Microbiological and Organoleptic Profiles of Mbuja: A Traditional Condiment Produced by Fermentation of *Hibiscus sabdariffa* Seeds in Cameroon

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Abstract: Twelve samples of Mbuja from four villages of Cameroon (Dzbam, Gouzda, Magoumaz and Midirey) were studied to determine their microbiological profile and sensory acceptability. The functional, faecal contaminant and pathogenic flora were screened and numbered using specific media. The results revealed the presence of *Bacillus* and lactic acid bacteria. However, coagulase-positive *Staphylococcus*, total coliforms, thermotolerant coliforms, spores of sulphite reducing anaerobes and yeast and moulds were not detected. Significant differences were observed within and between samples from villages. The pH values ranged from 4.73-6.53, while the water activity was between 0.58 and 0.72. Similar differences were noted for aerobic mesophilic flora, *Bacillus*, lactic acid bacteria. *Enterococcus* and *Bacillus cereus* were detected in only 4 and 3 samples, respectively. Gram staining and biochemical characterization revealed 8 *Bacillus* sp.: *Subtilis, Pumilus, Brevis, Polymyxa, Licheniformis, Laterosporus, Cereus* and *Circulans.* Three lactic acid bacteria species were identifie: *Lactobacillus brevis, Pediococcus pentosaceus* and *Leuconostoc mesenteroides* dextranicum. Five profiles (groups) were obtained on the basis of the functional flora of all samples (*Aerobic mesophilic* flora, *Bacillus*, lactic acid bacteria). The group 1, with the most important bacterial population and the highest pH was the most appreciated by tasters.

Key words: Seeds, *Hibiscus sabdariffa*, fermentation, *Bacillus*, lactic flora

INTRODUCTION

Traditional fermented foods are believed to be beneficial to their consumers. They contribute significantly to the food safety (FAO, 1998) and to the incomes of the populations by creating jobs for countries whose agriculture and the little developed industrial sector cannot absorb a labour force in constant increase (Conroy *et al.*, 1995). Fermented improve the nutritional status of the populations, while providing such micronutrients as vitamins (Steinkraus, 1992), improving the digestibility of foods through the production of enzymes like cellulases (Kovac, 1997) and inhibiting the pathogenic bacteria (Svanberg, 1992).

However in spite of their potential, their ancestral use and their supposed benefits, a number of traditional fermented products have not been the subject of scientific investigations yet. It is the case for Mbuja, a traditional condiment used as seasoning agent, produced by fermentation of cooked seeds of *Hibiscus sabdariffa* in Cameroon and in some other countries in Africa. Different names are given to the fermented seeds of *Hibiscus sabdariffa*: Datou, Bi-Kalga, Ganyiri-kolo (West Africa), Furundu (Sudan). In all these countries, the production remains traditional and the fermentation spontaneous.

With regard to fermented seeds of *Hibiscus sabdariffa*, studies carried out on Bi-Kalga in Burkina Faso showed an evident improvement of the nutritional value in particular of the protein content of the seeds (Bengaly *et al.*, 2003). But, very few information is available concerning the process of microbiological transformation of a product, which could be a good meat substitute at the disposal of populations with very low incomes.

The importance and the pre-eminence of *Bacillus* in the fermentation of many other proteinaceous seeds to produce traditional condiments were highlighted in several former studies (Odunfa, 1988; Steinkraus, 1991;

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Ndir *et al.*, 1997). *Bacillus* genera were isolated in homologous products like okpehe (Oguntoyinbo *et al.*, 2003) and Netetu or Dawa-dawa (Ndir *et al.*, 1997; Sakyi-Dawson, 2001). Their functional properties proved to be of important nutritional impact on the seeds (proteolytic and amylolytic activities, hydrolysis of the pectic substances and non-digestible sugars).

The present research, which is a first stage towards the control of the conditions of production of the condiment, aims at establishing a typology of Mbuja from different origins and producers on the basis of their microbiological profile (functional flora, pathogenic flora and quality indicators) and at selecting a typical condiment (on the basis of sensory acceptability) for further studies.

