

# Chapter 7

## Prebiotic and Probiotic Potential of Cereals



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### 7.1 Introduction

A food that improves human life and cures ailments is comprises of nutrient (prebiotic), which is processed by specialised bacteria (probiotic), and products (postbiotic) that are physiologically active, is one of the most intricate loops. Clinicians, microbiologists, dieticians, nutritionists, food technologists, and farmers are all interested in probiotics, their nutrients–prebiotics, and postbiotics. Probiotics play vital role for preventing and treating a variety of infectious and non-infectious diseases (Salmerón, 2017). Anti-diarrheal, anti-carcinogenic, anti-mutagenic, anti-inflammatory, antibacterial action, immune system activation, improvement in lactose metabolism, and suppression of gastrointestinal infections are all documented benefits. Mainly probiotic meals, which include microorganisms from the lactic acid bacteria family, are generally limited to drinks and cheese. Health-promoting foods are usually dairy-based, consisting of milk and its fermented products. Furthermore, lactose sensitivity and the cholesterol level of dairy products, together with the vegetarian tendencies of a wide range of third-world people, have

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pushed for the recent adoption of non-dairy beverages. On the other hand, several studies have focused on the probiotic capability of fermented cereal-based beverages, which are particularly popular in poor nations with low nutritional safety and a high frequency of gut infections (Mohammadi et al., 2021). Probiotic bacteria originate usually from humans or animals, however strains that are identified as probiotics can also be found in fermented non-dairy foods (Agrawal et al., 2000; Aidoo et al., 2006).

In current era, the chronic diseases, especially the non-communicable ones along with inflammatory disorders, usually associated with wrong habit of feeding the foods with poor fiber content in them and with higher content of refined carbohydrates and fats alongside. This resulted in increased number of diseases, making the researchers to focus on the importance of prebiotics which has active mechanism in controlling the process of infectious diseases (Rolim, 2015). Cereals, being one of the major components of diet possess dietary fiber along with their prebiotic potential. The effects of dietary fibers on human health are well documented and widely accepted as well, and prebiotic foods nowadays are getting much importance due to their beneficial effects and high nutritional status. According to solubility in water, dietary fibers are categorized as water soluble or water insoluble. The former one i.e. water soluble or water extractable dietary fibers includes  $\beta$ -glucan, arabinoxylans, fructo-oligosaccharides whereas the later one i.e. water insoluble or water unextractable dietary fibers include resistant starch, cellulose, etc. Water insoluble dietary fibers are generally more resistant to the colonic fermentation and therefore possess more prebiotic effects in contrast to water soluble dietary fibers. However, the potential of prebiotic depends upon the physicochemical properties and is affected by chemical structure, degree of polymerization and many more (Abdi & Joye, 2021).

Lactic acid fermentation of cereals is a widely adopted technique that is used to make a variety of foods throughout Asia and Africa, including porridge and beverages. Cereal grains, primarily sorghum and maize are soaked for 0.5–2 days in clean water. Soaking softens the grains, making it simpler to smash or wet-mill them into a slurry, from which hulls, bran particles, and germs can be sieved off. Mixed fermentations, including lactic acid fermentation, take place during the slurring or doughing stage, which lasts 1–3 days. During fermentation, the pH drops while the acidity rises as lactic and other organic acids accumulate as a result of microbial activity.

Lactobacilli and bifidobacteria have complicated dietary requirements that vary greatly from species to species, including vitamins, nucleic acid derivatives, fatty esters, peptides, amino acids, and carbohydrates (Severson, 1998). Starch, water-soluble or -insoluble dietary fibre components, and numerous free sugars such as arabinose, sucrose, maltose, fructose, xylose, stachyose, glycerol and glucose are the major component of cereals, determine the contents of these components. The ability of a probiotic starter to survive the acidic environment of the final fermented product, as well as the harsh conditions of the gastrointestinal tract, is a critical element in its selection. The metabolites generated by the starter, such as lactic acid and acetic acid, hydrogen peroxide, and bacteriocins, may affect the probiotic bacteria's viability in vitro (Saarela et al., 2000).

## 7.2 Why Cereal-Based Foods

Cereals, no doubt being a rich source of nutrition offers various challenges also, such as its swelling power upon cooking due to the presence of starch in them, limiting presence of amino acids in them along with limited and lower mineral bioavailability because of presence of number of anti-nutritional factors such as phytic acid, tannins, etc. Besides this, cereals are grown in more than 73% of the total harvested area globally and it contributes to more than 60% of the total production of food in the world. They are the major contributors of the protein, dietary fibers, vitamins, minerals and provide energy in optimum amount that is desired for the human health (Nout, 2009).

Therefore, the cereals or its constituents can possibly be employed for the formulation of functional foods, where the cereals can be used as the fermentable substrate for growing of probiotic microorganisms such as *bifidobacteria* and *lactobacilli*. These can also be used as the major source of dietary fiber for promoting the health benefits to the consumers. Also, due to the presence of specific content of non-digestible carbohydrates in them, they can either serve as prebiotic or as probiotic for increasing their stability (Charalampopoulos et al., 2002).

Various studies demonstrates that whole grain cereals play important role in protecting the body against various diseases such as cancer, cardiovascular disease, diabetes and other related diseases (Venn & Mann, 2004). This might be attributed to the presence of dietary fibers in whole grain cereals and due to presence of minerals in their outer fractions, which help them in fighting against the oxidative stress, carcinogenesis, hyperglycaemia and inflammatory disorders. Cereals with whole grain are carriers of micronutrients which include folates, zinc, selenium, manganese, betaine, sulphur amino acids, lignans, alkyl resorcinols, vitamin E, phenolic acids, iron, copper, carotenoids, choline, phytic acid, etc. (Das et al., 2012).

## 7.3 History of Cereal Based Beverages

Homo sapiens appear between 2000 and 1,000,000 years ago. The only drinks they drink are water and breast milk. About 11,000 years ago, the other beverages like animal milk, tea, beer, coffee, and afterwards fruit juices and soft drinks enter our diet. A large amount of archaeological evidence indicates that the production and consumption of beer was between 4000 and 3500 BC. At the same time, distilled alcoholic beverages were also developed. The Sumerians and Egyptians are considered to be the earliest winemakers. The domestication of crops has played a key role in beer brewing. Incorporating grains into the human diet is considered an important step in human evolution, because converting grains into staple food requires extraordinary technology and cooking skills (Wolf et al., 2008).

