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APPLICATIONS OF PROBIOTICS AND PREBIOTICS IN THE FOOD INDUSTRY: A REVIEW

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ABSTRACT

In the fascinating world of gut health, two terms often take the spotlight: probiotics and prebiotics. Probiotics and prebiotics have emerged as powerful tools in promoting gut health and overall well-being. Probiotics are live microorganisms that, when consumed in adequate amounts, confer health benefits to the host. They can be found in certain foods or taken as supplements. Probiotics have emerged as medical therapies for gastrointestinal and non-gastrointestinal diseases such as diarrhoea, constipation inflammatory bowel disease, irritable bowel syndrome urinary tract infections etc. They can also be incorporated into functional foods and beverages, such as probiotic-infused juices and smoothies, to promote a healthy gut microbiome. The global market of functional foods including probiotics is growing exponentially. Probiotics and prebiotics have received escalating attention in recent years in the scientific, healthcare, and public arenas. Prebiotics, on the other hand, are non-digestible substances that serve as food for beneficial bacteria in the gut. By nourishing these beneficial bacteria, prebiotics help support a healthy gut microbiome. Together, probiotics and prebiotics offer a holistic approach to improving digestion, boosting the immune system, and maintaining optimal gut health. In recent years, their applications have expanded across the food industry, with various products incorporating these ingredients to provide consumers with functional foods that promote a balanced and thriving gut microbiota. Prebiotics mostly consist of oligosaccharides but sometimes they also contain polysaccharides (e.g. inulin). Inulin and FOS (Fructo-oligosaccharide) are types of prebiotics which shown a great observability in food industry by replacing fat and sugar content in ice-creams resulting better quality of product. Since prebiotics are used extensively in various food applications there is scope for novel discoveries. Hence, the future of pre and probiotics in the food industry is bright, driven by the increasing recognition of their health benefits and the growing consumer demand for functional foods.
Keywords **:** Probiotics, prebiotics, lactic acid bacteria (LAB), oligosaccharides, food industry

Introduction

Probiotics are defined as 'live microorganisms which when administered in adequate amount confer health benefits to the host (FAO/WHO, 2002). The word "Probiotics" is derived from Latin (pro) and Greek (biotic) roots meaning "for life". Probiotics are defined as live bacteria with beneficial effects to the health of the host organism. Probiotics term coined in 1965 by Lilly and Stillwell.The concept of probiotics was first introduced in the 20th century by noble prize winner, Elie Metchnikoff (1845-1916).

Today, Lactobacillus and Bifidobacterium became the main probiotic groups, but the probiotic potential of Pediococcus, Lactococcus, Bacillus and yeasts have also been demonstrated (Soccol *et al*., 2010). In most cases, probiotics are produced directly by fermentation in foods such as yogurt, or are supplied through dietary supplements.

- Following are the requirements which have been identified for a microorganism to be defined as an effective probiotic:
- The probiotic should give positive effects on GIT of the host. It should be acid resistant and bile resistant.
- The adhesive capability of probiotics must be firm and faster.
- The probiotic should possess high survival rate & multiply faster.
- Exclude or reduce pathogenic adherence.
- Safe, non-invasive, non-carcinogenic and non-pathogenic to the host Produce acids, peroxide and bacteriocins, antagonistic for the growth of pathogens.
- Coaggregate to form a normal balanced flora.
- Probiotics should be durable enough to withstand the duress of commercial manufacturing, processing, packing and distribution so it can be delivered alive to the intestine (Behnsen, 2013).

Mechanism of action of probiotics : Probiotic bacteria have various effects on the host cells for example the prevention and treatment of a wide range of disorders. The main mode of probiotic actions Include: 1) enhancement of the epithelial barrier 2) increased adhesion to intestinal mucosa and simultaneous inhibition of pathogen adhesion 3) production ofanti-microbial substances 4) competitive exclusion of

pathogenic microorganisms 5) modulation of the immune system. Recent data exhibit the effect of probiotics in interference with quorum sensing.

