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Physical, chemical and microbiological characteristics of lafun produced in Benin

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Lafun is an African traditional fermented food product prepared from cassava. Whereas Nigerian lafun has been studied, characteristics of Beninese lafun are still unknown. This research was carried out to identify the characteristics of two types of lafun, Chigan lafun and ordinary lafun produced in Benin. The distinctive characteristics of Chigan lafun (the preferred type) were its lower solubility and fibre content and its higher hot paste viscosities compared to ordinary lafun. Whatever the type, lafun was found to be a dried and white product with a highly variable pH (4.5-8.8). Both types were rich in carbohydrates (76.0% of starch and 3.3% of crude fibres), poor in proteins (1.0%) and containing fat (0.4%) and ash (1.2%). The swelling power of the lafun flour (expressed by the quantity of water absorbed by 1 g of flour) was 28.9 g water/g for the both types. Lafun has a variable microbial load which levels ranged from 10^4 to 10^8 cfu/g and made up of *Bacillus* spp. (10^4 - 10^8 cfu/g), lactic acid bacteria (10^3 - 10^7 cfu/g), Enterobacteriaceae (10^3 - 10^7 cfu/g) and yeasts (10^2 - 10^7 cfu/g).

Key words: Cassava, characteristics, lafun, micro-organisms, retting, stiff porridge.

INTRODUCTION

Cassava is an important root crop often processed into various food products in tropical countries. In West Africa the most widely known cassava derived food is garri. Several research works have been carried out on the processing, nutritional and microbiological characteristics and quality improvement of garri (Westby and Twiddy, 1992; Nago, 1995; Igbeka, 1995; Giraud et al., 1995; Omonigho and Ikenebomeh, 2002; Kostinek et al., 2005). However, there are other traditional cassava products, like lafun, which are not so widely known.

Lafun is a fermented and sun-dried cassava pulp produced in Nigeria and Benin. The dried pulp is milled into flour which is used to prepare a stiff porridge called Oka usually consumed with tomato or vegetable soups. Two types of lafun are produced in Benin; ordinary lafun and Chigan lafun (Figure 1). Chigan lafun differs from ordinary lafun by its whiter colour and the high extensibility of its stiff porridge that determines the superiority of quality of root crops derived pastes in Benin (Hounhouigan et al., 2003).

The most important operations involved in both ordinary lafun and Chigan lafun processing are cassava root peeling, washing, retting, crumbling and drying (Figure 1). Chigan lafun production differs from ordinary lafun production by washing before and after peeling/cutting the tuber and by 2-3 washing operations after retting. According to the processors, washing after retting allows to reduce the strong aroma of the product, the toxic substrates of cassava and ensure a white colour to the lafun (unpublished information). Furthermore, for Chigan lafun production, retting is done in a covered container and retted cassava pulp is dried on a high platform to avoid dusts contrarily to the common practice. All the cassava varieties grown by the local farmers are suitable for lafun production. The use of previously prepared lafun as 'back

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Abbreviations: L^* , Lightness; ΔE , the total difference of colour from the white ceramic standard; RVU, rapid visco analyser unit; Wb, wet basis; db, dry basis; n, number of samples; cv1, cv2 and cv, coefficients of variation.

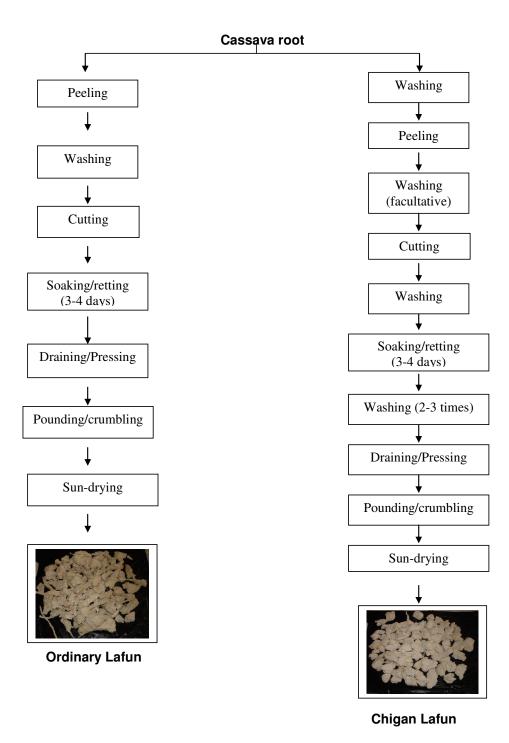


Figure 1. Procedure for preparation of ordinary lafun (left) and Chigan lafun (right).

slopping' for the fermentation process is a common practice among about 62% of the Beninese lafun processors (unpublished information).

