

Role of lactobacillus probiotics in gut health and food safety: Mechanisms for contaminant reduction and applications in food products

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a. University of Central Punjab, Avenue 1 Khayaban-e-Jinnah, Pir Mansur Johar Town, Lahore, Punjab, Pakistan **Abstract:** Lactobacillus probiotics, live bacteria that confer health benefits when administered in adequate amounts, have gained widespread acceptance, particularly due to their potential to influence and enhance general health by modulating the gut microbiota. In recent years, probiotics have been highlighted for their potential therapeutic applications in preventing and mitigating various diseases. Among these, *Lactobacillus spp.* represent one of the most extensively studied and utilized probiotic strains. This review explores the role of Lactobacillus in maintaining gut microbiota balance, producing bioactive metabolites such as short-chain fatty acids (SCFAs) and bacteriocins, and its involvement in food safety through the reduction of contaminants. Specifically, *Lactobacillus spp.* contribute to food safety by adsorbing, enzymatically degrading, or neutralizing harmful substances, including aflatoxins and heavy metals, thereby reducing their bioavailability and toxicity. The paper also discusses the practical applications of Lactobacillus-based probiotics in the food industry and the ongoing efforts to address associated safety concerns. Lactic acid bacteria provide a natural, non-chemical approach to food systems promotes a safer and more sustainable food supply. However, further research is required to optimize their applications and ensure their efficacy in diverse food matrices.

Key words: Lactic acid bacteria, Probiotics, *Lactobacillus casei*, Food contaminants, *Streptococcus thermophiles*, Food industry

1. Introduction

Lactic acid bacteria (LAB) are a group of beneficial microorganisms widely known for their ability to ferment sugars into lactic acid. LAB belongs predominantly to the phylum Firmicutes and is classified as Gram-positive, non-sporulating, and either rod or cocci-shaped (Miranda et al., 2021). They thrive in anaerobic or microaerophilic conditions, meaning they grow best in environments with little or no oxygen (T, 2018a). LAB is important in food preservation and contributes to fermented products flavor, texture, and safety. By producing lactic acid, they lower the pH of their environment, inhibiting the growth of spoilage microorganisms and pathogens. This acidification process is crucial to produce yogurt, cheese, sauerkraut, kimchi, pickles, sourdough bread, and various fermented beverages. LAB is also significant in producing probiotics, contributing to gut health by promoting a balanced intestinal microbiota (Pérez-Rivero and López-Gómez, 2023). Lactobacillus strains are amongst the most studied probiotics, given their diverse species and strains with distinct characteristics and functions. The beneficial effects of these strains include immune function reinforcement and decreased incidence of gastrointestinal illnesses, in addition to the beneficial impacts on digestion and nutrient absorption (Soomoro et al., 2022). One of the keyways that Lactobacillus demonstrates its probiotic effects is via its promotion of healthy microbiota in the gut (Soomoro et al., 2022). Lactobacillus use probiotic benefits to influence and modulate the gut microbiota, leading to beneficial impacts on human health such as inhibition of colonization and growth of pathogens in the gastrointestinal tract, production of antimicrobial compounds that have proven to be effective, promotion of cytokine production, and improvement of intestinal barrier integrity. The gut microbiota composition and function may be disrupted in connection with various gastrointestinal conditions and systemic diseases (Collado et al., 2007a). Probiotics, specifically Lactobacillus probiotics, have been studied intensively for their effect on influencing the gut microbiota and resulting health benefits to the host. This review focuses on the diverse habitats in which Lactobacillus species can be found, including the human gastrointestinal tract, dairy products, and fermented

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Corresponding email: <u>mohsin.barq@yahoo.com</u> (Mohsin Gulzar Barq) DOI: 10.61363/mapd4e52 products. Probiotic refers to live bacteria that have health-promoting properties to the host, and Lactobacillus is a significant probiotic genus. The beneficial potential of probiotics for health maintenance has been largely studied. *Lactobacillus* species exhibit several important characteristics that make them attractive probiotic candidates (Figure 1). Firstly, Lactobacillus species are present in fermented foods and as dietary supplements and are considered safe for human consumption.

They are a normal part of the human gastrointestinal microbiota, and their presence is associated with a healthy digestive system (Marcelino, 2013). Secondly, the survival and functionality of Lactobacillus species are ensured by their ability to withstand the extremely acidic environment of the stomach and reach the intestines in a viable form. The resistance of these bacteria is attributed to their strong cell walls and ability to tolerate acid. Thirdly, *Lactobacillus* species have the remarkable ability to attach to intestinal epithelial cells which allows them to colonize and improve the gut environment (Khatoon et al., 2023). This allows *Lactobacillus* species to outcompete pathogenic bacteria and prevent their colonization. Food contamination remains a critical issue globally, arising from microbial agents such as *Escherichia coli, Salmonella*, and *Listeria*, as well as chemical contaminants like heavy metals, pesticides, and industrial toxins (Hussain and Gooneratne, 2017). These contaminants compromise food safety and have severe public health implications. The consumption of contaminated food is one of the leading causes of foodborne diseases, which affect millions globally each year, leading to significant morbidity, mortality, and economic losses (Di Stefano and Avellone, 2014). Food contaminants can enter the food supply from farm to fork, through sources like air, water, soil, packaging materials, processing equipment, and intentional adulteration (Hussain and Gooneratne, 2017). With the global impact of foodborne diseases rising, preventative strategies are increasingly focused on reducing contamination risks (Rather et al., 2017).

