The Role of *Lactobacillus plantarum* in Producing Prebiotics and Evaluation Parameters of Prebiotic Properties for Health: A Review

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Abstract. *Lactobacillus plantarum*, a facultative anaerobic and microaerophilic lactic acid bacterium (LAB), is gaining popularity in research due to its multifunctional properties. This review aims to summarise information about the use of *L. plantarum* from prebiotics-producing fermentation, evaluation parameters of prebiotic properties, and its application in the health field. *L. plantarum* can potentially be used as a fermentation culture because it can produce pullulanase enzymes that can break 1,6-glycosidic bonds or amylopectin branch chains to produce straight amylose chains. The fermentation process, followed by heating and cooling, can retrograde and crystallise amylose fractions, thereby increasing the amount of prebiotic content in the form of resistant starch. Interestingly, the prebiotics were then evaluated using *L. plantarum* again to analyse several standard parameters, including the prebiotic index, prebiotic effects, and prebiotic activity. Moreover, *L. plantarum*, when used as probiotics, was incorporated into prebiotic-rich fermented food and beverage ingredients and then distributed to the market for promoting gastrointestinal health.

1 Introduction

Gastrointestinal infections are one of the major clinical problems. Every year, gastrointestinal infections cause significant morbidity and mortality worldwide. One of the most prevalent issues in tropical nations is intestinal disease, which typically presents as diarrhea, flatulence, abdominal pain, gastrointestinal bleeding, intestinal blockage, malabsorption, or malnourishment. One of the primary causes of morbidity and mortality in children is infectious diarrheal illnesses. The World Health Organization (WHO) estimates that diarrheal diseases are the second leading cause of death in children under five and caused the death of 370,000 children in 2019. This has sparked increased interest in the potential use of beneficial microorganisms, such as probiotics, in humans as a therapeutic approach for gastrointestinal disorders. Probiotics are live microorganisms that provide health benefits to the host by colonizing when given in sufficient amounts. Probiotics can alter the composition of human

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intestinal microorganisms and prevent the colonisation of pathogenic bacteria in the intestine. Additionally, probiotics have been shown to help the body establish a healthy barrier on the intestinal mucosal lining, thereby enhancing the intestinal barrier function and immune response. One of the most important groups of probiotics associated with several functional properties is Lactic Acid Bacteria (LAB). One of the bacterial species included in Lactic Acid Bacteria is *Lactobacillus plantarum*. Abdelazez et al. [1] reported that *L. plantarum*, proposed by Orla-Jensen in 1919, is a facultative heterofermentative lactic acid bacterium widely distributed in nature. *L. plantarum* can adapt to various environments. In addition, because it has the largest genome (~3.3 Mb) among the LAB group, *L. plantarum* can be isolated from a wide range of sources. *L. plantarum* is also very tolerant to various severe conditions, including stress on the digestive system, gastrointestinal tract, vagina, and urogenital. Recent studies indicate that interest in *L. plantarum* has increased, particularly in relation to its probiotic potential and applications in various fermented foods and beverages, including fermented dairy products, fermented meat products, and fermented vegetables. *L. plantarum* is gaining popularity in research because of its several multifunctional properties [2].

Fermentation is a traditional bioprocess widely used to improve the nutritional, functional, and sensory qualities of food. Microorganisms such as *Lactobacillus plantarum* play a pivotal role in these processes, contributing to the transformation of complex substrates into beneficial metabolites. Numerous studies have extensively reviewed the general role of lactic acid bacteria (LAB), including *L. plantarum*, in the production of fermented foods and the modulation of gut microbiota through probiotic activity [3]. However, a less explored aspect is the ability of *L. plantarum* to influence the formation of resistant starch (RS) during fermentation. Resistant starch is a non-digestible carbohydrate that escapes digestion in the small intestine and is fermented in the colon, where it serves as a substrate for beneficial gut microbes. Its prebiotic potential has been associated with improved glycemic response, enhanced mineral absorption, and reduced risk of colon cancer and other metabolic disorders. Recent studies suggest that microbial fermentation can modify the starch structure in ways that promote RS formation [4]. Yet, the specific contributions of individual strains, such as *L. plantarum*, in this context remain insufficiently characterised. Understanding this relationship is particularly important for developing functional foods with targeted health benefits. This review aims to explore the specific role of *Lactobacillus plantarum* in enhancing RS formation during fermentation. By synthesising existing findings and highlighting strain-substrate interactions, this paper aims to clarify the mechanisms through which *L. plantarum* contributes to the generation of compounds with prebiotic activity. In doing so, we address a specific niche in the broader field of fermentation research and propose practical implications for the development of gut health-promoting food products.