Poor water quality is one of the main factors for mankind to seek new beverages. Therefore, fermentation has been used to make various beverages, mainly for their

safety. Fermented alcoholic and non-alcoholic beverages were made in the Middle Ages. Most non-alcoholic cereal beverages are produced in Africa and South America (Altay et al., 2013). Some traditional African non-alcoholic beverages include Bushera from millet or sorghum, Kunan-Zaki from sorghum, Gowé from millet, Mahewu from corn, Obiolor from millet and sorghum and Obiolor from millet and sorghum, Borde from corn, millet, wheat, sorghum and barley. The last two beverages consist of several grains. South American non-alcoholic fermented beverages include Pozol, Agua-agria, Napú, Fubá, Acupe and Champuz. They are mainly made of corn. Kali as well as Ambil are the traditional fermented beverages from country India. The first one is made with millet and the second one is made with rice. Ambil uses yoghurt to ferment. Recently, in the past 200 years, beverage mixes made from malted grains have appeared. They are intended to be consumed with milk to increase its safety and flavor. The first malt powder was developed by William Horlick in the 1870s. By 1882, he had mastered the process of drying milk with malt and wheat to make the product easily soluble in water. The doctor recommended Horlick's food to their patients (Fernandesa et al., 2021).

## **7.4 Risks Associated with the Consumption of Dairy Products**

Many health risks are associated with the consumption of dairy based fermented foods, adhering to which people are now moving more towards cereal based fermented foods rather than dairy based fermented food products. Some of these issues include milk related allergies, lactose intolerance, high content of cholesterol in dairy based products, higher protein content, etc. Few of these are discussed in detail in the chapter below.

### ***7.4.1 Allergy Associated with Milk Proteins***

Atopic dermatitis is one of the diseases which is commonly associated with the allergy present in food amongst the children. The occurrence rate of atopic dermatitis in the initial first year lies between 2–3% (Høst, 2002; Jøhnke et al., 2006; Ricci et al., 2006). Various studies demonstrate that the use of some probiotics is beneficial in reducing the atopic dermatitis occurrence in human beings (Reid & Kirjaivanen, 2005). Also, it has been observed that not all of the probiotic preparations are used for the children who suffer from allergies associated with the milk of cow. However, by using the selective strains of probiotics the severity of the sensitivity can be reduced up to some extent in such children (Moro et al., 2006). Moreover, few studies suggests that the supplementation of probiotic has no significant effect on the any of the symptoms related to atopic dermatitis and in such cases,

there are chances of occurrence of increased allergen sensitization in the children suffering from atopic dermatitis (Lee et al., 2007; Moneret-Vautrin et al., 2006; Taylor et al., 2007).

### **7.4.2 Lactose Intolerance**

Lactose intolerance or mal absorption of lactose is one amongst the most common type of disorder related with mal absorption of carbohydrate. It is recognised when a person is not able to digest the lactose into its individual components i.e. galactose and glucose. This usually happens because of lack of enzyme responsible for digestion of lactose, which is known as lactase (Hauck et al., 2011). During birth, the activity of the enzyme lactase is at its highest, as the children consumes more of the milk as compared to adults and the activity of this enzyme decreases post weaning. In cases where lactose is not sub divided into its constituents, the colonic bacteria metabolizes the lactose and produces gases such as CH<sub>4</sub> (methane) and H<sub>2</sub> (hydrogen) along with the production of short chain fatty acids (Lee, 2014). The person starts facing the problems related to lactose intolerance just after consumption of milk or related products containing lactose after half an hour or 2 hours. The symptoms of lactose intolerance are recognized by cramping, loose stool, flatulence and bloating. In some cases there occurs irritability in the bowel syndrome also. The population of lactose intolerance people is highest among the Asian people, African Americans and Native Americans, however the lowest population of lactose intolerant people are found in Northern European.

### **7.4.3 High Cholesterol and Fat Content**

Milk is one the major sources of fats and the content of fat in ilk depend majorly on the source from where it is obtained. In case of cow milk, it has around 4–5% of the fat, while in case of buffalo milk; the fat content ranges from 7% to 8%. The ratio of poly unsaturated fatty acid to saturated fatty acid is 0.05. Consumption of large volume of milk generally enhances the low density lipoproteins cholesterol and levels of total cholesterol content in the blood. The saturated fat is majorly responsible for increasing the levels of plasma cholesterol, this in turn is responsible for increasing the risks associated with coronary heart disease. Such related risks can be reduced up to some extent by reducing the consumption of low density lipoproteins, which can be further be achieved by reducing the saturated fat content in diets. It has been observed that probiotics has potential beneficial effect on customers suffering from the hypercholesterolemia (Ebel et al., 2014).

## 7.5 Prebiotic Compounds in Cereal Dietary Fibers

### 7.5.1 Water Soluble Dietary Fibers

#### 7.5.1.1 $\beta$ -Glucan

It is one of the most important and major cereal water soluble dietary fiber. It is found in cell walls of endosperm in cereal grains.  $\beta$ -glucan (Fig. 7.1) comprises of D-glucopyranosyl units that are joined in a linear fashion to each other with a mixture of  $\beta$ -1-4 and  $\beta$ -1-3 glycosidic linkage. It has wide impact on the gastro intestinal health of the host due to variation in length and branching. Apart from cereal grains  $\beta$ -glucan is also observed in various marine plants, algae and mushrooms, with barley and oats having the highest content of  $\beta$ -glucan in them (Cloetens et al., 2012).

$\beta$ -glucan is getting attention from industrial point of view because of its unique properties like stabilizing, gelling and thickening. Besides this, it has biological effects due to its prebiotic property, thereby increasing the interests among the researchers to incorporate the dietary fibers in different foods. Also, due to variation in molecular weight of  $\beta$ -glucan, it becomes feasible to access the growth of microbes in colon. For instance, hydrolysates of 37 kDa, 150 kDa and 172 kDa promotes the growth of *Prevotella* and *Bacteriodes* species when tested in vitro, whereas the larger hydrolysates above 230 kDa had non-significant effect in enhancing the growth rate of the tested bacteria (Hamaker & Tuncil, 2014). Cancer prevention, anti-inflammatory action, skin protection, antioxidant, immunological modulation, and glycaemia and serum cholesterol reduction are only a few of the functional qualities that offer promise for human health. Any increase in arbinoxylan substitution resulted in bacterial population and short-chain fatty acid decrease. Because wheat has been used by humans for thousands of years, we hypothesised that the human gut microbiota has co-evolved to digest the ratio of dietary fibre

