From Kimchi to Kefir: An Exploration of Probiotics, Benefits, and Future

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Abstract. Live microorganisms known as probiotics, which have various beneficial claims, have undergone substantial research and commercial exploration in a wide range of goods across the world. Many scientific studies have demonstrated their advantages for both human and animal health. The two primary probiotic bacterial species are *Lactobacillus* sp. and *Bifidobacterium* sp. The multi-billion health food industry has employed probiotics with a variety of dietary matrices, which are briefly reported. The history of probiotics, their use in food and medicine, and the latest developments in probiotic processes such as microencapsulation and genetically engineered probiotics are all covered in this review.

Keyword. Antibiotics, Engineered probiotics, Fermentation, Freeze-drying, Lactic acid bacteria, Lactobacillus, Microencapsulation.

1 Introduction

Probiotics are essentially living microorganisms that help to maintain a healthy balance of gut bacteria, improving the overall health of the individual [1,2]. Lactobacillus and Bifidobacterium are the most common types of Gram-positive probiotic bacteria in which their primary roles involve regulating and preserving the health of the intestinal tract [3]. Probiotics are commonly added to a variety of foods, including traditional foods like dairy products and specialized food like yogurt, kombucha, and fermented veggie [4]. Probiotics can also be food and medical supplements [5]. According to Arora and Baldi, probiotics can be classified as pharmabiotics and nutria-biotics as shown in Figure 1 [6].

Pharmabiotics refer to probiotics that have particular health claims and are potentially able to improve physiological functions or treat diseases through pharmacological mechanisms [7]. On the other hand, nutribiotics are in the form of food or supplement to sustain and modify the physiological function to maintain human health [7,8]. With the beneficial properties and high demand for probiotics, this industry has appeared as one of the fastest expansions in food categories in recent years [2].

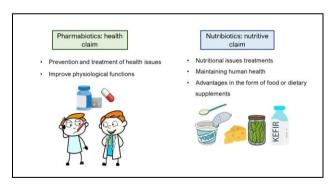


Fig. 1. Difference between pharmabiotics and nutribiotics.

Probiotics serve multiple crucial functions such as digestion and absorption of nutrients. They are also known to decrease the chances of infections and the production of toxins by harmful organisms, thereby reducing the potential for various human diseases [9, 10] and exhibit anticancer, anti-obesity, anti-diabetic, and immunomodulatory [11].

Probiotics contribute considerably to food preservation and enhancement, specifically via the fermentation process. This involvement allows probiotics to improve the taste, texture, and longevity of various food items including fruits, vegetables, baked goods, and meat products [9,12]. By fermentation,

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carbohydrates are changed into new products by microorganisms in a chemical reaction. In ancient times, people used the natural fermentation process to create a variety of goods, such as meals and medicines [13] as it was known to promote products' safety and preservation [14]. In addition, fermented or preserved food and beverages were associated with particular ethnic practices or cultures without recognizing the science behind utilizing the role of microorganisms. The fermentation processes involved in creating foods such as kimchi and kefir embody principles of sustainability and waste reduction. Fermentation is an ancient technique used to prevent food spoilage, and thus, reduce waste. For example, kimchi is traditionally made using cabbage and other vegetables that might otherwise be discarded due to seasonal overproduction. This process not only minimizes waste but also adds nutritional value to the original product. Kimchi is a common fermented vegetable that originated in Korea and is made primarily from cabbage and seasonings [15]. Kimchi has over 200 types with effective properties in battling cancer, aging, and inflammation [14]. Probiotics are not only limited to food but also cover beverages such as kefir. Similar to yogurt, kefir is a fermented milk generated by lactic acid bacteria and yeast contained in kefir grains [15]. It has antitumor and antiallergic features with the ability to reduce blood pressure [14].

Traditionally, when a scientific background was shaped about the important function of useful bacteria in fermented food, the category of dairy products dominated the probiotic foods in the market. However, nowadays, food industries have introduced several products globally, both dairy and non-dairy, as researchers have explored the beneficial impact of probiotic microbes [16]. Consequently, as illustrated in Figure 2, the probiotics market is predicted to grow from \$66.67 billion in 2022 to \$90.43 billion in 2026 a compound annual growth rate of 7.9% [17].