2. Materials and Methodology

This review article was written based on a literature study encompassing publications from 2012 to 2025. The data sources used were open-access sources, including ResearchGate, ScienceDirect, Scholar, SAGE, NCBI, and Nature. The literature study criteria included information on the classification and taxonomy of *Lactobacillus plantarum*, sources of *L. plantarum* bacteria, fermentation of *L. plantarum*, evaluation of prebiotic properties with *L. plantarum*, and the application of *L. plantarum* bacteria in the health industry.

3. Results and Discussion

3.1 Morphological and Physiological Characteristics of *Lactobacillus plantarum*

Fermentation is a process that utilises microbes to break down large organic molecules into smaller ones. Dietary components are frequently fermented by microbial or enzymatic action, resulting in favourable biochemical changes that lead to notable dietary alterations. Thus, microbial activity is crucial for food fermentation, which alters the food's chemical and physical characteristics [3]. Kumar et al. [4] reported that the microbes used for fermentation are lactic acid bacteria (LAB). LAB are divided into two groups based on their fermentation results, namely homofermentative and heterofermentative. In addition to lactic acid, heterofermentative bacteria also produce other substances, including ethanol, acetic acid, and carbon dioxide (CO_2). Homofermentative bacteria are those that produce lactic acid as their primary output. A number of antimicrobial substances, including organic acids, CO_2, hydrogen peroxide, diacetyl, reuterin, and bacteriocin, are produced by LAB. The organic acids produced by LAB are lactic acid and acetic acid, which can lower the pH of the cytoplasm because organic acid molecules enter the cell membrane. The types of LAB that play a role in the fermentation process are *Enterococcus, Streptococcus, Leuconostoc, Lactobacillus,* and *Pediococcus* [3].

The genus *Lactobacillus* comprises several groups, including various species that are utilised for fermentation and food preservation. Some *Lactobacillus* species are probiotics that aid in the digestion of living organisms, providing beneficial effects and promoting the health of their hosts. One type of *Lactobacillus* is *Lactobacillus plantarum* (Figure 1), as reported by Abdelazez et al. [1] L. plantarum is one of the most important microbes in the Gram-positive category, classified within the division of lactic acid bacteria (LAB). These bacteria are short, rod-shaped, have negative catalase, and do not form spores. *L. plantarum* is a facultative anaerobic and microaerophilic bacterium. Additionally, the low G+C content is acid-resistant. *L. plantarum* is grouped as a homofermentative *Lactobacillus*. The optimal temperature for the development of *L. plantarum* is generally lower than 37°C. Based on Behera et al. [2], the taxonomy of *L. plantarum* is included in the Kingdom Bacteria, Division Firmicutes, Class Bacilli, Order Lactobacillales, Family Lactobacillaceae, Genus: *Lactobacillus*, with Species: *Lactobacillus plantarum*. *L. plantarum* is a type that is often found in food microbiology, especially fermented food systems.

