

Review

Health-Promoting Effects of *Lactobacillus acidophilus* and Its Technological Applications in Fermented Food Products and Beverages

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Abstract: *Lactobacillus acidophilus* is a probiotic bacterium that possesses numerous health-promoting properties and has significant technological applications in the fermentation of a wide range of food products and beverages. This review discusses the health benefits of *L. acidophilus*, including its ability to enhance immunity; promote digestive wellness; and exhibit antioxidant, antitumor, and antimicrobial properties. This review also discusses the production of bioactive peptides and extracellular polysaccharides (EPS) by *L. acidophilus*. Factors, such as salinity, temperature, carbon sources, and nutrient availability, influence the growth of *L. acidophilus*, which can affect the survival and bioactive potential of fermented products. The proteolytic effects of *L. acidophilus* contribute to protein breakdown, which leads to the release of bioactive peptides with various health benefits. This review also discusses the applications of *L. acidophilus* in the fermentation of dairy products, cereal beverages, soymilk, fruit and vegetable juices, and other functional food preparations, highlighting its potential for improving the nutritional value, organoleptic properties, and probiotic delivery of these products. This review highlights the importance of understanding and controlling fermentation conditions to maximize the growth and health-promoting benefits of *L. acidophilus* in various food and beverage products.

Keywords: *Lactobacillus acidophilus*; food; beverages; fermentation metabolites; bioactive peptides; health-promoting properties

1. Introduction

Lactobacillus acidophilus is a Gram-positive, rod-shaped microorganism that exhibits several key characteristics. This bacterium is crucial for human well-being, as it aids in conditions such as lactose intolerance, nonalcoholic fatty liver disease, irritable bowel syndrome, and hypercholesterolemia and prevents *Helicobacter pylori* infections [1,2]. It

is commonly present in the gut and helps maintain optimal health. Specifically, it helps maintain the equilibrium of the microbiota, supports digestion, enhances nutrient absorption, and fortifies the immune system. Moreover, research has shown that *L. acidophilus* has the potential to decrease serum cholesterol levels, stabilize enteric microbiota, and combat cancer cells [3]. Additionally, *L. acidophilus* can inhibit the proliferation of pathogenic bacteria within the intestine, regulate the balance of intestinal flora, and enhance resistance, ultimately promoting growth and improving immunity [4]. Factors such as salinity, temperature, carbon sources, and nutrient availability can affect the growth of *L. acidophilus*. Studies have suggested that the survival and growth of *L. acidophilus* are influenced by salinity and temperature variations during fermentation [5]. The presence of essential nutrients such as amino acids, riboflavin, and minerals such as Mn^{2+} also plays a crucial role in enhancing the growth of *L. acidophilus* in fermented products [6]. Furthermore, optimizing the carbon source and concentration in the fermentation medium is essential for promoting the growth, phenolic production, and antioxidant activity of *L. acidophilus*. It is vital to understand and control these conditions to maximize the growth and bioactive potential of *L. acidophilus* during various fermentation processes [7]. *Lactobacillus acidophilus* exerts functional and proteolytic effects on various food products. It accelerates the growth of bacteria in milk, affecting lactic acid content and pH. *Lactobacillus acidophilus* exhibits proteolytic effects through its proteolytic system, involving components like CEPs (C-terminally encoded peptides) that cleave proteins into peptides, which are then internalized and degraded into amino acids by various peptidases. This system enables *L. acidophilus* to utilize proteins as a nitrogen source for growth, particularly in low-amino acid environments, such as milk. The proteolytic activities of *L. acidophilus* contribute to the breakdown of proteins, leading to the release of bioactive peptides with various health benefits, such as antioxidant [8], antibacterial [9], immunomodulatory [10], and angiotensin-I-converting enzyme (ACE) inhibitory activities [11]. Different strains of *L. acidophilus* may exhibit varying protein hydrolysis patterns that are influenced by factors such as CEP gene expression and enzymatic activity conditions.

Lactobacillus acidophilus has multiple applications in the food industry, including the fermentation of milk, enhancement of phenolic compounds, and extension of shelf life through microencapsulation. It is used in dairy products to increase nutrient absorption, adjust intestinal flora, and improve animal health [12]. *Lactobacillus acidophilus* fermentation enhances branched-chain amino acids and antioxidants in wheat bran, suggesting its potential application in functional cereal products to improve their nutritional value [13]. *Lactobacillus acidophilus* in cereal beverages may provide probiotic benefits such as those to gastrointestinal health, immune stimulation, and antimicrobial properties; enhance nutritional value; and potentially replace dairy probiotic foods [14]. It also enhances the antioxidant potential, viscosity, and isoflavonoid content of fermented soymilk, suggesting its beneficial application in vegan milk production [15]. Moreover, it is a novel probiotic for the fermentation of fruit and vegetable juice blends (artichokes), pineapple, squash, spinach, and cucumbers. Its application has been demonstrated in the development of functional beverages that enhance consumer acceptability and viability [16]. *Lactobacillus acidophilus* enhances probiotic delivery, increases antioxidant and antimicrobial capacity, and improves beverage bioactivity during the fermentation of apple juice [17]. *Lactobacillus acidophilus* in vegetable juices enhances probiotic content, antioxidant activity, and organoleptic properties, providing lactose-intolerant vegan consumers with healthy non-dairy options. *Lactobacillus acidophilus* has a wide range of applications in functional and edible probiotic preparations because of its resistance to acid and bile salts and its health-promoting properties in a variety of food products [18].

This review examines the health-promoting properties of *L. acidophilus*, particularly its ability to enhance immunity, improve gut health, and exhibit antioxidant, antitumor, and antimicrobial properties. This review also discusses the potential production of bioactive peptides and extracellular polysaccharides by *Lactobacillus acidophilus*. Furthermore, the effects of *L. acidophilus* on the fermentation of various food products such as milk, cheese,

yogurt, meat, and baking products have been reported. Finally, this review discusses the potential role of *Lactobacillus acidophilus* in the fermentation of plant-based beverages.

2. Health-Promoting Benefits of *L. acidophilus*

Lactobacillus acidophilus has various health benefits. Research has indicated that *L. acidophilus* plays a crucial role in improving cardiovascular health, lactose intolerance, cancer prevention, immune regulation, and gastrointestinal diseases [18]. Additionally, *L. acidophilus* exhibits antimicrobial activity against a wide range of pathogenic bacteria, contributing to its protective effects [1]. The immunomodulatory abilities of *L. acidophilus* can initiate anti-inflammatory responses, enhance phagocytosis, induce defensin production, and modulate intestinal permeability, all of which are essential for immunity enhancement and overall gut health. Clinical trials have shown that consuming acidophilus milk enriched with *L. acidophilus* can aid in managing conditions such as diarrhea [19], constipation [20], and Alzheimer's disease [21], showing its potential as an adjunct treatment for various health issues [22,23]. Furthermore, studies have highlighted the bioactive potential of *L. acidophilus* in fermented products, such as cheese, yogurt, fermented milk, and baking products, and its producing peptides with antihypertensive properties, thus emphasizing its role in enhancing the health benefits of food products [24,25]. The health-promoting effects of *L. acidophilus* on disease prevention, immunity enhancement, gut wellness, and the bioactivity of potential components are shown in Figure 1.

