Application of Postbiotics Produced By Lactic Acid Bacteria in the Development of Active Food Packaging

Seyed Ahmad Hosseini 1,2, Amin Abbasi 3,4, Sahar Sabahi 1,5, Nader Khani 3

1 Department of Nutritional Sciences, School of Allied Medical Sciences, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran
2 Nutrition and Metabolic Diseases Research Center, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran
3 Department of Food Science and Technology, Faculty of Nutrition & Food Sciences, Tabriz University of Medical Sciences, Tabriz, Iran
4 Drug Applied Research Center, Tabriz University of Medical Sciences, Tabriz, Iran
* Correspondence: Sabahi-s@ajums.ac.ir (S.S.);
Scopus Author ID 57222359418

Abstract: The use of postbiotics in the food industry is a common way to prevent food spoilage. Postbiotics are metabolites produced by probiotic bacteria that have many health effects. Non-toxicity and safety of postbiotics and their ability to inhibit microorganisms that cause food spoilage are the most important features of postbiotics in the use of these compounds in the food industry. In studies on postbiotics in the food industry, the use of these compounds as a way to control microbial spoilage of substances may interfere with the function (factors in the food matrix) in the function of postbiotics. Therefore, the use of postbiotics in the form of food packaging can be more effective. Therefore, due to their unique properties, postbiotics have received much attention in the food industry and can be used as a new approach in food packaging.

Keywords: probiotics; postbiotics; lactic acid bacteria; active packaging; antimicrobial activity.

© 2021 by the authors. This article is an open-access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https://creativecommons.org/licenses/by/4.0/).

1. Introduction

In recent years, the collection of synthetic plastics has led to the expansion of a thoughtful environmental challenge [1]. About eight million metric tons of plastic end up in the ocean every year, threatening the surrounding environment and human health. Current packaging materials consume our limited natural resources and lead to a different kind of waste. On the other hand, pathogenic microbial contamination and infections through surface contact of solid materials are major public health concerns. Today, biodegradable films and coatings are recognized as a new strategy to solving this obstacle by making inexpensive, sufficient, and renewable elements. In these contexts, there has been a growing interest in the food packaging field to detect novel and safe biopolymers and natural antimicrobial compounds [2,3]. Lactic acid bacteria (LAB) are a group of beneficial microbes whose derived metabolites possess a significantly vital role in food packaging. LAB is an essential part of food and is contained in the matrix of fermented food as safe and functional compounds, the consumption of which is directly related to health [4]. LAB in the matrix of fermented food uses suitable compounds (prebiotic-like elements) and then produces a variety of bioactive elements such as peptides, fatty acids, bacteriocins, and organic acids, which have important biological and physiological characteristics. The application of LAB in food packaging is a novel strategy to hinder the
growth and development of foodborne pathogenic germs. The antimicrobial mechanism of the lab in the food matrix is through the reduction of pH, the prevention of toxin production by pathogens, and the production of growth inhibitory compounds. In this regard, the main antimicrobial mechanisms of LAB are related to their unique action of secreting antimicrobial compounds (postbiotics) [4,5]. Some of the antimicrobial agents generated by LABs include lactic acids, acetic acids, hydrogen peroxide, reuterin, nisin, pediocin, or other bacteriocin-like substances, which among these antimicrobial agents, nisin has been approved by the Food and Drug Administration (FDA) and extensively apply in a various sector of the food industry as natural and safe antimicrobial agent. Despite the beneficial effects of LABs, the outcomes of some investigations have shown particular adverse clinical and technological influences of probiotics (e.g., metabolic disorders, including the generation of biogenic amines, the lack of clear clinical advice, and the lack of large and long-term clinical studies) [6]. In this regard, the use of metabolites LAB, which is known today as “postbiotic”, is a suitable alternative method. [7]. The term postbiotic has been points to the use of nonviable cells or cell fractions, which, when administered in adequate amounts, confer a health benefit to the host [8]. This study aims to highlight the definitions and characteristics of postbiotics and their application inactive food packaging.

2. Definition of Postbiotics

Postbiotic refers to compounds such as protein compounds, organic acids, peptides, vitamins, and short-chain fatty acids produced by probiotic bacteria in the gut when feeding on fibrous compounds [9,10]. In recent years, the terms biogenic, abiotic, metabolic, paraprobiotic, and postbiotic have been used to describe non-living cells and their metabolites [11,12]. The following biotics include the major part of inactivated microbial cells (cell body), cell fractions (Teichoic acid, cell-specific peptidoglycans, and morbid endogenous cells), Enzymes, bacteriocins, and organic acids. They are secreted when microbial cells release and decompose in the host's gut. If they are received in sufficient quantities, they have a beneficial effect on the host [13]. When a probiotic bacterium feeds on prebiotic compounds, it produces postbiotics which is in the state of a natural method [14,15]. However, today these compounds are also produced in a laboratory method. These manners include heat treatment (TT), ultraviolet radiation (UV), formalin inactivation (FI), ionizing radiation (IR), high pressure (HP), and sonication [13]. Postbiotics have beneficial characteristics such as certain chemical arrangements, safe resources, and higher shelf life, and have immunomodulatory, anti-inflammatory, cholesterol-lowering, antiproliferative, antioxidant, anti-hypertensive, and anti-obesity characteristics [16]. The use of postbiotics in the food industry is increasing due to their special biological properties. Here we examine the biological properties of postbiotics.

2.1. Safety aspects of postbiotics.

The multiple human microbiota system plays an important role in human health and disease status. It is known that the plentiful bacterial population (10^{14} CFU / g) in the intestine probably contributes to the immune system or metabolic roles and at the same time forms a defense system against various pathogens [17,18]. In addition, the high requirement for food and feed in reply to the increase in human population in developed countries makes the quality and safety of food and feed critical factors for the health and well-being of the community. However, probiotics are not considered completely safe for high-risk societies such as infants,
the elderly, or the immunocompromised, in whom probiotics can cause side effects, systemic infections and gastrointestinal manifestations, and the transmission of antibiotic resistance genes from probiotics to normal microbiota [19,20]. Currently, various recently published research suggests that the viability of probiotics is not a major factor in the health benefits of probiotic bacteria [21], which can be useful in avoiding the risks associated with probiotic consumption.

