MICROBIOLOGICAL PROPERTIES AND SPOILAGE INDICES OF NIGERIAN FERMENTED CEREAL GRUEL (SOY OGI) DURING STORAGE

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ABSTRACT: The microbiological properties changes and nutritional qualities of Soy ogi were evaluated during storage at room (28 $\pm 2^{o}$C) and refrigeration temperatures for 7 days to assess its shelf life. The result have indicated that the Total Heterotrophic Bacterial Count for refrigerated samples ranged from 3.2 $x10^3$ to 6.5 $x10^3$ cfu/g while Fungal Count ranged from 2.7 $x10^3$ to 5.9 $x10^3$cfu/g. Undecanted soy ogi samples stored at room temperature gave THBC values of 3.2 $x10^3$ - 8.4 $x10^3$ cfu/g and FC of 2.7 $x10^3$ - 1.0 $x10^4$ cfu/g, while the decanted samples stored at room temperature had THBC of 3.2 $x10^3$ to 7.4 $x10^3$ cfu/g and FC ranging from 2.7 $x10^3$ to 1.0 $x10^4$cfu/g. Changes in Total Coliform Count and Lactic Acid Bacterial Count were also noticed. The fermentation of the cereal gruel was carried out with diverse species of microorganisms but more predominated by *Lactobacillus* species (33.3%), *Streptococcus lactis* (30.6%) *and Saccharomyces* (38.7%). Their activities affected the pH, total titratable acidity, ammonia level, total reducing sugars, and colour of the stored cereal gruel. The changes (spoilage indices) were remarkable in samples stored at room temperature than refrigeration temperature. Therefore, storage with proper refrigeration is highly recommended due to its ability to retard fermentation, thereby preserving the quality of Soy ogi.

INTRODUCTION

In developing countries, the incorporation of high protein foods especially of plant origin into our diet is very necessary to combat malnutrition and the high cost of animal protein (Aminigo and Akingbala, 2004). Various plant proteins may be combined to obtain products with improve protein quality. Ogi or pap is a local name for a semi solid food made from cereals [commonly maize (*Zea mays*), sorghum (*Sorghum bicolor*) or millet (*Pennisetum glaucum*)]. It is commonly used as weaning food for babies and young children, as a standard breakfast cereal in many homes and food for invalids. Apart from human consumption, it also serves as a quick paper glue and free-range chicken feed supplement (Afolayan *et al,* 2010). The cereal consists of 12 to 14% water and ogi generally has been implicated in Kwashiokor among infants (Akanbi *et al.*, 2003). In most parts of West Africa, children are fed with mashed adult food of gelatinized cereal flavour slurries to complement breast milk from 4 to 6 months of age. These slurries absorb large quantities of water and swell up greatly when mixed with hot water. The foods are therefore bulky due to high viscosity. In addition, the protein content of maize is of poor quality, low in lysine, methionine and tryptophan, which are indispensables to the growth of young children (Odunfa *et al.*, 1994).

Efforts to improve the nutritional status of these staples have been based on fortification with legumes to provide the deficient amino acids (Osundahunsi and Aworh, 2003). Various attempts that have been made for nutrient restoration and fortification of ogi include combination with Soy flour, blends of roasted soybean and peanut meals (Aminigo and Ossai, 1999, Akanbi *et al.*, 2010) blends of cashew nut, African locust bean, sesame oil meals and okra seed meal (Aminigo and Akingbala, 2004). Though still largely regarded as a relatively new crop, soybean (*Glycine max*) has been successfully included in the diet of many Nigerians.

Soybean derivatives such as Soy garri, soy milk, soy ogi, soy-lafun etc have been developed and found to be acceptable to most Nigerians.

