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Review Article

Potential of food grade lactic acid bacteria and their bacteriocins in hurdle technology for enhanced food preservation

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Abstract

Lactic Acid Bacteria (LAB) have a long history of safe use in humans and are recognized as GRAS (Generally recognized as safe). Besides lactic and acetic acid which produced by LAB, other inhibitory substances are also produced among which bacteriocins are of high significance owing to their natural and nontoxic nature. Bacteriocins are the ribosomal synthesized antimicrobial protein metabolites produced by a wide variety of microorganisms. The bacteriocin producing attributes of a number of the LAB have been exploited in the potential application in food, agriculture and biopharma industry. In last few decades, the significance of LAB bacteriocins in food industry has attracted the attention of researchers, food manufactures and consumers who are looking for safe and natural products with no toxic properties and less side effects. The potential of LAB bacteriocin as natural and safe alternative are essentials not only due to their spectrum of inhibitory actions aimed towards industrially important spoilage microorganism, but also due to their role in enhancing the texture, quality and safety of the food products. More recently, the synergistic effect of bacteriocins in the presence of other preservation factors used in food preservation has highlighted their significance in hurdle technology. This technology has potential application in bakery, dairy and meat products. In this review we would discuss the role of LAB and their bacteriocin as bio-preservative in food industry and their role in presence and in combination with other hurdles.

Keywords: Bacteriocins, Bio-preservatives, Hurdle technology, lactic acid bacteria.

Introduction

Hurdle technology is defined as the combined effect of different microbial, physical, and chemical preservation factors (selected hurdles) that can control microbiological hazards and other microorganisms in food to achieve and retain the safety and suitability of the end product. In this technology, different preservation techniques are applied together to create hurdles which could overcome or inhibit the growth of unwanted microorganisms. These organisms contaminate food even after processing in various ways, including improper handling, packaging leakage, crosscontamination, and storage under undesirable

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conditions (Yap et al., 2022). Thus, hurdles can preserve foods, while other characteristics are least impacted, and also improve the stability, sensory quality, and nutritional properties of foods, as well as the overall cost.

In recent years, bacteriocins have gained great attention as a part of the hurdle technology. In 2000, Leistner stated that bacteriocins could be included as part of multiple antimicrobial factors or hurdles that could be applied in hurdle technology. The synergistic effect of several bacteriocins in combination with other antimicrobial agents, including chemical preservatives, natural phenolic compounds, and other antimicrobial proteins, is key for their selection in hurdle technologies. The application of different bacteriocins in food products could result in better preservation of the nutritional value and organoleptic properties of foods but could also reduce the content of chemical additives (Leistner, 2000). Additionally, use of different bacteriocins in combination might prevent the development of resistant strains (Huan et al., 2020). Another main advantage of using bacteriocins in combination with treatments, such as high-pressure processing or pulsed electric fields, is that they can provide an additional barrier to endospore-forming bacteria, resulting in more effective preservation of foodstuff and enhanced organoleptic and nutritional properties. Lactic acid bacteria (LAB) are the most commercialized cultures in food that can produce bacteriocins. In fact, use of bacteria as a starter culture for preserving foods has been used for centuries, especially in traditional fermented products, which has also improved the shelf life of foods (Leyva Salas et al., 2018).

Lactic acid bacteria

Lactic acid bacteria are Gram-positive, catalasenegative, non-spore-forming cocci or rods used to produce fermented foods (Mokoena et al., 2021). This group of bacteria are categorized as "generally regarded as safe" (GRAS) by the United States Food and Drug Administration (Wessels et al., 2004). The emphasis on LAB stems from the fact that this group of bacteria, which predominates the intestinal flora,