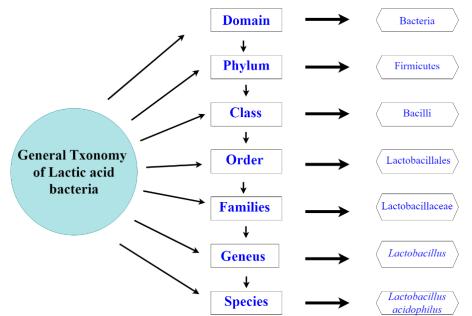


Figure 1. This shows the general taxonomy and key characteristics of Lactic acid bacteria

Traditional food preservation methods, though effective, have limitations in fully addressing these challenges. As a result, LAB and probiotics have emerged as promising tools in combating microbial contamination and degrading certain harmful chemicals in food. This review aims to explore the reduction potential of LAB and probiotics in eliminating food contaminants and to highlight their applications across various food products. By understanding their mechanisms of action and the diverse ways they can be integrated into the food supply chain, this review will underscore the role of these microorganisms in improving food safety and contributing to public health. Finally, the extensive variety of metabolic activities of Lactobacillus species, including the enzymes, and anti-microbial compounds that facilitate their antibacterial and immune-modulatory activities (Mezaini et al., 2013). These features collectively give Lactobacillus species high therapeutic potential as probiotics, and, as such, are highly regarded for their capacity to promote gut health and well-being.



2. Contaminants in food

Food contaminants present a serious global public health challenge, with biological, chemical, and physical contaminants contributing to various health risks. Preventing contamination at all stages of the food supply chain farming, processing, packaging, and storage is essential to ensure food safety. Regulatory bodies, such as the FDA (Food and Drug Administration) and WHO (World Health Organization), play a crucial role in monitoring and enforcing food safety standards to mitigate these risks. Moreover, consumers must practice safe food handling, cooking, and storage techniques to reduce the risk of contaminant exposure further (Table 1).

Table 1. This shows the contamination rate in different food products

Products	Contamination Rate	References
Ricotta	86.2 %	(Béjaoui et al., 2022)
Cheese	69.6 %	(Béjaoui et al., 2022; Serpe et al., 1999)
Raw milk	72.5 %	(Béjaoui et al., 2022)

2.1 Contamination by bacteria

Threats due to bacterial species are called Brucellosis which causes a foodborne disease in humans sometimes may be mild or chronic depending upon the severity of the infection. It mainly Spreads through animals or by consuming infected meats or products. Dairy products mainly ricotta, cheese, or raw milk may be contaminated by Brucella spp. like B. sius, B. melitensis and B. abortus. The well-affected areas due to Brucellosis are rural areas due to touching infected animals directly while people living in urban areas may ingest unpasteurized dairy products. The ingestion of contaminated products may be fatal and pose meningitis, endocarditis, or hepatitis (Béjaoui et al., 2022; Serpe et al., 1999). Illness caused by *Escherichia* produces Shiga toxin, which is present in fermented dairy products. Thus, causing foodborne diseases, especially in products like Kashk and Doogh. (Dehkordi et al., 2014). From bulk farm milk, market milk, fresh soft cheese, kareish cheese, and ice cream *Salmonella* spp. Can contaminate products mostly *S. typhimurium. S. enteritides* and *S. infantis* present (Omar et al., 2018). Two common species i.e. *S. Typhimurium & S. enteritides* pathogens of milk and its products mostly in raw sheep milk (Shaigan Nia et al., 2014).

According to several studies, in dairy products like cheese and milk (liquid), Gram-negative bacteria i.e. coliforms are mainly present as a contaminant (Hervert et al., 2017). Paenibacillus or Bacillus causes spoilage in milk, and it is tough to remove because it possesses characteristics of endospore-forming (Huck et al., 2007). Other pathogens that affect milk are Brucella abortus, Campylobacter, Clostridium, E. coli, Salmonella, Mycobacteria, and Streptococcus agalactiae (Msalya, 2017). Listeria monocytogenes also contaminates milk products. Cold raw milk favors the growth of these bacteria (Kabuki et al., 2004). Besides the presence of Gram-negative bacteria, Gram-positive aerobic bacteria are also present like Staphylococcus intermedius, Staphylococcus aureus, Streptococcus agalactiae, and Staphylococcus epidermis (Mdegela et al., 2004). Yogurt also becomes contaminated by these Gram-negative bacteria i.e. Enterobacteriaceae or also through E. coli. Through EB testing we can know whether yogurt is hygienic or not (Hervert et al., 2017). One of the common spoilage microorganisms Enterobacter cloacae, was likely to survive in cow milk. In soymilk Pseudomonas paucimobilis present and survive in yogurt (Canganella et al., 1999). Anaerobic facultative spoilage microbe i.e. Listeria monocytogenes present and cause the disease Listeriosis in humans which greatly affects the liver & spleen (Yang & Yoon, 2022). Grampositive bacteria i.e., Listeria monocytogenes contaminate the product, especially in fresh cheese processing plants, and harms humans' health if consumed. Also, Pseudomonas, Salmonella spp, E. coli, and Bacillus cereus are present (Al-Gamal et al., 2019). Cheese production from raw milk was mostly used, and common spoilage microbes are Verotoxigenic E. coli, S. aureus, and Salmonella (Table 2) (Costanzo et al., 2020).

Table 2. Food-borne bacteria and lactic acid	bacteria against them
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Pathogenic Bacteria	How LAB fights against them
Listeria monocytogenes	LAB produces bacteriocins like nisin and organic acids that lower the pH, making it difficult for Listeria to survive.