With the increasing interest in studying probiotics, various developments were achieved, for instance, engineered probiotics to yield products with desired and improved qualities, as well as microencapsulation technologies that intended to enhance the probiotics microbiome's viability during products' manufacturing and storage and within the harsh environment of the human gut. Lately, probiotics are being studied as a friendly antibiotic alternative [18] and to combat COVID-19 infections [11].

To maximize the potential of nutrition in maintaining health and preventing diseases, nutritionists, researchers, pharmacists, regulatory authorities such as the World Health Organization, and food producers are currently working together [19]. Hence, research on the possible positive impacts of probiotics on health has become a central area of study in the fields of nutrition, food, and medical sectors [19,20].

Since eating habits play a major role in people's health, nowadays, consumers have become increasingly aware of what they consume, then, there is a real need for the production of items, which offer extra health advantages beyond those of the original products [21] as well as outlining the work and studies done on such

food. This work shed light on providing useful information and exploring probiotics. It particularly aims to discuss types of probiotics, health benefits, latest developments, challenges, limitations, and the future of probiotics.

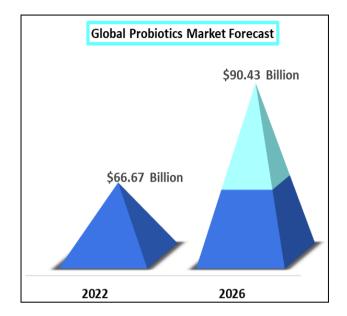


Fig. 2. 2026 forecast of the global probiotics market.

2 Literature Review

The word "probiotics," which means "for life," is a Greek word [22]. The classification of lactic acidproducing bacteria, also known as LAB, comprises such genera as Lactobacillus, Enterococcus, Streptococcus, and Bifidobacterium. Traditional fermented dairy products like Maasai milk, kurut, kefir, lassi curd, and buttermilk are the main source of these healthy probiotic microorganisms [23]. Regarding Gutiérrez et. al., [24], probiotics have been linked to a variety of therapeutic benefits, including improved immune defense, lowered cholesterol levels, shielding against gastrointestinal and urinary tract infections, cancer, and assisting in the treatment of atopic dermatitis, atherosclerosis, rheumatoid arthritis, and other conditions.

Table 1. Examples of disorders or diseases and probiotic strains that are used as treatment [14].

Disease or Disorder	Examples of probiotic strains
Acute and antibiotic associated	Lactobacillus rhamnosus, L.
diarrhea	bulgaricus, Enterococcus faecium
	W54, B. bifidum W23
Urinary or kidney	Oxalobacter formigenes, L.
stones	acidophilus LA-14.
Constipation	L. bulgaricus, L. acidophilus
	(NCFM), B. lactis
Acute viral	L. acidophilus, B. animalis ssp, L.
respiratory infection	rhamnosus,
Colon cancer	Lactic acid bacteria
Colic	B. breve, L. reuteri, L. casei
Diabetes and Obesity	L. gasseri, L. rhamnosus

Table 2. Probiotics are tested experimentally for the treatment and management of infections caused by foodborne pathogens [14].

Probiotic	Pathogen	Animal Model	Outcomes		
L. acidophilus	Salmonella	Rats	Prevention of		
heat killed	Salmo		Salmonella sp.		
			infection		
L. acidophilus,	E. coli	Sheep	Regular probiotic		
L. plantarum,	O157:		administration to sheep		
E. faecium, L.	H7		for seven weeks		
casei, and L .			considerably reduced		
fermentum			the overall number of		
			pathogens in excreta		
Probiotic E. coli	Salmonella	Turkey	After two weeks of		
Nissle	enteritidis		Salmonella sp.		
1917			exposure, the		
			administration of		
			probiotics dramatically		
			reduced the pathogen		
			load		

Around 1900, Elie Metchnikoff proposed that the healthy and long lives of Bulgarian peasants were due to their use of fermented milk, hence, the concept of probiotics was developed. He postulated throughout his research that the probiotics in yogurt shield the host from the negative effects of other pathogenic microbes [25]. It was identified that there are many lactobacilli strains found in yogurt, which gave them the scientific name *Lactobacillus bulgaricus*. In accordance with his research, the pure cultures of *L. bulgaricus* may be used to make fermented milk products. Metchnikoff proposed that pathogenic bacteria that produce toxins are removed from the gut by *Lactobacilli* [14].