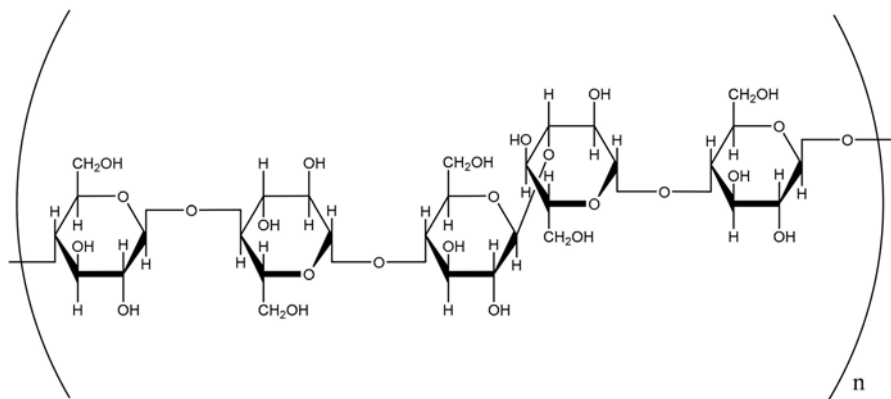


Fig. 7.1 Structure of  $\beta$ -glucan in cereals. (Abdi & Joye, 2021)

polysaccharides present in wheat more efficiently than other polysaccharide ratios. Beneficial bacteria have been demonstrated to preferentially ferment arabinoxylan over -glucan, with the ideal arabinoxylan:glucan ratio found in wheat (3:1) (Shoukat & Sorrentino, 2021).

### 7.5.1.2 Arabinoxylans

Arabinoxylans are the fragments of non-starch polysaccharides present in cell walls of cereals. It is composed of units of  $\beta$ -1,4 xylopyranose with moieties of arabinofuranose at C<sub>2</sub> position of xylopyranose units. Arabinoxylans are further divided into water extractable arabinoxylans and water unextractable arabinoxylans, on the basis of their solubility. Both types of arabinoxylans possess general structure. The water extractable arabinoxylans act as building blocks for the water unextractable arabinoxylans (Broekaert et al., 2011).

Arabinoxylans, being potential prebiotics are hydrolyzed by various hydrolytic enzymes of bacteria in large intestine. These enzymes include arabinofuranosidase and xylanase. The prebiotic activity of arabinoxylans is associated with its chemical structure. Various studies relating to the structural features of arabinoxylans claims that substitution of ferulic acid play important role in fermentation of arabinoxylans. There must exist a negative relation between fermentability of arabinoxylans and presence of esterified ferulic groups (Hopkins et al., 2003). Gut microbial fermentation can release ferulic acid found in arabinoxylans from various cereal grains. By stimulating the signalling pathways of Kelch-like ECH-associated protein-1 and nuclear factor E2-related factor-2, it can increase antioxidase activity and reduce reactive oxygen species levels. Cereal-derived arabinoxylans have been shown to have an important role in gut microbial population regulation, intestinal barrier function, immunological processes, and glucose and fatty acid metabolism. Activation of G protein-coupled receptors or suppression of histone deacetylase activity to reduce NF- $\kappa$ B signalling are currently the mechanisms of action of arabinoxylans on host health and metabolism, as mediated by short chain fatty acids generated by microbial fermentation. The gut bacteria ferment them to produce free ferulic acid and short-chain fatty acids. Ferulic acid activates the Nrf2 signalling pathway, which has pre-biotic effects on antioxidant capacity and inflammatory responses (Zhang et al., 2021).

Feruloylated arabinoxylan is not degraded by endogenous digestive enzymes in humans or animals, but it can be fermented by intestinal microbiota in the caecum and colon to create short chain fatty acids (Hald et al., 2016). As a result, insoluble ferulic acid is separated from cereal arabinoxylans in the intestine by microbial fermentation, which releases free ferulic acid, which is subsequently absorbed by the host. Bacteroides are the most common bacteria in the proximal colon, and they breakdown low-branched arabinoxylans into feruloylated xylo-oligosaccharide with low polymerization degrees (Pereira et al., 2021). When wheat arabinoxylan and barley -glucan were combined at a 3:1 ratio, the best prebiotic activity was identified, based on concentrations of total short chain fatty acids and increases in

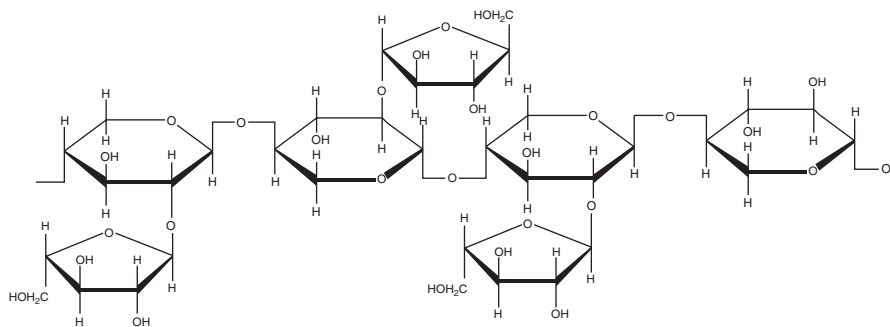
total bacteria as well as beneficial *Bifdobacterium*, *Clostridium coccoides*, and *Eubacterium* groups (Harris et al., 2019).

Short chain fatty acids, primarily lactic acid, acetic acid, propionic acid, and butyric acid, are produced by microbial fermentation of cereal-derived arabinoxylans. The production of short chain fatty acids by microbial fermentation is mediated by a set of fundamental processes mediated by the makeup and quantity of gut bacteria (Koh et al., 2016; Louis et al., 2014). Lactic acid is produced largely in the upper gut by microbial fermentation of soluble polysaccharides and indigested oligosaccharides, while acetic, propionic, and butyric acids are produced in the colon and cecum. Short-chain fatty acid synthesis is connected with a specific microbial population found in the host's foregut and hindgut. *Lactobacillus* is the most common lactic acid-producing bacterium; however, due to changes in the gut environment, such as pH and oxygen concentration, *Lactobacillus* abundance is reduced in the hindgut. Furthermore, the bacterial families *Prevotellaceae*, *Ruminococcaceae*, and *Lachnospiraceae* are abundant in the hindgut and are the most important producers of short chain fatty acids via microbial fermentation of cereal fibres (Liu et al., 2017). The chemical structure of Arabinoxylan is presented on Fig. 7.2.