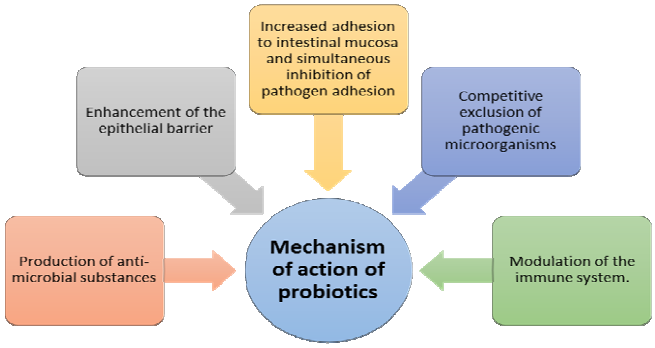

Fig. 1: Mechanism of action of probiotics

Enhancement of the Epithelial Barrier: The intestinal barrier is a major defense mechanism used to maintain epithelial integrity and to protect the organism from the environment. Defenses of the intestinal barrier consist of the antimicrobial peptides mucous layer, secretory IgA and the epithelial cells that form tight junctions (Ohland, 2010).

Increased Adhesion to Intestinal: Probiotic bacteria are able to adhere to epithelial cells, thereby, can block adherence of pathogens. The anti-adhesive effect may result of competition between probiotic strains and pathogens for the same receptor or the induction of mucin production by probiotics which is a complex glycoprotein mixture that is the component of mucous, thereby preventing the adhesion of pathogenic bacteria.

Competitive exclusion of pathogenic microorganisms: Competition for space to adhere between indigenous bacteria and exogenous pathogens result in the competitive exclusion of pathogenic bacteria. Exclusion is the result of different mechanisms and properties of probiotics to inhibit pathogen adhesion, including physical blocking of pathogenic bacteria colonization by probiotic bacteria from their favourite site such as intestinal villus, goblet cells and colonic crypts. First, the probiotic bacteria compete with pathogenic bacteria for nutrients and energy source thus, preventing them from acquiring energy required for growth and proliferation of pathogenic bacteria in the gut environment. Second, probiotics produce several organic acid and volatile fatty acids (VFA) as a result of their metabolism and fermentation. Consequently, the pH of the gut is lowered below that essential for survival of pathogenic bacteria such as *E. coli* and *Salmonella*.

Production of anti-microbial substances: Antimicrobial substances produced by probiotics can lead to inhibition of pathogen replication. These components are almost always low-molecular-weight (LMW) compounds such as short chain fatty acids (SCFA), antimicrobial peptides (AMPs) organic acids, and DE conjugated bile acids. Importantly, these LMW compounds are short chain fatty acids. Short chain fatty acids (SCFA), which include acetate, propionate and butyrate are produced by bacteria in the gut during fermentation of insoluble fibre from dietary plant matter.

Probiotics and the immune system: Probiotic bacteria can affect on numerous cell types involved in the innate and adaptive immune responses such as epithelial cells, monocytes/macrophages, dendritic cells, B cells, T cells, regulatory T cells and NK cells epithelial cells, dendritic cells, monocytes/macrophages and thereby exert their immunomodulatory effect. Probiotic bacteria can reduce Th1 response and suppress the production of pro-inflammatory cytokines, IL-12,TNF-α, andIFN-γ by dendritic cells (DC).

Health Benefits:

Probiotic impacts are strain particular the impacts depicted for one strain can't be specifically applied to others and every individual Probiotic bacterial strain has its own health benefits. The major beneficial effects are correlated against various disease conditions. Probiotics have a colossal criticalness and application in controlling different kinds of microbial infections. Nowadays such a benefit is usually interpreted as an increase in lactobacilli and/or bifidobacteria and a decrease in potentially pathogenic bacteria (De *et al*., 2006).

