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Probiotics and Traditional Fermented Foods: The Eternal Connection (Mini-Review)

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Abstract

Interactions of probiotic bacteria with the host and the impact of this interaction continue to fascinate microbial biotechnologists. Highlights of these influences of bacteria on human systems have been obtained from comparisons using germ free and conventional experimental animals. Where applied, outcomes and therapeutic uses are involved.There is a need to diversify the sources of isolation of probiotic bacteria to include fermented traditional foods of different societies. This approach invites more comprehensive research on different models to prove probiotics efficacy in a very well-defined patient groups pertaining to varied geographical locations, different ethnic communities and cultural values. Such invitation, no doubt, is motivated by the pressing need to develop and design alternatives to drugs causing bacterial resistance and risky side effects as well as complimentary treatments for other non-microbial diseases.

الملخص

إن تفاعل المحسنات الحيوية البكتيرية مع العائل وأثر ذلك كان وما زال يثير إعجاب علماء التقنيات الحيوية للأحياء الدقيقة وإن الآثار المميزة لهذه البكتيريا قد تم توثيقها من خلال دراسات مقارنة على حيوانات مخبرية. وأشارت نتائج هذه المقارنات لضرورة تنويع مصادر عزل هذه المحسنات الحيوية البكتيرية لتشمل أغذية مخمرة تقليدية لمجتمعات متباينة. ومن المؤمل أن تؤدي هذه المقاربة لدراسات شاملة على نماذج مختلفة لإثبات فعالية هذه المحسنات على مجموعات المرضى المحددة مكانياً وعرقياً ومع قيمها الحضارية. ولا شك بأن هذا التوجه سيتعزز بالحاجة الماسة لتطوير وتصميم بدائل لعقارات تؤدي لظهور المقاومة في البكتيريا إضافة للأعراض الجانبية كما يمكن أن تكون هذه المحسنات البكتيرية مكملة في معالجة أمراض غير بكتيرية.

Keywords: Probiotics, Traditional foods, Microbiota, Host-probiotic interactions.

1. Introduction

"Let food be thy medicine and medicine be thy food" as Hippocrates said, is the principle of today (Suvarna and Boby, 2005). Probiotics are one of the functional foods that link diet and health. Probiotics "For Life" are living, health-promoting microbial food ingredients that have a beneficial effect on humans (Chuayana *et al*., 2003).

The concept of probiotics have been first proposed by Nobel Prize winner Russian scientist Elie Metchnikoff, who suggested that the long life of Bulgarian peasants resulted from their utilization of fermented milk products (Mercenier *et al*., 2002; Chuayana *et al*., 2003; Tannock, 2003). Metchnikoff thought that when the fermented milk products were consumed, the fermenting *Lactobacillus* positively influenced the microflora of the gut, decreasing toxic microbial activities there (Mercenier *et al*., 2002; Chuayana *et al*., 2003).

As a result of more investigations in the probiotic field, its concept has been expanded to include bacteria from intestinal origin beside those bacteria isolated from fermented dairy products (Zeng *et al*., 2010). And we believe now that traditionally fermented foods are the untapped source for a wide variety of beneficial probiotic microorganisms.

Lactic acid bacteria (LAB) are the most common type of microorganisms used as probiotics. Strains of the genera *Lactobacillus*, *Bifidobacterium* (Yateem *et al*., 2008) and *Enterococcus* (Ljungh and Wadström, 2006) are the most widely used and commonly studied probiotic bacteria. The yeast *Saccharomyces boulardii* has also been studied as probiotics (Ljungh and Wadström, 2006).

All of these microorganisms have been considered as probiotics according to several criteria such as their total beneficial effect on the host, being non-pathogenic (Suvarna and Boby, 2005), and their ability to survive transit through the gastrointestinal (GI) tract (Saito, 2004; Crittenden *et al*., 2005; Liong and Shah, 2005).

