



## Ecology of Traditional Cereal Fermentation

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### Abstract

Cereals are globally number one food crops as well as substrates for fermentation. Fermentation of cereal-based foods is a common practice in Africa. It is a desirable process of biochemical modification of primary food matrix brought about by microorganisms and their enzymes. Traditional food fermentations represent an extremely valuable cultural heritage in most regions and harbour huge genetic potential of valuable but hitherto undiscovered strains. Microbial diversity of cereal-based fermented foods ranged from lactic acid bacteria to endospore-forming bacteria, amylolytic producing yeasts and filamentous moulds. Yeasts in cereal fermentation increases the protein content of fermented food products. The genus *Saccharomyces*, in particular *Saccharomyces cerevisiae* is strongly associated with the production of fermented cereal products for human Consumption.

The prominent group of bacteria with antifungal property is the lactic acid bacteria (LAB). LABs are large group of beneficial bacteria in the ecosystem of several fermentations as they have major potentials for use in biopreservation. Microorganisms engage in a wide variety of interactions. During the fermentation of sorghum, millet and maize, the pH of the medium decreased due to the production of carboxylic acid by fermenting microorganisms. Traditional food products arising from such fermentations include *pito*, *ogi*, *burukutu*, *kunu-zaki* etc. The predominant microorganisms for these fermentations are mainly LABs and yeast species. The ecological information helps to effectively manage the microbial growth and activities during fermentation.

**Key words:** Ecology, Cereal fermentations, Lactic acid bacteria, Yeasts, Cereal fermented food products.

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## 1. Introduction

Cereals are globally number one as food crops as well as substrates for fermentation. Well known products are beer, sake, spirits, malt vinegar, Ogi and baked goods made from dough leavened by yeasts or sourdough [1]. Cereals are a major component of human food in Africa. Nutritional experts have paid attention to cereal based foods from maize, sorghum and millet sources. These cereals have high content of soluble non-starch polysaccharides such as beta glucan which has a health promoting role. Clinical and epidemiological studies indicating that beta glucan from barley or oat based products control cardiovascular disease in humans have been reported [2-5]. Duchonova *et al.* [6] suggested that the multiple beneficial effects of cereal can be exploited in different ways hence design of novel cereal foods or ingredients can be targeted at a specific population.

When considering the multitude of foods made from cereals one has to recognize that their greater part has been subjected to fermentation processes taking place at least at one step of their generation. In general, fermentation is a process that proceeds under the influence of activities exerted by enzymes and/or microorganisms. In fact, both activities are important in cereal fermentation [7], but in the context of this treatise neither plain enzymatic processes are considered, nor those numerous fermentations in which cereals are mixed with substantial amounts of additional substrates, such as legumes or milk (e.g. soy sauce and miso in South-East Asia; idli and dosa in India; kishk in the Middle East) [8]. Like with any other fermentation process, the understanding of the microbial ecology of cereal fermentations needs the knowledge of the fermentation substrates, i.e. the grains or seeds of the various cereal plants, as well as the products obtained

thereof. This framework includes the characterization of the microbial associations and ecological factors, which govern the fermentation process and arise from the nature of the cereal substrate. In cereal fermentations endogenous enzymes, bacteria, yeast and moulds play roles either singularly or in combination, and contribute to the creation of a great variety of products. This work examines the ecological overview during cereal fermentation.

## 2. Fermentation of cereals

Fermentation of cereal-based foods is a common practice in Africa for food preservation. It is a technology that is simple; home based and has fed millions of people. Currently, a variety of fermented foods are produced from cereals at household and semi industrial scale. These foods are used as weaning foods for infants and children and also for adults [9 -11]. Fermented cereals are widely utilized as food in African countries and in fact cereals account for as much as 77% of total caloric consumption [12]. Fermentation is a chemical change brought about by enzymes of living microorganisms [13]. It is a desirable process of biochemical modification of primary food matrix brought about by microorganisms and their enzymes [14]. It occurs in yeasts and bacteria and also in oxygen-starved muscle cells as in the case of lactic acid fermentation. It is a form of food processing where microbes for example, lactic acid bacteria (LAB) are utilized. In the developing world, fermentation is one of the oldest technologies used for food processing and preservation. On this technology depend millions of people for preserving and often enhancing organoleptic and nutritional qualities of their food at costs available to the average consumer [15 -17]. Cereal grains such as maize, sorghum, millet, rice, barley, rye and wheat had been