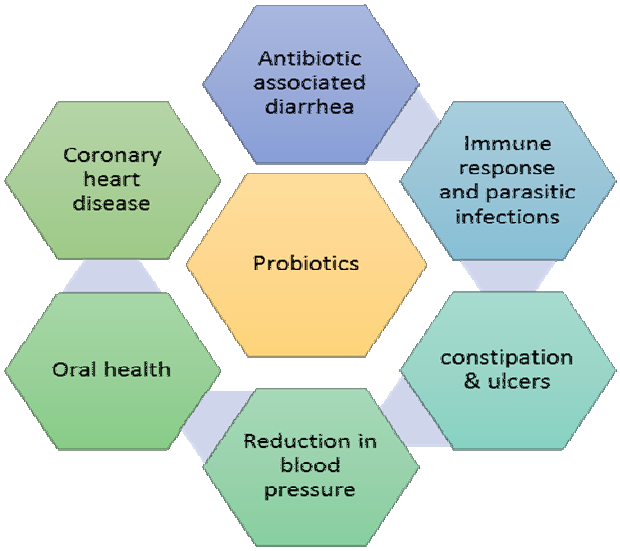

Fig. 2: Health Benefits of Probiotics

Types of Probiotic Bacteria:

Though there are many types of bacteria that can be considered probiotics, there are two specific types of bacteria that are common probiotics found in stores. These include:

- *Lactobacillus.*
- *Bifidobacteria.*
- *Streptococcus*
- *Enterococcus*
- *Leuconostoc*

Among all the strains Lactic acid bacteria (LAB) and *Bifidobacterium.* are the most significantly used bacteria in food industry particularly dairy industry. Other microorganisms such as yeast *Saccharomyces cerevisiae* and some *Escherichia coli* and *Bacillus* species are also used as probiotics. Lactic acid bacteria (LAB) which have been used for food fermentation since the ancient time, can serve a dual function by acting as food fermenting agent and potentially health benefits provider. LAB are GRAS (generalrecognized as safe) with no pathogenic, or virulence properties have been reported. For theuse of LAB as probiotics, some desirable characteristics such as low cost, maintaining its viability during the processing and storage, facility of the application in the products,resistance to the physicochemical processing must be considered.

Sources of probiotics : The most common source of probiotics is Yogurt. Yogurt consists of milk (usually from the cow, goat or sheep) fermented by bacteria that modify lactose into lactic acid. Lactic acid is responsible for giving yogurt its characteristics (sharp taste usually changed into good taste by using sweeteners and flavoring) and also denatures and precipitates casein, resulting in a semisolid consistency. ''Bioyoghurts'' are produced in a similar way, but bacteria used for fermentation are of different strains, usually *L. acidophilus*. Fermented milk and fortified fruit juice are common sources of probiotics (Iqbal, 2014).

Applications of probiotics in food:

1. **Potential use of bacillus coagulans in the food industry:** *Bacillus coagulans* (*B. coagulans*) was firstly isolated from spoiled milk. *B. coagulans* is a gram-positive, facultative anaerobic, non-pathogenic, spore-forming, lactic acid-producing bacteria. It is resistant to heat; the optimum growth temperature for *B. coagulants* is 35 to 50 °C and the optimum growth pH is 5.5 to 6.5. Although *B. coagulans* produces acid, it does not produce gas from maltose, raffinose, mannitol, and sucrose fermentation. It was reported that *B. coagulans* causes deterioration in dairy, fruit, and vegetable products due to acid production. In addition to lactic acid production, some strains also produce thermostable -amylase. For this reason, *B. coagulants* is important from an industrial point of view.
2. **Enzymes production from probiotics yeast that are added as food additives:** Among food components, enzymes serve as living catalysts for chemical reactions, which can be beneficial or detrimental during food processing. Some yeast enzymes have been introduced into the food industries. Although yeasts, in general, do not have the GRAS status but *S. cerevisiae* cells do (Verstrepen *et al*., 2006). Industrial enzymes are produced as heterologous proteins by recombinant methods. Certain yeasts have been developed to produce heterologous proteins including *Pseudozyma* spp. and other yeast.
3. **Probiotics in Dairy-based foods:** Milk and its products is good vehicle of probiotic strains due to its inherent properties and due to the fact that most milk and milk products are stored at refrigerated temperatures. Probiotics can be found in a wide variety of commercial dairy products including sour and fresh milk, yogurt, cheese, etc. Dairy products play important role in delivering probiotic bacteria to human, as these products provide a suitable environment for probiotic bacteria that support their growth and viability (Phillips *et al*., 2006).
4. **Probiotics in Cereal-based products:** Cereal-based probiotic products have health-benefiting microbes and potentially prebiotic fibres. Cerealsare good substrates for the growth of probiotic strains and due to the presence of non-digestible components of the cereal matrix may also serve as prebiotics (Salovaara *et al*., 2011). Champagne has listed number of cereal-based products that require a lactic fermentation, often in association with yeast or molds.
5. **Probiotics in fruits and vegetables:** Foods based on fruit and vegetables, such as fruit and vegetable juices, represent a new potential carrier and source of probiotic microorganism. LAB isolated from the same plant can be used as probiotics fruits and vegetables represent health promoting foods. Strains of LB have been use in this regard for the production of a beverage made of single and mixed fermented cereals. The microorganisms survived producing large amount of lactic acid and organic acids that shows the enrichment of food with probiotics but can also have negative effects on the sensory characteristics.
6. **Probiotics use as bio preservatives:** Use of microorganisms and their natural by-products for bio-preservation has been commonly practiced throughout the history of mankind, Lactic acid bacteria (LAB) are known to produce many antimicrobial compounds like organic acids, anti-fungal peptides, hydrogen peroxide and bacteriocins. They are being used in many dairy milk products. They are non-pathogenic, bile tolerant and salt tolerant. Bio-preservation adds to the extension of shelf life and improvement of food quality Using microbes or their metabolites. Bio-preservation is mainly used to preserve the spoilage of meat and meat products and they get higher chances of contamination.

Prebiotics :

A prebiotic was first defined as 'a non-digestible food ingredient that beneficially affects the host by selectively stimulating the growth and/or the activity of limited number of bacteria in the colon, and thus improves host health' (Gibson and Roberfroid 1995).

Table 1 : Different types of Prebiotics and their sources.

TYPE OF PREBIOTIC	FOOD SOURCES
Fructans (Inulin, Fructooligosaccharides)*	Chicory, artichoke, garlic, onions, leek, asparagus, banana
Galactooligosaccharides *	Chickpeas, beans, lentils, broccoli, Brussel sprouts
Resistant Starch*	Green bananas, plantains, cooked and chilled potatoes and rice, cold oatmeal, beans, peas, lentils and seeds
ß-Glucan*	Oatmeal, barley, mushrooms, yeasts, seaweed
Polyphenols	Many different fruits, particularly berries, tea (black, green or herbal), dark chocolate, red wine, globe artichokes, chicory, red onion, spinach, broccoli, herbs, spices
Omega-3 fatty acids (EPA and DHA)	Fatty fish such as salmon, trout, sardines, herring, tuna

Characteristics of ideal prebiotics :

- Selectively enrich for or a limited number of beneficial bacteria.
- Neither hydrolyzed nor absorbed by mammalian enzymes or tissues.
- Beneficially alter the intestinal micro flora and their activities.
- Beneficially alter luminal or systemic aspects of the host defense system.

Types of Prebiotics :

(a) Simple-Galacto-oligosaccharides (GOS), Fructo-oligosaccharides (FOS), Isomalto-oligosaccharides(IOS) and inulin.

