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Technological challenges for future probiotic foods

T. Mattila-Sandholm[a,*], P. Myllärinen[a], R. Crittenden[a], G. Mogensen[b], R. Fondén[c], M. Saarela[a]

[a] *VTT Biotechnology, P.O. Box 1500, FIN-02044 VTT, Finland*
[b] *GM-Consult, Thorsmosevej 16, 3200 Helsinge, Denmark*
[c] *Arla Foods, 10546 Stockholm, Sweden*

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Abstract

Modern consumers are increasingly interested in their personal health, and expect the food that they eat to be healthy or even capable of preventing illness. Gut health in general has shown to be the key sector for functional foods in Europe. The probiotic yoghurt market is well established but the key growth sector recently has been the probiotic drinks. The popularity of dose-delivery systems for probiotic drinks has also resulted in research efforts targeted to developing probiotic foods outside the dairy sector. New product categories, and thus novel and more difficult raw materials with regard to technology of probiotics, will certainly be the key research and development area for future functional food markets.

The viability and stability of probiotics has been both a marketing and technological challenge for industrial producers. Probiotic foods should contain specific probiotic strains and maintain a suitable level of viable cells during the product's shelf life. Unless strict demands are set on probiotic product definition and labelling their regulatory definition will remain obscure. The technological demands placed on probiotic strains are great and new manufacturing process and formulation technologies may often be required for bacteria primarily selected for their functional health properties. Before probiotic strains can be delivered to consumers, they must first be able to be manufactured under industrial conditions, and then survive and retain their functionality during storage as frozen or freeze-dried cultures, and also in the food products into which they are finally formulated. The probiotic strains should also survive the gastrointestinal stress factors and maintain their functionality within the host. Additionally, they must be able to be incorporated into foods without producing off-flavours or textures—they should be viable but not growing. The packaging materials used and the conditions under which the products are stored are also important for the quality of products.

Future technological prospects exist in innovations finding solutions for the stability and viability problems of probiotics in new food environments. Current research on novel probiotic formulations and microencapsulation technologies exploiting biological carrier and barrier materials and systems for enteric release provides promising results. Maintenance of low production costs will remain the challenge for future probiotic process and formulation technologies. Exploitation of food-grade raw materials such as native, and physically or enzymatically treated starches, is one example of future technology that has the potential to meet the challenge of broadening the range of food types into which probiotic ingredients can be successfully incorporated. Novel developments for control release systems in foods and pharmaceuticals will also provide new possibilities.

Keywords: Probiotics; Viability; Stability; Prebiotics; Starters; Lactic acid bacteria; Microencapsulation

1. Introduction

Probiotics are live microbial food supplements, which benefit the health of consumers by maintaining, or improving their intestinal microbial balance (Fuller, 1989). Due to their perceived health benefits probiotic bacteria have been increasingly included in yoghurts and fermented milks during the past two decades. Most commonly they have been lactobacilli such as *Lactobacillus acidophilus*, and bifidobacteria (Daly & Davis, 1998). A major development in functional foods pertains to foods containing probiotics and prebiotics, which enhance health promoting microbial flora in the intestine. There is growing scientific evidence to support the concept that the maintenance of healthy gut

*Corresponding author. Tel.: +358-9-4565200; fax: +358-8-4552103.

E-mail address: tiina.mattila-sandholm@vtt.fi (T. Mattila-Sandholm).

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microflora may provide protection against gastrointestinal disorders including gastrointestinal infections and inflammatory bowel diseases (Haenel & Bendig, 1975; Mitsuoka, 1982; Salminen, Ouwehand, & Isolauri, 1998). The use of probiotic bacterial cultures stimulates the growth of preferred microorganisms, crowds out potentially harmful bacteria, and reinforces the body's natural defence mechanisms (Salminen et al., 1998). Today, plenty of evidence exists on the positive effects of probiotics on human health. However, this has usually been demonstrated in diseased human populations only (Salminen et al., 1998). Thus there is an urgent need for evidence for probiotic health benefits in average (generally healthy) populations.

Before a probiotic can benefit human health it must fulfil several criteria. It must have good technological properties so that it can be manufactured and incorporated into food products without loosing viability and functionality or creating unpleasant flavours or textures. It must survive passage through the upper gastrointestinal (GI) tract and arrive alive at its site of action, and it must be able to function in the gut environment. To study the probiotic strain in the GI-tract, molecular techniques must be established for distinguishing the ingested probiotic strain from the potentially thousands of other bacterial strains that make up the GI ecosystem. Additionally, techniques are required to establish the effect of the probiotic strain on other members of the intestinal microbiota and importantly on the host. This includes not only positive health benefits, but also demonstration that probiotic strains do not have any deleterious effects. When this knowledge is available, the probiotics can enter human pilot studies that attempt to assess their health benefits to consumers (Mattila-Sandholm & Salminen, 1998; Mattila-Sandholm, Mättö, & Saarela, 1999).

