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Probiotics: Foods and health friendly microbes

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Abstract
Probiotics are live, beneficial microorganisms that offer a wide range of health-promoting effects to the host, such as inhibiting the adhesion and colonization of pathogenic bacteria, enhancing immune responses, neutralizing toxins, alleviating inflammatory bowel conditions, lowering cholesterol levels, contributing to cancer prevention, synthesizing essential vitamins, and producing antimicrobial compounds. Probiotic-enriched food products extend shelf life and improve resilience under adverse environmental conditions. This has led to growing interest in recognizing the vital role of specific microbial strains residing in the intestinal ecosystem. These strains are now widely regarded as probiotics—used both as biotherapeutic agents and as components of functional foods or nutraceutical supplements. This review aims to provide a comprehensive overview of the health benefits of probiotics and their role in disease prevention.

Keywords: Probiotics, prebiotics, synbiotics, food and health benefits

Introduction

History of Probiotics

Elie Metchnikoff, a Russian scientist, Nobel laureate, and professor at the Pasteur Institute in Paris, proposed more than a century ago that lactic acid bacteria (LAB) provided health advantages that could extend life. He proposed that altering the gut microbiota and substituting saccharolytic microorganisms for proteolytic microbes, which generate harmful compounds like phenols, indoles, and ammonia from the digestion of proteins could inhibit "intestinal auto-intoxication" and the ageing that follows. He created a diet consisting of milk fermented by the "*Bulgarian bacillus*" bacteria. Disorders of the intestinal tract were frequently treated with viable nonpathogenic bacteria to change or replace the intestinal microbiota. In 1917, before Sir Alexander Fleming's discovery of penicillin, the German professor Alfred Nissle isolated a nonpathogenic strain of *Escherichia coli* from the feces of a First World War soldier who did not develop enterocolitis during a severe outbreak of shigellosis. The resulting *Escherichia coli* strain Nissle 1917 is an example of a non-LAB probiotic. In order to treat infants with diarrhoea, Henry Tissier (of the Pasteur Institute) isolated a *Bifidobacterium* from a breastfed baby. According to his theory, it would displace the diarrhea-causing proteolytic bacteria. To combat diarrhoeal epidemics, Dr. Minoru Shirota discovered the *Lacticaseibacillus paracasei* strain Shirota in Japan. Since 1935, a probiotic product containing this strain has been sold commercially.

Introduction of Probiotics

Probiotic is a Greek word meaning "for life," coined by Lilley and Stillwell. Probiotics refer to microbes of non-pathogenic nature that are beneficial to their hosts (Soccol *et al.,* 2014) [85]. Probiotics have been in use for quite a long time as Romans and Greeks, the ancient civilizations developed fermented milk and used it as probiotics, even the bible mentions this sour milk so the concept of probiotics is not entirely new (Hosono, 1992) [41]. Probiotics improve the microbial balance of the Gastrointestinal (GI) tract. World Health Organization (WHO) defines probiotics as "Live microbes which confer a health benefit to their host when administered in adequate amounts" (Chen and Sears, 2015) [22]. *Lactobacillus, Bifidobacterium, Enterococcus, Lactococcus*, and *Streptococcus* are most commonly used as probiotics (de Sequeira *et al.,* 2022) [30]. Probiotics exert their beneficial effects through several key mechanisms, including enhancement of the epithelial barrier, promotion of microbial adherence to the intestinal mucosa, inhibition of pathogen adhesion, modulation of the immune system, and induction of biochemical changes that suppress the growth of pathogenic microorganisms (Bermudez-Brito *et al.,* 2012) [13]. Among these biochemical

changes is the production of antimicrobial compounds known as *bacteriocins*, which are proteinaceous in nature and exhibit targeted activity against specific pathogens. In addition to bacteriocins, probiotics also produce short-chain fatty acids (SCFAs), hydrogen peroxide (H_2O_2), and diacetyl compounds that collectively contribute to the modulation of intestinal microflora and confer positive health outcomes (Hawrelak and BNat, 2013) [39]. Nearly all strains of *Bifidobacteria* and *Lactobacilli* are capable of producing bacteriocins. Notably, in 2020, the genus *Lactobacillus* underwent significant taxonomic revision to better reflect the extensive diversity within the group.

Table 1: Notion with brief definitions

Notion	Definition
Probiotics	Live microorganisms that, when administered in adequate amounts, confer a health benefit on the host
Prebiotic	A selectively fermented ingredient that results in specific changes in the composition and/or activity of the gastrointestinal microbiota, thus conferring benefit(s) upon host health
Synbiotics	A mixture comprising live microorganisms and substrate(s) selectively utilized by host microorganisms that confer a health benefit on the host. There are two types of symbiotic as complementary synbiotics (mixtures of probiotics and prebiotics) and synergistic synbiotics (mixtures of live microbes selected to utilize a co administered substrate for a health effect)
Postbiotic	A preparation of inanimate microorganisms and/or their components that confers a health benefit on the host
Lactic acid bacteria (LAB)	A functional classification of nonpathogenic, non-toxigenic, Gram-positive, fermentative bacteria those are associated with the production of lactic acid from carbohydrates, making them useful for food fermentation. Species of *Lactobacillus*, *Lacticaseibacillus*, *Lactiplantibacillus*, *Limosilactobacillus*, *Levilactobacillus*, *Lactococcus*, and *Streptococcus thermophilus* are included in this group. Many probiotics are also LAB, but some probiotics (such as strains of *E. coli*, *Akkermansiamucinophila*, bacterial spore-formers, and yeasts used as probiotics) are not
Fermentation	A process by which a microorganism transforms food into other products, usually through the production of lactic acid, ethanol, and other metabolic end products