Storage or shelf life of Soy ogi is often less than ten days except when adequately refrigerated. Refrigeration is a good option in environments where electricity is available all the time and the family can afford the bill. Refrigeration slows fermentation and preserves the nutritional and physical properties of the soy ogi. In the rural areas of developing countries where access to electricity is either absent or inadequate the soy ogi is preserved for a few days by decanting the sour water and replacing with fresh water.
This study investigates the microbiological burden and changes in nutritional properties of decanted and undecanted soy ogi stored at room temperature as well as in refrigerator to assess its shelf life.

MATERIALS AND METHODS

Source of grain and Preparation of Soy ogi

Maize and soybean grains were purchased from the market in Uyo metropolis. The method of Adeyemi and Balogh (1985) was adopted for the processing of ogi (Fig I). Cleaned maize and dehulled steamed soybeans were separately steeped in water for 3 days for fermentation to take place. The steep water was drained, the grains washed thoroughly and rinsed in water. Thereafter, 70 parts of the maize were mixed with 30 parts of soybean and ground to paste using a grinder (model 6Xi Nissan). The paste was wet sieved with mushin cloth and allowed to settle in a plastic container to form a smooth slurry (soy ogi). The supernatant was decanted, the sediment transferred to a clean cheese cloth and squeezed to remove excess water.

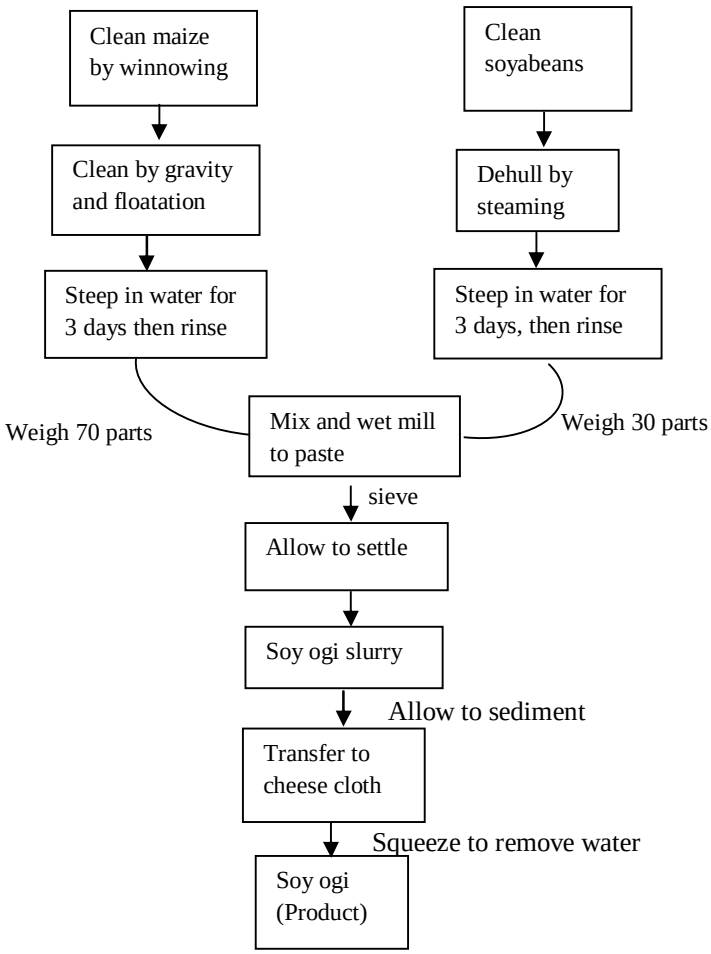

Figure I: Flow chart commonly employed in traditional soy ogi production in Nigeria (Adeyemi and Balogh, 1985.

Storage of Processed Soy ogi

Ten grams of the soy ogi samples were each placed in a sterile plastic container and 50ml of sterile water was introduced to cover the samples. The samples were prepared in triplicates and the experiment lasted seven days giving a total of 72 experimental samples. There were three treatments (namely room temperature (28 ± 2^oC) with decanting, room temperature without decanting and refrigeration temperature (4 to 8^oC).