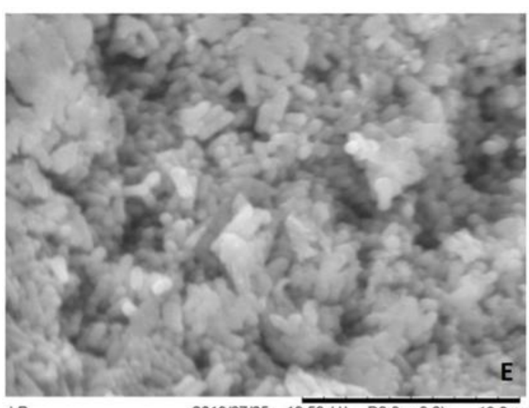

Fig. 1. *Lactobacillus plantarum* [5]

L. plantarum is a Gram-positive bacterium widely studied and utilized due to its probiotic properties and industrial significance. Found in a variety of environments, including fermented foods and the gastrointestinal tracts of humans and animals, this versatile microorganism has been extensively characterized in terms of its morphology and physiology [1]. Understanding its traits provides insight into its ecological roles, applications in biotechnology, and interactions with hosts. *L. plantarum* cells are rod-shaped, typically measuring 0.9 to 1.2 μm in width and 3 to 8 μm in length. They can occur singly or in short chains, depending on the growth conditions. The bacterium's cell wall is thick and composed of peptidoglycan, teichoic acids, and lipoteichoic acids, which confer structural integrity and resistance to osmotic pressure. This structural composition also plays a role in its interaction with the host's immune system. Unlike some bacteria, *L. plantarum* is non-motile and lacks flagella. The bacterium is non-sporulating, which means it does not produce spores for survival under harsh conditions [6]. Instead, it relies on physiological adaptations to endure stress. Some strains of *L. plantarum* can produce an extracellular polysaccharide capsule, which enhances their resistance to desiccation, bile salts, and acidic conditions, while also aiding in biofilm formation.

L. plantarum is a facultative heterofermentative organism. It primarily ferments sugars into lactic acid, contributing to the acidic environment in fermented foods and the gut. Under certain conditions, it can produce other byproducts, such as ethanol, acetic acid, and carbon dioxide. The bacterium's ability to utilize a wide range of carbohydrates, including glucose, fructose, and galactose, makes it highly adaptable to different environments. Although classified as a lactic acid bacterium, *L. plantarum* exhibits aerotolerance. It can grow in both aerobic and anaerobic conditions. The presence of manganese-dependent enzymes, such as superoxide dismutase, helps neutralize reactive oxygen species (ROS). *L. plantarum* thrives at temperatures between 30°C and 40°C, with an optimum around 37°C. It grows best at a pH of 5.5 to 6.5 but can tolerate acidic conditions down to a pH of 3.0, which is critical for its survival in the gastrointestinal tract and fermented foods. The bacterium's resilience to environmental stresses, such as high salinity, low pH, and bile salts, makes it suitable for probiotic use [1]. Stress-response proteins and changes in membrane composition facilitate this. *L. plantarum* has demonstrated significant probiotic effects, including modulating gut microbiota, enhancing immune responses, and producing antimicrobial compounds like bacteriocins and organic acids. *L. plantarum* is a prominent probiotic due to its ability to modulate gut microbiota, enhance mucosal barrier function, and exert immunomodulatory effects. Its resistance to gastric acids and bile salts facilitates its survival and colonization in the gastrointestinal tract. The bacterium is extensively used in the fermentation of dairy products, vegetables, and meat. Its metabolic flexibility and ability to produce antimicrobial compounds, such as bacteriocins, contribute to food preservation and flavour development [2]. The wide genetic diversity of *L. plantarum* enables it to colonise diverse habitats, ranging from plant surfaces to extreme environmental niches. This adaptability is attributed to its large genome, which encodes various transporters, stress response proteins, and metabolic pathways.

