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Review

Probiotic potential of spontaneously fermented cereal based foods – A review

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The realization that food has a role beyond provision of energy and body forming substances has shifted scientific investigations with growing interest in the research and development of functional foods. A lot of attention is being focused on probiotics due to the enormous support showing health benefits. Probiotics are associated with fermented foods and it is therefore of importance that spontaneously fermented foods, that are so common in Africa, be assessed for their probiotic attributes. These foods are within the economic means of the people and are widely accepted in populations where they are produced. The foods have relatively long shelf-lives under ambient temperatures (without spoiling), are widely accepted especially by the vulnerable groups such as children, expectant/breastfeeding mothers, the aged and are commonly served to the sick and recovering persons. These foods would therefore render an invaluable health benefit to communities when consumed and would impart an invaluable economic benefit to society. Probiotics are associated with lactic acid bacteria that are commonly found in fermented foods. This paper reviews studies that have been carried out to enumerate, isolate, characterize and identify the microorganisms involved in the spontaneous fermentations of cereal based products and their assessments for potential probiotic attributes.

Key words: Functional foods, fermented cereal foods, Lactic acid bacteria, probiotics, prebiotics, fructooligosaccharides, exopolysaccharides

INTRODUCTION

Fermentation is one of the oldest technologies used for food preservation. It is a technology depended upon by millions of people in the developing world for preservation of their food at costs available to the average consumer. At present, a variety of fermented foods are produced all over the world at household as well as industrial level; in both small scale and large commercial enterprises. In some regions, especially in Africa, these foods are used as weaning foods for infants and young children (Jakobsen and Lei, 2004; Kalui et al., 2008).

Little has been done to develop as well as upgrade fermented products and especially so in terms of quality, consistency, functionality, safety and shelf life (Rolle and Satin, 2000). There has been little exploration and exploitation of technologies used in their production with an aim of adding value to these fermented products.

Through research the fermentation process has been found to produce flavor enhancing compounds, useful enzymes and essential amino acids. Some fermentation microorganisms have been found to produce anti-microbial products that lead to safe and long storing of foods (Corgan et al., 2007; Kalui et al., 2009; Kalui et al., 2008; Parvez et al., 2006; Steinkraus, 2002). Associated with fermentation are useful microorganisms, referred to as probiotics, which have been shown to have health benefits to the human body. These health enhancing microorganisms bring about fermentation resulting in production of lactic acid and hence commonly referred to as Lactic acid bacteria (LAB) (Holzapfel and Schillinger, 2002; Shah, 2007). Most of them are generally regarded as safe (GRAS). It is imperative therefore that research and technology is now focusing a lot of attention on fermentation technologies and their products with an aim

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Abbreviations: GIT, Gastro-intestinal tract; **LAB,** lactic acid bacteria; **GRAS,** generally regarded as safe; **KSW,** koko sour water; **DP,** degrees of polymerization; **FOS,** fructooligosaccharide; **EPS,** expolysacharide.

of tapping into the possible associated health benefits.

The desire for quality life, the increasing cost of healthcare, the preference for naturally preserving as well as health enhancing foods are driving factors for research and development in the area of functional foods. Among a number of functional properties attributed to foods, probiotics (microorganisms providing health benefits to the host) take a centre stage (Grajek et al., 2005; Reid, 2008; Uyeno et al., 2008; Vasiljevic and Shah, 2008). In 1994, the World Health Organisation (WHO) deemed probiotics to be the next most important immune defence systems as a result of increasing antibiotic resistance of commonly prescribed antibiotics, (Levy, 2000). Probiotics are associated with fermented foods, latter having a long tradition of acceptability in communities where they are produced, safe use and the established as well as postulated claims of health benefits (Lei, 2008; Vasiljevic and Shah, 2008). Researches have been carried out on spontaneously fermented foods with an aim of identifying predominant microorganisms involved in the fermentation process. Some of these researches have further characterised these microorganisms for their probiotic potential. Considering the position of spontaneously fermented foods in Africa, there is need for information regarding the probiotic potential of these microorganisms associated with fermented traditional foods. This information can be utilized to explore and exploit this traditional technology with an aim of producing consistent, safe, long storing products that in addition to the normal functionality of foods, also impart health benefits to the consumers. This paper aims to provide a review of potential probiotic attributes of lactic acid bacteria isolated from spontaneously fermented products with an aim of providing basic information that could be utilized to advance further the research on microorganisms associated with spontaneous fermentations for the probiotic attributes, technical properties and food safety requirements.