Source: WGO, 2023

Prebiotics and Synbiotics

The Prebiotic Concept, first proposed by Gibson and Roberfroid in 1995, is more recent than probiotics. The key aspects of a prebiotic are that it is non-digestible by the host and that it leads to health benefits for the consumer through a positive influence on the resident beneficial microbes. Prebiotics typically consist of non-starch polysaccharides and oligosaccharides, although other substances are being studied as candidate prebiotics (Fig. 1) such as resistant starch, conjugated linoleic acid, and polyphenols. Sources of prebiotics i.e. Jerusalem artichoke, chicory roots, tomatoes, berries, onions, garlic, unrefined wheat, soybeans, asparagus, indigestible carbohydrates, etc. are reported by Pokusaeva *et al.*, 2011 [70]. Most prebiotics are used as food ingredients, in foods such as biscuits, cereals, chocolate, spreads, and dairy products. Commonly known prebiotics are: Oligofructose (fructooligosaccharide-FOS), Inulin, Galacto-oligosaccharides (GOSs), Lactulose and Breast milk oligosaccharides (human milk oligosaccharides or HMOs). Prebiotics can be defined as indigestible food components that provide health benefits by restoring the growth of beneficial microbes in the gastrointestinal tract (GIT). Prebiotics are known majorly for stimulating the activity and growth of good bacteria in the GIT. They stimulate the growth of bacteria present in the colon. Unlike other food components, they are hardly affected by hydrolyzing enzymes or acids which are present in our GIT, but are prone to fermentation by beneficial bacteria. There are certain prebiotics that exhibits several health benefits apart from modulating the growth of beneficial microbes. They perform as anti-inflammatory, anti-diarrheal and lower the risk of colon cancer. Alginate and agar have been derived from seaweed. In addition to being abundant in the polysaccharide known as Ulvan, Gelidium is also a unique prebiotic for the bacteria known as *Faecalis bacteriuria* (Saulnier *et al.*, 2009) [81]. They inhibit the growth of harmful bacteria and promote the growth and development of beneficial bacteria like *Bifidobacterium* (Lacerda *et al.*, 2020) [48]. The feasibility and effectiveness of probiotics are dependent on several factors including oxygen, moisture, stress, pH, etc. (Mazziotta *et al.*, 2023) [57].

The Synbiotics Concept was originally described as appropriate combinations of prebiotics and probiotics. More recently, the concept of synbiotics has evolved to consist of both complementary and synergistic synbiotics. A complementary synbiotics is defined simply as a mixture of probiotic(s) and prebiotic(s), where the two components meet the criteria defined for each, including proper characterization, and are used at a dose shown to provide a health benefit (Fig. 1). However, a synergistic synbiotics has been described as a mixture of a live microbe selected to utilize a co administered substrate, which together lead to a documented health benefit. The components of a synergistic synbiotics do not need to independently meet the criteria for a probiotic or prebiotic. Synbiotics not only improves the chances of survival of beneficial microbes, but also stimulates the growth and proliferation of the native bacterial population in the GI tract. Similarly synbiotics not only inhibit the growth of harmful pathogens, but also maintains intestinal biostructure and reduce the undesired metabolite concentration. Prebiotics improve the tolerance of probiotics to environmental factors like temperature, oxygenation and pH inside the GIT. But when prebiotics and probiotics are combined, efficiency of microbes and their tolerance to limiting factors get drastically improved causing a beneficial effect on the host's body (Manigandan *et al.*, 2012) [53]. Synbiotics are quite efficient in reducing blood fat and glucose levels, immune system modulation, osteoporosis prevention and in the treatment of neurodisorders arising from abnormal hepatic functions. *Lactobacilli*, *B. coagulans*, *Bifidobacteria* sp., *S. boulardi*, etc. are the most commonly used probiotic strains in synbiotic formulations along with oligosaccharide-based prebiotics (Pandey *et al.*, 2015) [66]. Synbiotics amalgamate probiotics (viable advantageous bacteria) with prebiotics (agents that facilitate their proliferation) to improve health (Fig. 2). The selection of probiotic and prebiotic strains is essential (Chan and Liu, 2022) [19]. The market applications of synbiotics are expanding as consumers become increasingly informed about gut health, probiotics, and

prebiotics. Primary applications encompass Synbiotics are commonly used into yogurts, smoothies, and dietary supplements (Rashidinejad *et al.*, 2022) [73]. These products provide convenient solutions for intestinal health. Moreover, Synbiotics may address dysbiosis related conditions such as gastrointestinal disorders, obesity, and metabolic syndrome. Additionally, microbiome research may provide tailored synbiotics formulations to address specific health requirements and microbiome characteristics. This could enhance the effectiveness of synbiotics for particular health outcomes (Chaturvedi and Chakraborty, 2021) [20]. A complementary synbiotics combines a prebiotic and a probiotic, which work independently to bring out one or more health benefits. The synergistic synbiotic is composed of a substrate that is utilized by the co-administered live microorganism, enhancing its functionality (Swanson *et al.*, 2017) [86].

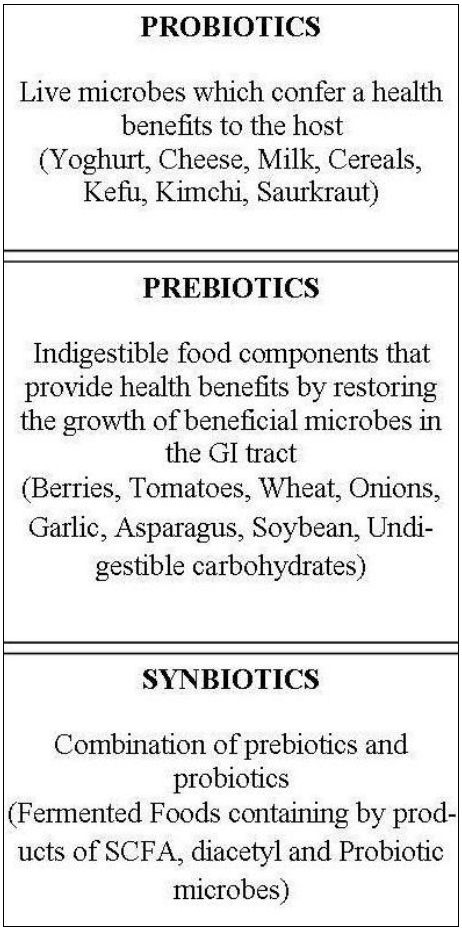
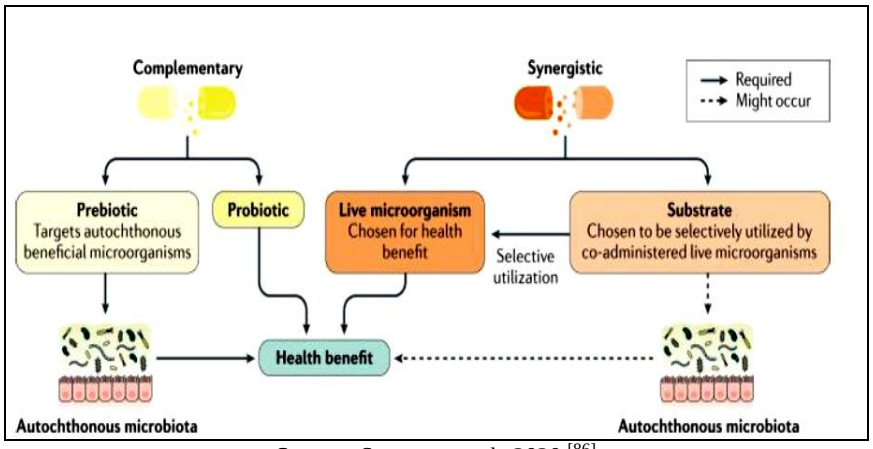