Several aspects, including safety, functional and technological characteristics, have to be taken into consideration in the selection process of probiotic microorganisms. Safety aspects include specifications such as origin (healthy human GI-tract), non-pathogenicity and antibiotic resistance characteristics. Functional aspects include viability and persistence in the GI-tract, immunomodulation, antagonistic and antimutagenic properties (Mattila-Sandholm et al., 1999; Saarela, Mogensen, Fondén, Mättö, & Mattila-Sandholm, 2000). Careful screening of probiotic strains for their technological suitability can also allow selection of strains with the best manufacturing and food technology characteristics. However, even the most robust probiotic bacteria are currently limited in the range of food applications to which they can be applied. Additionally, bacteria with exceptional functional health properties are often ruled out due to technological limitations. New process and formulation technologies will enable both expansion of the range of products into which probiotics can be applied and the use of efficacious strains that currently cannot by manufactured or stored with existing technologies.

The viability of probiotics has been both a marketing and technological concern for many industrial producers. The definition of live probiotic bacteria can and will stay obscure unless strict demands are set on product definitions. Probiotic foods should include specific probiotic strains at a suitable level throughout their shelf life. Factors related to the technological and sensory aspects of probiotic food production are of utmost importance since only by satisfying the demands of consumers can the food industry succeed in promoting the consumption of functional probiotic products in the future. The packaging materials used and the conditions under which the products are stored, are important for the quality of products containing probiotic bacteria.

2. Selection and production of probiotics

Good viability and activity of probiotics are considered prerequisites for optimal functionality. However, several studies have shown that non-viable probiotics can have beneficial effects such as immune modulation and carcinogen binding in the host (for a review see Ouwehand & Salminen, 1998; Salminen, Ouwehand, Benno, & Lee, 1999). Thus, for certain probiotic strains it might be sufficient that they grow well during initial production steps (to obtain high enough cell numbers in the product) but they do not necessarily need to retain good viability during storage. Factors influencing probiotic functionality are described in Fig. 1. Beneficial clinical effects of probiotics are listed in Table 1.

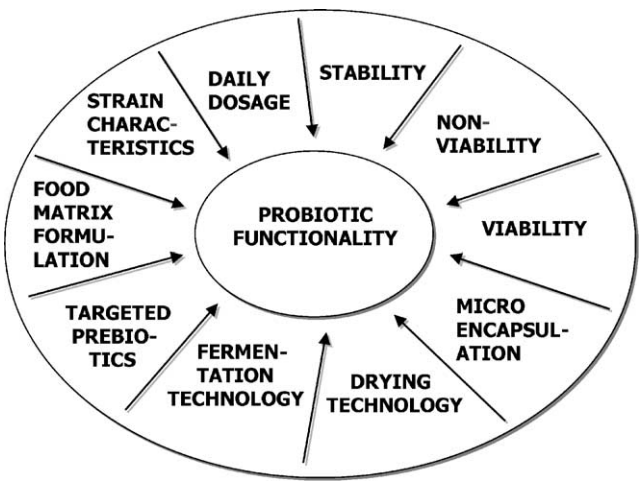

Fig. 1. Technological factors influencing the functionality of probiotics.

Table 1
Beneficial effects of probiotics detected in various clinical trials[a]

Modulation of intestinal flora
Modulation of immune response
Lowering faecal enzyme activities
Increase in faecal fatty acid content
Improvement of constipation
Alleviation of atopic dermatitis symptoms in children
Reduction, prevention or treatment of various diarrhoeal diseases (antibiotic associated, viral, *Clostridium difficile* associated, traveller's diarrhoea)
Positive effects on superficial bladder cancer and cervical cancer

[a] Note that not all beneficial effects can be attributed to all probiotic strains, but instead beneficial effects are strain-specific (Saarela et al., 2000).

The functional requirements of probiotics should be established by using in vitro methods and the results of these studies should be reflected in controlled human studies. While selecting a preferable probiotic strain several aspects have to be considered:

(a) Acid tolerance and tolerance to human gastric juice.
(b) Bile tolerance (an important property for survival in the small bowel).
(c) Adherence to epithelial surfaces and persistence in the human GI-tract.
(d) Immunostimulation, but no proinflammatory effect.
(e) Antagonistic activity against pathogens such as *Helicobacter pylori*, *Salmonella* sp., *Listeria monocytogenes* and *Clostridium difficile*.
(f) Antimutagenic and anticarcinogenic properties.

Feeding trials with different probiotic strains have shown that the probiotic strain usually disappears from the GI-tract within a couple of weeks after the ingestion is discontinued (Fukushima, Kawata, Hara, Terada, & Mitsuoka, 1998; Johansson et al., 1998; Alander et al., 1999, Donnet-Hughes, Rochat, Serrant, Aeschlimann, & Schiffrin, 1999). The role of the probiotic persistence in the human GI-tract has therefore been questioned. However, even temporary persistence, which has been noted for several ingested probiotic strains, may enhance their chances for beneficial functions in the GI-tract, and is therefore considered a desirable trait.