has a safe history of use and is considered noninvasive and nonpathogenic. In the late nineteenth century, microbiologists identified microflora in the gastrointestinal tract of healthy individuals that differed from those found in diseased individuals. The beneficial microflora found in the gastrointestinal tract are termed probiotics. These miracle bugs were defined by Havenaar (1992) as a "sufficient number of viable bacteria, fungi, or yeast exerting beneficial effects on the health of the host by improving the intestinal microbial balance." Later, the Food and Agriculture Organization (FAO) and World Health Organization (WHO) (2001) defined probiotics as "live microorganisms that, when administered in adequate amounts, confer health benefits to the host." This definition was later adopted by the International Scientific Association for Probiotics and Prebiotics (Reid et al., 2003). LAB, especially Lactobacillus and Bifidobacterium, are the most widely acknowledged probiotic bacteria commonly used in food products to control food spoilage and pathogenic microbes (Zielińska & Kolożyn-Krajewska, 2018). emphasis on LAB stems from the fact that this group of bacteria, which predominates the intestinal flora, has a safe history of use and is considered noninvasive and nonpathogenic. Many LABs produce a heterogeneous group of antimicrobial protein peptides called bacteriocins, which are widely acknowledged for their broad-spectrum inhibition (Alvarez-Sieiro et al., 2016; Darbandi et al., 2022). The effectiveness of bacteriocins is often dictated by environmental factors, such as pH, temperature, food composition and structure, as well as food microbiota (Silva et al., 2018).

Bacteriocin

Currently, there are numerous ways to preserve food to prevent food poisoning or spoilage, such as thermal treatment, water activity, and addition preservatives. In addition, bacteriocins produced by LAB are highly acknowledged for their safety and wide application in food preservation, especially in dairy products (such as milk, yoghurt, and cheese), eggs, meat, and vegetables to control pathogens (Zacharof and Lovitt, 2012).

Bacteriocins are ribosomal-synthesized peptides or proteins with antimicrobial activity that are active against other bacteria and are heat-stable peptides. Furthermore, these bacteriocins have been used directly in purified or semi-purified forms in bioactive films and coatings that are applied to the surfaces of food and packaging (Silva et al., 2018).

LAB bacteriocins can be divided into four classes. Class I includes lantibiotics, which contain amino acids, such as lanthionine and B-methyllanthionine, and several dehydrated amino acids (Guder et al., 2000). On the other hand, Lantibiotics are divided into two subgroups, A and B, based on mode of killing (Simons et al., 2020). Type A lantibiotics depolarize the cytoplasmic membrane of target cells, forming pores or leakage of the essential cell contents, leading to cell death similar to nisin (Bin Hafeez et al. 2021; Pérez-Ramos et al., 2021). Type B lantibiotics are smaller than type A lantibiotics and have a more globular secondary structure like mersacidin, which interferes with cell wall biosynthesis (Bin Hafeez et al. 2021). Class II LAB bacteriocins are also small (ranging in size from 30 to 60 amino acids) and heat stable (Fernandes & Jobby, 2022). They are also allocated into subgroups. Class IIa is the largest group with more activity against Listeria. Class IIa bacteriocins such as pediocin AcH (Bin Hafeez et al., 2021), sakacin A (Sc), and leucocin A (Simons et al., 2020) act through the formation of pores in the cytoplasmic membrane. Class III bacteriocins have large heat-labile proteins such as helveticins I and V (Bin Hafeez et al., 2021). Class (VI) require lipid or carbohydrate moieties for their activity. Little is known about the structure and function of this proposed class, such as leuconocin S (Hafeez et al., 2021).

Hurdle technology

Instead of using a single method for food preservation, multiple methods or hurdles provide better results. Hurdle technology involves intentionally combining methods or techniques to improve not only the stability of microbial and sensory quality of food but also the nutritional and economic properties of food (Leistner, 2000;

Guerrero et al., 2017). Hurdle preservation techniques include low temperatures (chills and frozen storage), low water activity (drying and curing), pH reduction (acidification), low oxygen concentrations (vacuum packing), raised carbon dioxide (modified atmosphere packaging), preservatives (sulfites and nitrites), salts, sugars, and spices. (Mechmechani et al., 2022). The influence of hurdles on the behavior of microorganisms in food, such as homeostasis, metabolic exhaustion, and stress reactions, should be considered (Leistner, 2000).

Role of LAB bacteriocins in hurdle technology

Hurdle technology can be achieved using both LAB and their antimicrobial metabolites, such as bacteriocins (Souza et al., 2022). LAB and bacteriocins have been extensively studied because they are natural biopreservatives (Souza et al., 2022). This can be an alternative to satisfy the increasing consumers demands for safe, fresh-tasting, ready-to-eat, minimally-processed foods and also to develop "novel" food products (e.g. less acidic, or with a lower salt content) (Naskar & Kim, 2021).

