

Applications of Bacteriocin Production for Food Processing Applications

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Abstract

Bacteriocins, ribosomally synthesized antimicrobial peptides produced by the lactic acid bacteria have garnered significant attention for their crucial role in food processing. This review highlights the current state of knowledge on bacteriocin production, emphasizing its application in enhancing food safety, extending shelf life, and improving nutritional quality. Furthermore, the review aims to stimulate further research and development of bacteriocin-based applications, highly contributing to the creation of safer, more sustainable, and higher quality food products. The usage of bacteriocins can be purified or on a crude form or through the use of a product that has passed through fermentation process with a bacteriocin-producing strain as a constituent in food processing or which can be integrated through a bacteriocin producing strain.

Keywords: Bacteriocins, Lactic Acid Bacteria, Food Preservation, Food Safety.

I. INTRODUCTION

The term "bacteriocins" describes ribosomally produced proteins or peptides which displays either lytic or inhibitory roles against bacterial cells, irrespective of the bacteria belonging to members of the same genus as the producing bacteria or related genera; in some circumstances, they can constitute wide range of microorganisms (Heilbronner et al., 2021). In 1925, a characterized bacteriocin was first reported. It was termed "colicin", which was named after *Escherichia coli*, the bacteria it originated from (Negash and Tsehai 2020). The majority of bacteriocins that are created are now known to originate from gram-positive bacteria. This does not fit the properties of colicin. Bacteriocins tend to be more active against gram-positive strains of bacteria (Cintas et al., 2006). A modified amino acid called lanthionine is present in certain bacteriocins, including nisin. This amino acid is created during the molecule's translation. Bacteriocins are peptides compounds having antimicrobial properties produced from both gram-positive and gram-negative bacteria. Bacteriocins can have cationic, anionic, and neutral properties (Karthikeyan and Santosh 2009). Bacteriocins are extracellular peptides or bioactive complexes released by the lactic acid bacteria. The majority of bacteriocins are produced by lactic acid bacteria. Small cationic peptides with 30 to 60 amino acid residues that are resistant to

heat are typically the bacteriocins released by lactic acid bacteria (Ferreira et al., 2007). The molecular weight of bacteriocins is within the range of 2000 Da to over 30,000 Da and are of proteinaceous nature which enable them to perform post-translational modification. Bacteriocins have many functions and provide lots of benefits. They can play roles as antagonists and as a result of this property that they are used as food biopreservatives. Bacteriocins are defined by several characteristics: (1) they possess a relatively narrow inhibitory spectrum of activity targeting mostly bacterial strains associated with the bacteriocin-producing species (2) the active compounds consist majorly of a protein fraction, (3) they exhibit bactericidal mode of action (4) they contain specific cell receptors that can recognize and inhibit or kill target bacterial cells and (5) the genes of their determinant are found in plasmids that function in production and immunity (Dimov et al., 2005).

In this review, the application of bacteriocin production for food processing to ensure enhanced food safety and quality will be discussed. In the subsequent sections, classification of bacteriocin, their mechanisms of action and structure, isolation and purification of bacteriocins, applications in food processing and limitations that are associated with resistance and purification will be analyzed.

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II. CLASSIFICATION OF BACTERIOCINS

The classification of bacteriocins has passed through significant revisions as a result of the vast expansion of knowledge and understanding about these biomolecules. From this research, the first group of classification can be related to the producing microorganism, which are the Gram-positive bacteriocins and Gram-negative bacteriocins, although some authors also accepted those ones that the archaea organisms produced like the halocins (Zimina et al., 2020).

➤ *Gram-Positive Bacteriocins*

Gram-positive bacteriocins are produced by the gram-positive bacterial genera such as *Lactobacillus* and *Staphylococcus*. They can be categorized into three classes based on the nature of their post-translation modifications: class I, class II and class III (Negash et al., 2020).

