Fermentation of cereals for specific purpose

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Summary
Cereal grains are considered to be one of the most important sources of dietary proteins, carbohydrates, vitamins, minerals and fibre for people all over the world. Fermentation can be defined as a desirable process of biochemical modification of primary food matrix brought about by microorganisms and their enzymes. Fermentation of cereals leads to a general improvement in the shelf life, texture, taste and aroma, nutritional value and digestibility and significantly lowers the content of antinutrients of cereal products. Lactic acid fermentation of cereals is a long-established processing method and is being used in Asia, Africa and other countries for the production of foods in various forms such as beverages (boza, bouza) gruels (togwa) porridge (ogi, yosa) and mixtures (tarhana, balao balao).

Keywords
fermentation; cereals; microorganisms; fermented food

Modern food processing is dependent on a range of preservative technologies to ensure that food is maintained at an acceptable level of quality from the time of manufacture through to the time of consumption. One of the oldest of these technologies is fermentation, a process dependent on the biological activity of microorganisms for production of a range of metabolites which can suppress the growth and survival of undesirable microflora in foodstuffs [1]. Its importance in modern-day life is underlined by the wide spectrum of fermented foods marketed both in developing and industrialised countries, not only for the benefit of preservation and safety, but also for their highly appreciated sensory attributes [2] (Tab. 1).

Fermentation may be useful strategy for reducing bacterial contamination of food. This method of preservation could help to reduce the prevalence of diarrheal diseases [3]. Fermentation is a low-cost and the most economical technique of production and preservation of foods [4]. Fermentation plays at least five roles in food processing [5]:

1. Enrichment of the human dietary through development of a wide diversity of flavours, aromas and textures in food.
2. Preservation of substantial amounts of food through lactic acid, alcoholic, acetic acid and alkaline fermentations.
3. Enrichment of food substrates biologically with protein, essential amino acids, essential fatty acids and vitamins.
4. Detoxification during food fermentation processing.
5. A decrease in cooking times and fuel requirements.

Fermented foods and beverages are defined as those products that have been subjected to the effect of microorganisms or enzymes to cause desirable biochemical changes [6]. Main food products produced by fermentation are listed in Tab. 2, together with the raw materials used and the type of culture employed [7].

FERMENTATION OF CEREALS

Cereals are grown over 73% of the total world harvested area and contribute over 60% to the world food production providing dietary fibre, proteins, energy, minerals and vitamins required for human health [8]. However, the nutritional quality of cereals and the sensorial properties of their products are sometimes inferior or poor in com-

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Tab. 1. The benefit of food fermentation.

Raw material	Stability	Safety	Nutritive value	Acceptability
Meats	++	+	–	(+)
Fish	++	+	–	(+)
Milks	++	+	(+)	(+)
Vegetables	+	(+)	–	(+)
Fruits	+	–	–	++
Legumes	–	(+)	(+)	+
Cereals		–	(+)	+

++ - definite improvement, + - usually some improvement, (+) - some case of improvement, – - no improvement.

Tab. 2. Types of fermented foods with a long history of use in large geographical areas of the world.

Product	Raw material	Starter culture
Beer	Cereals	Yeast
Wine	Grape juice	Yeast, lactic acid bacteria
Bread	Grains	Yeast, lactic acid bacteria
Soy sauce	Soybeans	Mould (*Aspergillus*), lactic acid bacteria
Sauerkraut, Kimchi	Cabbage	Lactic acid bacteria
Fermented sausages	Meat	Lactic acid bacteria
Pickled vegetables	Cucumbers, olives a.o.	Lactic acid bacteria
Fermented milks	Milk	Lactic acid bacteria
Cheese	Milk	Lactic acid bacteria, yeast, mould

parison with milk and dairy products. The reasons behind this are the lower protein content, deficiency of certain essential amino acids, the presence of determined antinutrients (phytic acid, tanins and polyphenols) and the coarse nature [6].

Fermentation can have multiple effects on the nutritional value of food. Microbial fermentation leads to a decrease in the level of carbohydrates as well as some non-digestible poly- and oligosaccharides. The latter reduces side effects such as abdominal distention and flatulence. Certain amino acids may be synthesised and the availability of B group vitamins may be improved [9]. Fermentation of cereals by lactic acid bacteria has been reported to increase free amino acids and their derivatives by proteolysis and/or by metabolic synthesis. The microbial mass can also supply low molecular mass nitrogenous metabolites by cellular lysis [10]. Fermentation has been shown to improve the nutritional value of grains such as wheat and rice, basically by increasing the content of the essential amino acids lysine, methionine and tryptophan [11]. Fermentation of rice by lactic acid bacteria enhances the flavour, nutritive value and available lysine content [12].

