Review article

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Lactobacillus acidophilus and Non-Digestible Carbohydrates: A Review

Haia Abobakr Al-kaf [1]*, Noor Azwani Zainol[2] Roslinda Abd Malek[2], Fahrul Huyop[1]

ABSTRACT

In the recent years, lactic acid bacteria species such as *Lactobacillus* are considering one of the important species of probiotics used in the food processing sector to produce fermented products. It plays a significant role for the transformation and preservation of food. Besides, there is a huge exploration of new molecules that promote health and exhibit potential for technological applications such as non-digestible carbohydrates. The non-digestible carbohydrates provide various health benefits such as balancing and sustaining the microbiota in the intestine and increasing the production of short chain fatty acids (SCFA). The aim of this review is to discuss some aspects of non-digestible carbohydrates as an enhancer for the growth of probiotics. These compounds can help in improving many characteristics of food such as sensory and textural properties.

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Introduction

Probiotics are live microorganisms and play an important role in the digestive system by keeping the gut healthy by balancing the beneficial microflora [1]. In last two decades, the use of probiotics has been increased significantly because of probiotic's ability in conferring many health benefits to human's digestive system like protecting the host from different harmful microorganisms and making the immune system stronger. Non-digestible carbohydrates are dietary fibers (prebiotics) and cannot be digested and absorbed by the small intestine. Prebiotic is defined as non-digestible food which helps in improving and balancing the growth of the beneficial microflora in the digestive system. Plants are considering the common source of these non-digestible carbohydrates and contain a mixture of polysaccharides which are the integral components in the cell wall of the plants [2]. Many

[1] Department of Biosciences, Faculty of Science, Universiti Teknologi Malaysia,81310 Johor Bahru, Malaysia
[2] Institute of Bioproduct Development (IBD), Universiti Teknologi Malaysia,81310 Johor Bahru, Malaysia
* Correspondence: haiaalkaff10@gmail.com/ fzhutm@gmail.com

researchers proved the ability of prebiotics (non-digestible carbohydrates) in inhibiting the attachment of pathogens to epithelial cells and it is depending on the secretion of the peptides which are responsible in enhancing the absorption of minerals, stimulating the appetite, and preventing from dangerous diseases. In 1995, the term "synbiotic" was introduced by Gibson and Roberfroid which referred to a combination of both probiotics and prebiotics [3]. Consuming synbiotic can stimulate and activate the metabolism of a physiological intestinal microbiota [4]. As a result, using synbiotic can help in overcoming problems related to the survival of probiotics in the gastrointestinal tract. The combination of both probiotics and prebiotics as a single product can lead to great influence compared of using probiotic or prebiotic alone [5]. Thus, current studies are focusing on studying and selecting carbohydrates components which can be utilized by probiotic strains in high levels to produce synbiotic products to enhance the growth of beneficial bacteria in the gastrointestinal tract.

Probiotics

In the recent years, many scientists explored about the broad advantages of using functional foods as a sustainable product for the health of the beneficial bacteria in the digestive system. Nowadays, lactic acid bacteria (LAB) species are widely used in food industries to produce fermented products. For a long time, many countries use LAB species in food processing industries to produce fermented milk. Species of probiotics are playing a big role in the colonization of the mucosal surface for the gastrointestinal tracts of humans [6]. In many years ago, researchers been working on the isolation of LAB naturally to be used as starter culture to produce fermented food [7]. LAB provide antimicrobial activities against food borne pathogens based on the production of different inhibitory mechanisms like organic acids, hydrogen peroxides, diacetyl and ammonia [8]. Probiotics can stand the harsh conditions such as the salivary enzymes, the acidic environment of the stomach, bile acids and salts that present in the gastrointestinal tract [9]. Probiotics exhibit many health benefits to the host if they are provided in adequate amounts. Studies indicated that probiotic bacteria can regulate the health of the gastrointestinal tract and vagina. Moreover, probiotics provide some essential mechanisms such as the regulation of the intrinsic defenses by increasing the antimicrobial peptide production through paneth cells and the mucin production through