Functional foods

Scientific investigations have changed the view of the role of food as being beyond the provision of energy and body forming substances to having the extra role of possessing active substances that impart health benefits to the consumer (Grajek et al., 2005). Foods are now known to contain bioactive substances that prevent the initiation, promotion and development of allergies, diseases such as cancer, cardiovascular diseases, diarhoea, osteoporosis, among others (Sanders, 2003, Pisulewski and Kostogrys, 2003; Parvez et al., 2006; Lei et al., 2008; Vasiljevic and Shah, 2008). This has led to the emergence of interest in functional foods which are defined as a part of an everyday diet and are demonstrated to offer health benefits and to reduce the risk of chronic diseases beyond the widely accepted nutritional effects. Functional foods include: i) conventional foods that contain naturally occurring bioactive substances such as dietary fiber, ii) foods enriched with bioactive substances such as probiotics, antioxidants, iii) synthesized food ingredients introduced to traditional foods such as prebiotics. Functional components in functional foods include probiotics, prebiotics, soluble fiber, polyunsaturated fatty acids, antioxidants, vitamins, minerals among others (Grajek et al., 2005). Functional foods are not prescribed but are consumed as part of a normal everyday diet. Health benefits associated with functional foods include reduction of the risk of cancer, improvement of cardiovascular health, boosting of immune system, improvement of gastrointestinal health, maintenance of urinary tract health, anti-inflammatory effects, reduction of blood pressure, antibacterial and antiviral activities, anti-obese effects, reduction of esteoporosis, maintenance of vision, among other benefits (Grajek et al., 2005; Nissen et al., 2009; Parvez et al., 2006; Shah, 2007). Biotechnology is playing a key role in development of functional foods and of these, probiotics have taken centre stage. Probiotics are mainly associated with fermented products.

Fermentation

Research findings have brought to light the invaluable attributes of fermented food products. It is now known that fermentation process leads to production of valuable products including flavour and aroma compounds; biomass proteins/amino acids; minerals; lipids; carbohydrates; vitamins and other products of the respiretory/biosynthetic process such as lactic acid, ethanol, acetylaldehydes, pyruvic acid, which help in altering the pH of food to levels that do not favor growth of pathogenic microorganisms (Au and Fields, 1981; Steinkraus, 1996; Baghel et al., 1985; Deshpande and Salunke, 2000; Beaumont, 2002; Annan et al., 2003; Kalui et al., 2009). This in turn enhances food safety and increases food shelf life hence aiding in food preservation (Yasmine, 2002).

The changes associated with the fermentation process are as a result of action of enzymes produced by microorganisms (Pederson, 1979; Steinkraus, 2002). Fermentation could lead to reduction of toxic products (Steinkraus, 1983; Dhanker and Chauhan, 1987) and has been reported to improve the bioavailability of minerals such as iron and zinc by significantly reducing the phytate compounds present in fermented cereals (Sankara and Deosthale, 1983). Fermentation leads to production of acids and probable bacteriocins that prevent growth of microorganisms hence increasing shelf life of fermented products (Mbugua and Njenga, 1991; Chen and Hoover, 2003; Kalui et al., 2009). This is a very valuable attribute especially in rural areas where advanced food preservation technologies such as refrigeration are not affordable and considering that people are beginning to appreciate more of naturally preserved than chemically preserved foods (Rolle and Satin, 2000).

Fermented foods are associated with 'good bacteria' referred to as probiotics (Patricia et al., 2002; Helland et al., 2004). Probiotics are beneficial bacteria in that they favorably alter the intestinal microflora balance, inhibit the growth of harmful bacterial, promote good digestion, boost immune function and increase resistance to infection (Reid, 1999; Patricia et al., 2002; Helland et al., 2004). People with flourishing intestinal colonies of beneficial bacteria are better equipped to fight the growth of disease causing bacteria (Reid et al., 2003; Holzapfel and Schillinger, 2002; Helland et al., 2004). Examples of probiotics that have found application in probiotic products include some strains of *Lactobacillus* genera (*L. plantarum, L. rhamnosus, L. acidophilus, L. reuteri, L. gasseri,* and *L. amylovorus*); *Bifidobacterium* genera *(B. adolescentis, B. animalis, B. bifidum, B. breve, B. infantis, B. lactis,* and *B. longum); Enterococcus (E. faecalis,* and *E. faecium)* (Hozapfel and Schillinger, 2002). Species of the genera *Lactobacillus* are the most widely studied for probiotic attributes (Mishra and Prasad, 2005).