Probiotic bacteria actions include: adherence and colonization of the host gut (Sanders, 2003), suppression of growth and invasion by pathogenic bacteria (Reid and

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Burton, 2002), production of antimicrobial substances such as bacteriocins (Nowroozi *et al*., 2004), improvement of intestinal barrier function, and stimulation of host immunity (Tannock, 2003).

Evidence from *in vitro* systems, animal models and humans suggests that the therapeutic use of probiotics has been considered very successful in the cases of lactose intolerance (Suvarna and Boby, 2005), inflammatory bowel disease (Daniel *et al*., 2006), colon cancer, *Helicobacter pylori* infection (Ouwehand *et al*., 2002), reduction of allergy, irritable bowel syndrome (Ljungh and Wadström, 2006), and certain diarrheal disease including antibiotic-associated diarrhea in adults, travelers' diarrhea, and diarrheal diseases in young children (Reyed, 2007). Additionally, probiotics may improve intestinal mobility and relieve constipation, especially in seniors (Ouwehand *et al*., 2002; Crittenden *et al*., 2005). They also reduce the cholesterol levels in serum (Liong and Shah, 2005) and no doubt many other benefits are there to be found by deligent research efforts.

Nowadays, probiotics are available in a variety of food products, dietary supplements (Parvez *et al*., 2006) and drugs (Sanders, 2003). In the United States, food products containing probiotics are almost exclusively dairy products - fluid milk and yoghurt- due to the historical association of lactic acid bacteria with fermented milk (Schillinger, 1999). The most frequently used bacteria in these products include *Lactobacillus* and *Bifidobacterium* (Sanders, 2003).

Probiotics are commonly not long-term colonizers of the GI tract, although they can adhere temporarily to the epithelium (Bezkorovainy, 2001). Instead, they may divide very slowly in the intestine, while remaining metabolically active (Marco *et al*., 2006). Therefore, daily consumption of these bacteria is probably the best way to maintain their effectiveness (Champs *et al*., 2003).

Isolation and characterization of novel strains of lactic acid bacteria from uninvestigated niche could have the dual advantage of revealing taxonomic characteristics and obtaining strains with interesting new functional traits that may be useful for probiotic application (Ortu *et al*., 2007). Among resources of probiotic bacteria are traditional fermented foods which vary according to societies and social habits of dieting (Reddy *et al*., 2007).

2. Present status of the art

Searching literature demonstrates the rapid growth of probiotics research with most published articles dates after 2003 (Reid *et al*., 2006). The type of research is being extended to include discovering events taking place at the interface between mucosal lining and microbiota, links between probiotics use and effects at distant body sites. Applications in the fields of allergy, inflammation, cardiovascular diseases and cancers are top priority targets hoping to benefit mankind, the emphasis being upon therapy rather than health improvements only. Recent research results were successful in using certain probiotic bacteria as a delivery system of endostatin for cancer gene therapy (Hu *et al*., 2009). The biological activity of some probiotics is attributed to fermentation end products including among others organic acids, CO_2, H_2O_2, diacetyl, and bacteriocins (Müller *et al*., 2009; Sanahan, 2010). In the majority of cases, this biological activity is a combination of all these factors (Todorov and Dicks, 2005). Probiotic induced-pathogen inhibitor may provide significant protection against pathogen spread in the gastrointestinal tract leading to enhanced human health (Collado *et al*., 2007a) through inhibition, displacement or competition with the pathogen (Gueimonde *et al*., 2006). Among the most studied bacteria as a probiotic included mainly isolates of the genera *Lactobacillus* and *Bifidobacterium*. Other bacteria of less potential as probiotics are isolates of the genus *Bacillus* such as *Bacillus subtilis*, *Bacillus clausii*, *Bacillus pumilus* and *Bacillus coagulans* (Hong *et al*., 2005; Patel *et al*., 2009). Reports claiming that probiotics use strengthened immune system, and helped in combating allergies, excessive alcohol intake and other diseases (Nichols, 2007; Sanders, 2003) encouraged researchers to search for novel probiotic bacteria in traditionally fermented foods which is usually linked to good health of people who consume such foods regularly (Salminen *et al*., 1998).