### 7.5.1.3 Fructans

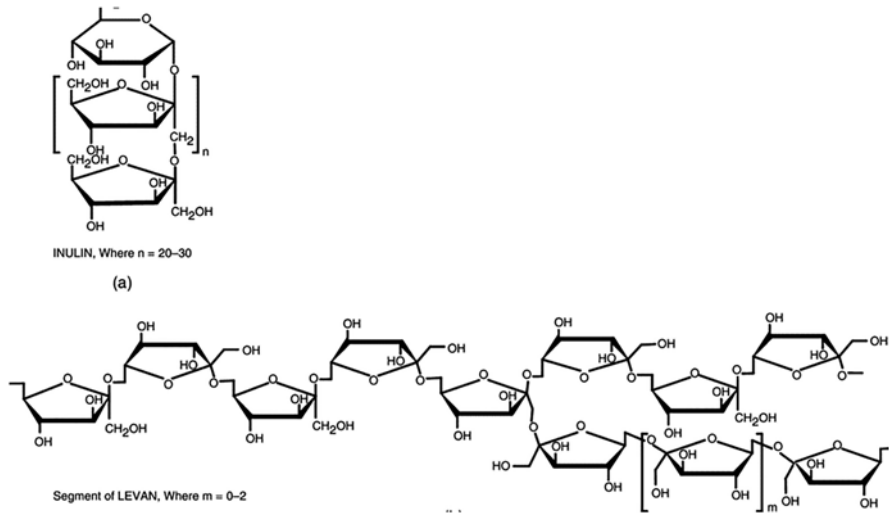
Cereals and their derivatives are the major source of fructans in our daily food. Although eating large amounts of cereal products can cause fructans to have a huge impact on colon health, eating large amounts of fructans (for example, up to 20 grams per day) may also cause bloating and mild flatulence depending up on consumer health and sensitivity. The fructan content of immature grains of most grains is significantly higher than that of mature grains. For instance, in the immature grains of triticale, barley, rye and wheat the reported fructan content is about 23.7–39.0 g/100 g of dry flour. In wheat grains, fructans are reported to accumulate during cell division and expansion stages (Van Loo, 2006).

Both *in vitro* and *in vivo* studies have proved that such fructans have the potential to promote health and have been regarded as prebiotics. Fructans are resistant to



**Fig. 7.2** Chemical structure of Arabinoxylan (Sinha et al., 2011)





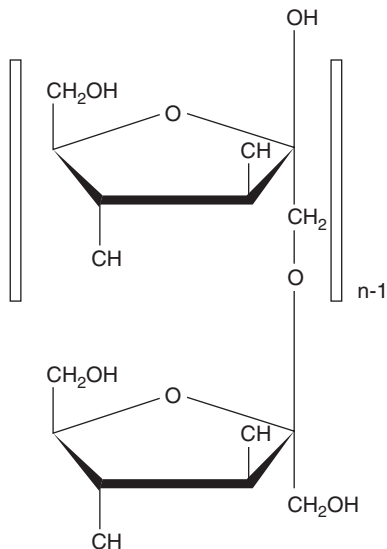
**Fig. 7.3** Chemical structure of two fructans: inulin and levan (Izydorczyk et al., 2005)

enzymes that occur in the upper gastrointestinal tract and they are easily hydrolyzed by specific hydrolytic enzymes of certain bacterial species in the lower gastrointestinal tract, so they are fermented there and produce fatty acids (short-chain) or gases. The positive health effects of inulin-like fructans are not limited to their effects on colonic bacteria, because recent evidence suggests that these fructans have a direct immunomodulatory effect on the innate immune system. Fructans arouse Toll-like receptors and are accountable for activating cellular responses, therefore, they seem to trigger a protective effect against oxidation *in vitro* (Pasqualetti et al., 2014). Figure 7.3 represents the chemical structure of Fructans.

#### 7.5.1.4 Fructooligosaccharides

Fructooligosaccharides are low molecular-weight oligosaccharides of fructose with degree of polymerization  $\leq 10$ . These are produced by sucrose's transfructosylation and contain 2-,4- $\beta$ ,2-1 linked fructosyl segments. Fructooligosaccharides are found in plant tissues in the form of osmoregulators or source of carbohydrates. Among different plants, the level of fructooligosaccharide is 10 times more in durum wheat. Also, the levels of fructooligosaccharide in immature wheat are comparatively higher as compared to the matured one. Fructooligosaccharides are well known for their potential to exhibit bifidogenic effects, however these effects are influenced by the polymerization length (Yeung et al., 2005). The structure of fructooligosaccharide is presented in Fig. 7.4.

**Fig. 7.4** Structure of fructooligosaccharide (Yeung et al., 2005)



## 7.5.2 Water Insoluble Dietary Fibers

### 7.5.2.1 Resistant Starch

Maltodextrin or resistant starch, is the starch that resists digestion in small intestine by an enzyme called pancreatic amylase. This undigested starch therefore reaches the colon. The behaviour of resistant starch is similar to fermentable fibers, say guar gum, which results in lowering of colonic pH and increasing the faecal matter. It also improves the bowel health, glycaemic control, thereby behaves like compounds known as dietary fibers. Several factors are responsible for the resistant starch that makes them resist to digestion which includes the fragment size of starch, its conformation and structure of the starch granule. Resistant starch is commonly found in cereals, seeds, nuts and vegetables (Fuentes-Zaragoza et al., 2011). These resistant starches are used as encapsulating material for reduced gastric pH. The structural representation of resistant starch is presented in Fig. 7.5.

### 7.5.2.2 Cellulose

Cellulose, a polysaccharide consists of  $\leq 3000$  D-glucose units linked with  $\beta$ -1,4 linkage. Due to the lack of cellulase in the digestive tract, cellulose cannot be ingested in monogastric animals. It is one of the basic components of the cell walls of plant. Cells with higher cell content of cellulose in them possess stronger as well as thicker walls. Cellulose is basically straight chain polymer with no branching and coiling at all (Ummartyotin & Manuspiya, 2015). The structure of cellulose is presented in Fig. 7.6.