Microbial Analysis of Processed Soy ogi

Microbial counts were made on selective media after decimal dilution of the samples using the pour plate method. Total heterotrophic bacterial count was determined on Nutrient agar, total coliform count on MacConkey agar, faecal coliform count on Eosin Methylene Blue agar, lactic acid bacterial count on Man Rogosa Sharpe (MRS) agar and total fungal count on Sabouraud's Dextrose agar containing 100ml streptomycin. Colonies which developed were counted and expressed as colony forming units (cfu/g or 100g). Plates for bacterial counts were incubated at 37^oC for 24 to 48 hours while fungal plates were incubated at room temperature (28 ± 2^oC) for 5 to 7 days. MRS plates were incubated in anaerobic jars using BTL gas pak.

Determination of Changes in Nutritional Attributes of Storage Soy ogi

The parameters used to monitor spoilage during storage of soy ogi as described by Teniola and Odunfa (2002) were pH, total titratable acidity, total reducing sugars, ammonia level and colour change. pH was determined using pH meter (Griffin, England). Titratable acidity was measured by titrating a mixture of 3g of soy-ogi slurry and 27 ml of distilled water to pH 8.5 using 0.1M sodium hydroxide solution (Kingamkono *et al.,* 1994). The result was expressed as g lactic acid/100g sample.

Sensory Evaluation

Sensory evaluation of the samples was carried out using a semi-trained panel consisting of ogi porridge consumers who were familiar with ogi porridge quality. Selection was based on interest and availability. All the samples were rated acceptable based on a 9 point scale (with 9 = highly acceptable and 1 = highly unacceptable.

RESULTS

The microbial load of soy ogi stored at refrigerated temperature is on Table I. The results revealed that the total heterotrophic bacterial count (THBC) of the fermented cereal gruel ranged from 3.2 $x10^3$ cfu/g initially to 6.5 $x10^3$cfu/g after seven days of fermentation. Total coliform count (TCC) ranged from an initial 2.8 $x10^3$cfu/g to 5.2 $x10^3$cfu/g, lactic acid bacterial count (LABC) increased from 2.9 $x10^3$cfu/100g to 5.5 $x10^3$cfu/100g, while the total fungal count (FC) increased from 2.7 $x10^3$cfu/g to 5.9 $x10^3$cfu/g. For undecanted soy ogi stored at room temperature, the results revealed that THBC increased from 3.2 $x10^3$cfu/g to 1.6 $x10^4$cfu/g on day 4, followed by decrease in the density of heterotrophic bacteria in the course of fermentation. Similar trends were observed for TCC and LABC. However fungal growth was progressive throughout the period (Table 2).

Table 3 shows that decanting the cereal gruel before storage at room temperature influences the microbial activities therein. There was increase in THBC from Day 1 to Day 5, while a decline in bacterial load was noticed on the 5^{th} day of fermentation. Similarly the total coliform count increased from 2.8 $x10^3$ cfu/g to 9.5 $x10^3$cfu/g on Day 4 before decreasing to 8.2 $x10^3$cfu/g on the 7^{th} day. Similar trends were observed for the lactic acid bacterial count, while the total fungal load increased throughout the period of fermentation. The fermentation process generally retard the proliferation of feacal coliforms, as no viable cells of *Escherichia coli* was detected in all fermented samples after storage in 24 hours. Table 4 shows the prevalence of diverse species of microorganisms isolated from fermented soy ogi. The dominant isolates were *Streptococcus lactis* (30.6%), *Lactobacillus* sp (33.3%) and *Saccharomyces* (38.7%). The activities of the microorganisms affected the pH, total titratable acidity, ammonia, total reducing sugar levels of the fermented cereal gruel. It also affected the colour and general

acceptability of the fermented soy ogi (Table 5). For refrigerated samples, there was a decrease in pH from 5.22 to 5.19 after 7 days. Similar observation was recorded for samples stored at room temperature. There was an increase in the total titratable acidity of the refrigerated samples, while the total reducing sugars initially increased but decreased with increase in the duration of storage. The ammonia levels of the refrigerated samples and the decanted samples stored at room temperature increased throughout the storage period. However, for undecanted samples stored at room temperature, the ammonia level decreased after 5 days of storage. In the refrigerated samples, there was no colour change whereas in the samples stored at room temperature without decanting, colour change was observed from the 4^{th} day of storage. The refrigerated samples and decanted samples stored at room temperature were acceptable up to the 7^{th} day, whereas the acceptability of the undecanted samples stored at room temperature was low after 4 days of storage.