In selecting starter microorganisms reliable acid-forming ability is one of the most important characteristics. However, when selecting probiotics the criteria should be connected to the impact on human health and well being. As the environment within the GI-tract and the food might be quite different the probiotic is often not suitable as a starter organism (Oberman & Libudzisz, 1998; German et al., 1999). The growth rate might be too slow and they might give off-flavours (Svensson, 1999). To improve the suitability of the food as a substrate for the probiotic, energy sources (e.g. glucose), growth factors (e.g. yeast extract and protein hydrolysates) or suitable antioxidants, minerals or vitamins can be added into it (Kurmann, 1988; Ishibashi & Shimamura, 1993; Dave & Shah, 1998; Gomes, Malcata, & Klaver, 1998). However, even if such an adjustment may improve the performance of the probiotic as a starter, it is often not enough. By use of a starter in addition to a probiotic preparation this problem can usually be solved (Fondén, Grenov, Reniero, Saxelin, & Birkeland, 2000).

Several technological aspects have to be considered in probiotic selection. These include the following:

(a) Good sensory properties.
(b) Phage resistance.
(c) Viability during processing.
(d) Stability in the product and during storage.

Most leading starter culture manufacturers today produce lactic acid bacteria and bifidobacteria commercially (Mogensen & Friis, 1997). Commercially available probiotic cultures may consist of a single strain or a mixture of several strains. In most cases the probiotic properties are affected by the way in which the strain or culture has been produced (for a review see German et al., 1999). Therefore, specific information on strain-specific properties should be available for the process optimisation. Probiotic cultures may be incorporated in special formulations like capsules or tablets, or they may be used in the production of a large variety of fermented food products. In some cases the cultures may be added to a food to contribute specific probiotic or functional properties.

Given the many uses of lactic acid bacterial (LAB) cultures, there is considerable commercial interest in the production of stable starter and probiotic LAB that contain a large number of uninjured, viable cells. The use of liquid and frozen concentrates have been used extensively in the past, but large savings in the costs of transport and storage, and improvements in culture stability, can be made using freeze-dried and spray-dried preparations (Champagne, Gardner, Brochu, & Beaulieu, 1991; Gölker, 1993). Despite the fact that spray-drying is more economical than freeze-drying, especially on a large scale (Gölker, 1993; Johnson & Etzel, 1993), many LAB cannot tolerate the relatively high temperatures that are used during spray-drying (Porubcan & Sellars, 1979). As a consequence, freeze-drying is the most popular method for the production of dried LAB preparations. Although freeze-drying is less destructive to microorganisms than spray-drying (Porubcan & Sellars, 1979), protectants are usually added to the cultures to be dried in order to prevent, or at least mitigate, cell injury during drying and subsequent

storage (Champagne et al., 1991; Souzu, 1992). The most common protectants used at industrial scale are lactose or sucrose, monosodium glutamate (MSG), and ascorbate in milk or in water base (Mäyrä-Mäkinen & Bigret, 1998).

Most commercial probiotic culture preparations are supplied in highly concentrated form, and most of them are constructed for direct vat set (DVS) applications (Honer, 1995). Use of these highly concentrated DVS cultures is common due to the difficulties involved in propagating probiotic microorganisms at the production site. The DVS cultures are supplied either as highly concentrated frozen cultures or as freeze-dried cultures. Usually deep-frozen cultures contain more than 10^{10} cfu/g, whereas freeze-dried cultures typically contain more than 10^{11} cfu/g (Oberman & Libudzisz, 1998). The cell concentration per gram of product varies with the culture and the type of organisms used.

3. Probiotic interaction with starter bacteria

In fermented probiotic products it is important that the probiotic culture used contributes to good sensory properties. Therefore, it is quite common to use probiotic bacteria together with other types of bacteria (starters) suited for the fermentation of the specific product. The interactions between probiotic and starter might have an impact on the product quality. It has been shown that it is possible to produce fermented dairy products with excellent sensory properties and good survival of the bacteria by using starter and probiotic organisms together (Fondén et al., 2000). Suitable starters might be *Streptococcus thermophilus*, yoghurt cultures and mesophilic starters with different combinations of *Lactococcus* strains. The most suitable combination of starter and a specific probiotic bacteria has to be determined using a screening process evaluating the impact of different starters on the sensory properties and on the survival of the probiotic strain (Ishibashi & Shimamura, 1993; Samona, Robinson, & Marakis, 1996). In selecting a suitable starter the negative impact on probiotic survival in vitro and in vivo should also be taken into consideration. The survival of the probiotic bacteria might be influenced by the metabolites formed by the starter such as lactic acid, hydrogen peroxide and bacteriocins.