goblet cells, and degradation of the luminal for both antigens and toxins. Probiotics was firstly isolated from many food products [10]. The common species of probiotics are *Bacteriodes, Bifidobacterium, Enterococcus, Escherichia, Lactobacillus, Lactococcus, Leuconostoc, Pediococcus, Ruminococcus, Saccharomyces* and *Streptococcus* [11]. Many studies showed that probiotics can provide positive effects and play an effective role in fighting some of the gastrointestinal diseases such as irritable bowel syndrome, elimination of *Helicobacter pylori*, inflammatory bowel disease, and allergic disease. Also, several clinical studies proved the efficiency of probiotics in treating some diseases like obesity, insulin resistance syndrome, diabetes type 2, and non-alcoholic fatty liver disease [5]. In addition, the common strains of Gram-positive bacteria and yeast used in probiotic products are the genus of *Bacillus* and the genus of *Saccharomyces* respectively [12]. In the few years, there was a significant increase in the production of dairy products that contain probiotic bacteria like fermented milk, baby food, cheese, and ice cream. Commonly, probiotic food produced via fermentation process using fruits, vegetables, and cereals as substrates. The effectiveness of the consumed probiotic food products is depending on the number of active cells that been consumed by the individuals. Thus, increasing shelf-life of probiotics must be maintained during the production of probiotics foods. The addition of *Bifidobacterium* and *Lactobacillus acidophilus* to probiotic foods was reported to increase the nonspecific immune phagocytic activity of the circulating blood granulocytes. A study showed that the intake of yogurt containing a probiotic strain can help in stimulating the production of cytokines by blood mononuclear cells [13].

Lactobacillus acidophilus in food industries

Lactobacillus acidophilus is a type of lactic acid bacteria which is capable of fermenting sugars and yield lactic acid as a final product. *L. acidophilus* is a homofermentative, Gram-positive, rod in shape bacteria. This strain can exhibit many useful benefits such as providing thermostability, maintaining the growth activity at a wide pH range, and offering a strong inhibition actions against spoilage of food and pathogenic bacteria. These properties make *L. acidophilus* an important class of bio-preservatives and widely used in many fermented food products. Table 1 shows some common examples of *L. acidophilus* fermented products. Consumption of *L. acidophilus* can help in decreasing the blood cholesterol, reducing the

risk of mutagenicity and carcinogenicity and it can help in decreasing constipation, diarrhea and lactose intolerance. In addition, for a long-time *L. acidophilus* has been introduced into many dairy products, commonly yoghurt and sweet *acidophilus* milk by inoculating *L. acidophilus* into the milk [14]. Furthermore, the mechanism of *L. acidophilus* makes it a good competitor, and able to adhere easily to epithelial cells of the intestine and enhance the immunity and it is identified as nontoxic strain [15]. Moreover**,** there is no difference between *acidophilus* milk and non-fermented but from a nutritional perspective, consuming *acidophilus* milk is more beneficial especially for those who have less lactase (lactose-digesting enzyme). Meanwhile, lactose can be hydrolyzed by *β*-galactosidase in *L. acidophilus*. Thus, it is important to increase the shelf-life of *L. acidophilus* in the fermented products by providing efficient substrates to be utilized fully as an energy source. Besides, final pH and storage temperature are critical factors to be consider for the stability determination and the survival of *L. acidophilus* [8].

Table 1 Examples of common available Lactobacillus acidophilus products [15]

Meat-based products	**Dairy products**	**Non-dairy products**
Sausage	*acidophilus* milk	Soy milk
Salami	Yogurt	Boza
Ground beef	Miru-miru	Fermented tomato juice
Dry fermented pork loins	Kefir	Kombucha

Non-digestible carbohydrates

The use of prebiotics particularly dietary fibers (non-digestible carbohydrates) attracted many scientists. The dietary fibers can be metabolized by the intestinal microbiota and this will result in the production of short-chain fatty acids. Besides, the mechanism of non-digestible carbohydrates during fermentation can offer an anti-inflammatory and immunomodulatory abilities to treat many pathological conditions [16]. The non-digestible carbohydrates are dietary fibers which are not digested and absorbed by human's small intestine such as cereals, legumes, fruits and vegetables. The categorization of dietary fibers is mainly based on sources, solubility and fermentation properties. Many studies highlighted

about the importance of using non-digestible carbohydrates because of their advantages to human's health such as lowering the blood cholesterol and enhancing the function of the large bowel. Non-digestible carbohydrates are comprising of a carbohydrate polymers which are the components of plant cell walls such as cellulose and hemicelluloses [17]. Moreover, non-digestible carbohydrates are used as energy source for the growth of probiotics based on fermentation in order to stimulate the microbiotas activities and keep the host healthy [18]. Non-digestible carbohydrates can be found in many natural sources such as asparagus, sugar beet, garlic, grains and onion. Generally, non-digestible carbohydrates can be synthesized by different reactions such as hydrolysis, isomerization and fructosyltransfer [19]. The use of non-digestible carbohydrates can help in improving the quality of final products based on the sensory properties, texture, and physicochemical characteristics [20]. Prebiotics (non-digestible carbohydrates) can also regulate obesity by the expression of intestinal anorectic hormones by reducing the number of orogenic hormones [21].

