ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

A Review On Probiotic Yeast Strains from Food and Environmental Origin

Nilanjana Das*.

School Bio Sciences and Technology (SBST), VIT University, Vellore: 632014, Tamil Nadu, India.

ABSTRACT

Probiotics are defined as pharmaceutical preparations containing living non–pathogenic microorganisms which upon ingestion in certain numbers exert health benefits including pathogen interference, colonization resistance, immunostimulation and immunomodulation, anticarcinogenic and antimutagenic activities, alleviation of symptoms of lactose tolerance, reduction in serum cholesterol, reduction in blood pressure, decrease of incidence of diarrhea, prevention of vaginitis and maintenance of mucosal integrity. Most of the studies on commercialized probiotics contain bacteria and very few of them present yeast in its composition. *Saccharomyces boulardii* is the only yeast commercialized as probiotic for human so far. However, few studies have suggested the use of other yeast species or genera based essentially on in vitro assays and very few clinical trials. The purpose of this review is to update the present status of probiotic yeast strains from food and environmental origin.

Keywords: Acid tolerance, bile tolerance, cholesterol removal, probiotics, yeast

Corresponding author

ISSN: 0975-8585

INTRODUCTION

During the last few years, a large number of food products have been launched in the food market attracting attention and interests among the consumers due to health benefits. Products defined as probiotics are now well described from a scientific point of view and are recommended for the enhancement of human health towards protection against several diseases or for the supply of essential metabolites with dietary and therapeutic characteristics [1]. In this context, the products containing probiotic microorganisms play an important role in the development of knowledge of the complex microflora.

The term 'Probiotic' is usually defined as live microorganisms that exhibit a beneficial effect on the host's health when they are ingested. In recent years, attention has been paid towards designing the functional foods which contain probiotic microorganisms responsible for health benefit in the host. Some important criteria are mainly used for selection of probiotic organisms which include their survival in the gastrointestinal environment and they must be able to present beneficial functions as colony resistance, immunomodulation or nutritional contribution of the normal gastrointestinal microbiota when ingested by human and animal hosts [2]. Most of the probiotics commercialised today have been selected from bacteria. Reports are less using yeast strains as probiotics.

There are many prerequisite conditions that must be met for a microorganism to be considered a true probiotic, the most important of which is the survival of the potentially beneficial microorganism in the human gut [3]. When probiotic microorganisms are absorbed with food into the body, they reach the gastrointestinal tract, which is a very low pH environment followed by exposure to bile acid as well as digestive enzymes from the pancreas. Therefore, tolerance against low pH and bile acid is very important criteria. Moreover, probiotic organism must be able to attach at enterocytes so as to proliferate in the intestines [4].

PROBIOTIC PROPERTIES

Acid and Bile tolerance

The major criterion for selecting a probiotic strain is the assessment of its resistance to low pH and bile exposition since the main biological barriers to be overcome after ingestion to reach its place of action are the acid in the stomach and bile salts in the intestine[5]. The first main barrier that the microorganisms meet after ingestion is gastric juice, in which the inhibitory effect is strictly related to the pH and to the concentration of hydrochloric acid [6]. The pH of the excreted HCl is 0.9, but the presence of food items raises the pH to 3.0 [7]. Moreover, the presence of food or other components buffer the probiotics ingested, conferring some protective effect on the microbial cells in the stomach [8] . Based on these facts, pH 2.5 has been considered as satisfactory value for the selection of acid resistant yeast strains.

To be considered as probiotics, microorganisms have to survive under the acidic conditions of the stomach and also have to survive intestinal secretion and the bile salts in the duodenum [7]. The bile salts released in to the upper small intestine have a detergent like function and play a role in specific [9] and non specific [10] defence mechanisms of the gut. Therefore, bile is the critical for the cell membranes of microorganisms that are composed of lipids and fatty acids [7] and the efficacy of its inhibitory effect is primarily determined by bile salt concentration[11] . Although the bile salt concentration of GI tract varies, the mean intestinal bile concentration used for screening of a resistant probiotic strain is 0.3 % [12].