Safety issues and administrative conductance related to postbiotics and related functional foods should be anticipated due to potential risks and safety issues. As is known, most probiotic foods include *Lactobacillus* spp. and/or *Bifidobacterium* spp., which have seldom been recognized in the etiology of clinical infections in humans [22]. Consequently, probiotic products on the market today are considered safe foods despite the risks associated with certain groups of people. With all of the above hazards connected with bacteria in food in mind, there are still beneficial bacteria such as LAB and *Bifidobacteria* that could work as valuable agents against the appearance of perishable pathogens and microorganisms as they compete with pathogenic bacteria and with the generation of biologic compounds can improve food safety and shelf life [23]. Considering all the reports available on the positive and negative effects of various bacteria, the postbiotic was introduced as a novel theory of functional food constituents to overcome the adverse influences of probiotics. However, it may be necessary to consider the probiotic cells as a safe resource and fermentation in the production of postbiotics [24], and the safety features when consuming postbiotics. Since the postbiotic application is a relatively novel strategy, to the best of our knowledge, there is no clinical or epidemiological proof of hazards correlated with its use, but in theory, there is no risk of infection due to the nonexistence of live microorganisms. Several *in vitro* and in vivo investigations assessed the influence of postbiotics on various cells, blood parameters, metabolic biomarkers, and intestinal mucosa, as shown below, with bacteria and microbial products [16,25]. Such investigations elucidate the potential mechanism of action and can also form a template for additional animal and human investigations to distinguish the use of postbiotics. Due to the restrictions in the transferability of *in-vitro* studies and the possibility of species-specific effects [26], *in-vivo* studies are therefore urgently recommended to verify the *in-vitro* effects of postbiotics. The *in vitro* model using the cancer cell lines Caco2 and HT29 showed that the heat and postbiotics killed cells of a native strain of *Lactobacillus plantarum* A7 and a commercial strain of *L. rhamnosus* GG at concentrations of 2.5, 5, and 10 mg/ml reduced the growth rate of the examined cell lines [27]. The cytotoxicity of the cell extracts of *L. acidophilus* LA102 and *L. casei* LC232 was evaluated in an *in vitro* model of Caco2, and HRT18 cell lines, and the outcomes revealed that the rate of inhibition of proliferation was 37% (Caco2) and 68.5% (HRT18). For LA102 the inhibition rate was an additional 48% (Caco2) and 45.7% (HRT18) for LC232 at a concentration of 100 µg/ml. The results obtained approved the IC50 values as follows: 1.6 and 2.5 µg/ml LA102 and 15.4 and 6.2 µg/ml LC232 against Caco2 and HRT18 [28]. In a further *in vitro* study, the cytotoxicity of individual postbiotics generated by *L. plantarum* strains (I UL4, TL1, RS5, RI11, RG11, and RG14) in a concentration range of 0.47% to 30% (v/v) was assessed on the various cancer cell lines. The results demonstrated the ability to induce inhibitory effects such as antiproliferative induction and apoptosis in malignant cells that were dose and time-dependent [4]. With regard to the molecular signaling mechanisms, the antimetastatic and antiapoptotic effects of the HKSON fraction of *L. reuteri* were measured in a cancer stem cell model. The effects against metastases and apoptosis could be described by the downregulation...
of MMP9 and COX2 in accordance with the upregulation of TIMP1 so that it could be concluded that secretory macro-molecules are accountable for the recognized effects through modulation of various gene expressions [29]. A study by Cui et al. (2019) in a mouse model revealed that postbiotics (0.2 ml oral probe) from L. reuteri ZJ617 can ameliorate the acute damage induced by lipopolysaccharides in the liver. The ZJ617 supernatant used as a postbiotic was able to reduce inflammatory liver infiltration and serum biomarkers. The use of freshly weaned lambs as animal models showed that postbiotic supplementation (0.9% postbiotic L. Plantarum RG14) was able to increase weight gain, feed intake, nutrient uptake, and nutrient digestibility, which reflects the ability of postbiotic to improve rumen fermentation and microbial activity in the gut [30]. Izuddin et al. reported that postbiotic supplementation (0.9%) of L. plantarum RG14 in the diet of lambs after weaning led to a higher production of mucosal antibodies and antimicrobial peptides in the intestinal papillae, while the population of intestinal pathogens was lower than in the control group without postbiotics. The effects mentioned could be beneficial in reducing the cost of treatments and production losses because the animals' immune system is better prepared against several pathogens [31]. In a poultry type (broiler chicken), which was stimulated with the application of Cl. perfringens, Johnson et al. (2019) mentioned that the management of postbiotics (7.5 g/L of clean ingesting water) diminishes pro-inflammatory responses and production of homeostatic-like responses [32]. Another investigation with broiler chickens confirmed that the inulin and postbiotic (0.15% and 0.45% RG14) in the weight loss programs beneficially affected the animal's overall performance. Hence, the development in increase overall performance and populace of useful bacteria, in step with a discount withinside the populace of Enterobacteria and E. coli, become distinguished. Additionally, the extended acetic acid attention with related changes in ileal cytokine expression become additionally acquired in handled chickens [33]. Apart from various animal models for postbiotics, the peptides, peptidoglycan, cell-surface proteins, short-chain fatty acids, EPS, vitamins, lipopolysaccharides, and teichoic acids have been diagnosed because of the ability options materials for infectious illnesses withinside the discipline of aquaculture systems [33]. It is thought that the intestine epithelial barrier creates the primary protection barrier in opposition to the outside global related to unique pathogens, antigens, and dangerous substances. Consequently, any disorder withinside the barrier capability of the intestine epithelium (described as the leaky intestine) may want to open the gate and disclose the inner of the frame closer to more than one bad material and organism. Therefore, any materials with the potential to reinforce the intestinal barrier can be categorized as health-selling materials, and therefore, postbiotics can be taken into consideration as feasible candidates [34]. The supply of the useful outcomes of fermented ingredients at the gastrointestinal tract now no longer simplest comes from stay microorganisms due to the fact the goods from fermentation also can exert health-selling outcomes [35]. The underlined mechanisms regarding the outcomes of postbiotics on intestine barrier integrity have been addressed in various studies. The purification of two proteins known as p40 and p75 (based on the molecular weight) of postbiotics from L. rhamnosus GG resulted in explaining the possible mechanism of action associated with postbiotics. These proteins may serve as important players in different aspects like preventing TNF-induced damages, inhibition of apoptosis, and stimulation of intestinal epithelial cells proliferation [36]. Different pieces of evidence confirmed the importance of p40 and p75 as fundamental proteins in the function of probiotics and their metabolites [37,38]. Several studies had been designed for toddlers and adolescent volunteers due to the fact the hobby in probiotics and prebiotics began out to boom, stimulating
the investments of the latest dietary techniques aiming at modulating the intestine microbiota and using postbiotics on dietary perspectives. Clinical reviews primarily based totally on the fermented infant's formulation with merchandise from *B. breve* C50 and *S. thermophilus* in a mixture with short-chain galactooligosaccharides and long-chain fructooligosaccharides showed the protection of the formulation in wholesome period newborns [39]. The nonviable *lactobacilli* merchandise is taken into consideration as a large part of postbiotic proposals in adults. Some research proved the protection of the usage of *lactobacilli* merchandise in adults, and the bulk of those research describes the useful results of postbiotics on gastrointestinal functioning and remedy of diarrhea [40]. The inactivated lifestyle medium from *Lactobacillus* and *Lactococcus* (drugs consistent with day) in adults (suggest age 53.4 ± 17.three years) with diarrhea confirmed higher overall performance indicated with the aid of using decrease ache scores, bloating, and the first-class of existence stepped forward after remedy. However, this takes a look at lacked the manipulate organization, and the authors in comparison it with the baseline statistics after 1 month of remedy [41]. Another take a look at centered on wholesome adults (20 to 70 years old) with a bent in the direction of constipation or extra actions of the bowel to check the results of postbiotics from *Lactobacillus gasseri* CP2305 as opposed to a placebo. The centered markers had been Bristol stool scale scores, output, and color tone. All the critiques confirmed nice consequences in postbiotic remedies and the organization with extra constipation confirmed higher consequences in comparison to the diarrhea organization. The evaluation of fecal samples indicated growth in propionic acid, butyric acid, and valeric acid, and Clostridium cluster IV in postbiotic-treated groups [40]. In conclusion, postbiotics may contribute to the host's wellbeing and convey the beneficial effects via different mechanisms, even though the underlined mechanism of action has not been fully understood. The aforementioned studies confirmed and suggested the safety of using postbiotics in appropriate doses and concentrations.

2.2. Antimicrobial effects of postbiotics.

Besides, the significant antimicrobial effect of postbiotics is another vital factor in its use in food packaging. The postbiotics used in the food packaging composition prevent the spoilage of food inside the package by microorganisms by preventing it from entering the food matrix [42]. Since most food spoilage occurs by bacteria, studies have focused on bacteria. Studies have shown that each postbiotic compound has a specific mechanism in inhibiting spoilage bacteria [42-44]. The primary antibacterial mechanism of postbiotics consists of acidifying the mobile cytoplasm and stopping power law and production, defeating the growth of pathogenic microorganisms through the formation of holes in cellular membranes, morphological and purposeful adjustments of touchy additives consisting of proteins and peptides through growing acidity within the bacterial cell membrane in addition to inducing the oxidation of bacterial cells. The production and use of antimicrobial and bioactive food films for food packaging is a growing research field [30,42]. The postbiotics cocktail includes a huge type of biologically active metabolites creating synergistic antimicrobial actions within the films and food products. Moreover, the purification procedure of postbiotics metabolites (e.g., bacteriocins) includes more expensive methods, which make the active film greater pricey for the food producers and consumers [45,46]. Therefore, postbiotics can serve as the ideal antimicrobial compound, and there may be a developing hobby within the fabrication of postbiotics-included antimicrobial active films at some point in current years. Among the postbiotic compounds of bacteriocins and exopolysaccharides, it has been used
most in the production of packaging films. Studies in recent years show a good relationship between postbiotics and food packaging. For example, Sharaf et al. (2019) prepared chitosan nanoparticles containing postbiotics produced from seven LAB (L. plantarum, L. helveticus, L. rhamnosus, L. reuteri, Streptococcus thermophiles, Enterococcus faecium, and L. lactis) and confirmed the antibacterial and antifungal activity of all active nanoparticles in Egyptian cheese [47]. In another study, Salvucci et al. (2019) produced postbiotics from E. Faecium and also their semi-purified bacteriocin extract as an antimicrobial agent in the formulation of triticale flour films.

When the postbiotics combination turned into integrated into the films, considerable growth in water solubility and opacity of films turned into found; nevertheless, the components did now no longer alternate the films' mechanical characteristics and water vapor permeability. Incorporating 1% (v/v) of postbiotics or bacteriocin-wealthy extract created suitable antibacterial interest in triticale flour films. At the identical concentration, no considerable distinction was found among the antibacterial interest of postbiotics and bacteriocins extract-loaded films [48]. Divsalar et al. (2018) organized cellulosic paper lined with chitosan-ZnO nanocomposite and used nisin (500 and 1,000 μg/mL) as an antimicrobial agent in its structure. The incorporation of nisin furnished a robust antimicrobial impact on Listeria monocytogenes. Moreover, during 1-month storage at 4 °C, no significant decrease was observed in the antimicrobial activity of films. They also evaluated the effect of the active films for the packaging of ultra-filtered white cheese and reported that films with 1,000 μg/mL of nisin were able to completely inactivate the initial counts of L. monocytogenes on the surface of the cheese after 14 days of storage at 4 °C [49-51].