L. plantarum breaks down complex compounds into simpler compounds, resulting in lactic acid. Lactic acid can produce a low pH on the substrate, creating an acidic atmosphere and increasing the substrate's acidity by 1.5 to 2%. In acidic conditions, *L. plantarum* can inhibit the growth of pathogenic microorganisms and toxin producers [7]. *L. plantarum* can also produce hydrogen peroxide and bacteriocins, which are bactericidal protein compounds as antibacterial substances. By producing lactic acid, *L. plantarum* creates an acidic microenvironment that inhibits competitors while maintaining intracellular pH homeostasis through the action of proton pumps. Adaptations, such as bile salt hydrolase activity, enable it to deconjugate bile acids, thereby aiding its survival in the gastrointestinal tract [1]. The production of compatible solutes, such as glycine betaine and trehalose, helps the bacterium manage osmotic pressure. The morphological and physiological characteristics of *L. plantarum* underscore its ecological versatility and functional significance. Its robust structure, metabolic flexibility, and stress tolerance make it a key player in both natural ecosystems and industrial applications. As research continues to explore its genetic and functional potential, *L. plantarum* remains a model organism for understanding the efficacy of probiotics and microbial adaptability.

3.2. *Lactobacillus plantarum* as Probiotic

Lactobacillus plantarum has a wide range of applications, for example, as a probiotic bacterium that consumes prebiotics as a carbon source and substrate. *L. plantarum* was isolated from starchy wastes like cassava starch, and it is a potential source of lactic acid. *L. plantarum* can also improve the texture and taste of bread and sausages [2]. *L. plantarum* facilitates the bioconversion of substrates into potentially prebiotic compounds, and the specific metabolites produced depend on the nature of the substrate. During fermentation, *L. plantarum* produces the enzyme pullulanase, which can break the 1,6-glycosidic bond or amylopectin branch chain, allowing a straight chain of amylose to be produced. The longer the fermentation, the greater the growth of *L. plantarum*, resulting in higher enzyme production, which can increase amylose levels. The higher the amylose content, the more the autoclaving-cooling process of a starch suspension at gelatinisation temperature can cause the breaking (dissociation) of hydrogen bonds within the amylopectin double helix structure, the melting of the crystalline part, and the release of amylose from its granules (amylose leaching).

Table 1. Modification techniques for increasing resistant starch by fermentation of *L. plantarum* in various food materials

Food Materials	Source of *L. plantarum*	Fermentation procedure	RS control	RS Modified	References
Porang	*L. plantarum* IIA-1A5 culture	Starch fermented by *L. plantarum* in an incubator at 32°C for 24 h, followed by autoclaving and cooling for one cycle	53.33 %	55.16 %	[8]
Chinese yam	*L. plantarum* Xi'an Jushengyuan Biotechnolog y	The starch pressure cooker is maintained at 121 °C, refrigerated at 4°C, and then enzymatic hydrolysis is performed. RS3 added to *L. plantarum* liquid, maintaining a total bacterial count of 10^9 CFU/mL, then incubated at 37 °C for 4, 8, 12, and 16 h	39.43 ± 0.25 %	45.37 ± 0.25 %	[7]
rice	*L. plantarum* YI-Y2013	Starch fermented by *L. plantarum* (10^8 CFU/mL) at 37 ºC for 12, 24, 36, 48, and 60 h, then freeze-dried	18.49 ± 2.14 %	27.30 ± 0.04 %	[9]
Plantain	*L. plantarum* A6 (LMG 18053)	Heated at 95 ºC for 15 min under stirring. Gelatinised slurries were then cooled to 30 ◦C and inoculated with *L. plantarum*	20.7 ± 7.2 (g/100 g)	19.3 ± 3.7 (g/100 g)	[10]
Breadfruit			20.2 ± 2.9 (g/100 g)	26.0 ± 4.3 (g/100 g)	
Sweet potato			18.4 ± 5.5 (g/100 g)	27.5 ± 2.1 (g/100 g)	
Wheat	*L. plantarum* ELB75	Starch fermented by adding *L. plantarum* at two different fermentation temperatures (25 ºC and 30 ºC)	2.86 ± 0.65 (g/100 g)	16.93 ± 0.71 (g/100 g)	[11]

Furthermore, the starch paste is cooled (cooling), which can cause the amylose fraction to undergo retrogradation. The repeated autoclaving-cooling process can lead to the formation of more retrograded or crystallised amylose fractions, increasing the resistant starch content with each additional cycle [7]. Table 1 contains various studies on *L. plantarum* as a fermentation-producing prebiotic. Modification with *L. plantarum* fermentation can increase the levels of resistant starch in food materials, as proven by RS Modified being higher than RS control in all food materials.