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Salmonella spp.	They create organic acids and compete for nutrients, effectively lowering the pH and limiting <i>Salmonella</i> 's growth.
Escherichia coli O157	LAB produces organic acids, hydrogen peroxide,
Staphylococcus aureus	and bacteriocins, all of which help to combat E. coli. They generate reuterin, along with organic acids
	and bacteriocins, to inhibit Staphylococcus growth.
Clostridium perfringens	LAB produces organic acids and bacteriocins that
	help reduce the presence of <i>Clostridium</i> in food
Bacillus cereus	By producing bacteriocins and lowering the pH,
	LAB can inhibit the growth and spore formation of
	Bacillus.

2.2. Contamination by Fungi

In fermented products highly, toxigenic molds are present which represents mycotoxins of them. Airborne infection, improper use of equipment, or handling refers to a source of contamination in products (Pei et al., 2021). It can be affected by Aflatoxin especially when milk is in liquid form which is produced by the species *Aspergillus flavus* and causes a contagious effect on the health of humans (Darsanaki, 2014). Dairy products may be affected by it even after passing through pasteurization. Collection of milk safely, storage, and temperature level are important in protecting products free from MI (AFM1) (Figure 2).

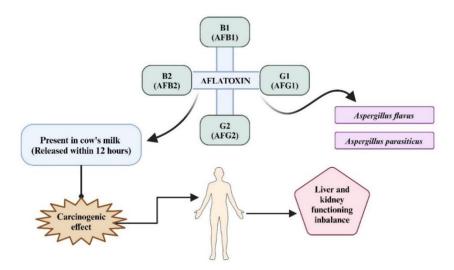


Figure 2. Types of aflatoxin and their impact on the Human Body

However due to its growth rate at low temperatures, fermentation process between product ingredients like lactose and sucrose, hydrolysis of protein, or formation of lactic acid in products these characteristics may lead to the formation of aflatoxin. Another species *Aspergillus parasiticus* also has a carcinogenic effect on the liver, kidney, and its functioning. Different applications are applied to remove this but cannot give 100 % accuracy. In aflatoxin compound M1 (AFM1) causes dangerous effects on the health of humans (Bayat, 2021). Aflatoxin sometimes produces mycotoxin which is referred to as ochratoxin which has hazardous effects not only on humans but also on animals (Ogheneoruese Onoharigho et al., 2022). Yeast and mucor are also present in it (Bastam et al., 2021a). Some other species are also important according to a study conducted in Isfahan shows that *Acremonium* and *Cladosporium* are present which contaminate milk and its related products (Table 3) (Fallahi, 2014).



Product	Fungal Isolates	Contamination Rate	Refereences
Peanut butter	Helmintosporium	47.44%	(Zamblé and B. O. L. I,
	Penicillium	41.54%	2016)
	Aspergillus sp	20.22-51.65%	
Cooking butter	Aspergillus	2.8%	(Ismail, 2001)
U U	Geotrichum	78%	(Zamblé and B. O. L. I,
	Penicillium	3.9%	2016)
	Mycosphaerella	1.4%	<i>`</i>

It may be affected by spoilage due to yeast which serves as a topmost contaminant in it. As yogurt is stored at low temperatures for its proper formation and its acidic environment yeast is more likely to grow especially *Saccharomyces* or *Candida kluveromyces* (Al-Gamal et al., 2019). Common spoilage microorganisms that are involved in the spoilage of yogurt are *Penicillium monilia, Alternaria, Rhizopus, Micelia sterilis,* and *Cladsporium*. Contamination through air is very common and it makes products spoil especially through *Penicillium* or *Clostridium*. For this purpose, certain food additives like Potassium sorbate added in it. Bio preservatives enhance product quality as well as increase its life span. Different LABs are used to make fruits, vegetables, or dairy products free from contaminants (Buehler et al., 2018). Contaminants that affect butter mainly molds or yeast change the color of the product and make its flavor bitter (El-shafe et al., 2017). Butter is mainly spoiled by *Penicillium spp.* and *Cladosporiodes spp.* Butter is mainly affected by *Geotrichum candidum* and about 78% of it constitute cooking butter (Ismail, et al., 2001). Yeast caused spoilage in cheese. Moreover, it changes its texture, smell, color, and taste etc (El-shafe et al., 2017). Depending upon the type of cheese various microorganisms spoil it like soft cheese *Aspergillus, Mucor*, and *Penicillium* cause spoilage in this product. Cheddar cheese was commonly affected by *Aspergillus, Penicillium*, or *Fusarum* (Table 4) (Ismail, 2001).

Product Name	Countries	Common Spoilage Microorganisms	References
Processed Cheese	Egypt	Aspergillus	(Ismail, 2001)
		Penicillium	
Hard Cheese	Egypt	Rhizopus	(Ismail, 2001)
		Penicillium	
		Aspergillus	
		Geotrichum	
		Candida	
		Other yeast	
Cheese	New Zealand	Penicillium commune	(Ismail, 2001)
	and	P. requefortii	
	Australia	P. chrysogenum	
		P. expansum	
		P. solitum	
		P. viridicatum	
	_	P. brevicompatum	
Packaged Cheese	Europe	P. commune	(Ismail, 2001)
		P. verrucosum	
		P. solitum	
		P. roquefortii	
		P. nalgiovense	
Cheddar Cheese	Egypt	Aspergillus	(Ismail, 2001)
	07 Г	Penicillium	()
Soft Cheese	Egypt	Candida	(Ismail, 2001)
	OJ I	Penicillium	<pre></pre>

Table 4. shows the different types of fungal species that spoil the different kinds of cheeses.

