Production and Quality Assessment of "Ori-ese" (a fermented sorghum based food in Nigeria)

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Abstract: "Ori-ese" – a fermented sorghum based food was produced, the microbiological and physicochemical quality was investigated. The microbial isolate during steeping and slurry fermentation consisted of lactic acid bacteria (*Lactobacillus plantarum, Lactobacillus cellobiosus, Lactobacillus acidophilus, Lactobacillus dextranicum, Lactobacillus fermentum, Lactococcus lactis, subsp cremoris, Leuconostoc messenteroidis and Pediococcus acidulactis*), coliforms (*Escherichia coli and Enterobacter aerogenes*), other bacteria (*Proteus spp, Corynebacterium sp, Bacillus cereus and Bacillus subtilis*), molds species belonging to two genera (*Aspergillus and Penicillium*) and yeast were mainly species of *Saccharomyces* and *Candida*. Both lactic acid bacteria ($2.1 \times 10^5 - 3.3 \times 10^8$ cfu/g) and yeast ($5.0 \times 10^5 - 4.5 \times 10^6$ cfu/g) populations increased with fermentation time and reached the peak during fermentation of slurry ($3.9 \times 10^7 - 9.0 \times 10^8$ cfu/g) which resulted in total elimination of coliform and moulds. There was an increased in lactic acid production during steeping and slurry fermentation which ranged from 13.65 - 72.64mg and 72.64 - 101.69mg and a concomitant reduction in pH which ranged from 5.3 - 4.2 and 4.2 - 3.0 during steeping and slurry fermentation respectively. The crude protein, crude fiber, crude fat of "ori-ese" was found to be higher than that of sorghum grains. Remarkable increase was observed in Ca, K, Na and P and reduction in Fe content was observed

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1. INTRODUCTION

Fermentation of food substrates is a major source of nourishment for rural populations, and contributes significantly to food security by increasing the range of raw materials which can be used in the production of edible products and food varieties. Sorghum (Sorghum bicolor (L.) Moench) belonging to the family of Poaceae, is one of the most popular crops in Africa. Asia and Latin America (Anglani, 1983). Among the fermented foods, sorghum based products are largely being used in African countries. Above 35% of sorghum is grown directly for human consumption. The rest is used primarily for animal feeds, alcohol production and industrial products (FAO, 1995; Awka, 2004). Sorghum is one of the main staples foods for the world's poorest and most insecure people. It is a key staple in many parts of the developing world, especially in the drier and marginal areas of the semiarid tropics. In Nigeria, people consume sorghum in fermented form mainly as "ogi", "burukutu" and "pito" (brewed) from malted sorghum (Ekundayo, 1969). Fermentation of this staple raw material has been reported to contribute to enhanced nutritional value of the fermented products (Eka, 1980; Odunfa, 1995 and Sanni, 1989). However fermented staple foods in the sub-region are been prepared traditionally, therefore the fermentation process is usually done by natural microfloral which is spontaneous and uncontrolled. Therefore, there is no literature on production of "ori-ese", a fermented sorghum based food important to the people of Ekiti, South-Western Nigeria. Therefore, this work reports the traditional method of preparing the food, associated microorganism during fermentation and the physicochemical quality of the product.

2. MATERIALS AND METHODS 2.1 Production of "Ori –ese"

Sorghum (sorghum bicolor) grains brown variety was obtained from Uyo market, South Eastern Nigeria. Broken and moldy seeds were removed manually. 500gm of the grain were washed thoroughly in tap water during which the floating seeds were discarded. The washed grain were steeped in water for three days, wet milled, the slurry was allowed to ferment for 24hours after which it was boiled for 20 minutes. The resulting dough was molded into а ball shape, wrapped in Thaumatococcus daniellii Benth ("ewe iran") leave tied with rope and properly cooked for one hour. The resulting thick porridge is "Ori-ese."