Fig 1: Prebiotics, Probiotics and Synbiotics with their sources



Source: Swanson *et al.*, 2020 [86]

Fig 2: Composition of Complementary and Synergistic Synbiotics

Genera, Species, and Strains used as Probiotics

A probiotic strain is identified by the genus, species, subspecies (if applicable) and an alphanumeric designation that identifies a specific strain. In the scientific community, there is an agreed nomenclature for genus, species, and subspecies names. Strain designations, product names, and trade names are not controlled by the scientific community. According to the guidelines of the World Health Organization (WHO) and Food and Agriculture Organization (FAO), probiotic manufacturers should deposit

their strains in an internationally recognized culture collection. Such depositories will give an additional designation to strains (WGO, 2023). Few examples of commercial strains and the names associated with them given here (Table 2).

Colonizing Microbiota

The functions of both probiotics and prebiotics for gastrointestinal endpoints are interwoven with the microbes that reside in the human gut. The intestine contains a large number of microbes, located mainly in the colon and comprising hundreds of species. Estimates suggest that over 40 trillion bacterial cells are harbored in the colon of an adult human being (including a small proportion of Archaea, less than 1%). Fungi and protists are also present, with a negligible contribution in terms of cell numbers, whereas viruses/phages may outnumber bacteria cells. Gut microbes add an average of 600,000 genes to each human being (Li *et al.*, 2014). At the level of species and strains, the microbial diversity between individuals is quite remarkable: each individual harbors his or her own distinctive pattern of bacterial composition, determined partly by the host genotype, by initial colonization at birth via vertical transmission, and by dietary habits. In healthy adults, the fecal composition is stable over time. In the human gut ecosystem, the two bacterial divisions *Bacteroidetes* and *Firmicutes* predominate and account for more than 90% of microbes. The rest are *Actinobacteria, Proteobacteria, Verrucomicrobia*, and *Fusobacteria*. The normal interaction between gut bacteria and their host is a symbiotic relationship. An important influence of intestinal bacteria on immune function is suggested by the presence of a large number of organized lymphoid structures in the mucosa of the small intestine (Peyer's patches) and large intestine (isolated lymphoid follicles). The epithelium over those structures is specialized for the uptake and sampling of antigens, and they contain lymphoid germinal centers for induction of adaptive immune responses. In the colon, microorganisms proliferate by fermenting available substrates from diet or endogenous secretions and thereby contribute to host nutrition. Many studies have shown that populations of colonizing microbes differ between healthy individuals and others with disease or unhealthy conditions. However, researchers are not able to define the composition of healthy human microbiota (Table 3). Certain commensal bacteria (such as *Roseburia*, *Akkermansia*, *Bifidobacterium*, and *Faecalibacterium prausnitzii*) seem to be associated more commonly with health, but it is a current active area of research to determine whether supplementation with these bacteria will improve health or reverse disease.

Table 2: Nomenclature used for Probiotic Microorganisms

Genus	Species	Subsp.	Strain designation	International strain depository designation	Strain nick name	Product name
Lacticaseibacillus	*rhamnosus*	None	GG	ATCC 53103	LGG	Culturelle
Bifidobacterium	*animalis*	*lactis*	DN-173 010	CNCM I-2494	*Bifidus regularis*	Activia yogurt
Bifidobacterium	*longum*	*longum*	35624	NCIMB 41003	*Bifantis*	Align

Table 3: Human Intestinal Microbiota

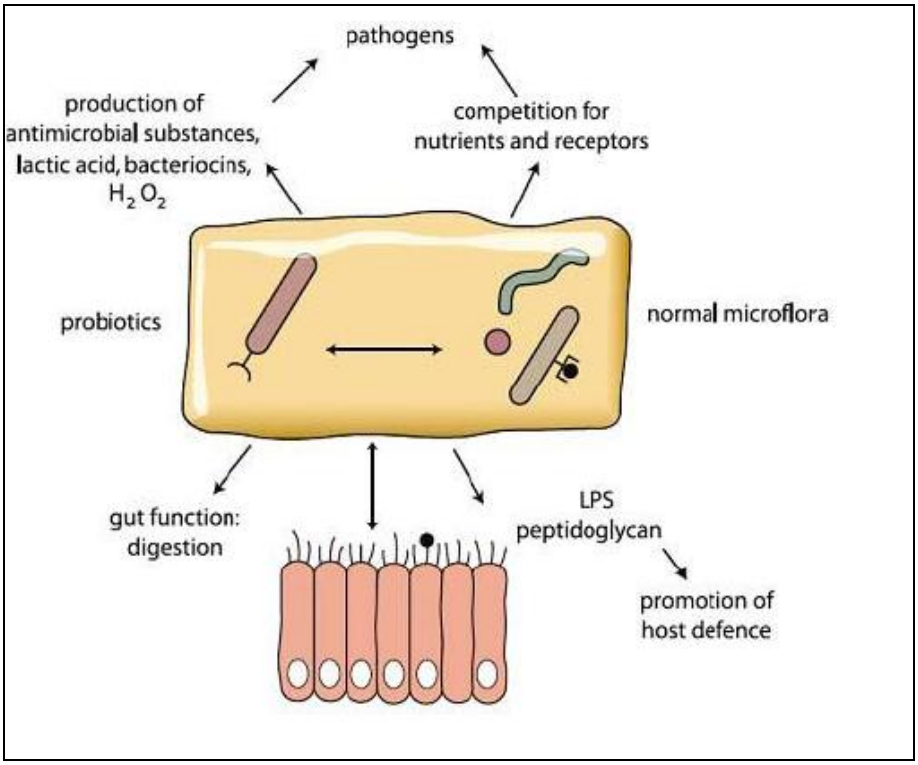
Stomach and duodenum	• Harbor very low numbers of microorganisms: < 103 cells per gram of contents • Mainly lactobacilli and streptococci • Acid, bile, and pancreatic secretions suppress most ingested microbes • Phasic propulsive motor activity impedes stable colonization of the lumen (also true for the small intestine)
Jejunum and ileum	Numbers progressively increase from 104 in the jejunum to 107 cells per gram of contents in the distal ileum
Large intestine	Heavily populated by anaerobes: up to 1012 cells per gram of luminal contents