4. Demonstration on technological criteria for probiotics

The EU FAIR project, "Demonstration of the Nutritional Functionality of Probiotic Foods", prepared statements on the technological properties of probiotic foods with new and existing strains, which fulfil the demands of the consumer. By employing standard methods suitable culture concentrates could be produced with high cell concentrations and good survival during storage at low temperatures. It was demonstrated that for some strains and manufacturing conditions it is possible to use only the probiotic strain as the acid producing strain. However, in many cases the use of a supporter starter is preferable. Products produced by both methods were shown to have good organoleptic properties and the survival of the probiotic organisms was excellent. Some of the strains were also used in products presently on the market. By comparison it was shown that the commercial products have even better technological properties as a result of industrial optimisation. It was concluded that it is possible to produce probiotic foods by different processes containing high levels of specific probiotics throughout the storage time in combination with acceptable organoleptic properties (Fondén et al., 2000). In the production of probiotic dairy products, selected probiotic cultures should be used to (Fondén et al., 2000):

1. Produce concentrated cultures of each specific strain in levels above 10^{10} with good storage properties at low temperature.
2. Produce probiotic foods with the help of a supporter culture such as a yoghurt culture or a pure *Streptococcus* strain.
3. Ferment milk together with at least some supporter cultures without inhibition of the growth of any of the added strains.
4. Produce probiotic foods with levels of the specified probiotic strain up to 10^8 cells/g product.
5. Produce probiotic foods with high and constant levels of the probiotic strain when stored at low temperature for three weeks.
6. Produce probiotic foods with an acceptable taste and flavour throughout the storage time.
7. Produce probiotic foods with an acceptable stability and viscosity in many cases even improved in comparison to just using the supporter culture.

5. Manufacturing of non-dairy probiotic foods

Application of probiotic cultures in non-dairy products and environments represents a great challenge. Probiotic viability in the food matrix depends on factors such as pH, storage temperature, oxygen levels, and presence of competing microorganisms and inhibitors. In products like probiotic-containing baby foods or confectionery, it is important that the formulation maintains the activity and viability of the probiotic for extended periods of time. Since the probiotic cultures are included as ingredients to these kinds of products, they do not usually multiply, which sets great demands

for the probiotic stability. Factors like water activity, oxygen tension and temperature becomes increasingly important when dealing with these kinds of products. Storage at room temperature, which is common for many types of non-dairy products such as cereal products, drinks, confectionary etc., can create an overwhelming challenge for probiotic stability. Using probiotic encapsulation technology to ensure the probiotic viability can sometimes solve this problem (Myllärinen et al., 2000). Stable probiotic-containing baby food formulations and confectioneries have been developed and are currently on the market (Langhendries et al., 1995; Fukushima, Hara, Terada, & Mitsuoka, 1997).

6. Formulation of probiotics—good results in starch encapsulation of lactic acid bacteria

Starch is a dietary component having an important role in colonic physiology and functions and a potential protective role against colorectal cancer (Cassidy, Bingham, & Gummings, 1994; Silvi, Rumney, Cresci, & Rowland, 1999). Resistant starch is starch that is not digested by pancreatic amylase in the small intestine and reaches the colon. Starch has been classified into three types by Englyst, Kingman, and Gummings (1992): RS1, starch entrapped within food matrix, RS2, granular starch structure and RS3, retrograded starch formed by food processing. Resistant starch can be fermented by human and animal gut microflora. In a study where rats were fed with native potato starch (RS2) an increase in the intestinal population of bifidobacteria, lactobacilli, streptococci and enterobacteria was demonstrated. The fermentation of carbohydrates by anaerobic bacteria produced short-chain fatty acids and lowered the pH in the lumen (Macfarlane & Gummings, 1991; Kleessen et al., 1997; Le Blay, Michel, Blottière, & Cherbut, 1999).

VTT Biotechnology initiated studies on starch-encapsulation of probiotic bacteria in the four-year Functional Food Research Programme in 1996. The aim of the research has been to stabilise LAB and formulate new types of foods fortified with encapsulated health-promoting bacteria that are only released upon reaching the human gut. Currently VTT is developing a carbohydrate based stabilisation/capsulation method for probiotics in order to enhance their viability in products and in the human intestine. The main focus is on developing a feasible starch based technology for probiotic microencapsulation by performing both the bacterial production and their capsulation in one batch process. In this technology, large potato starch granules (50–100 μm), which are enzymatically treated to obtain a porous structure, are used as a carrier. Subsequently, amylose, the linear polymer of starch, is solubilised, cooled and precipitated over the bacteria-filled starch granules. Finally the whole product, together with the growth media, is freeze-dried to a powder form (Myllärinen et al., 2000) (Fig. 2).