In the 1930s, the first clinical studies examining how well probiotics relieve constipation were conducted. Subsequently, a probiotic product was approved by the United States Department of Agriculture as a therapeutic agent for managing *E. coli*-induced scouring in pigs. A variety of probiotic species have been employed in the prevention and treatment of many diseases such as colon cancer, kidney or urinary stones, constipation, acute diarrhea, obesity, diabetes, etc. Table 1 shows several illnesses with examples of probiotic strains used as a cure [14].

As per the data from the Centers for Disease Control and Prevention, foodborne diseases are responsible for around 48 million cases annually, 128 thousand of them requiring hospitalization with 3 thousand fatalities. With the increasing number of moralities, foodborne diseases are becoming a major threat worldwide [26]. Multiple biological, physical, and chemical methods such as antibiotics, thermal treatment, and preservatives, respectively, were used to diminish foodborne diseases. Nevertheless, these approaches were not effective and resulted in side effects including altering the appearance and organoleptic properties of food, antibiotic resistance in foodborne pathogens, as well as chemical intolerance in the body. Thus, alternatives are needed to manage and monitor foodborne illnesses. As demonstrated in Table 2, several studies have proven experimentally the effectiveness of probiotics as an alternative to preventing infectious diseases [14]. These and other studies along

with the advantages of probiotics opened the door for many opportunities for further applications in medical fields and food industries.

3. Latest Development

In the past decades, the main focus of scientists and food technologists besides understanding the mechanism of action of probiotic bacteria is developing probiotic food and supplements. As a result, consumers have witnessed a significant increase in probiotic-based products which include pharmaceutical probiotic formulations, fermented dairy products, and non-dairy products as illustrated in Table 3.

A few years ago, the majority of the probiotics on the market were dairy based. However, high demand for non-dairy-based products such as fermented fruits and vegetables, herbs and tea, and cereals, has appeared. This is attributed to consumers with allergies to milk and its products and lactose intolerant or owing to dairy products' high cholesterol and saturated fatty acid content [14].

Table 3. Classification of probiotics [14].

Food formulations Probiotic Pharmaceutical formulations	Food	Diary based products	Lassi, Yogurt, Yakult, Cheese, Buttermilk, Kefir, Icecream, Chocolate		
			Vegetables	Pickles	
			Fruits	Jam, Puree	
			Soy and Pulses	Flour, Soy milk, Tempeh	
	formulations	Non- diary- based	Cereals	Koji, Bread, and bakery	
	products	Meat and Fish	Salami, Sausage, Fermented meat		
			Herbs	Kombucha, Tea	
	l t	Tablet			
		Freeze-dried powder			
		Syrup			
		Capsule			

3.1 Microencapsulation

All forms of probiotics including powder, suspensions, capsules, and embedded into food share the same issue of losing their viability upon processing and storing as they contain live bacteria [27]. With the recent explosion of probiotic-based food in the market, ensuring the survival and retention of the probiotics' health-boosting properties throughout the manufacturing process, technological food handling, and storage until the product's expiration date is a crucial precondition for their application. Furthermore, given that probiotics need to be alive and functional when they arrive at their

intended site within the host, it's essential that they possess the ability to resist the host body's natural defenses against ingested bacteria. Microencapsulation, a recently developed highly effective technique, has been shown to improve the survival rate of probiotics during food processing and their journey through the upper parts of the digestive system [28]. It refers to enclosing solids, liquids, or gases within tiny, sealed capsules that can be released at predetermined rates in response to particular conditions. The ultimate goal microencapsulation is to shield the probiotics from unfavorable conditions in the environment. In the food industry, this technique is applied for the following reasons:

- 1. Protection of the probiotic (core) from degradation caused by environmental factors such as temperature, oxygen, light, and moisture.
- 2. Reduction of core material evaporation.
- 3. Improvement of material handling by altering physical characteristics.
- 4. Masking of flavor, color, or unwanted taste.
- 5. Achieving even dispersion upon dilution.