2.3. Reduction and degradation of chemical contaminants.

Recently studies have shown that postbiotics have the potential to destroy chemical contaminants such as bisphenol A, pesticides, and mycotoxins. In this sense, several works are attentive to bisphenol A, which is a group of low-molecular-weight compounds responsible for possible adverse reactions after food use [52,53]. The bisphenol-a, accumulated in fermented foods, is mostly produced by microbial decarboxylation of amino acids[54]. For example, precursor amino acids such as tyrosine, lysine, ornithine, histidine, and tryptophan produce cadaverine, histamine, tyramine, putrescine, and tryptamine, respectively. Putrescine can be transformed into spermidine that can form spermine[55]. Bisphenol-a is usually found in foods such as fish, vegetables, meat, cheese, and wines. Some bisphenol-a, especially histamine, tyramine, putrescine, and cadaverine, are involved in variant physiological and toxicological problems in the human body. The main microbial groups associated with bisphenol-a making are some Gram-negatives (Enterobacter spp. and Pseudomonas spp.) and Gram-positive bacteria, for example, Staphylococcus spp and probiotics strains[43]. Some species of Streptococcus spp., Lactococcus spp., Pediococcus spp., and Lactobacillus spp. generally have decarboxylase enzymes and have been cited as accountable for bisphenol a, accumulation in fermented foods[56]. The bisphenol-a formation/accumulation in foods could be decreased by (a) limiting microbial growth utilizing irradiation, controlled atmosphere packaging, hydrostatic pressures, or food additives, and (b) using an amine-negative starter culture and/or amine oxidase [57]. On the other hand, many published works showed the bisphenol-a-degrading capability of probiotics strain [58], however as previously mentioned, the direct use of probiotics strain has some drawbacks. In another study, the researchers have been using postbiotics from probiotics to evaluate the bisphenol-a degradation on the food matrix and in
in vitro
capability of postbiotics may be connected to (a) prohibition of the growth of bisphenol A producing LAB, (b) direct degradation of bisphenol A by postbiotics, and (c) decrease of bisphenol A formation by changing environmental conditions (e.g., pH) [59,60]. To understand the first strategy, the antimicrobial performance of postbiotics should be investigated by in vitro tests (i.e., agar disk and well 3406 diffusions, broth microdilution). The third scenario may be related to diamine oxidase and monoamine from LAB, which can be present in postbiotics and permeation of the bisphenol A degradation [61]. In another study, the researchers purified the thermostable amine oxidase mixture from L. plantarum CAU 3823 with a moderated possibility to degrade 40% of investigated bisphenol A (histamine, tyramine, putrescine, and cadaverine) in Chinese rice wine[56]. Amine oxidase was also identification in L.casei, L.plantarum [62], and L.curvatus [63]. Xie, Wang, Deng, and Xu (2016) investigated the performance of postbiotic and heat-treated postbiotic from L. plantarum on cell growth and diamine manufacture by four amine-positive bacteria (Enterobacteria species). Both types of postbiotics remarkably reduced cell growth and diamine production in all species except for Enterobacter cloacae. When the initial pH value reduced from 6.5 to 4.5, significant decreases in four amine-positive bacteria were found (except for E. cloacae in pH 6.5). In another study, postbiotics comprising diamine oxidase were more impressive than the lactobacillus bacteria strain (L. sakei) to degrade HIS in both model systems (phosphate buffer) and tuna soup [64]. L. curvatus G-1 was proposed as a potential candidate for bisphenol A control in fermented meat because it exhibited low BAs production and high bisphenol A degradability (more than 40%) [65]. Therefore, the application of starter culture with amine oxidase capacity in food may be considered the main factor for the bacterial choice for the fermentation process. It is also recommended to monitor the amine oxidase amount of postbiotics before using them in degradation experiments. García-Ruiz et al. (2011) investigated the biodegradation capabilities of some wine-associated lactobacillus and postbiotics on some bisphenol A in culture media and wine. Most of the isolates (n = 80), chiefly belonging to the LABs were able to degrade at least two of the three studied bisphenol A (HIS, TY, and PU) simultaneously[65]. They also reported that postbiotics from a LAB with high bisphenol A -degrading ability also revealed activity at an optimal pH of 4.7. Moreover, to avoid bisphenol A manufacture, postbiotics must be obtained from non-decarboxylase action of LAB [59], since decarboxylase enzymes may exhibit activity even later cell lysis and then, they may resist the commonly used food treatments (e.g., freezing, smoking, and heating) [56]. Toy et al. (2015) examined the BAs discount functionality of postbiotic (50% and 25% concentrations) of four LAB strains in some foodborne pathogens in tyrosine decarboxylase broth. Postbiotics of Streptococcus thermophiles at each concentration suppresses the BAs manufacturing through Salmonella paratyphi A, while the best 50% of postbiotics acquired from P. acidophilus avoided the TY formation. Interestingly, the simultaneous utility of postbiotics from Streptococcus thermophilus and L. lactis subsp. lactis at 50% concentrations decreased TY manufacturing through Staphylococcus aureus. It appears that, in this case, the short discount in pH might also additionally inhibit TY generation through the pathogens. Similarly, the PU-discount pastime of four LAB strains (P. acidilactici, L. mesenteroides, S. thermophilus, and L. lactis subsp. lactis) changed into monitored in ornithine decarboxylase-enriched broth [59]. BAs reduction potential varied depending on the used strains, but ≥a 65% reduction in accumulated PU was reported in all investigate postbiotics. The presence of AAs and reduction in pH are the main mechanisms behind PU decline. Owing
to the protective function of some polyamines, the quantity of BAs increased in the presence of postbiotics [66]. In the food model, the combination of postbiotics from *Pediococcus acidilactici* with thyme extract exhibited varied activity on BAs formation in the fish fillet, depending on the type of BAs and storage time [67]. The outcomes of the works on this area confirmed that the capabilities of postbiotics vary drastically primarily based totally on the sort and concentrations of postbiotics, the form of BAs, and the BAs-generating pathogen [68]. Therefore, it is essential to accurately select the postbiotics from LAB strains in keeping with the preferred impact of generating BAs.

### 2.4. Antibiofilm effect of postbiotics.

Biofilms are a group of one or greater kinds of microorganisms that may develop at distinct levels. A biofilm is a complicated microbial network enclosed inside a polysaccharide or protein matrix [69]. Biofilms may result from microorganisms which include fungi and microorganisms. Both gram-positive and gram-negative bacteria revel in such ability [70]. Bacterial resistance within the biofilm section to antimicrobials is a main worldwide issue. The formation levels encompass a reversible and irreversible attachment to the outside and micro clone formation with the generation of exopolysaccharides [71]. In the food industry, irreversible biofilms and colony ingredients are very vital, and managing them is crucial for meals safety [72]. Biofilms shaped within the meals enterprise are greater immune to cleansing and disinfection processes. *L. monocytogenes*, *Yersinia enterocolitidis*, *Campylobacter jejuni*, *Staphylococcus aureus*, and *Bacillus cereus* are vital biofilm-forming microorganisms within the meals enterprise [73]. Many strategies were used to manipulate and damage the biofilms shaped through microorganisms. Using postbiotics to kill biofilms is a brand new approach. In addition to having antimicrobial properties, postbiotics additionally have the belongings of destroying biofilms shaped through the microorganism. In current years, the impact of postbiotics on the removal of bacterial biofilms has been studied, which has yielded positive results [74]. In one study, the antibiofilm impact of postbiotics derived from the probiotic microorganism *Lactobacillus* LA5, *L. casei* 431, and *L. salivarius* on a biofilm shaped through *L. monocytogenes* at the polystyrene surfaces turned into observed. It validated that postbiotics damage biofilm formation. The authors set up that the presence of bacteriocin- and natural acid-primarily based postbiotics are the primary reason for the biofilm discount of *L. monocytogenes*. Therefore, postbiotics may be used as a device to manipulate and do away with biofilm formation through microorganisms within the meals enterprise [75].

### 3. Postbiotic Food Packaging

In the past, postbiotics were added directly to food, and the direct addition of postbiotics to food to increase shelf life was very common [76]. Despite the excessive antimicrobial ability of postbiotics, their direct use of the food matrix to increase its shelf life has a few disadvantages. The interplay of the complexation of postbiotics with the food additives can lessen their efficiency [77]. Additionally, the lower consistency of a few antimicrobial metabolites reasons their degradation and inactivation at some stage in meals processing. Lower miscibility of a few antimicrobial compounds with meals matrix is every other shortcoming that limits postbiotics applicability within the meals formulation. Economically, the embedding of a huge amount of the preservative compound within the indoor components of the meals matrix isn't suitable, due to the fact the microbial deterioration
mainly the mold growth, initiates from the outside of meals [78,79]. A promising technique proposed to overcome those obstacles is using food packaging to prolong the shelf life of the foods. Active packaging is a system wherein the meal's product, the packaging material, and the environment engage positively to increase the shelf life of meals [80]. Postbiotics additionally have characteristics that make their use as a brand new compound in meal packaging favorable, together with being (a) maximum of them are usually identified as secure substances (GRAS); (b) they're now no longer poisonous on eukaryotic cells; (c) they may be without difficulty inactivated through digestive proteases and thus, do now no longer affect the intestine microbiota; (d) they're living in a huge variety of pH; (e) they have got sturdy antimicrobial results in opposition to many foodborne pathogens and spoilage microorganisms even at low concentrations; and (f) most of the antimicrobial postbiotics have excessive thermal stability and their incorporation to the polymer matrix is possible even via extrusion and different heat-based processing methods [9,81]. Postbiotics contain various compounds such as organic acids, proteins, peptides, enzymes, and exopolysaccharides (EPS). Since most food spoilage occurs by microorganisms, especially bacteria, and due to the antibacterial properties of postbiotics, any of these compounds can be used in food packaging. Each of the postbiotic compounds inhibits food spoilage bacteria by a specific mechanism. The efficacy of postbiotics in food packaging is dependent on (a) the postbiotic type used in the packaging composition, (b) the type of target microorganism or contaminant, (c) concentration and form of application and (d) the food matrix characteristics. Among the above indicators, the type of postbiotics is very important.

