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Research Article

THE IMPROVING THE CONSERVATION OF THE « ATTIEKE » BY SOLAR DRYING

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ABSTRACT

In Côte d'Ivoire, Cassava roots are mainly processed into *attiéké*, a kind of fermented cassava semolina, dried and steamed. The *attiéké* is the most consumed product from cassava transformation, but has a short preservation time. The main objective of this study is to extend the shelf life of *attiéké* by solar drying. In this work, it is necessary to dry fresh *attiéké* in an improved and hygienic dryer. The dried *attiéké* is then packed in polythene bags and stored at room temperature. Some biochemical, microbiological and organoleptic parameters were monitored during storage to assess the stability of dried *attiéké*. This results in an increase in the moisture content (8.5 ± 0.08 to $13.86 \pm 0.23\%$), protein (3.64 ± 0.24 to $4.60 \pm 0.17\%$) and acidity (67.70 ± 9.01 to 126.04 ± 1.80 méq/100g). Also, a decrease in carbohydrate (86.49 ± 0.83 to $80.00 \pm 0.33\%$), lipid (0.89 ± 0.01 to $0.83 \pm 0.02\%$), ash (0.71 ± 0.01 to $0.68 \pm 0.00\%$), and pH (6.84 ± 0.00 to 6.41 ± 0.02) was observed. Microbiological analyzes showed the presence of germs (GAM: 4.6.105, Yeast and mold: 2.0.105), but eliminated after reconstitution of dried *attiéké*. Organoleptic tests have shown that reconstituted *attiéké* has the same sensory characteristics as ordinary *attiéké* with the exception of the acidic parameter. Dried *attiéké* is less acidic than fresh *attiéké*. Finally the dehydrated *attiéké* can be kept for 42 days.

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INTRODUCTION

In Côte d'Ivoire, cassava is the second most important food crop for roots and tubers after yams, and is grown in almost all of Côte d'Ivoire (N'zué *et al.*, 2004, Kouadio *et al.*, 2010). It is a perishable commodity and, to limit its losses due to physiological deterioration 24 to 48 hours after harvest, the cassava root is transformed. It is transformed into placali (Koko *et al.*, 2012), gari (James *et al.*, 2013), attoukpou (Nevry *et al.*, 2007), and particularly *attiéké* (Assanvo *et al.*, 2002, Tetchi *et al.*, 2012; Krabi *et al.*, 2015; Yao *et al.*, 2015). *Attiéké* is the most fermented cassava product that is widely consumed (Kakou, 2000). It is also known in neighbouring countries (Aboua, 1995). *Attiéké* is a steamed granular cassava meal ready-to-eat, couscous-like product, with slightly sour taste and whitish colour (Djeni *et al.*, 2011). *Attiéké* is a typically Ivorian food which was originally prepared and consumed exclusively by ethnic groups living in the lagoon area such as Ebrié, Adjoukrou, Alladjan, Avikam. This food, a simple dish as only a product of self-consumption, *Attiéké* has now acquired the statute of annuity product, nourishing big urban markets. Thus, there has been a change from family production to commercial

production stimulated by the increasing urban demand and the formation of small cooperatives (Diop, 1992). Its moisture content is enough raised (40% f.m) and its preserving does not exceed one week. However *attiéké* is a meal much appreciated by the inhabitants of the Ivory Coast, particularly by the coastal population of the country for which it constitutes the staple food (Kouadio *et al.*, 1991). This dish has now conquered African, European, American and Asian countries (Kakou, 2000). The main objective of this study is to extend the shelf life of *attiéké* in order to ensure the food security of developing countries like Côte d'Ivoire, which are subject to famine.

MATERIAL AND METHODS

Material

Plant Material

The work presented in this manuscript was done on fresh *attiéké* (Figure 1)

Drying Equipment

Drying was carried out at room temperature in an improved dryer (Figure 2a and 2b)

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Table I Evolution of the biochemical characteristics of samples of dried attiéké during the Storage