Fermentation is a metabolic process through which *L. plantarum* converts carbohydrates into organic acids, gases, and other byproducts. This process is primarily anaerobic, although the organism can tolerate oxygen, making it a facultative anaerobe. The fermentation pathways in *L. plantarum* are critical for its survival and industrial utility, especially in the production of fermented foods and beverages [1]. The fermentation process is divided into two categories. Firstly, the homofermentative Pathway: *L. plantarum* predominantly employs the Embden-Meyerhof-Parnas (EMP) pathway to metabolize glucose. This glycolytic pathway breaks down glucose into two molecules of pyruvate, which are subsequently reduced to lactic acid. The overall reaction can be summarised as follows: Glucose → 2 Lactic Acid + 2 ATP. This homofermentative process is energetically efficient, ensuring a high yield of lactic acid, which makes it valuable for food preservation and flavour enhancement. Secondly, heterofermentative Capability: Although primarily homofermentative, *L. plantarum* exhibits heterofermentative behaviour under specific conditions, such as when alternative carbon sources like pentoses are metabolized [3]. In this scenario, the phosphoketolase pathway (PKP) is activated, leading to the production of lactic acid, acetic acid, ethanol, and carbon dioxide. Pentose → Lactic Acid + Acetic Acid + CO_2 + ATP.

L. plantarum exhibits remarkable metabolic diversity, which allows it to utilise a wide range of carbohydrates and adapt to varying environmental conditions. Some of the critical metabolic pathways include four categories. Firstly, carbohydrate Metabolism: *L. plantarum* can metabolise a variety of sugars, including glucose, fructose, galactose, and arabinose. The EMP pathway is central to its carbohydrate metabolism, ensuring efficient ATP production and reducing power in the form of NADH [3]. Secondly, Amino Acid Metabolism: Amino acid metabolism plays a vital role in the survival and growth of *L. plantarum*, especially in nutrient-limited environments. For example, the organism can convert glutamate to γ-aminobutyric acid (GABA), a bioactive compound with potential health benefits. Thirdly, lipid Metabolism: While not a primary pathway, lipid metabolism in *L. plantarum* supports membrane biosynthesis and maintenance. The organism synthesises short-chain fatty acids and modifies membrane lipids to adapt to environmental stresses, such as changes in pH and temperature. Last one, Stress Response Mechanisms: *L. plantarum* employs metabolic adaptations to counteract environmental stresses. For instance, it can produce exopolysaccharides (EPS) to enhance biofilm formation and resistance to desiccation or osmotic stress. *L. plantarum*'s homofermentative properties make it essential for making fermented foods, including sourdough bread, cheese, yogurt, sauerkraut, and kimchi. The lactic acid produced not only imparts a tangy flavour but also acts as a natural preservative by lowering the pH and inhibiting the growth of spoilage organisms [2]. *L. plantarum* is widely recognised as a probiotic due to its ability to survive gastric transit and colonise the gut. It contributes to gut health by producing antimicrobial compounds, such as bacteriocins, modulating the immune response, and enhancing the gut barrier function. The metabolic pathways of *L. plantarum* have been harnessed for industrial applications, including the production of bioactive compounds, bioplastics, and biofuels. Its ability to produce GABA and other functional metabolites underscores its importance in the field of nutraceuticals. The fermentation and metabolic pathways of *L. plantarum* reflect its adaptability and utility in diverse applications. Its homofermentative efficiency, coupled with its metabolic versatility, enables it to play a crucial role in food preservation, human health, and biotechnological innovations [3]. Continued research into its metabolic pathways holds promise for enhancing its applications and unlocking new benefits in various fields.