Different amylases have been tested using a range of conditions to establish the optimal method for hollowing starch granules to produce an internal space for the encapsulated bacteria. It was found that freeze-dried starch granules were up to 70% hydrolysed and native granules only 25% hydrolysed by Megazyme bacterial α-amylase. Malt amylase provided similar results using the same conditions. Fungal and pancreatic amylases hydrolysed much less potato starch than bacterial or malt based amylases. The size of the starch granules was over 50 μm before hydrolysis and did not change even after 70% of the amylose had been hydrolysed. The fact that the enzyme attacked the inside of the freeze-dried granules, making them porous, was evident when the hydrolysed granules were examined in microscopy cross sections. The use of a simple precipitation process to produce a stable and resistant coating for the capsules is at the moment the most convenient coating technique (Myllärinen et al., 2000).

Several probiotic strains, including *Lactobacillus rhamnosus* VTT E-97800, have been used in starch encapsulation studies. The viability of encapsulated *L. rhamnosus* E-97800 stored at room temperature under normal atmospheric humidity has been at least six months, and when frozen, at least 18 months. Use of an in vitro GI transit model showed that the capsule material is resistant in the upper intestine (only 10% degradation), and in an in vivo test it was seen that encapsulated *L. rhamnosus* VTT E-97800 cells survived passage through the human GI-tract (unpublished results). Recently, investigation into the adhesion of bifidobacteria to the surface of different starch granules has commenced. Some bifidobacterial strains demonstrated very good adhesion to the surface of potato starch granules (Fig. 3). A strain with strong adhesion was also found to be able to hydrolyse the raw starch (Crittenden et al., 2001).

Since the project aims were to develop a method to economically produce food-grade probiotic capsules, a strong emphasis was placed on the development of inexpensive, edible growth media for probiotic bacteria. During the project, a new edible, rye-based growth medium was developed for lactic acid bacteria. In the future, research will be focused on the scale-up and optimisation of the process, and on new technological innovations. Novel polysaccharides capable of enhancing the bacterial adhesion will be tested. An interesting approach will be the use of starch granules that naturally form aggregates, such as small barley starch granules. Furthermore, new hydrophobic materials will be used to promote resistance of the capsules to high humidity. Formulation of capsulated