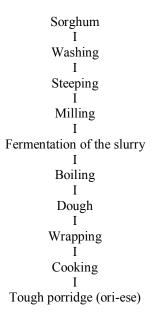


Fig. 1. Flow chart for traditional production of "ori-ese"

2.2 Microbiological and physicochemical analyses

At different stages of fermentation process. samples were collected (steeping water at 24, 48 and 72hrs) and during fermentation slurry at 0hr, 12hrs and 24hrs. Isolation was made from serial dilution of 1ml of steeping water and 1g of the fermented samples. The dilution were made using peptone water, after appropriate dilution, 0.1ml of the diluents were pour plated in triplicate plates on nutrient agar for total plate count, Mac Conkey agar for Coliform count, Salmonella / Shigella agar (SSA agar) for Salmonella/Shigella count, Sabouraud Dextrose agar (SDA) with Chloramphenicol (250mg/100ml) was used for fungi count and De Man Rogosa Sharpe medium (MRS) for lactic acid bacteria count while for yeast count the medium was adjusted to pH 3.5 with tartaric acid. All plate were incubated for 48hours at 30°C except for SDA which was incubated at 26°C for 6 days and MRS plates were incubated under anaerobic condition (BBL gas Pak Anaerobic system, Cockeyskonlk, USA). Colonial counts were made using digital illuminated colony counter (Gaalen Kamp Model), for yeast count the medium was adjusted to pH 3.5 with tartaric acid. Pure culture of each isolate obtained by streaking the specific colonies on suitable media and incubated appropriately, these were maintained in an agar slants in McCartney bottles. The isolated colonies were randomly picked, purified and the organism was identified following the Scheme of Sneath et al., (1986) based on morphological, physiochemical and biochemical characteristics. Sugar fermentation was done. The associated fungi were then identified with

reference to Fawole and Oso's Laboratory Manual (1988) while the yeast was identified using the method of Beech et al., (1986) and Lodder, (1970). The pH of the samples was determined using a pH meter (Titrimeter U9N model). The amount of lactic acid produced in the fermenting medium was determined by the titration procedure of the Spicher and Stephen (1982). Acid equivalent was the amount of NaOH consumed in ml, while each ml of NaOH is equivalent to 90.08mg of lactic acid. Proximate compositions were carried out according to the method of A.O.A.C (1990). This includes determination of pH, moisture content, ash content, crude protein, and fiber, fat, total carbohydrate contents and mineral content such as potassium, calcium, phosphorus and iron.

3. RESULTS

The total count of microbial group during steeping of sorghum and slurry fermentation for "oriese" production is shown in Table 1. The total microbial count at different stages of steeping of sorghum and slurry fermentation was in order of 10^2 to 10^8 cfu /ml. The lactic acid bacteria count ranged from 2.1 x $10^5 - 1.70$ x 10^6 cfu /ml and 3.9 x 10^8 to 9.0 x 10^8 during steeping and slurry fermentation respectively. The total plate count ranged from 5.0 x $10^5 - 7.5 \times 10^6$ cfu /ml and 1.6 x 10^3 to 1.0 x 10^2 cfu /ml during steeping and slurry fermentation. The coliform count ranged from 1.0×10^2 to 1.0×10^2 and $<10^{2}$ cfu /ml during steeping and slurry fermentation. There was reduction in coliform count after 72hrs of steeping after which they were not detected. The moulds count ranged from 4.0 x 10^2 to $<10^2$ during steeping and slurry fermentation. Salmonella / Shigella were not detected during steeping and slurry fermentation. Yeast count ranged from 5.0 x 10^4 to 4.5 x 10^6 cfu /ml during steeping and slurring fermentation.

The microflora found associated with sorghum grain fermentation during "ori ese" production consisted of Enterobacteria, lactic acid bacteria, moulds and veasts. Moulds and coliforms constitute very low proportions of the population. However as the fermentation progressed the lactic acid bacteria and yeast count increased and reach the peak at 72hrs steeping, and their number increased during slurry fermentation. Different microbial species encounter during fermentation of grain and slurring may be due to uncontrolled and spontaneous nature of the fermentation. Similar result was reported by Halm et al., (1993) during the steeping and fermentation stages of maize grain for "kenkey" production. The growth of LAB increased consistently throughout the fermentation period which gradually dominating the microflora with total

elimination of coliform and mould during slurry fermentation due to lactic acid production.

The identities of the associated microorganisms during steeping and slurry fermentation stages of "ori-ese" production are shown in Table 2. The isolates were lactic acid bacteria (Lactobacillus plantarum, Lactobacillus cellobiosus, Lactobacillus acidophilus, Lactobacillus dextranicum, Lactobacillus fermentum, Lactococcus lactis, subsp cremoris, Leuconostoc mesenteroidis Pediococcus acidulactic), Coliform and

Escherichia coli and Enterobacter aerogenes), Other *bacteria* such as *Proteus*, *Corynebacterium*, *Bacillus cereus*, *Bacillus subtilis*, *klebsiella* sp., Moulds species belonging to general (*Aspergillus* and *Penicillium*) and yeast were *Saccharomyces* and *Candida*. During steeping and slurry fermentation a succession in the growth of lactic acid bacteria was observed. *L. plantarum*, *L. fermentum* and *Leu*. *messenteroides* and *Lactococcus lactis subsp cremoris* were isolated after steeping for 48hrs.