The viability and survival of a probiotic cell that is encapsulated rely on the concentration of the wall material, size of the microcapsule, and type of bacteria strain. The encapsulation process involves enclosing cells in a wall material using various processes such as spray-drying, freeze-drying, emulsification, extrusion. The selection of encapsulation is extremely important. During industrial processing and storage, inadequate encapsulation can expose sensitive probiotics to the external environment, causing cell wall damage and decreased nutritional properties. Commercially, freeze-drying and spry-drying have been successfully employed [29]. Among the encapsulation method, freeze-drying achieved the highest encapsulation efficiency of 98% according to Palanivelu et. al., [30]. Freeze-drying (Figure 3), also known as lyophilization, is a standard method used to maintain probiotics in a dehydrated state for extended periods, however applying to freeze-dry for encapsulation is relatively new [29]. Commonly, water is extracted from probiotic bacterial cells as a standard practice to ensure the stability along the storage period [27].

During World War II, the freeze-drying method was first made commercially available to preserve penicillin and blood plasma. Subsequently, the French virologist Charles Merieux preserved vaccines using the freezedrying technique. In 1938, the scientist Max Mortgenthaler pioneered the development of freezedried coffee, a breakthrough that paved the way for the creation of various powdered food items [27]. Probiotic cells are mixed with an aqueous solution of wall material during the process of this method of microencapsulation.

The mixture is subsequently frozen at low temperatures, and the frozen water is then sublimated under vacuum conditions. The main drawback of this process is the elevated cost and time consumed, and it may damage cell membranes reducing their activity [29]. Despite this, several techniques have been developed to increase the survivability of probiotic cultures using this useful and popular drying process. Pulse-spouted

microwave freeze drying is an example that is capable to decrease drying time [27].

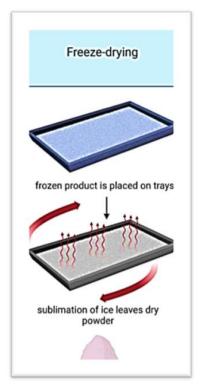


Fig. 3. Schematic of Freeze-drying [27].

3.2 Engineered Probiotics

Although microencapsulation can enhance and protect probiotic microbial strains under harsh conditions, there are other challenges in creating easily accepted probiotic-based foods, determining appropriate doses, and evaluating the effectiveness of different strains. Researchers have recently studied the design of novel probiotics with desired features through genetically modifying probiotics. Genetically engineering bacteria strains can improve their properties, safety, and effectiveness in therapeutic applications [31]. Besides, this process allows further applications like vaccine and drug delivery [32]. There are various applications of genetically modified (GM) probiotics.

One property of probiotics that grabbed the attention antimicrobial of scientists is their activity. Conventionally, infectious diseases are treated using antibiotics. However, antibiotics are known to kill all types of bacteria including the beneficial ones. Moreover, antibiotics can cause adverse side effects at the patient level and allow for the development of antimicrobial-resistant pathogens [18]. Therefore, there is a pressuring need for alternatives in antimicrobial therapy [3], particularly creating novel antimicrobial agents that are risk-free, environmentally benign, and effective against bacteria that are resistant to antibiotics [31]. GM probiotics can generate molecules that are similar to antibiotics. These molecules are called antimicrobial peptides (AMPs) and have antimicrobial action without antibiotic resistance phenomena [33].

Infections caused by *Clostridium difficile* cause traveler's diarrhea which leads to severe clinical issues. In the last decade, probiotics have been proposed probiotic as an effective approach to treating it instead of using antibiotics [34].