3.1. Postbiotic food packaging based on organic acids.

Organic acids-based compounds are considered suitable antimicrobial factors [82]. Organic acids are referred to as one of the key postbiotics. Lactic acid (produced via way of means of bacterial fermentation processes) is to be had in isomers; L and D, the previous of that are powerful in inhibiting pathogenicity [83]. Also, citric acid and acetic acid hinder the growth of pathogens via way of means of growing an acidic environment. Among natural acids, lactic acids (pka= 3.86) and acetic acids (pka=4.76) hinder the growth of pathogens via way of means of lowering pH cost under in vitro or/and in vivo conditions [84]. The inhibitory impact of natural acids is associated with their impact on bacterial cell membranes. The foremost mechanisms right here include reducing the intracellular pH and membrane integrity [85]. The antimicrobial activity of natural acids may be related in ways. Acidity of cellular cytoplasm and prevention or/and the regulation of energy generation [84].

3.2. Postbiotic food packaging based on the peptide.

Antimicrobial peptides are produced through microorganisms. Peptides break microbes via pleiotropic (more than one action) mechanisms, along with microbial membrane degradation and the hindrance of macromolecule production [86]. Antimicrobial peptides are divided into ribosomal and non-ribosomal types. Peptides produced through the microorganism are ribosomal [87] and display robust antimicrobial interest in vitro through disrupting microbial membranes [88]. Peptides are typically found in all microorganisms. As mentioned, the primary goal of a few peptides is the cell membrane, while for others, it is the cytoplasm and delicate arrangements of microorganisms [89]. Antimicrobial mechanisms of the peptides include (a) developing acidity within the bacterial cell membrane, (b) developing physical
holes that leak cellular content, (c) activating deadly strategies along with inducing hydrolases which have unfavorable results on the cell wall, (d) and damaging sensitive intracellular elements of the microbes [90].

3.3. Postbiotic food packaging based on bacteriocins.

Bacteriocins are peptides or proteins with antimicrobial activity produced by various bacteria, such as Archaeabacteria and Eubacteria [85]. Bacteriocins have an excessive antimicrobial interest used for many years by people in fermented foods [91]. Bacteriocins are divided into steps with size, mechanism of action, and inhibitory spectrum. Bacteriocins have many useful effects, which include hindering the increase and improvement of gastrointestinal pathogens and being heat- and pH-resistant. According to the outcomes of studies, the principal interest of bacteriocins is within the bacterial cytoplasmic membrane [92]. The antimicrobial mechanism of bacteriocins is directly related to their effects on the structure and function of bacterial peptides and their inhibitory activities on spores and pore formation on pathogenic cell membranes.

4. Forms of Using Postbiotics in Active Food Packaging

There are numerous sorts of the use of postbiotics in food active packaging systems such as (a) coating or adsorbing of a slimy layer of postbiotics onto the polymer surface; (b) immobilization of individual postbiotic (e.g., bacteriocins and enzymes) on polymers with the aid of using ion or covalent linkages; (c) direct incorporation of postbiotics in a packaging polymer matrix; and (d) lamination of the postbiotic-loaded active film among outside layers, which improve the postbiotics consistency and manage their migration [93].

4.1. Application of individual postbiotic.

Frist form of using postbiotics in food active packaging is the application of individual postbiotic produced by different LABs. Bacteriocins, as bioactive peptides with antimicrobial action generated through LAB are the maximum regularly used postbiotics metabolite within the meals industry. Usually, the low yield and excessive production charges restrict the application of natural bacteriocins at manufacturing levels [81]. Due to those reasons, researchers have proven high-quality hobbies in generating and comparing bacteriocin-loaded active packaging systems over the past decade. Among hundreds of present bacteriocins, nisin is the most famous and appreciably used bacteriocin within the fabrication of active antimicrobial films produced through a few unique strains of L. lactis. Divsalar et al. (2018) organized a cellulosic paper covered with chitosan-ZnO nanocomposite and used nisin (500 and 1,000 μg/mL) as an AA in its structure. The incorporation of nisin supplied a sturdy antimicrobial impact on L. Monocytogenes. Moreover, in the course of the 1-month storage at 4 °C, no considerable lower change into located within the antimicrobial pastime of films. They additionally evaluated the impact of the active films for the packaging of ultra-filtered white cheese and mentioned that films with 1,000 μg/mL of nisin have been capable of absolutely inactivating the preliminary counts of L. monocytogenes at the surface of the cheese after 14 days of storage at 4 °C. Soto, Hernández-Iturriaga, Loarca-Piña, Luna-Bárcenas, and Mendoza (2019) organized nisin-loaded amaranth protein isolate/pullulan nanofibers films and located whole bactericidal activity in opposition to S. Typhimurium, L. monocytogenes, and L. mesenteroides in apple juice and fresh cheese [21]. Pediocin produced through Pediococcus
spp is every other broadly studied bacteriocin, that's used withinside the instruction of antimicrobial films. The antimicrobial activity of nisin and pediocin-loaded starch–halloysite nanocomposite films in opposition to *L. monocytogenes* and *Cl. Perfringens* have been investigated [94]. According to their results, the antilisterial activity of pediocin changed better than the nisin, however, nisin exhibited a more antagonistic function on *Cl. perfringens*. They additionally stated that incorporating halloysite helped manipulate the bacteriocins diffusion and better the antimicrobials retention withinside the polymer matrix. There are a few reviews on the usage of lactations withinside the system of active films. Enterocins of *Enterococcus* spp. are every other institution of purified individual postbiotic used withinside the fabrication of antimicrobial films. Gelatin films have been fabricated containing a combination of enterocins A, B, and P synthesized through *Enterococcus faecium* SM21 and prunin laurate as herbal antimicrobials[85]. According to their results, the enterocins-loaded film changed into powerful in opposition to *L. monocytogenes*, *S. aureus*, and *B. cereus*, and in all cases, a synergistic inhibitory action changed into located, while prunin laurate and enterocins have been concurrently introduced to the film. They located that the mechanical, thermal, and barrier characteristics of gelatin films have been no longer substantially laid low with active compounds incorporation.