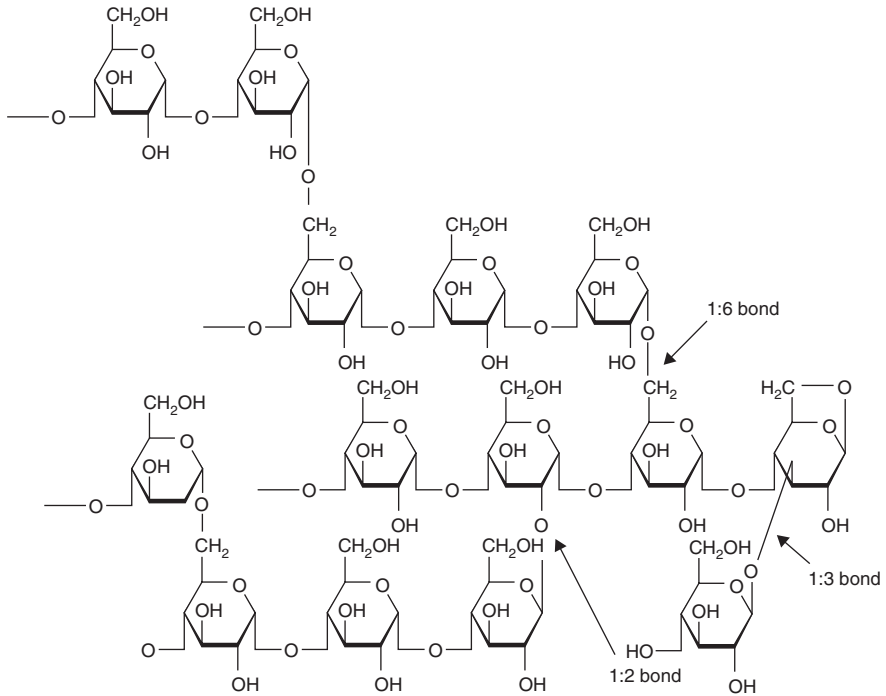
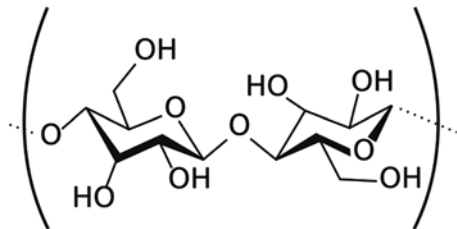


Fig. 7.5 Structure of starch (Slavin et al., 2009)

Fig. 7.6 Chemical structure of cellulose (Visanko et al., 2016)



## 7.6 Effect of Processing on Pre-biotic Potential of Cereals

### 7.6.1 Fermentation

Processing techniques such as flour pre-processing or wholemeal pre-processing generally improves the prebiotic potential of the cereal crops. It has been observed that certain techniques such as sprouting or pre fermentation also affect the prebiotic characteristics of cereal grains. Fermentation is one amongst the traditional technique used for preservation of food. However, fermentation nowadays is becoming the centre of attraction for the scientists as it has ability to enhance the functional as well as nutritional properties of the product, say, the products made from

sour dough after fermentation has positive effects on intestinal health via various mechanisms such as, it changes the fermentability and dietary fiber population, produces exopolysaccharides and other secondary metabolites which ultimately affects the bacteria present in gut. It is found that the fermentation of whole-wheat starter products will enhance the wheat bran arabinoxylan solubility. Hence, the fermentation of starter can affect the prebiotic properties of arabinoxylan by making it easier for the growth of beneficial intestinal bacteria. Using yeast or more specifically, pre-fermenting bran with yeast and lactic acid bacteria may be a simple tool to increase the prebiotic properties of cereal ingredients. The fermentation effect is believed to be caused by changes in enzyme activity and pH during the process, which will change the fermentation mode of dietary fiber. In a study, hydrolase from *Trichoderma* successfully converted the insoluble dietary fiber in durum wheat into soluble dietary fiber with prebiotic potential. In this study, the enzymatic treatment of insoluble dietary fiber resulted in the production of soluble dietary fiber, which supports the growth of lactobacilli and bifidobacteria in the intestinal model (Lappi et al., 2010; Napolitano et al., 2009; Noori et al., 2017).

### 7.6.2 Germination or Sprouting

Sprouting/ germination are another ancient traditional grain pretreatment technology, which can be regarded as a green tool to improve the health characteristics of grains. According to various studies, durum wheat varieties germinated in an *in vitro* digestion model significantly enhanced the prebiotic index. Though, this increase depends on the wheat genotype. The influence of germination on prebiotic index is related to alter of nutritional characteristics (Singh et al., 2015). In the study of wheat kernels, it was observed that during the prolonged germination period of 168 hours, total dietary fiber and soluble dietary fiber increased significantly while the content of insoluble dietary fiber was halved. Noori et al. (2017) determined the potential prebiotic activity of germinated rye in an *in vitro* study. Their results showed that the concentration of germinated rye extract was positively correlated with the vigor of *Lactobacillus acidophilus* and *Bifidobacterium animalis*.

### 7.6.3 Baking

Bread is a common way to consume cereal grains, however breads are primarily made from wheat. By utilising flours with higher amounts of dietary fibre or adding fibre-rich fractions, the amount of dietary fibre in bread can be increased. Endogenous enzymes prevalent in wheat have the ability to modify the  $\beta$ -glucan polymer structure dramatically (Andersson et al., 2004). Andersson et al. (2008) reported a steady decline in molecular weight of  $\beta$ -glucan as fermentation duration increased within the first several minutes. Endogenous enzymes were also important for enhancing  $\beta$ -glucan polymer extractability by degrading other cell wall polysaccharides in the

grain. When compared to native flour, short fermentation times doubled extractability, while extended fermentation times lowered extractability to values below natural extractability. The content of water-extractable  $\beta$ -glucans was unaffected by the level of endogenous enzyme activity. Finally, both the molecular weight and extractability of  $\beta$ -glucan were unaffected by oven baking. As a result, the impact of bread processing on  $\beta$ -glucan related viscous behaviour will be greatly influenced by the fermentation process. According to other study, some barley cultivars are more resistant to these fermentation-related alterations, which open up new possibilities for the production of  $\beta$ -glucan enriched breads (Djurle et al., 2018).

Tosh et al. (2008) reported on the self-aggregation of enzymatically formed  $\beta$ -glucan oligomers on muffin preparation with  $\beta$ -glucanase. Extractability of  $\beta$ -glucans was altered to variable degrees by partial depolymerization caused by enzyme addition: extractability increased with decreasing molecular weight and then fell following the self-assembly of gel networks in solutions caused by partially depolymerized  $\beta$ -glucans. At low molecular weight, the lowest extractability values were 17%, whereas at high molecular weight, they were 54%. The baking of treated muffins (30–60 minutes at 180 °C) had no effect on the  $\beta$ -glucans, as previously indicated for bread.