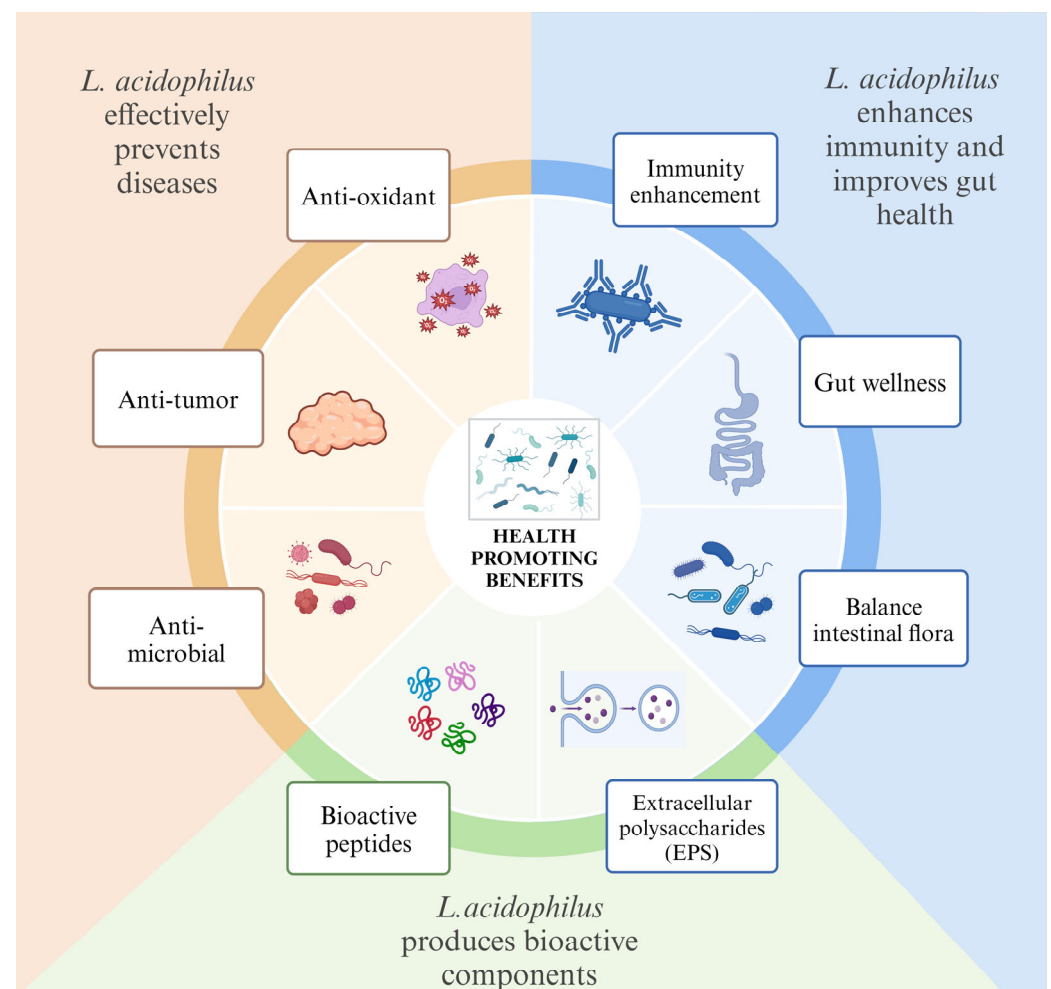


Figure 1. Health-promoting effects of *L. acidophilus* on disease prevention, immunity enhancement, gut wellness, and bioactivity of potential components.

2.1. Immunity Enhancement

Lactobacillus acidophilus has been extensively studied for its role in immunity enhancement. Research indicates that *L. acidophilus* supplementation can improve immune responses by increasing humoral immunity levels, such as with IgG and IgA, and enhancing cellular immunity, leading to a strengthened immune system [26,27]. *Lactobacillus acidophilus* plays a crucial role in enhancing immunity through various mechanisms. Studies have shown that *L. acidophilus* can modulate immune responses by increasing CD4+ and CD4+/CD8+ levels, enhancing IgA production, and reducing IL-6 and TNF- α levels [28,29]. Moreover, studies have demonstrated the immune system-boosting properties of *L. acidophilus* in poultry. In particular, it has been shown to elevate antibody levels and enhance the immune response to vaccinations, including those against the Newcastle disease virus, suggesting its potential as an immune booster in vaccination programs [29].

2.2. Gut Wellness

Lactobacillus acidophilus LA-05 has demonstrated encouraging potential in the treatment of gastrointestinal disorders by regulating the immune system, enhancing lactose tolerance, and providing relief from conditions such as colitis [30]. Studies have highlighted the ability of LA-05 to regulate intestinal flora, produce metabolites that inhibit pathogenic bacterial growth, and enhance anti-inflammatory effects by modulating TLR expression. *L. acidophilus* can produce organic acids like acetic and lactic acid, which lower pH levels and inhibit the growth of pathogenic bacteria, contributing to a healthy gut environment [2]. *Lactobacillus acidophilus* has demonstrated efficacy in reducing the duration and severity of diarrhea in individuals, particularly children. Research has shown that *L. acidophilus* can effectively combat acute gastroenteritis by decreasing the frequency and duration of diarrheal episodes. This probiotic strain has been found to positively affect the immune system, enhance both specific and non-specific immunity, and regulate the body's response to pathogens. The probiotic properties of *L. acidophilus* extend to maintaining intestinal homeostasis, supporting intestinal epithelial integrity, and ameliorating gastrointestinal disorders. Furthermore, *L. acidophilus* has been shown to modulate immune responses by restoring the balance of intestinal microbiota [31,32].

2.3. Antimicrobial Activity

Lactobacillus acidophilus exhibits significant antimicrobial activity against various pathogens and spoilage microorganisms. Research indicates that *L. acidophilus* inhibits the growth of pathogens such as *Clostridium perfringens*, *Escherichia coli*, and *Staphylococcus aureus* by producing antibacterial substances including lactic acid and bacteriocins [1,33]. Moreover, studies have shown that *L. acidophilus* inhibits gas production via *Cl. perfringens*, further highlighting their antimicrobial properties [26]. In addition, *L. acidophilus* was found to have a strong inhibitory effect on *Candida* spp. [12]. Researchers isolated 32 types of good bacteria, lactic acid bacteria (LAB), from 13 honey samples sold in Malaysia, six of which were identified as *Lactobacillus acidophilus*, which showed the ability to kill harmful bacteria that are resistant to many antibiotics. The antibacterial activity of these LAB strains was tested against three types of harmful bacteria: *S. aureus*, *S. epidermis*, and *Bacillus subtilis*, which showed significant inhibition zones, indicating that they could effectively inhibit the growth of these bacteria [9]. Antimicrobial activity is often attributed to the production of organic acids, which lower pH and create an unfavorable environment for pathogens, as well as the secretion of bacteriocin-like inhibitory substances [34]. The cell-free supernatant (CFS) of *Lactobacillus acidophilus* has shown significant antimicrobial activity against various harmful bacteria, including *Staphylococcus aureus* and *Escherichia coli*. This indicates that the substances produced by *L. acidophilus* can inhibit the growth of these pathogens. *L. acidophilus* has been found to prevent the formation of biofilms by harmful bacteria like *E. coli* and *S. aureus*, making it harder for these pathogens to establish infections [35]. The antimicrobial properties of *L. acidophilus* are not only beneficial for food preservation and safety but also for therapeutic applications, such as reducing the

risk of gastrointestinal diseases and inhibiting the growth of intestinal pathogens. Overall, the diverse antimicrobial mechanisms of *L. acidophilus* make it a valuable probiotic for improving human health and food safety [3,36].