Table 1: Microbial load (cfu/g) of Soy ogi stored at refrigeration temperature

Days	THBC	TCC	FCC	LABC	FC
0	3.2×10^3	2.8×10^3	2.6×10^3	2.9×10^3	2.7×10^3
1	3.5×10^3	3.1×10^3	-	3.2×10^3	3.3×10^3
2	3.9×10^3	3.5×10^3	-	3.6×10^3	3.5×10^3
3	4.1×10^3	3.8×10^3	-	4.0×10^3	3.9×10^3
4	4.5×10^3	4.1×10^3	-	4.1×10^3	4.4×10^3
5	5.7×10^3	4.5×10^3	-	4.9×10^3	4.9×10^3
6	5.8×10^3	4.9×10^3	-	5.1×10^3	5.2×10^3
7	6.5×10^3	5.2×10^3	-	5.5×10^3	5.9×10^3

THBC= Total heterotrophic bacterial count, TCC= Total coliform count, FCC= Feceal coliform count, LABC= Lactic acid bacteria count, FC= Fungal count, - = not detected.

Table 2: Microbial load (cfu/g) of undecanted Soy ogi stored at room temperature

Days	THBC	TCC	FCC	LABC	FC
0	3.2×10^3	2.8×10^3	2.6×10^3	2.9×10^3	2.7×10^3
1	8.8×10^3	5.2×10^3	-	6.8×10^3	6.7×10^3
2	1.0×10^4	5.7×10^3	-	7.7×10^3	7.2×10^3
3	1.3×10^4	7.5×10^3	-	8.2×10^3	7.9×10^3
4	1.6×10^4	9.5×10^3	-	8.7×10^3	8.2×10^3
5	1.0×10^4	9.2×10^3	-	8.2×10^3	8.7×10^3
6	9.2×10^3	8.2×10^3	-	7.8×10^3	9.2×10^3
7	8.4×10^3	8.2×10^3	-	7.2×10^3	1.0×10^4

THBC= Total heterotrophic bacterial count, TCC= Total coliform count, FCC= Feceal coliform count, LABC= Lactic acid bacteria count, FC= Fungal count, - = not detected.

Table 3: Microbial load (cfu/g) of decanted Soy ogi stored at room temperature

Days	THBC	TCC	FCC	LABC	FC
0	3.2×10^3	2.8×10^3	2.6×10^3	2.9×10^3	2.7×10^3
1	8.8×10^3	5.2×10^3	-	6.8×10^3	6.7×10^3
2	1.0×10^4	5.7×10^3	-	7.7×10^3	7.2×10^3
3	1.3×10^4	7.5×10^3	-	8.2×10^3	7.9×10^3
4	1.6×10^4	9.5×10^3	-	8.7×10^3	8.2×10^3
5	1.0×10^4	9.2×10^3	-	8.2×10^3	8.7×10^3
6	9.2×10^3	8.2×10^3	-	7.8×10^3	9.2×10^3
7	7.4×10^3	8.2×10^3	-	7.2×10^3	1.0×10^4

THBC= Total heterotrophic bacterial count, TCC= Total coliform count, FCC= Feceal coliform count, LABC= Lactic acid bacteria count, FC= Fungal count, - = not detected.