According to Reid, *et al.,* 2006, the understanding of probiotic microorganisms has expanded in the last 20 years after incepting the term in research circles. The term now implicates exact speciation of the microorganism, safe and effective use of probiotic formulations, exhibition of health and or therapeutic benefits on experimental models and probably volunteers and finally safe manufacture and retail. Unfortunately, these requirements are not fully observed worldwide, resulting in marketing too many of the so-called probiotics that do not meeting the proper criteria of a true probiotic. Table 1 presents few examples of bacterial probiotic strains which belong mainly to species of the genera *Lactobacillus* and *Bifidobacterium* with exceptions pertaining to *Escherichia coli* and the yeast *Saccharomyces cerevisiae*. Such information provide us with substantial evidence that in the last decade probiotics are beginning to be a convincing complimentary rout to traditional drug-based therapies. This trend, will with no doubt pave the way for more sophisticated approaches to develop genetically modified microorganisms which harbour new traits in constructed probiotics for special therapeutic purposes; such thoughts were entertained by Lartigue *et al*., (2007) in his article about genome transplantation in bacteria.

Table 1. Probiotic microorganisms exhibiting positive clinical responses.

Strain	Probiotic clinical response
Lactobacillus rhamnosus GG	Adhesion displacement of several pathogens (Collade *et al*., 2007b).
Bifidobacterium animalis BB12	Prevention and treatment of diarrhea (Weizman *et al*., 2005).
Bifidobacterium infantis 35624	Relieve abdominal pain (O'Mahony *et al*., 2005).
Lactobacillus paracasei LP-33	Relieve of allergic rhinitis (Wang *et al*., 2004a).
Lactobacillus acidophilus La5	*Helicobacter pylori* inhibition (Wang *et al*., 2004b).
Lactobacillus reuteri ATCC 55730	Production of CD4- positive T- lymphocytes (Valeur *et al*., 2004).
Bifidobacterium longum BL1	Serum cholesterol reduction (Xiao *et al*., 2003).
Saccharomyces cerevisiae Lyo	Reduced diarrhea (Kurugol and Koturoglu, 2005).
Escherichia coli Nissle 1917	Colitis treatment (Tromm *et al*., 2004)
Lactobacillus plantarum LP31	Inhibition of food pathogens (Müller *et al*., 2009).
Bifidobacterium longum	Antitumor effect (Hu *et al*., 2009).
Lactobacillus salivarius UCC118	Colon cancer reduction in mice (O'Mahony *et al*., 2001).
Lactobacillus buchneri P2	Serum cholesterol reduction (Zeng *et al*., 2010).
Lactobacillus casei	Reduction of bladder cancer (Ohashi *et al*., 2002).

3. Bioprospecting for probiotics in traditional fermented foods

The majority of infectious diseases caused by pathogenic bacteria and fungi represent a real challenge for current efforts to combat it through chemical and pharmacological research (Sanders *et al*., 2003). Rapid emergence of antibiotic resistance by many pathogens, along with increased toxicity of in field antibiotics prompts the continuous search and development of new anti-infective and antipathogenic materials (Sleator and Hill, 2006). Antipathogenic and probiotic bacteria would be the corner-stone in the search for new and effective alternative to traditional prophylactic means in a variety of clinical cases and settings (Sleator and Hill, 2008). In the last decade, it was observed that the selection of probiotics is based mainly on their ability to adhere and colonize the gastrointestinal epithelia and to compete with pathogens for binding sites and nutrients (Collado *et al*., 2007a; Ouwehand and Salminen, 2003). The functional activity of probiotics have been associated with managing the diarrheal and gastrointestinal infections by probiotics strains of different species of Lactics isolated mainly from fermented dairy products and related sources available in the western hemisphere (D'Souza *et al*., 2002). In addition, there is ample evidence suggesting the use of probiotics to treat and prevent urinary tract infection (Velraeds *et al*., 1998), rotavirus diarrhea (Szajewska *et al*., 2001), recurrence of certain cancers (Ohashi *et al*., 2002)), and reduction of allergies (Gill and Guarner, 2004)and recently, probiotic prophylaxis against asthma and eczema is being entertained (Sleator and Hill, 2008).