4.2. Application of postbiotics mixture.

The postbiotics solution carries a huge type of biologically energetic metabolites stimulating synergistic antimicrobial activities withinside the films and food products [95]. Moreover, the purification procedure of postbiotics metabolites (e.g., bacteriocins and EPS) entails more expensive methods, making the active film greater highly priced for the food producers and consumers. Accordingly, postbiotics can act as the ideal AA, and there's a developing hobby withinside the fabrication of postbiotics-integrated antimicrobial active films throughout current years. As the primary file on postbiotics-integrated meals packaging films, Beristain-Bauza, Mani-López, Palou, and López-Malo (2016) delivered postbiotics from *L. rhamnosus* NRRL B-442 (6, 12, or 18 mg/mL) in whey protein isolate and calcium caseinate films. According to their outcomes, the postbiotics at a concentration of 18 mg/mL had major antimicrobial hobby towards *E. coli*, *L. monocytogenes*, *S. aureus*, and *S. Typhimurium* without any unfavorable consequences at the bodily residences of each film. The water vapor permeability and puncture power of films decreased, and brown-colored films were acquired after postbiotics incorporation. In the second one, studies stated via way of means of the equal group, whey protein isolate–alginate films supplemented with postbiotics from *L. sakei* NRRL B-1917 become fabricated for packaging of beef cubes inoculated with *E. coli* or *L. monocytogenes* [96]. During the refrigerated storage, 1.4- and 2.3-log10 CFU/g discounts of *L. monocytogenes* and *E. coli* have been found after a hundred and twenty and 36 hr, respectively. Sensorial reviews via way of means of panelists verified that there had been no meaningful variations among wrapped and unwrapped grilled red meat samples. In some other studies, approximately the utility of postbiotic active films on meat products, Rivas, Cayré, Campos, and Castro (2018) produced natural and synthetic casings (ovine, porcine, bovine, collagen, and cellulose casings) as vectors of postbiotics from *L. curvatus* ACU-1 and used for wrapping sausage meat paste. All postbiotics-loaded casings had appropriate antimicrobial activity towards *L. monocytogenes*. The authors proposed that each casing has been a powerful carrier of postbiotics and the active casings maintained their activity until the end of the trial. Recently, an antimicrobial meat wrapping nanopaper primarily based totally on bacterial
nanocellulose integrated with postbiotics from *L. plantarum* has developed [42]. The nanocellulose films have been dipped into postbiotic answers with exceptional concentrations (5% to 35%) and at exceptional impregnation times (5 to 60 min). The immobilization of postbiotics at the nanocellulose matrix produced a coherent community with reduced porosity. A meaningful reduction in tensile power and a growth in elongation capabilities of nanocellulose become found via way of means of the incorporation of postbiotics. The antimicrobial activity of nano papers towards *L. monocytogenes* elevated via way of means of growing postbiotics awareness and dipping time as much as 21.21% and 28 min, respectively. The optimized nanopaper and managed nanocellulose become used to percent ground meat. The counts of *L. monocytogenes* within the floor meat extensively decreased (5 log cycles) after nine days of storage at 4 °C. Moreover, the ground meat packed via way of means of postbiotics-activated nanocellulose reduced the whole mesophilic and psychrophilic bacterial counts and thiobarbituric acid values of meat samples. Postbiotics integrated active films have additionally been evaluated for the packaging of dairy products. Marques *et al.* (2017) produced a starch-primarily based antimicrobial active film containing postbiotics from *L. curvatus* P99 at concentrations (MIC: 15.6 μL/mL and minimal bactericidal awareness (MBC): 62.5 μL/mL). In agar antagonistic assay, the postbiotics-loaded casing found out robust bactericidal activity towards *L. monocytogenes*. After that, they packaged sliced "Prato" cheese with action films and studied the kinetics of antimicrobial movement of films throughout 10 days of storage. The outcomes indicated that each film has been powerful to lessen the *L. monocytogenes* Scott A counts in packaged "Prato" cheese, and the film containing the minimal bactericidal concentration (MBC) concentration of postbiotics remained underneath the restrict of detection (2.7 logs CFU/g) after 10 days of storage at 4 °C, without displaying distinguishable consequences on sensory characteristics [97]. Sharaf *et al.* (2019) organized chitosan nanoparticles containing postbiotics constituted of seven LAB (*L. plantarum*, *L. helveticus*, *L. rhamnosus*, *L. reuteri*, *S. thermophiles*, *E. faecium*, and *L. lactis*) and showed the antibacterial and antifungal activity of all active nanoparticles in Egyptian cheese. The postbiotics from *L. lactis* ATCC 11454, a nisin manufacturer strain, have been used as an AA to prepare sodium alginate/collagen or sodium carboxymethylcellulose/collagen-primarily based energetic films [98]. According to their outcomes, the incorporation of postbiotics adversely affected the physical characteristics of the films. The films' tensile power, extensibility, and transparency were reduced after postbiotics incorporation, and brown-colored films with better water solubility have been acquired. However, the films exhibited sturdy antimicrobial activity on *L. monocytogenes*, *S. aureus*, and *E. coli*. Heat treatment of postbiotics (30 min at 90 °C), earlier than including it in the film, precipitated a discount of 14% to 26% within the overall antimicrobial performance of the films. They attributed such discount to the presence of heat-touchy bacteriocin-like inhibitory materials within the postbiotics. Salvucci *et al.* (2019) produced postbiotics from *E. faecium* and additionally their semi-purified bacteriocin extract as an AA within the components of triticale flour films. When the postbiotics combination becomes integrated into the films, meaningful growth in water solubility and opacity of films becomes found; however, the components no longer alternate the films' mechanical residences and water vapor permeability. Incorporating 1% (v/v) of postbiotics or bacteriocin-rich extract created accurate antibacterial activity in triticale flour films. At the equal concentration, no meaningful distinction between postbiotics' antibacterial activity and bacteriocins extract-loaded films is found. Generally, the literature evaluation shows that despite the fact that sturdy antimicrobial active films may be organized
via way of means of incorporation of postbiotics to the packaging materials, the physicochemical residences of films can be adversely laid low with this incorporation and require an optimized postbiotics concentration to attain the right stability among antibacterial activity and thermomechanical residences of films [48]. To face this challenge, Bagde and Nadanathangam, (2019) used cellulose nanocrystals for the immobilization of postbiotics from \( P.\ acidilactici \) and \( E.\ faecium \) and integrated the activated cellulose nanocrystals into corn starch films. They advised higher stability of bacteriocins throughout the storage after immobilization on cellulose nanocrystals. In addition, the unfavorable impact of postbiotics at the mechanical and barrier residences of starch films become decreased after immobilization on cellulose nanofiller. They proposed that accurate antimicrobial packaging films with better thermal, mechanical, and barrier residences will be produced by immobilizing AAs on appropriate nano reinforcements earlier than including them into the film matrix [99].

5. Microencapsulation of Postbiotics

Protection of postbiotics towards damaging environments, which includes antimicrobial agents, chemicals, active oxygen in case of compulsory anaerobic microbes, bile salts, and excessive acidity, might be finished via using microencapsulation methods. Also, using strategies that include fluidized mattress drying, spray cooling, extrusion, chilling, molecular inclusion, spray drying, co-crystallization, and co-accretion might be possible to expand the processing of a forming capsule [54,100]. Choosing the method of the hobby depends on the material type, application, and release mechanism. Compounds that include carbohydrates, proteins, and lipids may be used to microencapsulate postbiotics [101]. Materials used for the microencapsulation of postbiotics should be non-toxic, highly soluble, heat-resistant, oxygen-permeable via the meals matrix, acid-resistant, and volatile at pH above 6 [102,103]. In the manner of encapsulating postbiotics, a biocompatible matrix should be used to encapsulate postbiotics towards elements that include pH and excessive temperature. The biocompatible matrix acts as a semi-permeable membrane and lets in the switch of postbiotics in directions. Studies in the latest years at the encapsulation of postbiotics have proven that encapsulation is an appropriate approach to guard those compounds towards irrelevant elements. In this regard, Le et al. (2019) encapsulated postbiotic (bacteriocin) produced via way of means of \( Lactobacillus\ plantarum \) isolated from Vietnamese fermented yogurt in alginate-gelatin (ALG-GEL). Also, its antimicrobial consequences within the presence of things which includes incubation temperature, mild pH, and surfactants (Ethylene diamine tetraacetic acid (EDTA), sodium dodecyl sulfate (SDS), and twin) towards five indicator organisms, which include \( E.\ coli,\ Salmonella,\ S.\ aureus,\ L.\ monocytogenes,\ \)and \( Bacillus\ subtilis \) had been evaluated in meat. They determined that encapsulating postbiotics within the presence of those elements should save you the spoilage of red meat via way of means of pathogens. It appears that the microencapsulation of postbiotics may be a desirable manner to guard postbiotics. Using microencapsulation technology, postbiotics might be utilized in meals uncovered to excessive temperatures and low pH (e.g., vegetables) [104].

6. Conclusions

The use of postbiotics in the food industry is a common way to prevent food spoilage. Postbiotics are metabolites produced by probiotic bacteria that have many health effects. Non-toxicity and safety of postbiotics and their ability to inhibit microorganisms that cause food
spoilage are the most important features of postbiotics in using these compounds in the food industry. In studies on postbiotics in the food industry, the use of these compounds as a way to control microbial spoilage of substances may interfere with the function (factors in the food matrix) in the function of postbiotics. Therefore, the use of postbiotics in the form of food packaging can be more effective. Therefore, due to their unique properties, postbiotics have received a lot of attention in the food industry and can be used as a new approach in food packaging. Future head-to-head trials are necessary to distinguish appropriate strains of parent cells, active compounds, biological function, optimal dosages and to examine their activities in developing active food packaging.

**Funding**

This research received no external funding.

**Acknowledgments**

The authors would like to express their thanks to the Research vice-chancellor of Ahvaz Jundishapur University of Medical Sciences for this study's financial support.

**Conflicts of Interest**

The authors declare that they have no conflicts of interest.

**References**

1. Alizadeh-Sani, M.; Ehsani, A.; Kia, E.M.; Khezerlou, A. Microbial gums: introducing a novel functional component of edible coatings and packaging. *Applied microbiology and biotechnology* **2019**, *103*, 6853-6866, [https://doi.org/10.1007/s00253-019-09966-x](https://doi.org/10.1007/s00253-019-09966-x).

2. Majeed, M.; Majeed, S.; Nagabhushanam, K.; Mundkur, L.; Rajalakshmi, H.; Shah, K.; Beede, K. Novel Topical Application of a Postbiotic, LactoSporin®, in Mild to Moderate Acne: A Randomized, Comparative Clinical Study to Evaluate its Efficacy, Tolerability, and Safety. *Cosmetics* **2020**, *7*, 70, [https://doi.org/10.3390/cosmetics7030070](https://doi.org/10.3390/cosmetics7030070).

3. Pop, O.L.; Pop, C.R.; Dufrechou, M.; Vodnar, D.C.; Socaci, S.A.; Dulf, F.V.; Minervini, F.; Suharoschi, R. Edible Films and Coatings Functionalization by Probiotic Incorporation: A Review. *Polymers* **2020**, *12*, 12, [https://doi.org/10.3390/polym12010012](https://doi.org/10.3390/polym12010012).

4. Wong, K.E.; Ngai, S.C.; Chan, K.-G.; Lee, L.-H.; Goh, B.-H.; Chuah, L.-H. Curcumin nanoformulations for colorectal cancer: a review. *Frontiers in pharmacology* **2019**, *10*, 152, [https://doi.org/10.3389/fphar.2019.00152](https://doi.org/10.3389/fphar.2019.00152).

5. Akbani, M.R.; Haghighi, H.R.; Chambers, J.R.; Brisbin, J.; Read, L.R.; Sharif, S. Expression of antimicrobial peptides in cecal tonsils of chickens treated with probiotics and infected with Salmonella enterica serovar typhimurium. *Clinical and Vaccine Immunology* **2008**, *15*, 1689-1693, [https://doi.org/10.1128/CVI.00242-08](https://doi.org/10.1128/CVI.00242-08).

6. Tomar, S.K.; Anand, S.; Sharma, P.; Sangwan, V.; Mandal, S. Role of probiotics, prebiotics, synbiotics and postbiotics in inhibition of pathogens. *The Battle against Microbial Pathogens: Basic Science, Technological Advances and Educational Programs; Méndez-Vilas, A., Ed* **2015**, 717-732.