Fermentation organisms

Changes occur during fermentation that are as a result of the activity of microorganisms: bacteria, yeast and moulds (Steinkraus, 2002). The most important group of microorganisms involved in the spontaneous or natural fermentation of foods are the lactic acid bacteria (Steinkraus, 2002; Jakobsen and Lei, 2004). Lactic acid bacteria are Gram positive, non-spore forming catalase negative cocci or rods that are anaerobic, micro-aerophilic or aero-tolerant (Wood and Holzafel, 1995). These microorganisms produce lactic acid as the sole, major or an important product from the energy yielding fermentation of sugars (Wood and Holzapfel, 1995; Steinkraus, 2002). They include the genus *Lactobacillus, Lactococcus, Pediococcus, Leuconostoc, Bifidobacterium,* some *Enterococcus* and some *streptococcus (*Wood and Holzafel, 1995; Steinkraus, 2002). Some yeast such as saccharomyces and moulds such as penicillium, aspergillus and botrytis too produce lactic acid (Wood and Holzafel, 1995).

The lactic acid fermentations provide foods that have a variety of flavors, aromas, textures in addition to the foods being safe and having a long shelf-life. However, not all lactic acid bacteria are useful; some are involved in food spoilage due to action of proteinases and lipases that degrade proteins and lipids, respectively, producing by-products that cause off-flavours. *Pediococcus damnosus* has been reported to produce off-flavour in beer while *L. bifermentans* and *L. alimentarius* are associated with spoilage of refrigerated and packaged foods. Some are pathogenic especially species belonging to the genera *Streptococcus* (cause throat infections*)* and *Enterococcus* (eg. *E. faecum causes urinary tract infections and abdominal septis*) (Wood and Hozapfel, 1995).

Lactic acid bacteria important in food technology include those of the genera *Lactobacillus, Lactococcus, Pediococcus,* and *Leuconostoc* (Harrigan and McCance, 1990). *L. fermentum, Pediococcus pentosaceus, W. confusa, L. plantarum, L. salivarius, L. casei, L. acidophilus,* and *Leuconostoc* spp are some species that have been reported isolated from cereal based fermented foods (Mensah, 1990; Olasupo et al., 1997; Anthony and Chandra, 1997; Jacobsen and Lei, 2004; Achi, 2005; Kalui et al., 2009). Examples of *Lactobacillus* spp involved in lactic acid bacteria fermentation of cereal based fermented foods include *L.* p*lantarum, L. casei, L. sakei, L. acidophilus,* and *L. salivarius* among others (Jacobsen and Lei, 2004; Achi, 2005; Kalui et al., 2009). Kalui et al. (2009) reported isolation of *L. Fermentum, P. Pentosaceus, L.* P*lantarum, W. confusus* and *L. Rhamnosus* from ikii, a traditional fermented maize porridge in Kenya. In this product, *L. Fermentum* was the predominant species (43% of isolates), followed by *P. Pentosaseus* which formed 38% of the isolates; *L.* P*lantarum* formed 10%, *W. Confusus* formed 8% and least was *L. Rhamnosus* which formed 1% of the total isolates.

Lactobacillus fermentum have been reported as the predominant species in kisra a Sudanese sorghum fermented flat bread (Hamad et al., 1992) and kenkey a Ghanaian maize dough (Halm et al., 1993). *L. fermentum* and *L. plantarum* have been reported to be the most commonly associated lactic acid bacteria species with spontaneous lactic acid fermentations of cereal products (Kunene et al., 2000; Jacobsen and Lei, 2004). In their studies of koko, Jacobsen and Lei (2004) reported levels of *P. pentosaceus* ranging from 8.3 to 33.3%. *Lactobacillus confusus* has been isolated in low numbers from mawe (Hounhouigan et al., 1994), togwa (Mugula, 2003) and wheat sourdough (Corsetti et al., 2004) and bushera (Muyanja et al., 2002). Jacobsen and lei (2004) reported isolation of *Lactobacillus fermentum, Pediococcus pentosaceus, Lactobacillus confusus and Lactobacillus paraplantarum* from koko, a fermented millet product.