one of the earliest sources of food [18]. One way of processing the grains into food is through fermentation [19]. The grains are fermented, milled and mixed into starch mash by mixing with water and then cooked. Many indigenous cereal fermentations involve the combined action of bacteria, yeast and fungi. Some micro flora may participate in parallel while others may participate in a sequential manner with a changing dominant flora during the course of the fermentation. The soaking of the grains in excess water allows the selection of desirable microorganisms such as lactic acid bacteria, yeasts and moulds [20, 21].

### 3. Fermented cereal foods

Culturable and non-culturable microorganisms naturally ferment majority of global fermented foods and beverages. Traditional food fermentation represents an extremely valuable cultural heritage in most regions, and harbors a huge genetic potential of valuable but hitherto undiscovered strains. Holistic approaches for identification and complete profiling of both culturable and non-culturable microorganisms in global fermented foods are of interest to food microbiologists. The application of culture-independent technique has thrown new light on the diversity of a number of hitherto unknown and non-cultural microorganisms in naturally fermented foods as reported by Jyoti, Koichi and Wilhelm [22].

Fermented cereal foods play an important socio-economic role in developing countries as well as making a major contribution to the protein requirements of natural populations. In general, traditional fermented foods are made under primitive conditions, which result in low yield and poor quality. Their microbiota which is dominated by lactic acid bacteria has been extensively investigated. The

relation between microbial diversity and product characteristics are linked between the food microbiota and health benefits. The beneficial effects are the preservation of foods and the increase in their organoleptic characteristics because of the production of lactic acid and other metabolites synthesized by lactic acid bacteria [23]. The production of fermented sugar rich foods, namely cereals, roots and fruits are worldwide since the oldest record [24, 25]. Several of mankind's milestones such as the dawn of agriculture are closely linked with the production of some types of alcoholic beverages. Similar processes of fermentation emerge undependably in many civilizations across the globe. Interestingly, the main players of the whole process are relatively few, most yeast from *Saccharomyces* genus and lactic acid bacteria [26, 27]. Nowadays, such microorganisms have significant roles in several industrial relevant processes, including the production of beer, wine, bread, cheese, kunu, ogi, burukutu and other cereal products [28].

Historically, the classical separation of microbiological research between the bacteriologists has led to the study of bacteria and fungi in axenic settings. The compartmentalization has overlooked the fact that in many environments, bacteria-fungi coexist and interact. These bacteria-fungi interactions (BFIs) often have important ramifications for the biology of the interacting partners. These interactions harbour properties distinct from those of the single component [29]. This leads to the production of antibiotics [30], modification of the physical environment such as pH [31-36]. A bacteria-fungi interaction is applied in fermentation and brewing as determined by the taste, quality and safety of a wide range of foods [37].

Fermented cereal foods are foods which have been subjected to the actions of microorganisms or enzymes so that desirable biochemical changes cause

a significant modification to the food [8]. By fermentation, food is made more digestible, nutritious and safer or has better flavour [38]. In developing countries, fermented foods are produced primarily at the household and village level where they find wide consumer acceptance. Fermented beverages serve as food supplement like the use as a weaning food to supplement breastfeeding. They made available the diet required in human body [39]. Traditional fermentation processes used in the production of these foods are uncontrolled and are dependent on microorganisms from the environment or the fermentation substrate for initiation of the

fermentation processes [40]. Fermentation is a natural process that unavoidably affects human food supply worldwide. Many common food products contain fermentation microorganisms and their products. When consumed by humans, it often introduced micro floral which inhabit the human body [41]. The classes of fermented food produced in different regions of the world reflect the diet of the region and also the availability of the raw material. Some fermented cereals, their products and microorganisms involved are shown in Table 1.