3. Reduction mechanism of bacterial contaminants in food

The general aim of shelf-life extension is to keep foods safe and stable. Generally, this can be achieved by controlling the growth of spoilage microorganisms and pathogenic bacteria. In treating and controlling pathogenic growth, an antimicrobial agent, such as nisin, or two or more antimicrobial agents could be used synergistically against the target organism. The majority of reported foodborne illnesses are caused by pathogenic bacteria such as *Campylobacter jejuni, S. aureus, L. monocytogenes, E. coli*, and *Salmonella spp*. Consumer demand for minimally processed foods has obligated the food industry to search for new methods of ensuring food safety. It can no longer rely on traditional heat treatment methods to create microbiologically safe foods. On the other hand, fruits, and vegetables, have been found to harbor pathogenic bacteria. LAB may enhance the nutritional quality of food and contribute to intestinal health due to the production of antimicrobial agents. There are various mechanisms for foodborne pathogens prevention and elimination of food spoilage bacteria, like the production of antimicrobial substances that may prevent adherence of pathogens to epithelial and mucosal surfaces. LAB produces different compounds that help to reduce the contamination rate by bacterial species in food. It includes organic acids, bacteriocin production, hydrogen peroxide production, competitive exclusion, and Stimulation of Host Defenses (in fermented foods).

LABs are also able to inhibit the fungi (molds) responsible for food contamination and mycotoxin production. LABs contain bacteriocin-like substances and produce organic acids with typical fungistatic and fungicidal properties and inhibit fungi and yeast such as *Aspergillus versicolor*, *Penicillium expansum*, *Fusarium culmorum*, *Candida parapsilosis*, *Aspergillus niger*, and *Penicillium chrysogenum*. Some species of the several genera of fungi produce mycotoxins which are *Aspergillus*, *Fusarium*, *Penicillium*, and *Alternaria*. These fungi genera produce different Mycotoxins, including aflatoxins, fumonisins, ochratoxin, patulin, tricothecenes, and zearalenone, are derived as secondary metabolites. These metabolites have potential carcinogenic, teratogenic, immunotoxic, neurotoxic, hepatotoxic, and nephrotoxic effects. Among approximately 400 compounds identified as mycotoxins, 30 of them are considered dangerous to human health (Bayat, 2021). Thus, myco-toxin contamination should be controlled either by preventing their formation or by detoxification. The control of mycotoxins is more difficult because the normal cooking method cannot destroy all of them. Therefore, food processing techniques are necessary to remove mycotoxins. LABs (*Lacticaseibacillus casei* and *Limosilactobacillus reuteri*) are known to efficiently bind toxins Such toxins include AFs (aflatoxin) in aqueous solutions. Thus, the control of these microbes using LAB is a natural way of food preservation (Figure 3).

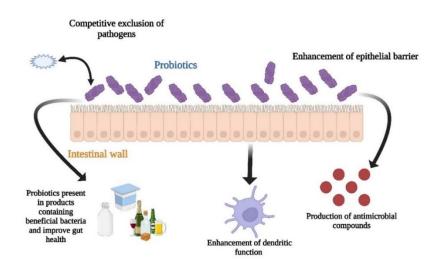


Figure 3. Mechanism of reduction of contaminants with Lactobacillus and their role in food products.

3.1 Production of organic acid

Lactic acid bacteria (LAB) play a crucial role in keeping food safe using several natural mechanisms. They produce compounds like lactic acid, acetic acid, and reuterin which lower the pH of the food making it harder for harmful bacteria to grow. LAB also competes with pathogens for nutrients quickly consuming what is available and creating an acidic environment that further stops the growth of harmful microbes. Additionally,



they can break down glycerol releasing antimicrobial substances that help preserve food. Beyond just keeping food safe LAB also improves the taste and extends the shelf life acting as a natural defense against spoilage. The wide variety of LAB strains brings an even broader range of protective benefits, making them highly effective in preventing foodborne illness (Gänzle, 2015).

Lactic acid bacteria (LAB) play an important role in protecting food from various harmful food-borne bacteria such as *Listeria monocytogenes* and *Staphylococcus aureus*. LAB also excels at competing for nutrients, quickly using up available resources and producing organic acids that inhibit the growth of harmful bacteria like *Salmonella spp*. and *Escherichia coli* O157:H7 (Cálix-Lara et al., 2014). Additionally, they release hydrogen peroxide and other antimicrobial compounds that further strengthen their protective effects. The lowered pH disrupts bacterial cell membranes, inhibits enzyme activity, and reduces the growth of spoilage organisms and pathogens like *Escherichia coli*, *Salmonella*, and *Listeria*.

3.2 Production of bacteriocin

These LAB-produced bacteriocins are antimicrobial peptides that can inhibit many other bacteria, such as pathogens and spoilage organisms. These include *Lactobacillus, Lactococcus, Streptococcus,* and *Enterococcus,* among the various species of LAB, which spontaneously produce bacteriocins as part of a defense mechanism for competition in the gut or food products. Of specific interest are these bacteriocins due to their potential application as natural preservatives for foods and as alternatives to antibiotics (Parada et al., 2007). There are two major classes of bacteriocin Class I bacteriocin is called Lactococcin are small heat-stable peptides that don't require post-translation modifications. When LAB produces this type of bacteriocin it directly targets inhibiting the peptidoglycan synthesis of other contaminants present in food. In this way, contamination reduction can easily performed. The other class of bacteriocin is Class II bacteriocin which is called Lantibiotics which function by directly destabilizing the cytoplasmic membrane of the contaminants present in food with the help of the creation of pores that allow bacteriocin molecules to enter into the bacteria and the cells death. In this way, microbial contamination can easily be reduced by the help of different bacteriocins (Table 5) (Ibrahim et al., 2021).