MATERIALS AND METHODS

Characteristics of the samples of Mbuja: The samples of Mbuja come from 4 localities of Cameroon: Dzbam (13.73' long East; 10.88' Lat. North), Gouzda (13.82' long East; 10.85' Lat. North), Magoumaz (14' long East; 10.78' Lat. North) and Midirey (13.86' long East; 10.81' Lat. North). All sampling villages are situated in the main region of production o f Mbuja, around the town of Mokolo (13.8' long East; 10.7' Lat. North). The production is realised according to a traditional method shown in Fig. 1 small scales family production units. Three different production units were selected for each locality of sampling. They were immediately packed in sterile bags to avoid any recontamination and stored at 25°C, the average temperature in the sampling area at the time of experience.

Physicochemical analyses: Two physicochemical parameters, chosen for their influence on microbial growth, were studied: pH and the water activity. The pH was read using a pH meter LPH 330T (Grosseron, France) on suspensions of 10 g of each sample in 150 mL of de-ionised water and the water activity (a_w) was determined with an a_w-meter FA-ST/1 (GBX scientific instruments).

Microbiological analysis
Screening of microorganisms: Two types of micro organisms were screened on specific media (Table 1). pathogenic bacteria and quality indicators on one hand and functional flora on the other. The screening and counts of the various bacterial populations were carried on using decimal dilutions of 'Mbuja'. Ten grams of each sample were first diluted in 90 mL of tryptone-salt (tryptone-salt 9, 5 g diluted in 1 L of distilled water)
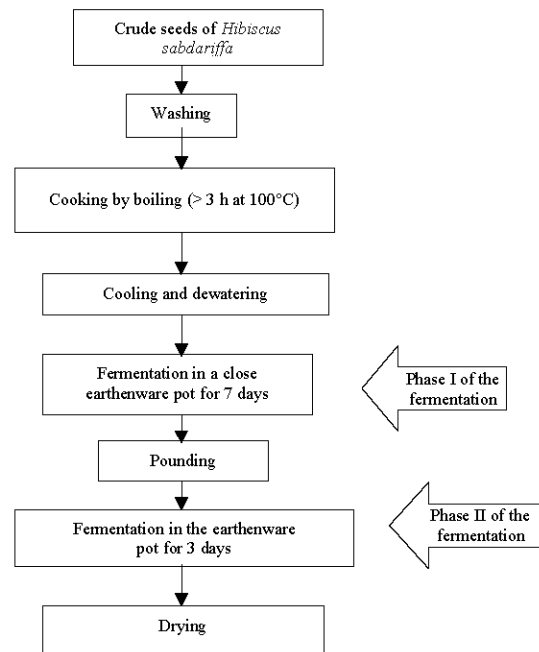

Fig. 1: Flow diagram of 'Mbuja' production

using a mix 1 stomacher (AES laboratoire). This suspension was thereafter, used for the preparation of decimal dilutions of 10^{-2}-10^{-6}.

Surface plating was carried out using a spiral (spiral system and interscience) for the screening of aerobic mesophilic flora (on PCA medium), lactic bacteria (MRS), spores of *Bacillus* after heat treatment of the samples (10 min with 80°C) (glucose agar with BCP), *Bacillus cereus* (Mossel medium), yeasts and moulds (yeast and malt extract agar + streptomycine and chloramphenicol).

For the screening of coagulase + staphylococci (Baird Parker medium), total coliforms (medium VRBL at 30°C), thermotolerant coliforms (VRBL at 44°C) and enterococci (Bile Esculine Azide), plating was made by inclusion, while flora of Anaerobes sulfite-reducing anaerobes were analyzed in tubes of VF-sulfite medium. The conditions of plating and incubation are shown in Table 1. The enumeration was carried out by counting the characteristic colonies in CFU (Colonies Forming Units).

Characterization and identification of the strains: The identification of the isolated strains of *Bacillus* was carried out by biochemical characterization according to the modified dichotomic key of Gordon *et al.* (1973). The following physiological and biochemical tests were performed on each presumed *Bacillus*: Vosges-Proskauer reaction, mobility, fermentation of the mannitol, use of citrate, hydrolysis of starch, hydrolysis of gelatine, casein

Table 1: Plating condition used for the screening of different microflora studied according to methods by Afnor (1999)