3.3 *Lactobacillus plantarum* as A Parameter for Evaluation of Prebiotic Properties

Prebiotics are indigestible food ingredients that can reach the large intestine and serve as a substrate for the endogenous bacteria in the colon. These prebiotics are useful for stimulating and enhancing the function of digestive bacteria in the large intestine, allowing them to maintain colon health. This indirectly provides the host with energy, metabolic substrates and essential micronutrients. Based on Kumar et al. [4], prebiotics, in general, have been observed to cause reactions that can increase the concentration of lactic acid and acetic acid. This is shown by the fermentation carried out by lactic acid bacteria (*Lactobacillus* sp.) and *Bifidobacteria*. Lactate, the primary product of homofermentative fermentation, is produced in the fermentation process. In heterofermentative fermentation, other compounds include acetate, ethanol, CO_2, formic acid, and succinate. Prebiotics, which are substrates that host bacteria preferentially utilise to provide health benefits, have garnered considerable interest in nutritional science and medical research. The evaluation of prebiotic properties involves multiple dimensions, including chemical composition, functional effects on gut microbiota, and demonstrable health benefits. Prebiotics typically consist of non-digestible carbohydrates, such as oligosaccharides, resistant starches, and dietary fibres. These substances pass through the upper gastrointestinal tract and enter the colon undigested, where they serve as food sources for beneficial microbes. The selection of candidate prebiotics involves two criteria. Firstly, prebiotics must remain structurally intact through the stomach and small intestine. Secondly, the compound should be selectively utilised by beneficial bacteria, particularly *Bifidobacteria* and *Lactobacilli* [12]. The prebiotics must retain their functional properties during processing and storage.

Despite advancements, challenges in prebiotic evaluation remain. These include variability in individual microbiota responses, the complexity of gut microbiota interactions, and the need for standardised testing protocols. Future research should focus on several key areas. Firstly, tailoring prebiotics to individual microbiota profiles for optimised benefits. Secondly, exploring new compounds, including non-carbohydrate prebiotics, with targeted health effects, and investigating the molecular mechanisms underlying prebiotic-microbiota interactions. Evaluation of prebiotic properties using *L. plantarum* (Table 2) can be seen from several important parameters, including prebiotic index, prebiotic effects and prebiotic activity [5]. The prebiotic index (PI) reflects the ability of a particular substrate to support the growth of an organism compared to other organisms and growth on non-prebiotic substrates, such as glucose or other sugars used as controls [13]. Food ingredients are considered good sources of prebiotics if they have a prebiotic index of 2.0 or higher. The prebiotic index formula assumes that an increase in the numbers of *Lactobacilli* and *Bifidobacteria* is beneficial, while a rise in *Bacteroides* and *Clostridium* (subgroup *Histolyticum*) is detrimental. The prebiotic effect shows an absolute increase in the population of probiotic bacteria, while the prebiotic index considers the concentration of the prebiotics [8].

Table 2. Some methods of evaluating prebiotic properties use *L. plantarum*

Food Materials	Source of *L. plantarum*	Prebiotics Evaluation Procedure	Prebiotic Effect	Prebiotic Index	Prebiotic activity	References
Porang	*L. plantarum* IIA-1A5 culture	adding *L. plantarum* culture into m-MRSB with 2,5% (w/v) of modified porang flour	3.07	2.46	0.02	[8]
Corn	*L. plantarum* IIA-1A5	observing the change in the number of *L. plantarum* colonies on m-MRSB medium with 2.5% starch	0.76	0.6	1.96	[5]
Cempedak Seed	*L. plantarum* 1-S27202	adding *L. plantarum* 1-S27202 (7 log CFU/ml) into fermentation media (m-MRSB)	0.270	0.680	n	[14]
Daluga	*L. plantarum* BSL	adding 5% (v/v) of *L. plantarum* BSL culture in m-MRSB media containing 2.5% (w/v) autoclaved daluga heat moisture treatment	0.97	3.74	0.17	[12]