Biochemical parameters	Storage time (day)			
	0	14	28	42
Titratable acidity (méq/100 g)	67.70 ± 9.01 ^a	81.24 ± 5.41 ^b	92.70 ± 4.77 ^c	126.04 ± 1.80 ^d
Ash (%)	0.71 ± 0.01 ^e	0.69 ± 0.01 ^e	0.69 ± 0.01 ^e	0.68 ± 0.00 ^e
Carbohydrate (%)	86.49 ± 0.83 ^f	82.68 ± 0.28 ^g	81.79 ± 0.75 ^g	80.00 ± 0.33 ^h
Humidity (%)	8.50 ± 0.08 ⁱ	11.80 ± 0.26 ^j	12.51 ± 0.11 ^k	13.86 ± 0.23 ^l
Lipid (%)	0.89 ± 0.01 ^m	0.87 ± 0.02 ^m	0.85 ± 0.01 ^m	0.83 ± 0.02 ^m
pH	6.84 ± 0.00 ^f	6.72 ± 0.00 ^g	6.49 ± 0.01 ⁱ	6.41 ± 0.02 ⁱⁱ
Protein (%)	3.64 ± 0.24 ^v	3.98 ± 0.14 ^v	4.51 ± 0.25 ^w	4.60 ± 0.17 ^w
Energetic value (Kcal/100 g)	368.53	354.47	352.85	345.87

Values with the same letters in the line are not significantly different at the 5% threshold according to the Tukey HSD test.

Table II Organoleptic characteristics of the rehydrated attiéké and the one bought on the market.

Paramètres	Rehydrated attiéké	Market attiéké
Acidity	1.66 ± 1.09 ^a	2.66 ± 0.70 ^b
Sweet taste	2.37 ± 0.76 ^c	2.08 ± 0.77 ^c
Firmness	2.41 ± 0.88 ^d	2.70 ± 0.90 ^d
Color	2.70 ± 1.04 ^f	2.83 ± 0.96 ^f
Global appreciation	2.87 ± 0.85 ^g	2.75 ± 0.53 ^g

Values with the same letters in the line are not significantly different at the 5% threshold according to the Tukey HSD test.

Table III Microorganisms counted (in CFU/g) in attiéké before drying, after 42 days of storage and after rehydration.

	Mesophilic aerobic bacteria	Total Coliforms	Fecal coliforms	Salmonella sp	ASR	B. cereus	S. Aureus	Yeasts & molds
Before drying	0	0	0	0	0	0	0	0
After 42 days of storage	4.6.10 ⁵	0	0	0	0	0	0	2.0.10 ⁵
After rehydration	0	0	0	0	0	0	0	0

ASR : Anaerobic Sulphito-Reducers
 B. Cereus : Bacillus Cereus
 S. Aureus : Staphylococcus Aureus
 CFU : Colony Forming Units



Figure 1 Fresh attiéké

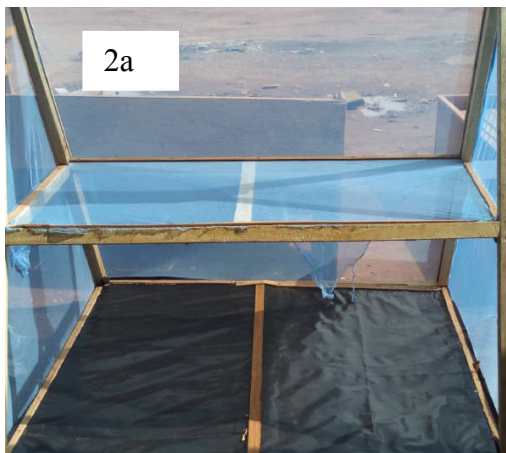


Figure 2 (2a and 2b) Improved dryer

Methods

Description of the Experiment and sampling

Drying Phase

Ten (10) kg of *attiéké* were taken just after cooking and packaged in plastic packaging at the production site of the village of Akoupé Zeudji (Anyama, Côte d'Ivoire). The *attiéké* batch was spread in an improved dryer to be sun-dried in the village of Allokoï (Anyama, Côte d'Ivoire). Every two (02) days approximately 20 g are taken for the determination of the moisture content. Indeed, the aim was to achieve a moisture content of less than 10% in order to maintain *attiéké*. After about five (5) days of drying the objective was reached with a moisture content of 8.5%.

Conservation Phase

After drying, the *attiéké* was packed in polyethylene bags and stored at room temperature for preservation. Thus, every two weeks, approximately 300 g are taken for biochemical analyzes for 42 days. Regarding the microbiological analyzes, they were carried out on the first day and the 42nd day of conservation. The organoleptic tests consisted of comparing the reconstituted *attiéké* and the ordinary *attiéké* bought on the market by hedonic test.