**Table 1.** Common fermented cereal foods, products and microorganisms of fermentation.

Cereal	Product	Fermenting microorganism	Reference
Wheat, Rye	Bread	<i>Saccharomyces cerevisiae</i> , Yeasts, <i>Lactic acid bacteria</i> (LAB)	42
Maize, Millet, Sorghum	Ogi	LAB, <i>Enterobacter</i> sp, <i>Cephalosporium</i> , <i>Fusarium</i> , <i>Aspergillus</i> sp, <i>Penecillium</i> sp, <i>S. cerevisiae</i> , <i>Rhodotorula</i>	8, 19, 39,42,43, 45
Sorghum	Burukutu	<i>Saccharomyces</i> sp, <i>Leuconostoc mesenteroides</i> , <i>Acetobacter</i> sp, <i>Lactobacillus</i> sp, <i>Rhodotorula</i> , <i>Cryptococcus</i>	46, 47
Maize, Millet, Sorghum	Mahewu	<i>Lactobacillus delbruckii</i> , <i>L. bulgaricus</i> , <i>Debaromyces</i> sp, <i>Saccharomyces</i> sp	42, 48, 49, 59, 60
Maize, Sorghum, Millet	Uji	LAB	42
Rice	Idli	<i>Leuconostoc</i> , <i>L. delbruckii</i> , <i>L. fermentum</i> , <i>Lactococcus lactis</i> , <i>Pediococcus cerevisiae</i>	50-52
Sorghum	Beer	<i>S. crevisiae</i> , <i>S. calbergensis</i>	52, 53
Maize, Sorghum	Kinky	LAB, Yeast	42, 54
Sorghum	Pito	<i>Geotrichum</i> sp, <i>Lactobacillus plantarum</i> , <i>Rhizopus oryzae</i> , <i>S. cerevisiae</i>	47, 58
Maize	Kunu	LABs, <i>S. cerevisiae</i>	61-64

#### 4. Microbial diversity and succession during fermentation

Microbial diversity of cereal based fermented foods range from lactic acid bacteria to endospore forming bacteria, amylolytic producing yeasts and filamentous moulds. Raw cereals contain surface micro flora which are activated when water is added to the substrates

and micro floral which are also derived from various sources such as raw materials, utensils/equipments and food matrix [54]. The establishment of a particular micro flora in the substrate depends on water activities, pH, food matrix composition, salt concentration and method of preparation. During spontaneous fermentation, microbes either occur in

succession or co-exist with other microbial group in synergistic effect. For example, yeasts multiplication is favoured by acid environment developed due to metabolic activities of LAB, while bacteria growth is favoured by the yeasts activities as it provides several growth factors such as vitamins, minerals and nitrogen compounds during fermentation [65].

However, dominance of particular strains during fermentation results due to competitive abilities of the strains in the substrate. The predominant organisms reported in all cereal fermentations are lactic acid bacteria and Yeasts. Steinkraus [28] reported that the association of LAB and Yeasts during fermentation may also contribute metabolite which may impart taste and flavour to food. Naut [65] and Mensah [66] suggested that the production of lactic acid (in gruel) during fermentation may promote or improve the microbiological safety and stability of the product. Lactic acid bacteria strains are capable of lowering the pH of cereal fermentation to below 4.0 thus preventing spoilage of food and contribute largely to aroma formation in cereal products. Due to these properties, other non-acid producing organisms were eliminated making lactic acid bacteria as predominant organisms in most of the fermentation processes [8]. Four different genera of lactic acid bacteria dominated during cereal fermentation, they are; *Lactobacillus*, *Lactococcus*, *Leuconostoc* and *Pediococcus* [28]. Yeasts are found with lactic acid bacteria in almost all cereal fermentations [8]. They contribute significantly for structural quality and organoleptic characteristics of the product, ethanol. Their significance is also recorded in ogi, kenkey, beer, burukutu. Yeast strains are reported to exhibit wide range of enzymatic activities to produce flavour compounds such as alcohols, acids, esters terpenes and lactones during cereal fermentation. The predominant strains reported

from fermented cereals were *Saccaromyces cerevisiae*, *Geotrichum candidum*, *Candida krusei* and *Candida tropicalis* [67, 68].

