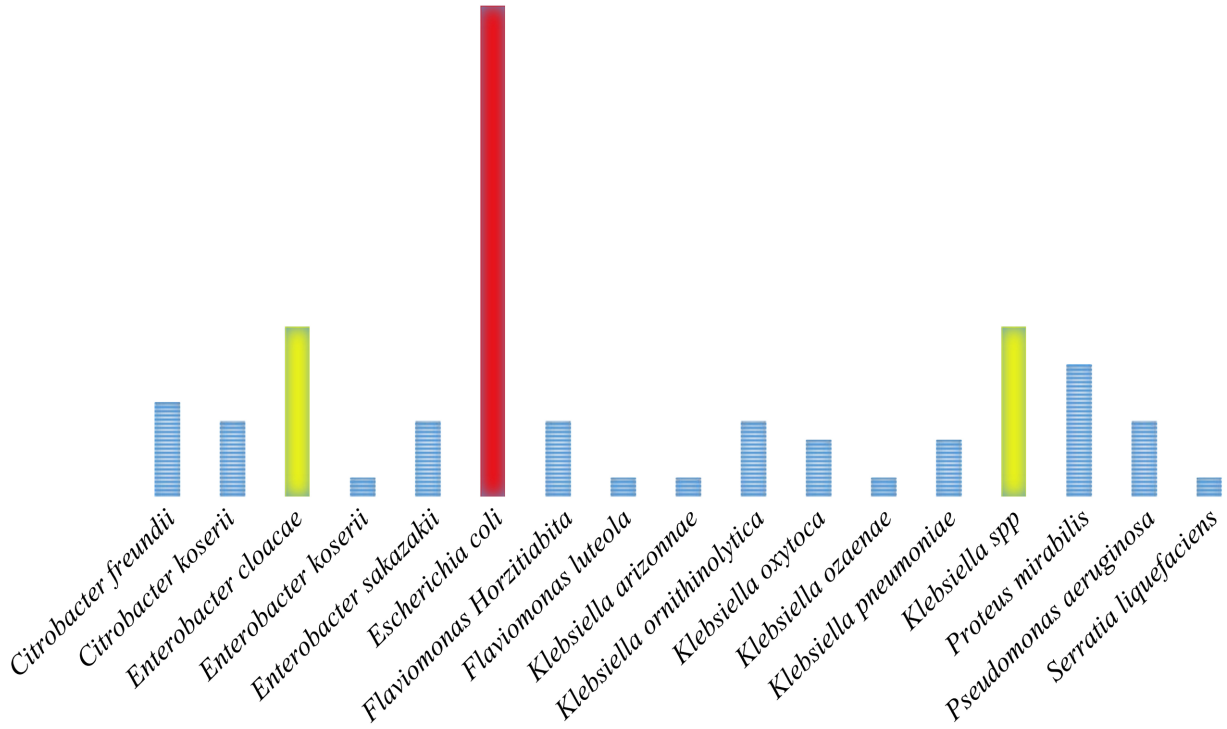

Figure 8. Different bacterial species isolated in peanut pastes.

Escherichia coli strains were more isolated in peanut pastes (29.9%), followed by Enterobactere cloacae and *Klebsiella Spp* strains with a prevalence of 10.3% each.

5. Discussion

Samples of peanut pastes sold in the markets of the city of Bangui were collected and analyzed at the National Laboratory of Clinical Biology and Public Health according to ISO standards [15] [16]. The questionnaire administered to sellers showed that the marketing of peanut pastes in the Central African Republic was carried out 96.3% by women and 3.8% by men. The level of education of sellers was very low. A prevalence of 65.6% of sellers had dropped out of primary school, 3.8% in the first cycle and 24.4% in the second cycle. The results of the studies carried out by Lango yaya *et al.* in the Central African Republic in 2020, Nanadoum *et al.* in Chad and AKA *et al.* in Ivory Coast in 2006 on sales of traditional drinks are really the same with regard to the age groups of sellers, the sex ratio and the level of education [17] [25] [26]. This similarity of results could be explained by the correlation of African cultural diversity which prohibits the practice of certain activities reserved for people of a given sex. The peanut paste manufacturing process is made up of four major steps: sorting, grilling, blanching and