Sweet potato

Sweet potato (*Ipomoea batatas*) belongs to *Convolvulaceae* family (*Morning glory*). It is one of the important agricultural crops. It supplied with dietary fibers and vitamins and one of the best favourable products because of the various properties it has such as its versatility and its strong resistance to different environmental conditions [22]. In general, the bioactive compounds of sweet potato are metabolized steadily in intestine where there are variety of different probiotic floras, hence these floras are helping in releasing and transforming bioactive compounds and yield bioavailability through fermentation. An analysis was carried and it showed that few individuals were having problems related to digestion and were not able to digest the nutrients in sweet potato. Thus, fermentation of this nutrients by *L. acidophilus* is believed to act as a pre-digestion stage; helping in increasing free form bioactive compounds of the sweet potato and increase the bioavailability by reducing the content of sugar [23]. Since sweet potato is comprising of many carbohydrates, this make it a good substrate to be fermented by *L. acidophilus*. Hence, this leads in releasing high levels of bound bioactive compounds during fermentation along with the formation of free secondary metabolites [24]. Previously, it was reported that sweet potato has the potency to act as prebiotic source and utilized by *Lactobacillus* species based on *in vitro* fermentation

[25]. The amount of dietary fibers in sweet potato are not constant and subjected to the genetic conditions of the crops and the analytical methods used. The main compounds of carbohydrates source in sweet potato root are sucrose, maltose, and glucose and these compounds give the sweetness taste for the sweet potato root [26]. Furthermore, studies on monkey kidney cell (CV-1) indicated that fermented sweet potato can inhibit pheochromocytoma derived cancer cell (PC-12) along with the amount of cytotoxicity to normal [24]. It helps in bringing up a novel technique for the utilization of sweet potato by *L. acidophilus* to improve the anticancer capabilities needed for health.

Yam

Yam which known as (*Dioscorea spp.*) is one of the high demanding food recently and has a significant role for small and marginal rural families in the world [27]. There are a lot of yam processed products such as pounded yam, boiled yam, fufu, amala, and elubo. Yam has high contents of sugars which are sucrose, glucose, fructose, and maltose. All these sugars are very suitable to be used in microbial fermentation. Furthermore, it also has amounts of polyphenols, allantoin, and diosgenin which contribute many health benefits. For instance, Allantoin is used to protect the skin, while diosgenin provides benefits like antitumor and lipid metabolism [28]. In addition, *Dioscorea* tubers have many nutritious benefits compared to other root crops [29]. Yam is rich with carbohydrates and it is a good source of prebiotics and has bioactive components; if it is administered into probiotic products it will help in increasing and balancing the growth of the microbiota in human's gut [30]. In addition, it contains prebiotic properties since it is rich with polysaccharides; this make the dietary fibers in yam resistant to the hydrolysis of the human digestive enzymes present in the intestine which are specific for glycosidic bonds as shown in Figure1.

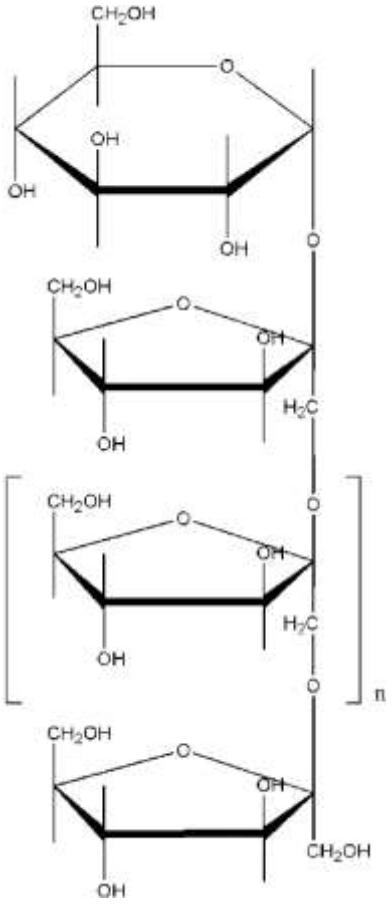

Fig 1 Structure of inulin, a linear fructosyl polymer linked by *β*-(2,1) bonds [31]

In addition, *Lactobacillus* was capable to ferment yam as energy source *in vitro* and the level of lactic acid produced was 8.1 g/L after 6 h of fermentation [32]. Yam possess many benefits to individuals and it is very useful in the pharmaceutical industries to produce dietary supplements and some cosmetic products. It can also regulate the production of sex steroid hormones [33].