7. Callon, C.; Arliguie, C.; Montel, M.-C. Control of Shigatoxin-producing Escherichia coli in cheese by dairy bacterial strains. *Food microbiology* **2016**, *53*, 63-70, [https://doi.org/10.1016/j.fm.2015.08.009](https://doi.org/10.1016/j.fm.2015.08.009).

8. Homayouni Rad, A.; Aghebati Maleki, L.; Samadi Kafil, H.; Abbasi, A. Postbiotics: A novel strategy in food allergy treatment. *Critical reviews in food science and nutrition* **2021**, *61*, 492-499, [https://doi.org/10.1080/10408398.2020.1738333](https://doi.org/10.1080/10408398.2020.1738333).
9. Rad, A.H.; Aghebati-Maleki, L.; Kafil, H.S.; Abbasi, A. Molecular mechanisms of postbiotics in colorectal cancer prevention and treatment. *Critical reviews in food science and nutrition* 2020, 1-17, https://doi.org/10.1080/10408398.2020.1765310.
10. Abbasi, A.; Sheykhsaran, E.; Kafil, H.S. *Postbiotics: Science, Technology and Applications*; Bentham Science Publishers 2021.
11. Homayouni Rad, A.; Aghebati Maleki, L.; Samadi Kafil, H.; Abbasi, A. Postbiotics: A novel strategy in food allergy treatment. *Critical reviews in food science and nutrition* 2020, 1-8, https://doi.org/10.1080/10408398.2020.1738333.
12. Abbasi, A.; Aghebati-Maleki, L.; Homayouni-Rad, A. The promising biological role of postbiotics derived from probiotic Lactobacillus species in reproductive health. *Critical Reviews in Food Science and Nutrition* 2021, 1-13, https://doi.org/10.1080/10408398.2021.1935701.
13. Rad, A.H.; Abbasi, A.; Kafil, H.S.; Ganbarov, K. Potential pharmaceutical and food applications of postbiotics: a review. *Current pharmaceutical biotechnology* 2020, https://doi.org/10.2174/1389201021666200516154833.
14. Rad, A.H.; Maleki, L.A.; Kafil, H.S.; Zavoshti, H.F.; Abbasi, A. Postbiotics as novel health-promoting ingredients in functional foods. *Health promotion perspectives* 2020, 10, 3-4, https://doi.org/10.15171/hpp.2020.02.
15. Aghebati-Maleki, L.; Hasannezhad, P.; Abbasi, A.; Khani, N. Antibacterial, Antiviral, Antioxidant, and Anticancer Activities of Postbiotics: A review of Mechanisms and Therapeutic Perspectives 2021, https://doi.org/10.33263/BRIAC122.26292645.
16. Fathi-zavoshti, H.; Douroud, N.; Shabazi, N.; Abbasi, A. Evaluating the role of postbiotics as a new generation of probiotics in health and diseases. *Journal of Ardabil University of Medical Sciences* 2020, 19, 381-399. https://jarums.arums.ac.ir/browse.php?a_code=A-10-14621&slid=1&slc_lang=en.
17. Chong, C.Y.L.; Bloomfield, F.H.; O’Sullivan, J.M. Factors affecting gastrointestinal microbiome development in neonates. *Nutrients* 2018, 10, 274, https://doi.org/10.3390/nu10030274.
18. Mosca, F.; Gianni, M.L.; Rescigno, M. Can postbiotics represent a new strategy for NEC? *Probiotics and Child Gastrointestinal Health* 2019, 37-45, https://doi.org/10.1007/5584_2018_314.
19. Sotoudegan, F.; Daniali, M.; Hassan, S.; Nikfar, S.; Abdollahi, M. Reappraisal of probiotics’ safety in human. *Food and Chemical Toxicology* 2019, 129, 22-29, https://doi.org/10.1016/j.fct.2019.04.032.
20. Abbasi, A.; Aghebati-Maleki, A.; Aghebati-Maleki, L.; Yousefi, M. Probiotic intervention as a potential therapeutic for managing gestational disorders and improving pregnancy outcomes. *Journal of Reproductive Immunology* 2020, 103244, https://doi.org/10.1016/j.jri.2020.103244.
21. Aguilar-Toalá, J.; García-Varela, R.; García, H.; Mata-Haro, V.; González-Córdova, A.; Vallejo-Cordoba, B.; Hernández-Mendoza, A. Postbiotics: An evolving term within the functional foods field. *Trends in Food Science & Technology* 2018, 75, 105-114.
22. Plumed-Ferrer, C.; Kivelä, I.; Hyyönen, P.; Von Wright, A. Survival, growth and persistence under farm conditions of a Lactobacillus plantarum strain inoculated into liquid pig feed. *Journal of applied microbiology* 2005, 99, 851-858, https://doi.org/10.1111/j.1365-2672.2005.02666.x.
23. Gaggia, F.; Mattarelli, P.; Biavati, B. Probiotics and prebiotics in animal feeding for safe food production. *International journal of food microbiology* 2010, 141, S15-S28, https://doi.org/10.1016/j.ijfoodmicro.2010.02.031.
24. Morniroli, D.; Vizzari, G.; Consales, A.; Mosca, F.; Gianni, M.L. Postbiotic Supplementation for Children and Newborn’s Health. *Nutrients* 2021, 13, 781, https://doi.org/10.3390/nu13030781.
25. Angiari, S.; Runtsch, M.C.; Sutton, C.E.; Palsson-McDermott, E.M.; Kelly, B.; Rana, N.; Kane, H.; Papadopoulou, G.; Pearce, E.L.; Mills, K.H. Pharmacological activation of pyruvate kinase M2 inhibits CD4+ T cell pathogenicity and suppresses autoimmunity. *Cell metabolism* 2020, 31, 391-405. e398, https://doi.org/10.1016/j.cmet.2019.10.015.
26. Karl, J.P.; Hatch, A.M.; Arcidiacono, S.M.; Pearce, S.C.; Pantoja-Feliciano, I.G.; Doherty, L.A.; Soares, J.W. Effects of psychological, environmental and physical stressors on the gut microbiota. *Frontiers in microbiology* 2018, 9, 2013. https://doi.org/10.3389/fmicb.2018.02013.
27. Sadeghi-Aliaabadi, H.; Mohammadi, F.; Fazeli, H.; Mirlohi, M. Effects of Lactobacillus plantarum A7 with probiotic potential on colon cancer and normal cells proliferation in comparison with a commercial strain. *Iranian Journal of Basic Medical Sciences* 2014, 17, 815.
28. Awaisha, S.; Obeidat, M.; Al-Tamimi, H.; Assaf, A.; El-Qudah, J.; Rahahleh, R. In vitro cytotoxic activity of probiotic bacterial cell extracts against Caco-2 and HRT-18 colorectal cancer cells. *Milk Science International-Milchwissenschaft* **2016**, *69*, 33-37.

29. Maghsoud, F.; Johari, B.; Rohani, M.; Madanchi, H.; Saltanatpour, Z.; Kadivar, M. Anti-proliferative and antimetastatic potential of high molecular weight secretory molecules from probiotic Lactobacillus reuteri cell-free supernatant against human colon cancer stem-like cells (HT29-ShE). *International Journal of Peptide Research and Therapeutics* **2020**, *1*-13, https://doi.org/10.1007/s10989-020-10049-z.

30. Izuddin, W.I.; Loh, T.C.; Foo, H.L.; Samsudin, A.A.; Humam, A.M. Postbiotic L. plantarum RG14 improves ruminal epithelium growth, immune status and upregulates the intestinal barrier function in post-weaning lambs. *Scientific reports* **2019**, *9*, 1-10, https://doi.org/10.1038/s41598-019-46076-0.

31. Izuddin, W.I.; Humam, A.M.; Loh, T.C.; Foo, H.L.; Samsudin, A.A. Dietary postbiotic lactobacillus plantarum improves serum and ruminal antioxidant activity and upregulates hepatic antioxidant enzymes and ruminal barrier function in post-weaning lambs. *Antioxidants* **2020**, *9*, 250, https://doi.org/10.3390/antioxidants9030250.

32. Johnson, C.N.; Kogut, M.H.; Genovese, K.; He, H.; Kazemi, S.;Arsenault, R.J. Administration of a postbiotic causes immunomodulatory responses in broiler gut and reduces disease pathogenesis following challenge. *Microorganisms* **2019**, *7*, 268, https://doi.org/10.3390/microorganisms7080268.

33. Kareem, K.; Loh, T.; Foo, H.; Asmara, S.; Akit, H. Influence of postbiotic RG14 and inulin combination on cecal microbiota, organic acid concentration, and cytokine expression in broiler chickens. *Poultry science* **2017**, *96*, 966-975, https://doi.org/10.3382/ps/pew362.

34. Anderson, R.C. Are Postbiotics the Long Sought—After Solution for a Leaky Gut? *The Journal of nutrition* **2019**, *149*, 1873-1874, https://doi.org/10.1093/jn/nzx171.

35. Maguire, M.; Maguire, G. Gut dysbiosis, leaky gut, and intestinal epithelial proliferation in neurological disorders: towards the development of a new therapeutic using amino acids, prebiotics, probiotics, and postbiotics. *Reviews in the Neurosciences* **2019**, *30*, 179-201, https://doi.org/10.1515/reveneu-2018-0024.

