Journal of Medical and Applied Biosciences
ISSN: 2277 - 0054
Volume 8, Number 1, 2016

Effect of Fermentation Period on the Microbial Profile during the Production Starter Based Kunun-Zaki

[1]Mohammad Abubakar Birnin Kebbi, [1*]Musliu Abdulkadir & [2]Oluwafemi, O. Fasiku,
[1]*Department of Science Laboratory Technology, College of Science and Technology*
[2]*Department of Preliminary Studies, College of Science and Technology*
Waziri Umaru Federal Polytechnic, Birnin-Kebbi, Kebbi State.
Email: musille2003@yahoo.com

ABSTRACT

The present study was conducted to determine the microbial profile during starter based kunun zaki production. The starter culture used were Lactic acid bacteria collected from the microbiology laboratory, department of science laboratory technology, Waziri Umaru Federal Polytechnic, Birnin Kebbi. Microbial counts (total bacteria, yeast count and enteric bacteria) were done after 8 hours of fermentation then after every 4hrs of fermentation. The highest bacteria count obtained was (4.4×10^5) in SFK (spontaneous fermented kunun) while the lowest count was 2.4×10^2 in LFK (Lactobacillus plantarum kunun). Lactic acid bacteria range from 6.2×105 to 1.0×103. The highest enteric bacteria count was observed in SFK (9.0×104) so also the highest yeast counts (1.3×105). No growths were recorded for yeast and enteric bacteria after 12 to 24 hours of fermentation for the starter based products. The results of this research study showed that the LAB species used as starter controlled the growth of unwanted microorganisms.

Keywords: Kunun Zaki Production, Lactic Acid Bacteria, Microbial Counts

INTRODUCTION

Kunun-zaki' is fermented non-alcoholic cereal beverage whose popularity in Nigeria is due to its characteristic sweet-sour taste typical of other lactic acid bacterial fermented foods of African origin such as mahewu and baganiya (Dirar, 1993). However, the short shelf-life (24 - 48 h) of this product is of major concern to its manufacturers and consumers (Efiuvwevwere and Akoma, 1995). The use of starter cultures does not find widespread application in the traditional fermentation for foods in Africa. However, instances exist where backslopping and microbially-impregnated cloth such as an inoculation belt are used to initiate and control food fermentation (Holzapfel, 2002). For example, in

the fermentation of cassava dough into agbelima, pretreated cassava chunks are wrapped in a piece of cloth that has been used on the previous occasion to produce an inoculum. This is done before the chunks are allowed to ferment into an inoculum, which is subsequently used to ferment cassava mash into agbelima (Amoa-Awua and Jakobsen, 1995).

In recent years, attempts are being made in laboratories in several African countries to develop starter cultures along the lines used in modern food processing establishments to upgrade the production of various indigenous African fermented foods.Lactic acid Bacteria (LAB) are Gram positive fastidious, acid tolerant, generally non-sporulating, catalase negative devoid of cytochrom and non-respiring rod or cocci that are characterized by their common metabolic and physiological characteristics. They produce lactic as major or sole product of fermentative metabolism. Lactic acid bacteria have been used for fermentation of food and feed products since ancient days and today their major applications are still in the food and feed industry as starter culture (Oshoma *et al.,* 2009).

Fungi, Gram-positive and Gram-negative bacteria had been isolated from sampled Kunun-zaki. It had been discovered that microorganisms especially *E. coli* can grow and survive during the storage of Kunun-zaki at different temperature (Oshoma *et al.,* 2009). It is possible that the high counts of spoilage and pathogenic microorganisms in Kunun-zaki could be reduced if starter cultures are employed in its fermentation process as done in the developed world (Agarry *et al.*, 2010). The aim of this research is to evaluate the effect of fermentation period on the microbial profile during the production of starter based kununzaki