2.4. Antitumor Activity

Lactobacillus acidophilus exhibits anticarcinogenic and antitumor effects through various mechanisms. Research suggests that probiotics such as *L. acidophilus* can inhibit carcinogens, activate the host immune system, and reduce intestinal pH, potentially preventing tumor formation [37]. Additionally, *L. acidophilus* exopolysaccharides have been shown to suppress inflammation through the TLR2/STAT-3/P38-MAPK pathway, offering protection against hepatocellular carcinoma [38]. *Lactobacillus acidophilus* has demonstrated significant anticarcinogenic and antitumor effects in various studies. Studies have shown that *L. acidophilus* strains, such as KLDS1.0901 and CICC 6074, inhibit the proliferation of colon cancer cells, such as HT-29, and induce apoptosis through mechanisms involving mitochondrial membrane potential loss, reactive oxygen species generation, and the modulation of apoptotic signaling pathways [39,40]. Al-Asfour et al. [41] aimed to observe the suppression of oral carcinoma cells by *Lactobacillus acidophilus*. They exposed human oral cancer cells to *Lactobacillus acidophilus* and *Streptococcus mutans*, both separately and together, to observe their effects on cancer cell growth and survival. The results showed that *Lactobacillus acidophilus* significantly reduced the number of cancer cells, and this effect was linked to an increase in a protein that helps trigger cell death in cancer cells. This study suggests that *Lactobacillus acidophilus* may be useful in treating oral cancer as it can reduce cancer cell growth through a specific cell death mechanism, and this effect is not hindered by the presence of *Streptococcus mutans*.

2.5. Antioxidant Activity

Several studies have demonstrated the antioxidant activity of *Lactobacillus acidophilus*. Rianingsih and Sumardianto [7] demonstrated that the antioxidant activity of *L. acidophilus* is higher than that of *L. plantarum* during fermentation, which may be attributed to the production of antioxidant compounds. Furthermore, cell-free extracts of *L. acidophilus* displayed antiproliferative and antioxidant activities against the HT-29 cell line, reducing cell viability and inducing apoptosis through intrinsic and extrinsic pathways [42]. Additionally, the fermentation of seaweed extracts with *L. acidophilus* resulted in increased antioxidant activity, demonstrating the potential of this probiotic strain (LA-05) to enhance antioxidant properties [7]. Moreover, the development of antioxidant activity in whey drinks containing *L. acidophilus* LA5 has been observed, with free bacteria showing increased antioxidant activity over time, indicating the role of *L. acidophilus* in promoting antioxidant effects during fermentation processes [43]. Collectively, these findings highlight the antioxidant potential of *Lactobacillus acidophilus* under various experimental settings. Studies on the antioxidant and antitumor properties of *Lactobacillus acidophilus* are detailed in Table 1.

Table 1. Antioxidant and antitumor properties of *Lactobacillus acidophilus*.

Strain	Properties	Subjects	Findings	References
ATCC 43121	Antiproliferative, proapoptotic, and antioxidant effects	HT-29 cell line	Intrinsic pathway-dependent apoptosis was induced. Cell viability was significantly reduced to $42.2 \pm 0.01\%$ and $19.40 \pm 0.01\%$ by 5 and 8 mg ml ⁻¹	[42]
LA-5	Antioxidant activity	Wistar rats	Improve the antioxidant defenses Inhibited the tumor volumes by 59.16%, 28.29%, and 63.39%. Acidophilus milk	[44]
LA-5	Antitumor effects	Mouse xenograft breast cancer model	and PE combination significantly enhanced the ConA-, LPS-, and PHA-induced splenocyte proliferation	[37]

Table 1. Cont.

Strain	Properties	Subjects	Findings	References
LA	Antiproliferation and anti-angiogenesis properties	Gastric (AGS) and bladder (J253) cancer cell lines	Downregulated COX2 expression in AGS by 70 % and 95 %; antiproliferation and anti-angiogenesis of LA against gastric cancer by downregulating COX2 expression	[45]
ATCC 4356	Immunomodulatory and antitumor activities	Rats	ATCC 4356 exopolysaccharides suppress mediators of inflammation through the inhibition of TLR2/STAT-3/P38-MAPK pathway	[38]
CICC 6074	Anti-cancer effect	HT-29 cells induced-mouse model	CICC 6074 induced colon cancer apoptosis by up-regulating Bax, down-regulating Bcl-2, releasing Cyt c from the mitochondria into the cytoplasm, and activating Caspase-3 and Caspase-9	[40]
KLDS1.0901	Anti-cancer effect	Colon cancer cells (HT-29, Caco-2, and IEC-6 cells)	Inhibited the proliferation of HT-29 and Caco-2 cells, reduced the mitochondrial membrane potential of HT-29 cells	[39]
ATCC 4356	Anti-cancer effect	Oral squamous cell carcinoma cells (HNO97 cell line)	Exhibits antiproliferative activity against OSCC cells possibly partially via a TRAIL-induced mechanism of apoptosis	[41]

2.6. Bioactive Peptides

Lactobacillus acidophilus is a prolific producer of bioactive peptides with diverse therapeutic potentials, including antioxidant, antiviral, antifungal, and antimicrobial activities. The antioxidant properties of peptides derived from *L. acidophilus* are significantly enhanced through controlled fermentation processes because of their ability to produce antioxidant peptides during milk and meat fermentation, which can significantly enhance the therapeutic potential of fermented products [46]. Okon et al. [47] found that the use of the bacterium *Lactobacillus acidophilus*, especially in dry-cured loins, increased the antioxidant activity of proteins during fermentation and storage, with small peptides showing the highest antioxidant capacity. The results revealed that the antioxidant peptides SAGNPN, AAAAG, IHSGSVG, NVLVG, NAAKL, and GLAGA exhibited the highest antioxidant activities. Antimicrobial peptides (AMPs) produced by *Lactobacillus acidophilus* are gaining attention because of their potential applications in combating multi-resistant bacteria and food preservation. AMPs, such as the newly discovered peptide Doderlin with the sequence PTHLLKAFSKAGF, exhibit significant antimicrobial activity against pathogens such as *Candida albicans*, showing their potential in the pharmaceutical and food industries. The production of AMPs by *L. acidophilus* can be optimized under specific conditions, such as temperature, fermentation time, and yeast extract concentration, which enhance their inhibitory effects against pathogens such as *Listeria monocytogenes* [48]. The functional bioactive peptides produced by the *Lactobacillus acidophilus* strains are listed in Table 2.

Table 2. Functional bioactive peptides produced by *Lactobacillus acidophilus*.

Strain	Bioactive Peptides	Food System	Bioactivities	References
NCDC-15	KVLPVPQK (β -CN f169–176) YQEPVLGPVRGPFPIIV (β -CN f193–209)	Fermented milks	Angiotensin-I-converting-enzyme (ACE)-inhibitory peptides	[11]
DPC6026	IKHQGLPQE, VLNENLLR, and SDIPNPIGSENSE	Bovine α_{s1} -casein	Antibacterial activity against pathogenic strains <i>Enterobacter sakazakii</i> ATCC 12,868 and <i>Escherichia coli</i> DPC5063	[49]
LA-5	GVSKVKEAMAPK	Bovine β -CN	Antioxidant	[50]
LA-5	DVENLHLPLPL	Bovine β -CN	ACE-inhibitory activity	[51]
LA-5	GLDIQKVAGT, GLDIQKVAGTW	Bovine β -LG	ACE-inhibitory activity, antibacterial activity	[52]
LA	NEPTHLLKAFSKAGFQ	Milk, yogurt, cheeses	Antimicrobial activity	[48]
LA	Acidocin 4356	Whey	Antimicrobial activity	[53]

Table 2. Cont.