36. Cicenia, A.; Scirocco, A.; Carabotti, M.; Pallotta, L.; Marignani, M.; Severi, C. Postbiotics activities of lactobacilli-derived factors. *Journal of clinical gastroenterology* **2014**, *48*, S18-S22, https://doi.org/10.1097/MCG.0000000000000231.

37. Bermudez-Brito, M.; Plaza-Díaz, J.; Muñoz-Quezada, S.; Gómez-Llorente, C.; Gil, A. Probiotic mechanisms of action. *Annals of Nutrition and Metabolism* **2012**, *61*, 160-174, https://doi.org/10.1159/000342079.

38. Karimi, N.; Jabbari, V.; Nazemi, A.; Ganbarov, K.; Karimi, N.; Tanomand, A.; Karimi, S.; Abbasi, A.; Yousefi, B.; Khodadadi, E. Thymol, cardamom and Lactobacillus plantarum nanoparticles as a functional candy with high protection against Streptococcus mutans and tooth decay. *Microbial pathogenesis* **2020**, *148*, 104481, https://doi.org/10.1016/j.micpath.2020.104481.

39. Wegh, C.A.; Geerlings, S.Y.; Knol, J.; Roeselers, G.; Belzer, C. Postbiotics and their potential applications in early life nutrition and beyond. *International journal of molecular sciences* **2019**, *20*, 4673, https://doi.org/10.3390/ijms20194673.

40. Sawada, D.; Sugawara, T.; Ishida, Y.; Aihara, K.; Aoki, Y.; Takehara, I.; Takano, K.; Fujiwara, S. Effect of continuous ingestion of a beverage prepared with Lactobacillus gasseri CP2305 inactivated by heat treatment on the regulation of intestinal function. *Food Research International* **2016**, *79*, 33-39, https://doi.org/10.1016/j.foodres.2015.11.032.

41. Tarrerias, A.; Costil, V.; Vicari, F.; Letard, J.; Adenis-Lamarre, P.; Aisene, A.; Batistelli, D.; Bonnaud, G.; Carpenter, S.; Dalbies, P. The effect of inactivated Lactobacillus LB fermented culture medium on symptom severity: observational investigation in 297 patients with diarrhea-predominant irritable bowel syndrome. *Digestive Diseases* **2011**, *29*, 588-591, https://doi.org/10.1159/000332987.

42. Yordshahi, A.S.; Moradi, M.; Tajik, H.; Molaei, R. Design and preparation of antimicrobial meat wrapping nanopaper with bacterial cellulose and postbiotics of lactic acid bacteria. *International journal of food microbiology* **2020**, *321*, 108561, https://doi.org/10.1016/j.ijfoodmicro.2020.108561.

43. Molaei, R.; Tajik, H.; Moradi, M. Magnetic solid phase extraction based on mesoporous silica-coated iron oxide nanoparticles for simultaneous determination of biogenic amines in an Iranian traditional dairy product; Kashk. *Food Control* **2019**, *101*, 1-8, https://doi.org/10.1016/j.foodcont.2019.02.011.
44. Rad, A.; Abbasi, A.; Javadi, A.; Pourjafar, H.; Javadi, M.; Khaleghi, M. Comparing the microbial quality of traditional and industrial yoghurts. Biointerface Research in Applied Chemistry 2020, 10, 6020-6025.

45. Cazón, P.; Vázquez, M. Bacterial celluose as a biodegradable food packaging material: a review. Food Hydrocolloids 2020, 106530, https://doi.org/10.1016/j.foodhyd.2020.106530.

46. Abbasi, A.; Hajipour, N.; Hasannezhad, P.; Baghbanzadeh, A.; Aghebati-Maleki, L. Potential in vivo delivery routes of postbiotics. Critical Reviews in Food Science and Nutrition 2020, 1-39, https://doi.org/10.1080/10408398.2020.1865260.

47. Sharaf, O.M.; Al-Gamal, M.S.; Ibrahim, G.A.; Dabiza, N.M.; Salem, S.S.; El-ssayad, M.F.; Youssef, A.M. Evaluation and characterization of some protective culture metabolites in free and nano-chitosan-loaded forms against common contaminants of Egyptian cheese. Carbohydrate polymers 2019, 223, 115094, https://doi.org/10.1016/j.carbpol.2019.115094.

48. Salvucci, E.; Rossi, M.; Colombo, A.; Pérez, G.; Borneo, R.; Aguirre, A. Triticale flour films added with bacteriocin-like substance (BLIS) for active food packaging applications. Food Packaging and Shelf Life 2019, 19, 193-199, https://doi.org/10.1016/j.fpsl.2018.05.007.

49. Divsalar, E.; Tajik, H.; Moradi, M.; Forough, M.; Lotfi, M.; Kuswandi, B. Characterization of cellulose/zinc oxide nanocomposite containing nisin and its application in packaging of UF cheese. International journal of biological macromolecules 2018, 109, 1311-1318, https://doi.org/10.1016/j.ijbiomac.2017.11.145.

50. Homayouni Rad, A.; Samadi Kafil, H.; Fathi Zavoshti, H.; Shahbazi, N.; Abbasi, A. Therapeutically effects of functional postbiotic foods. Clinical Excellence 2020, 10, 33-52.

51. Abbasi, A.; Rad, A.H.; Ghasempour, Z.; Sabahi, S.; Kafil, H.S.; Hasannezhad, P.; Rahbar Saadat, Y.; Shahbazi, N. The biological activities of postbiotics in gastrointestinal disorders. Critical Reviews in Food Science and Nutrition 2021, 1-22, https://doi.org/10.1080/10408398.2021.1895061.

52. Czajkowska-Myslek, A.; Leszczyńska, J. Risk assessment related to biogenic amines occurrence in ready-to-eat baby foods. Food and Chemical Toxicology 2017, 105, 82-92, https://doi.org/10.1016/j.fct.2017.03.061.

53. Leylabadol, H.E.; Heravi, F.S.; Soltani, E.; Abbasi, A.; Kafil, H.S.; Parsaei, M.; Sanaie, S.; Ahmadian, Z.; Ghotaslou, R. The role of gut microbiota in the treatment of irritable bowel syndrome. Reviews in Medical Microbiology 2021.

54. Mah, J.-H.; Park, Y.K.; Jin, Y.H.; Lee, J.-H.; Hwang, H.-J. Bacterial production and control of biogenic amines in Asian fermented soybean foods. Foods 2019, 8, 85, https://doi.org/10.3390/foods8020085.

55. Papageorgiou, M.; Lambropoulou, D.; Morrison, C.; Kłodzińska, E.; Namieśnik, J.; Plotka-Wasylko, J. Literature update of analytical methods for biogenic amines determination in food and beverages. TrAC Trends in Analytical Chemistry 2018, 98, 128-142, https://doi.org/10.1016/j.trac.2017.11.001.

56. Barbieri, F.; Montanari, C.; Gardini, F.; Tabanelli, G. Biogenic amine production by lactic acid bacteria: A review. Foods 2019, 8, 17, https://doi.org/10.3390/foods8010017.

57. Naima, A.; Flint, S.; Fletcher, G.; Bremer, P.; Meerdink, G. Control of biogenic amines in food—existing and emerging approaches. Journal of food science 2010, 75, R139-R150, https://doi.org/10.1111/j.1750-3841.2010.01774.x.

58. García-Ruiz, A.; González-Rompinelli, E.M.; Bartolomé, B.; Moreno-Arribas, M.V. Potential of wine-associated lactic acid bacteria to degrade biogenic amines. International journal of food microbiology 2011, 148, 115-120, https://doi.org/10.1016/j.ijfoodmicro.2011.05.009.

59. Ozogul, F.; Tabanelli, G.; Toy, N.; Gardini, F. Impact of cell-free supernatant of lactic acid bacteria on putrescine and other polyamine formation by foodborne pathogens in ornithine decarboxylase broth. Journal of agricultural and food chemistry 2015, 63, 5828-5835, https://doi.org/10.1021/acs.jafc.5b02410.

60. Homayouni-rad, A.; Oroojzadeh, P.; Abbasi, A. The Effect of Yeast Kluyveromyces marxianus as a Probiotic on the Microbiological and Sensorial Properties of Set Yoghurt during Refrigerated Storage. Journal of Ardabil University of Medical Sciences 2021, 20, 254-268.

61. García-Ruiz, J.M.; López- Moreno, J.I.; Vicente-Serrano, S.M.; Lasanta–Martínez, T.; Beguería, S. Mediterranean water resources in a global change scenario. Earth-Science Reviews 2011, 105, 121-139, https://dx.doi.org/10.1016/j.earscirev.2011.01.006.
62. Fadda, S.; Vignolo, G.; Oliver, G. Tyramine degradation and tyramine/histamine production by lactic acid bacteria and Kocuria strains. *Biotechnology Letters* **2001**, *23*, 2015-2019, [https://doi.org/10.1023/A:1013783030276](https://doi.org/10.1023/A:1013783030276).

63. Li, M.; van Esch, B.C.; Henricks, P.A.; Folkerts, G.; Garssen, J. The anti-inflammatory effects of short chain fatty acids on lipopolysaccharide-or tumor necrosis factor α-stimulated endothelial cells via activation of GPR41/43 and inhibition of HDACs. *Frontiers in pharmacology* **2018**, *9*, 533, [https://doi.org/10.3389/fphar.2018.00533](https://doi.org/10.3389/fphar.2018.00533).