#### 7.6.4 *Cooking*

Cooking also affected the dietary fibre content of the cereal grains. Foods that have been demonstrated to have high dietary fibre include oat bran, baked porridge, muffins, and morning cereals. The cooking influence of the dietary fibre may also be modulated by the physical format of the ingredient. For example, Beer et al. (1997) found that heating oat bran into porridge had no effect on the extractability of  $\beta$ -glucans, but that cooking rolled oats had a modest increase in extractability.

According to Johansson et al. (2007), adding oat flakes to boiling water and simmering for 10 minutes enhanced the amount of soluble dietary fibre ( $\beta$ -glucans). Similarly, Åman et al. (2004) found that using a large bran particle size and a short fermentation time can reduce  $\beta$ -glucan breakdown during bread making. According to Beer et al. (1997), freezing reduced the extractability of  $\beta$ -glucans in muffins but had no effect on molecular weight. The extractability was lowered by 50% after 8 weeks of freezer storage. These alterations were attributable to molecular organisation during storage, which caused water to be repelled from the polysaccharide matrix, according to the authors.

#### 7.6.5 *Extrusion*

Cooking by extrusion is a popular method of processing dietary fibre rich foods, in addition to muffins and bread making. The high pressure and high shear that will be applied to the product distinguishes extrusion from typical porridge cooking or

dough baking. Depending on the settings used, extrusion can affect both molecular weight and extractability. Authors Gaosong and Vasanthan (2000) measured increased extractability that was both cultivar- and process-parameter-dependent when processing two different barley cultivars (waxy and regular) using twin-screw extrusion at different moisture levels (20–50% moisture) and temperatures (90, 100, 120, and 140 °C). At each extrusion temperature examined, solubility of the waxy material declined as moisture content increased. At each temperature level, the regular cultivar demonstrated higher extractability following extrusion and increased extractability with increasing moisture. Extrusion of the waxy cultivar revealed some fragmentation, which was particularly apparent around 120 °C. Extrusion seems to be a problem for the standard cultivar.

Zhang et al. (2011) used oat bran alone and found that extrusion parameters (feed moisture and temperature) had a substantial impact on the soluble dietary fibre content. On hulled barley husks, an improvement in extractability was also recorded (up to 8% at high temperature–low moisture extrusion conditions). As a result, traditional processing of -glucan-rich foods will always result in a change in dietary fibre structure, extractability (cell wall release or aggregation), or both. Even if some general trends emerge, these changes are difficult to forecast since they are highly process-dependent. Furthermore, the physical source of dietary fibre such as bran, flour, or rolled grain, as well as the type of the processed recipe, will influence the structural change (Sharma & Gujral, 2013).

## 7.7 Cereal-Based Probiotics Products

Probiotics are usually selective live microorganisms present in different foods that provide promising health benefits by stabilizing the gut micro flora upon consumption. They are well known for their potential to exhibit preventive properties against number of non-infective and infective disorders. They play important role in reducing the levels of cholesterol serum, improves lactose metabolism, protein digestibility, stimulates the immune system, absorption of calcium and possess various anti carcinogenic properties, anti-mutagenic properties, anti-diarrhoeal properties as well as aid in curing of bowel disease. Unfortunately, the majority of probiotic foods available in market such as cheese, beverages are usually milk based or confined to its fermented products, comprising of mainly the live microorganisms belonging to the family of lactic-acid bacteria (Enujiugha & Badejo, 2017). However, there are few beverages and probiotics containing products which are cereal based despite of the fact that cereals are widely distributed throughout and has rich nutritive value as well. But due to emerging demand of different tastes among the consumers and also because of vegan consumers, the researchers are now focussing towards development of cereal based probiotic products. Such products are not only boon for the vegan people but also for the people who suffers from allergies associated with milk or milk products, especially for the ones who are intolerant to lactose. Cereal based probiotic products have further advantages over

the milk based probiotic products in terms of longer shelf life and nutritional properties (Hassan et al., 2012). Various cereal based probiotic beverage products are enlisted in Table 7.1 along with their probiotic microorganisms. In addition, various labs are still developing more of the cereal based products to exploit the potential of

**Table 7.1** Cereal based probiotic beverages

Sr. No.	Product name	Substrate	Microorganism/specie	Protocol or conditions	Countries	References
1.	Akamu	Millet, Maize, Sorghum	<i>L. fermentum</i> , <i>L. plantarum</i> , <i>S. lactis</i>	Steeping of grains at room temperature for 2–4 days Souring Sieving Wet milling	Nigeria	Enujiugha and Badejo (2017)
2.	Pozol	Maize	<i>Lactobacillus</i> , <i>Leuconostoc</i> , <i>Lactococcus</i> , <i>Pediococcus</i>	Maize is cooked in 1% w/v of lime solution Washing is done using water Grinding is done to till dough is formed Balls off dough are made wrapping is done in banana leaves Fermentation for upto 4 days at ambient temperature.	Mexico	Prado et al. (2008)
3.	Gowe	Sorghum	<i>L. fermentum</i> , <i>P. acidilactici</i> , <i>Lb. mucosae</i> , <i>W. confusa</i>	Blending of non malted and malted Sorghum flour Fermentation to be done where the surrounding has moisture content of 52–87%.	Benin Republic	Vieira-Dalodé et al. (2008)
4.	Boza	Millet, Rye, Maize, wheat	<i>L. plantarum</i> , <i>L. brevis</i> , <i>Lb. acidophilus</i> , <i>L. mesenteroides</i> , <i>L. fermentum</i> , <i>L. ewffinolactis</i> , <i>L. coprophilus</i>	Cereals are boiled in water Cooling and sieving Addition of sugar	Bulgaria, Romania, Albania, Turkey	Blandino et al. (2003)

(continued)

**Table 7.1** (continued)

Sr. No.	Product name	Substrate	Microorganism/specie	Protocol or conditions	Countries	References
5.	Bushera	Millet, Sorghum	<i>L. lactis</i> , <i>Lb. brevis</i> , <i>L. mesenteroides</i> , <i>S. lactis</i>	Mixing of grains with boiling water Cooled to ambient temperature Germinated flour is added to the mixture Mixture is allowed to ferment for 1–6 days at room temperature.	Uganda	Muyanja et al. (2003)
6.	Symbiotic functional drink	Oats	<i>Lb. plantarum</i> B28	Fermentation was carried out for 8 hours with the help of sucrose at 37 °C.	Bulgaria	Angelov et al. (2006)
7.	Mahewu	Millet malt/ Sorghum, corn meal	<i>Lb. brevis</i> , <i>L. lactis</i> , <i>Lb. bulgaricus</i>	Maize porridge is mixed with water Addition of cereal flours Allowed to ferment at ambient temperature.	African and Arabian Gulf nations	Prado et al. (2008)
8.	Grainfields	Grains, seeds (maize, alfalfa, oats, rice, mung beans, barley, millets, rye, wheat)	<i>S. boulardii</i> , <i>Lb. acidophilus</i> , <i>Sc. Cerevisiae</i> , <i>Lb. delbreukii</i>	Natural organic fermentation	Various countries	Prado et al. (2008)