Natural fermentation of maize increased total soluble solids and non-protein nitrogen and slightly increased protein content. Sixteen hour-fermented dough had higher levels of the albumin plus globulin fraction, indicating that natural fermentation of maize results in improvement in the nutritional value of the grain. Also maize protein digestibility is elevated [13].

Improvement in starch digestibility during fermentation can be related to enzymatic properties of fermenting microflora that brings about the breakdown of starch oligosaccharides. The enzymes bring about cleavage of amylase and amylopectin to maltose and glucose. Reduction in amylase inhibition activity may also be responsible for the starch digestibility. Similarly an improvement in protein digestibility of fermented products is mainly associated with an enhanced proteolytic activity of the fermenting microflora.

Fermentation is such an important process which significantly lowers the content of antinutrients (phytates, tanins, polyphenol) of cereal grains [14]. Fermentation also provides optimum pH conditions for enzymatic degradation of phytate which is present in cereals in the form of complexes with polyvalent cations such as iron, zinc, calcium, magnesium and proteins. Such a reduction in phytate may increase the amount of soluble iron, zinc, calcium several folds [6, 9].

During fermentation, the reduction of phytic acid content may partly be due to phytase activity as it is known to be possessed by a wide range of microflora. Optimal temperature for phytase activity has been known to range between 35 °C and 45 °C [14].

Tanin levels may be reduced as a result of lactic acid fermentation, leading to increased absorption of iron, except in some high tanin cereals, where little or no improvement in iron availability has been observed [9]. Diminishing effect of fermentation on polyphenols may be due to the activity of polyphenol oxidase present in the food grain or microflora [14].

Traditional fermented foods prepared from most common types of cereals (such as rice, wheat, corn or sorghum) are well known in many parts of the world. Some are utilized as colorants, spices, beverages and breakfast or light meal foods, while a few of them are used as main foods in the diet [6].

Lactic acid fermentation of cereals is a long-established processing method and is being used in Asia and Africa for the production of foods in various forms such as beverages, gruels and porridge. Fermented cereal based foods produced in Africa are presented in Tab. 3 [7]. Although differences exist between regions, the preparation procedure could be generalised. Cereal grains, mainly maize, sorghum, or millet grains are soaked in clean water for 0.5–2 days. Soaking softens the grains and makes them easier to crash or wet-mill into slurry from which hulls, bran particles, and germs can be removed by sieving procedures. During the slurrying or doughing stage, which lasts for 1–3 days, mixed fermentation including lactic acid fermentation takes place. During fermentation, the pH decreases with a simultaneous increase in acidity, as lactic and other organic acids accumulate due to microbial activity.

The traditional foods made from cereal grains usually lack flavour and aroma [8]. During cereal fermentations several volatile compounds are formed, which contribute to a complex blend of flavours in products. The presence of aromas represented by diacetyl, acetic acid and butyric acid makes fermented cereal-based products more appetizing (Tab. 4) [6]. The proteolytic activity of fer-

Tab. 3. Fermented cereal-based foods in Africa.

Name of product	Associated microorganisms	Substrate	Area of products
Ogi	*Lactobacillus plantarum, Lactobacillus fermentum, Lactobacillus* spp.*, Saccharomyces cerevisiae, Candida mycoderma, Rhodotorul*a spp.*, Aerobacter cloacae, Candida krusei, Debaromyces hasenu, Klebsiella* spp., *Staphylococcus* spp.	Maize, sorghum or millet	West Africa
Kolo and Kenkey	*Lactobacillus* spp. and yeasts	Maize, sorghum or millet	Ghana
Mahewu (magou)	*Lactobacillus delbrueku* and *Lactobacillus bulgaricus*	Maize, sorghum or millet	South Africa
Uji	*Lactobacillus* spp.	Maize, sorghum or millet	East Africa
Kisra	Information not available	Sorghum	Sudan
Enjara	*Candida guillienmandi*	Sorghum	Ethiopia

Tab. 4. Compounds formed during cereal fermentation.