• *Class I:*

They can as well be called lantibiotics, they have a molecular weight of less than 5kDa, they are thermostable in nature and with a large rate of post-translation modifications. They have large quantity of amino acid structures which can be found within their structure. This includes lanthionine and methyl-lanthionine (from which the name was derived) and also unsaturated amino acids. This composition of these amino acids enables them to form ring structures that are intramolecular in nature, generally through the di-sulfide bonds (Zimina et al., 2020). An example of bacteriocin in this category is the nisin. This class can be further subdivided into the following:

- ✓ Class IA: consists of structures that are polar in nature having a net charge that is positive.
- ✓ Class IB: they are globular peptides that lack a net charge or have a net charge that is negative in nature.

• *Class II:*

This class of bacteriocins of gram-positive bacteria entails equally small molecules, that is usually <10 kDa and can be further classified into the following (Antoshina et al., 2022).

- ✓ Subclass IIA: consists of peptides whose activity is against *Listeria* (food-borne disease pathogenic bacteria) Example include: leucocin B-TA1 Ia and pediocin PA-1/AcH.
- ✓ Subclass IIB: correlate with peptides that are confined in a dimeric arrangement, whereby two peptides that are unaltered behave in a synergistic manner in order to achieve the antimicrobial effect. For example lactococcin G and lactococcin (Sharma et al., 2021).
- ✓ Subclass IIC: includes peptides that are circular in nature e.g. lactococcin B

- ✓ Subclass IID: consists of linear peptides whose activity is not against *Listeria*. This subclass contains two-component bacteriocin, enterosine L50 and the single peptide enterosine Q, produced by *Enterococcus faecium* L50, and Aurosin A70, produced by *Staphylococcus aureus* A70.
- ✓ Subclass IIE: They are bacteriocins with cyclic peptides. Contrary to linear bacteriocins, these bacteriocins are cyclic in nature by forming a head-tail peptide binder. Example: AS-48, gassericin A, and circularin A
- ✓ Subclass IIF: These are other unmodified bacteriocins. This group contains other class II bacteriocins which do not take the properties of the structure and motifs of any of the listed subclasses.

• *Class III:*

Composed of bacteriocins that are proteinaceous in nature with a high molecular weight that is >30 kDa, they have the potential to have complex structure that is connected to their function, this enables them to be thermolabile. For example, helveticin J, lactacins A and lactacin B. This group has privilege of containing some bacteriocins that are also produced by gram-negative bacteria under specific conditions, such as klebeicin (Negash et al., 2020).

➤ *Gram-Negative Bacteriocins*

There are only two groups present in these classification which is according to the bacteriocins produced by gram-negative microorganisms. This group of bacteriocins are mostly isolated from producing strains of *E. coli* or from other enterobacteria. The two types of bacteriocins present in this group are Colicins and microcins (Negash et al., 2020).

- Colicins: These are proteins that have antibacterial nature with a molecular weight of 30–80 kDa, generally produced by strains of *Escherichia coli* that contain a plasmid known as colicinogenic. Some researchers suggested the subdivision of this group into two classes: Colicins produced by *E. coli* specifically, which is as a result of the type of plasmid from which they are gotten from, and the second group are the colicins produced by other member of *Enterobacteriaceae*; but this classification has not yet been fully accepted by all authors (Negash et al., 2020; Perez-Ramos et al., 2021).
- Microcins: These include low molecular weight bacteriocins which are about 1-10 kDa with a highly active and stable molecular structure and a wide pH range. They are insensitive to proteases function (a highly desirable property present in microbiomes like the digestive system of humans) also they are resistant to temperature changes (Perez-Ramos et al., 2021).

Table 1 Examples of Bacteriocins that are Produced by Lactobacilli (Rodriguez et al., 2003; Chen and Hoover, 2003).

| Bacteriocin | Bacteriocin producing strain |
|---|---------------------------------|
| Lactacin F | Lactobacillus johnsonii spp |
| Lactocin 705 | Lactobacillus casei spp. |
| Lactocin G | Lactobacillus lactis spp. |
| Lactococcin MN | Lactococcus lactis var cremoris |
| Nisin | Lactococcus lactis spp. |
| Leucocin H | Leuconostoc spp. |
| Plantaricin EF, Plantaricin W Plantaricin JK, Plantaricin S | Lactobacillus plantarum spp |

III. MECHANISM OF ACTION OF BACTERIOCINS

Some of the mechanism of action of bacteriocins to inactivate bacteria is so related to the activities performed by traditional antibiotics, and they include the following (Reis et al., 2012; Maryam et al., 2017).