(continued)



**Table 7.1** (continued)

Sr. No.	Product name	Substrate	Microorganism/specie	Protocol or conditions	Countries	References
9.	Kunun zaki	Sorghum, Millet	<i>Lb. fermentum</i> , <i>L. lactis</i> , <i>Lb. plantarum</i>	Steeping of grains or millets Wet milling Addition of spices usually pepper or ginger Wet sieving Addition of sugar Fermentation is carried out for upto 48 hours.	Nigeria	Agarry et al. (2010)
10.	Koko	Millet	<i>Lb. paraplantarum</i> , <i>Lb. confusa</i> , <i>Lb. fermentum</i> , <i>P. pentosaceus</i>	Steeping of grains overnight Milling Addition of water and mixing Sieving Fermentation for 2–3 hours Addition of sugar	Kenya	Lei and Jakobsen (2004)
11.	Togwa	Finger millets, Maize	<i>Lb. fermentum</i> , <i>W. confusa</i> , <i>Lb. plantarum</i> , <i>Lb. cellobiosus</i> , <i>Lb. brevis</i>	Cooking of cereal flour in water Cooled at 35 °C Addition of culture of togwa along with cereal flour Fermentation is done till pH reaches at 4.0	Africa	Oi and Kitabatake (2003)
12.	Borde	Finger millets, Maize, tef, Barley	<i>Lb. fermentum</i> , <i>Lb. acidophilus</i>	25% unmalted and 75% malted cereals are allowed to ferment for 4 upto 4 days in four different phases.	Ethiopia	Abegaz et al. (2002)

(continued)

**Table 7.1** (continued)

Sr. No.	Product name	Substrate	Microorganism/specie	Protocol or conditions	Countries	References
13.	Mageu	Wheat, Maize	<i>Lb. fermentum</i> , <i>Lb. lactis</i>	Boiling of Maize in water Addition of wheat flour Allowed to ferment for 24 hours at 35 °C	South Africa	Enujiugha and Badejo (2017)
14.	Kwete	Millet, Maize	<i>L. lactis</i> , <i>Lb. plantarum</i> , <i>L. brevis</i>	Soaking of grains for upto 48 hours Allowed to germinate upto 3 days Subjected to sun drying for 1 to 2 days Souring is performed at 24 to 30 °C for 1 day. Allowed to ferment for 24 hours Filtration	Uganda	Muyanja & Namugumya (2009)

probiotics in the field of cereals, for instance, Kedia and others have made probiotic beverages based on malt; different oat based beverages are developed by Angelov and others in their experiment.

## 7.8 Other Products

Apart from cereal based probiotic beverages, several efforts have been made by researchers to impart the benefits of probiotics in traditional fermented foods so that it gets easily acceptable amongst the consumers, the examples of this includes the traditional food products such as Adai made of legumes and cereals by using lactic acid bacteria (LAB); Atole, made from maize with LAB as the probiotic microorganisms; Ben saalga, made of pearl millet using LAB, etc. The common traditional foods which are popularly consumed as breakfast or dinner in Southern India and in other countries like Sri Lanka i.e. as Dosa, Idli, are made up of Bengal gram, rice, legume or cereals can be inoculated with different probiotic microorganisms such as *L. mesenteroides*, LAB, *Lb. fermentum*, *S. cerevisiae* (Agrawal et al., 2000; Aidoo et al., 2006; Farnworth, 2005; Tou et al., 2006). Other traditional fermented foods

include Tempeh, Uji, Saurkraut, Ilambazi lokubilisa, etc. which can be turned into probiotic foods after incorporation of live probiotic microorganisms in them. However there are several probiotic products that are not dairy based but are fruits or vegetable based, soy based or cereal based and are available in market as well. These include puddings based on cereals (Helland et al., 2004), Yosa or pudding based on oat bran (Blandino et al., 2003), rice based yogurt (Wongkhalaung & Boonyaratanakornkit, 2000).

Several probiotic products are produced globally based on the cereal matrices. These includes cereal bars which are available by different commercial names viz. Effi foods probiotic care bars, vega one, Good! Greens bars, all in one Meal bars. These are originated in United States of America (USA) by using single probiotic microbe i.e. *B. coagulans*. PROBAR meal bars is another example of baking mix made by using culture of *B. coagulans*. Other products such as Yog active probiotic cereals (origin- Germany, probiotic microbe- *Lb. acidophilus*), probiotic granola bars (commercial name- Udi's Gluten free, origin- USA, organism- *B. coagulans*), oatmeal bar (commercial name- Pop culture probiotic, origin- California, organism- *B. coagulans*), cereal bar (commercial name- Welo Probiotic bar, origin- Canada, organism- *B. coagulans*), Poppers (commercial name- Brad's Broccoli peppers, origin- Pipersville, organism- *B. coagulans*), Muesli (commercial name- Nutrus Slim Muesli, origin- India, organism- *B. coagulans*), Muesli (commercial name- Something to crow about- probiotic Muesli, origin- New Zealand, organism- *B. coagulans*), Burritos (commercial name- Probiotic Burritos, origin- USA, organism- *B. coagulans*) and many more (Dey, 2018).

## 7.9 Health Benefits of Pre-biotic and Probiotic

Probiotics serves both, the health as well as the nutritional benefits upon their consumption. Enzymatic hydrolysis of bacteria escalates the protein bioavailability, which in turn is beneficial for the consumer if he or she is suffering from the deficiency of production of endogenous protease. This happens because of increased free amino acid production due to enzymatic hydrolysis by bacteria as discussed earlier. Various strains of lactic acid bacteria are also responsible for increasing the content of vitamins, especially B-complex in various fermented foods.

Probiotics exhibit the beneficial effects when consumed in sufficient quantity or concentration, say, as lower as  $10^8$  colony forming unit per gram of the product and this must will be capable of surviving the harsh conditions present in human stomach such as, low pH of the gastric juice and then it reaches the small intestine and last to the colons. The best effect of probiotics is believed to be achieved when microorganisms colonize themselves in intestinal epithelium. At this time, they are capable of affecting the immune system of intestine by providing antioxidants and anti-mutagens, and also by displacing the enteric pathogens.