### 5. Alcoholic fermentation of cereals

Alcoholic fermentation ecosystem is dominated by one genus of yeast; *Saccharomyces*. Beer is produced by fermenting maltose from malted grains [69]. Pito and Burukutu are brewed by fermentation of malted and germinated cereals [52]. Alcohol fermentation is the conversion of sugar into CO<sub>2</sub> and ethyl alcohol. The process is carried out by yeast cells using a range of enzymes. It is a complex series of conversions that bring about conversion of sugar to CO<sub>2</sub> and alcohol.

### 6. Yeasts in cereal fermentation

Yeasts are unicellular organisms (fungi) being the simplest eukaryotes present in a great number of environment. They can be found not only in decomposing fruit, trees and soils but also in commercial relationship with higher eukaryotes, humans included and even salt water [70]. Some yeasts are adapted to extreme environments such as high salt concentrations, low pH or extremely cold temperature. The genus *Saccharomyces*, particularly *Saccharomyces cerevisiae*, is strongly associated with the production of fermented cereal products for human consumption, namely; bread, wine, beer e. t. c. Yeasts in cereal fermentation had been shown to increase the protein content of the fermented product [71]. The possible functions of yeasts in fermented cereal foods are fermentation of carbo hydrate (formation of alcohol), production of aroma compounds (ester, alcohols, organic acid, carbonyl), stimulation of lactic acid bacteria providing essential metabolites, inhibition of mycotoxin producing moulds (nutrient competition, toxic compounds e.t.c.),

degradation of mycotoxin, degradation of cyanogenic glucosides (linamarase activity), production of tissue degrading enzymes (cellulase and pectinase) and probiotics. The interaction of yeast in cereal fermentation may contribute metabolite which could impact flavour and taste to the food [14].

### 7. Bacteria in cereal fermentation

Lactic acid bacteria are group of gram positive bacteria, non-respiring, non-spore forming, cocci or rods, which produce lactic acid as the major end product of fermentation of carbohydrate. Their primary activity is to convert carbohydrate to a desired metabolite such as alcohol, acetic acid, lactic acid or carbon (IV) oxide (CO<sub>2</sub>). while their secondary reactions lead to the formation of flavour and texture [42]. These bacteria ferment sour dough bread, sorghum and barley for beer; maize, sorghum, millet for ogi, kunu zaki, and burukutu [11, 63, 72]. Despite their complexity in ecosystem, the whole basis of lactic acid fermentation is centered on the ability of lactic acid bacteria to produce acid which then inhibit the growth of other non-desirable organisms [73]. As microorganisms co-exist the environment, they may influence one another. The prominent group of bacteria with antifungal activity is the lactic acid bacteria. The biocontrol imposed by lactic acid bacteria over mould may result from competition for space and nutrients, production of antifungal compounds and metabolite that may also influence the production of mycotoxins [74]. Lactic acid bacteria are large group of beneficial bacteria in ecosystem of several fermentations as they have major potential for use in biopreservation because they are generally recognize as safe for consumers [75]. Their significant role is noticed in the nutritional value, safety, shelf-life and acceptability of cereal based foods [71, 76-77].

### 8. Microbial Interaction

Most food harbour a mixture of microorganisms which include different species of bacteria, yeasts and filamentous fungi as well as strains within these species [78]. Microorganisms engage in a wide variety of social interactions.. A cooperation behavior is one that benefits an individual (the recipient) other than the one performing the behavior (the actor) [79]. The ecological theory describes the range of interaction associations (their type of cooperation) as mutualism, commensalisms, parasitism or predation, amensalism or antagonism, competitive [80-82]. This could occur within different microbial groups such as bacteria-bacteria; yeast-yeast; bacteria-yeast; bacteria-fungi. All of these interactions regardless of the outcome occur through a diverse set of mechanisms by which genetic and molecular information is transferred. Antagonism is probably the best known microbial interaction in food ecosystem because it can be applied as a natural biocontrol strategy to enhance food quality and safety [83-84]. Bacteria cell growing in liquid media undergo regular physical interactions with each other and when grown on solid media are in tight contact [85]. More complex bacteria cell aggregates composed in several species such as consortia of methanogenous and methylotrophic bacteria are also found in natural habitats. This kind of cooperation is beneficial for bacteria because the product secreted by one species can serve as substrate for another species [86].