Strain	Bioactive Peptides	Food System	Bioactivities	References
CGMCC1.1878	SLPS	Yogurt	Inhibited <i>Staphylococcus aureus</i> growth	[54]
ATCC 4356	SLA	Kasar cheese	Increased the bioactivity of the cheese	[10]
NX371	CAAATCAGTAATATGGAAAATC	Milk and cheese	Damaged the cell wall, and disrupted the membrane structure, resulting in leakage of intracellular ATP.	[55]

2.7. Extracellular Polysaccharides Production

The production and application of EPS from *L. acidophilus* and other LAB strains holds significant promise for enhancing food quality and promoting human health through their multifunctional properties [56]. Extracellular polysaccharide production by *Lactobacillus acidophilus*, particularly strain BCRC 10695, has been extensively studied for its beneficial effects and potential applications in food and pharmaceutical industries. The production process involves optimizing the composition of the culture medium, including carbon sources such as sucrose, nitrogen sources such as yeast extract, and surfactants such as polysorbate 80. Using response surface methodology (RSM), the optimal concentrations were determined to be 10.15 g/L sucrose, 25 g/L yeast extract, and 2 g/L polysorbate 80, resulting in an EPS yield of 923.7 mg/L after 96 h of cultivation [57]. *Lactobacillus acidophilus* is known for its production of extracellular polysaccharides (EPS), which have various functional properties, such as enhanced taste, texture, and health benefits, including immunomodulatory and anticancer activities. Recently, in Deepak et al. [58], EPS was found to be effective under both normal- and low-oxygen conditions, increasing the expression of certain genes related to cancer inhibition. EPS production was optimized using specific design methods, achieving a peak EPS concentration of 597 mg/L during batch cultivation. Abedfar et al. [59] isolated *Lactobacillus acidophilus* from rice bran sourdough, which produces a special substance (EPS) with high carbohydrate content and antioxidant properties. They used various methods to identify and analyze EPS, confirming its structure and components such as glucose, galactose, and maltose. The EPS from *Lactobacillus acidophilus* showed significant antioxidant activity, suggesting that it could be useful as a functional ingredient. Overall, the production and application of EPS from *L. acidophilus* holds significant promise for enhancing food quality and promoting human health through their multifunctional properties.

3. Applications of *L. acidophilus* in Fermented Food Products

3.1. Cheese

Lactobacillus acidophilus plays a crucial role in various cheese types because of its probiotic properties and bioactive potential. It is a Gram-positive lactic acid bacterium that can produce antimicrobial peptides, inhibit the growth of pathogenic bacteria, and enhance the overall quality of dairy products [60]. *Lactobacillus acidophilus* plays a crucial role in cheese production as a probiotic microorganism, enhancing the survival and growth of beneficial bacteria in dairy products. Research has shown that incorporating *L. acidophilus* into cheese formulations can increase probiotic counts, improve viability during storage, and provide health benefits for consumers. Studies have demonstrated that the addition of *L. acidophilus* to cheese can protect against spoilage organisms, enhance the texture and sensory properties of the product, and contribute to the development of synbiotic cheeses containing prebiotics such as pectin, inulin, and maltodextrin [4,10,61]. Neto et al. [62] developed and tested *L. acidophilus* microparticles with L-cysteine hydrochloride and/or ascorbic acid for use in Reino cheese. They monitored the survival of these microorganisms for more than 60 days and analyzed the best formulation in terms of size and shape. Three cheese types were prepared: one without *Lactobacillus acidophilus*, one with free microorganisms, and one with microparticles. The cheeses were tested for microbiological quality and *Lactobacillus* viability after 1 and 25 d of ripening. The best microparticle

formulation showed over 80% encapsulation yield and 33% lactobacilli survival after 60 d. Cheese containing these microparticles had the highest viability of *lactobacilli*, indicating better protection and potential for industrial use. Diniz-Silva et al. [4] determined the effect of *Lactobacillus acidophilus* along with oregano and rosemary essential oils in the production of Minas Frescal cheese during refrigerated storage. The essential oils improved the efficiency of probiotics, which reduced *E. coli* counts in the first 15 days of storage, and this reduction was linked to the presence of terpenes in the cheese. Over time, the terpenes decreased, but improved the aroma, flavor, and overall appeal of cheese. Together, these essential oils could help control *E. coli* in probiotic cheese, but the amounts need to be carefully chosen to avoid negative taste effects. In a previous study, Lopes et al. [60] evaluated the effects of *L. acidophilus* on Ricotta cheese prepared from goat milk. The addition of probiotics made it softer, stickier, and more cohesive, changed its color, and increased its yield. Microencapsulation helped the probiotics survive better and improved the texture and flavor of cheese. Cheese with microencapsulated probiotics exhibited no moisture loss, less breakdown of proteins and acids, and a better taste with fruity and floral notes while maintaining high probiotic levels even in simulated digestion. Mojaddar Langroodi et al. [61] examined the effects of *Anethum graveolens* essential oil (DEO) and *Lactobacillus acidophilus* to prevent *Escherichia coli* O157 in cheese. Different concentrations of DEO and *L. acidophilus* reduced *E. coli* O157 counts and improved taste and pH. The combination of DEO and *L. acidophilus* strongly inhibited *E. coli* O157, thereby confirming DEO's positive role in dairy products. Furthermore, the survival of *L. acidophilus* during the fermentation and ripening stages of cheese production is essential for maintaining probiotic viability and ensuring the desired health effects for consumers.

3.2. Yogurt

Lactobacillus acidophilus plays a crucial role in yogurt production by binding aflatoxin B1, enhancing probiotic properties, and improving the quality of the final product. Research indicates that *L. acidophilus*, when combined with inulin and fructooligosaccharides (FOS), can increase coagulation time, extend the shelf life of yogurt, and positively affect sensory attributes [63,64]. Furthermore, incorporating *L. acidophilus* into yogurt formulations has been shown to increase the number of viable bacteria, improve viscosity, and enhance physicochemical properties during storage [65]. Additionally, probiotic yogurt fortified with *L. acidophilus* has been found to effectively reduce lactose intolerance symptoms, making it a recommended treatment option for lactose intolerant individuals [66]. Mousavi et al. [67] prepared synbiotic yogurt with *Lactobacillus acidophilus* and flaxseed, focusing on optimizing probiotic count, texture, and taste. They used a central composite design (CCD) with flaxseed concentration (0–4%) and storage time (1–28 d) as variables. The results showed that flaxseed concentration and storage time significantly affected probiotic viability, texture, and the sensory attributes of yogurt. Moreover, the addition of flaxseed increased *L. acidophilus* growth, viscosity, hardness, cohesiveness, gumminess, and water-holding capacity while reducing syneresis and adhesiveness. However, high flaxseed levels reduce sensory appeal. Overall, 4% flaxseed in probiotic yogurt resulted in a functional product with 76.8% desirability that maintained good properties for approximately 13 days during refrigerated storage. Masoumi et al. [65] prepared tested probiotic yogurt with *L. acidophilus* in 55 lactose-intolerant patients. The experimental group had fewer symptoms and lower hydrogen levels than the control group, indicating that probiotic yogurt can help to treat lactose intolerance. Patients were divided into two groups: one group received non-probiotic yogurt, and the other received probiotic yogurt for one week. These results suggest that probiotic yogurt is a safe and effective treatment to reduce lactose intolerance. Hasani et al. [68] examined the effect of different concentrations of barley bran on the growth of *Lactobacillus acidophilus* and the physical and taste properties of low-fat yogurt over 28 days. The results showed that the addition of barley bran increased the bacterial count and thickness of yogurt compared to the control group. High barley bran levels (1.2%) reduced taste scores but increased thickness. The 0.6% barley bran level maintained