Mechanism	Description
Production of Antimicrobial Compounds	LAB naturally produces substances like lactic acid, acetic acid, and reuterin, which lower the pH of food and help inhibit the growth of harmful bacteria.
Competitive Exclusion	LAB compete with harmful bacteria by quickly using up available nutrients, producing acids that create an environment where pathogens struggle to survive.
Utilization of Glycerol	LAB can break down glycerol, releasing antimicrobial compounds in the process. This helps to preserve food and keep it free of harmful microbes.
Impact on Food Quality and Safety	The metabolic activities of LAB not only help prevent the growth of pathogens but also enhance the taste and shelf life of food products.
Diversity of LAB	The metabolic activities of LAB not only help prevent the growth of pathogens but also enhance the taste and shelf life of food products.

 Table 5. Mechanisms of lactic acid bacteria in enhancing food safety and quality

4. Enhancing Food Products with Lactic Acid Bacteria

Lactic acid bacteria (LAB) especially Lactobacillus, play a big role in making food healthier, tastier, and safer. LAB is essential for the process of fermentation where they turn sugars into lactic acid. This gives food a unique flavor and texture and naturally preserves it by lowering the pH which stops harmful bacteria from growing. This process helps extend the shelf life of many foods while keeping them safe to eat. LAB helps your body to absorb nutrients more easily. For instance, dairy products break down lactose which means people who are lactose intolerant can enjoy milk and yogurt without discomfort. So, they make these foods more accessible and easier to digest (Al-Gamal et al., 2019).

Lactobacillus strains are known for their probiotic benefits. They help keep the balance of good bacteria in your gut which is important for digestion and immune health. Adding these probiotics to foods like yogurt or fermented veggies offers extra health perks. Additionally, LAB produces natural substances that protect food from spoilage. By creating antimicrobial compounds, they prevent the growth of harmful bacteria and fungi making food safer and helping to avoid foodborne illnesses. These helpful bacteria also make food taste and feel better. The flavors in fermented foods like yogurt, cheese, and pickles give them their signature tang and texture that people love. These probiotics support digestion, strengthen immunity, and even have anti-inflammatory effects making them a favorite for health-conscious consumers (Zhang et al., 2018).

5. The Role of *Lactobacillus* Strains in Health and Disease Prevention

Lactobacillus strains that is a key group of probiotics have demonstrated significant health benefits particularly in promoting gut health and preventing various diseases.

5.1 Lactobacillus salivarius CECT 5713 and Digestive Health

This strain plays a crucial role in preventing digestive issues, such as indigestion. Studies have shown that infants who were fed formula containing Lactobacillus salivarius for up to six months experienced a notable increase in fecal lactobacilli (Maldonado et al., 2010). This boost in beneficial gut bacteria helps maintain a balanced microbiome which is essential for proper digestion and overall gut health.

5.2 Lactobacillus rhamnosus GG and Infection Prevention

Lactobacillus rhamnosus GG is widely used in fermented milk products and has been shown to reduce the risk of respiratory and gastrointestinal infections prevalent in developing countries affecting 5-44% of the population. Regular consumption of products containing this probiotic strain can significantly lower the incidence of such infections by supporting the immune system and maintaining gut integrity (Hojsak et al., 2010).

5.3 Lactobacillus acidophilus and Lactose Intolerance

Lactobacillus acidophilus is particularly effective in enhancing lactose utilization in individuals who are lactose intolerant. These individuals lack lactase, the enzyme responsible for breaking down lactose often leading to symptoms like diarrhea and abdominal discomfort. By consuming *L. acidophilus*-rich products lactose digestion is improved. (Kim & Gilliland, 1983).

5.4 Yogurt and Gut Health

Lactobacillus casei is commonly used to treat diarrhea, especially in children. Fermented yogurt containing *L. casei* has been found to significantly improve gut health by restoring the balance of beneficial bacteria in the gut, thus reducing the severity and duration of diarrhea. (PEREG et al., 2005).

5.5 Gut Microbiota and Protection Against Harmful Bacteria

Probiotics such as *Lactobacillus casei, L. acidophilus,* and *Bifidobacterium* strains, naturally present in the human intestine contribute to maintaining a healthy gut environment. These strains produce organic acids and bacteriocins, which inhibit the growth of harmful bacteria by creating an inhospitable environment for pathogens. This antimicrobial activity is essential for promoting a healthy gut and preventing infections (Figure 4) (Mazahreh et al., 2009).