FERMENTATION IN FOOD PROCESSING

Fermentation in food processing is the conversion of food carbohydrate to alcohols and carbon dioxide or organic acid using yeasts, bacteria, or a combination of them under anaerobic conditions. Fermentation usually implies that the action of microorganism is desirable. The science of fermentation is also known as Zymology or Zymurgy. The term "Fermentation" is sometimes used to specifically refer to the chemical conversion of sugar into ethanol, a Process which is used to produce alcoholic beverages such as wine, beer, and acid. Fermentation is also employed in the leavening of bread (CO_2 Produced by yeast activities). In preservation techniques to produce lactic acid in sour foods such as sauerkraut, dry sausages, kimch and yoghurt. And in

picking of food with Vinegar (Acetic acid). Locally fermented foods are produced by the activities of microorganisms, which while forming only a small proportion of the total weight of the foods, have profound effects on the character of the food. Locally fermented foods in Nigeria are grouped into tubers (e.g. gari, and fufu), cereals (e.g. ogi and pito), legumes (e.g. dawadawa and iru), milk (e.g. local cheeses) and beverages (e.g. palm wine). The mechanism of fermentation is biochemical and it involves lactic acid fermentation to yield products such as organic acids, alcohols, aldehydes and ketones. Some species of microorganisms involved in fermentation are *Lactobacillus, Lactococcus, Leuconostoc, Enterococcus, Streptococcus, Penicillium*and *Saccharomyces*. Locally fermented foods are significant in provision of employment opportunities, market improvement, availability of food supplement and poverty alleviation. Some advantages of locally fermented foods are enhancement of organoleptic and preservative properties, provision of nutritional quality, detoxification and production of antibiotics. However, they contribute to food spoilage and health defects. Some factors influencing production of locally fermented foods are temperature, water activity, hydrogen ion concentration (pH), oxygen availability and substrate used for food fermentation process. (Nwachukwu *et al.*, 2010).

The primary benefit of fermentation is the conversion of sugars and other carbohydrates to usable end products. According to Steinkraus (1995), the traditional fermentation of foods serves several functions, which includes: enhancement of diet through development off flavour, aroma, and texture in food substrates, preservation and shelf-life extension through lactic acid, alcohol, acetic acid and alkaline fermentation, enhancement of food quality with protein, essential amino acids, essential fatty acids and vitamins, improving digestibility and nutrient availability, detoxification of anti-nutrient through food fermentation processes and a decrease in cooking time and fuel requirement (Dirar, 1993). Fermentation can produce important nutrients or eliminate anti-nutrients. Food can be preserved by fermentation, since fermentation uses up food energy and creates conditions unsuitable for spoilage microorganisms. For instance, in pickling, the acid produced by the dominant organism inhibits the growth of all other microorganisms.

Fermenting makes foods more edible by changing chemical compounds, or predigesting, the foods for us. There are extreme examples of

poisonous plants like cassava that are converted to edible products by fermenting. Some coffee beans are hulled by a wet fermenting process, as opposed to a dry process. Reduction in anti-nutritional and toxic components in plant foods by fermentation was observed in a research which showed " Cereals, legumes, and tubers that are used for the production of fermented foods may contain significant amounts of antinutritional or toxic components such as phytates, tannins, cyanogenic glycosides, oxalates, saponins, lectins, and inhibitors of enzymes such as alpha-amylase, trypsin, and chymotrypsin (Dirar, 1993).

LACTIC ACID BACTERIA

Lactic acid bacteria (LAB) are group of Gram -positive bacteria that are devoid of *cytochromes* and preferring anaerobic conditions, fastidious, acid-tolerant and strictly fermentative. They are usually non-motile and non-sporulating bacteria that produce lactic acid. This bacterial group contains both rods (Lactobacilli and *Carnobacteria*) and Cocci (*Streptococci)*. Different species of lactic acid bacteria (such as *Streptococcus*, *Leuconostoc, Pediococcus, Aerococcus*, *Enterococcus, Vagococcus, Lactobacillus*, *Carnobacterium*) have adapted to grow under widely different environmental conditions. They are found in the gastrointestinal tract of various animals, dairy products, seafood products, soil and on some plant surfaces (Salmon *et al.,* 2008)Although lactic acid bacteria are not dominant in the normal intestinal micro biota, several trials have been undertaken to induce an artificial dominance of lactic acid bacteria. Based on their carbohydrate metabolism LAB are divided into two distinct groups. The homo-fermentative group utilizes the Embden-Meyerhof-Parnas (glycolytic) pathway to transform a carbon source chiefly into lactic acid. Hetero -fermentative bacteria produce equimolar amounts of lactate, CO_2, ethanol or acetate from glucose exploiting phosphor ketolase pathway.