Table 4: Prevalence of Microorganisms associated with fermented cereal gruel during storage

Isolates	**Frequency**	**Percentage occurrence (%)**
Bacteria		
Leuconostoc spp	3	8.3
Staphylococcus aureus	2	5.6
Lactobacillus spp	12	33.3
Streptococcus lactics	11	30.6
Enterobacter spp	7	19.4
Corynebacterium spp	1	2.8
Fungi		
Verticilium spp	6	19.4
Aspergillus spp	7	22.6
Cladosporium spp	4	12.9
Penicillium spp	2	6.4
Saccharomyces spp	12	38.7

DISCUSSION

The fermentation of corn kernel attracted diverse microflora of bacteria, yeasts and molds. This is however a general feature of fermentation of plant materials whereby diverse microflora contribute their activities in increasing values of some vital mineral elements useful to the body (Oboh *et al.,* 2002), reducing anti-nutrient substances harmful to the body system (Achinewhu *et al.,* 1998) and adding aroma, taste and texture for desired and acceptable product. Akanbi *et al* (2010), investigating the quality of selected cereal soybean mixtures during 'ogi' production revealed the presence of *Bacillus, Staphylococcus, Enterobacter, Pseudomonas, Citrobacter, Corynebacterium* and *Micrococcus.* In the present study, *Lactobacillus* sp*, Streptococcus lactis* and *Saccharomyces* sp were found to be the dominant organisms associated with the process. *Lactobacillus* sp has been implicated in several reports as the most important/dominant microorganism involved in fermentation of maize during 'akamu' production (Amusa *et al,* 2005). According to Ijabadenyi (2007), the presence of *Lactobacillus* makes ogi a good source of bacteriocins. Other workers (Chukwuemeka, *et al,* 2006, Olasupo *et al.,* 1997) also identified *Lactobacillus* isolates as important microflora of African fermented foods. Bacteria such as *Bacillus* and *Staphylococcus,* isolated from the fermented cereal gruel are of public health importance. The presence of these microbes indicates possible contamination resulting from handling and the processing environment. The presence of *Bacillus* and *Pseudomonas* is probably responsible for rancidity and ropiness associated with fermented ogi (Amusa *et al,* 2005).

Osungbara (2009) reported that during the fermentation of ogi surface microflora of fermenting maize are eliminated within 6 hours of steeping. Others mainly yeasts (*Candida, Saccharomyces* and *Rhodotorula*) are actively involved in the fermentation process, results of the present study have shown fermentation as a process that can effectively retard or eliminate *Escherichia coli* in cereal gruel. The presence of moulds such as *Aspergillus niger, Penicillium* sp, *Rhizopus* sp on the surface of raw maize grains and during the early stages of fermentation has been reported (Omemu *et al,* 2007). They are most likely part of the grain surface microflora that are undesirable in many foods because of their ability to produce mycotoxins such as aflatoxin B1, fumonisin B1, ochratoxin A, trichothecenes and zearalenone, which are classified as possible carcinogens to humans (Vainio *et al.,* 1993). They are also implicated in the spoilage of stored wet ogi (Onyekwere *et al.,* 1989). The observed discoloration of the stored soy ogi coincided with growth of moulds in the samples.

Table 5: Changes in some nutritional attributes (spoilage indices) of soy ogi stored under room and refrigerated temperatures

Fermentation period	Spoilage indices (mg/100ml)																	
	Refrigerated gruel						Undecanted gruel stored at room temperature						Decanted gruel stored at room temperature					
	pH	TTA	AL	TRS	CL	OA	pH	TTA	AL	TRS	CL	OA	pH	TTA	AL	TRS	CL	OA
1	5.22	0.53	0.02	35.09	-	9.2	5.25	1.04	0.42	38.37	-	9.0	5.26	0.60	0.25	31.57	-	9.1
4	5.21	1.54	0.42	32.57	-	8.5	5.15	1.64	0.34	7.82	+	5.2	5.15	0.77	0.34	30.22	-	8.2
5	5.20	1.87	0.55	31.73	-	7.8	5.16	1.84	0.31	0.79	++	4.3	5.14	0.89	0.36	23.86	-	7.3
7	5.19	2.38	0.75	29.22	-	7.1	5.21	1.24	0.27	0.11	+++	4.0	5.10	1.07	0.41	9.51	+	5.1