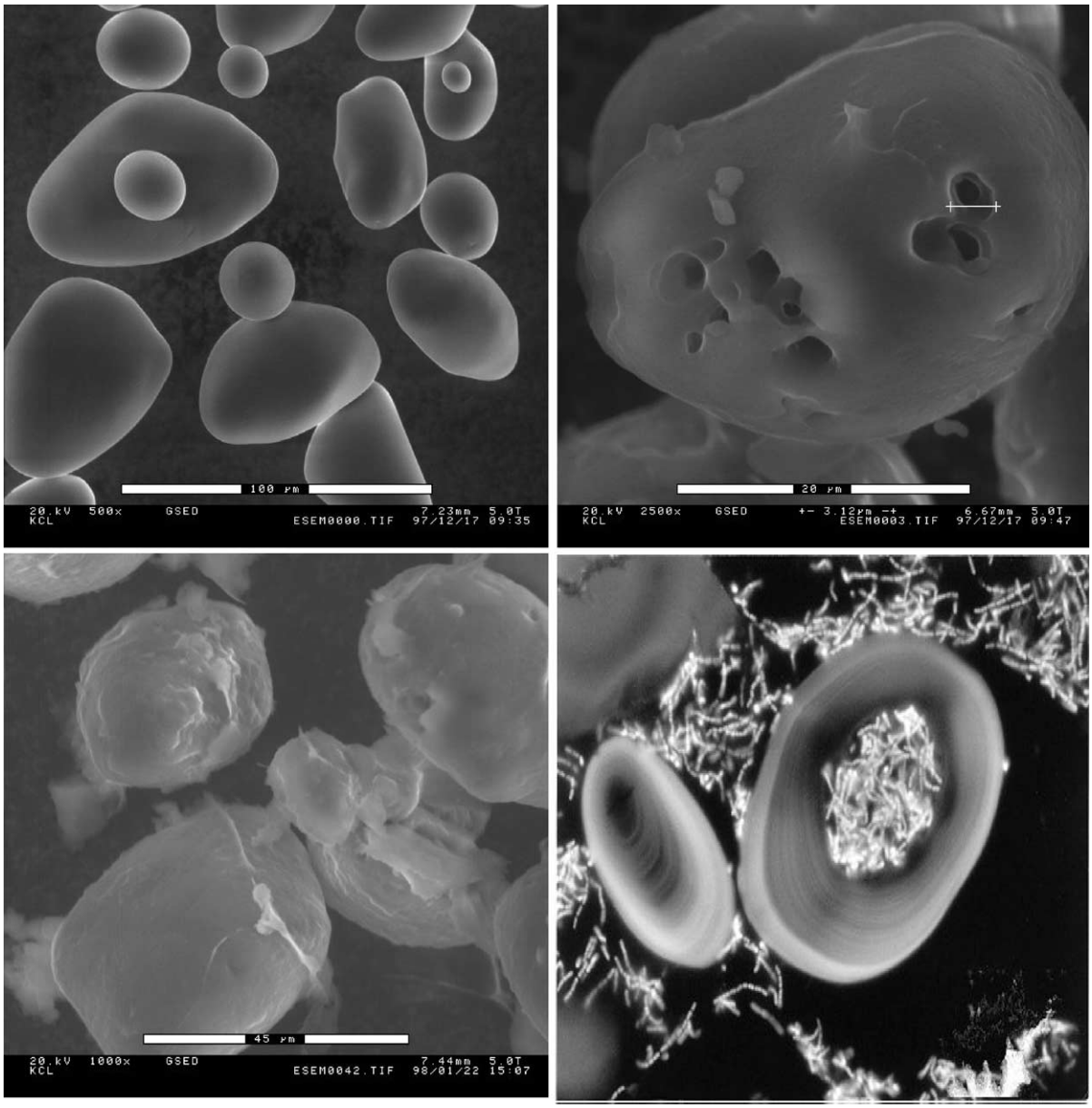

Fig. 2. Potato starch granules (up left), hydrolysed granules with pores on the surface (up right), amylose coated potato starch granules (bottom left), and microscopy cross section of a bacteria-filled potato starch granule (bottom right, 1450 ×).

probiotics into different food matrices is currently in progress.

7. Functional prebiotic ingredients—promoters for probiotics

In addition to the probiotic approach of directly introducing live bacteria to the colon through dietary supplementation, another approach to increase the number of beneficial bacteria such as bifidobacteria in the intestinal microbiota is through the use of prebiotics. Prebiotics are non-digestible dietary components that pass through to the colon and selectively stimulate the proliferation and/or activity of populations of desirable bacteria in situ (Gibson & Roberfroid, 1995; Van Loo et al., 1999). Due to the potential synergy between probiotics and prebiotics, foods containing a combination of these ingredients are often referred to as synbiotics (Gibson & Roberfroid, 1995; Collins & Gibson, 1999). Prebiotics might influence the growth and survival of the probiotic by influencing the growth and metabolites of both the probiotic and the starter. This has to be kept in mind while considering interactions between probiotics and starters in fermented dairy products. Interaction between the probiotic and the