5.6 Fermented Beverages: Their Role in Health and Production

Fermented beverages have gained popularity due to their health benefits and are commonly produced by introducing lactic acid bacteria (LAB) or yeast during fermentation. These beverages not only improve sensory qualities like flavor and texture but also serve as a source of probiotics that enhance gut health. Lactic acid beverages are primarily prepared using milk or dairy products. Key bacterial strains involved in their production include Lactobacillus acidophilus, Lactobacillus rhamnosus, and Bifidobacterium animalis. Research indicates that these beverages contain around 51% lactic acid, contributing to their distinct taste and health benefits (Almeida et al., 2009). Factors such as the physical, sensory, and chemical properties of the beverage are crucial in optimizing production and enhancing quality. These drinks are a rich source of live microorganisms that can colonize the human gut helping prevent diseases by promoting a balanced microbiome and improving digestive health. Dairy-based fermented beverages typically consist of a mixture of whey and milk (usually from buffalo or cows). Strains like Lactobacillus bulgaricus and Lactobacillus acidophilus are commonly used in their production. Whey, a by-product of cheese production has a reputation for promoting health due to its high nutrient content, including calcium (Ca), iron (Fe), magnesium (Mg), and phosphorus (P), while maintaining low cholesterol levels. However, improper disposal of whey from the dairy industry can pose environmental hazards. These dairy beverages are particularly beneficial for treating infections caused by gut microbiome imbalances further highlighting their probiotic properties (Simões da Silva et al., 2020). Recent studies have explored the combination of fruit juices and whey to create probiotic beverages (Kidist Fikre Worku et al., 2019). For example, pineapple juice mixed with whey in a 65:35 ratio, inoculated with 1% Lactobacillus acidophilus, has been shown to promote the growth of beneficial gut bacteria. Additionally, Lactobacillus plantarum is known to enhance gut health when used in fermented fruit beverages. A notable example is the Cornelian cherry, which possesses unique antimicrobial and antimalarial properties making it a valuable ingredient in probiotic drinks (Mantzourani et al., 2018).

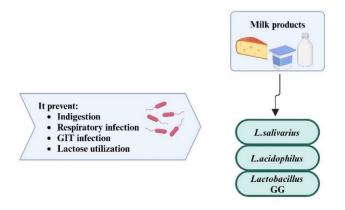


Figure 4. Role of Lactobacillus in milk products and enhancing human health.

6. Industrial applications of LAB

LABs have many modern applications because of their exceptional metabolic properties, security, and capacity to flourish in different conditions. In the emerging field of biotechnology, LABs have arisen as an overlooked yet truly great individual using their flexible capacities across modern applications. From the development of matured food varieties to the pharmaceutical industry and then some to the textile industry, these have proved to an excellent microorganism in every industrial application. They are broadly utilized as starter societies in the maturation of dairy items like cheddar, yogurt, and aged milk, as well as in meat, fish, organic products, vegetables, and cereal products. LAB contributes altogether to the flavor, surface, and healthy benefits of aged food varieties, going about as assistant societies and speeding up cheddar (cheese) development. Additionally, they produce bacteriocins and antifungal mixtures, prompting the use of bio-defensive societies in food preservation (Miranda et al., 2021b; Zapaśnik et al., 2022).

6.1 Commercial use of LAB in Food industry

Many researchers have reported the good potential of LAB on humans or animals when they consume fermented food that then shows good benefits on their health. The fermentation processes, bio-synthetic ability, and metabolic activities are the key elements that work with the use of LAB as microbial starters for creating, delivering, as well as expanding explicit gainful mixtures in aged (fermented) food (López and Spano, 2023). LAB ferments carbohydrates to Lactic acid, so it is mostly used in the fermented food industry. There are several commercial uses of LAB in specific food industries in different processes.

6.1.1 Versatility in food fermentation

LAB plays a crucial role in dairy and non-dairy food products, contributing to their fermentation, flavor, and potential health benefits. These bacteria commonly found in fermented dairy foods, include genera like Lactobacillus, Lactococcus, etc. LAB strains isolated from dairy environments have shown protection from low pH, bile salts, and recreated gastric and gastrointestinal circumstances, along with beneficial enzymatic activities and genes that indicate probiotic potential (Coelho et al., 2022a; Colombo et al., 2018). LAB in dairy products can enhance lactose digestion, stimulate the immune system, and even stop the growth rate of pathogens like Staphylococcus aureus, Escherichia coli, and Salmonella typhimurium (Ağagündüz et al., 2021). Their presence in dairy products contributes to the rich flavor, texture, and potential health-promoting effects associated with these products. (Coelho et al., 2022a). LAB additionally has well-being-advancing properties, making them significant as probiotic societies in the food industry (Zapaśnik et al., 2022). Furthermore, LAB exhibits antimicrobial action against foodborne microorganisms, adding to sanitation by hindering the development of destructive microbes, yeasts, and growths through the creation of natural acids like lactic acid, acidic acid, and propionic acid (Wang et al., 2021). Their capacity to kill mycotoxins and produce metabolites, for example, bacteriocins further upgrades their job in food preservation (Zapaśnik et al., 2022). Generally, LAB's metabolic qualities and antimicrobial properties make them fundamental for further developing the taste, surface, security, and period of usability of different food items in the business.