Table 1- Total count of microbial groups during steeping of sorghum and slurry fermentation for "Ori-ese" production

Fermentation period	Lactic	acid	Salmonella	Coliform	Yeast	Total plate	Moulds/Yeast
(hr)	Bacteria	count	count (cfu/g)	count (cfu/g)	count	count (cfu/g)	count (cfu/g)
During steeping	(cfu/ml)				(cfu/g)		
0	2.1×10^5		-	$4.2 ext{ x10}^{3}$	5.0 x10 ⁴	7.5x10 ⁵	1.0x10 ⁵
24	5.6 x10 ⁶		-	$1.0 \text{ x} 10^2$	7.1 x10 ⁵	6.3 x 10 ⁵	$5.0 \text{ x} 10^3$
48	$2.50 \text{ x}10^7$		-	-	$4.5 ext{ x10}^{6}$	5.5x10 ⁵	$7.2 \text{ x}10^2$
72	3.3 x 10⁷		-	-		5.0x10 ⁵	$4.0 \text{ x}10^2$
During slurry	Lactic	acid	Salmonella	Coliform	Yeast	Total plate	Moulds/Yeast
fermentation (hr)	Bacteria	count	count (cfu/g)	count (cfu/g)	count	count (cfu/g)	count (cfu/g)
	(cfu/g)				(cfu/g)		
0							
12	6.0 x10 ⁸		-	-	$2.1 \text{ x} 10^7$	$1.1 \text{ x} 10^2$	
24	9.0 x10 ⁸		-	-	8.6 x10 ⁷	$1.0 \text{ x} 10^2$	

However *L. plantarum* had the highest frequency of occurrence and dominated until the end f the slurry fermentation. This result is in agreement with the report of Oyewole and Odufa (1990) during cassava fermentation for "laafun" production. During the fermentation *Aspergillus* and *Penicillium* species were recognized as contaminants because they do not play any important role during fermentation. The presence of moulds has been reported during fermentation of maize for "kenkey" production (Jespersen *et al.*, 1994). Changes in pH and lactic acid production may be due to metabolic activity of these microflora which created a favorable condition for lactic acid bacteria and yeast which can tolerate the acidic environment and in turn inhibit the growth of other organisms such as enteric organisms.

Table 2- Identities of isolated microorganisms during steeping of sorghum and slurry fermentation stages of	
"Ori-ese" production	

Fermentation time (hr)	Lactic acid bacteria	Coliform	Moulds	Yeast	Salmonella /Shigella	Other Bacteria
Steeping of sorghum	Pediococcus spp.	E.coli, E. aerogenes	Mucor spp. Aspergillus niger	Candida crusei, Candida tropicalis	-	Proteus sp Klebsiella oxytoca
24	L. plantarum	E. aerogenes	A. niger	Saccharomyces cerevisiae		Corynebacterium sp
48	L. lactis Susp cremoris L.acidophilu	-	Penicillium sp	Saccharomyces cerevisiae	-	-
72	L. plantarum, L. fermentum,L. mesenteroides		-	Saccharomyces cerevisiae	-	-
Slurry fermentation 0	L. plantarum, L.brevis, L. acidophilus	-	-	Saccharomyces cerevisiae	-	B. subtilis
12	L.cellobiosus L.plantarum	-	-	Saccharomyces pombe, Saccharomyces cerevisiae	-	B. subtilis
24	L.plantarum, L. acidophilus	-	-	Saccharomyces cerevisiae, Saccharomyces sp	-	B. subtilis

Inhibition of Coliforms or *Enterobacteriacea* by species of lactic acid bacteria was in agreement with the work of Lonner *et al.*, (1986). The reason for this may be due to the fact that lactic acid bacteria and yeast outnumber the *Enterobacteriaceae* which resulted in faster acid production. Therefore in contrast to finding for other indigenous fermented foods (Adegoke and Babalola, 1988) other workers have also reported the inhibition of different strains of entero - pathogens in lactic fermented cereal gruel (Kingamokono *et al.*, 1995 and Svanberg *et al.*, 1992).