Barley

Barley (*Hordeum vulgare L*.) is believe to be one of the most important cereal in the world beside maize, rice, and wheat. Barley can adapt easily to any environmental conditions. The great increase of barley use in food area is because of the functional components that barley has which is the *β*-glucans which contribute to many health benefits such as lowering the

level of blood cholesterol and control the glycemic index [34]. The *β*-Glucans in barley forms the water-soluble fraction of many cereals and are stored in the cell walls of the aleurone and subaleurone layer of barley [35]. Figure 2 shows the structure of *β*-glucans in barley.

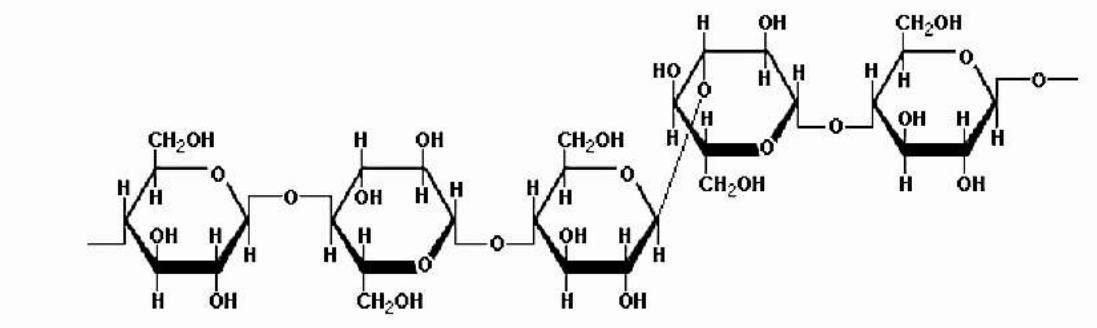

Fig 2 The structure of *β*-glucans in barley [36]

It was reported that fermentation of barley exhibit a good enhancement for food's quality. Based on previous studies, results showed that fermentation of barley with *Lactobacillus* species possess a great influence on the level of phenolics and *β*-glucan compounds [37,38]. Moreover, the *β*-glucan in barley, was characterized by groups of contiguous (1 → 4)-*β*-linkages and isolated (1 → 3)-*β*-linkages and it was reported that there were positive effects recorded on the health of animals and humans, and this showed the ability of barley in stimulating and modulating the beneficial bacteria in the immune system, pathogen inhibition, anti-tumor and anti-cancer activity, and reduce cholesterol levels [39]. Some of the *in vitro* studies have shown that (1→3) (1→6)-*β*-glucans can improve the activity of macrophages, besides activate the antimicrobial activity for both mononuclear cells and neutrophils and this result helps in making the immune system much stronger due to the increase of pro-inflammatory cytokine levels and chemokine [40].

Garlic

Garlic (*Allium sativum*) is mainly used as a treatment and as a preventer from diseases across the world [41]. Garlic can promote many benefits and in the past it was used in ancient Egypt as a therapeutic plant to cure some diseases. Garlic allicin is an antibiotic and antifungal which gives the specific smell in garlic. The chemical structure of allicin is shown in Figure 3 [42].

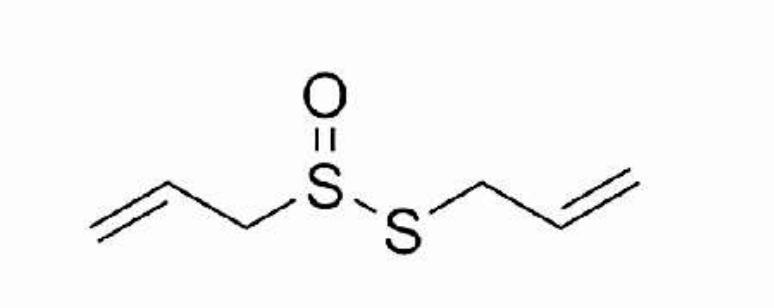

Fig 3 The chemical structure of allicin [43]

Based on a previous study [42], it was reported that the addition of garlic as a prebiotic source for lactic acid bacteria can enhance its growth and contribute a significant role to improve the growth and activity for some strains in the human's gut. The effectiveness of the antibacterial activity in garlic makes it important in fighting various kinds of Gram-negative or -positive bacteria [44]. Moreover, *Staphylococcus*, *Mycobacteria*, and *Proteus* species were recognized to have high sensitivity towards the antibacterial activity present in garlic, hence this will help in fighting pathogenic bacteria if garlic administered in high levels [45].