Lactic acid bacteria may be homofermentative and/or heterofermentative. *Lactococcus* are homofermentative, producing only lactic acid from glucose. They also produce ammonia from arginine and examples include *L. lactis* subsp *lactis, L. lactis subsp diacetylactis and L. cremoris. Leuconostoc* are heterofermentative producing lactic acid, carbon dioxide and aroma compounds, examples of which incude *Leuconostoc mesenteroides, L. cremoris, L. dextranicum* among others. *Lactobacillus* is both homofermentative and heterofermentative. Some require low while others require high temperatures for optimum growth.

Lactic acid bacteria have been claimed to have health benefits. These include antimicrobial effects against pathogenic bacteria, anti-tumor effects and protection against diarrhea associated with antibiotics or food allergy (Holzapfel and Schillinger, 2002; Desai et al., 2004). They have been reported to be acid and bile tolerant and hence are able to survive in the gastro-intestinal tract (GIT). Lactic acid bacteria can therefore help enhance the gut bacterial population, estimated to be at a concentration of 5 x 10^{11} bacterial cells per gram (Holzapfel and Schillinger, 2002). Gut flora are associated with health benefits to the host. As a result, these health enhancing bacteria have been termed as 'probiotics'. A lot of attention is being given to lactic acid bacteria fermented foods as potential sources of 'probiotics' that can enhance the balance of intestinal flora hence increasing the health benefits associated with them (Holzapfel and Schillinger, 2002; Helland et al., 2004; Jacobsen and Lei, 2004; Desai et al., 2004). Some examples of species that have been reported to possess probiotic attributes include *L. plantarum, L. acidophilus, L. rhamnosus, L. reuteri* and *L. casei* (Parves et al., 2006).

Antimicrobial Activity

In the FAO/WTO draft guidelines of 2002 on evaluation of probiotics in food, antimicrobial activity against potentially pathogenic microorganisms was one of the recommended attributes for potential probiotic strains. Lactic acid bacteria have been reported to produce antimicrobial products and exert a strong antagonistic activity against food contaminating microorganisms (De martinis et al., 2001; Ogunbanwo, 2003; Hernandez et al., 2004). These antimicrobial products include organic acids, hydrogen peroxide, diacetyl, antifungal fatty acids, phenyllactic acid, bacteriocins and bacteriocin-like products (Lavermicocca et al., 2000; Messens and De Vuyst, 2002; Corsetti et al., 2004). As a result, lactic acid bacteria fermentations produce foods that may be free of pathogenic microorganisms (hence safe) and that can naturally be preserved or stored for long (Ogunbanwo et al., 2003; Corsetti et al., 2004). In the presence of acids and a low pH, spoilage and pathogenic microorganisms that may invade the food product are less able to do so (Steinkraus, 2002).

In our study of ikii, a traditional fermented maize porridge in Kenya, the pH was found to decrease to 3.9, initial count of coliforms of 4.17 X 10^{3} decreased and were not detected by the end of 24 h of the fermentation process. In another study (Kalui et al., 2009), *Lactobacilli* isolates from ikii showed antimicrobial effect against *E. faecalis, S. aureus* and *E. coli*, all of which are food contaminants and pathogens. Antagonism was observed to be highest against *E. faecalis* which is a lactic acid bacteria and least against *E. coli* which is gram negative and not a lactic acid bacteria. Some strains did not show any antagonism against *E. coli. A*ntimicrobial activity is reported to be highest against closely related species (De martinis et al., 2001; Ogunbanwo, 2003; Hernandez et al., 2004) and this may have been the reason for antagonism being highest against *E. faecalis* and least for *E. coli*. Jacobsen and Lei (2004) reported a decrease in pH to a level of 3.7 and little or no antimicrobial activity against *L. sakei and L. innocua*, in Koko, a spontaneously fermented porridge in Ghana. Muyanja et al. (2002) reported production of organic acids including lactic, citric, pyruvic, succinic, acetic and pyroglutamic acids in bushera, a traditional fermented sorghum based beverage in Uganda. Mbugua (1991) reported a decrease of coliforms to non-detected levels in fermented Uji.