Table 3 shows the physicochemical changes during steeping of sorghum and slurry fermentation for "Oriese" production. During steeping there was a gradual reduction in the pH from 5.3(0h) to 4.2 (72hrs) which resulted in an increase in acid and lactic acid equivalent at the same interval from 0.15 to 0.8ml and 13.63 to 72.64mg (Table 3). During the fermentation of the slurry (24hrs) after milling, there was a gradual reduction in pH from 4.2 (0h) to 3.8(24hrs) and a concomitant increased in lactic acid development from 72.64 to 101.69mg.

The proximate composition of "Ori ese" and sorghum is shown in Table 4. From the result some changes in the level of nutrient and mineral content during fermentation was observed. The percentage protein content, fat and crude fiber contents ranged from 11.80 - 14.17%, 3.20 - 4.31% and 1.10 - 3.86% respectively in which "Ori-ese" samples had the highest. The percentage carbohydrate content ranged from 72.33 - 82.59% in which sorghum samples had the highest.

Table 5 shows the mineral composition of the "Ori-ese". The percentage Na, K, Fe, Ca and P content ranged from 49.20 - 51.6 6mg/100ml, 50.90 - 86.40mg/100ml, 1.72 - 5.32mg/100ml, 24.40 - 65.87mg/100ml and 350 - 362.68mg/100ml respectively. "Ori-ese" had the highest mineral composition except for Fe in which the highest was recorded in sorghum samples.

Fermentation time(hr)	pH	Acid equivalent(ml)		Lactic acid(mg)		
	During steeping of sorghum					
		\A	В	А	В	
0	5.3	0.15	0.12	13.65	13.66	
24	4.8	0.65	0.65	59.02	59.04	
48	4.3	0.70	0.72	63.56	63.57	
72	4.2	0.80	0.82	72.64	72.63	
During fermentation of slurry						
0	4.2	0.80		72.64	72.66	
12	3.8	0.95		85.98	85.99	
24	3.0	1.12		101.69	102.67	

Table 3. Physicochemical changes during steeping a d slurry fermentation for "ori ese" production

Table 4. Proximate composition of sorghum grain and "ori -ese"

	P	Proximate composition (%)			
Parameters	Sorgh	Sorghum grains		-ese"	
	Α	В	Α	В	
Moisture	12.1	12.3	91.62	91.60	
Ash	1.57	1.58	5.35	5.34	
Fiber	1.10	1.11	3.85	3.86	
Carbohydrate	82.62	82.59	72.33	72.33	
Fat	3.21	3.20	4.31	4.30	
Protein	11.50	11.52	14.16	14.17	

Table 5. Mineral composition of sorghum grain and "ori –ese"

Mineral composition(mg/100g)						
Na	49.20	49.22	51.66	51.60		
K	50.90	50.89	86.40	86.40		
Fe	5.32	5.32	1.78	1.72		
Ca	24.60	24.40	65.87	65.86		
Р	350	352	362.67	362.68		

Some changes in the level of nutrient and mineral contents during fermentation were observed which indicate that the biological value of "ori ese" was superior to that of the raw sorghum grain. There was an increase in crude protein content, this may be due to the metabolic effect of the microflora on the substrate during fermentation and the fact that protein in sorghum grain which are located in the testa and germ were retain during processing since "ori ese" production did not involve sifting which can result in loss of protein unlike "ogi" in which its traditional processing leads to losses of nutrients especially protein (Banigo and Muller, 1972). Protein is a limiting factor in the diet of the people in under developed and developed areas of the world. It indicates that consumption of this product would significantly increase the protein intake of the consumer. Similar result was obtained by Graham et al., (1986) during the production of ".nasha". There was an increase in crude fiber, crude fat, ash content during fermentation of sorghum grain for "ori ese" production. Remarkable increase was obtained in Ca, K, Na and P content where there was reduction in Fe content. Increase in mineral content confirmed the fact that fermented foods contribute a product of microbial decomposition resulting in the mineralization of the higher organic compounds to release these elements.

In conclusion fermentation of sorghum grain for "ori ese" production improve the nutritive quality of the product. In addition to lactic acid bacteria, yeasts were very important in sorghum fermentation for "ori ese" production as it has been reported for other indigenous food. Further research should be directed towards improved method of production and development of starter cultures for commercial production of "ori ese"

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