Yeast isolates from infant feaces and Feta cheese was found to grow well at pH 3.0 and 5.0. The strains of *I.orientalis, C. parapsilosis and C. albicans* , two strains of *D. hansenii* , one strain of K. *marxianus* and one strain of *K. lactis* grew at pH 1.5 and 2.0 . The isolates from faeces were more resistant to low pH and bile than those from Feta cheese [6].

The potential of yeasts viz. *Candida humilis, Debaromyces occidentalis, Kluyveromyces lactis, Kluyveromyces loddrae, Kluyveromyces marxinus, Saccharomyces cerevisiae and Yarrowwia lipolytica* of dairy origin as probiotics was evaluated [13]. All species other than *K. marxinus* and *C. humilis* were resistant to acidic conditions. Irrespective of the presence of bile acid, growth was observed for all stains when incubation was carried out at 27° C. Limited growth was observed for *D. hansenii* and *K. lodderae*, irrespective of presence of bile acid.

Pennacchia et al. [1] isolated 20 acid and bile tolerant yeasts from foods. By using molecular methods, five strains were identified as *Saccharomyces cerevisiae*, three *Candida* spp., one as *Candida pararugosa* and one as *Pichia spp*. Almost all the tested yeast strains showed a survival percentage > 90 % when exposed to an acidic conditions at pH 2.5 for 2.5 h. In their study, human intestinal fluid was simulated for selecting resistant yeast strains by adding 0.3 % 0f bile salts and 0.1 % of pancreatin. Ten strains showed satisfying tolerance to bile salts and pancreatin exposure.

The yeast species viz. *Candida krusei, Kluveromyces marxianus, Candida tropicalis, Cadsida rugosa, Candida fabianii, Candida norvegensis* and *Trichosporon asahii* were identified during fermentation of fura , a West African spontaneously fermented cereal and characterized with probiotic potential [14]. All the yeasts isolates survived in pH 2.5 or in the presence of bile salts (0.3 % w/v) oxgall). Yeast strains viz. *S. Cerevisiae* AAV2, isolated from wine and *Issatchenkia orientalis* CL1132 isolated from fruit showed higher tolerance to pH and bile salts [15].

Heat stress tolerance

Temperature tolerance is one of the important characters of probiotic organisms [15]. The microorganisms used as probiotics must confront an adverse condition such as mild heat shock (mammal body temperature i.e. 37 °C). Psomas et al [6] reported that all the probiotic yeast isolates grew in broth at 25°, 37°, 42° C. Forty four out of 50 yeast isolates from Feta cheese grew at 25° C, 37° C and 43° C. Tolerance to elevated temperature is rarely found in yeast from non pathogenic sources. *Sachharomyces boulardii*, a non pathogenic yeast, was isolated from lychee fruit in Indochina and grew at the unusually high temperature of 37 °C [16] and this is the only yeast commercialised as probiotic for human so far. Pederson et al [14] reported the growth of probiotic yeasts viz. *Candida krusei, Kluveromyces marxianus, Candida tropicalis, Cadsida rugosa, Candida fabianii, Candida norvegensis* and *Trichosporon asahii* at 37° C independently which were determined as indicators of the survival potential of the isolates during passage through the human gastro intestinal tract. Six isolates of *Saccharomyces* yeasts along with reference strain *S. Boulardii* NCDC363 were subjected to heat tolerance studies [15]. All the strains exhibited increased growth at a temperature of 37° C whereas three strains grew at 42° C.