(b) Complex- Human milk oligosaccharides, Arabinoxylan, Resistant starch Oligosaccharide is the main constituent of prebiotic food products. It stimulate the growth of

beneficial bacteria and increase the resistance to invading pathogens.

Mechanism of action of prebiotics: Prebiotics mechanism involves various steps like 1) Modulation of the Gut Microbiota 2) Immune System 3) Anti-pathogenic activity 4)Mineral Absorption.

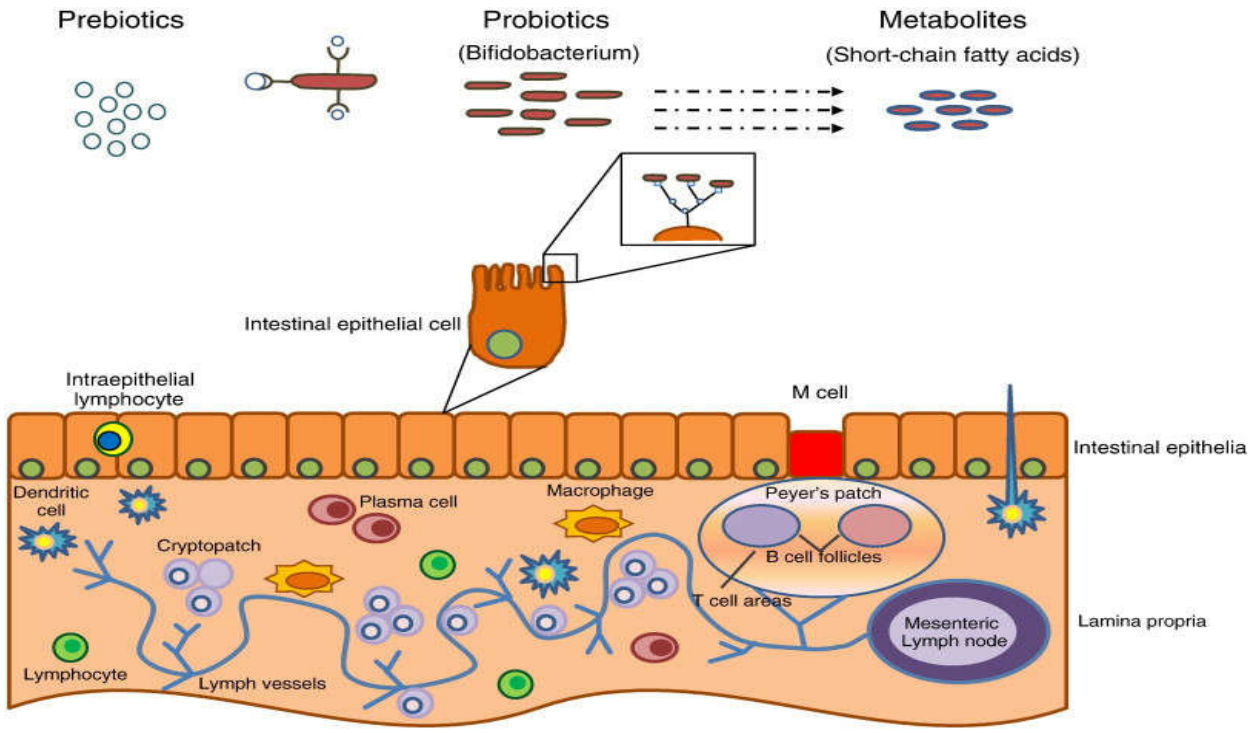

Fig. 3: Mechanism of action of prebiotics

Modulation of the Gut Microbiota: Evidence from human feeding trials has shown that prebiotics affect the composition of the gut microbiota, leading to an increase in health promoting organisms such as bifidobacteria and lactobacilli. These bacteria are generally safe because they mainly ferment carbohydrates, are not pathogenic and are non-toxigenic, while they have a role in colonization resistance and frequently manifest immuno modulatory properties in the host. Some species are also able to ferment prebiotics to SCFA such as acetate and butyrate, which are important sources of energy for the host. While bifidobacteria do not produce butyrate, they have been shown to stimulate butyrate producing bacterial species such as eubacteria in the gut.