Source: WGO (2023)

(The gut microbiota form a diverse and dynamic ecosystem, including bacteria, Archaea, Eukarya, and viruses that have adapted to live on the intestinal mucosal surface or within the gut lumen)

Mechanisms of Action of Probiotics and Prebiotics

Prebiotics affect intestinal bacteria by enhancing the numbers or activities of beneficial bacteria. This may result in decreasing the population of potentially pathogenic microorganisms or reducing potentially deleterious metabolic activities of host microbiota. Prebiotics may also impact immune function. Probiotic strains may mediate health effects through one or more of several identified mechanisms. Probiotics may affect the intestinal ecosystem by impacting mucosal immune mechanisms, by interacting with commensal or potential pathogenic microbes, by generating metabolic end products such as short-chain fatty acids, and by communicating with host cells through chemical signaling (Fig. 3; Table 4). These mechanisms can lead to antagonism of potential pathogens, an improved intestinal environment, bolstering the intestinal barrier, down-regulation of inflammation, and up-regulation of the immune response to antigenic challenges. These phenomena are thought to mediate most beneficial effects, including reduction of the incidence and severity of diarrhea, which is one of the most widely, recognized uses of probiotics.



Source: WGO, 2023

Fig 3: Mechanisms of Microbiota and Probiotic Interaction with the Host

Table 4: Mechanisms of Probiotic and Prebiotic Host Interaction

Probiotics	
Immunologic benefits	• Activate local macrophages to increase antigen presentation to B lymphocytes and increase secretory immunoglobulin A (IgA) production both locally and systemically • Modulate cytokine profiles • Induce tolerance to food antigens
Nonimmunologic benefits	• Digest food and compete for nutrients with pathogens • Alter local pH to create an unfavorable local environment for pathogens • Produce bacteriocins to inhibit pathogens • Scavenge superoxide radicals • Stimulate epithelial mucin production • Enhance intestinal barrier function • Compete for adhesion with pathogens • Modify pathogen-derived toxins
Prebiotics	
• Metabolic effects: production of short-chain fatty acids, absorption of ions (Ca, Fe, Mg) • Enhancing host immunity (IgA production, cytokine modulation, etc.)	

(Symbiosis between microbiota and the host can be optimized by pharmacological or nutritional interventions in the gut microbial ecosystem using probiotics or prebiotics)

Safety Criteria of Probiotics, Potential Risks and Safety Concerns for Specific Populations

The safety of probiotics is crucial due to their consumption by diverse populations, including infants, the elderly and immune compromised individuals. The subsequent safety criteria are typically evaluated. Incorporating with Probiotic strains must be accurately characterized to ensure identification, purity, and viability (Damián *et al.*, 2022; Mazziotta *et al.*, 2023) [26, 57]. Probiotic products must undergo testing for harmful bacteria, mycotoxins, and heavy metals. The final product must be free from hazardous contaminants to ensure customer safety (Tegegne and Kebede, 2022) [89]. Manufacturers must rigorously oversee quality during the whole production process. This entails maintaining the potency, stability, and shelf life of probiotic products while adhering to superior manufacturing practices (Al-Rashidi *et al.*, 2022) [4].

Probiotics Classification

At present, there are a variety of microbes that are being used as probiotics (Hawrelak and BNat, 2013) [39]. Bacteria belonging to the *Lactobacillus* genus are gram-positive bacilli that are capable of producing lactic acid in the GIT and GUT (Genitourinary tract), they are anaerobes that can improve uptake and bioavailability of minerals and reduce intestinal permeability. In fact, some strains of this genus exhibit anti-cancer and hypolipidemic activity (Cichonska and Ziarno, 2022) [23] as well as highly prone to acidic environments and high temperatures (Cao *et al.*, 2020) [17]. Some of the bacterial strains (Elhossiny *et al.*, 2023) [31] those are commonly used as probiotics given here (Table 5).

Table 5: Some Important Bacterial Strains Commonly Used as Probiotics

Genus	Species
Lactobacillus spp.	*Acidophilus, rhamnosus, fermentum, johnsonii, lactis, reuteri*
Bifidobacterium spp.	*Breve, infantis, longum, lactis, thermophilum*
Bacillus spp.	*coagulans*
Streptococcus spp.	*thermophilus*
Enterococcus spp.	*faecium*
Saccharomyces spp.	*cerevisae*

- ***Lactobacillus acidophilus:*** The strain *Lactobacillus acidophilus* has been widely utilized due to its potential health advantages. It possesses the capacity to cling to diverse intestinal cells, exhibits tolerance to bile, and demonstrates resistance to acid, which are essential attributes for a probiotic strain. Commercially accessible strains of *Lactobacillus acidophilus* are LA-1, LA-5, NCFM, DDS-1, and SBT-2026 (Elhossiny *et al*, 2023) [31].

- ***Lactobacillus rhamnosus:*** The strain *Lactobacillus rhamnosus* has developed distinctive adaptations that enable its survival in the acidic and basic environments present in the human body. The capacity of *L. rhamnosus* to attaches and colonize the intestinal walls enables it to potentially provide extended advantages. Consequently, it is frequently used into yogurts, cheeses, milk, and other dairy products to augment probiotic levels. Additionally, *L. rhamnosus* plays a pivotal part in the process of cheese ripening, so boosting the overall flavor. Moreover, some strains of *L. rhamnosus* have demonstrated advantageous effects on both adults and children, specifically in the treatment of irritable bowel syndrome (IBS), eczema, allergies, and immune system support (Gao *et al.*, 2022) [35].