good taste and had a bacterial count higher than the minimum acceptable level, making it the recommended amount for making symbiotic yogurt. Ertem et al. [69] tested the effects of *Lactobacillus acidophilus* and dry white mulberry and walnut paste (Gobdin) on probiotic yogurt properties. Six types of yogurt were prepared with different amounts of Gobdin and bacterial combinations, and then stored at 4 °C. The highest *L. acidophilus* count was observed in 5% Gobdin yogurt on day 7, whereas the lowest count was observed in control yogurt on day 21. Despite this decline, all probiotic yogurts maintained beneficial bacterial levels above 8 log CFU/g for 21 days. The 5% Gobdin yogurt showed the highest acceptability on day 1; however, all yogurts scored well overall. The study concluded that yogurt containing 5% Gobdin and *L. acidophilus* is a viable new functional food. Overall, the presence of *L. acidophilus* in yogurt contributes to its health benefits, quality, and consumer acceptance.

3.3. Fermented Milk

Fermented milk is a dairy product produced through a fermentation process involving lactic acid bacteria, such as *Lactobacillus acidophilus* and *Lactobacillus casei*, which play a crucial role in the development of the characteristic flavors of the product. When combined with other probiotics such as *Bifidobacterium* and *Lactobacillus acidophilus*, fermented milk can act as a therapeutic agent for gastrointestinal disorders, offering antimicrobial, anticarcinogenic, and antioxidant activities [70]. The fermentation of milk with specific strains, such as *L. acidophilus* LA-5, can lead to various health benefits, including improved digestion, immune response stimulation, and potential protection against intestinal diseases. *Lactobacillus acidophilus* plays a crucial role in milk fermentation by enhancing various properties and health benefits. It has been shown to reduce serum cholesterol levels, stabilize the microbiota, stimulate the immune system, improve lactose digestion, and potentially combat cancer cells [71]. *Lactobacillus acidophilus* LA-5 is a probiotic used in fermented milk, but grows slowly in milk, which is not ideal for commercial production. Therefore, understanding its nutrient requirements can help speed up the fermentation process. The essential nutrients for *L. acidophilus* LA-5 include amino acids such as Asn, Asp, Cys, Leu, and Met; vitamins such as riboflavin; and nucleotides such as guanine and uracil, along with the metal ion Mn^{2+} . The addition of these nutrients to milk can significantly reduce fermentation time by up to 9 h, which is beneficial for producing fermented milk more quickly and efficiently. When these nutrients are added, the viable cell count of *L. acidophilus* LA-5 increases, indicating that more live bacteria are present in the final product, enhancing its probiotic benefits [6]. Dabaj et al. [70] examined the effects of two *Lactobacilli* strains, *L. casei* 431 and *L. acidophilus* La-5, on fermented milk. They identified 105 different compounds, with ketones being the most common, and found that *L. acidophilus* La-5 produced more key flavor compounds. This research suggests that *L. acidophilus* La-5 could be a good starter culture for producing tasty fermented dairy products because of its ability to produce important flavor compounds. Li et al. [72] found that adding onion juice to milk helps the bacteria *Lactobacillus acidophilus* grow better and produce more acid. This leads to higher cell counts and better storage stability, likely due to the presence of polyphenols, sulfur compounds, fructans, and minerals in onion juice. Onion juice also slightly improves the antioxidant capacity of fermented milk, resulting in shorter fermentation times, better antioxidant properties, and richer flavors, making it useful for industrial applications.

3.4. Fortified Milk Products

Fortified milk products are dairy beverages enriched with additional nutrients or bioactive compounds, which enhance their nutritional value and health benefits. Research has shown that fortifying milk with extracts, such as pomegranate peel [8] and date extract [73], and probiotic strains, such as *Lactobacillus acidophilus* LA-05, ATCC 4356, and LA-308 [74], can significantly increase the antioxidant capacity, total phenolic content, and probiotic content of the final product. These fortified milk products undergo specific preparation methods, including fermentation with beneficial bacteria, such as *L. acidophilus*, to

ensure the incorporation of added nutrients while maintaining the quality and safety of the beverage. Elkot et al. [75] developed a synbiotic ice cream using camel milk and black rice powder (BRP), enhancing physicochemical properties and *Lactobacillus acidophilus* viability during storage. Black rice powder was incorporated into ice cream blends at different levels, resulting in increased overrun, viscosity, and melting resistance, while decreasing the freezing point. Sensory evaluation favored treatments with 25% BRP, showing an improved viability of probiotic bacteria over 60 d of storage. By fortifying milk with various ingredients, manufacturers can offer consumers a diverse range of functional dairy products that not only provide essential nutrients but also offer additional health-promoting properties.

3.5. Meat Products

Lactobacillus acidophilus CRL641, a probiotic strain commonly used in meat fermentation, has been shown to inhibit spoilage bacteria in refrigerated meat systems, thereby enhancing shelf life and quality [76]. Studies have demonstrated that the incorporation of *L. acidophilus* into meat products can improve flavor, texture, and color, making them more acceptable to consumers [77,78]. Additionally, the use of *L. acidophilus* LA-308 in fermented meat spreads has been linked to enhanced tenderness and flavor of the meat, demonstrating the potential of probiotics to improve meat quality and its sensory characteristics [79]. Furthermore, the fermentation process involving *L. acidophilus* has been found to reduce protein oxidation levels, increase water- and oil-holding capacities, and positively impact the texture of meat analogs, highlighting the multifaceted benefits of probiotic fermentation in meat products. Arief et al. [76] assessed the effects of probiotic lactic acid bacteria, specifically *Lactobacillus acidophilus* IIA-2B4, on the pH, texture, color, fatty acid profiles, flavor compounds properties of the fermented beef sausages. The results indicated significant differences in these attributes, highlighting the potential influence of probiotics on the overall quality and characteristics of the sausages. The bioprotective extracts of *L. acidophilus* and *Lactobacillus curvatus* CRL705 exhibited antimicrobial activity against *Lactobacillus sakei* CRL1407 in refrigerated meat models, BE-1 completely inhibited the spoilage strain, and BE-2 exerted bacteriostatic effects. The antimicrobial activity correlated with inhibitory treatment efficacy, while flow cytometry revealed a high percentage of dead cells with BE-1 treatment, indicating the potential of lactic acid bacteria as biocontrol agents to extend the shelf life of fresh meat [80].