In many environments, bacteria and fungi coexist and interact as reported by Steinkraus [28] during the fermentation of Ogi. These bacteria-fungi interaction often have important ramification for the biology of the interacting partners. Studies have revealed that fungi and bacteria often form physically and metabolically interdependent consortia that harbour

properties distinct from those of their single components [29]. Bacteria-fungi interaction result in the production of food such as sourdough, wine, beer and other cereal fermented products [87 - 89]. Metabolite exchange in the bacteria-fungi interactions (BFI) result in the formation or degradation of a molecule that neither partner can produce alone. Several complex food products require BFIs for their production like wine, beer Ogi, cheese e.t.c., and each partner contribute to the synthesis and organoleptic qualities of the final end product [90].

### 9. Microbiology of Ogi fermentation

During the fermentation of maize, sorghum or millet for ogi production, the pH of the medium decreases due to the production of carboxylic acids by microorganisms. Studies undergone by Akinrele [19]; Odufa [44] and Okafor [91] showed that the bacteria count is usually higher than that of the fungi indicating the predominance of bacteria in maize fermentation. *Lactobacillus plantarium* and *Corynebacterium* species are principally responsible for production of organic acid as reported by Inyang and Idoko [92]. The overall effect of steeping and souring is the development of the characteristic flavour through the production of carboxylic acids especially lactic, butyric and acetic acids [93]. The interaction of yeasts "*Saccharomyces cerevisiae* *Candida* and *Mycoderma*" lead to flavour development.

The predominant microorganism in Ogi fermentation include *Cephalosporium* species, *Fusarium* species, *Aerobacter*, *Cloacae*, *Sacharomyces cerevisiae*, *Lactobacillus plantarum*, *Pediococcus*, *Candida*, *Mycoderma*, *Lactobacillus fermentum* and *Lactobacillus cellibiose* [43].

### 10. Microorganisms of pito fermentation

Pito is a traditional beer produced in West Africa. The production involved soaking cereal grains (maize, sorghum or combination). During soaking, the microorganisms involved in the fermentation are *Geotrichum candidum*, *Lactobacillus* sp. and *Candida* sp. They are responsible for the alcoholic fermentation. Pito processing involves several stages which are malting, mashing, filtering, and fermenting. At each of the processes, there exist bacteria-fungi interaction with the substrate which break down the starch to fermentable sugars principally maltose and glucose [94] and further breakdown to release metabolite which could impact taste and flavour to food. Again the lactic acid produced promotes the microbiological safety and stability of the products [66] [95]. Yeasts and lactic acid bacteria are predominant in the fermentation.

### 11. Microorganisms in kunu fermentation

Kunu-zaki is a traditional non-alcoholic, fermented beverage in Nigeria [64] [96 - 97] processed mainly from Millet, Sorghum and Maize [98 - 99]. Kunu is rich in carbohydrate, vitamins and minerals but low in protein as reported by Gaffa *et al.* [100].

Microorganisms associated with the production of kunu-zaki using sorghum grains were isolated and characterized. They consisted mainly of lactic acid bacteria, *Lactobacillus plantarum*, *Leuconostoc mesenteroides*, *Corynebacterium*, *Lactococcus lactis*, *Pediococcus cerevisiae* and yeast *Saccharomyces cerevisiae*. The organisms occurred in various numbers at different stages of production but persisted to the end of the production process. The growth of lactic acid bacteria brought about a gradual decrease in pH of the steeping medium from 6.87 to 4.78 in the final product. Although it is clear that

specific microorganisms are involved in the production process, the chances of contamination seem to be unavoidable, as the process is not standardized and the processing environment is unhygienic [101].