- ***Lactobacillus fermentum:*** *Lactobacillus fermentum* is a probiotic bacterium that exhibits abilities in adhesion and anti-infective properties. It is classified as non-starter lactic acid bacteria (NSLAB) within specific cheese varieties. These characteristics suggest that *Lactobacillus fermentum* has the potential to effectively combat infections and promote health in the urinary-reproductive tract. Additionally, it has been observed that *Lactobacillus fermentum* strains exhibit a notable capacity for auto-aggregation, a crucial factor in their ability to cling to epithelial cells and facilitate the production of biofilms within the gastrointestinal tract (Anjum *et al.*, 2014) [5].

- ***Lactobacillus johnsonii:*** *Lactobacillus johnsonii* is utilized in Nestlé's LC-1 yogurt products and possesses the capacity to augment immune responses, survive diverse conditions such as bile salts and antibiotics, combat antimicrobial multidrug resistance microorganisms, and maintain a high level of probiotic viability in food items. Furthermore, *Lactobacillus johnsonii* has demonstrated the ability to decrease the adhesion and activity of pathogenic strains, impede the growth of gut pathogens, and shorten the duration of diarrhea and enterocolitis (Tavasoli *et al.*, 2022) [87].

- ***Lactobacillus lactis:*** *Lactobacillus lactis* (lactic acid bacterium) has a good potential of probiotic capabilities in enhancing immune system function and alleviating inflammatory bowel illness. The anti-inflammatory capabilities of some strains of *Lactobacillus lactis*, such as *L. lactis* ML-2018, have been recognized, particularly in their ability to inhibit the production of inflammatory factors triggered by lipopolysaccharides (LPS) (Nazia *et al.*, 2014 [65]; Zanjani *et al.*, 2017) [104].

- ***Lactobacillus reuteri:*** *Lactobacillus reuteri* is encompassing the prevention and management of urogenital disorders and bacterial vaginosis in females, atopic disorders, food hypersensitivity, and the prevention of dental caries. Furthermore, it has been examined for its capacity to prevent colitis and decrease contacts between P-selectin-associated leukocytes and platelets with endothelial cells, emphasizing its significance in intestinal illnesses (Assimos, 2020) [9].

- ***Bifidobacterium breve:*** *Bifidobacterium breve* is a symbiotic organism that resides in the human intestines. It is used for the treatment of several ailments such as constipation, diarrhea, irritable bowel syndrome, and even the common cold and flu (Zanjani *et al.*, 2017) [104].

- ***Bifidobacterium infantis:*** *Bifidobacterium infantis* naturally inhabits the mouth and digestive system. It belongs to the same group as *Lactobacillus* and is a type of lactic acid bacteria that is essential for keeping a healthy digestive system. *Bifidobacterium infantis* 35,624 has been specifically examined for its potential in treating irritable bowel syndrome (IBS). Its efficacy in alleviating symptoms such as bloating, bowel issues, pain, and gut dysbiosis in infants has been demonstrated (Sanders *et al.*, 2019) [78].

- ***Bifidobacterium longum:*** *Bifidobacterium longum* resides in the gastrointestinal system and is widely recognized as a prominent constituent of the human gut microbiota. It is particularly prevalent in the baby gut, where it is the most numerous species. It demonstrates the synthesis of bioactive compounds and the interaction between bifidobacterial surface-associated molecules and the host organism. Its ability to modulate immune cells, and its potential to reduce allergic reactions in mice models and treat inflammatory bowel disease (Azad *et al.*, 2018) [11].

- ***Bifidobacterium lactis:*** *In vitro* testing has extensively established the strain characteristics and processes of *Bifidobacterium lactis*, demonstrating its great stability in meals and freeze-dried powders. Clinical evidence has demonstrated that *Bifidobacterium lactis* HN019

promotes gut health, digestion, and immunological function (Sanders *et al.*, 2019) [78].

- ***Bifidobacterium thermophilum:*** *Bifidobacterium thermophilum (*aero tolerant bacterium) possessing the ability to endure and flourish in oxygen-depleted settings, rendering it a promising contender for probiotic application. *Bifidobacterium thermophilum* demonstrates bacteriocins like antimicrobial properties against various pathogens, including *Listeria* spp., *Salmonella* spp., *Campylobacter jejuni* in broiler and rota virus infection.

- ***Bacillus coagulans:*** *Bacillus coagulans* (spore-forming probiotic bacteria) renowned for its exceptional resilience to adverse settings and its multitude of probiotic traits, enabling it to remain inactive under harsh conditions (elevated gastric acidity). Additionally, it has the ability to control the host's symbiotic microbiota and hinder the proliferation of harmful bacteria, so promoting general gastrointestinal well-being and providing support to the digestive and immune systems. In natural food sources, such as fermented foods like sauerkraut, kimchi, and yogurt, *Bacillus coagulans* can be found. Furthermore, it finds utility in several probiotic food additives, showcasing its suitability for industrial implementation in the food sector (Ma *et al.*, 2021) [51].

- ***Streptococcus thermophilus:*** *Streptococcus thermophilus* is frequently used in the manufacturing of many dairy commodities, such as cheeses and yogurt. It aids in the hydrolysis of lactose in milk, leading to the distinctive flavor and consistency of yogurt. It has been linked to a range of health advantages, such as bolstering the immune system and mitigating inflammation in the gastrointestinal and urogenital systems as well, in combating viral, fungal, and parasitic infections (Qu *et al.*, 2023) [71].

- ***Enterococcus faecium:*** *Enterococcus faecium* possesses a distinct advantage in enduring the process of digestion and thriving in the gastrointestinal tract, fostering a harmonious gut milieu by engaging in competition with detrimental species for nutrients and adhesion sites. Moreover, strains of *Enterococcus faecium* exhibit promising therapeutic properties, including the ability to prevent and treat diarrhea in domesticated animals, as well as block the proliferation of pathogenic *Listeria* spp. (Tilwani *et al.*, 2022) [91].