Lafun has been extensively studied in Nigeria for the analysis of chemical composition (Oke, 1965; Longe, 1980), evaluation of different drying methods (solar, oven and sun-drying) on the quality (Sanni et al., 1998) and identification of cassava preparation of quality lafuns preferred by consumers (Oyewole and Afolami, 2001). More recently, Nwabueze and Odunsi (2007) reported that lafun yield, colour and sensory attributes or the reduction of its cyanogenic potentials could be maximized through the optimization of inoculum volume, fermentation time and drying temperature. Nwachukwu and Edwards (1987) and Oyewole and Odunfa (1988) studied the micro-organisms associated with cassava fermentation during Nigerian lafun production. Ijabadeniyi (2007) reported that a wide spectrum of bacteria and filamentous fungi occurred in lafun sold in Nigerian market. However, there is no information about the quality characters of the two types of lafun produced in Benin. Moreover, since lafun is used as a 'starter', there is a need to investigate its microbiological profile. This study aimed to determine the physico-chemical and microbiological characteristics of the different types of the Beninese lafuns as they were proposed to the rural and urban consumers.

MATERIALS AND METHODS

Samples collection

Lafun samples were collected from processors and sellers of the most important production areas in Benin (Ketou and Pobe). Twenty-two (22) samples of 500 g each including 15 ordinary lafun and 7 Chigan lafun were collected aseptically with a sterile spoon in sterile stomacher bags (Lab Blender, Model 4001, Seward Medical, London, UK) and stored at 5 °C. They were later used for physical, chemical and microbiological analysis. The microbiological analysis was conducted at the latest 48 h after sampling. Two samples were taken for each analysis.

Physical analysis

Colour parameters (Lightness L* and the total difference of colour from the white ceramic standard ΔE) of lafun were determined with a Minolta Chroma Meter CR-210b (Minolta Camera Co. Ltd, Osaka, Japan) in L*a*b* system (McLaren, 1976) with a white ceramic plate used as standard (D₆₅ Y_{94.8} x $_{.3150}$ y $_{.3324}$). Titratable acidity and pH of the suspension obtained from 10 g of Lafun flour in 90 ml of distilled water was measured (Nout et al., 1989). Solubility and swelling power of lafun flour was determined according to Mestres et al. (1997). A water suspension containing 4% (w/w) of lafun flour was heated with a Rapid Visco Analyser (RVA, Newport Scientific, Naraben, Australia) from 35 to 70 °C and held at this temperature for 3 min. Pasting temperature, peak of viscosity and final viscosity of lafun flour were recorded on RVA, as described by Padonou et al. (2005) for cassava flour, with a water dispersion that contained 8% dry mater. This suspension was heated from 35 to 80°C, held at 80 °C for 3 min and cooled to 50 °C.

Proximate analysis

Water content of the samples was determined after heating the flour at 105 °C for 24 h [Method 44-01, American Association of Cereal Chemists (AACC) 1983]. Protein content was calculated by the Kjeldahl-Nitrogen analysis procedure, using 6.25 as conversion factor (Method 46-11A, AACC 1983). Lipids content was determined after extraction with petroleum ether (Method 30-25, AACC 1983) and ash content was evaluated by Method 08-01 (AACC 1983). Starch content was assessed by the modified polarimetric method (Akissoé, 1992; personal commucation). Total fibre content was determined as described by Osborne and Voogt (1978).

Microbiological analysis

Microbial cultivation was carried out basically following the approach described by Amoa-Awua and Jakobsen (1995). Aerobic

mesophilic counts (AMC) were made on Plate Count Agar medium (PCA; Oxoid CM 325). The morphological characteristics of colonies on PCA were examined and each morphotype was observed by microscopy (x1000, Axiostar plus, Carl Zeiss, Göttingen, Germany). The number of colony forming units (cfu) of morphotypes which displayed bright spores on microscope was recorded separately. These colonies were also examined for Gram reaction (Gregersen, 1978) and catalase production. Lactic acid bacteria (LAB) were enumerated on de Man, Rogosa and Sharp agar plates (MRS agar; Oxoid CM 361) while Enterobacteriaceae and total coliforms were counted respectively on Violet Red Bile Glucose Agar (VRBG; Oxoid CM 485) and Violet Red Bile Agar (VRBA; Oxoid CM 107) and later subcultured in Brillant Green Bile Broth (Oxoid CM 31) containing inverted Durham tube. Yeasts were enumerated on Malt Yeast Glucose Peptone (MYGP) agar medium with antibacterial agents added as described by Vieira-Dalodé et al. (2007). PCA and MRS agar plates were incubated at 30 °C for 2 - 3 days. Enterobacteriaceae and total coliforms were incubated at 37 °C for 1 day while yeasts were incubated at 25 °C for 7 days.