Immune System: Evidence suggests that prebiotics can have significant effects on the immune system. unknown if these are direct or indirect effects resulting from stimulation by immunomodulatory bacteria, or production of SCFA, which are known to have immunomodulatory properties, and can bind to SCFA G protein coupled receptors on immune cells within gut-associated lymphoid tissues (GALT). Addition of FOS and lactulose to the diet has been shown to increase mucosal immunoglobulin production, mesenteric lymph nodes, Peyer's patches and altered cytokine formation in the spleenand intestinal mucosa.

Anti-pathogenic activity: Prebiotics in diet protect the GIT from infection and inflammation by inhibiting attachment and/or invasion of pathogenic bacteria or their toxins to colonic epithelium in various studies. This attachment is mediated by glycol conjugation glycoproteins and lipids present on the microvillus membrane. Prebiotic especially GOS contain structures similar to those found on microvillusmembrane that interfere with the bacterial receptor by binding them and thus prevent bacterial attachment to colonic epithelium.

Mineral Absorption: The ability to improve calcium, magnesium, iron and zinc absorption, and the attendant enhancement of bone mineralization is the most significant health effects of prebiotics on mammalian physiology and the attendant enhancement of bone mineralization. Several mechanisms have been proposed for prebiotic action in mineral absorption.

Functional oligosaccharides contain:

I. **Fructo-oligosaccharides (FOS):** It is a group of oligosaccharides or a group of connected simple sugars. It helps in stimulate production of beneficial bacteria, minimal effect on blood sugar, also help strengthen the immune system. Sources- onions, garlic and chicory roots. FOS is also linked with improving stool frequency and consistency in adults (Yu *et al*., 2017). Fibre isolated from orange seed, pulp and peel was used as a fat replacer in ice cream. The fibre being rich in soluble fiber, insoluble fibre and total phenolics allowed for 70% fat reduction in the product (de Moraes Crizel *et al*., 2013).

II. **Inulin:** It is a dietary fibre that is found in many common foods such as leeks, kiwi, asparagus, onions, garlic, bananas, wheat, rhubarb , dairy products etc. There are several benefits of inulin apart from weight management, it even increases calcium absorption, improved bone health. It can also be use to replace sugar and fat as a food additive to improved taste. The use of inulin as a fat replacer in ice-cream at increasing

levels of 5%, 7% and 9% resulted in enhanced consistency and stickiness. The sensory profile was smooth because of less grittiness and hardness (El-Nagar *et al*., 2002). However, the use of inulin in full fat ice-cream as a sugar replacer at 10% and 30% sucrose replacement levels resulted in high consistency coefficient, pseudoplasticity and apparent viscosity.

III. **Soybean meal oligosaccharides(SMO):**They are found in soy milk, soy oil, bean curd, ice cream. Promote growth of Bifidobacteria in the intestinal tract lowering cholesterol.

IV. **Galacto-oligosaccharides:** Galacto-oligosaccharides (GOS) are found naturally in breast milk but are added to foods such as infant formula, fruit drinks, dairy products, breakfast cereals and biscuit crackers. Increased the good bacteria boosts the immune system. Promote intestinal health by keeping unfriendly bacteria such as *E.coli*, vaginal, urinary infections at bay. The high solubility of GOS increases its use in dairy products, e.g. dried buttermilk (Curda *et al*., 2006). One of the major applications is in infant nutrition where a substitute of breast milk has to stimulate the bifidobacteria and lactobacilli.

V. **Xylo-oligosaccharides (XOS):** They are naturally present in fruits, vegetables, bamboo, milk and honey. It helps to improve blood sugar levels and fat absorption, re-establish normal colonic flora, it also increase mineral absorption and vitamin B creation.