3.6. Baking Products

Lactobacillus acidophilus has a significant effect on fermented bread, especially when encapsulated in alginate/fish gelatin, as shown in various studies. The encapsulation of *L. acidophilus* in alginate/fish gelatin capsules can enhance its viability during baking and storage, with a 3% matrix exhibiting the highest protective effect [81]. Additionally, the addition of xanthan gum and maltodextrin to the alginate matrix improves the encapsulation yield and provides better protection for the probiotic during baking and storage, ensuring its viability [82]. Furthermore, the double-layered encapsulation of *L. acidophilus* using sodium alginate and chitosan enhanced the survival of the bacteria, particularly during storage, indicating its potential as a beneficial component of bread products [83]. Overall, these findings highlight the positive effects of *L. acidophilus* on the quality and viability of probiotics in fermented bread, thereby demonstrating its potential as a health-promoting ingredient in baking products. Bread is popular because of its variety and ease of use. Truong et al. [82] prepared probiotic cream bread using microencapsulated *Lactobacillus acidophilus* with different combinations of alginate, maltodextrin, and xanthan gum. Xanthan gum improved the encapsulation yield by more than 92% for some samples. During baking, the viability of *L. acidophilus* decreased less in samples containing xanthan gum and maltodextrin than in other samples. In simulated stomach and intestinal conditions, the combination of xanthan gum and maltodextrin provided the best protection for probiotics, acting as buffers and protective agents, and improving probiotic survival. Mirzamani et al. [83] prepared the probiotic bread using *Lactobacillus acidophilus* ATCC 4356 to improve

its resistance to stomach conditions and heat during bread baking. The first layer, 1% alginate, exhibited the best survival under stomach-like conditions. The second layer used 0.5% chitosan, which made the probiotics more heat resistant. The study found that higher levels of alginate and xanthan reduced the encapsulation efficiency; however, 1% xanthan resulted in the best yield. Chitosan provided a smoother surface, which protected probiotics. In bread, 1% chitosan in the outer layer helped the probiotics survive better, both when fresh and after 24 h. An overview of *Lactobacillus acidophilus* fermentation for the development of food products is presented in Table 3.

Table 3. Overview of *Lactobacillus acidophilus* fermentation for the development of food products.

Strains	Fermentation Temperature and Time	Food Products	Findings	References
LA-5	At 22 °C for 18 h	Milk products	Used as a bacterial supplement in fermented products	[84]
LA-5	At 37 °C for 15 h	Sweet whey and skim milk	Skim milk microparticles allowed an increase in the viability of the probiotic	[84]
LA-5	At 37 °C for 8 h	Fermented milk	Regulated the growth of probiotics in fermented milk prepared by a single probiotic strain	[6]
LA-5	At 37 °C for 48 h	Yogurt	The addition of microencapsulated <i>Lactobacillus acidophilus</i> LA-5 improved the physiochemical properties of the yogurts	[85]
ATCC 4356	At 42 °C for 4 h	Low-fat yogurt	High levels of barley bran (1.2%) decreased sensory prosperity scores and led to a viscosity increment; the amount of <i>L. acidophilus</i> and viscosity in samples containing barley bran was significantly higher than the control group	[68]
ATCC 4356	At 37 °C for 18 h	Cheese whey	The production of valuable organic acids including pyruvate, propionate, acetate, lactate, formate, and butyrate	[86]
LA-3	At 37 °C for 18 h	Reino cheese	Showed the highest lactobacilli viability (8.49 ± 0.08 Log CFU/g), provided additional protection to the <i>L. acidophilus</i> microorganism, benefiting microbial cell survival, and therefore resulted in a ripened Reino cheese	[62]
LA-308	At 37 °C for 24 h	Meat analogs	Improved water-holding capacity and sensory properties of meat analogs, as well as reducing hardness and protein oxidation levels	[79]
CRL641	At 37 °C for 24 h	Refrigerated meat	Inhibited <i>Latilactobacillus sakei</i> CRL1407; improved sensorial effects of both extracts	[87]
LA-5	At 37 °C for 24 h	Chicken meat spread	Showed positive effects on its sensory attributes, resulting in an appreciable-quality probiotic chicken meat spread	[78]
IIA-2B4	At room temperature for 24 h	Fermented beef sausage	The development of unique flavor compounds including acid, alcohols, aldehydes, aromatics, ketones, sulfur, hydrocarbons, and terpenes	[76]
ATCC 4356	At 37 °C for 24 h	Probiotic bread	The combination of maltodextrin and xanthan gum in the alginate matrix provides the best survivability during storage	[82]
LA-5	At 37 °C for 24 h	Bread	The application of alginate and chitosan in the microcapsules can protect the <i>L. acidophilus</i> and it is considered as an effective method in probiotic bread production	[83]

4. Applications of *L. acidophilus* in Fermented Plant-Based Beverages

4.1. Fruit Juices

The effect of *Lactobacillus acidophilus* on fruits varies depending on fruit type and fermentation process. Research has shown that *L. acidophilus* can improve the nutritional value, antioxidant activity, enzymatic browning, and sensory attributes of fruit juices such as apple [17], yellow mombin [88], papaya [89], and pear [90]. Fermentation with *L. acidophilus* can lead to changes in pH, organic acids, sugars, and the production of beneficial compounds such as lactic acid, enhancing the quality of the final product. Additionally, the presence of *L. acidophilus* in fruit juices can contribute to improved microbial stability, nutritional value, and potential health benefits, owing to its probiotic nature. Different fruits exhibit varying responses to *L. acidophilus* LA-02 fermentation, highlighting the impor-

tance of considering a specific fruit matrix when utilizing this probiotic bacterium in fruit products [88,91]. Peng et al. [91] tested different types of *Lactobacillus acidophilus* bacteria and fermented nine kinds of cloudy apple juice. Changfu contained the most bacteria and acetic acid, Golden Delicious contained the most sugar, and Qinguan contained the most lactic acid and esters. Changfu, Qinguan, and Golden Delicious were the best for producing tasty, aromatic, and highly bacteria-fermented cloudy apple juices. Ribeiro et al. [88] aimed to test yellow mombin juice as a carrier for the probiotic *Lactobacillus acidophilus* LA-02. The best conditions for making the juice were found to be a pH of 6.4 and 16 h of fermentation, resulting in high probiotic viability. The juice had good chemical properties, including a high phenolic content and antioxidant activity. It also showed antibacterial effects against certain harmful bacteria and remained stable for 28 days at 4 °C, with no significant loss of probiotic viability under simulated stomach conditions. Karbasi et al. [92] unbleached date syrup to make a non-dairy probiotic drink with *Lactobacillus acidophilus* and *Lactobacillus rhamnosus*. They examined growth, pH, acidity, sugars, organic acids, phenolic content, antioxidant activity over 48 h at 37 °C, and cell viability for 4 weeks at 4 °C. Both bacteria grew well in date syrup, especially *L. acidophilus*, which showed better growth and higher sugar consumption. *L. acidophilus* produced more gluconic acid than lactic acid and maintained higher cell viability after 4 weeks. The total phenolic and antioxidant activities were similar for both the bacteria. Li et al. [90] fermented browned pear juice with *Lactobacillus acidophilus* CH-2 to reduce browning effects. Various tests have been conducted to analyze color changes and antioxidant activity. The results showed a significant reduction in browning and color difference after fermentation. The degree of browning and color difference decreased notably, and the fermentation supernatant exhibited high antioxidant activity. The reduction in browning is likely due to two factors: the production of strong antioxidant metabolites by *L. acidophilus* CH-2 and the breakdown of browning products during fermentation. Sheng et al. [93] reported that mixed cultures of *Lactobacillus acidophilus*-26 improved the quality of fermented red globe grape juice more than single cultures. They increase the number of viable bacteria, reduce total sugars, and enhance antioxidant properties, pH, and functionality. Mixed cultures also enriched the juice aroma with esters, acids, alcohols, aldehydes, and ketones. They significantly increased the content of compounds, such as acetic acid, ethyl acetate, and 2-hexenol, contributing to the unique flavor of the juice.