Flora analyzed	Culture medium and references	Temp. (°C) of incubation	Incubation time (h)
Total coliforms	VRBL (Merck)	30	24
Thermotolerants coliforms	VRBL (Merck)	44	24
Spores of sulfite reducing anaerobes	VF sulfite (AES laboratoire)	30	24
Coagulase + staphylococci	Baird Parker	37	24-48
Pathogenic bacteria and quality indicators			
Enterococci	Bile Esculine Azide (AES laboratoire)	37	24
Spores of *Bacillus cereus*	Mossel (AES laboratoire)	37	48
Aerobic mesophilic flora	PCA (Merck)	30	48
Spores of *Bacillus*	Glucose agar +BCP (AES laboratoire)	30	48
Functional flora			
Lactic flora	MRS (AES laboratoire)	30	48
Yeast and moulds	Yeast and malt extract agar + streptomycine and chloramphenicol (LBEM, ESMISAB)	25	2-5 days

and lecithin, production of acid and gas from glucose, reduction of nitrates, growth at 50 and 65°C, growth in NaCl 7% and pH of the cultures in VP medium.

The isolates of lactic bacteria were characterized and identified on the basis of morphology, gram staining, catalase test and their biochemical profiles obtained by the API 50 CHL galleries (BioMérieux SA, France) were analyzed using APILAB plus software. *Pediococcus* were identified according to the biochemical characteristics described in the Bergey's manual of determinative bacteriology (Holt *et al.*, 1994).

Sensory evaluation: Sensory analysis was carried out on groups of samples obtained by the principal component analysis based on their microbiological composition. A sample of each group of condiments having the same microbiological characteristics was submitted to a panel made up of 13 women from 35-42 years-old, selected on the basis of their frequent use (use of the condiment in the preparation of at least one of the three daily meals) and continuous use (consumption since the childhood) of the Mbuja. They were asked to mark the samples for the taste, the colour, the flavour and the general acceptability of the product.

A 9 points hedonic scale was used and only one characteristic was evaluated at the same time. The tasters were held to rinse their mouth after each elementary tasting before the sample, according to a definite order.

Statistical analysis of the results: The results obtained for the various treatments were compared by Analysis of Variances (ANOVA) followed by the test of Duncan, where significant differences were noted, using STATgraphics Plus 5.1 software (Manugistic Inc. Software, Rockville the USA). The correlations between the parameters and their significance were obtained with statistica for windows (STATSoft Inc., Tulsa USA). The typology of the samples according to their microbiological

composition was established using a Principal Component Analysis (CPA) carried out with STATBOX 6.6 (Grimmersoft, France).

RESULTS

Pathogenic flora and quality indicators: The pathogenic bacteria and the quality indicators were analyzed in order to determine the safety and hygienic quality of 'Mbuja'. As shown on Table 2, total coliforms, thermotolerant coliforms, spores of sulfite-reducing anaerobes and coagulase + staphylococci were absent or lower than the limits of detection. However, the enterococci and *B. cereus*, whose faecal origin is possible, were detected in 4 and 3 samples out of 12 analyzed, respectively. The counts of enterococci varied from 3.5×10^4-4.5×10^5 CFU g^{-1} of 'Mbuja' produced by Dzbam 1 and Gouzda 2. *Bacillus cereus* were detected with concentrations of 3×10^4, 3×10^5 and 4×10^5 spores g^{-1}, respectively Dzbam 1, Gouzda 2 and Magoumaz 3.

Functional flora
Aerobic Mesophilic Flora (AMF): The AMF was evaluated with an aim of determining the total bacterial load of the 'Mbuja'. It varied between 8.1×10^5 and 2.7×10^8 CFU g^{-1} (Table 2). The sample Dzbam 1 had the greatest total bacterial load while that of Magoumaz 1 was the weakest. A significant difference was observed between the samples from various origins (p≤0.05). This difference was mainly due to the producer. However, the two samples with the weakest AMF (Magoumaz 1 and 2) did not present a significant difference.

Bacillus: The counts of the spores of *Bacillus* in the 'Mbuja' revealed densities ranging between 2.6×10^5 and $6.21 0^7$ spores g^{-1} (Table 2). The greatest number of spores of *Bacillus* was detected on the sample of Dzbam 1, whereas the weakest was detected on that of Magoumaz 1. However, no significant difference was noted on the five samples with weak *Bacillus* flora