Simulated gastric and intestinal tolerance

Pepsin resistance of microbes is considered as one of the most prerequisite properties of probiotics [17]. Pepsin and pancreatic enzymes are the major digestive enzymes, which are active at highly acidic and basic pH respectively. Lohith and Appaiah [15] reported that *Pichia kudriavzevii* P2 grew well in simulated gastric and pancreatic juice compared to the reference strain *Saccharomyces. boulardii* NCDC363 . Sourabh et al [18] reported a decrease in viability of *S. cerevisiae* in simulated gastric juice and small intestinal juices after 4 hours exposure. *S. boulardii* maintained 75 % of viability in simulated gastric environment [19] . Exposure of non-Saccharomyces yeast *Pichia kudriavzevii* exhibited high resistance in gastric juice [20] .

Cholesterol removal

All probiotic bacterial strains possess varying degree of cholesterol removal capacities from the media through several mechanisms including cholesterol assimilation, incorporation of cholesterol into cellular membrane, binding of cholesterol to cells and bile salt deconjugation [21]. Eight yeast strains isolated from infant feces and the traditional Greek Feta cheese, selected for their probiotic properties, were tested along with a commercially available strain of *Saccharomyces boulardii* for their ability to remove cholesterol from a growth medium (yeast extract glucose peptone broth) supplemented with 0.3% Oxgall. The amount of cholesterol removed during 72 h of growth at 37°C revealed significant variations among the yeast strains examined [22].

Adhesion, auto aggregation and surface hydrophobicity

Adhesion to intestinal epithelial cell is another important parameter which helps in colonization and proliferation of cells. Auto- aggregation and cell surface hydrophobicity is an in vitro method to test the adhesive capacity of probiotics [15]. Sourabh et al [18] reported auto-aggregation ranged from 18.7 % to 67.65

% for different *S. Cerevisiae* strains. Lohith and Appaiah [15] reported that all the *S. Cerevisiae* strains showed good auto- aggregation capacities ranging from 83.8 % to 86.03 % at 24 hours. The yeast cells exposed to chloroform showed higher percentage of microbial adhesion. To hydrocarbon, strains *S. Cerevisiae* AAV2 and *Issatchenkia orientalis* CL1132 showed around 93 % of adhesion.

Antagonistic properties

An important property of probiotic organisms is their antagonistic activity against pathogegenic bacteria either by competitive exclusion, decrease of redox potential, interbacterial aggregation, or production of antimicrobial substances including organic acids, other primary metabolites such as hydrogen peroxide, and special compounds like bacteriocins and antibiotics.

There is report on *Sacharomyces boulardii* which inhibited the growth of *Staphylococcus. aureus* and also decreased the number of bacterial counts of *Pseudomonas auruginosa* [23-24] . However, Martin et al [25] did not notice any antagonism capacity of *S. boulardii* against pathogenic *E. Coli* and *Bacillus cereus*. Lohith and Appaiah [15] showed inhibition zone against *E. coli, L. monocytogenes* and *S. aureus* due to secretion of organic acids such as caproic acid, caprylic acid and capric acid affecting the growth of pathogenic bacteria [26].

Antagonism assays were performed using 25 yeast specimens [27]. Among 14 indicator strains (13 bacteria and one yeast) used in antagonistic assays, *P.aeruginosa*, *E faecalis* and *C. perfringens* were the most sensitive. Intermediate sensitivity was observed for *C. difficile* and *B. cereus* and low sensitivity for *S.typhi*, *S. typhimurium*, *C. albicans* and *S. sonnei* was noted.

CONCLUSION

The present review report indicates that yeast strains also have noteworthy probiotic properties such as acid and bile tolerance, heat stress tolerance, ability of cholesterol removal, proficient adhesion properties and antagonistic properties like bacteria. In view of these positive properties, yeast strains can function as a probiotic very efficiently. However, *in vitro* studies do not allow direct extrapolation on the behaviour of probiotics. So, further in vivo validations are needed to confirm the potential of the selected probiont.

ACKNOWLEDGEMENT

The author gratefully acknowledges VIT University for providing facility to access Scopus indexed journals for preparation of the manuscript.