Bacteriocins have been associated with lactic acid bacteria in fermented food products. Bacteriocins are antimicrobial peptides or small proteins which inhibit microorganisms usually of closely related species to the producer strain (Holzapfel, 2002; Ogunbanwo et al., 2003; Corsetti et al., 2004). Many bacteriocins have been found to be active against food-borne pathogens such as *Listeria monocytogenes, Staphylococcus aureus, Bacillus cereus* and *Clostridium dificile* (Hozapfel, 2002). Bacteriocins are classified into three categories: Class 1 consisting of small heat-stable peptides and referred to as lantibiotics due to possession of thioether amino acids such as lanthionine; Class 11, small hydrophobic heat-stable non-modified and consisting of a single peptide, two or more polypeptides; Class 111 consisting of large, hydrophilic, heat-labile proteins. Examples of class 1 bacteriocins include nisin, lactocins and pediocin PA-1/ACH produced by *L. lactis, L. sake* and *P. acidilacti,* respectively; class 11 include lactociccin G plantaricin-s, plantaricin-jk and enterococcin produced by *L. lactis, L. plantarum* and *E. facecum,* respectively; class 111 include helveticin V-1829 and helveticin J produced by *L. helveticus* (Chen and Hoover, 2003; Corsetti et al., 2004).

Several types of bacteriocins from food-associated lactic acid bacteria have been identified and characterized, some of which include nisin, diplococcin, acidophilin, bulgarican, helveticins, lactacins and plantaricins. Nisin is now commercially available and has a long history of safe use in food preservation due to its effectiveness against gram -positive food pathogens and spoilage agents (Chen and Hoover, 2003; Ogunbanwo et al., 2003). Bacteriocins have been considered as potential natural food preservatives and a lot of attention is being focused on these especially in this era of preference for naturally to chemically preserved foods (Chen and Hoover, 2003).

Some lactic acid bacteria such as *Leuconostoc mesenteriodes susp cremoris* are able to produce bacteriocin-like inhibitory compounds. Reuterin is a well known bacteriocin-like antimicrobial substance produced by *Lactobacillus reutiri* and inhibitory against a wide spectrum of bacteria, moulds and yeasts. Plantaricin c is a bacteriocin produced by *L. plantarum* and has been reported active against different species of *Enterococcus, Lactobacillus, Streptococcus*, *Bacillus,* and *Clostridium*

(Hozapfel, 2002; Chen and Hoover, 2003). In our study of antimicrobial effect of isolates from ikii (Kalui et al., 2009), the strains showed antimicrobial activity, which in some instances may have been beyond the effect of organic acids. Perhaps some strains were able to produce bacteriocins since there was a demonstration of loss of growth inhibition in the presence of proteolytic enzymes. The findings however necessitated further studies to be carried out to ascertain the possibility of production of bacteriocins by microorganisms involved in spontaneous fermentation of ikii since there was no antimicrobial activity observed in this study after neutralization of supernatant to pH 7 irrespective of absence of proteolytic enzymes.

Probiotics

Probiotics have been defined as living bacteria and supportive substances that have beneficial effects on the host by improving the bacterial balance in the intestine (Fuler, 1991). This definition was later expanded to include living bacteria or mixed bacteria that have beneficial effects on the gastrointestinal and respiratory system of the host by improvement of the balance of intestinal flora (Salminen et al., 1998). Recently, probiotics have been more widely defined as bacteria that work to maintain the host's health (Hozapfel, 2002; Saito, 2004; Grajek et al., 2005).

Lactobacillus and *Bifidobacteria* are examples of genera of which some of the species are promising probiotics (Saito, 2004). These microorganisms are gram-positive lactic acid producing bacteria that constitute a major part of the normal intestinal microflora in animals and humans (De Simone et al., 1993; Smirnov et al., 1993). Others with probiotic characteristics include some species of the genera *Enterococcus, Lactobacillus rhamnosus GG* and a variant of *L. caseis* spp. *Rhamnosu,* which is an extensively studied probiotic that has been shown to be effective in reducing the severity and duration of diarrhea (Jakobsen and Lei, 2004; Saito, 2004).

Probiotics play a key role in enhancing resistance to colonization by exogenous, potentially pathogenic organisms (Reid et al., 1999; Elmer et al., 1996; Holzapfel and Schillinger, 2002; Helland et al., 2004). They do this by producing compounds such as lactic acid, hydrogen peroxide and acetic acid that increase acidity of the intestine and inhibit the reproduction of many harmful bacteria. Probiotics also produce bacteriocins, which act as natural antibiotics that kill undesirable/pathogenic microorganisms. They are also known to out compete the pathogenic microorganisms hence preventing latter's survival in the GIT (Reid et al., 1999; Holzapfel and Schillinger, 2002; Helland et al., 2004).