Table 2: Microbial screening and counts for the 12 sample of Mbuja

Micro-organisms screend	Average microbial count (log CFU g^{-1} ±SD)					
	Dzbam			Gouzda		
	Dz 1	Dz 2	Dz 3	Ga 1	Ga 2	Ga 3
Total coliforms	ND	ND	ND	ND	ND	ND
Thermotolerant coliforms	ND	ND	ND	ND	ND	ND
Spores of sulphite reducing anaerobes	ND	ND	ND	ND	ND	ND
Coagulase + staphylococci	ND	ND	ND	ND	ND	ND
Enterococci	4.54±0.09^a	ND	ND	ND	5.65±0.07^d	ND
Spores of *bacillus*	5.46±0.21^a	ND	ND	ND	ND	5.96±0.07^b	ND
Aerobic mesophilic flora	8.43±0.10^f	6.86±0.19^{ab}	7.39±0.08^{bc}	7.10±0.02^{ab}	7.75±0.01^c	7.65±0.03^{bc}
Spores of *Bacillus*	7.79±0.00^e	6.20±0.16^a	5.97±0.03^a	6.44±0.07^a	7.21±0.03^d	7.12±0.02^{cd}
Lactic flora	8.82±0.05^c	7.19±0.57^a	7.35±0.06^a	6.37±0.08^a	6.90±0.16^a	6.27±0.26^b
Yeast and moulds	ND	ND	ND	ND	ND	ND

	Magoumaz			Midirey		
	My 1	My 2	My 3	Mz 1	Mz 2	Mz 3
Total coliforms	ND	ND	ND	ND	ND	ND
Thermotolerant coliforms	ND	ND	ND	ND	ND	ND
Spores of sulphite reducing anaerobes	ND	ND	ND	ND	ND	ND
Coagulase + staphylococci	ND	ND	ND	ND	ND	ND
Enterococci	ND	5.02±0.03^b	ND	ND	ND	5.36±0.08^c
Spores of *Bacilluscereus*	ND	ND	4.46±0.21^c	ND	ND	ND
Aerobic mesophilic flora	5.91±0.00^a	6.44±0.02^a	8.04±0.03^d	8.29±0.04^c	7.62±0.12^{abc}	7.53±0.20^{abc}
Spores of *bacillus*	5.42±0.05^a	6.40±0.12^a	7.77±0.04^e	7.07±0.04^c	7.04±0.04^{bc}	6.89±0.10^b
Lactic flora	5.70±0.12^a	7.31±0.03^a	7.06±0.21^a	7.61±0.14^a	7.08±0.10^a	8.69±0.05^b
Yeast and moulds	ND	ND	ND	ND	ND	ND

Different letters in the same line indicate a significant statistical defference according to test (p<0.05)

(Dzbam 2 and 3, Gouzda 1, Magoumaz 1 and 2). Similarly, there were not significant difference between the two stronger values of *Bacillus* counted in the samples Dzbam 1 and Magoumaz 3.

Twenty 6 strains of *Bacillus* were isolated and purified. Observations of the shapes of cells after gram staining as well as the biochemical characterization and their use in the dichotomic key of Gordon *et al.* (1973) revealed the occurrence of 8 species of *Bacillus*: *B. subtilis* (7% of the isolates), *B. pumilus* (19%), *B. brevis* or *Brevibacillus brevis* (15%), *B. polymyxa* (4%), *B. licheniformis* (15%), *B. laterosporus* or *Brevibacillus laterosporus* (27%), *B. cereus* (7%) and *B. circulans* (6%).

Lactic flora: The most abundant flora was counted in the sample of Dzbam 1 (6.6×10^7 CFU g^{-1}) and the least important in that of Magoumaz 1 (5.8×10^5 CFU g^{-1}) (Table 2). However, 10 of the 12 samples did not present a significant difference between them with regard to the lactic flora (Dzbam 2 and 3; Gouzda 1-3; Magoumaz 1-3; Midirey 1 and 2).

Eight strains of gram positive and catalase negative bacteria were isolated on MRS medium. Their biochemical characterization on API 50 CHL galleries and the biochemical tests described in Bergey's manual of determinative bacteriology (Holt *et al.*, 1994) helped in identifying these strains. Three species were therefore, identified: *L. brevis*, *L. mesenteroides* dextranicum and *P. pentosaceus*.

pH and activity of the water of the various samples of Mbuja: Measurements of the pH of the samples of Mbuja of different origins (Fig. 2) provided low pH values ranging between 4.73 and 6.53. The most important pH was obtained for the sample of Dzbam 1 and the lowest for that of Midirey 2. The difference between most samples was significant (p≤0.05).