LAB have long been used as components of hurdle technologies because of their ability to rapidly decrease the pH of fermented foods and competitively inhibit the growth of other microorganisms, partly due to the production of broad-spectrum bacteriocins (Renye & Somkuti, 2015). In addition, several LAB bacteriocins have potential applications in food preservation, and the use of bacteriocins in the food industry can help reduce the addition of chemical preservatives as well as the intensity of heat treatments, resulting in foods that are more naturally preserved and rich in organoleptic and nutritional properties (Gálvez et al., 2007). Bacteriocin is a natural antimicrobial with the ability to combat pathogenic and spoilage bacteria. It possesses low toxicity, high potency, and a narrow target-specific effect mechanism and is effective at nanomolar concentrations. Some studies have focused on the use of bacteriocins, which inhibit Gram-positive bacteria (Pérez-Ramos et al., 2021; Gálvez et al., 2007). Since bacteriocins have shown limited effects against Gram-negative pathogens, they

could be applied in foods in combination with other hurdles or treatments to induce cell damage and partial disorganization of the outer membrane protective layer of Gram-negative bacteria (Li et al., 2019).

In addition to the available commercial preparations of nisin and pediocin PA-1/AcH, other bacteriocins (such as lacticin 3147, enterocin AS-48, and variacin) also offer promising perspectives. Bacteriocins can be added to foods in the form of concentrated preparations, such as food preservatives, shelf-life extenders, additives, or ingredients, or they can be produced in situ by bacteriocin genic starters, adjuncts, or protective cultures (Gálvez et al., 2007). Broad-spectrum bacteriocins present potential wider uses, whereas narrow-spectrum bacteriocins can be used more specifically to selectively inhibit certain high-risk bacteria in foods, such as Listeria monocytogenes, without affecting harmless microbiota. Therefore, they can be used in combination with other antimicrobial agents, including chemical preservatives, natural phenolic compounds, and other antimicrobial proteins to synergistically exert their effects. Moreover, this method, as well as the combined use of different bacteriocins, can effectively inhibit microbial resistance (Yu et al., 2023; Gradisteanu Pircalabioru et al., 2021). Successful applications have been reported for bacteriocins used with various nonthermal hurdles, including high hydrostatic pressure (HHP), pulsed electric fields (PEF), and irradiation (Table 1). Commonly used hurdles include high short time temperature, (HTST), ultra-high temperature (UHT) pasteurization, refrigeration, salting, pH reduction due to LAB fermentation, and decreased water activity (de Oliveira et al., 2015).

Combination of bacteriocin and physical hurdles against pathogens

Bacteriocin and heat

Heat processing is one of the oldest and most commonly used technologies for controlling pathogens. Heat affects the protein structure, resulting in the inactivation of microorganisms

(Sehrawat et al., 2021). For the first time, heat was used in canning methods to inhibit spore growth and increase shelf life. Because the use of high temperatures can destroy some nutrients, the combination of heat with other treatments, such as bacteriocins, was considered to be more effective. For instance, the use of nisin (2000 IU/mL) in skim milk before heating at 103 °C increases the number of control spores of B. cereus and thermophilic B. stearothermophilus ATCC 12980 (Wandling et al., 1999). Furthermore, heating can overcome the narrow spectrum of activity of bacteriocins; in this case, P. aeruginosa and Salmonella were not sensitive to nisin, whereas the use of this bacteriocin with heating at 55 °C resulted in the sensitivity of these pathogens to nisin. Similar spores of Clostridium sporogenes ATCC 7955 were not inactivated by heating at 90 °C, whereas the use of 16 µg/mL nisin at 90°C inactivated rapidly (Hofstetter et al., 2013).