4.2. Vegetable Juices

Lactobacillus acidophilus ferments vegetable juices by utilizing the sugars present in the juices to produce lactic acid, which lowers the pH and increases the acidity of the juice, thereby enhancing its shelf life and safety. This process also results in the production of beneficial organic acids and vitamins, which contribute to the nutritional value of juice [94,95]. The fermentation process involves bacteria metabolizing glucose and other sugars, which leads to a reduction in glucose levels and an increase in lactic acid concentrations, as observed in various studies [96,97]. The addition of nutrients such as glucose, lactose, and prebiotics can further accelerate the fermentation process, improve the growth of *Lactobacillus acidophilus*, and inhibit the growth of contaminating microorganisms, thus enhancing the flavor and taste of juice [98]. Lavinia et al. [99] tested cucumber, white cabbage juices, and red cabbage juices for growing *Lactobacillus acidophilus* to make probiotic products. Cabbage juice showed a rapid drop in pH and a large increase in microbes, whereas cucumber juice had the fastest acidification rate. All juices complete fermentation within approximately 6 h. Using statistical methods, the study grouped variables into three clusters and used Principal Component Analysis (PCA) to simplify the data, explaining 80.43% of the total variation. Tenguria et al. [100] explored the use of probiotic bacteria in non-dairy products like carrot juice to avoid issues like milk allergies and lactose intolerance. Carrots are nutritious and popular particularly in India. The study tested carrot juice with *Lactobacillus acidophilus* for 30 days, comparing it to juice with benzoic acid and a control with no additives. They evaluated properties such as color, smell, texture, pH, and total soluble solids. The results

showed that carrot juice with probiotics maintained a better quality than the control and was similar to that of fresh juice. This suggests that carrot juice is a good medium for delivering probiotics.

4.3. Grains and Nuts Beverages

Lactobacillus acidophilus plays a significant role in enhancing the nutritional value and bioavailability of bioactive compounds in various beverages. Research indicates that the fermentation of beverages with *Lactobacillus acidophilus* LA-20079 leads to an increase in total phenol and flavonoid content, contributing to the antioxidant activity of the beverage. Additionally, the amino acids released during fermentation affect the flavor profile and biological value of the beverage, with specific amino acids such as glutamic acid, aspartic acid, threonine, serine, and alanine being significantly higher in fermented beverages [101]. Igwebuike et al. [102] evaluated the properties of a drink made from partially hydrolyzed tigernut milk and beetroot juice fermented with the probiotic *Lactobacillus acidophilus* LA-02. The drink was tested in different beetroot juice concentrations (10%, 20%, and 30%) and compared to a control without beetroot juice. The results showed that hydrolysis decreased starch content and increased sugar content. The pH decreased over time, and the titratable acidity increased. All samples supported the growth of *L. acidophilus* LA-02, with a significant increase in the bacterial counts. Sensory evaluation indicated varying degrees of likeness for color, aroma, mouthfeel, taste, and overall acceptability. A 20% beetroot juice blend was recommended based on sensory preferences, and the drink showed potential as a probiotic.

4.4. Bean Milk

Lactobacillus acidophilus LA-05, a probiotic strain, has been extensively studied for its beneficial effects on various food products. Research has indicated that *L. acidophilus* thrives in bean milk, contributing to the fermentation process and enhancing the properties of the product. Studies have shown that the addition of bean components, such as faba bean, can positively affect pH reduction and the fermentative capacity of the microflora, leading to improved outcomes in terms of bacterial counts, lactic acid content, and overall product quality [103–107]. Furthermore, optimizing the carbon source and concentration in the fermentation medium is crucial for the growth and phenolic production of *L. acidophilus*, highlighting the importance of the medium composition in maximizing the potential of probiotics in bean milk. Additionally, the growth kinetics and survival of *L. acidophilus* in different types of milk, including black rice milk, further emphasize the adaptability and efficacy of the strain in diverse food matrices, demonstrating its potential in bean milk applications [5]. Boudjou et al. [103] tested whole ground faba bean in kefir to observe bacterial growth during storage. The addition of 4% faba bean flour increased the abundance of bacteria, increased acidity, and lowered pH over 28 d at 4 °C. Faba bean flour was found to significantly enhance the survival and growth of probiotic bacteria like *Lactobacillus acidophilus* and *Bifidobacterium lactis* during the storage of kefir, which is a fermented milk drink. This is crucial to maintain the health benefits of kefir over time. The supplementation of kefir with faba bean flour extended its shelf life by maintaining the viability of probiotic bacteria and preventing spoilage. This makes it a practical option for both producers and consumers. Aduol et al. [108] fermented cowpea milk using *Lactobacillus acidophilus*. The fermentation process led to a significant decrease in crude fat (except for one culture) and carbohydrates, whereas crude fiber was not detected. Protein content increased slightly in each culture. Microbial growth increased during the first two weeks of storage at 4 °C and then decreased on the 28th day. Sensory attributes, such as taste, texture, and overall acceptability, showed no significant differences, but aroma and appearance showed significant differences. This study showed that fermenting cowpea milk improved its nutritional quality by developing a yogurt-like product. This suggests the need for further research to enhance sensory acceptability and explore the potential health benefits through in vivo studies. Recent studies on *Lactobacillus acidophilus* in the

fermentation of plant beverages are tabulated in Table 4. Recent research on the application of *L. acidophilus* in the fermentation of different food products and plant-based beverages is shown in Figure 2.

Table 4. Recent studies on *Lactobacillus acidophilus* in the fermentation of plant beverages.

Strains	Fermentation Temperature and Time	Plant Beverage	Findings	References
ATCC 314	At 37 °C for 12 h	Pigeon pea (<i>Cajanus cajan</i>) product	Higher viability and good sensory attributes; it should be considered suitable for a pigeon pea-based fermented probiotic product	[109]
La-05	At 37 °C for 24 h	Incorporation in vegan milks	The utilization of chitosan coating in the alginate microparticle is recommended only for increasing the survival of the probiotic cultures in vegan milks	[110]
LA-02	At 37 °C for 17 h	Different fruit juices	In apple juice, there is an increase in probiotic viability. In orange juice, microencapsulation also showed satisfactory results, as only microencapsulated probiotics were able to survive for 63 days, showing high viability	[111]
PTCC 1643	At 37 °C for 24 h	Peach juice	The biological activities of peach juice including Maillard reaction inhibition, superoxide anion radical-scavenging activity, Fe-reducing power, and anti-inflammatory activity were markedly increased during the fermentation period	[112]
CH-2	At 37 °C for 24 h	Pear juice	Metabolites were produced with strong antioxidant activity via the fermentation by <i>L. acidophilus</i> CH-2 for the degradation of browning products by fermentation	[90]
CICC®20709	At 36 °C for 24 h	Loquat juice	Significantly enhanced the antioxidant activity of loquat juice	[113]
LA-26	At 36 °C for 24 h	Grape juice	Better tastes, such as sourness, and aromas but also decreased the amount of bitterness and levels of sulfur compounds	[93]
LA-20079	At 37 °C for 48 h	Fermented beverage	Highest pH decreases and sugar consumption showed a much better growth rate	[92]
TISTR 2365	At 36 °C for 24 h	Fermented rice (khao mak) sap beverage	Significant increase in total phenolic contents and DPPH radical scavenging activities	[101]