Lactic Acid Bacteria as Functional Starter Cultures

A starter culture can be defined as a microbial preparation of large numbers of cells of at least one microorganism to be added to a raw material to produce a fermented food by accelerating and steering its fermentation process. The group of lactic acid bacteria occupies a central role in these processes, and has a long and safe history of application and consumption in the production of fermented foods and beverages. They cause rapid acidification of the raw material through the production of organic acids, mainly lactic acid. They also produce acetic acid, ethanol, aroma compounds, bacteriocins,

exopolysaccharides, and several enzymes. In this way, they enhance shelf-life and microbial safety, improve texture, and contribute to the pleasant sensory profile of the end product. The earliest production of fermented foods was based on spontaneous fermentation due to the development of the microflora naturally present in the raw material. The quality of the end product was dependent on the microbial load and spectrum of the raw material. Spontaneous fermentation was optimized through backslopping, i.e., inoculation of the raw material with a small quantity of a previously performed successful fermentation. Hence, backslopping results in dominance of the best adapted strains. It represents a way, be it unconsciously, of using a selected starter culture to shorten the fermentation process and to reduce the risk of fermentation failure. Backslopping is still in use, for instance in the production of sauerkraut and sourdough, and particularly for products which the microbial ecology and the precise role of successions in microbial population are not well known. Today, the production of fermented foods and beverages through spontaneous fermentation and backslopping represents a cheap and reliable preservation method in less developed countries, whereas in Western countries the large-scale production of fermented foods has become an important branch of the food industry. Moreover, the Western consumer appreciates traditionally fermented products for their outstanding gastronomic qualities (Huili *et al.,* 2011). The direct addition of selected starter cultures to raw materials has been a breakthrough in the processing of fermented foods, resulting in a high degree of control over the fermentation process and standardization of the end product. Strains with the proper physiological and metabolic features were isolated from natural habitats or from successfully fermented products. However, some disadvantages have to be considered.

In general, the initial selection of commercial starter cultures did not occur in a rational way, but was based on rapid acidification and phage resistance. These starters are not very flexible with regard to the desired properties and functionality of the end product. Originally, industrial starter cultures were maintained by daily propagation. Later, they became available as frozen concentrates and dried or lyophilized preparations, produced on an industrial scale, some of them allowing direct vat inoculation. Because the original starter cultures were mixtures of several undefined microbes, the daily propagation probably led to shifts of the ecosystem resulting in the

disappearance of certain strains. Moreover, some important metabolic traits in lactic acid bacteria are plasmid-encoded and there is a risk that they are lost during propagation. It is further likely that loss of genetic material occurred due to adaptation to the food matrix. The biodiversity of commercial starters has therefore become limited. This often leads to a loss of the uniqueness of the original product and the loss of the characteristics that have made the product popular.

In contrast, the fermentation of traditional fermented foods is frequently caused by natural, wild-type lactic acid bacteria that originated from the raw material, the process apparatus, or the environment, and that initiate the fermentation process in the absence of an added commercial starter. Moreover, many traditional products obtain their flavour intensity from the non-starter lactic acid bacteria (NSLAB), which are not part of the normal starter flora but develop in the product, particularly during maturation, as a secondary flora. Pure cultures isolated from complex ecosystems of traditionally fermented foods exhibit a diversity of metabolic activities that diverge strongly from the ones of comparable strains used as industrial bulk starters. These include differences in growth rate and competitive growth behaviour in mixed cultures, adaptation to a particular substrate or raw material, antimicrobial properties, and flavour, aroma, and quality attributes. Wild strains need to withstand the competition of other microorganisms to survive in their hostile natural environment, so that they often produce antimicrobials such as bacteriocins. In addition, they are more dependent on their own biosynthetic capacity than industrial strains and harbour more amino acid converting enzymes that they play a key role in flavour formation. Such findings underline the importance of the Protected Designation of Origin (PDO) of many of these products, which is crucial from an economical point of view since they contribute to the survival of small-scale fermentation plants in a world of ongoing globalization. A recent trend exists in the isolation of wild-type strains from traditional products to be used as starter cultures in food fermentation (Fujitoshi *et al.,* 2005).