Bacteriocins and pulsed electric fields

PEF is commonly used for persevering liquid food and is categorized as a non-thermal method of preservation. Viedma et al. (2009) analyzed combination of enterocin AS-48 with high-intensity pulsed-electric field (HIPEF) against cider-spoilage, exopolysaccharide-producing strain Lactobacillus diolivorans in apple juice. They indicated that a higher concentration of bacteriocin (2.0 mg/mL) with a higher HIPEF (1000 µs) resulted in the highest pathogen inactivation (4.87 log). Sobrino-Lopez et al. (2008), found adding only bacteriocin like nisin and lysozyme was not reduced the count Staphylococcus aureus effectively, but when combined with HIPEF, can decreased up to 6.2 log of target bacteria. The reduction was achieved using 1,200-us HIPEF, 1 IU/mL nisin, and 300 IU/mL lysozyme at pH 6.8. In a subsequent study, they analyzed the effect of AS-48 with nisin, lysozyme, or both, as well as with the use of HIPEF on Staph. aureus in milk (Sobrino-Lopez et al., 2009). Decreased counts of S. aureus was observed when treatment with AS-48 plus use of HIPEF as well as addition of AS-48, nisin and HIPEF. The best concentration was obtained by using 28 AU/mL of AS-48,20 IU/mL of nisin, and treatment with HIPEF for $800 \mu s$, which resulted in a reduction of over 6 logs in the bacteria. They also indicated that addition of bacteriocin before HPEF treatment had a greater effect than addition of bacteriocin after this treatment. Similarly, Viedma et al. (2010) showed that an enterocin AS-48 concentration of 0.613 AU/mL in combination with an HPEF treatment time of 1,000 ms decreased the *pediococci* by 6.6 log in apple juice. Novickij et al. (2018) evaluated the combination of nisin with nanosecond in *E. coli*. They found that a PEF duration of 20–30 kV/cm, ranging from 500 to 900 ns, was adequate for permeabilization of the target bacteria. They also showed that 2.5 min a time resolution is effective for controlling the time used for PEF.

A moderate electric field (MEF) is another electric field technology, in comparison with PEF, a lower electric field (1 V/cm to 1 kV/cm) is used in the MEF, whereas in the PEF, the electric field is generally 10–

14 kV/cm (Mok et al., 2020). The inhibitory effect of the combination of shear-stress MEF (SS-MEF) and nisin on *E. coli* K12 and *L. innocua* in apple juice has been analyzed (Mok et al., 2020). They showed that this combination could reduce harmful bacteria by 5-log less than 5 min, whereas the color and pH of the juice did not change.

Bacteriocins and high-pressure processing

Another technique categorized as nonthermal processing is high-pressure processing (HPP), which uses water as the transmission medium of pressure while maintaining the nutritional, sensory, and functional properties of food.

This technique is commonly used for processing of fruit juice and meat (Liu et al., 2022). HPP has a greater effect on the microbial membrane, and the

Table 1. Combined antimicrobial effect of bacteriocin with other hurdles

Bacteriocin	Other treatment	Target microorganism	Food matrix	Reference
	Pulsed electric fields	L. innocua	Liquid whole egg	Calderón-Miranda et al. (1999)
	Pulsed electric fields + Carvacrol	B. cereus	Skim milk	Pol et al. (2001)
	Pulsed electric fields	S. aureus	Skim milk	Sobrino-Lopez and Martín-Belloso (2006)
Nisin	Pulsed electric fields	L. innocua	Whey	Gallo et al. (2007)
	High pressure homogenization	E. coli; L. innocua	Apple and carrot juice	Pathanibul et al. (2009)
	High pressure	Spores of <i>B. subtilis</i> and <i>B. cereus</i>	Milk	Black et al. (2008)
	Carbon dioxide, NaCl and low temperature	L. monocytogenes	Cold-smoked salmon	Nilsson et al. (1997)
Enterocin AS-48	High-intensity pulsed- electric field	S. enterica	Apple juice	Viedma et al. (2008)
	High hydrostatic pressure	L. monocytogenes, S. enterica and S. aureus	A low acid fermented sausage	Ananou et al. (2010)
Enterocin LM-2	High hydrostatic pressure	L. monocytogenes and S. enteritidis	refrigerated shelf life of sliced cooked ham	Liu et al. (2012)
Lactocin AL705	High hydrostatic pressure	L. innocua	ready-to-eat sliced cured- cooked pork	Dallagnol et al. (2017)

sensitivity of bacteria to this method varies and includes Gram-negative bacteria more than Grampositive bacteria and spores (Liu et al., 2022).