Key: - No colour change, + colour change, TTA= Total titratable acidity, AL= Ammonia level, TRS= Total Reducing Sugars, CL= Colour change, OA= Overall Acceptability

Supplementation with soybean in the production of soy ogi increased microbial population, protein and lactic acid contents (Akanbi *et al.,* 2003). Soybean is a rich medium containing proteins, vitamins and importantly fermentable sugars such as sucrose and oligosaccharides, raffinose and stachytose. In the present study, there was a general increase in population of microbial types as storage period progressed.

Major fermentation products include organic acids such as lactic acid, acetic acid, propionic and butyric acids (Gabriel and Akharaiyi, 2007). Similar decline in pH from 6.3 to 3.16 was reported by Egounlety and Syarief (1992) during the studies of ogi supplemented with tempe. Owusu-Kwarteng *et al.,* (2010) observed that free fatty acids from soybeans could contribute to total acids (expressed as lactic acid) during soy-cereal fortification. This possibly explained the increase in acidity of the soybean fortified cereal gruel during their investigation. In this study, the pH decreased as storage period increased. The level of titratable acidity of soy ogi observed under different storage conditions revealed a gradual increase in total titratable acidity. Similar observation was reported by Egounlety and Syarief (1992) with an enhanced acidity from an initial value of 0.0685% to 0.097% during the production of ogi supplemented with tempe. The total reducing sugars decreased with increasing days of storage. Under room temperature with decanting, the sharp decline in total reducing sugars and increased discoloration observed agrees with the report that when the sour water is not changed, the shelf life of wet ogi is less than 7 days at room temperature (Olasupo *et al.,* 1997). The change in colour at 6^{th} day of storage and a sharp decline in total reducing sugars at 7^{th} day of storage observed in decanted samples stored under room temperature agrees with the report that decanting the sour steep water results in loss of nutrients (Aremu, 1993). However, no significant changes were observed in samples under refrigeration.

CONCLUSION

From this study, there is ample evidence to show that storage without refrigeration could deteriorate the quality of soy ogi. Therefore, storage with proper refrigeration is highly recommended due to its ability to retard fermentation thereby preserving the quality of soy ogi.

REFERENCES

Achinewhu, S.C., Barber, L. I. and Ijeoma, I.O. (1998). Physiochemical properties and gavification (gari yield) of selected cassava cultivars in Rivers State, Nigeria. *Plant Foods Hum. Nutr*., 52: 133-140.

Adeyemi, I.A. and Balogh, E., (1985). Biotechnology in food processing prospects in Nigeria. *Nigerian Food Journal*, 2: 33-39.

Afolayan, M.O., Afolayan, M. and Abuah, J.N. (2010). An investigation into sorghum based ogi (Ogi-Baba) storage characteristics. *Advanced Journal of Food Science and Technology,* 2(1): 72-78.

Akanbi, C.T., Ade-Omowaye, B.I., Ojo, A. and Adeyemi, I.A., (2003). Effect of processing factors on rheological properties of Ogi. *International Journal of Food Properties*, 6(3): 405-418.

Akanbi, W.B., Togun, A.O., Adediran, J.A. and Ilupeju, E.A.O., (2010). Growth, dry matter and fruit yield components of okra under organic and inorganic sources of nutrients. *American- Eurasian Journal of Sustainable Agriculture*, 4(1): 1-13.

Aminigo, E. R. and Akingbala J.O. (2004). Nutritive composition and sensory properties of ogi fortified with okra seed meal. *J. Appl. Sci. Envir. Managt.,* 8 (2): 23 –28.

Aminigo, E.R and Ossai, G.E.A., (1999), Production and evaluation of high-protein roasted maize meal. *J. Appl. Sci. Environ. Mgt.,* 3: 17 – 22.