Nowadays, the consumer pays a lot of attention to the relation between food and health. As a consequence, the market for foods with health-promoting properties, so called functional foods, has shown a remarkable growth over the last few years. Also, the use of food additives is regarded as unnatural and unsafe. Yet, additives are needed to preserve food products from spoilage and to improve the organoleptic properties. The demand

for a reduced use of additives and processing seems contradictory with the market preference for products that are fresh, safe, tasty, low in sugar, fat, and salt, and easy to prepare. In cheese-making, for instance, the use of raw milk permits the manufacture of high-value traditional artisan varieties but brings about safety risks, e.g. the development of *Listeria monocytogenes*. On the other hand, pasteurization of the milk results in loss of flavour and gives end products that are perceived by the consumer as "boring". These market trends put the food industry under pressure to look for alternatives. In food fermentation, one of the key points for intervention seems to be on the level of the starter culture. Unfortunately, industrial starter cultures lack the necessary characteristics for product diversification, and the commercial availability of new interesting starter cultures is limited. The increased understanding of the genomics and metabolics of food microbes opens perspectives for starter improvement. Through molecular biology it is now possible to express desirable and suppress undesirable properties of starter culture.

Recently, the use of functional starter cultures in the food fermentation industry is being explored. Functional starter cultures are starters that possess at least one inherent functional property. They can contribute to food safety and/or offer one or more organoleptic, technological, nutritional, or health advantages (Fujitoshi *et al.,* 2005).

LABORATORY FERMENTATION OF KUNNUN ZAKI

Kunnu-zaki is a cereal food drink that has become a popular refreshing non-alcoholic beverage in Nigeria, particularly, in the North and South West. Millet, sorgum and maize grains are the three principal cereals from which Kunun-zaki can be produced (Adeleke *et al.,* 2004). It is usually flavoured with such spices as ginger, black pepper and tamarind for improvement in its taste and aroma, and also to serve as purgative and cure for flatulent conditions. The final product is claimed to enhance lactation in nursing mothers (Efiuvwevwere and Akoma, 1997). Like other cereal foods, Kunun-zaki also protects the body against cholesterol and bile acid metabolism related diseases such as gallstones and certain forms of heart diseases (Bangert, 1989). It was observed that Kunun-zaki elevates lymphocyte counts obtained in the blood samples of animals fed with Kunun-zaki which is indicative of its medicinal attributes, a concept widely believed by its numerous consumers (Akoma *et al.,* 2006). Kunun-zaki is supposed to be self-

sanitizing considering the generation of such fermentation products as organic acids, hydrogen peroxide, antibiotic-like substances and lowered oxidation-reduction potential (Bangert, 1989).