The synergistic effect of high pressure and bacteriocin on the death rates of *S. aureus* and *E. coli* in meat was investigated by Garriga et al. (2002). Although S. aureus was the least sensitive to HPP, addition of nisin led to a significantly lower count of target bacteria at 4 °C. In addition, E. coli was inactivated when nisin was used, and the counts of the remaining bacteria remained unchanged for 61 days at 4 °C. The inactivation of *E. coli* in cheese was higher when HPP was applied at 300 MPa on day 50, whereas adding lacticin 481-, nisin A-, bacteriocin TAB 57-, or enterocin AS-48 decreased the number of E. coli 0157:H7 to levels below 2 log units on day 60 of storage (Rodriguez et al., 2005). They also indicated that the use of HPP (300 MPa) with bacteriocin on day 50 completely inactivated *E. coli* in 60-day-old cheese. Similarly, Arqués et al. (2005a) showed that the use of HPP on day 51 was more effective than on day 2 in decreasing the count of *Listeria monocytogenes* in cheese. The count of *L.* monocytogenes in cheese treated with bacteriocin and 300MPa was 3.83-5.43 log cfug⁻¹, whereas use of 500 MPa on day 2 reduced up to 1.81 log cfug-1. In addition, L. monocytogenes was not detected in cheese samples treated with HPP on day 50. Similar results showed that the use of HPP (300 and 500 MPa) with bacteriocin reduced S. aureus in cheese samples (Arqués et al., 2005b). The combined effect of HPP and enterocin LM-2 on the shelf life of refrigerated sliced ham and cooked ham was evaluated by Liu et al. (2012). They found that HHP at 400 MPa with enterocin 256 or 2560 AU/g could extend the shelf life of products to 70 and 90 days, respectively. They also indicated that *L. monocytogenes* and *S. enteritidis* were reduced in sliced cooked ham stored at 4 °C after treatment of HPP with bacteriocin. Oner (2020) indicated that the combination of nisin and HPP was more effective against E. coli than against L. innocua in green juice. They also found that nisin combined with HPP had no effect on pH, whereas the color of the green juice changed.

Bacteriocins and nanotechnology

Nanotechnology involves the manipulation of functional materials and structures to nanoscale sizes (with diameters ranging from 1 to <1000 nm) (Klaessig et al., 2011). Nanoparticles are the basic elements of nanotechnology, but most of these elements do not show antimicrobial activity, except silver, which has a wide range of antimicrobial activities (Sulthana & Archer, 2021). The combination of bacteriocin with conjugated nanoparticles has various advantages, such as lowering the level of bacteriocin, which protects bacteriocin from proteolytic enzymes and can also survive better during long-term storage (Sulthana & Archer, 2021).

Sharma et al. (2012) showed that enterocin-coated silver nanoparticles had a wide broad of inhabitation against food borne pathogens. In addition, AgNPs with bacteriocin from Lactobacillus paracasei showed higher antimicrobial activity against S. aureus and P. aeruginosa (Gomaa, 2019). Similar results showed that two bacteriocin from Lactobacillus strains capped with silver nanoparticles had higher activity against S. aureus and Shigella flexneri (Sidhu & Nehra, 2020). The inhibition zone of bacteriocin-selenium nanoconjugate against P. aeruginosa, Candida albicans and Candida tropicalis were 15.625 µg mL-1 and $31.25 \,\mu g \, \text{mL}^{-1}$, $52 \,\text{mm}$ and $47 \,\text{mm}$ (±SD), respectively (Al-Shimmary & Al-Thwani, 2024).

Industrial preservatives recently used in hurdle technology

Microencapsulation

Microencapsulation has recently been used in the food industry and can be used as an additive, enzyme, and bioactive compound, resulting in improved nutritional quality (Shamloo et al., 2019). Microencapsulation is defined as active packaging in small compartments that releases their components gradually during extended periods (Muthuvelu et al., 2023). Since bacteriocins can be affected by proteolytic degradation in the food matrix, nanoencapsulation with metal nanoparticles, chitosan, nanofibers, and liposomes can overcome this limitation and improve their activity.

The growth inhibition of *L. monocytogenes* by various concentrations of microencapsulated nisin (16, 31, and 63 µg/mL) with alginate-cellulose nanocrystals was studied by Hug et al. (2014). They found that different concentrations of nisin significantly reduced L. monocytogenes (16, 31, and 63 µg/mL nisin decreased by 2.65, 1.50, and 3.04 log CFU/g, respectively) after 28 d of storage. The antimicrobial activity of chitosan-nisin microcapsules against Bacillus subtilis CICC10275 was higher at pH 5 and 6 than 7.4 (Hu et al., 2017). Moreover, the 1% chitosanmicrocapsules exhibited the nisin antimicrobial activity. The activity of bacteriocin produced by *Pediococcus acidilactici* ITV26 encapsulated in liposomes was evaluated by García-Toledo et al. (2019). These results indicate that the encapsulation efficiency was approximately 88%, and the inhibition activity was higher than that of the control. Additionally, bacteriocin release occurred slowly over 120 h at various pH values (4, 5,6 and 6.8). Ramalho et al. (2023) showed that microencapsulation of enterocin from Enterococcus durans in whey powder had more activity against E. coli, L. monocytogenes, L. innocua, and L. ivanovi compared to unencapsulated enterocin as control. They also found that encapsulated enterocin prevented Listeria sp. within 12 h compared to the control samples.