The water activity of the whole samples varied from 0.58 (Midirey 2) to 0.72 (Magoumaz 3) and differed significantly (p≤0.05) between the samples (Fig. 3).

Profile of the samples according to the functional flora: The samples of 'Mbuja' analysed differed with regard to the functional component of their microflora. The Principal Component Analysis (PCA) showed that >95% of the variation is explained by the principal plan (F1, F2). The axis F1 explains 75.9% of total variability and is correlated with *Bacillus* sp. (Bsp.) and with the aerobic mesophilic Flora (FT), while the axis F2 explains 19.8% of variability and is correlated with the lactic Flora (FL).

The PCA revealed 5 profiles or goups with the following characteristics (Fig. 4).

Group 1: High concentrations in lactic Flora (FL), total Flora (FT), *Bacillus*. This group included the two repetitions of the sample Dzbam 1 (Dm 1A and B).

Group 2a: Average concentrations in FL, FT, *Bacillus*. It comprised the samples Dzbam 2A (Dm 2A) and Dzbam 3A and B (Dm 3A and B).

Group 2b: High concentrations in FL and low concentrations in FT and *Bacillus*. It was made up of samples Midirey 3A and B (My 3A and B).

Group 3: Low concentrations in FL and high concentrations in FT and *Bacillus*. This group was the opposite of group 2 and included the samples Gouzda 1 (Ga 1A and B), Gouzda 2 (Ga 2A and B), Gouzda 3 (Ga 3A and B), Midirey 2 (My 2A and B) and Magoumaz 3 (Mz 3A and B).
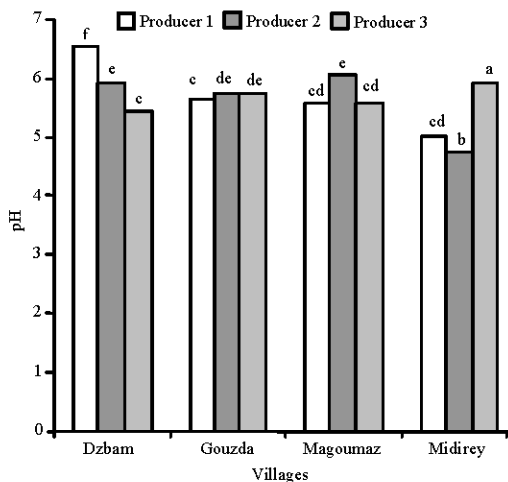

Fig. 2: The pH of samples from different villages and producers Different letters on histograms indicate a significant statistical difference according to Duncan test (p ≤ 0.05)
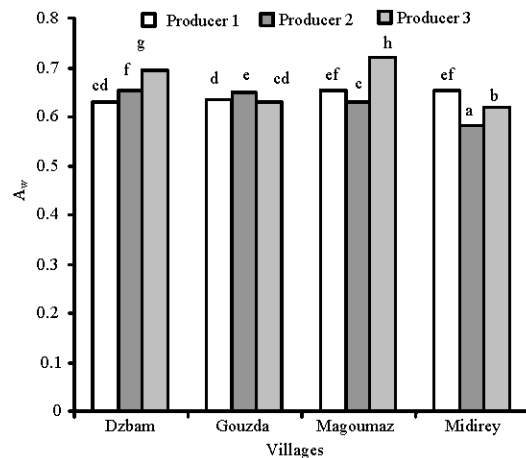