Organic acids		Alcohols	Aldehydes and ketones	Carbonyl compounds
Butyric	Heptanoic	Ethanol	Acetaldehyde	Furfural
Succinic	Isovaleric	*n*-propanol	Formaldehyde	Methional
Formic	Propionic	Isobutanol	Isovaleraldehyde	Glyoxal
Valeric	*n*-Butyric	Amyl alcohol	*n*-Valderaldehyde	3-Methyl butanal
Caproic	Isoburyric	Isoamyl alcohol	2-Methyl butanol	2-Methyl butanal
Lactic	Caprylic	2,3-Butandieol	*n*-Hexaldehyde	Hydroxymethyl furfural
Acetic	Isocaprilic	β-Phenylethyl alcohol	Acetone	
Capric	Pleagronic		Propionaldehyde	
Pyruvic	Mevulinic		Isobutyraldehyde	
Palmitic	Myristic		Methyl ethyl ketone	
Crotonic	Hydrocinnamic		Butanone	
Itaconic	Benzylic		Diacetyl	
Lauric			Acetoin	

mentation microorganisms often in combination with malt enzymes may produce precursors of flavour compounds, such as amino acids, which may be deaminated or decarboxylated to aldehydes and these may be oxidized to acids or reduced to alcohols [15]. Knowledge of biochemical pathways leading to flavour production can help in making the right choice of starter culture. However, the end product distribution of lactic acid fermentations depends also on the chemical composition of the substrate (carbohydrate content, presence of electron acceptors, nitrogen availability) and the environmental conditions (pH, temperature, aerobiosis/anaerobiosis), controlling of which would allow specific fermentations to be channelled towards a more desirable product [8].

MICROORGANISMS FOR CEREAL FERMENTATIONS

The basic fermentation process involves the enzymatic activities of lactobacilli, leuconostoc, pediococci, yeasts and moulds. Their metabolic activities result in the production of short chain fatty acids such as lactic, acetic, butyric, formic and propionic acids. The pH of these foods is reduced to values of 4 or less [16]. Acids formed during fermentation process, lower the pH thus inhibiting the growth of spoilage organisms [17]. The pH requirements of a number of pathogens are shown in Tab. 5 [16].

Lactic acid bacteria are industrially important microbes that are used all over the world in a large variety of industrial food fermentations [18]. The primary activity of the culture in food fermentation is to convert carbohydrates to desired metabolites such as alcohol, acetic acid, lactic acid or CO_2. The cultures used in food fermentation are, however, also contributing by secondary reactions to the formation of flavour and texture [7]. Lactic acid bacteria are considered to have several beneficial physiological effects, such as antimicrobial activity, enhancing of immune potency [19] and prevention of cancer and lower serum cholesterol levels [20]. The proposed health and nutritional benefits of *Lactobacillus* species are [21]:

- enzyme (lactase) formation,
- colonization and maintenance of the suitable intestinal microflora,
- competitive exclusion of undesirable microorganisms,
- microbial interference and anti-microbial activities,
- pathogen clearance,
- immuno-stimulation and modulation,
- cholesterol reduction/removal.

Lactobacillus spp. play a significant role in most of the fermented cereals [17]. The *Lactobacillus bulgaricus* and *Streptococcus thermophillus* are suitable bacteria for rice fermentation because of their lack of amylase, which is necessary for saccharification of rice starch [12]. It was shown that *Lactobacillus plantarum* and *Pediococcus* spp. dominate the latter stages of maize dough fermentation and may therefore be responsible for the rapid acidification of inoculated dough [22]. Mixed culture of lactic acid bacteria and propionic bacteria appeared to be a potential alternative along with conventional starters in rye sour dough fermentation, providing for an improved mould-free shelf-life of bread [23].

In case when the cereal grain are used as natural medium for lactic acid fermentation, amylase need to be added before or during fermentation or amylolytic bifidobacteria need to be used because these microorganisms contain enough amylase which is necessary for saccharification of the grain starch [24]. The use of amylolytic lactic acid bacteria as a starter culture offers another alternative by combining both amylase production and acidification in one microorganism [25].

Fermentation of cereals with pure yeast cultures has been shown to increase the protein content of the fermented products [26]. The possible function of yeasts in fermented foods and beverages are [27]: – fermentation of carbohydrates (formation of alcohols etc.),

- production of aroma compounds (esters, alcohols, organic acids, carbonyls, etc.),
- stimulation of lactic acid bacteria providing essential metabolites,
- inhibition of mycotoxin-producing moulds (nutrient competition, toxic compounds, etc.),
- degradation of mycotoxins,

Tab. 5. Approximate pH tolerance of some microorganisms.

Organism	Minimum pH	Maximum pH
Escherichia coli	4.4	9.0
Salmonella typhi	4.5	8.0
Campylobacter jejuni	2.3	
Shigella sp.	4.5	8.0
Streptococcus lactis	4.3–4.8	
Lactobacillus sp.	3.0	7.2
Yeasts	1.5	8.0–8.05
Moulds	1.5–2.0	11.0

- degradation of cyanogenic glucosides (linamarase activity),
- production of tissue-degrading enzymes (cellulases and pectinases) and
- probiotic properties.