Biochemical and Organoleptic Analyzes of Attiéké

Titration of Titratable Acidity

The titratable acidity was determined according to the AOAC (2000) method. This assay consists in measuring the titratable acidity of a product with a solution of sodium hydroxide (NaOH), normality 0,1N, in the presence of phenolphthalein as color indicator. To do this, a mass P of 10 g of finely ground *attiéké* is diluted in 50 ml of distilled water, then allowed to macerate and the supernatant is filtered; 3 drops of phenolphthalein are added to a volume V = 10 ml of this filtrate and the determination is carried out by pouring the NaOH solution (0.1N) until the appearance of a pink color. Let V2 be the volume of the NaOH solution poured, the normality N1 of the filtrate taken is obtained by the formula:

$$N1 = \frac{N2 \times V2}{V1}$$

N1: Normality of the filtrate taken ; *N2*: Normality of the NaOH solution ; *V1*: volume of the dilution ; *V2*: volume of the NaOH solution poured.

This normality has been converted to milliequivalence per 100 g of fresh sample (meq / 100g) using the formula:

$$\text{Acidity (meq / 100g)} = \frac{N1 \times 10^5}{P}$$

P: Fresh sample mass

Determination of the Hydrogen Potential (pH)

pH was measured with a digital pH meter (Consort P107, Belgium). Ten (10) g of *attiéké* are ground in 50 ml of distilled water. The ground material is centrifuged at 3000 rpm for 30 min; the supernatant is collected in a jar. The pH is read on a digital display by directly immersing the pH meter electrode in the solution after calibrating the pH meter.

Determination of Moisture content and dry Matter

Humidity was determined by gravimetry after drying in an oven according to the method of **Favier (1977)**. Five (5) gram of *attiéké* crushed material is introduced into a pre - weighed nacelle. The nacelle is then located in the oven (Memmert), setting at 105°C for 24 hours. After cooling the capsule in a desiccator, the nacelles are new resources to the electronic scale (OHAUS analytic plus). The operation is resumed until a constant weight is obtained (within 0.2%). The experiment is done in triplicate and the average is determined. Thus, the moisture content was given by the following formula:

$$\% \text{Humidity} = \frac{PE - (Pf - Po)}{PE} \times 100$$

$$\% \text{Dry matter} = 100 - \% \text{Humidity}$$

PE = Test portion in gram ; *Po* = empty weight of the nacelles in gram ; *Pf* = final weight (nacelles + PE) in gram.

Assay Rate of Ash

The crude ash content is determined by gravimetry after furnace burning according to the method of **Favier (1977)**. In a previously calibrated crucible, 5 g of each sample are weighed. The crucible containing the sample is then introduced into the Nabertherm furnace. The temperature gradually increases to 550°C. Incineration is at 550°C for 3 hours (American Oil Chemists Society (AOCS), 1990). After cooling to the desiccator, the crucible is weighed again on the electronic scale. The operation is repeated until a constant weight of 0.5% is obtained.

The percentage of ash expressed as a percentage is given by the following formula:

$$\% \text{ash} = \frac{(P3 - P1)}{(P2 - P1)} \times 100$$

P1: Weight of the empty crucible ; *P2*: Empty crucible weight + test sample ; *P3*: Weight of empty crucible + ashes.

Determination of the Protein Content

The protein content is determined according to the Kjeldahl method for the determination of total nitrogen (American Oil Chemists Society (AOCS), 1990). In a Kjeldahl matrix, 0.1 g of attack is introduced, a catalyst pellet. Kjeldahl (3.5 g of K₂SO₄

and 0.4 of CuSO₄) as well as 10 ml of concentrated sulfuric acid. The mineralization is carried out on a heating block at a progressive temperature up to 500°C. It is followed for 3 to 5 hours until a clear (greenish) solution is obtained. The mineralizate is cooled and its volume is made up to 50 ml with distilled water before being neutralized with a solution of NaOH (10N) in the presence of phenolphthalein until the solution is brown. The neutralized solution is distilled and the ammonia solution is supplied in a 250 ml container containing 20 ml of boric acid (8 g of acid dissolved in 500 ml of distilled water + a few drops of helianthine and green bromocresol). The distillation is stopped when the final volume in the Erlenmeyer flask reaches 150 ml. Using a burette contains 0.1N sulfuric acid solution; the distillate is then titrated until the transfer of the indicator from green to pink. A blank is treated according to the same procedure, except that the sample is replaced by distilled water. The protein content is an application of the following formula:

$$\% \text{ Protein} = \frac{(Ve - Vb)}{PE} \times N \times 0,014 \times 6,25 \times 100$$

Ve: drop of the burette (sample) ; *Vb*: drop of the burette (white) ; *N*: normality of the sulfuric acid used for the determination (0,1N) ; *PE*: test portion ; 0.014: molar mass of nitrogen × 0.001 ; 6.25: conversion factor.