Also, the three spices (ginger, pepper and tamarind) have been shown to possess varying levels of antimicrobial property (Bankole *et al.,* 1999), contrary to a much earlier report that these harboured bacteria and toxigenic fungi. Utensils, water source, product handling and non-regulated production and dispensing methods are factors that can encourage microbial contamination of Kunun-zaki with its attendant objection-able degradative changes. (Peters and Odeyemi, 1990). Fungi, Gram-positive and Gram-negative bacteria had been isolated from sampled Kunun-zaki (Adeleke *et al.,* 2004). It had been discovered that microorganisms especially *E. coli* can grow and survive during the storage of Kunun-zaki at different temperature (Oshoma *et al.,* 2009). It is possible that the high counts of spoilage and pathogenic microorganisms in Kunun-zaki could be reduced if starter cultures are employed in its fermentation process as done in the developed world (Agarry *et al.*, 2010). Kunun-saki, a type of Sorghum beer is a traditional beverage produced and consumed locally in Nigeria and some African countries. It is produced mainly from starchy grains such as guinea corn, millet and maize. These crops are produced in the tropical regions of Africa and particularly in the Northern Guinea Savanna areas of Nigeria (Ettasoe, 1972; Asiedu, 1989). The process of kunun-zaki production basically involves malting, mashing, boiling, fermentation and maturation in which variations may occur depending on local practices (Ekundayo, 1969). The basic characteristics of kunun-zaki include a sour taste due to the presence of lactic acid, a pH of 3.3-3.5 and opaque color because of suspended solids and yeasts. It contains vitamins, iron, manganese, magnesium, phosphorous and calcium and also contains about 26.7g of starch and 5.9g of proteins per litre (Kingsley and Victor, 2007). The production of kunun-zaki includes: a 12- 36h spontaneous mixed fermentation involving lactic acid bacteria and yeasts (Sefa-Dedeh, 1991; Sefa-Dedeh *et al.,* 1999). Bacteria of the genera *Lactobacillus* spp and *Leuconostoc mesenteriodes*are the major contributors to the acidity of kunun-zaki during the initial souring stage. Most of the acid produced is lactic acid with only traces of acetic and formic acid being present (Sefa-Dedeh, 1991; Kolawole, 2007).

In the fermenting medium, the yeast mainly isolated are *Saccharomyces cerevisae* and *Saccharomyces chavalieria* (Achi, 2005). The

diversity of the associated yeast micro flora of traditional alcoholic beverages in sub Saharan Africa was attributed to the spontaneous nature of the fermentation, sources, and types of ingredients used (Sanni, 1993; Sanni and Lonner, 1993). Glover *et al.* (2005) also reported that *Saccharomyces cerevisiae* predominate the alcoholic fermentation of sorghum wort during production of pito within eight geographical regions of Ghana and dolo at four production sites in Burkina Faso. From the previous reports (Sanni and Oso, 1988) fermented foods and beverages constitute a major portion of the Nigerian diet. However, most of these foods and beverages are still products of traditional family art and fermentation process is initiated by chance inoculation. The raw materials are low in protein, vitamins and mineral (Sanni and Oso, 1988). Spontaneous fermentation are difficult to control, are predictable in terms of length of fermentation and quality of product; can produce unwanted products or products with short shelf life. To overcome these problems, the most predominant microorganisms found in an acceptable product can be isolated, purified and used as starter cultures to initiate the fermentation (Togo *et al.*, 2002).

METHODOLOGY

Collection of Sample

Sorghum and millet were obtained from New-Market Brining Kebbi, Kebbi State , the samples were collected in a sterile polythene bag and carefully transported to the Laboratory for Further Processing, Starter culture obtain form the Department of Science Laboratory , Microbiology Unit, Waziru Umaru Federal Polytechnic Brinin Kebbi.

Media Preparation

DeMann Rogosa and Sharpe medium (MRS) for the growth of Lactic acid bacteria, Nutrient agar for bacteria count, MacConkey and EMB agar for enteric bacteria count and yeast extract agar for yeast count. All media were aseptically prepared using manufactures instructions and standard operating procedure.

MICROBIOLOGICAL ANALYSIS

Isolation media used were Nutrient Agar (NA), Malt Extract Agar, MacConkey Agar, DeMann Rogosa and Sharpe Medium (MRS), Eosine Methylene Blue Agar (EMB), and Plate Count Agar (PCA). The Kunun zaki samples under fermentation were being taken at intervals for microbial enumeration starting from 0 to 24 hours during fermentation.1ml of the sample was transferred into the test tubes containing sterile distilled water to make dilution of 10^{-1} and shaken to dislodge microorganisms. The

process was continued until 10^{-7} dilution was obtained. After serial dilution has been done, 1ml each from dilutions10^{-4}, 10^{-5} and 10^{-6} were pipetted into sterile Petri dishes. The molten forms of the media were poured into separate plates containing the inoculum. This was done for Nutrient agar, Malt Extract agar, MacConkey agar and MRS respectively. Nutrient agar and MacConkey agar were incubated at 37°C for 24hours. MRS at 37°C under anaerobic condition for 48hours and Malt extract at 30°C for 5days.After incubation, the plates were counted as colony forming units (cfu/ml) and sub-cultured to obtain pure cultures (Fawole and Oso, 2007).