In addition, the following properties and functions have been attributed to probiotics: they adhere to host epithelial tissue; they are acid resistant and bile tolerant; they are safe, non-pathogenic and non-carcinogenic; they cause improvement of the intestinal microflora; they have a cholesterol lowering, immunostimulating and allergy lowering effect; synthesize and enhance the bioavailability of nutrients (Ouwehand et al., 2002; Saito, 2004; Grajek et al., 2005; Parvez et al., 2006).

Potential probiotic lactic acid bacteria therefore attribute to these characteristics. They should be safe, viable in delivery vehicles, resistant to acid, tolerant to bile, have ability to produce antimicrobial substances, adhere to epithelial tissue, colonize the GIT, stimulate a host immune response and influence metabolic activities such as vitamin production, cholesterol assimilation and lactose activity (Soomro et al., 2002). Probiotic prepara-tions of *L. reuterii* have been found beneficial in the prevention and treatment of infantile viral diarrhea and antibiotic associated diarrhea (Salminen et al., 1998; Pariyaporn et al., 2003). *Lactobacillus casei* when administered orally was found to reduce recurrence of superficial bladder carcinoma in humans (Aso et al., 1995). There have been reports of improved production of immune factors such as immunoglobulins, interferons, interleukins, tumour necrosis factors and an increased phagocytic activity by some administered LAB bacteria (Kaila et al., 1992). Examples of most commonly used strains used in probiotic compositions include those of species *L. plantarum, L. rhamnosus, L. acidophilus, L. salivarius, l. reuteri and E. faecum* (Parvez et al., 2006).

There is need to carry out research regarding probiotic attributes of spontaneously fermented foods. More than 2 litres of gastric juice is secreted each day into the stomach (Morelli, 2000). This gastric juice renders the stomach pH to approximately 2.0 (Murthy et al., 2000). For strains to survive and colonize the gastrointestinal tract, microorganisms should express tolerance to acid and bile salts (Gibson et al., 1998). Potential probiotics should therefore be able to tolerate a low pH of 2. Researchers have suggested that the survival of probiotics both *in vitro* and *in vivo* is strongly influenced by the food used for their delivery (Charteris et al., 1998; Erkkila and Petaja, 2000; Dunne et al., 2001; Charalampopoulos et al., 2002a; Patel et al., 2004). Morelli (2000) suggested that food intake could protect bacteria during gastric passage. The buffering capacity, pH as well as the physical and chemical characteristics of a food carrier have been shown to have significant influence on microbial survival of potential probiotics (Patel et al., 2004). Charteris et al. (1998b) proposed the need for consideration of inclusion of foods during *in vitro* tests. Huang and Adams (2004) reported a significantly enhanced survival of potential probiotic dairy propionibacteria strains in pH 2.0 on addition of soymilk.

Jacobsen and Lei (2004) and Kalui et al.(2009) reported acid and bile tolerance of LABs isolated from koko and ikii, respectively, both of which are spontaneously fermented cereal based porridges. This indicates that the LAB isolates are able to withstand the physiological challenges of the GIT and have the potential of surviving and colonizing the GIT. There is need to carry out further investigations as to the ability for LABs involved in spontaneous fermentations to survive and colonize the GIT. This will necessitate investigations such as simulated stomach duodenum passage, hydrophobicity characteristics, adhesion to human cell linings and binding characteristics to human extracellular matrix.

Lei et al. (2006) carried out a research on effect of koko sour water (KSW) on diarrhea and though they reported children that received KSW being better than those that did not, any effect in reducing diarrhea by use of spontaneously fermented cereal foods is yet to be proven. Amylase digested and fermented porridge was found to be more effective than conventional porridge in the repair of mucosal damage after acute diarrhea (Willumsen et al., 1997). The prevalence of feacal enteric bacteria, such as *Salmonella, Shigella, and E. coli,* in young children was found to be significantly less in children receiving fermented maize gruel than those who were not (Nout et al., 1991; Mensah et al., 1991; Tetteh et al., 2004). There is convincing evidence that certain probiotic strains are effective in preventing and treating acute diarrhea hence there is prospective potential of producing a traditional fermented product with diarrhea treatment attributes. The improved keeping qualities, elimination of coliforms during fermentation process indicate that traditionally fermented foods have an important role in preventing acute diarrhea. Perhaps there will be a need to determine specific probiotic cultures that affect diarrhea and utilize as starter cultures in these spontaneously fermented products. The availability of traditional fermented foods in populations with high prevalence of acute diarrhea necessitate further investigations as to the potential of these products being able to alleviate the problem of diarrhea.