Banana

Banana which known as (*Musa acuminata*) is a common consumed fruit in the world and it is rich with vitamin B6, vitamin C, potassium and resistance starch; whereby individuals not able to digest it in the small intestine thus, it can pass to the large intestine and it produces short chain fatty acids, carbon dioxide, methane and hydrogen in it. The produced fatty acids offer health benefits to human such as reducing the pH of the colon, protect the gut from colon cancer, inhibit the formation of any carcinogenic amines, and enhance the process of feces discharging [46]. For prebiotic properties banana is consisting of 60 to 80% of non-digestible carbohydrates [47,48]. These facts make banana a good source of prebiotic for *Lactobacilli* species. Based on the previous research [49], the addition of banana powder to certain species of gut microbiota was capable to regulate the microflora in the intestine and produced different short-chain fatty acids (SCFAs) through *in vitro* fermentation process after 24 h. Figure 4 demonstrates the chemical structure of the SCFAs produced.

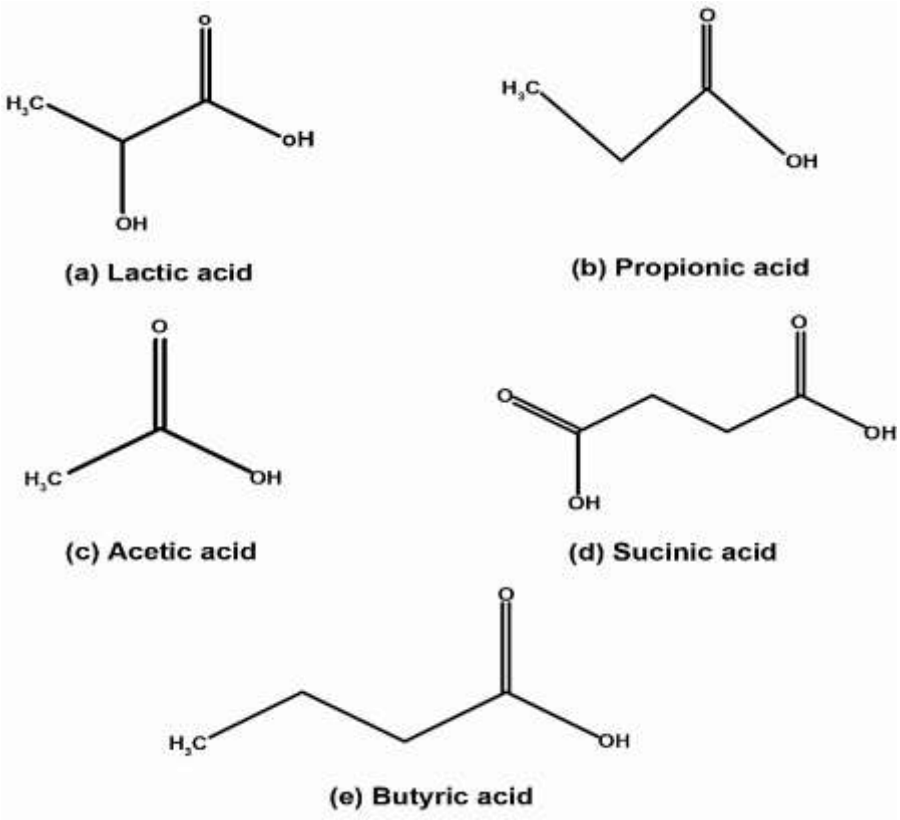

Fig 4 The chemical structures of SCFA [50]