Fig. 3: Water activity of samples from different villages and producers Different letters on histograms indicate a significant statistical difference according to Duncan test (p ≤ 0.05)
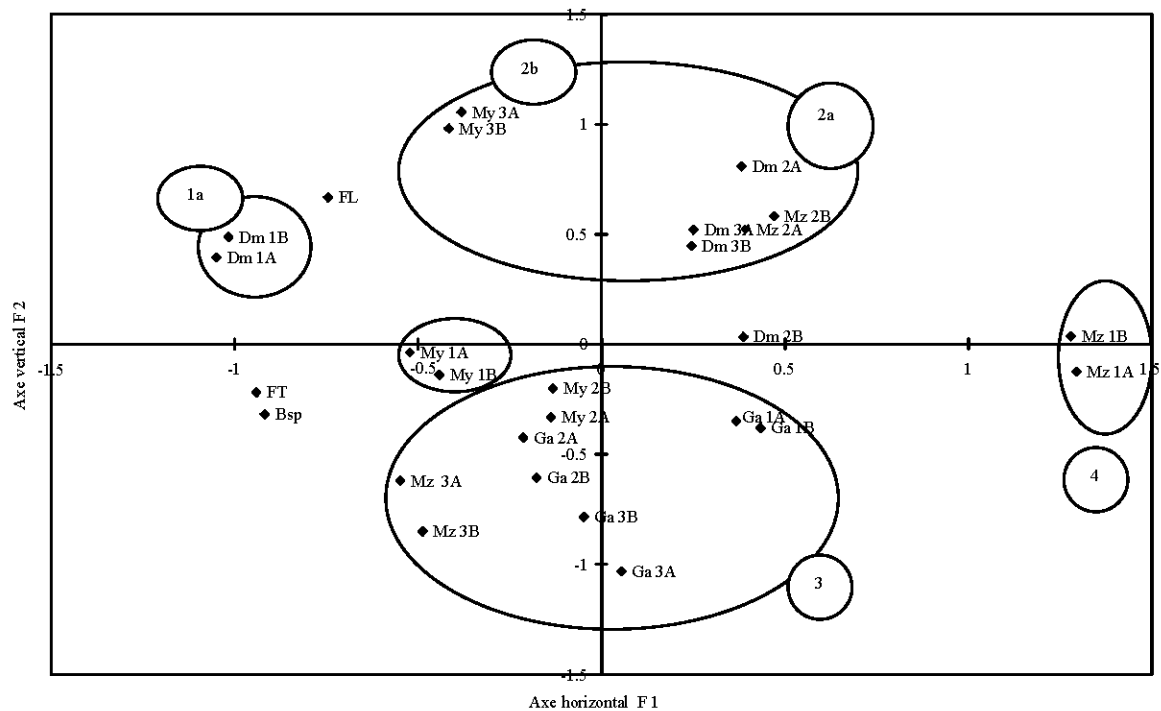

Fig. 4: The PCA profile of samples based on the functional flora: lactic Flora (FL), aerobic mesophilic Flora (FT) and *Bacillus* (Bsp.)
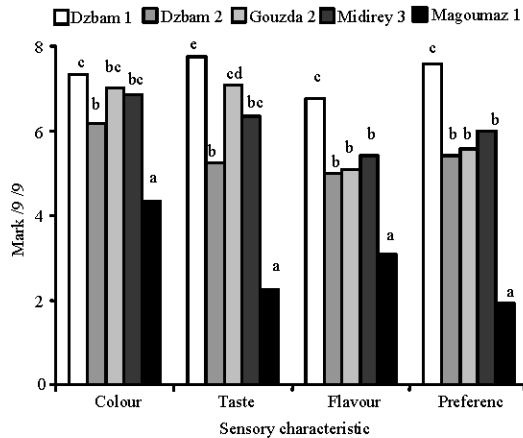
Fig. 5: Sensory scores of 'Mbuja' from different groups (means over 9 expressed in the hedonic scale). Different letters on histograms indicate a significant statistical difference according to Duncan test (p≤0.05)

Group 4: Low concentrations in FT, FL and *Bacillus*. This group was the opposite of group 1 and had the 2 repetitions of the sample Magoumaz 1 (Mz 1A and B).

Sensory analysis: The organoleptic parameters (colour, taste and flavour) and the preference were evaluated to determine the most appreciated 'Mbuja' from the five different microbiological profiles established by the principal component analysis. The panelists perceived a significant difference between all the sensory parameters tested (Fig. 5). The flavour, the colour and the taste were significantly correlated with the consumer's choice for a sample of Mbuja (R = 0.76; 0.65; 0.81, respectively at p≤0.05). The sample Dzbam 1 (group 1) obtained the best preference score. On the other hand, those of the 2a groups, 2b and 3 did not present significant difference as regards the preference whereas the sample group 4 (Magoumaz 1) was the least preferred, with a score of 2 out of 9 in the hedonic scale).

DISCUSSION

The data collected made it possible to draw up a first inventory of the categories of bacteria isolated in the Mbuja and to try to check if the microbial typology of a product resulting from a given locality were connected to organoleptic characteristics.