Prebiotics

Associated with probiotics are prebiotics. Prebiotics are non-digestible food ingredients that have a beneficial effect on the host by selectively stimulating growth of health-promoting bacteria (Desai et al., 2004). They are assimilated by beneficial bacteria such as *Bifidobacteria* hence improving their growth activity leading to an enhanced intestinal balance. For a food to qualify as a prebiotic, it: has to be non-digestible by human enzymes; has to undergo selective fermentation by potentially beneficial bacteria in the colon; should cause an alteration in the composition of the colonic microbiota towards a healthier composition and all these changes should lead to a beneficial health to the host (Pariyaporn et al., 2003). Some researches with prebiotics have reported reduction in putative risk factors for colon cancer and control of serum triglycerides as well as cholesterol (Roberfroid, 2001).

Examples of prebiotic substrates incude inulin, lactulose, fructo-oligosaccharides, sugar alcohols such as lacto and xylitol (Salminen et al., 1998b). Most of these are obtained from natural sources or synthesized naturally from sucrose (Roberfroid, 1998). Inulin and raftilose are extracted from chicory roots or Jerusalem artichoke and have degrees of polymerisation (DP) ranging from 2 to 50. Nutraflora is a commercially marketed fructo-oligosaccharide (FOS) that consists of a glucose monomer linked at ά1,2 to two or more β-2,1-linked fructosyl units. FOS have been self affirmed by manufacturers as GRAS and are added to food products as food supplements (Desai et al., 2004). Synbiotics are a combination of probiotics and prebiotics that improve the survival of probiotic strains (Brink et al., 2002). Fyos (Nutricia) is an example of a fermented synbiotic product consisting of a probiotic culture *L. casei*, a prebiotic oligofructose and inulin (Soomro et al., 2002).

Ability of probiotic bacteria to ferment oligosaccharide is an important characteristic (Kaplan and Hutkins, 2000). This is because the availability of carbohydrates that escape metabolism and adsorption in the small intestine have a major influence on the microbiota that becomes established in the large intestine. These prebiotics could therefore be selective for probiotic microorganisms. A range of oligosaccharides have gained lots of attention as concerns being assessed for their prebiotic effects (Gibson, 1998; Roberfroid, 2001; Desai et al., 2004). Due to increased awareness of health benefits associated with probiotics, there is an increased trend in focus on the potential of fermented foods being modified and developed into probiotic foods that contain prebiotics (Helland et al., 2004; Grajek et al., 2005).

Exopolysaccharides

Lactic acid bacteria strains have been documented to produce expolysacharides (EPSs) that have received a lot of attention due to their contribution to improvement of texture and viscosity of fermented food products (Patricia et al., 2001; Savadogo et al., 2004). The lactic acid bacteria produce EPS probably as a protective function in their natural environment such as against desiccation, phagocytosis, phage attack, osmotic stress, antibiotics or toxic compounds (Patricia et al., 2001). The EPS may have a role in cell recognition, adhesion to surfaces and formation of biofilms that facilitate colonisation to various ecosystems (Hugenholtz, 2001). This is a beneficial attribute for probiotics in their endeavor to colonize the GIT.

Health benefits have been attributed to some exopolysaccharides. They have been reported to possess antitumor, anti-ulcer, immunomodulating and cholesterol lowering effects (De Vuyst and Degeest, 1999). Kazitawa and Ittoh (1992) reported an increase in B-cell dependent mitogenic activity induced by slime material products from *L. lactis subsp. cremoris*; water soluble EPS from kefir grains were shown to retard tumor. In our work (Kalui et al., 2009), we assessed production of EPS and amounts ranging from 298.53 to 431 mg/l were produced by *L. plantarum* and *L. rhamnosus* isolate strains from ikii, a spontaneously fermented maize porridge. Spontaneously fermented products therefore have potential and it is therefore necessary to assess them for production of EPS with an aim of tapping into the health benefits associated with EPS.