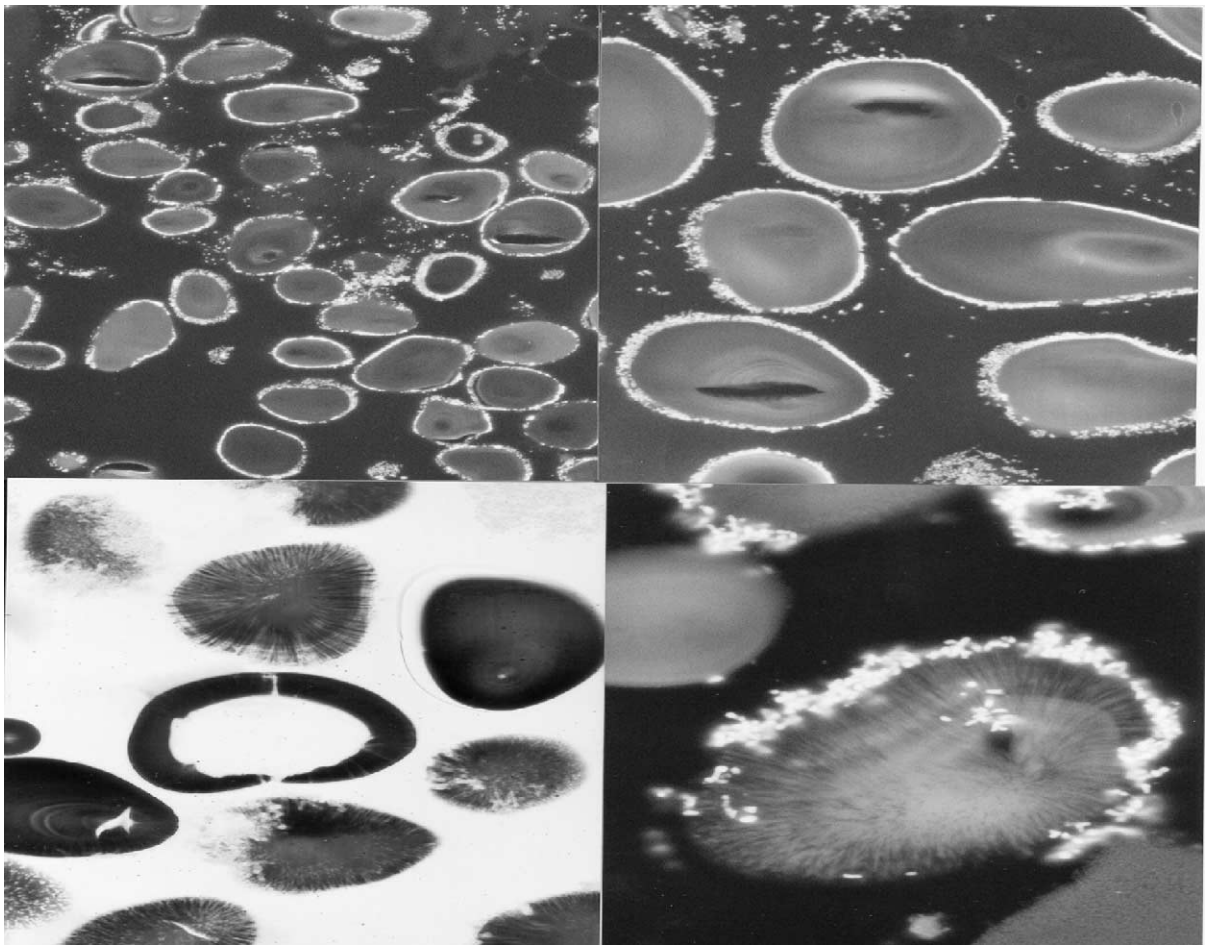

Fig. 3. Adhesion of bifidobacteria on the surface of large potato starch granules (DAPI and iodine stain, microscopy cross section, thickness 4 μm, 290× and 725× (upper panel), 725× and 1450×).

prebiotic in vivo might be favoured by an adaptation of the probiotic to the prebiotic substrate prior to consumption. This might result in a competitive advantage for the probiotic if it is consumed concurrently with the prebiotic (Fig. 4).

The prebiotics identified thus far are non-digestible carbohydrates including lactulose, inulin, and a range of oligosaccharides that supply a source of fermentable carbohydrate for beneficial bacteria in the colon (Crittenden, 1999). Some starches also escape complete digestion during passage through the human small intestine and arrive in the colon as fermentable carbohydrate sources for intestinal bacteria (Cummings & Macfarlane, 1997a, b). Granular starches synthesised by a number of food plants provide examples of these resistant starches, being incompletely digested due to their size and molecular conformation (Vonk et al., 2000). In animal models, the inclusion of resistant starches in the diet has been shown to increase the numbers of bifidobacteria in the intestinal tract (Brown et al., 1997; Brown, Wang, Topping, Playne, & Conway, 1998; Kleessen et al., 1997; Silvi et al., 1999; Wang, Brown, Evans, & Conway, 1999).

The benefits of using resistant starch extend beyond traditional prebiotics, since resistant starch can be used to ensure the viability of probiotic populations from the food to the large intestine. Resistant starch offers an ideal surface for adherence of the probiotics to the starch granule during processing, storage and transit through the upper regions of the gastrointestinal tract, providing robustness and resilience to environmental stresses. Bacterial adhesion to starch may also provide advantages in new probiotic technologies to enhance delivery of viable and metabolically active probiotics to the intestinal tract (Crittenden et al., 2001). This includes the previously mentioned technology to encapsulate probiotics within starch granules that are then coated with amylose (Myllärinen et al., 2000). Binding of adhesive strains to the resistant starch core may facilitate encapsulation of the bacteria using this technology. Encapsulation of starch-utilising bifidobacteria together with starch also ensures physical association