Probiotics also posses anti microbial properties which might be attributed to the production of number of inhibitory compounds such as bacteriocins,

hydrogen peroxide and organic acids, and to its competition towards the nutrients. The organic acids such as acetic acids and lactic acid produced by the bacteria are responsible for lowering the pH and thereby become responsible for exhibiting the bacteriostatic effect or sometimes bactericidal effect also. However, the role of probiotic strain in the inhibition of pathogen is limited as the bacteriocins that are present since ages, do possess the inhibitory action against the closely related species like that of *Clostridium* (spore-forming), *Bacillus* (spore-forming) and other species of *Lactobacillus*. The other metabolites especially, the low molecular weight ones which includes acetic acid, hydrogen peroxide, various aroma compounds and lactic acid along with different secondary metabolites are considered to be the most important since they exhibit wide range of inhibitory spectrum against number of harmful organisms such as *Chlostridium*, *E. coli*, *Salmonella*, *Helicobacter*, etc. Also, the lactic acid bacteria from *Lactococcus* genera, *Lactobacillus* genera and *Pediococcus* genera are responsible for producing the di-acetyl that is rarely found in the food fermentations is one of the major contributor of anti-bacterial activity (Enujiugh & Badejo, 2017).

## **7.10 Effect of Prebiotic Dietary Fibers on Health**

### ***7.10.1 Effect on Composition of Hind Gut Bacteria***

The genera of these bacteria are the well-known common markers for the health of microbiota that are responsible for targeting the dietary stimulation. *Lactobacillus* has been shown to down-regulate the inflammation of the gastrointestinal mucosa. *Lactobacillus* plays a role in helping patients with lactose intolerance to digest the lactose, relieve constipation, improve symptoms of IBS (irritable bowel syndrome), and may help to prevent diarrhea in case of travellers. *Bifidobacterium* naturally exists in gastrointestinal tract of humans and possess strong affinity for fermenting certain oligosaccharides, thereby establishing them as common marker to have prebiotic ability. It has been observed that *Bifidobacterium* does not produce any carcinogens in the body. The level of bifidobacteria is negatively correlated with weight gain and obesity. The decrease of bifidobacteria and the decrease of bacterial diversity are related to the increase of IBS and inflammation. Though, various studies reveal the beneficial nature of these bacteria towards health, however, the mechanism of the same is not yet clear (Carlson et al., 2018).

### ***7.10.2 Effects on the Absorption of Mineral***

Prebiotics are well known in improving and maintaining the bone structure of elder people and adolescents in the era where the bone fracture and osteoporosis is a common problem. Intake of prebiotics usually increases the calcium absorption along

with its bioavailability, thereby reduces the risk of osteoporosis. Distal intestine, the primary site for the absorption of calcium, which is escalated by various chemical changes and increased acid-fermentation of prebiotic dietary fibers with the help of various bacteria.

### ***7.10.3 Effects on Production of Metabolites***

Various metabolites, primary as well as secondary metabolites are formed by indirect or direct fermentation of various selective compounds that are correlated with number of beneficial effects on human health. Various SCFA (short chain fatty acids) are produced upon fermentation of amino acids, nutrients, carbohydrates and other different compounds by gut microbiota, which is usually absorbed in the small intestine. These include butyrate, acetate and propionate, which contribute up to 95% of the SCFA that are produced in colon. SCFAs are known to possess number of positive outcomes in humans. It has been observed that inulin type fructans when fermented, increases the levels of hippurate, which is microbial a co-metabolite, usually present in higher concentration in lean people as compared to the obese ones. Its increased levels are known to possess beneficial effect on humans upon fermentation of inulin after consumption.

### ***7.10.4 Protein Fermentation***

Protein fermentation from endogenous or undigested protein sources occurs in the absence of fermentable carbohydrates, can lead to the accumulation and formation of harmful metabolites, such as ammonia, sulphides, phenols and amines. Also, at the same time, the concentration of SCFAs decreases and the pH value of the environment increases, thereby providing a favorable environment (distal colon) for protein fermentation to occur efficiently. This in turn leads to the production of branched-chain fatty acids and various indoles and phenols, which is unique to the bacterial metabolism.

### ***7.10.5 Effects on Risks Associated with Allergy***

Microbial diversity of gut is responsible for development of inflammatory conditions such as diseases related to allergy. Allergies during the first 5 years of life are generally associated with the decreased level of Lactobacilli and bifidobacteria. Dietary oligosaccharides have immuno-modulating effects against development of rhinoconjunctivitis and eczema. They also possess various allergy protective effects.

## 7.11 Future Perspectives

Cereals are one amongst the most suitable substrate that can be used for growing the probiotic strains, which are basically human derived. Irrespective of the number of differences in the performances in between the specie, complexity of substrates, a stable systematic approach is to be implemented which is capable of enhancing the survival and the growth of the probiotic strain. Also, the functionality can be improved by addition of various non digestible components of cereal based matrix which can act as prebiotics. Novel functional ingredients are advantageous for the manufacturing companies as they can add more of value to the products. The development of such novel products requires campaigns so as to make consumers aware of the products and its benefits. Also, consumer needs some additional adaptation time to accept the new product. Therefore, the approach of making fortifying the traditional cereal based food products are of more advantageous as these are easily acceptable among the consumers. Cereals are capable enough to deliver the probiotic lactic acid bacteria to human guts and can act as substrate for the growth of bacteria as well.

## 7.12 Summary

Cereal-based foods have been consumed since ancient times, and their popularity continues to grow due to their consumer acceptance and health benefits. However, due to advancements in food technology and interest in the production of novel goods with improved health qualities, their design has evolved. Probiotics, prebiotics, and synbiotics have all had a lot of success in the field of functional foods over the previous two decades. Various cereal-based products can now be formulated with synbiotic features that include additional health-improving molecules without altering their physicochemical, rheological, or sensory properties. Food businesses can add additional value to items that consumers are already familiar with by developing new functional ingredients. New food development necessitates greater marketing campaigns, and consumers may require time to adjust to the new product. Nutritional food components enable producers to meet and exceed the demands of today's health-conscious consumer by either inventing new and inventive products or simply reformulating existing ones. Cereals contain possibly pre-biotic compounds whose activity as well as the ability to develop and transfer probiotic lactic acid bacteria to the human gut should be investigated.

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