- ***Saccharomyces cerevisiae:*** *Saccharomyces cerevisiae*, specifically the variant *S. boulardii*, is widely recognized for its impacts on gastrointestinal well-being in both human and animal populations. It is commonly employed as an adjunctive measure against gastrointestinal tract disorders, including inflammatory bowel disease, and for the management of diverse forms of diarrhea (Fernandez-Pacheco *et al.*, 2018) [34].

Microbial Strains for Next Generation Probiotics (NGPs)

Human are recognized as "Superorganisms", and closely associated with microbiomes (Salvucci, 2019; Tawseefa Jan *et al.*, 2024) [76, 88]. The human gastrointestinal tract (GIT), is home to 100 trillion microorganisms (bacteria, fungus, viruses, and protozoa), as revealed by research carried out by the human microbiome study (Anwar *et al.*, 2019) [8].

Role of Probiotics in Food Products

Probiotic foods contain a high quantity of microorganisms, and a daily intake of 108 and 1011CFU/ day is suggested, present in dairy and non-dairy products (Markakiou *et al.*, 2020; El-Sohaimy and Hussain, 2023) [55, 32]. As antibiotic therapy generally affects the intestinal microbiota composition, regular consumption of probiotic foods is supportive of improving the balance of beneficial microbiota composition in the intestine (Dahiya and Nigam, 2023; Leeuwendaal *et al.*, 2022) [25, 49]. The global probiotic food product market is rapidly growing due to consumer awareness, and its impact on food and host health. Today probiotic product in the functional food market ranges from 60% - to 70% (Pimentel *et al.*, 2021) [69]. Probiotic foods and drinks in the global market in 2011 were approximately 24.8 billion euros, 31.1 billion euros in 2015, and in 2020 approximately 43 billion euros. Functional foods are defined as foods containing health-enhancing factors beyond traditional nutrients and are also known as medicinal foods, therapeutic foods, designer foods, nutraceuticals, and superfoods (Hammam, 2019) [38]. Probiotic cultures are incorporated into different food products like milk, milk powder, milk-based drinks, butter, cheese, ice cream, mayonnaise, vegetables, fruits, meat, cereals, and powder products (Ranadheera *et al.*, (2017) [72]. Probiotics incorporated into vegetable products, called vegan probiotics can have hypercholesterolemia and anti-carcinogenic effects in vitro, and Lactiplantibacillus plantarum-1 and *Lacticaseibacillus rhamnosus* GG incorporated into the blueberry bagasse provided effects on cholesterol reduction (Verma and Rout, 2022; Vijayaram *et al.*, 2024) [95, 97]. Some of the important food products from probiotics are given here (Fig. 5).

- **Dairy Products:** Probiotics are used in cheese, yogurt, ice cream, and other dairy products (Shori, 2021; Kariyawasam *et al.*, 2021) [84, 46]. Production of lactic acid by probiotic bacteria in the dairy fermentation process plays a significant role in food preservation, and preventing spoilage caused by *Campylobacter*, *Clostridium botulinum*, *Escherichia coli* O157:H7, *Listeria monocytogenes*, *Norovirus*, *Salmonella*, *Staphylococcus aureus* and *Shigella* (Campos *et al.*, 2023) [16].

- **Probiotic Yogurt:** Yogurt is commonly used due to its taste and health benefits and is in some cases the main meal or snack during the day [Daniel *et al.*, 2022] [27]. Yogurt and fermented milk give an excellent source of fatty acids like conjugated linoleic acid [CLA], and today CLA is attracting a lot of interest due to immunomodulatory, anti-diabetogenic, apoptotic, anti-obesity, anti-carcinogenic, anti-atherogenic, and osteosynthetic properties (Mei *et al.*, 2022) [58]. The production of yogurt requires milk to acidify, and the acidification process mainly depends on bacteria such as *L. acidophilus, L. casei, Lactobacillus delbrueckii, L. paracasei, Streptococcus thermophilus*, and *B. lactis* (Nadelman *et al.*, 2019) [63]. Yogurt provides positive impacts on human health like reducing cholesterol

levels in blood serum and lowering blood pressure and heart rate, including antihypertensive effects (Shafi, *et al.*, 2019; Ahmadian *et al.*, 2022) [83, 3].

- **Probiotic Milk:** Several probiotic bacteria are reported in fermented milk; B. *longum*, *Enterococcus faecium*, *L. plantarum*, *Leuconostoccremoris*, *Lactobacillus helveticus*, *Lentilactobacillus hilgardii*, *Lactobacillus alimentarius*, *Lactobacillus bifermentans*, *L. paracasei*, *Lactobacillus kefiri*, *Lactobacillus kefiranofaciens*, *Lactobacillus pentosus*, *Lactococcus lactis*, and *Leuconostoc pseudomesenteroides* (Terefe, 2022) [90]. Fermented milk provides a positive response on plasma cholesterol levels, decreases low-density lipoprotein levels in hyper cholesterolemic persons, reduces blood pressure, and prevents hypertension (Yilmaz *et al.*, 2022) [103]. The consumption of kefir notably reduces total cholesterol, triglycerides, and LDL cholesterol, reduction in blood glucose level and HbA1C, and enhances insulin resistance (Salari *et al.*, 2021 [75]).

- **Probiotic Cheese:** Probiotic-incorporated cheese products increase the nutritional value and consumer awareness of foods and health, and probiotic cheese products improve sensory properties and health claims like reducing oxidative stress in the liver, lungs, and intestine (Grom *et al.*, 2020; Mukhtar *et al.*, 2020; Mohammed *et al.*, 2021; Homayouni *et al.*, 2022) [37, 62, 60 40].

- **Probiotic Butter:** Supplementations of probiotic strains have also been incorporated in butter play by modulating the health of the human, because a balanced intestinal microbiota reduces the risk of development of various diseases such as cancer, colitis, lactose intolerance, heart diseases, and obesity (Kalicka *et al.*, 2019) [45].

- **Probiotic Ice Cream:** Ice cream supplemented with probiotics provided positive effects, improved human health, and enhanced the nutritional value of the ice cream product, and probiotics viability level is better in ice cream products compared to other functional dairy and non-dairy products based on avocado (Acu *et al.*, 2020 [2]; Krawęcka *et al.*, 2021) [47].