The first point was related to the pathogenic flora and the hygienic quality indicators whose detection would be a handicap for the development of this product and would require a control of the risk. The results revealed the good

hygienic quality of the products studied. These findings were different from those obtained by Ndir *et al*. (1997) for Netetu, a condiment obtained by fermentation of *Parkia biglobosa* seeds. On 9 samples various sources, these researchers showed that 6 contained >10^3 CFU g^{-1} Streptocoques D; 7 had >10 CFU g^{-1} sulfite reducing *Clostridium* and all were contaminated by pathogenic *Staphylococci*, with less 100 CFU g^{-1} however. The fair good hygienic quality of Mbuja could be explained by the initial heat treatments of seeds (cooking by boiling at 100°C for >3 h) and sun-drying upon fermentation, but also partially by acidic pH, sometimes lower than 5. The competition of the other microbial species (lactic bacteria and *Bacillus*), production of antibacterial substances by some of these species combined with the inhibiting effect of the acidic pH and the weak a_W on studied enteropathogens and by extension on *Salmonella* and *Shigella* can explain the relative good hygienic quality of the products tested.

This is confirmed by the absence of quotation of food poisoning cases related to Mbuja. However, indices of lack of hygiene (presence of *Enterococcus*, detection of *Bacillus cereus*), probably due to a recontamination by the operators are noted in some products.

Indeed the 'Mbuja' proved to be slightly acidic (pH ranging between 4.73 and 6.53). Acidic pH (ranging between 5.6 and 5.8) were also obtained for Furundu, a meat substitute prepared by fermentation of cooked *Hibiscus sabdariffa* seeds in Sudan (Yagoub *et al*., 2004). These pH indicate hydrolysis of sugars, proteins and lipids ending in the release of organic acids, amino acids and fatty acids. However, for other fermented products, the pH values were equal or higher than 8. It is the case of Bi-Kalga obtained also by fermentation *Hibiscus sabdariffa* of seeds (Bengaly *et al*., 2003) and of many other traditional condiments obtained by fermentation of proteinaceous seeds like Dawa dawa (seeds of *Parkia biglobosa*), Ogiri (seeds of *Citrillus vulgaris*), Ogiri-saro (seeds of *Sesamum indicum*) etc. whose pH is equal to or higher than 8 (Steinkraus, 2002).

Two groups of bacteria implied in fermentation were detected and counted: *Bacillus* and lactic flora. The number of *Bacillus* in the 'Mbuja' was variable but remained high in all samples. The identified species exhibited an important metabolic activity: hydrolysis of proteins and starch in particular. The phenotypical identification of *Bacillus* revealed the occurrence of 8 species of which the more represented was *B. laterosporus* or *Brevibacillus laterosporus* (27% of the isolated strains) whereas *Bacillus polymyxa* was the least frequent, on the contrary to the majority of the African traditional condiments, where *Bacillus subtilis* was the dominant species (Sanni *et al*., 2000).

The prevalence of the *Bacillus* genera was earlier noted in other fermented products like Netetu, Dawa-dawa and the Ogiri (Ndir *et al.*, 1997; Oguntoyinbo *et al.*, 2003; Sakyi-Dawson, 2001). The apparent the variability in *Bacillus* counts in the different samples could be in relation with the unstandardized conditions of fermentation.

On the other hand, the present analyses are in favour of a bigger role of the lactic flora in the production of 'Mbuja'. Such a role has not been described in the production of other condiments obtained by fermentation of proteinaceous seeds (Steinkraus, 1991). Whatever, the source of the 'Mbuja', an important lactic flora was detected. *Pediococcus pentosaceus, Lactobacillus brevis* and *Leuconostoc mesenteroides* dextranicum were the species identified in this condiment. The proportion between these species depended on the samples but all the species were always counted.

CONCLUSION

A Principal Component Analysis (PCA) based on the decimal logarithms of bacterial counts taking into account the aerobic mesophilic flora, the lactic flora and *Bacillus*, showed that the samples constituted five profiles or groups. However, the profiles were made up only on a quantitative basis since the same type of flora was met in all samples.

The organoleptic tests carried out on representatives of each group confirmed a clear separation of the groups. The metabolic activity of the various bacterial species probably generated different sensory profiles for each sample. The sensory preference was affected by the parameters taste, colour and flavour. A recombination of the groups could be considered if the preference of tasters was taken into account: group 1 (more accepted) constitute one group, groups 2a, 2b and 3 form another group and group 4 (less accepted) the last group. The taste, the colour and the flavour were significantly correlated with the general preference of the sample of the 'Mbuja', with the flavour and the taste having the strongest correlation. The development of the organoleptic properties thus resulted from complex phenomena generated by the microflora during the fermentation. For this aim, the analysis of the aromatic compounds in relation to the flora implied in fermentation would better elucidate the influence of the biochemical processes on the development of these sensory properties of 'Mbuja'. The analysis of the dynamics of the bacterial populations in the fermentation process would also bring useful precise details suited to better determining the importance of certain bacteria and in the

long term to better control the production of the 'Mbuja'. In effect, the technology of production described in the same way on the various sites could be variously applied.