Most research articles pertaining to the aforementioned observations were dependent upon bacterial probiotics originating mainly from fermented dairy products and to less extent from human body indigenous microflora such as vaginal isolates or infants faeces (Reid, 2005). While the probiotic market is growing rapidly, it becomes eminent to diversify our sources in the search for new and novel probiotic microorganisms. This trend is encouraged by remarks stating that using different strains from different sources present interesting situations since differences between strains of the same species do exist (Reid, 2005; Weizman *et al*., 2005).

4. Oriental sources for probiotic strain selection

With the increased data about benefits of probiotics for human health and treatment and since most isolated and patented strains are of western origin, it is greatly inviting to try and isolate such probiotics from the untapped source examplified by a wide variety of different fermented foods of the orient (Table 2). For example, Keshik which is a Jordanian traditional fermented food made up of barboiled dried wheat and butter milk is of interest (Tamime and O'Connor, 1995). The product is similar to Tarhana (Turkish traditional fermented food) which proved to be a rich source of probiotic Lactic acid bacteria (Sengun, 2009). Among other potential foods also is the fermented eggplant (locally named Makdoos) made up of baby Aubergine stuffed with ground wall-nut, garlic, parsley and fermented in olive oil. Jameed which is solar-dried curd of sheep or goat naturally-fermented milk prepared and used traditionally by Jordanian Beduins (the old desert dwellers) is another unique source of probiotic bacteria. In a preliminary screen (un-published data) we were able to isolate a wide selection of lactic acid bacteria from these foods to be tested further for their suitability and unique probiotic properties. Recently, probiotic lactic acid bacteria have been isolated from unpasturized natural camel milk with superior probiotic characteristics (Yateem *et al*., 2008; Khedid *et al*., 2009). These are just few examples of traditional foods in Jordan as part of the Middle East and many other countries of the orient would probably be richer especially traditional fermented foods of South East Asia and India, which invites more research in this direction. The long shelf-life of such traditional foods could probably indicate the presence of

antimicrobial compounds among other characteristics exhibited by the indigenous bacteria which makes it a good probiotic candidate. Linking this to the increased consumer demand for natural and additive free foods would definitely maximize the interest in search for new sources of probiotic strains (Müller *et al*., 2009).

Table 2. Some oriental traditional foods of East Mediterranean countries including Jordan.

Traditional food	Ingredients/ natural fermentation
Aubergine Makdoos	Aubergine, wall-nuts, garlic, parsley, salt fermented in olive oil.
Green pepper Makdoos	Sweet green pepper, shredded tomato, parsley, salt garlic fermented in water or olive oil.
Pickled Green or Black Olives	Olives, salt, water fermented or pickled in olive oil.
Vine leaves	Vine leaves, pickled or packed to ferment naturally.
Keshik	Boiled wheat, thick curd of goat or sheep yoghurt or fermented milk, salt subjected to solar drying where slow fermentation takes place.
Jameed	Goat or sheep thick yoghurt curd, salt and then solar dried where slow fermentation improve the taste.
Tarhana	Dried mixture of cracked wheat yoghurt or fermented milk.
Turkish Tarhana	Cracked wheat or flour, yoghurt and vegetables fermented and then dried.
Cyprus Tarhana	Cracked wheat and fermented milk flavoured with bay leaf, thyme and fennel seed.
Shatta	Ground dried miniature chilies fermented in salt water and vegetable oil.