Determination of Fat Content

The determination of the fat content was made by the Soxhlet extraction method using hexane as the solvent. The balloons are washed beforehand and dried. The empty weight of the balloons (*Po*) is noted. Then 5 g of *attiéké* (*PE*) are introduced into the extraction cartridges, then corked with cotton and placed in the soxhlet. The balloons are filled with about 300 ml of hexane and then connected to the soxhlet. All connected to a refrigeration system and connected to a cryostat to condense the solvent vapors for driving the lipids. The extraction lasts 4 hours. Hexane is separated from lipids by rotavapor evaporation. The flasks are then dried in an oven at 105°C. After one hour in the oven, the flasks are cooled in a desiccator and then weighed. The gross fat content is determined according to the following formula:

$$\text{lipid content} = \frac{(Pf - Po)}{PE} \times 100$$

Po: mass of the empty balloon in gram ; *Pf*: mass of the balloon containing the lipids in grams ; *PE*: test portion.

Determination of Total Carbohydrate Content

The total carbohydrate content was determined according to the formulas described by Bertrand and Thomas (1910):

Total carbohydrates (%) = 100 - (% moisture +% protein +% lipid +% ash).

Determination of the Energy Value

The energetic value was determined by calculation, according to the Coleman (1970) formula using the Atwater and Rosa (1899) coefficients.

E (calories) = (4 x% proteins) + (4 x% total carbohydrates) + (9 x% lipids).

Sensory Analysis

Sensory analysis was performed according to the method proposed by Lateur *et al.* (2001). The organoleptic parameters of two types of *attiéké*, the one bought at the market and the reconstituted dried *attiéké* were evaluated. A scale of 1 to 5 was used to indicate: 5 = excellent, 4 = good, 3 = average, 2 = bad and 1 = very bad. Indeed, each panelist receives two or three samples of each type of *attiéké*. He judges, the sweet taste, the acidity, the firmness, the color of the *attiéké* while chewing for certain descriptors and by simply observing for others. It then assigns a note to the sample based on the descriptors.

Microbiological Analyzes

The microbiological analysis focused on the enumeration and identification of the bacteria usually sought to locate the microbiological quality of a food.

Ten (10) g of *attiéké* sample are slurried in 90 ml of sterile buffered peptone water, thus obtaining a dilution of 10^{-1} . Subsequent dilutions are then performed until the 10^{-8} dilution. The search and enumeration of the total aerobic germs are carried out on Plate Count Agar (PCA) medium. After homogenization and cooling, the Petri dishes are aerobically incubated for 72 hours at 30°C.

Total coliforms are counted by adding 1 ml of double-layer inoculum on Violet Red Bile Lactose (VRBL) agar medium incubated at 30°C for 24-48 hours.

Research and enumeration of staphylococci coagulase+ is performed on Baird-Parker base agar supplemented with Rabbit Plasma Fibrinogen (RPF) incubated at 37°C for 24-48 hours. Inoculation is performed on the surface by spreading 0.1 ml. Two petri dishes were seeded by dilution. A first reading took place after 24 hours and the second after 48 hours.

The search for *Bacillus cereus* (BC) consisted in inoculating on the surface Petri dishes containing the "MOSEL" medium. The incubation is carried out at 30°C for 24 to 48 hours. Identification uses different morphological and biochemical criteria such as mobility, NO_3^- reduction, and penicillin resistance.

Research and enumeration of *Clostridium perfringens* under anaerobic conditions was also performed on tryptic sulfite cycloserine agar (TSC). Incubation was at 46°C for 18-24 hours.

The search for yeasts and molds is done on sabouraud medium. The seeding technique used is that of spreading. About 12 ml of the supercooled chloramphenicol sabouraud agar is poured into the Petri dishes and then allowed to solidify. After solidification of the medium, 0.1 ml of diluted *attiéké* juice (10^{-1} and 10^{-2}) is spread with a rake on the medium. Once seeding is complete, the incubation is at 25°C for 72 h.