Safety

Safety is one of the recommended attributes in the FAO/WHO guidelines (2002) on evaluation for probitics. The gastrointestinal tract (GIT) is lined with an epithelium layer of cells and a mucoid lining. Haemolysis activity would therefore break down the epithelial layer while gelatinase activity would derange the mucoid lining. These would interfere with the normal functioning of these very important linings across which many physiological substances are exchanged and would cause pathways for infections. A deranged gut environment and mucosal lining is recognized as a source of allergic autoimmune diseases as well as acute chronic infections (Bengmark, 2003). It is therefore important that a probiotic strain should not have hemolytic and gelatinase activity. Isolates of *L. plantarum* and *L. rhamnosus* from *ikii* showed negative hemolysis and gelatinase activity (Kalui et al., 2009).The isolates assayed for gelatinase activity produced a mucoid substance around the colonies. This may be an indication that the strains are actually able to produce a mucoid substance that enhances the mucosal lining of the GIT hence adding on to the benefits of the mucoid layer of the GIT.

An important safety aspect in assaying for probiotic potential is antibiotic resistance. This is because antibiotic resistant genes, especially those encoded by plasmids could be transferred between microorganims. The potential strains need to be assayed for their antibiotic resistance to prevent the undesirable transfer of resistance to other endogenous bacteria. The risk of gene transfer depends on the nature of the genetic material (plasmids, transposons), the nature and concentrations of the donor and recipient strains and their interactions and the environmental conditions. The presence of an antibiotic may facilitate the growth of antibiotic resistant mutants (Marteau, 2001). Further safety assessments may include the determination of production of biogenic amines.

Current development in cereal based fermented products

Due to the invaluable aspects attributed to fermented foods, a lot of research is being carried out in respect to fermented food. The microbiota of many traditional African fermented maize products such as *mawe* (Hounhouigan et al., 1994), *kenkey* and *ogi* (Olasupo et al., 1997), *Bushera* (Muyanja et al., 2002), *koko* (Jakobsen and Lei, 2004) have been studied. The microbiota reported in these studies have been found to belong to the lactic acid bacteria group and mainly of the genera *Lactobacillus, Pediococcus, Leuconostoc and Enterococci*. Some studies have involved development of a probiotic fermented maize porridge (Helland et al., 2004).

Studies on lactic acid bacteria isolated from fermented foods in Africa have demonstrated an antimicrobial activity of these LABs towards pathogenic bacteria (Jacobsen and lei, 2004). The inhibition mechanism of Lactic acid bacteria is attributed to low pH as a result of presence of lactic acid (among other metabolite acid products) and antibiotic substances produced by these microorganisms (Mbugua and Njenga, 1991; Jacobsen and lei, 2004; Parvez et al., 2006; Vasiljevic and Shah, 2008; Reid, 2008; Kalui et al., 2009).

Lactic acid bacteria involved in fermentation of African foods have been documented as having the health benefit of shortening periods of diarrhea (Gaundalini et al., 2000; Rosenfeldt et al. 2002a, b; Jacobsen and lei, 2004). Diarrhea is a major cause of morbidity and mortality of millions of children in the developing world (Reid et al., 1999; Ribeiro, 2000). Foods that contain lactic acid bacteria and are able to shorten, alleviate or even prevent diarrhea need be developed in the developing world. Jacobsen and Lei (2004) investigated on one such Nigerian food, *koko* sour water (obtained from fermented millet) claimed to have anti-diarrhea attributes. In their study, microorganisms involved in production of the porridge showed antimicrobial activity against *Listeria innocua*, which is a pathogenic microorganism.

Lactic acid bacteria involved in production of African fermented foods are being investigated widely for their probiotic potential (Jacobsen and lei, 2004; Helland et al., 2004). African foods found to have potential probiotics shall have the health benefits associated with probiotics. These foods shall in addition be cheap, accessible and acceptable to the African indigenous people (Jacobsen and lei, 2004).

Conclusion

Lactic acid bacteria isolated from spontaneously fermented foods have shown combinations of potential probiotic attributes. These strains could be further assayed *in vitro* and *in vivo* to ascertain their probiotic effects as guided by the 2002 FAO/WHO guidelines for evaluation of probiotics. Such studies may involve but not be limited to: safety; evaluation of response to simulated stomach duodenum passage; susceptibility to antibiotics; hydrophobicity characteristics; adhesion to human cell linings; binding characteristics to human extra-cellular matrix; cholesterol reduction ability inclusive of other attributes claimed for probiotics.

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