5. Probiotic delivery and persistence:

In order to secure full functionality of a probiotic, an efficient delivery system, in vivo survival and clear clinical efficacy must be met (Doleyres and Lacroix, 2005). In this context, it is implicated that increasing the stresses' tolerance level of the probiotic microorganism is of utmost importance (Sheehan *et al*., 2007). The novel thoughts of improving probiotic functions may be seen through modifying the strain tolerance to different stresses on the genetic level. This thought has been tested using some probiotic strains of *Bifidobacterium breve* and *Lactobacillus salivarius* (Sheehan *et al*., 2006), but further investigations are needed such as interference with the ability of a probiotic microorganism to enhance its compatible solutes productivity which is another approach of arming probiotics against different stresses (Termont *et al*., 2006), an area which also needs further exploration in the quest for the best probiotic formulations. The third prospective approach involves what is referred to as designer probiotics i.e. which are basically tailored-probiotics that would be very specific in targeting pathogens or intoxications where by doing this a pronounced prophylactic and therapeutic effects are realized (Laurel and Berger, 2005; Paton *et al*., 2005). Genetic engineering has been thought of as a useful approach for the design of probiotic bacteria that counteract the symptoms of genetic and age related diseases (Saier and Mansour, 2005). In combination therapy, components of pathogenic and nonpathogenic food-related bacteria are currently being evaluated as candidates for oral vaccines (Amdekar *et al*., 2009; Gill and Prasad, 2008; Collado *et al*., 2007b). It is believed now that as our knowledge of microbial variations is refined, tailoring of human associated microorganisms is rational and may lead to remodeling of their functions (Predis and Versalovic, 2009). Roos *et al*., (2010) reported improved humoral immune response against *E. coli* and bovine herpes vaccines in lambs through feeding with probiotic *B. cereus* and *Saccharomyces boulardii* strains.

6. Side effects and risks:

Some microorganisms have a long history of use as probiotics without causing illnesses in consumers. However, some probiotics safety has not been fully studied and understood scientifically. More information is especially needed on how safe they are for infants, children, old people and those with compromised immune systems.

Probiotics side effects, if they exist, would probably be mild digestive disturbances including gas and bloating. More serious contraindications have been observed in some incidences. Probiotics might in theory cause infections that need antibiotic treatments especially with people underlying health problems. They may also cause unhealthy metabolic activities i.e. too much stimulation of the immune system or even gene transfer among cells (Nerstedt, 2007).

Side effects and unsubstantiated claims are possible with prophylactic agents including probiotics. Probiotics are microorganisms, and we know the unpredictability of microorganisms in terms of how it will react to the existing or indigenous microbiota located in the target host. Gould and Shrot (2008) pointed out that particular illnesses such as post-antibiotic diarrheae are very responsive to probiotic treatment. However, they emphasize that many studies on humans have been disappointing although so

many of therapeutic potentials of probiotics still need great deal of substantiation before being recommended for medicinal applications. With these in mind, probiotics might be considered as one facet in a patient's health maintenance strategy. To conclude, we would say that since each human family and each individual in that family harbour a different combination of gut microflora, there may be some genetic propensities for and against certain probiotic strains. More investigations are needed to find out which group may benefit from which probiotic candidates and probably the source-effect of that probiotic microorganism. Further along the same concept, it is usually accepted to say that many studies of probiotics are carried on experimental animals rather than human subjects where lab results and animal studies cannot always be generalized to results in humans. In this context, it is observed that a single bacterial strain colonizing the gut of a gnotobiotic usually reaches a much higher counts than it does in a conventional animal, where the bacteria is faced with competition from others (Tannock, 2004). It is also noted that in the complex conventional ecosystem, the process of up and down regulation of host genes expression induced by different bacterial species vary tremendously with the presence of varying species and even strains (Hooper *et al*., 2001).

7. Conclusion

Probiotics are receiving great deal of attention in different fields of microbial biotechnology applications including health improvement and therapeutic purposes. This necessitates further diversification of sources of isolation to include traditional fermented foods of different cultures and geographical settings. Further works are needed to elucidate the functional characteristics of probiotic microorganisms. Designer probiotic for specific treatment is also an emerging field of research which would strengthen the drive of using probiotics for the treatment of different ailments and human health improvements.

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