The search for *Salmonella* is done after pre-enrichment which consists of inoculation of 25 g of *attiéké* sample in 225 ml of buffered peptone water (EPT) incubated at 37°C for 16 to 20 h. This step is followed by enrichment of the culture broth obtained on two selective media, Rappaport Vassiliadis (RV) and selenite cystine (SC). 0.1 ml of the culture broth obtained after pre-enrichment are transferred into 10 ml of Rappaport

Vassiliadis (RV) medium. The medium is incubated at 37°C for 16 to 20 hours. In parallel, 2 ml of the culture broth obtained after pre-enrichment are added to 20 ml of Selenite Cystine Broth (SC). The medium is incubated at 42°C for 18 to 24 hours. Isolation consists of inoculation with the culture broth from the enrichment stage of two other different culture media, namely Chromogen *Salmonella* (OSCM) and Xylose Agar, Lysine and Deoxycholate (XLD). Both media are incubated at 37°C for 24-48 hours. A second enrichment is carried out on both Muller-Kauffmann medium and Rappaport Vassiliadis (RV) broth. The two media, inoculated with the broth obtained from the pre-enrichment, are incubated at 42°C for 20 to 24 hours. Isolation of strains is carried out on Hektoen medium incubated at 42°C for 20 to 24 hours. The identification of the different strains obtained is done on API *Salmonella*.

The number of sprouts is determined according to the following formula:

$$N = \frac{\sum C}{(V \times 0,1 \times d)}$$

N = total number of germs (CFU/g);

$\sum C$ = the sum of the colonies in the dilutions considered;

d = dilution rate considered;

v = volume of seeded inoculum.

Statistical Analysis

Statistical exploitation of the results was performed using the STATISCA 7.1 software. The one-way analysis of variance (ANOVA) was performed to compare variables analyzed on *attiéké* during conservation. The differences were considered significant for the values of $p \leq 0.05$. To separate the different samples, tests of multiple comparison (Tukey HSD) were conducted.

RESULTS AND DISCUSSION

Biochemical analyzes showed a significant increase in acidity in dried *attiéké* ($p \leq 0.05$) during its storage (Table I). The acidity values ranged from 67.70 ± 9.01 meq / 100g on day 0 to 126.04 ± 1.80 meq/100g on day 42. The microorganisms (yeasts and molds) identified in *attiéké* during drying are responsible for the increase of acidity in *attiéké* during its conservation. Indeed, these microorganisms by feeding on sugars and water, are full of energy to multiply. They produce metabolites that are among others acid, carbon dioxide, the alcohol responsible for the increase of the acidity (Lucile *et al.*, 2016). The evolution of the ash content is constant ($p \leq 0.05$) when storing dried *attiéké*. The values recorded in this study (0.71-0.68%) are consistent with those of Sotomey *et al.* (2001) found in the *Attiéké* of Benin a content of $0.68 \pm 0.09\%$. The Ivorian standard (CODINORM, 2006) requires values for mineral salts of less than 1.4% dry matter. The *attiéké* sample dried and stored has ash values below this standard. Ashes or total mineral salts represent the non-organic matter contained in the sample, obtained after incineration. They are dependent on the raw material and the value can be influenced by processing processes and good hygiene and manufacturing practices (dust contamination) (Guira, 2013). The total carbohydrate level decreases significantly ($p \leq 0.05$) when storing dried *attiéké*. Carbohydrate values ranged from $86.49 \pm 0.83\%$ on day 0 to

80.00 ± 0.33% on day 42. The Ivorian standard (CODINORM, 2006) does not specify the level of carbohydrate. Total carbohydrates but recommends a starch content of 75 to 80% dry matter in attiéké. Drying as a processing and preservation technology is also a process for concentrating certain substances such as sugars (Belem *et al.*, 2017); this could explain the high carbohydrate contents of dried attiéké. In addition, the reduction of the carbohydrate content during the conservation of dried attiéké is due to a reduction of the water content, thus favoring the development of a microflora (GAM, yeasts and molds). These microorganisms present in dried attiéké are responsible for the loss of total carbohydrates. Indeed, microorganisms transform starch into protein (Zouménou, 1994).