64. Naila, A.; Flint, S.; Fletcher, G.; Bremer, P.; Meerdink, G. Histamine degradation by diamine oxidase, Lactobacillus and Vergibacillus halodonitrificans Nai18. *Journal of Food Processing and Technology* **2012**, *3*, [https://doi.org/10.4172/2157-7110.1000158](https://doi.org/10.4172/2157-7110.1000158).

65. Freiding, S.; Gutsche, K.A.; Ehrmann, M.A.; Vogel, R.F. Genetic screening of Lactobacillus sakei and Lactobacillus curvatus strains for their peptidolytic system and amino acid metabolism, and comparison of their volatilomes in a model system. *Systematic and Applied Microbiology* **2011**, *34*, 311-320, [https://doi.org/10.1016/j.syapm.2010.12.006](https://doi.org/10.1016/j.syapm.2010.12.006).

66. Özogul, F.; Toy, N.; Özogul, Y. The impact of the cell-free solution of lactic acid bacteria on cadaverine production by Listeria monocytogenes and Staphylococcus aureus in lysine-decarboxylase broth. *International Journal of Nutrition and Food Engineering* **2015**, *9*, 309-317.

67. Kuley, E.; Durmus, M.; Ucar, Y.; Kosker, A.R.; Tumerkan, E.T.A.; Regenstein, J.M.; Özogul, F. Combined effects of plant and cell-free extracts of lactic acid bacteria on biogenic amines and bacterial load of fermented sardine stored at 3±1° C. *Agriculture Food Bioscience* **2018**, *24*, 127-136.

68. Özogul, F.; Toy, N.; Özogul, Y.; Hamed, I. Function of cell-free supernatants of Leuconostoc, Lactococcus, Streptococcus, Pediococcus strains on histamine formation by foodborne pathogens in histidine decarboxylase broth. *Journal of Food Processing and Preservation* **2017**, *41*, e13208.

69. Urish, K.L.; DeMuth, P.W.; Kwan, B.W.; Craft, D.W.; Haider, H.; Tuan, R.S.; Wood, T.K.; Davis, C.M. Antibiotic-tolerant Staphylococcus aureus biofilm persists on arthroplasty materials. *Clinical Orthopaedics and Related Research* **2016**, *474*, 1649-1656, [https://doi.org/10.1007/s11999-016-4720-8](https://doi.org/10.1007/s11999-016-4720-8).

70. Miao, J.; Liang, Y.; Chen, L.; Wang, W.; Wang, J.; Li, B.; Li, L.; Chen, D.; Xu, Z. Formation and development of Staphylococcus biofilm: with focus on food safety. *Journal of Food Safety* **2017**, *37*, e12358, [https://doi.org/10.1111/jfs.12358](https://doi.org/10.1111/jfs.12358).

71. Przekwas, J.; Wiktorczyk, N.; Budzyńska, A.; Gospodarek-Komkowska, E. Ascorbic Acid Changes Growth of Food-Borne Pathogens in the Early Stage of Biofilm Formation. *Microorganisms* **2020**, *8*, 553, [https://doi.org/10.3390/microorganisms8040553](https://doi.org/10.3390/microorganisms8040553).

72. Andrade, J.C.; João, A.L.; Alonso, C.d.S.; Barreto, A.S.; Henriques, A.R. Genetic Subtyping, Biofilm-Forming Ability and Biocide Susceptibility of Listeria monocytogenes Strains Isolated from a Ready-to-Eat Food Industry. *Antibiotics* **2020**, *9*, 416, [https://doi.org/10.3390/antibiotics9070416](https://doi.org/10.3390/antibiotics9070416).

73. Shi, X.; Zhu, X. Biofilm formation and food safety in food industries. *Trends in Food Science & Technology* **2009**, *20*, 407-413, [https://doi.org/10.1016/j.tifs.2009.01.054](https://doi.org/10.1016/j.tifs.2009.01.054).

74. Sharma, V.; Harjai, K.; Shukla, G. Effect of bacteriocin and exopolysaccharides isolated from probiotic on *P. aeruginosa* PAO1 biofilm. *Folia microbiologica* **2018**, *63*, 181-190, [https://doi.org/10.1007/s12223-017-0545-4](https://doi.org/10.1007/s12223-017-0545-4).

75. Moradi, M.; Mardani, K.; Tajik, H. Characterization and application of postbiotics of Lactobacillus spp. on Listeria monocytogenes in vitro and in food models. *LWT* **2019**, *111*, 457-464, [https://doi.org/10.1016/j.lwt.2019.05.072](https://doi.org/10.1016/j.lwt.2019.05.072).

76. Gómez-Sala, B.; Herranz, C.; Díaz-Freitas, B.; Hernández, P.E.; Sala, A.; Cintas, L.M. Strategies to increase the hygienic and economic value of fresh fish: Biopreservation using lactic acid bacteria of marine origin. *International journal of food microbiology* **2016**, *223*, 41-49, [https://doi.org/10.1016/j.ijfoodmicro.2016.02.005](https://doi.org/10.1016/j.ijfoodmicro.2016.02.005).

77. Hamad, G.; Botros, W.; Hafez, E. Combination of probiotic filtrates as antibacterial agent against selected some pathogenic bacteria in milk and cheese. *Int J Dairy Sci* **2017**, *12*, 368-376, [https://doi.org/10.3923/ijds.2017.368.376](https://doi.org/10.3923/ijds.2017.368.376).

78. Dunand, E.; Burns, P.; Binetti, A.; Bergamini, C.; Peralta, G.H.; Forzani, L.; Reinheimer, J.; Vinderola, G. Postbiotics produced at laboratory and industrial level as potential functional food ingredients with the capacity to protect mice against Salmonella infection. *Journal of applied microbiology* **2019**, *127*, 219-229, [https://doi.org/10.1111/jam.14276](https://doi.org/10.1111/jam.14276).
97. de Lima Marques, J.; Funck, G.D.; da Silva Dannenberg, G.; dos Santos Cruxen, C.E.; El Halal, S.L.M.; Dias, A.R.G.; Fiorentini, Â.M.; da Silva, W.P. Bacteriocin-like substances of Lactobacillus sakei cell-free supernatant on fresh beef. Food Microbiology 2017, 62, 207-211, https://doi.org/10.1016/j.fm.2016.10.024.

98. Waghu, F.H.; Idicula-Thomas, S. Collection of antimicrobial peptides database and its derivatives: Applications and beyond. Protein Science 2020, 29, 36-42, https://doi.org/10.1002/pro.3714.

99. Makarova, K.S.; Wolf, Y.I.; Karamycheva, S.; Zhang, D.; Aravind, L.; Koonin, E.V. Antimicrobial peptides, polymorphic toxins, and self-nonself recognition systems in Archaea: an untapped armory for intermicrobial conflicts. MBio 2019, 10, e00715-00719, https://doi.org/10.1128/mBio.00715-19.
monocytogenes in cheese. *Food microbiology* 2017, 63, 159-163, https://doi.org/10.3390/molecules25051134.

98. Peluzio, M.d.C.G.; Martinez, J.A.; Milagro, F.I. Postbiotics: Metabolites and mechanisms involved in microbiota-host interactions. *Trends in Food Science & Technology* 2020.

99. Bagde, P.; Nadanathangam, V. Mechanical, antibacterial and biodegradable properties of starch film containing bacteriocin immobilized crystalline nanocellulose. *Carbohydrate polymers* 2019, 222, 115021, https://doi.org/10.1016/j.carbpol.2019.115021.

100. Silva, D.R.; Sardi, J.d.C.O.; de Souza Pitanguí, N.; Roque, S.M.; da Silva, A.C.B.; Rosalen, P.L. Probiotics as an alternative antimicrobial therapy: Current reality and future directions. *Journal of Functional Foods* 2020, 73, 104080, https://doi.org/10.1016/j.jff.2020.104080.

101. Gbassi, G.K.; Vandamme, T. Probiotic encapsulation technology: from microencapsulation to release into the gut. *Pharmaceutics* 2012, 4, 149-163, https://doi.org/10.3390/pharmaceutics4010149.

102. Rathore, S.; Desai, P.M.; Liew, C.V.; Chan, L.W.; Heng, P.W.S. Microencapsulation of microbial cells. *Journal of Food Engineering* 2013, 116, 369-381, https://doi.org/10.1016/j.jfoodeng.2012.12.022.

103. Anwar, F.; Altayb, H.N.; Al-Abbasi, F.A.; Al-Malki, A.L.; Kamal, M.A.; Kumar, V. Antiviral effects of probiotic metabolites on COVID-19. *Journal of Biomolecular Structure and Dynamics* 2020, 1-10, https://doi/10.1080/07391102.2020.1775123.

104. Koirala, R.; Gargari, G.; Arioì, S.; Taverniti, V.; Fiore, W.; Grossi, E.; Anelli, G.M.; Cetin, I.; Guglielmetti, S. Effect of oral consumption of capsules containing Lactobacillus paracasei LPC-S01 on the vaginal microbiota of healthy adult women: a randomized, placebo-controlled, double-blind crossover study. *FEMS microbiology ecology* 2020, 96, fiaa084, https://doi.org/10.1093/femsec/fiaa084.