Statistical analysis

Comparison of means by Student t-test was performed using Minitab 14 (Minitab Sarl, Paris, France) software.

RESULTS AND DISCUSSION

Proximate analyses showed that Lafun was very poor in proteins, in lipids and in ash (Table 1). Its main component is starch which amount was 75.5% for Ordinary lafun and 77% for Chigan lafun (dry basis). However, the amount of proteins, lipids, ash and starch in lafun flour was low compared with unfermented cassava flour as determined by Ketiku and Oyenuga (1976), Longe (1980), Ihekoronye and Ngoddy (1985), Sylvestre (1987) and Padonou et al. (2005). The wastes that occur during retting and washings could explain this phenomenon (Oyewole and Odunfa, 1989; Oyewole and Afolami, 2001). Moorthy and Mathew (1998) also noted a decrease of proteins and lipids content during lafun processing. During cassava retting, the cell walls of cassava root were lysed releasing cells components especially starch grains and free sugars into the retting juice (Brauman et al., 1996). These carbohydrates were probably used for microbial activities.

The water content of the samples was variable but the mean values obtained were 15.1% for ordinary lafun and 13.8% for Chigan lafun (Table 1) In fact, the lafun sold in Benin markets is always exposed without any package in opened bowls. Since the collected samples were subjectted to the same conditions, the high level of water content could be explained by the environmental humidity in this part of Benin which is about 70 - 90% (Adam and Boko, 1983).

The water activity of the lafun samples probably affectted their microbiological characteristics. Microbial counts revealed the presence of an important population of micro-organisms in the Beninese Lafun (Table 2). The aerobic mesophilic counts for all the samples investiga-

Parameter	Ordinary lafun (n = 15)		Chigan lafun (n = 7)	
	Mean	cv (%)	Mean	cv (%)
рН	6.2 ^a	18.7	5.9 ^a	25.8
Titratable acidity (% lactic acid)	0.3 ^b	66.1	0.4 ^b	67.0
Water (% wb)	15.1°	14.4	13.8 ^c	17.5
Proteins (% db)	1.0 ^d	24.7	0.9 ^d	20.4
Lipids (% db)	0.3 ^e	76.3	0.5 ^e	60.4
Ash (% db)	1.4 ^f	43.3	1.0 ^f	25.4
Starch (% db)	75.5 ^g	6.5	77.0 ^g	6.3
Fibres (% db)	3.6 ^h	26.0	2.5 ⁱ	39.1

 Table 1. Chemical characteristics and nutritional composition of ordinary lafun and Chigan lafun.

Values with different letters (a, b, c,...) on a line are statistically different (P < 0.05).

Groups of micro- organisms	Ordinary lafun (n=15)	Chigan lafun (n=7)
Aerobic mesophilic count	$4.3 \times 10^5 - 8.9 \times 10^8$	$1.5 \times 10^4 - 9.1 \times 10^7$
<i>Bacillus</i> spp.	3.1 x 10 ⁶ – 5.5 x 10 ⁸	7.1 x 10 ⁴ – 9.5 x 10 ⁷
Lactic acid bacteria	$4.7 \times 10^4 - 5.3 \times 10^7$	3.6 x 10 ³ – 3.8 x 10 ⁵
Enterobacteriaceae	$1.1 \times 10^5 - 3.0 \times 10^7$	4.5 x 10 ³ – 1.9 x 10 ⁷
Total coliforms	$< 10^2 - 2.3 \times 10^5$	$< 10^2 - 1.4 \times 10^4$
Yeasts	2.5 x 10 ² – 5.3 x 10 ⁷	$3.1 \times 10^2 - 3.7 \times 10^5$

 Table 2. Microbial counts of ordinary lafun and Chigan lafun (cfu/g).

ted were variable and ranged from 10^4 to 10^8 cfu/g. *Bacillus* spp. were present at high levels from 10^4 to 10^8 cfu/g. They were Gram positive, catalase positive and had phase bright spores. Some species (*Bacillus cereus*) were reported to be pathogenic if present in foodstuff at levels of 10^5 - 10^7 cfu/g (Michelet et al., 2006). Lactic acid bac-teria, Enterobacteriaceae and yeasts counts ranged from 10^2 to 10^6 cfu/g. However, about 55% of all the analysed samples showed less than 10^2 cfu/g as total coliforms (data not shown) which were positive for gas production in Brillant Green Bile Broth (Oxoid CM 31).