In addition, the moisture content increases significantly ($p \leq 0.05$) during the preservation of dried attiéké. Moisture values ranged from 8.50 ± 0.08% on day 0 to 13.86 ± 0.23% on day 42. This increase in moisture content would be due to a reabsorption of water by dried attiéké. Indeed, the moisture content of the atmosphere around the product being about 70% and that of attiéké packed by polyethylene being 8.5%, would explain this recovery of water by attiéké dehydrated. These observations are confirmed by Genin (1963) who showed that no plastic film is suitable for pack a food requiring significant barrier properties. He also showed that for many applications it is necessary to combine two or more different materials to obtain satisfactory barrier properties as per for example, protection against light, permeability to gases and water vapor. It is clear that dehydrated attiéké should be packaged in barrier water vapor or a combination of packaging (laminated packaging) to prevent recovery of water.

The fat content of dried attiéké remains constant ($p \leq 0.05$) during its conservation. These results are confirmed by those of Yao *et al.* (2015) who found a constant value of fat in the cassava cultivar pellets kept for 24 months. PH values ranged from 6.84 ± 0.00% (day 0) to 6.41 ± 0.02% (day 42), with an average value of about 6.6. This value is above the pH range admitted for the quality of attiéké as defined by the CODINORM NI 484 (2013) standard which is also between 4 and 5. Also, it is much higher than that obtained by Gnagne *et al.* (2016) for fresh attiéké (4,4) and for dried and reconstituted attiéké (5,6). However, she is identical to the pH value obtained by Yao *et al.* (2015) which is 6.55 for the attiéké obtained with dried native cassava pellets not preserved and kept for 6 to 24 months. This difference could be explained by the effect of the concentration of solutes by drying. By elsewhere, during storage, there is water reabsorption due to the use of polyethylene packaging not suitable for such a food. This reabsorption of water would explain the decrease in pH of dried attiéké during storage due to dilution of solutes. Protein content values range from 3.64 ± 0.24% on day 0 to 4.60 ± 0.17% on day 42. The average value of protein of dried attiéké is approximately 4.18. This content is much higher than that of the Ivorian standard for attiéké which defines a range of 1 to 2% of protein relative to the dry matter in attiéké. This increase in protein content in dried attiéké is due to the presence of microorganisms that would transform starch into protein and thus improve the content by about 20% (Zouménou, 1994).

In addition, the energy value decreases significantly ($p \leq 0.05$) during the preservation of dried attiéké. Values range from 368.53 kcal/100g on day 0 to 345.87 kcal/100g on day 42. This

decrease is due to the decrease in carbohydrate in dried attiéké during the conservation. In fact, attiéké is an energetic food, it is low in protein, in lipid and most of its calorific value comes from carbohydrates (Guira, 2013). The results of the hedonic tests showed that the attiéké dried and then rehydrated has the same organoleptic characteristics as the fresh attiéké purchased from a market in the exception of the acidity parameter (Table II). Dried attiéké is less acidic than that bought at market, this drop in acidity is due to the effect of drying. This is confirmed by the work of Yao *et al.* (2015) who obtained after reconstitution of native cassava pellets and cooking, products less sour than those prepared with fresh cassava. According to the same authors, drying removed 10% of residual HCN contained in the cassava paste after pressing. Microbiological examination assessed the level of contamination of attiéké dried by microorganisms (Table III). The high load of mesophilic aerobic germs would promote a strong alteration and constitute a risk for consumption (Kasse *et al.*, 2014). This alteration flora, consisting of the total aerobic mesophilic flora with the highest content ($4,6 \cdot 10^5$ ufc/g) would be of the same order of magnitude as $18,47 \cdot 10^4$ ufc/g reported by Fall *et al.* (2014), in Guedji, a condiment made from fermented and dried fish. To this one, would be added some yeasts and molds attached to the plant kingdom by their structure cellular. On the other hand, a reheating has eliminated all the flora of attiéké dried. The study indicated the absence of coliforms, Salmonella, Sulfito-Reducing agents, Bacillus cereus, and Staphylococcus aureus.

CONCLUSION

At the end of this work, the results indicate a decrease of the carbohydrate, the pH during conservation. Also, an increase in moisture, protein and acid. The values of the ash and lipid content remain constant during the conservation. The results of the hedonic tests showed a conservation of organoleptic characteristics except the acidity which is weak than that of the ordinary attiéké. Microbiological analyzes showed the presence of germs during conservation, but removed after reconstitution of attiéké. This work indicates that dried attiéké remains stable and fit for consumption beyond one month (42 days). It also shows that dried attiéké should be kept in barrier the water vapor or, a combination of packaging to prevent the water intake and increase thus the shelf life.

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