Ikpoh *et al.* [102], reported that bacteria such as, *Escherichia coli*, *Staphylococcus aureus* *Streptococcus* sp., *Pseudomonas* sp., *Bacillus* sp., and fungi such as *Penicillium* sp., *Fusarium* sp., *Aspergillus* sp., *Candida albicans*, and *Rhizopus nigricans* were isolated in which their interaction (bacteria-fungi) cause spoilage of the drink as a result of fermentation which is one of the major factors considered during storage. If these organisms are present in large quantity they could cause the spoilage of Kunu beverages [103]. The presence of *Saccharomyces cerevisiae* was observed by Aboh and Oladosu [104] and that the high bacteria load in the drink was attributed to poor hygienic practices of the handlers and possible contamination from the utensils and water used for processing [105]. However, the micro flora of the finished product depends on the processing and storage conditions. The dominant microorganisms in the fermentation of kunu are *Saccharomyces cerevisiae* *Klebsiella* sp., *Staphylococcus aureus*, *Aspergillus niger*, *Lactobacillus fermentum* and *Penicillium* sp., reported by Aboh and Oladosun [104].

### 12. Microorganisms of Agidi fermentation

Agidi is a gel-like traditional fermented starchy food item produced from maize, millet or sorghum. The microorganisms of fermentation involve the interaction of bacteria and yeasts as reported by Osungbaro [105] Ogiehor, Ekundayo and Okwu,(128). The bacteria involved are *Bacillus* sp., *Staphylococcus* sp., *Streptococcus* sp., *Lactobacillus*, *Leuconostoc*, *Pseudomonas* and *Alcaligenes* while fungi involved are *Aspergillus*, *Penicillium*, *Alternaria*, *Fusarium*,

*Rhizopus*, *Mucor* and *Geotrichum* sp. These interactions lead to the release of metabolite (organic acid) by the breakdown of carbohydrate [106 - 107]. The interactions of these organisms also cause spoilage given rise to the release of off odour. Biosin and Wanley [106] and Hounhouigan *et al* [107] reported that mould such as *Aspergillus niger*, *A. flavus*, *Penicillium* sp., and *Rhizopus* sp., are responsible for agidi spoilage.

### 13. Microorganisms of Burukutu fermentation

Burukutu is a popular alcoholic beverage of vinegar-like flavour consumed in Northern Guinea savannah region of Nigeria, in the republic of Benin and Ghana. The processing involves malting, mashing and fermentation. The malted grains are sundried and grounded with part of water. The pH of the fermenting mixture decreases from about 6.4 to 4.2 within 24 h of fermentation and decreases further to 3.7 after 48 h. The predominant organisms of the fermentation are *Acetobacter* sp., and *Candida* sp. The interaction of these organisms with the substrate led to the production of metabolite which lower the pH and destroy organisms that could not withstand it, especially the pathogenic bacteria [105].

### 14. Conclusion

Ecology deals with the study of the interaction of organisms with each other and their environment. This gives the insight or ways in which microorganisms and food depend on one another. The ecological information helps to effectively manage the growth and activities of microorganisms in foods. The importance of ecological concepts in understanding the occurrence of growth of microorganisms in foods is well recognized by food microbiologists. These ecological principles are the foundation upon which

modern quality assurance, predictive modeling and risk analysis strategies have been developed to prevent outbreak of food spoilage and food born diseases. These are also based on the functional use of microorganisms in the production of fermented foods and beverages and for their use as probiotics and biocontrol agents.

Understanding the ecology of cereal fermentation reveals the dynamics of growth, survival and biochemical activities of microorganisms in food which is as a result of stress reactions in response to the changing physical and chemical conditions in the food microenvironment and the ability to colonize the food matrix and to grow into spatial heterogeneity, and the *in-situ* cell-to-cell ecological interaction which happens in a solid phase.

#### 15. Conflict of Interest

The author(s) report(s) no conflict(s) of interest(s). The author along are responsible for content and writing of the paper.

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NA

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