RESULT AND DISCUSSION

Result

Table 1: Bacteria Counts (CFU/ml) at Different Fermentation Period

Product code	Fermentation hour				
	0hrs	8hrs	12hrs	16hrs	24hrs
LPK	3.0×10^5	2.5×10^3	1.9×10^3	1.8×10^2	1.5×10^2
LFK	2.5×10^5	1.3×10^5	3.2×10^3	9.0×10^2	2.4×10^2
LPFK	4.0×10^5	2.0×10^3	3.2×10^2	1.6×10^2	4.0×10^1
SFK	4.4×10^5	2.2×10^3	3.5×10^3	1.4×10^5	4.2×10^5

Table 2: Total Lactic Acid Count (CFU/ml) at Different Fermentation Period

Product code	Fermentation hour				
	0hrs	8hrs	12hrs	16hrs	24hrs
LPK	1.6×10^4	3.9×10^5	4.8×10^5	6.2×10^5	5.7×10^5
LFK	1.5×10^4	4.0×10^5	5.8×10^5	5.4×10^5	4.8×10^5
LPFK	1.5×10^4	1.0×10^3	1.2×10^5	1.6×10^5	1.2×10^5
SFK	2.5×10^5	1.2×10^5	2.4×10^5	1.4×10^5	1.0×10^5

Table 3: Yeast Count (CFU/ml) at Different Fermentation Period

Product code	Fermentation hour				
	0hrs	6hrs	12hrs	18hrs	24hrs
LPK	5.0×10^4	3.0×10^3	NG	NG	NG
LFK	1.0×10^5	9.0×10^3	NG	NG	NG
LPFK	1.1×10^5	1×10^4	NG	NG	NG
SFK	1.3×10^5	1.1×10^5	1.6×10^5	1.8×10^2	2.4×10^2

Table 4: Enteric Bacteria Count (CFU/ml) at Different Fermentation Period

Product Code	Fermentation Hour				
	0hrs	6hrs	12hrs	18hrs	24hrs
LPK	7.0×10^4	6.0×10^2	NG	NG	NG
LFK	8.0×10^4	6.0×10^4	NG	NG	NG
LPFK	9.0×10^4	5.0×10^4	NG	NG	NG
SFK	1.5×10^5	2.0×10^5	2.4×10^3	3.0×10^2	2.0×10^2

DISCUSSION

Table (1) shows bacterial counts at different fermentation period. The fermentation was done for 24hr and viable colony count was carried out at six hour interval after the first 8 hours of fermentation. The counts obtained varies, there were decreases in bacterial population for the starter based fermentation as the fermentation period progresses; for LPK the bacteria count obtained is as follow (3.0×10^5 2.5×10^3 1.9×10^3 1.8×10^2 1.5×10^2) while for LFK (2.5×10^5 4.3×10^5 3.2×10^3 9.0×10^2 2.4×10^2), this agree with result obtained by Adriana *et al.* (2002) that studies the effect of Lactic acid bacteria in microbiological quality of fermented food. For spontaneous fermentation (SFK) there was decrease in bacteria growth for the first 8hrs of fermentation period but later increase as the fermentation progresses to 24hrs the bacteria (4.0×10^5 2.2×10^3 3.5×10^3 1.4×10^5 4.2×10^5) this result is also agrees with the work of Adriana *et al.* (2002), and Amankawah *et al.* (2009).