Immobilization

Immobilization is another technique for preservatives that are used in different products, such as confectioneries, syrups, favors, fruit beverages, milk products, yeasts for baked goods, and whey lactose hydrolysates (Muthuvelu et al., 2023). Immobilization techniques are categorized as antimicrobial packaging systems that help to release bacteriocin from food slowly and extend shelf life.

The antimicrobial activity of bacteriocin produced by *Lactobacillus sakei* immobilized on bacterial cellulose membranes produced by Gluconacetobacter xylinus against *L. monocytogenes* was higher than that of free bacteriocin (Malheiros et al., 2018). In contrast, both free and entrapped bacteriocins could inhibit the target bacteria after 5 days. In another study, the stability of bacteriocins from *Enterococcus faecium*

and *Pediococcus acidilactici* immobilized by physical adsorption onto cellulose nanocrystals from cotton linters was analyzed (Bagde & Vigneshwaran, 2019). The stability of immobilized bacteriocins increased by up to 50% in terms of antimicrobial activity. Kumari et al. (2021) compared the antimicrobial activity of bacteriocin produced by L. plantarum by two methods include immobilized and freeze dried in pineapple wine and then tested the antimicrobial activity against L. acidophilus, A. aceti, S. cerevisiae, E. coli, S. aureus and B. subtilis. They found that the inhibition zone of immobilized bacteriocin was higher than that of freeze-dried bacteriocin, and the activity was effective at pH 3.5. In addition, the organoleptic properties of immobilized bacteriocin were better than those of freeze-dried bacteriocins.

Biopolymers

Biopolymers are used as intelligent and active polymer systems in the food industry (Muthuvelu et al. 2023). This technique is more effective for micronutrients and antioxidant compounds, and therefore can improve the nutritional value and shelf life (Chiralt et al., 2020).

In a previous study, bacterial control in fish packaging using a film prepared with Pectin, Gellan, EDTA, and bacteriocin-like compounds produced Streptococcus infantarius was evaluated against E. coli, S. aureus, and L. monocytogenes (Pérez-Arauz et al., 2021). Whilst Streptococcus infantarius alone cannot show inhabitation activity against E. coli and S. aureus, but this film blend can inhibit the growth of all three target bacteria. They also assumed that EDTA, which acts as a permeabilization agent in the outer membrane, is more sensitive to bacteriocins. Contessa et al. (2021) designed active packaging using biopolymers mixed with a bacteriocin extract of Lactobacillus sakei. They found that the mechanical properties and water vapor permeability of this packaging were unchanged, whereas the swelling property decreased. Moreover, visible light protection was enhanced, and when used in cream cheese packaging, microbiological stability increased. A bioactive biopolymer based on Latilactobacillus curvatus 54M16 producing bacteriocins showed antimicrobial activity against L. innocua C6 after 28 days of storage at various temperatures (4 °C, 10 °C, 20 °C, and 30 °C) (Giello et al., 2023). In addition, the count of *L. innocua* on fennels processed with the active coating showed a significant decrease of about 2 log cycles compared to control samples (3.42 to 4.13 log CFU/cm²) at the end of storage.

Conclusion

Foods should be regarded as complex ecosystems where microbial interactions significantly influence the microbial equilibrium and the proliferation of either beneficial or harmful bacteria. Recent advancements in molecular microbial ecology offer insights into the global effects of bacteriocins within food ecosystems, and the examination of bacterial genomes may uncover new sources of bacteriocins. Currently, hurdle technology is employed in both industrialized and some developing countries as a gentle, reliable, and cost-effective method for food preservation. Comprehensive investigations into the impact of food preservation techniques on the physiology and behavior of microorganisms in foods, specifically their homeostasis, metabolic exhaustion, and stress responses, have led to the development of the novel concept of multi-target food preservation.

Conflicts of Interest

Authors have no conflict of interest.

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