6.1.2 Yogurt and Cheese

One of the most important products of the dairy industry is Yogurt which is the best example of a fermented product in which LAB is used and gives us the best form of yogurt (Chen et al., 2017). The rate of consumption of yogurt is very high in the whole world. It helps to improve our gut and health. It is delivered through the fermentation of milk by unambiguous types of microorganisms, principally Lactobacillus bulgaricus and Streptococcus thermophiles, albeit extra probiotic microbes may likewise be incorporated relying upon the ideal qualities of the yogurt (Colombo et al., 2018) (Mazahreh et al., 2009). During fermentation, these microbes convert lactose, the regular sugar present in milk, into lactic acid. This fermentation cycle makes the milk proteins coagulate, bringing about the thick, smooth surface attribute of yogurt. The aging likewise adds to the tart flavor and particular smell of yogurt (T, 2018a). Yogurt is valued for its taste and surface as well as for its various medical advantages. It is a rich wellspring of fundamental supplements like calcium, protein, nutrients B6 and B12, riboflavin, and potassium. National Yogurt Association rules characterize the dynamic culture yogurt as a result that contains live (LAB) microbes in sum > 108 cells/g toward the end season of the production (Mazahreh et al., 2009). Moreover, the live microscopic organisms' societies present in vogurt, frequently alluded to as probiotics, can assist with keeping a good arrangement of stomach microorganisms, help in processing, support resistant capability, and possibly mitigate side effects of lactose bigotry. Limited research on humans concerning the gut-brain axis focuses on LAB which is linked to brain functioning, psychology, and immunology. For instance, fermented milk products containing specific LAB strains improved cognitive performance and modulated immune responses in different populations. In milk fermentation, the Lacticaseibacillus case DN-114001 strain of LAB which is mostly present in yogurt helps to improve our immune system (Marcos et al., 2004). While consumption of unpasteurized milk increased Lactobacillus levels and influenced microbiome profile, no direct link between LAB amount and mental or psychological measures has been established, indicating a need for further research on the direct effects of dairy LAB on the gut-brain axis (Table 6) (Ağagündüz et al., 2021;Butler et al., 2020).

Table 6. Shows the different parameters on which Lactic acid bacteria isolates grow and the number of bacterial isolates present in different dairy food products.



Industrial product Fermented dairy products	LAB strains	Salt concentration, temperature, and pH	Bacterial amount in the end product	References
Yogurt	Lactobacillus bulgaricus Streptococcus thermophiles Lactobacillus Acidophilus Bifidobacterium Lactobacillus casei	Tolerating environments with a low pH 2% or eventually 4% of salt, 42°C is the ideal growth temperature for a starter culture	About > 108 cells/g	(Colombo et al., 2018; Marcelino, 2013) (Mazahreh et al., 2009) (Hosken et al., 2023)
Cheese	Lactococcus lactis S. salivarius subsp. Thermophilus Lb.helveticus, Lb.delbrueckii subsp Delbrueckii subsp. bulgaricus Ln. lactic Ln. cremoris S. salivarius subsp. Thermophilus	The temperature for cheese ripening is 15°C pH is about neutral, high activity of water Good source of nutritional The mesophilic starter culture temperature is 30 °C and 40 and 45 °C is for Thermo tolerant species	2.3×10 ⁷ CFU per gram.	(Hosken et al., 2023) (Marcelino, 2013) (Coelho et al., 2022b; Nicosia et al., 2023)
Kefir	LAB strains: Lactobacillus kefiranofaciens, Lactobacillus kefir, Lactobacillus plantarum, Lactococcus lactis ssp. lactis, Kluyveromyces marxianus, Lb. acidophilus Lb. parakefir Lb. casei Lb. rhamnosus, Lb. fructivorans, Lb. hilgardii Yeast strains: Zygosaccharomyces sp. Candida lipolytica, C.holmii Saccharomyces cerevisiae S. fragilis S. lactis	The temperature for LAB Strains to ferment kefir is 37°C	10 ⁸ CFU/mL of lactobacilli and lactococci and 10 ⁵ cfu/mL of yeasts Acetic acid bacteria (10 ⁵ CFU/g)	(Arslan, 2015; Prado et al., 2015; Tan et al., 2022)
Fermented non-dairy products Sausages	Lactobacillus sakei Lactobacillus plantarum, Lactobacillus paracasei subsp. paracasei, Weissella viridescens, Lactobacillus coryniformis	15 to 35°C for 1 to 5 days in the U.S for lab fermentation 20 to 32°C for 2 to 5 days in Europe pH for lab fermentation in Sausages is typically below 4.7	LAB present at end of sausages is about 5- 7 log CFU/g of sausages	(Dé et al., 1999; Zamfir et al., 2022) (Mohammadpourfard et al., 2021) (Messens et al., 2003)

LAB plays a crucial role in the food industry, particularly in cheese production. These bacteria are extensively used for various purposes in cheese making, contributing to the fermentation, flavor development, and quality of cheeses. In the food industry, LAB is majorly employed as starter cultures in cheese production, aiding in curd acidification and influencing the appearance and flavor of the product. Additionally, LAB is utilized as a non-starter LAB (NSLAB) in cheese ripening, further enhancing the sensory characteristics of cheeses (Better et al., 2023; T, 2018b; Valdiviezo-Marcelo et al., 2023). In previous studies, 16S rRNA genome sequencing plays an important role in studying the genome of ripening cheese which is carried out by LAB strains. These research give us the microbiota synthesis of smear-aged cheddar (cheese), and to decide how individual bacterial species and microbial gatherings change over the long run inside the cheese grid. The microbiota progression was checked during the industrial cheese production, the temperature of ripening and storing as shown in the table which included maturation and started maturing at 15 °C, and during after production of cheese, which included putting away the result in the market at 4 °C for an ensuing time of as long as 40 days (Korena et al., 2023; Lovayová et al., 2015). The uses of LAB in cheese production which is held in the food industry are multifaceted. LAB is essential for acidifying milk, initiating fermentation, and producing specific flavors and textures in cheeses. Moreover, these bacteria contribute to the safety and quality of cheeses, playing a vital role in the ripening process and overall maturation of the product. LAB also has probiotic potential, offering additional health benefits to consumers when present in cheeses (Ahansaz et al., 2023).