Applications of prebiotic in food industry:

Table 2 : Applications of prebiotic in food industry

Applications	Functional properties
Dairy products	Fat or sugars replacement, texture and mouth feel, fiber and prebiotic
Frozen desserts	Fat or sugars replacement, texture and mouth feel, melting behaviour
Fruit preparations	Sugar replacement synergy with intense sweeteners, body and mouth feel, fiber
Beverages and drinks	Fat or sugar replacement, mouth feel, foam stabilization and prebiotics
Baked goods and breads	Sugar replacement, moisture retention, fiber and prebiotics
Breakfast cereals and extruded snacks	Sugar replacement, crispiness and expansion, fiber and prebiotics
Filling	Fat or sugar replacement, texture and mouth feel.
Dietetic products	Fat or sugar replacement, fiber and prebiotic
Sugar confectionary	Sugar replacement, fiber and prebiotics
Chocolate	Sugar replacement, heat resistance and fiber
Soups and sauces	Sugar replacement and prebiotics
Meat products	Fat replacement, texture stability and fiber

Source: Wang, (2009)

1. **Prebiotics in cheese:** Addition of prebiotics to a probiotic cheese (made using starter and probiotics) fastest fermentation and high lactic acid production. Addition of probiotic compound in probiotic cheese is advantageous so far as functional CLA compound are concerned plus an enhanced nutritional quality. Cheese made from the combination of different prebiotics and probiotics shown potential functional petit-Suisse cheese.
2. **Prebiotics in dairy fruit beverages:** Probiotic oat bran is added to a dairy fruit beverage was well accepted by consumers.
3. **Prebiotics in processed fruits and vegetables:** Prebiotics have been incorporated in edible coatings of apples which reduced the loss of bioactive and volatile compounds from apples. The alginate coating which was a carrier for FOS and inulin enhanced the nutritional benefits and shelf life of freshly cut apple wedges (Robsle *et al*., 2010).
4. **Prebiotics in bakery products:** In bakery products, prebiotics can act as fat and sugar replacer. The effects observed on addition of prebiotics like inulin in various bakery products such as bread, biscuits and cakes. The addition of beta-glucan (a prebiotic with high-affinity for water) in bread resulted in underdevelopment of gluten network and less steam production during baking (Gill *et al*., 2002).
5. **Prebiotics in fermented milk:** The total solid content of the prebiotic fermented milk is higher within the addition of insulin, also increased its acidity. The probiotic fermented milk has greater thixotropy and the hysteresis got lowered with an increase in the temperature. The inulin addition in a product resulted with a greater tendency toward a greenish coloration.
6. **Prebiotics in meat industry:** Dietary fibers from oats, soy, sugar beet, fruits such as apple were incorporated in meat sausages and patties. Similarly, inulin was used in sausages and cooked ham (Jime´nez-Colmenero *et al*., 2001).
7. **Prebiotics in cereals:**Prebiotic activity of cereal derived arabinoxylan oligosaccharides (AXOS). Prebiotic gluten free breads have softness, traditional bread aroma, sweetness and crumb color are the most desired sensory properties. Biscuits added with FOS have a significant increase in faecal bifidobacterial numbers. FOS addition to cereals was tolerated up to doses as high as 3.00 g FOS/day (Moore *et al*., 2003).

Conclusion

Probiotics and prebiotics have a noteworthy effect on human health and have greater potential for incorporation into a broader range of common foodstuffs as they have both technical as well as nutritional properties.The integration of pre and probiotics into the food industry also aligns with the growing consumer interest in health and wellness. Consumers are increasingly seeking products that promote

digestive health, enhance immunity, and contribute to overall well-being. This shift in consumer preferences presents a vast market potential for innovative pre and probiotic offerings in order to revolutionize the food industry and pave the way for a healthier and more personalized approach to nutrition.

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