Note: n = not determined

Based on Figueroa-Gonzalez et al. [13], prebiotic activity is related to the selectivity of prebiotics in supporting the growth of probiotics, but it does not occur in pathogenic bacteria. Probiotic bacterial growth is supported by substrates with high prebiotic activity ratings, and cell numbers (CFU/mL) are equivalent to those grown on glucose. However, the development of *E. coli* on prebiotics is expected to be significantly lower compared to probiotics. Therefore, the prebiotic activity score can be determined relative to a particular strain by quantitative calculations. Suppose a group of bacteria shows a relative increase greater than the total bacterial population. In that case, the increase is considered > 1, but otherwise, it is considered < 1. This value is then added to the PI calculation to produce an overall score [8]. Therefore, carbohydrates have a positive activity score if probiotic strains metabolise them and are selectively metabolised by probiotics but not by other gut bacteria [13].

Based on Table 2, the prebiotic effect and prebiotic index showed positive values in all food materials, indicating that the resistant starch research has a positive effect on the growth of *L. plantarum*. These food materials have the potential to be prebiotics because they meet the second requirement for prebiotics, namely, fermentation by intestinal microflora, as indicated by positive effects and prebiotic index values [12]. The data is supported by prebiotic activity, which yields positive values, indicating that the growth of *L. plantarum* is higher compared to that of pathogenic bacteria. Evaluation of prebiotic properties with *L. plantarum* proves that it can be done on food ingredients, so that it is no longer necessary to use other LABs. The use of *L. plantarum* for fermentation and evaluation sequentially can make the research more efficient. However, several studies still compare the growth of *L. plantarum* with other probiotics, such as *L. rhamnosus* [14]. The evaluation of prebiotic properties is a multifaceted process requiring rigorous scientific methodologies. By enhancing our understanding of their chemical, functional, and clinical attributes, prebiotics hold immense potential in improving human health. As research progresses, the development of novel prebiotics and personalised interventions will likely redefine the role of these compounds in nutrition and medicine.

3.4 Benefits of *Lactobacillus plantarum* for Health and Optimization with Prebiotics

After being used in fermentation to produce prebiotic compounds and reused for evaluation of its prebiotic activity, *L. plantarum* has the potential to be used as a probiotic. The combination of *L. plantarum* and prebiotics (such as resistant starch) can be formulated into synbiotic products based on fermented foods or beverages, such as yogurt, which can then be marketed commercially. A synbiotic is a product that combines both probiotics (live beneficial microorganisms) and prebiotics (non-digestible food components that selectively stimulate the growth or activity of beneficial bacteria in the gut) in a way that promotes synergistic health benefits, particularly for gastrointestinal and immune health [13]. In recent years, the concept of synbiotics, a combination of probiotics and prebiotics, has gained attention for its potential to enhance gut health synergistically. Synbiotics aim to improve the survival and implantation of live microbial dietary supplements in the gastrointestinal tract by selectively stimulating the growth and activity of beneficial bacteria, such as *Lactobacillus plantarum*, through the presence of specific prebiotic substrates. The potential health benefits of *L. plantarum* as a probiotic for human health include immune system regulation, lowering cholesterol levels, maintaining stable intestinal microorganism balance, and reducing tumour risk.

Table 3. Mechanisms and health benefits of *Lactobacillus plantarum* and optimisation with prebiotics