grinding. These production stages are in line with those described by Jean-Luc Knoden *et al.* in the peanut butter manufacturing manual and Woodroof *et al.* on the peanut paste production process who also described these four major steps [3]. The peanut paste processing technique described in our study is almost the same in most African countries. The results of the HACCP risk assessment in this study showed that only 3% of vendors have knowledge of food hygiene, 27% of food preservation technology. For conservation, 61.25% of sellers adopted humidity conservation technology. The proportion of contamination depending on the shelf life was of the order of 76.23% for pasta preserved for more than 3 days and 26.25 for pasta preserved for less than three days. The storage time factor is a risk for contamination of peanut pastes with Chi2 = 0.002. The sales environment had 26.5% dirty spaces, 17% space marked by the presence of flies and 62% covered with puddles of water and 47% dusty space. The personal hygiene of sellers was 14% of sellers with dirty clothes, 57.50% of hair and clothes not covered and 11.88% of lesions located on the upper part of the body. The contamination of peanut pastes is due in most cases to preservation methods (Chi 2 = 0.002 significant) and the hygienic conditions of sellers (Chi 2 = 0.034 significant). The results of this study are inconsistent with the recommendations relating to food hygiene rules and good practice applicable to foodstuffs intended for human consumption described by Buisson *et al.* in 2008 [7]. The method of sale of peanut pastes was of the order of 48.3% of sale only of peanut pastes and 56.3% of sale associated with other food condiments. The results thus obtained corroborate with those of the studies carried out by Raveloson *et al.* in 2004; Razafindramanana *et al.* in 1995 and RAZAFY *et al.* in the city of Antananarivo who obtained in their studies the two modalities such as the sale associated with other food products and the sale only of peanut paste by some sellers [27] [28]. The modality of sale associated with other food products could be explained as the possible strategy to gain and maintain customers. In this study the bacterial load varied 56219 $\times 10^6$ CFU/g for mesophilic aerobic germs; 69 $\times 10^6$ - 71 $\times 10^6$ CFU/g for total coliforms; 43 $\times 10^6$ - 88 $\times 10^6$ CFU/g for fecal coliforms; enterococci were 3.65 $\times 10^6$ - 111.8 $\times 10^6$ CFU/g. Our results are consistent with those obtained by Mejrhit1 *et al.* in Morocco in 2015 in peanuts who obtained similar results [1]. The high number of bacterial load could be explained by the non-compliance with hygiene rules in the production process up to sale; unsuitable sales locations, sales materials as well as lack of knowledge of critical points in the contamination chain. The proportion of germ contamination is very high with mesophilic aerobic germs in the Boy Rabe Market, which means, 219 $\times 10^6$ CFU/g; Miskine Market of 148 $\times 10^6$ CFU/g and 160 $\times 10^6$ CFU/g. For total coliforms, contamination is high at the Combattant Markets of 88 $\times 10^6$ CFU/g; 76.6 $\times 10^6$ CFU/g; Miskine of 71 $\times 10^6$ CFU/g and Pétévo. Fecal coliforms are from 69 $\times 10^6$ to 71.1 $\times 10^6$ CFU/g at the Boy-rab, Combattant and Sango markets. Enterococci of 82.9 $\times 10^6$ CFU/g at the Miskine Market and 111.2 $\times 10^6$ CFU/g at the Pétévo Market and finally sulfite-reducing anaerobes of 26.7 $\times 10^6$ CFU/g at the Mamadou Mbaïki Market and 70.8

$\times 10^6$ CFU/g at the Sango Market. The results obtained are inconsistent with those of the study conducted in Morocco by Euloge S. *et al.* in Benin in 2012 on mycotoxins and microflora contaminating peanuts [29]. The high concentration obtained in certain markets could be explained by the environmental situation of these markets or the places of sale which do not meet the conditions required for the protection and reduction of risks relating to the contamination of peanut pastes for human consumption. Our results are in line with those of a study conducted by Ezzchiel *et al.* in 2012 in Nigeria on the bacteriological quality of peanut butter in local markets in Nigeria, who also obtained a high microbial load of high proportion of coliforms [30]. The overall contamination profile by isolated and identified enterobacteria is 54% prevalence of contamination. The isolated germs were of the order of 3.4% *Klebsiella pneumonae*. 4.6% *Citrobactere koserii, Enterobactere sakazakii, Flaviomonas horzitiabita, Klebsiella oxytoca* and *Pseudomonas aeruginosa*. 5.7% *Citrobactere freundii.* 8% Proteus mirabilis. 10.3% *Enterobactere cloacae* and *Klebsiella spp* and 29.9% *Escherichia coli.* Our results corroborate those of the studies carried out by Euloge S *et al.* in 2012 in Benin; Ezzchiel *et al.* in 2012 and Odu *et al.* in 2001 in Nigeria who isolated and identified pathogenic germs in peanuts and its derivatives [30]. The prevalence of contamination by Market was 35% at the Pétévo Market, 40% at the Centrale and Sango Markets, 60% at the Mamadou Mbaïki Market, 70% at the Ouango Market and 75% at the Boyrab Market. These results could be explained by the fact that the sales environment is often unsanitary from one market to another.

6. Study Limitation

The choice of markets for our sampling was not representative of all markets in the Central African Republic. Few studies have been carried out in this area, which has made it difficult to discuss our results.

7. Conclusion

The results obtained from this study reveal that the consumption of traditionally processed peanut pastes exposes consumers to a serious public health problem. Failure to comply with hygiene rules, lack of knowledge of the concepts of toxi-infection and food poisoning and the unsanitary nature of the sales environment, allows an increase in germs indicating fecal contamination and of medical interest such as *Enterobacteriaceae koserii*, *Flaviomonas luteola, Klebsiella arizonnae, Klebsiella ozaenae, Seratia liquefasciens, Klebsiella pneumonae, Citrobactere koserii, Enterobacter sakazakii, Flaviomonas horzitiabita, Klebsiella oxytoca* and *Pseudomonas aeruginosa, Proteus mirabilis, Enterobacter cloacae* and *Klebsiella spp* and *Escherichia coli*. The high microbial load and the pathogens isolated in this study may pose a risk of poisoning by toxins produced by these microorganisms, foodborne infectious diseases for the health of consumers. We suggest that the Central African Ministry of Public Health monitor foodstuffs intended for human consumption and that sellers strictly comply with the hygiene rules applicable to food-

stuffs to guarantee the health of consumers.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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