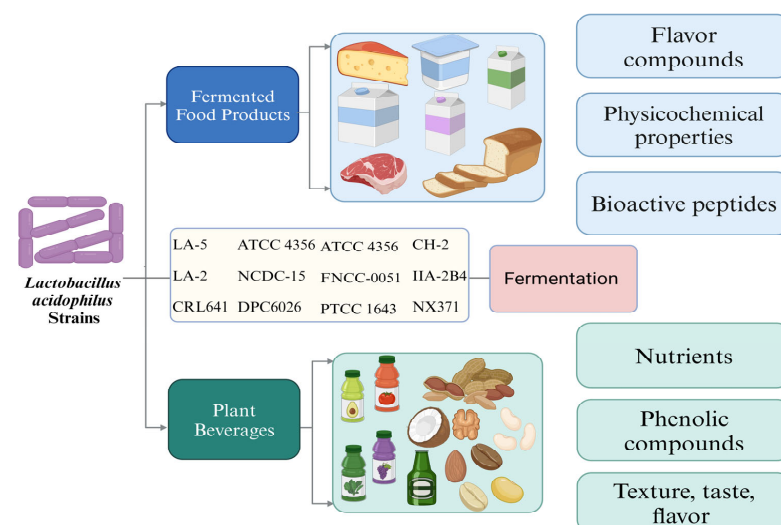


Figure 2. Application of *L. acidophilus* in the fermentation of different food products and plant-based beverages.

5. Safety of *Lactobacillus acidophilus*

Lactobacillus acidophilus is widely recognized for its safety and beneficial effects as a probiotic, particularly in the dairy industry and for human consumption. Studies have shown that *L. acidophilus* is a genetically stable and low-diversity species, with commercial strains essentially identical at the sequence level, indicating a high degree of safety and consistency in its use [114]. The Norwegian Scientific Committee for Food Safety assessed various strains of *L. acidophilus*, including W37, DDS-1, La-5, and La-14, and concluded that these strains are unlikely to cause adverse health effects in a generally healthy population with a mature gastrointestinal tract. However, they noted a lack of data on long-term adverse effects in infants and young children, suggesting that prolonged daily consumption by individuals with immature gastrointestinal tracts could potentially have unknown long-term effects [115]. Further research involving healthy adult volunteers consuming high doses of *L. acidophilus* 821-3 demonstrated that the strain persisted in the gastrointestinal tract for several days post-consumption without causing severe adverse effects, indicating its potential as a safe probiotic [116]. The FAO/WHO guidelines emphasize the need for comprehensive safety assessments of lactic acid bacteria, including *L. acidophilus*, to ensure consumer safety, particularly regarding antibiotic resistance and toxin production [117]. Despite the extensive use of lactic acid bacteria in fermented foods globally, which has not shown general harm to consumer health, it is crucial to consider that no live bacteria can be guaranteed to pose zero risk, especially in immunocompromised individuals. Specific strains such as *L. acidophilus* BIOTECH 1900 have demonstrated strong antagonistic activity against pathogens and favorable safety profiles, including susceptibility to most antibiotics and a lack of hemolytic activity, making them suitable for probiotic use [118]. Although isolated cases of lactobacillemia have been reported in at-risk populations, the overall biological risk of *L. acidophilus* remains negligible, and new guidelines have proposed granting a 'long-standing presumption of safety' status to the *Lactobacillus* genus based on its historical use [119]. Additionally, the long-term use of probiotics such as *L. acidophilus* has shown potential benefits in modulating gut microbiota and improving metabolic health, further supporting its safe consumption [120]. Overall, the extensive body of research supports the safety of the long-term consumption of *L. acidophilus*, and ongoing studies are needed to address specific concerns in vulnerable populations.

6. Challenges and Opportunities

Lactobacillus acidophilus (*L. acidophilus*) presents significant challenges and promising opportunities for commercial food production. One of the primary challenges is its complex nutritional requirements and low cell density during industrial production, which restrict its broader application. Optimizing fermentation conditions, such as adjusting the carbon source and nutrient composition, has been shown to significantly enhance cell density and growth rates, as demonstrated by the increased viable counts of *L. acidophilus* IMAU81186 under optimized conditions [121]. Additionally, the fastidious nature of *L. acidophilus* in terms of its nutritional needs and sensitivity to environmental conditions, such as metal ion stress, further complicates its industrial use. Research has shown that, while certain metal ions such as Zn and Mn do not affect growth, others such as Cu can be highly toxic, necessitating the selection of stress-resistant strains or adaptation techniques to improve tolerance and productivity [122].

Despite these challenges, *L. acidophilus* offers numerous opportunities, particularly for sustainable food production. Its ability to ferment food enhances safety, prolongs shelf life, and augments nutritional value by producing lactic acid and bacteriocins, which inhibit foodborne pathogens and spoilage organisms [2,123]. Moreover, *L. acidophilus* can be utilized in the production of single-cell protein (SCP) from organic waste, providing a sustainable protein source while addressing waste management issues [124]. The probiotic properties of *L. acidophilus* also present significant health benefits, particularly in gastrointestinal health, as seen in products such as acidophilus milk, which has been shown to prevent various digestive tract problems and improve overall gut health [125]. Further-

more, the development of genetically modified strains, although challenging owing to regulatory and technical barriers, has the potential to enhance the functional properties and industrial applicability of *L. acidophilus* [126]. Studies have also shown that different strains of *L. acidophilus* exhibit varying degrees of resistance to antibiotics and gastrointestinal conditions, which can be leveraged to select the most suitable strains for specific applications [127,128]. The adaptive laboratory evolution (ALE) approach has also been successful in increasing the acid tolerance of related strains, suggesting a potential method for improving the robustness of *L. acidophilus* in acidic environments, which is crucial for its application in various fermentation processes [129]. Overall, while the commercial production of *L. acidophilus* faces several challenges, ongoing research and technological advancements continue to unlock new opportunities, making it an invaluable asset in the food industry.

7. Conclusions

Lactobacillus acidophilus is a probiotic bacterium with several advantages for human health and technological applications in food and beverage fermentation. Some of its benefits include enhanced immunity; the promotion of gut health; and antioxidant, antitumor, and antimicrobial effects. The growth of *L. acidophilus* is influenced by various factors such as salinity, temperature, carbon sources, and nutrient availability, which can affect the survival and bioactive potential of fermented products. *L. acidophilus* also has proteolytic effects, which help break down proteins and release bioactive peptides with various health benefits. This bacterium is used in the fermentation of dairy products, cereal beverages, soymilk, fruit and vegetable juices, and other functional food preparations to improve nutritional value, taste, and probiotic delivery. To obtain the most *L. acidophilus* from various food products and beverages, it is essential to control the fermentation conditions. Future research should focus on enhancing their viability and probiotic characteristics through various encapsulation techniques, and in vivo studies on the synergistic use of prebiotics, which can improve the survival and functionality of the digestive system.

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