Table (2) shows Lactic acid bacteria count at different fermentation periods generally in the sample the counts increased with fermentation time. The yeast the enteric count were recorded only at early hours of fermentation but later reduced into zero level at 12 to 24hrs of fermentation period but for SFK there were growth at the end of fermentation period (Table 3 and 4) these results is not in satisfy with the result obtained by Amankawah *et al.* (2009) which numerous growth was observed during and at the end of the fermentation period. The international microbiological standard recommended that limit of contamination for food should be less than 1.5×10^6CFU/g (Anon, 1974). The reduction and the total disappearance of the yeast and enteric bacteria may result from the lactic acid bacteria used as starter culture.

The nutritional characteristics of the formulated weaning foods after 12hours and 24 hours fermentation as presented in Table 5 and Table 6

respectively. The analysis of the proximate composition of all the starter fermented formulated sorghum, soya bean and tiger nuts blends indicates that the protein content increase with increase in fermentation periods. The highest protein content (35.63%) was observed in samples SBFR2 after 24h fermentation time, while the lowest protein content of 19.80% was observed in sample SPFR1 at 12h fermentation time.

CONCLUSION

This study showed that the starter cultures used either singly or in consortium has a grate influence in the control and reduction of microbial load reduction most especially the yeast and enteric bacteria as the fermentation period progresses.

REFERENCES

Adeleke, O.E., Olaitan, J.O. and Olubile, O. (2004). Microbial Isolates from KUNNU-Zaki and their Antibiotic Sensitivities. *Adv. Food Sci.* **26(**4): 168-170.

Adriana, L.B., Amelia, F.G., Rita, P.S. and Jorge, L.P. (2002). Effect of Lactic acid Bacteria Culture in the Microbiological Quality of Fermented. *Pakistan Journal of Nutrition* **4** (8) 30-40

Agarry O.O., Nkama I., Akoma O. (2010). Production of 'Kunun-Zaki' (Nigerian Fermented Cereal Beverage) using Starter Culture. *Int. Res. J.Microbiol.,* 1(2)18-25

Akoma, O., Jiya, E.A., Akumka, D.D. and Mshelia, E. (2006). Influence of Malting on the Nutritional Characteristics of Kunun-Zaki. *Afr. J. Biotechnol.* 5(10):996-1000.

Amankwah, E.A., Barimah, A.K.M., Nuamah, J.H., Oldman, W. and Nmaji C.O. (2009). Formulation of Weaning Food from Fermented Maize Rice, Soybeans and Fish Meal *Pakistan Journal of Nutrition* **8** (11): 1747 – 1752.

Amoa-Awua, W.K. and Jakobsen, M. (1995). The Role of Bacillus Species in the Fermentation of Cassava. *Journal of Applied Bacteriology*. **79**: 250– 256.

Asiedu, J.J. Processing *Tropical Crops: a Technological Approach London:* Macmillan Washington DC, Pp:22: 113-114

Bangert, B.A. (1989). *The* Quality of Cereal Foods. *Food Chem.* **33**: 15-26.

Dirar, H.A. (1993). The Indigenous Fermented Foods of the Sudan. A Study in African Food and Nutrition. *CABS International Wallingford.* pp.111

Efiuvwevwere, B.J.O., Akoma O. (1995). The Microbiology of 'Kunun-Zaki' a Cereal Beverage from Northern Nigeria during the Fermentation (Production) Process. *World J. Microbiol. Biotechnol.* **11**: 491-493.

Ettasoe, C. (1972). *Sorghum and Pearl Millet: In Leaky CL, Wills J.B. (Eds) Food Crops of Lowland Tropics.* Oxford University Press, Great Britain. Pp **191-1922.**

Fujitoshi, Y., Yi-Sheng, C. and Takashi, S. (2005). Isolation and Characterization of Lactic Acid Bacteria from Soils in Vineyard, Institute of Enology and Viticulture, University of Yamanashi. *Journal of General Applied Microbiology* **51,** 313-318.

García-Ruiz, A., Bartolomé, B., Martínez-Rodríguez, A.J., Pueyo, E., Martín-Álvarez, P.J., Moreno-Arribas, M.V. (2008). Potential of Phenolic Compounds for Controlling Lactic Acid Bacteria Growth in Wine. *Food Control* **19**(9) 835-841.