Current trends of probiotics and prebiotics

Nowadays, functional food is receiving greater attention by many people in the world. This attention is due to the advances in food technology particularly in food processing sector which offered many positive effects in the manufacturing of processed products that are associated with health benefits. However, some of the functional ingredients can alter the properties of the products such as the texture of food. In addition, the use of probiotics strains and prebiotics is increasing and many studies focusing on producing functional products with better textural properties and able to contribute positive effects to human health [51]. Consumers now are more cautious in purchasing their food products and more knowledgeable about the unhealthy properties or ingredients in food like artificial additives. Therefore, they are trying to look for products that contain more natural and healthy features [52]. In addition, the production of functional foods is offering many benefits like fighting

against common health disorders specifically the chronic diseases such as obesity, cancer, diabetes, and cardiovascular problems [51]. The use of probiotics and prebiotics in the processing of functional ingredients is believed to provides many health benefits and alters the food texture to makes it more safely to consume and beneficial to enhance the sensorial pleasant of the products [52]. Texture in food production is important as it is a sensory characteristic for food products and it plays a role for the perception and enjoyment in terms of food quality. The addition of prebiotics mainly non-digestible carbohydrates into the ingredients of food products can alter the structure of the food because of the fibers content in which can change the textural properties depending on the degree of polymerization (dp) and the chain length, since it is important for the solubility of fibers in water and interaction with other food compounds such as proteins [53]. Moreover, some probiotics strains can change the food texture as well, depending on the production of exopolysaccharides by lactic acid bacteria. The studies that contributed in texture changes by the addition of probiotics can be seen in Table 2. Generally, for the texture alteration of food products it is important to choose strains and prebiotics which are effective for fermentation process such as the use of *L. acidophilus* strain which is commonly used in many food industries.

Table 2 Effects of probiotics in the texture of food products

Study	**Probiotic strain**	**Food product**	**Effects**
[54]	*Lactobacillus rhamnosus*	Cream cheese	-Influence the firmness due to the acid development and proteolysis activity.
[55]	Exopolysaccharide producing lactic acid bacteria	Carrot puree	-Observed texture was associated with the types of produced EPS. -The fermentation using different bacteria strains resulted in changing the product texture.
[56]	*Lactobacillus casei 01*	Ice cream	-The ice cream produced had a lower fat destabilization and a huge increase was reported in viscosity, however, the melting time was reduced.
[57]	*Lactobacillus rhamnosus GG Bifidobacterium breve*	Panela cheese	-The cheese produced showed greater consumer acceptance based on the compactness, hardness, moisture, and softness characteristics.

Furthermore, in the past few years, functional food products that contain both prebiotics and probiotics (synbiotic) were highly demanded. Food industries nowadays are trying to develop non-dairy synbiotic products due to the various health benefits that they can provide such as enhancing the growth of beneficial bacteria in the gut. Thus, the addition of probiotic strains into fruit matrices which are rich in carbohydrates and dietary fibers can contribute to provide health benefits to the host. For dairy products, a problem is always occurring; which is the low survival rate of the probiotic bacteria used during storage however, many studies reported about the overcoming of this problem by the addition of non-digestible carbohydrates along with probiotics strains; because non-digestible carbohydrates contain high amounts of fibers which can be utilized by bacteria in order to increase its growth and shelf-life. Consumption of synbiotic food can help in modifying the microbiota composition and metabolic activities in the gut. In addition, it is reported that synbiotic can help in increasing the levels of *Lactobacilli* and *Bifidobacteria*, boosting the functions of the liver, helping in the prevention of bacterial translocation, and reducing susceptibility to pathogens in surgical patients [58]. As synbiotic is a combination of probiotics and prebiotics, therefore it can provide both prebiotic and probiotic effects. Probiotics help in improving the growth of beneficial bacteria, while prebiotics stimulate the growth of a specific group of bacteria present in the gastrointestinal tract. The use of synbiotic is extremely recommended since probiotic without its prebiotic food cannot survive longer in the digestive system. Accordingly, probiotics need an efficient source of prebiotic to be used for their growth and this will result in gaining better intolerance for oxygen, low pH, and temperature. Combining both prebiotics and probiotics into synergy will result in increasing the number of beneficial bacteria [59].

Future Prospect

The current trends dictated by the increase of interest towards purchasing healthier food which result in offering an opportunity for the use of novel, economy and technological matrices for the improvement of the functional products [60]. These potential heterogeneous matrices are held to be opened to innovate and need evaluation for both efficacy and safety

to ensure the consumer's acceptance. The notable feature in the marketing strategy will be based on the exclusive health claims and the consumer's understanding [61].

Conclusion

The use of probiotics in the last years has received a great attention to produce fermented products that contribute and offer many benefits to the health. This review has highlighted about some of non-digestible carbohydrates to be used as carbon sources and enhancer to promote the growth of *Lactobacillus acidophilus* and other LAB strains.

Abbreviations
LAB: lactic acid bacteria; dp: degree of polymerization; SCFA: short chain fatty acids; h: hours

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