REFERENCES

Afnor, 1999. Microbiologie Alimentaire, Tome 1: Méthodes Horizontales, 7th Édn., pp: 663.

Bengaly, M.D., A.B. Bere and A.S. Traore, 2003. Etude de la composition du Bi-Kalga: Un condiment traditionnel riche en protéines obtenu par fermentation des graines d'oseille de Guinée (*Hibiscus sabdariffa*). Voies alimentaires d'amélioration des situations alimentaires en Afrique de l'Ouest 23-28 Novembre. Ouagadougou, Burkina Faso.

Conroy, C., A. Gordon and A. Marter, 1995. Development and Dissemination of Agro Processing Technologies, Natural Resources Institute, UK.

FAO, 1998. Food Agriculture Organization. Fermented fruits and vegetables. A global perspective. FAO Agricultural Services. Bulletin No. 134.

Gordon, R.E., W.C. Haynes and C.H.N. Pang, 1973. The Genus *Bacillus*, Agriculture Handbook No. 427. US Department of Agriculture, Washington DC.

Holt, J.G., N.R. Krieg, P.H.A. Sneath, J.T. Staley and S.T. Williams, 1994. Bergey's Manual of Determinative Bacteriology. 9th Edn. Williams and Wilkins, Baltimore, Maryland 21202, USA.

Kovac, B., 1997. The use of the Mould *Rhizopus oligosporus* in food production. Food Technology and Biology.

Ndir, B., R.D. Gningue, N.G. Keita, M. Souane, L. Laurent, C. Cornelius and P. Thonart, 1997. Caractéristiques microbiologiques et organoleptiques du nététu du commerce. Cahiers Agricultures, 6: 299-304.

Odunfa, S.A., 1988. Review: African fermented foods, from arts to science. Mircen J., 4: 255-273.

Oguntoyinbo, F.A, A.I. Sanni, W.H. Holzapfel and C.M.A.P. Franz, 2003. Caractérisation génomique et propriétés fonctionnelles d'espèces de *Bacillus* isolées de l'Okpehe, un condiment fermenté traditionnel. Voies alimentaires d'amélioration des situations alimentaires en Afrique de l'Ouest 23-28 Novembre. Ouagadougou Burkina Faso.

Sakyi-dawson, E.O., 2001. Feasibility of the use starter cultures in the production of soydawadawa. IFT Annual Meeting-New Orleans, Louisiana, USA.

Sanni, A.I., G.S. Ayernor, E. Sakyi-dawson and S. Sefa-dedeh, 2000. Aerobic spore-forming bacteria and chemical composition of some Nigerian fermented soup condiments. Plant Foods Human Nutr., 55: 111-118.

Steinkraus, K.H., 1991. African Alkaline Foods and Their Relation to Similar Foods in Other Parts of the World. In: Westby, A. and P.J.A. Reilly (Eds.). Traditional African Foods-Quality and Nutrition. Stockholm, Sweden: International Foundation for Science, pp: 87-92.

Steinkraus, K.H., 1992. Lactic acid fermentations in applications of biotechnology to traditional fermented foods, report of an Ad Hoc panel of the Board on Science and Technology for the International Development, National Academy Press, Washington DC, USA.

Steinkraus, K.H., 2002. Fermentations in world food processing. In: Comprehensive Reviews in Food Sci. Food Safety, 1: 23-32.

Svanberg, B., 1992. Fermentation of cereals: Traditional Household Technology with Nutritional Benefits for Young Children, IDRC Currents 2, Canada.

Yagoub, A.E.A., B.E. Mohamed, A.H.R. Ahmed, A.H. El-Tinay, 2004. Study on furundu, a traditional sudanese fermented roselle (*Hibiscus sabdariffa* L.) seed: Effect on *in vitro* protein digestibility, chemical composition and functional properties of the total proteins. J. Agric. Food Chem., 52: 6143-6150.