García-Ruiz, A., Moreno-Arribas, M.V., Martín-Álvarez, J.P. and Bartolomé, B. (2011). Comparative Study of the Inhibitory Effects of Wine Polyphenols on the Growth of Enological Lactic Acid Bacteria. *International Journal of Food Microbiology* **145**(2-3), 426-431.

Glover, R.L., Abaidoo, R.C., Jakobsen, M. and Jespersen, L. (2005). Biodiversity of Saccharomyces Cerevisiae Isolated from a Survey of Pito Production Sites in Various Parts of Ghana. *Systematic and Applied Microbiology* **28** (8): 755-761.

Harper, L. 2003. *Development of Weaning Food Formulations Base on Cereal, International J. Food Sci and Technol., 24-28.*

Holzapfel, W.H., (2002). Appropriate Starter Culture Technologies for Small-Scale Fermentation in Developing Countries. *International Journal of Food Microbiology*. **75,**.197– 212.

Kingsley, C.E. and Victor, E.E. (2007). *A* Kinetic Study of Burukutu Fermentation *Journal of Engineering and Applied Sciences* **2(**7):1193-1198.

Nwachukwu, E., Achi, O.K. and Ijeoma, I.O. (2010). Lactic Acid Bacteria in Fermentation of Cereals for the Production of Indigenous Nigerian Foods. *African Journal of Food Science and Technology* **1**(2): 021-026.

Oshoma, C.E., Aghimien, M.O. and Bello, Z.O. (2009). Growth and Survival of Escherichia Coli in Kunun Zaki during Storage. *World J. Agric. Sci.* **5**(4): 494-497

Peters, H.I., Odeyemi, O. (1990). Bacteriological Quality of Mukuro Dam in Ile-Ife, Nigeria. *Nig. Food J.* **8**: 92-100. Press Ltd.1989 pp **301-3143**.

Salmon, K. Idris, T. Usman, D.M. (2008) Production of Weaning Food from Fermented Cereal – Base using Maize *African Journal of Food Science;* 58 (20) 520-536.

Sanni A.I., Oso B.A. (1988). Nutritional Studies on Agadagidi. Die Nahrung for Upgrading Traditional Fermented Foods through Biotechnology. *African Journal of Biotechnology*. **32**(2): 169-17226

Sanni, A.I., A.A Onilude and T.I. Ibidapo, 1999.*Biochemical Composition of Infant Weaning Food Fabricated from Fermented Blends of Cereal and Soybean, J. Food Chem,. 65:35-39.*

Sanni, A.I. (1993).The Need for Process Optimization of African Fermented Foods and Beverages. *International Journal of Food Microbiology.***18**: 85-95

Sanni, A.I., Lonner, C. (1993). Identification of Yeasts Isolated from Nigerian Traditional Alcoholic Beverages. *International Journal of Food Microbiology.* **10**: 517-523.

Sefa-Dedeh, S. (1991). Cereal Based Traditional Beverages from Ghana: Process and Product Characteristics of Pito. In: Westby, A., Reilly, P.J.A. (Eds.), Proceedings of a Regional Workshop on Traditional African Foods— Quality and Nutrition, 25–29 Nov. 1991. International Foundation for Science, Stockholm, Pp. 19– 22.

Sefa-Dedeh, S., Sanni, A.I., Tetteh, G. and Sakyi-Dawson, E. (1999). Yeasts in the Traditional Brewing of Pito in Ghana. *World Journal of Microbiology and Biotechnology.* **15:** 593-5977.

Togo, C.A., Feresu, S.B. and Mutukumira, A.N. (2002). Identification of Lactic Acid Bacteria Isolated from Opaque Beer (Chibuku) for Potential Use as a Starter Culture. *Journal of Food Technology of Africa*. **7**(3): 93-97

Reference to this paper should be made as follows: Mohammad Abubakar Birnin Kebbi; et al. (2016), Effect of Fermentation Period on the Microbial Profile during the Production Starter Based Kunun-Zaki. *J. of Medical and Applied Biosciences,* Vol. 8, No. 1, Pp. 1 – 15.