Mechanism	Health Benefit	Role of Prebiotics in Optimisation	References
Modulation of Gut Microbiota	Inhibits the growth of pathogens, enhances beneficial bacteria	Serve as selective substrates for *L. plantarum*, promoting growth and colonisation	[15]
Strengthening of Intestinal Barrier	Reduces gut permeability, prevents leaky gut, protects against endotoxin leakage	Enhances microbial activity, leading to short-chain fatty acid (SCFA) production that supports epithelial integrity	[15]
Production of Bioactive Metabolites	SCFAs (e.g., acetate, lactate) regulate gut pH, support colonocyte health	Increased fermentation of prebiotics results in higher short-chain fatty acid (SCFA) production	[7]
Immunomodulation	Boosts mucosal immunity, balances pro- and anti-inflammatory cytokines	Prebiotics enhance probiotic activity, leading to more effective immune signalling	[15]
Antioxidant and Anti-inflammatory	Reduces oxidative stress, lowers the risk of chronic inflammation	Prebiotics stimulate the metabolism of polyphenols and antioxidant precursors	[15]
Cholesterol Reduction	Helps lower LDL cholesterol and improve lipid profiles	Enhanced fermentation activity leads to greater bile salt hydrolase activity	[15]
Resistant Starch Interaction	Promotes RS formation, supports digestive and metabolic health	RS acts as both substrate and prebiotic, enhancing *L. plantarum* activity synergistically	[9]
Nutrient Bioavailability	Improves absorption of minerals (e.g., calcium, magnesium)	Short-chain fatty acids (SCFAs) from prebiotic fermentation improve mineral solubility and uptake	[15]

Among *Lactobacilli*, *L. plantarum* is a flexible and widespread species. Its dual role as a native human intestinal inhabitant and a protected starter culture in food fermentation provides good stability, allowing it to be used as part of food fermentation [1]. *L. plantarum* reduces total cholesterol and LDL cholesterol concentrations as well as serum fibrinogen levels after consumption. *L. plantarum* has been given to patients with irritable bowel syndrome and has been detected to reduce symptoms such as pain and flatulence. Administration of *L. plantarum* mitigates the adverse effects of antibiotics on colonic fermentation and may offer additional benefits in patients with recurrent diarrhea associated with *Clostridium difficile*. Consumption of *L. plantarum* may have a preventive effect on milder gastrointestinal symptoms during antibiotic treatment.

L. plantarum as a probiotic carries out prebiotic metabolism through the glycolysis process with the help of the pyruvate kinase enzyme to form pyruvic acid, which is then converted into short-chain fatty acids such as acetic acid, propionate and butyrate. Prebiotics that are not utilised by probiotic bacteria will then be utilised by the human digestive tract as a source of fibre that will help the process of water absorption in the colon, thus forming faeces [15]. Prebiotics stimulate the growth and activity of probiotic bacteria in the colon, such as *Bifidobacteria* and *Lactobacillus,* thereby inhibiting the growth of pathogenic bacteria, increasing calcium absorption, protecting against colon cancer, lowering cholesterol, and enhancing immunity. Prebiotics, such as resistant starch, inulin, raffinose, FOS, and GOS, can be utilised by probiotic bacteria as a carbon source to increase their growth. The products of prebiotics fermentation by probiotic bacteria are short-chain fatty acid (SCFA) compounds such as acetic acid, propionate, and butyrate [1]. The presence of SCFA will lower the pH of the intestinal mucosal environment. These short-chain fatty acids cause the intestines to become acidic, ultimately suppressing the growth of pathogenic bacteria that cause intestinal inflammation, such as enteropathogenic *Escherichia coli* (EPEC) and *Clostridium perfringens*. SCFA can also lower intestinal pH, increase calcium absorption, and reduce ammonia and amines absorption, thereby preventing high blood pressure.

4. Conclusion

Lactobacillus plantarum plays a crucial role in fermentation processes by converting specific substrates into metabolites with potential prebiotic properties, including short-chain fatty acids and oligosaccharides. While it does not produce prebiotics directly, its metabolic activity contributes to the formation of compounds, such as resistant starch, that support the growth of beneficial gut microbiota. This interaction between microbial metabolism and substrate type is crucial in designing functional foods that aim to improve gut health. By enhancing the levels of resistant starch, L. plantarum indirectly supports various health benefits, including improved mineral absorption, modulation of the immune system, inhibition of pathogenic bacteria, and potential protection against chronic diseases. These findings underscore the significance of selecting appropriate microbial strains and substrates to optimise prebiotic outcomes in food applications.

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