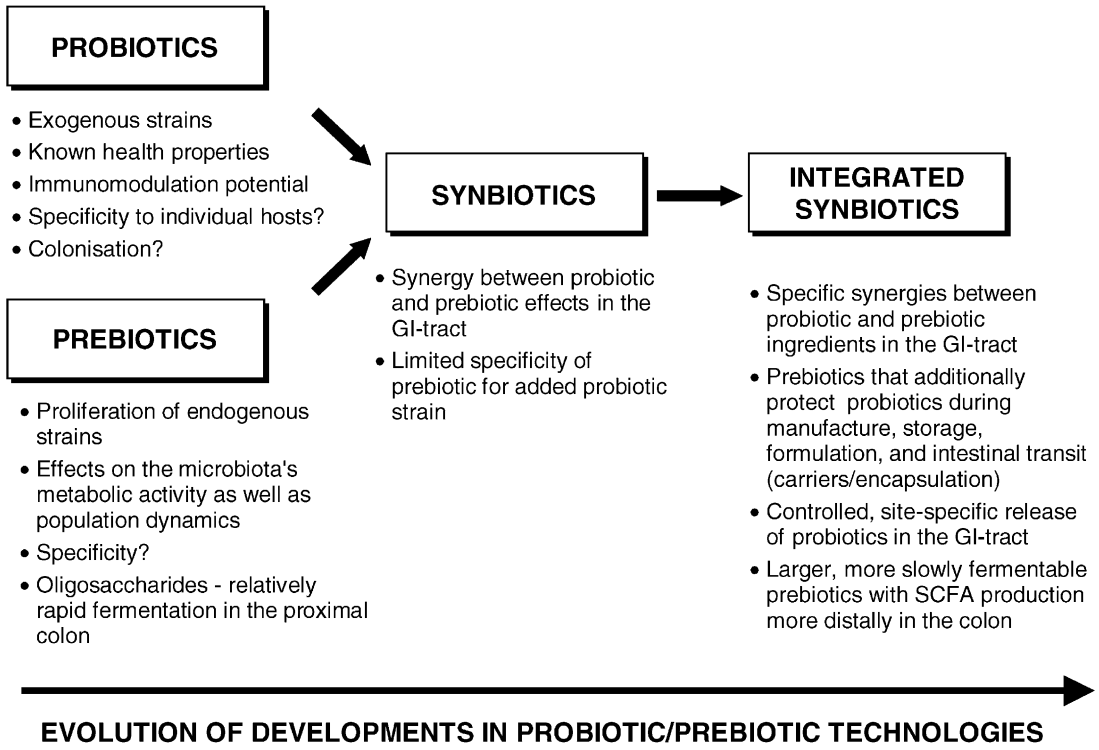

Fig. 4. Evolution in developments of probiotic/prebiotic technologies.

between the probiotic and prebiotic. This may enable relatively selective utilisation of the substrate by the probiotic, providing a selective competitive advantage for the added probiotic within the intestinal tract.

8. Research prospects

Diet is a major focus of public health strategies aimed at maintaining optimum health throughout life, preventing the early onset of chronic diseases such as GI disorders, cardiovascular disease, cancer, osteoporosis, as well as promoting healthier ageing. Although the highly complex relationship of food and health is still poorly understood, recent research advances in a variety of different disciplines provide promising new approaches to improve our understanding. The growing demand for 'healthy' foods is stimulating innovation and new product development in the food industry internationally. The food industry has a central role in facilitating healthier eating practices through the provision and promotion of healthy foods.

Continuously increasing consumer health consciousness and expenditure are socio-economic factors responsible for the expanding worldwide interest in functional foods. Considerable confusion and scepticism, however, exists between consumers, consumer organisations, scientific communities and media about the claims associated with probiotic products. Recent EU projects have demonstrated that, with co-ordinated efforts towards communication and a scientific approach to selecting and applying probiotics, functional food products can be developed with measurable health benefits for consumers. Probiotic strains can be successfully manufactured and incorporated into highly acceptable food products where they can retain their viability and functionality. There are many strain to strain variations, not only in their technological properties but also in their effects on human health (Mattila-Sandholm et al., 1999).

Currently, industrial demand for technologies ensuring probiotic stability in foods remains strong. The encapsulation technology for probiotic or protective cultures provides promising prospects for improved culture performance. However, development of this technique is still in the research phase and far from a final optimised commercial process. Valid techniques for ensuring probiotic stability and for optimising fermentation procedures, including development of inexpensive, non-dairy, edible culture media for probiotics are important. Furthermore, pilot- and commercial-scale production of new anaerobic probiotic/protective cultures will continue to be challenging. The stress factors influencing the viability and functionality of organisms need to be explored and controlled. Research toward in situ diagnostic tools for quality control of probiotic stains would be a huge step towards well-controlled product development.

The probiotic concept is today widely spread in the scientific and industrial fields. However, further scientific input is required. Important target research areas, including GI-tract diagnostics and immunology, methodology, biomarkers, and functionality, will lead to tools and scientifically sound methods for well-designed informative human studies. Controlled human studies are essential for the success of probiotic functional foods, and they should be tailored for specific population groups such as the elderly and babies. Future research on probiotic bacteria will centre on selecting new and more specific strains for the well being of the host (age groups, healthy populations, disease specific). The future scientific and technological research trends will be:

- To study the mechanisms of action of probiotics in the GI-tract, and develop diagnostic tools and biomarkers for their assessment.
- To examine the effects of probiotics on GI-diseases, GI-infections, and allergies.
- To ensure the stability and viability of probiotic products by developing feasible technologies (e.g. process and material development for microencapsulation).
- To develop technology for non-dairy, novel or artificial probiotic applications.
- To evaluate the role of probiotics in healthy consumer groups and to address consumer aspects.

An EU cluster on probiotics and prebiotics called ''Food, GI-tract functionality and human health'' was launched at the beginning of 2001, and will continue for 3.5 yr. The cluster consists of 42 laboratories from 12 countries and includes the pre-eminent probiotic research expertise within Europe. Significant achievements will be established within pre-competitive research areas such as biomarkers, mechanisms of action, new diagnostic tools for specific detection of gut microflora, as well as population dynamics, prebiotic modification methodology and designed health microbes with targeted effects. The cluster programme includes scientific, industrial and consumer platforms, disseminating research innovations to targeted audiences.

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