It has been suggested that the proliferation of yeasts in foods is favoured by the acidic environment created by lactic acid bacteria while the growth of bacteria is stimulated by the presence of yeasts, which may provide growth factors, such as vitamins and soluble nitrogen compounds. Association of lactic acid bacteria and yeasts during fermentation may also contribute metabolites, which could impart taste and flavour to fermented food [28].

EXAMPLES OF FERMENTED CEREAL FOODS

Tarhana

Popular Turkish fermented wheat-yogurt mixture produced from white wheat flour, yogurt, onions, tomato puree, yeast (baker's yeast), salt, paprika, dill, mint, tarhana otu (*Echinophora sibthorpiana*) and water. The mixture is fermented for several (1–7) days at 30 °C. The final pH of tarhana varied in the range 4.3–4.8 [29].

Togwa

Fermented gruel or beverage prepared either from cassava, maize, sorghum, millet or their combinations [28]. Togwa is widely produced in Tanzania for use directly as a weaning food or diluted for use as refreshment. The fermentation is spontaneous [15] or the lactic acid bacteria (*Lactobacillus brevis, Lactobacillus cellobiosus, Lactobacillus fermentum, Lactobacillus plantarum, Pediococcus pentosaceus*) and yeasts (*Candida pelliculosa, Candida tropicalis, Saccharomyces cerevisiae*) are used at concentration 10^9 CFU.ml^{-1} and 10^7 CFU.ml^{-1} [28]. Fermentation is carried out at ambient temperature for 9–24 h (to pH ≤ 3.8) [15].

Ogi

Sour porridge with a flavour resembling yogurt. It is made by lactic acid fermentation of corn, sorghum or millet. Soybeans may be added to improve nutritive value [30]. It is considered the most important weaning food for infant in West Africa although it is also consumed by adults. Ogi is a natural fermentation product [6]. Lactic acid bacteria, yeasts and moulds are responsible for the fermentation although *Lactobacillus plantarum* is the predominant microorganism. Other bacteria such as *Corynebacterium* hydrolyse corn starch, and then yeasts of the *Saccharomyces cerevisiae* and *Candida* species also contribute to flavour development [6, 30]. Optimum pH for ogi is 3.6–3.7. The concentration of lactic acid may reach 0.65% and that of acetic acid 0.11% during fermentation [30].

Yosa

New snack food made from oat bran pudding cooked in water and fermented with lactic acid bacteria and *Bifidobacteria.* After fermentation, the matter is flavoured with sucrose or fructose and fruit jam. It is mainly consumed in Finland and other Scandinavian countries. Oat fibre is a good source of β-glucan, which can lower the cholesterol levels in the consumer blood, which in turn can reduce the risk of heart disease [6].

Balao balao

Philippine balao balao is a lactic acid fermented rice/shrimp mixture prepared by mixing boiled rice, whole raw shrimp and solar salt (about 3% w/w), packing in an anaerobic container and allowing the mixture to ferment over several days or weeks [5]. The chitinous shell becomes soft and when the fermented product is cooked, the whole shrimp can be eaten [30].

Boza

Highly viscous traditional fermented Turkish beverage made from cereals such as maize, rice and wheat flours. The preparation of boza is normally carried out by natural fermentation involving mixed cultures of lactic acid bacteria and yeasts [31].

Alcoholic cereal based fermented beverages can be classified into wines (sake-rice wine consumed in Japan and China, bouza-alcoholic wheat beverage consumed in Egypt and Turkey) and beers (European barley beer, rice beer in the Asia-Pacific countries) [6].

Bouza

Alcoholic wheat beverage common in Egypt. It is a thick, pale yellow liquid with 3.8–4.0% v/v alcohol and a pH 3.1–4.0. The microorganisms responsible for the fermentation are yeasts and bacteria [17].

Lactic fermentation of bread dough improves the keeping quality and flavour of the baked products. It also enhances the palatability of bread made from low-grade flours and under-utilized cereals [32]. The basic biochemical changes that occur in the sour-dough bread fermentation are:

1. acidification of the dough with lactic and acetic acids produced by the lactobacilli and
2. leavening of the dough with CO_2 produced by the yeast and the lactobacilli [30].

CONCLUSION

Fermented foods can be defined as foods in which microbial activity plays an essential role in conferring the required stability, safety and sensory properties to the product [33]. A benefit of nutritional effects of lactic acid fermentation (degradation of anti-nutritional factors and increased mineral bio-availability, improvement of protein digestibility of tannin-rich cereals, and degradation of flatulence-causing oligosaccharides) is clearly evident. Reduction of viscosity of starchy porridges is reached by addition of germinated cereal grains; this enables the preparation of porridges of increased nutrient density that are still sufficiently liquid to be swallowed by infants. Moreover, lactic acid fermentation enhances considerably sensory properties of food resulting in a variety of tastes [9].

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