REFERENCES

[1] Pennacchia C, Blaiotta G, Pepe O, F. Villani F. J. Appl. Microbiol. 2008 ; 105:1919 -1928.
[2] Martins FS, Nardi RMD, Arantes RME, Rosa CA, Neves MJ, Vicoli JR. J Gen Appl Microbiol. 2005 ; 51 : 83–92.
[3] Fuller R. J. Appl. Bacteriol. 1989 ; 66 : 365-378.
[4] Ouwenhand AC, Salminen S, Isolauri E. Antonie Van Leeuwenhoek., 2002; 82; 279–289.
[5] Gueimonde M, Salminen S. *Digest Liver Di* , 2006; 38 : S242–S247.
[6] Psomas E, Andrighetto C, Litopoulou-Tzanetaki E, Lombardi A, Tzaneta N. Int.J. Food. Microbiol. 2001; 69 : 125-133.
[7] Erkkila S, Petaja E. Meat Sci., 2000 ; 55 : 297–300.
[8] Prasad J, Gill H, Smart J, Gopal PK. Int Dairy J., 1998 ; 8 : 993–1002.
[9] Marteau P, Minekus M, Havenaar R, Huis In't Veld JHJ. J Dairy Sci. 1997; 80 : 1031–1037.
[10] Kalambaheti T , Cooper GN, Jackson GDF. Gut., 1994; 35 : 1047–1052.
[11] Charteris WP , Kelly PM, Morelli L, Collins JK. J Food Prot , 2000; 63 : 1369–1376.
[12] Gilliland SE, Stanley TE, Bush LJ, J Dairy Sci. 1984, 67 : 3045–3051.
[13] Kumara H, Tanoue Y, Tsukahara M ,Tanaka T, Shimazaki K. J Dairy Sci., 2004; 87: 4050-4056.
[14] Pedersen LL, Owusu-Kwarteng J, Thorsen L, Jespersen L, Int J Food Microbiol. 2012 ; 159 : 144–151.
[15] Lohith K, Anu Appaiah KA.. Int. J. Probiotics and Prebiotics.2014; 9 : 58-63.
[16] McFarland,LV, Bernasconi P. Microb. Ecol. Health Dis. 1993; 6 : 157–171.

[17] Latha S, Vinothini G, Devadasan JDC, Dhasekaran D.J. Biosci Bioeng. 2016 : 121 : 124-131.
[18] Sourabh A, Kanwar SS, Sharma OP. J. Yeasts Fungal Res. 2011; 2 : 117-126.
[19] Fietto JLR , Araujo RS, Valadao FN, Fietto LG, Brandao RL, Neves MJ, Gomes CO, , Nicoli JR, Castro IM, Can J Microbial. 2004; 50 : 615 -621.
[20] Chen L, Ma Y, Maubois J , He S, Li L,Chen. H., Dairy Sci. Techn. 2010; 90 :537-548 .
[21] Miremadi F, Ayyash M, Sherkat F, Stojanovska L, J Functional Foods. 2014; 9 : 295-305.
[22] Psomas EI, Fletouris DJ, Litopoulou – Tzanetaki E, Lombardi A, Tzanetakis N, J Dairy Sci. 2003 ; 86 : 3416 -3422.
[23] Bornet, E. Bergogne-Berezin M. Science Aliments, 1996; 6 : 63-73.
[24] Rajkowska K, Kunicka-StyczyBska A, Rygal´a. A., Food Technol. Biotechnol., 2012; 50 : 230–236.
[25] Martins FS, Silva AA, Vieira AT, Barbosa FH, Arantes RM, Teixeira MM, Nicoli JR, Arch Microbiol. 2009; 191: 623-630.
[26] Murzyn A, Krasowska A , Stefanowicz P , Dziadkowiec D, Lukaszewicz M, Plos One. 2010; 5: 1-7.
[27] Tiago FCP,Martins FS,Rosa CA, Nardi RMD, Nicoli JR. World J Microbiol. Biotechnol. 2009 ; 25: 657 – 666.