- **Cereal Food Products:** Cereals such as wheat, maize, oat barley, and other grains are rich fibre sources, and they revealed beneficial effects, these products are used to improve the survival and viability of probiotics in the GI tract (Budhwar *et al.*, 2020 [14]; Pavalakumar *et al.*, 2023) [67]. Cereal fermentation by probiotics can decrease non-digestible carbohydrates, increase the level of lysine, and availability of vitamin B group, degradation of phytates, and release minerals i.e manganese, iron, zinc, and calcium (Mishra and Panda, 2022) [59]. Cereals, bread, biscuits, and beverages fermented with LAB revealed pH reduction in the intestine, improvement of antimicrobials, enzymes, and vitamins, reduction of serum cholesterol, regulation of the immune system, revival and restore the gut microbiota after diarrhoea, reduce food allergens, antioxidative activity and reduce lactose malabsorption symptoms (Abdi and Joye, 2021) [1]. *L. acidophilus*, *Lactobacillus bulgaricus*, *B. bifidum*, and *S. thermophilus* are used to develop rice-based food product applications [Chaturvedi and Chakraborty (2021) [20]. In addition, supplementation of LAB and bifidobacteria in the fermentation of soy milk significantly increased antioxidant activities and serum lead levels but in rats no significant effect on blood parameters such as red blood cell counts, haematocrit and haemoglobin levels, and superoxide dismutase (SOD) activity and malondialdehyde (MDA) were revealed (Iraporda *et al.*, 2022) [43].

- **Fruit Food Products:** Different probiotics are used as supplements to fruits and vegetables like pineapple, cranberry, strawberry, sweet lime, mango, grapes, cashew apple, olive, carrot, beetroot, and oranges (Natt and Katyal, 2022) [64]. Durian fruit, a product called "tempoyak" manufactured in Malaysia is fermented with *Levilactobacillus brevis*, *Leuconostoc mesenteroides* and *L. fermentum* (Jain *et al.* (2021) [44]. Different types of fruits and vegetables are used to make turşu products, and the LAB tursu can be of help against cirrhosis and diarrhoea (Şanlıbaba, 2023) [79]. *B. longum* and *L. plantarum* are the suitable carrier delivery for non-juice as well as these products considerably increased antioxidant level, reduce cholesterol level and enhance product quality (Chaudhary *et al.*, 2019) [21]. Cornelian cherry juice fermented with L. *plantarum* ATCC 14917 evaluated the viability in the juice during cold storage, but no sensory variations were noted between the fermented and non-fermented samples (Mantzourani *et al.*, 2019) [54].

- **Vegetable Food Products:** Vegetables contain carbohydrates, vitamins, minerals, and phytochemicals, and fermented vegetables are the new way to improve plant-based products like beet, carrot, radish, artichoke, cabbage, broccoli, celery, aloevera, soy, almond, and walnut (Valero-Cases *et al.*, 2020; Ansari and Pourjafar, 2021) [93, 7]. Vegetable juices are of help for lactose intolerant people who cannot intake probiotics via milk and milk products and people on a fat-free diet as the juices contain zero fat and high fibre content (Barbu *et al.*, 2020) [12].

- **Meat Products:** The most frequently used probiotics in fermented meat products are *L. plantarum*, *L. sakei*, *L. paracasei*, *L. casei* and *L. rhamnosus* (Das *et al.*, 2020; Montanari *et al.*, 2022) [28, 61]. The inclusion of probiotics in meat products considerably increased taste, flavour, quality, and moisture conditions (Sbehat *et al.*, 2022) [82]. However, successful incorporation is dependent on viability during processing and product shelf life as some technological advancements can affect the survival of the probiotics in fermented meat products like pH, acidity, water activity, processing and storage temperature, presence of other microorganisms, the concentration of additives (salt, sugar, nitrite, and nitrate), and composition of the protein matrix (Todorov *et al.*, 2022) [92].

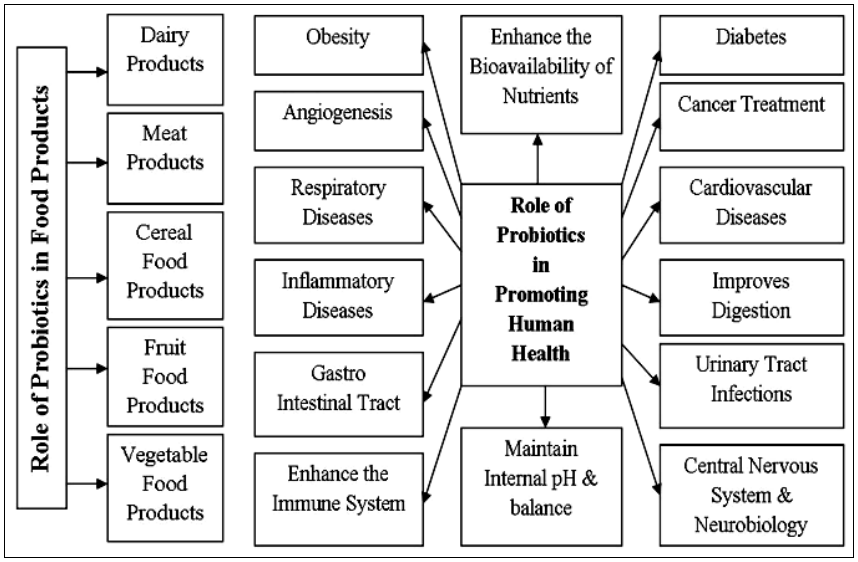


Fig 5: Role of Probiotics in Food Products and Promoting Human Health

Role of Probiotics in Promoting Human Health

Probiotics are used for the management of an array of disorders and unusual physiological conditions (Sarita *et al.*, 2025) [80]. Probiotics are now under investigation for their potential in treating obesity, metabolic syndrome, respiratory infections, and Covid-19. Some of the health benefits from probiotics are given here (Fig. 5).