➤ Action on Cell Wall Synthesis

Lactococcus lactis produces Nisin A, which is a bacteriocin that function by inhibiting the synthesis of

cell wall in two ways, first is by the attachment to lipid II in membrane which act as a precursor in synthesis of cell wall or by producing pores in membrane which can lead to cell lysis. The first mode of action is bacteriostatic activity and the other action is bactericidal activity. Nisin therefore plays a crucial function in antibiotic resistance against some gram-positive bacteria thereby rendering it beneficial both in the pharmaceutical and food industries (Allard et al., 2007; Reis et al., 2012).

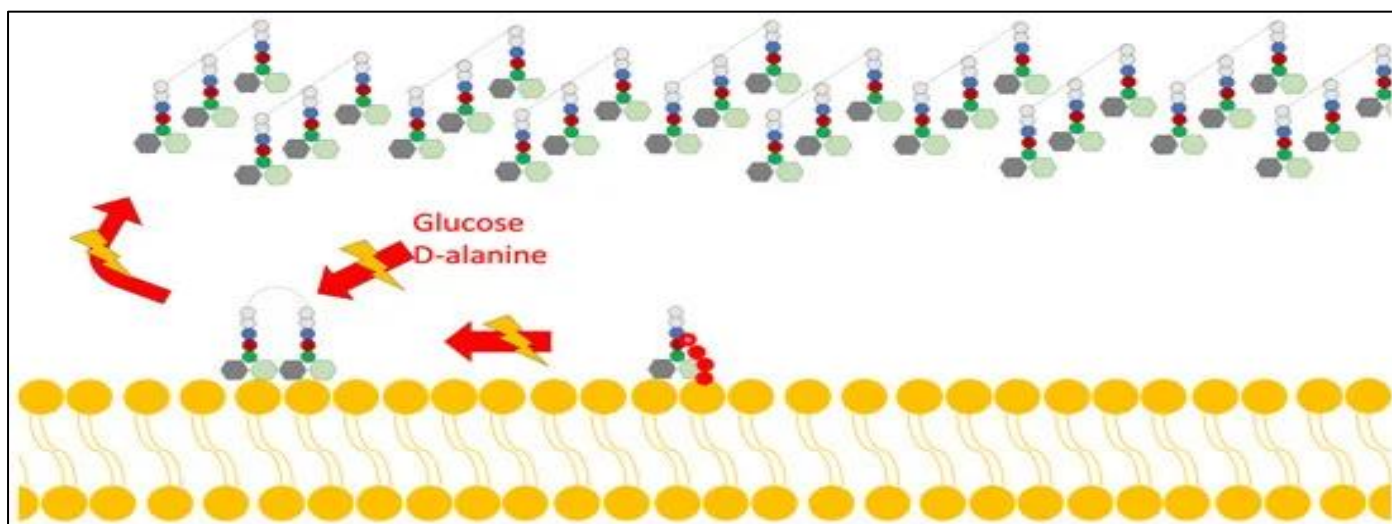


Fig 1 Scheme of the Mechanism of Action of Lantibiotics via the Inhibition of cell wall Synthesis (Sharma et al., 2021)

➤ Inhibiting DNA Synthesis

There is a structural difference between DNA gyrase of human and bacterial DNA and therefore a major region for antimicrobial action. Quinolones which are group of antibiotics inhibit the synthesis of DNA by preventing the unwinding of DNA during replication. Bacteriocins such as microcin B17 also have this type of similar inhibition activity, but are not accepted yet for use by humans (Collins et al., 2013).

➤ Interference with Protein Synthesis

Bacteriocins such as colicins, function in the inhibition of the synthesis of protein by interfering with translation of proteins at different stages. Bacteriocins including colicins E3-E6 and cloacin DF13 exhibit rRNase activity at 16S. Group E colicins have endonuclease mode of action and they prevent translation by performing their function on 30'end of the coding sequence and attaching to the 16S rRNA. Some colicins

such as D and E5 are tRNases perform the function of prolonging the exhaustion of tRNA leading to limited action of protein synthesis (Akutsu et al., 1989; Ogawa et al., 2006; Ng et al., 2010).