Furthermore, engineered probiotics offer anticancer therapeutics. Uncontrolled cell division, or cancer, is usually discovered only when it has progressed to an advanced stage. Once cancerous cells have spread, they eventually require several developmental stages for a tumor to become malignant [35]. According to the World Health Organization (World Health Organization, 2020), it is predicted that by 2040 cancer cases will reach around 30 million. Cancer is typically treated by chemotherapy and multiple drugs supplying. These treatments are found to damage both cancer cells and healthy cells, leading to resistance that affects future treatments of patients [35]. Recently, engineered probiotics have been found to target and replicate within tumor cells, offering a safer and potentially more effective alternative to traditional cancer treatments. Specifically, certain bacteria like Clostridia sp. and Salmonella sp. have been found to be helpful in treating cancer [31]. The use of these probiotics could also help combat drug resistance and reduce the side effects of chemotherapy. Furthermore, non-pathogenic strains, such as E. coli, have the ability to multiply within tumors and nearby tissues, making them valuable in treating melanomas (associated with skin cancer) [36]. As scientists continue to study the mechanisms of these bacteria, it may be possible to explore various combinations of probiotics to treat different types of cancer.

4. Current Challenges and Future Prospects

Despite the facts associated with the health benefits of probiotics, there are still a number of obstacles that could prevent their widespread use [37]. Maintaining probiotics' viability in edible products at the time of consumption and improving bioavailability in the human gut are the two most difficult probiotic delivery steps. Making up food matrices augmented with probiotics is still an interesting research topic, especially regarding their interaction with carbohydrates, proteins, and flavoring agents as they have been shown to alter or change probiotics' efficacy and capability. As much literature has been developed over the years, this is still the case [38]. Although microencapsulation is utilized to assure probiotics' viability during the manufacture, treatment, and storage of food items, it faces some challenges such as developing microencapsulation equipment, creating beads or capsules from polymers, choosing non-toxic materials for encapsulation, and figuring out the proper mechanism for probiotic release. Cost is one of the significant difficulties since finished items that have been encapsulated can be quite expensive. This is due to the requirement of financial resources and time for their development [39].

Even though clinical trials provide us with significant insights, the precise ways in which bacterial probiotics

work and their effectiveness in easing different health conditions still pose numerous unanswered questions. In the future, further clinical study is thus required, as foods containing bacterial probiotics have the potential to minimize drug consumption and prevent specific diseases when combined with a balanced diet and lifestyle. Also, as the majority of microencapsulated have been applied to fermented foods, particularly dairy products, more study is required as they can be incorporated into non-diary food. As a result, the food business as well as research in the fields of nutrition, health, and food science are presented with tremendous potential [35]. Future research should also focus on understanding the long-term safety and gastrointestinal tolerance of products containing probiotics, as well as the illnesses that can be caused by certain probiotics, such as opportunistic pathogens like Lactobacilli as such information is limited especially in new-borns [39].

There is no doubt that probiotics have various health claims. It is known that premarket safety approval of drugs is necessary; Nonetheless, such requirements are still not mandated for nutritional and dietary supplements. For instance, in the United States, nutritional supplements (capsules or tablets) do not fall under the same manufacturing and quality control testing requirements as medications [40]. As a result, both consumers and healthcare providers lack assurance about the quality standards of probiotic products as well as the safety of their constituents [31,40]. To address probiotic concerns, such as safety, effectiveness, claims, and labeling, it is recommended that probiotic products be given worldwide regulatory status. Moreover, the probiotic formulations that have been demonstrated to provide the host with specific health benefits must describe these benefits in detail, along with an efficient control system for evaluating any negative side effects and tracking the probiotic formulations' overall health benefits. Genetically modified probiotics necessitate a strict safety protocol to prevent the spread of strains and genetic alterations.

5. Conclusion

Probiotics have gained more attention in the food and feed sectors over the past several decades as a result of rising consumer demand for wholesome and functional meals, or foods that do more than just provide essential nutrients for survival. Because of their well-established functional probiotic qualities, LAB strains are the most commonly used probiotics. Nevertheless, there is still a need to find new probiotic strains with superior healthpromoting properties compared to the current ones. A plethora of advantages are associated with encapsulation when it comes to preserving live microorganisms. The functional components can be encapsulated using a variety of techniques. The greatest encapsulation efficiency was achieved using conventional freezedrying methods. Genetically modified probiotics are a promising and novel option for treating infections. Engineered probiotics have the potential to restore health effectively, easily, and with site specificity. Eventually, a detailed safety assessment should be carried out from production reaching the end user consumption.

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