- **Probiotics in Cancer Treatment:** Cancer is undoubtedly one of the leading causes of death around the globe. Probiotics play an important role in reducing the risk of colon and bladder cancer. *Helicobacter*, *Pseudomonas* and *Acinetobacter* are responsible for tumor formation in the colon which ultimately leads to colon cancer. They proliferate easily in absence of beneficial bacteria. Probiotics can play a major role in modulating the intestinal and gut microbiome. *Lactobacillus acidophilus* and *L. caseishirota* are the most commonly used strains (Dasari *et al.*, 2017; Maleki *et al.*, 2016) [29, 52]. Probiotics have the potential to inhibit the proliferation and growth of colorectal cancer (CRC), which are essential metabolites of probiotics, serve as a source of energy for the colonic mucosa, strengthen the intestinal protective barrier, regenerate the colonic epithelium, regulate the pH of the intestinal lumen, inhibit the proliferation of cancer cells, and encourage the death of cancer cells through the process of apoptosis (Wong and Yu, 2019) [99]. Suitable probiotic-related treatments may be utilized for the purpose of preventing colorectal cancer (CRC) prior to the development of the disease (Ezeji *et al.*, 2021) [33]. Probiotics are currently being used as an adjuvant therapy in the treatment and care of colorectal cancer (CRC), primarily for the purpose of mitigating surgical complications and alleviating the adverse effects of chemotherapy (Zhao *et al.*, 2023) [105].

- **Probiotics for Treatment of GI Tract Issues:** Probiotics can be effectively used for the treatment of lactose intolerance, gastrointestinal infections, ulcerative colitis, gastrointestinal tumors and Chrohn's disease. Probiotics compete with pathogens for the binding site at epithelial tissue and some synthesize biochemical that inhibits the growth of pathogens. *Lactobacillus plantanum* effectively prevents bloating and abdominal pain, *S. boulardii*is used for the treatment of diarrhea and for improving overall gut functioning (Iannitti and Palmieri, 2010) [42].

- **Probiotics for Diabetes:** Probiotics can effectively modulate gut hormones, the hormones are known for controlling homeostasis, their modification neutralizes the resistance to insulin which is the major cause of type 2 diabetes. There are some probiotics that can reduce the growth of adipocytes which aids in the prevention of a range of metabolic disorders (Westfall *et al.*, 2018) [98].

- **Probiotics for UTI:** Urinary Tract Infections (UTI) is quite common in both elder and young women. There are over 50 probiotics that can effectively treat UTI, all these are based on *Lactobacillus* spp. i.e. *Lactobacillus brevis*, *L. reuteri*, *L. vaginalis*, *L. rhamnosus* (Van *et al.*, 2023) [94].

- **Probiotics for Obesity:** Genetic variability, energy intake and expenditure imbalance are the major reasons for obesity which is a growing issue in these times. Adiponectin and leptin are present in adipocyte tissues and these are majorly responsible for obesity, *Lactobacillus gasseri* BNR17 inhibits their growth. Probiotics stimulate the adrenergic nervous system which generates a thermogenic response, this facilitates weight loss. Certain probiotics like *Lactobacillus acidophilus*, *L. casei*, *Bifidobacterium longum* exhibit hypocholesterolemic activity, they reduce the level of triglycerides, Low Density Lipids (LDL) and High Density Lipids (HDL) (George *et al.*, 2018) [36].

- **Probiotics for Central Nervous System (CNS) and Neurobiology:** *Lactobacillus plantanum* shows a promising effect to children with autism. Certain strains of *Lactobacillus* like *L. helveticus*, *L. casei*, *L. rhamnosus* which are known to reduce psychological distress, anxiety symptoms, autism associated symptoms, respectively. The relationship between the

brain and the gastrointestinal tract is well-established (Victoria Obayomi *et al.*, 2024) [96]. A novel, natural therapy for mental disorders utilizing prebiotics, probiotics, and synbiotics to regulate the central nervous system with minimal side effects of mental disorder (Cryan *et al.*, 2019; Ansari *et al.*, 2023) [24, 6] and identified positive effects of prebiotics, probiotics, and synbiotics on anxiety, depression, stress, sleep, and Alzheimer's disease (Centurion *et al.*, 2022) [18].

- **Probiotics for Angiogenesis:** New vessels are regenerated from old vessels via angiogenesis, it aids in faster wound healing. If not done in a proper way, it may lead to cancer and diabetic retinopathy. *S. boulardii* is known for protecting the host body against inflammation and injury by decreasing visceral hypersensitivity and modifying inflammatory cytokine profile (Xu *et al.*, 2024) [101].

- **Probiotics for Respiratory Diseases:** Bronchitis, sinusitis, pharyngitis, rhinosinusitis, otitis is some of the most common respiratory disorders. Probiotics exhibit both anti-inflammatory and anti-microbial properties owing to which they can be used for the prevention of a number of respiratory disorders. For example: for controlling episodes of pneumonia in patients with cystic fibrosis, *Lactobacillus rhamnosus* can be administered. *Lactobacillus fermentum*, *L. casei*, and *Bifidobacterium longum* are some of the common probiotics used for the treatment of respiratory issues (Soccol *et al.*, 2014) [85].

- **Probiotics for Cardiovascular Diseases:** Angiotensin-Converting Enzyme (ACE) is a key enzyme behind hypertension. *Lactobacillus helveticus* and *Saccharomyces cerevisiae* are known to synthesize peptides that can inhibit the activity of ACE (Mayta-Tovalino *et al.*, 2023) [56].

- **Probiotics for Inflammatory Diseases:** Probiotics can be used for the treatment of Ulcerative colitis and Chrohn's disease. They result in inflammation of GIT, both aerobic and anaerobic bacteria are responsible. *Lactobacillus*, *Enterobacter*, and *Bifidobacillus*are used for the treatment of inflammation (Roy *et al.*, 2023) [74].

Conclusion

Natural mediated functional foods awareness is the major development for probiotic functional foods as probiotic functional foods provide numerous health-beneficial effects, and dairy and non-dairy products are suitable carriers for the delivery and growth of probiotics. Dairy products used to be the most popular functional food, however these days there are additionally non-dairy functional foods. Probiotics have been shown in numerous studies to have potential uses in lowering illness risk. In recent years, the usage of probiotics has grown a lot since they may be delivered in many ways with little side effects. Numerous probiotics have been authorised for mass sale in order to reduce illness symptoms. Future research should look into other methods of encapsulating probiotics to increase their stability and viability, enhance the quality of functional products, and provide health advantages.

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