Overall, the industrial application of LAB in cheese production in the food industry is extensive and essential for ensuring the desired characteristics, flavors, and quality of cheeses. LAB are versatile microorganisms that significantly impact the cheese-making process, making them indispensable in the food industry. The significance of LAB in cheddar creation is because of the occurrence of starter societies and nonstarter LAB. Starter societies, fundamentally Lactococcus lactic or S. thermophilus, are liable for changing over lactose into lactic acid at a controlled rate (Hosken et al., 2023; Marcelino, 2013). In the cycle, it brings about a progressive reduction in pH, which essentially affects different parts of cheddar creation and eventually decides the cheddar's structure and standard. During the beginning phases of many cheeses maturing, L. delbrueckii or L. helveticus (Coelho et al., 2022b; Nicosia et al., 2023) assume a basic part, separating proteins, utilizing lactose, creating fragrant mixtures, and giving substrates that can be additionally consumed by other microbial gatherings, for example, NSLAB. NSLAB primarily incorporates into the facultative heterofermentative Lactobacillus sort, trailed by Pediococcus pentosaceus (Marcelino, 2013) (Coelho et al., 2022b). They can affect the cheddar taste and surface because of the development of mixtures from the catabolism of amino acids, for the most part, methionine, fragrant amino acids, and stretched chain amino acids, notwithstanding the amalgamation of EPS. Moreover, bacteriocins, hydrogen peroxide, diacetyl, and CO₂ are likewise created by NSLAB, going about as bio preservatives, and adding to the cheddar security (Table 7) (Marcelino, 2013) (Coelho et al., 2022b).

Name of Products	Bacteria	References
Milk	Bacillus	(Huck et al., 2007)
	Campylobacter	(Msalya, 2017) (Kabuki et
	Clostridium	al., 2004) (Mdegela et al.,
	Salmonella	2004) (Bastam et al.,
	S. aureus	2021b)
	S. intermedius	
	S. epidermis	
	S. agalactiae	
	Listeria monocytogenes	
	Mycobacteria	
Yogurt	E. coli	(Hervert et al., 2017)
5	Enterobacteriaceae	(Canganella et al.,
	P. paucimobilis	1999)](Yang & Yoon,
	E. cloacae	2022)
	L. monocytogenes	,

 Table 7. Demonstrate the bacterial isolates present in different milk products.



Cheese	Listeria monocytogenes Pseudomonas Salmonella Bacillus	(Marrakchi et al., 1993) (Al-Gamal et al., 2019) (Costanzo et al., 2020)
	S. aureus	

6.1.3 Food preservation

LAB assumes an urgent part in food safeguarding in the food business because of its capacity to hinder the development of a wide assortment of food decay organic entities. They are known for their power to deliver restraining specialists, which brings about a reduction of pH and improves the nature of food. (Negm, 2018; Soomoro et al., 2022). LAB is added to food as societies and is by and large viewed as innocuous or even helpful for human wellbeing in the US, where they are recorded as GRAS (Generally Recongnized as Safe). LAB produces antimicrobial mixtures called bacteriocins, which stand out enough to be noticed because of their possible handiness as regular substitutes for synthetic food additives. Bacteriocins-conveying social orders have been precisely applied to thwart *Listeria monocytogenes* and *Clostridium spp*. in various developed types of meat, vacuum-packaged things, and vegetable-based food assortments (Mokoena et al., 2021). Nisin A is one of the most important bacteriocins used as a food preservative (Arauz et al., 2009). The spread of immunizing agent poison hindrance and interest in food things with fewer added substances requires a search for new decisions to avoid the abuse of accommodating serum poisons.

LAB withdrawn from uniquely designed developed vegetables produces antibacterial substances against both Gram-positive and Gram-negative ordinary foodborne bacterial microorganisms (Mokoena et al., 2021; Negm , 2018). This extensive variety of restriction suggests that the LAB strains have the potential as should be expected bio preservatives in various food things, and to fight foodborne microorganisms. Bacteriocins are generally bactericidal, while some are bacteriostatic, conveying support in the food and medication regions, especially moreover where development is awful. Bacteriocins are fruitful against Gram-positive toxigenic and pathogenic organisms, acting by outlining pores in the movies of target microorganisms. Heterofermentative *Lactobacillus spp.* have been shown to keep out squandered organic entities in cheddar processing. Bacteriocins can restrain the expansion of food decay microorganisms and foodborne microbes, making them a promising option in contrast to compound additives in the dairy business (Mezaini et al., 2013; Negm & G. E. S, 2018).

7. Conclusion

LAB and probiotics are a non-chemical strategy for reducing food contamination through several mechanisms of action. The essential, organically important acids, bacteriocins, and other inhibitory compounds that LAB and probiotics produce, and their competition with harmful microorganisms distinguish them from other existing cultures and position them as an essential part of food safety strategies. Because, as for the above, the successful application of LAB and probiotics in many food products, from dairy and meats to fermented vegetables and beverages, underlines their benefits: enhancing the quality, shelf life, and prevention of disease in foods. Increased consumer demand for natural, safe preservation requires deeper integration of LABs and probiotics in the food chain. Importantly, there is a need to investigate the best uses of these additives in different food systems and how they interact with various contaminants. Ultimately, this can sharply enhance the benefits of using LABs and probiotics in creating a much safer, sustainable, and healthy food supply chain.

CRediT authorship contribution statement

We, the authors, declare that the manuscript titled "Role of lactobacillus probiotics in gut health and food safety: Mechanisms for contaminant reduction and applications in food products" is an original work and has not been submitted elsewhere for publication. The authors – Mohsin Gulzar Barq, Amna Razzaq, Manal Yasin, Khadija Nawal, and Tayyaba Qureshi – have all contributed significantly to the preparation of this review.

Conflict of interest statement

The authors declare no conflicts of interest related to this work.

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