According to Tsav-Wua et al. (2004), microbial counts of the traditional fermented cassava flour (Kpor Umilin) produced in Nigeria ranged from 2.7 x 10^3 to 1.2 x 10^7 cfu/g. ljabadeniyi (2007) also reported that the total viable counts of lafun sold in Oja Oba market (Nigeria) was about 1.5 x 10⁶ cfu/g. Lactic acid bacteria, *Bacillus* spp., yeasts and filamentous fungi were previously identified as micro-organisms that play important roles in the traditional fermentation of cassava for lafun production (Oyewole and Odunfa, 1988). Several authors (Oyewole and Odunfa, 1988; Amoa-Awua et al., 1996; Kostinek et al., 2005; Coulin et al., 2006) reported the role of lactic acid bacteria in the souring of cassava fermented products through the lactic acid production. Some Bacillus species have shown their ability to break down cassava tissue during the fermentation process (Amoa-Awua and Jakobsen, 1995; Obilie et al., 2003). Furthermore, AmoaAwua et al. (1997) demonstrated the contribution of some yeasts and filamentous fungi in the breakdown of cassava tuber tissue by cellulase production. The textural breakdown of cassava is assumed to allow a more intimate interaction between the indigenous linamarase and cyanogenic compounds of cassava. Lactic acid bacteria, yeasts and filamentous fungi contributed to cassava detoxification (Okafor and Ejiofor, 1986; Amoa-Awua et al., 1996, 1997). They are also involved in the build up of the aroma compounds during fermentation (Caplice and Fitzgerald, 1999; Oyewole, 2001). These advantages could explain the use of previously produced Lafun flour as 'back slopping' by the processors in the production area. However, attention should be paid to the microbiological safety of lafun.

At P = 0.05 level, Chigan lafun group was significantly different from ordinary lafun group for most of the physical characteristics (Table 3). But nearly all the chemical characteristics were similar, except the fibre content (Table 1). The significant lower fibre content of Chigan lafun comes probably from the fibre removal by processors during the thorough washing after retting and during the spreading out of the fermented cassava mash to dry. In addition, the ranges of microbial counts for the bacteria and yeast enumerated overlapped even if Chigan lafun counts were lower ten to hundred times (Table 2). It may suggest that apart from the physical characteristics, the other characteristics were not affected by the changes

Characteristics	Ordinary lafun (n = 15)		Chigan lafun (n = 7)	
	Mean	cv (%)	Mean	cv (%)
L*	89.9 ^a	2.2	92.8 ^b	0.7
ΔΕ	12.1 ^c	14.6	8.7 ^d	12.1
Pasting temperature ($^{\circ}$ C)	78 ^e	2.1	77 ^e	2.0
Solubility (mg/ml)	3.3 ^f	20.9	2.3 ^g	29.5
Swelling power (g water/g)	29.3 ^h	8.3	28.2 ^h	2.0
Peak of viscosity (RVU)	162.4 ⁱ	12.7	189.5 ^j	11.6
Final viscosity (RVU)	144 ^k	12.9	174 ¹	14.9

Table 3. Physical characteristics of ordinary lafun and Chigan lafun.

Values with different letters (a, b, c,...) on a line are statistically different (P < 0.05).

introduced in Chigan lafun technology. However, the washings, the retting condition and the drying system probably helped to reduce the microbial load of Chigan lafun.

Chigan lafun technology allowed to obtain a wither product. By colour parameters measurement it appeared that Chigan lafun was significantly (P < 0.05) more lightly and whiter than ordinary lafun. Moreover, solubility of Chigan lafun flour was significantly (P < 0.05) lower while its viscosities were higher. Probably, washing operations reduced the soluble matters content of Chigan lafun especially the free sugars which are said to lower the gel viscosity (Trémolières et al., 1977). For all samples (Table 3), solubility of the flour was ten times lower than the one of unfermented cassava flour (Padonou et al., 2005). This difference is probably due to the retting operation which allowed soluble matters to move from the roots to the retting water (Brauman et al., 1996). In contrast, pasting temperature, peak of viscosity and final viscosity values of lafun flour were higher than those found with unfermented cassava flour. Fermentation may be responsible of this difference as previously reported by Moorthy and Mathew (1998) for native starch. However, Mestres and Rouau (1997), in contrary, reported that the viscosity of hot dispersion of sour cassava starch decreased. It points at a need for more research to understand the pasting behaviour of fermented cassava derived products.

In conclusion, Chigan lafun differs fundamentally from ordinary lafun by the physical characteristics particularly its wither and more lightly colour. The colour characterristics depend mostly on washing, retting and drying conditions. However, there is a poor relationship between physical, chemical, microbiological of lafun and textural characteristics of the stiff porridge. Further studies are needed to establish the quality determining parameter of lafun.

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