Amusa, N.A, Ashaye, O. A and Oladapo, M. O. (2005). Microbiological quality of ogi and soy-ogi (A Nigerian fermented Cereal Porridge) widely consumed and notable weaning food in Southern Nigeria. *Journal of Agriculture, Food and Environment*, 3: 81-83.

Aremu, C.Y. (1993). Nutrient composition of corn *ogi* prepared by a slightly modified traditional technique. *Food Chem.*, 46: 231-233.

Chukwuemeka, U.N, Anyanwu B.N and Ibekwu, V.I, (2006). Nutritional, microbiological and organoleptic qualities of maize-based pap 'akamu' enriched with odudu (*Sphenostylis sternocarpa*). *Nig. J. of Microbiology*, 20(3): 1453-1459.

Egounlety, M. and Syarief, R., (1992). Study on supplementation of ogi with *Tempe*. *Nigerian Food Journal*, 10: 92-102.

Gabriel, R.A.O. and Akharaiyi, F.C., (2007). Effect of spontaneous fermentation on the chemical composition of thermally treated jack beans (*Canavalia ensiformis* L.). *Int. J. Biol. Chem.*, 1: 91-97

Ijabadenyi, A.O., (2007). Microorganisms associated with Ogi traditionally produced from three varieties of maize. *Research Journal of Microbiology*, 2(3): 247-253

Kingamkono, R., Sjogren, E., Suvanberg, U. and Kaijser, B. (1994). pH and Acidity in lactic–fermenting cereal gruels: Effects on viability of Enteropathogenic microorganisms. *World Journal of Microbiology and Biotechnology***,** 10: 664- 669.

Oboh, G., Akindahunsi, A.A. and Oshodi, A.A. (2002). Nutrient and anti-nutrient contents of *Aspergillus niger* fermented cassava products (flour and garri). *J. food Comp. Anat.*, 15: 617-622.

Odunfa, S.A. Nordstrom, J. and Adeniran, S.A., (1994). *Development of Starter Cultures for Nutrient Enrichment of Ogi, a West African Fermented Cereal Gruel.* Report submitted to HBVC Research grants program. USAID, Washington U.S.A

Olasupo, N.A, Olukoya, S.A and Odunfa, S.A., (1997). Assessment of a bacteriocin-producing *Lactobacillus* strain in the control of spoilage of a cereal-based African fermented food. *Folia microbiology*, 42:31-34.

Omemu, A.M., Bankole, M,O., Oyewole, O.B. and Akintokim, A.K., (2007). Yeasts and moulds associated with ogi- A cereal based weaning food during storage. *Research Journal of Microbiology* , 2 (2): 141-148.

Onyekwere, O.O, Akinrele, I.A, Koleoso, A.O., (1989). Industrialization of ogi fermentation In Industrialization of indigenous fermented foods; Steinkraws, K. H, ed; Marcel Dekker Inc. New York, pp, 329–361.

Osundahunsi, O.F. and Aworh, O.C., (2003). Nutritional evaluation, with emphasis on protein quality, of maize-based complementary foods enriched with soya bean and cowpea *Tempe. Int. J. Food Sci. Technol.*, 38: 809-813.

Osungbaro, T. O., (2009). Physical and nutritive properties of fermented cereal foods. *African Journal of Food Science*, 3(2): 23-27.

Owusu-Kwarteng, J., Tano-Debra, K., Glover, R.L.K. and Akabanda, F., (2010). Process characteristics and Microbiology of *fura* produced in Ghana. *Nature and Science*, 8(8): 41-51.

Teniola, O.D. and Odunfa, S.A., (2002). Microbial assessment and quality evaluation of ogi during spoilage. *J. Microbiol. Biotechnol.*, 18(8): 731-737.

Vainio, H., Hesseltine, E. and Wilbourn, J., (1993). Report on an IARC working group meeting on some naturally occurring substances. *Int. J. Cancer*, 53: 535-537.