➤ Disrupting Membrane Structure

Bacteriocins can make membrane integrity destruction to occur by attaching to the lipid II site and creating holes which can lead to leakage of cell content. Therefore, one important bacteriocin is nisin, and some others are Bac-GM17, PlnJ/K, PlnE/F, Pep5, I and epidermine. Some bacteriocins, such as lactacin Q, do not attach to lipid II, but perform their function by producing toroidal pores from which lipid can alter movement resulting in the leakage of protein and other constituents of the cell causing lysis of cell. Also, carnocycline A also relates directly with bilayer section of the lipid and creates ion-specific membrane pores (Yoneyama et al., 2009; Li et al., 2013).

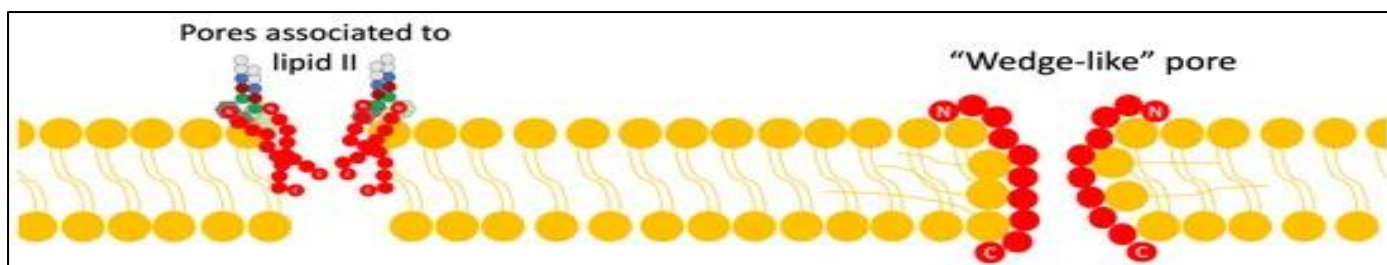


Fig 2 Scheme of the Mechanism of Action of Lantibiotics via pore Formation. (Sharma et al., 2021).

➤ *Formation of Septum*

Septum formation is another innovative mode of action performed by bacteriocins that take place in the replication of the surface of mucopeptide and cytoplasmic membrane during mitosis. During cytokinesis, bacteriocins play a key role as an effect of forming bulges and actively destruction of cell cycle. This nature of antimicrobial inactivation is not discovered yet by two bacteriocins. The two bacteriocins consist of lactococcin 972 which function against *Lactococcus* that are of closely related species and also garvicin that can inhibit *Lactococcus graviae* strains (Martinez et al., 2000; Maldonado-Barragan, et al., 2013).

IV. ISOLATION OF BACTERIOCINS FOR THEIR APPLICATIONS

For the isolation and application of bacteriocins, the procedures for detecting and determining the spectrum of action, isolation, and additional characterization have been advancing alongside with the understanding of these molecules. After the first assessment, detecting strains that produce a bacteriocin with great biotechnological function is performed through several procedures, some of which are: the point inoculation method, oxford cup method, cross-streaking method, radial-streaking method, diffusion-well method, disk diffusion method, agar insert

method (Yadav et al., 2023). After identifying the strain of bacteria known to produce bacteriocin of interest, next is the extracting and purification step. The first step is by cultivating the producing strain in a proper liquid medium, where we can get the CFS from, where several metabolic products released by the bacteria, like the bacteriocins, are present (Ye et al., 2021).

In addition, this CFS has experienced numerous purification approaches in order to acquire the bacteriocin of interest in its pure state. The most used methods include are Ion exchange and gel chromatography, HPLC, reverse-phase chromatography, and solvent fractionation, among others that makes sure these components of interest to be separated from other impurities that may interfere, as well as other particles of the culture medium. All these approaches have been proven to work effectively after thorough research, as reported by Ye and other authors (Ye et al., 2021). In conclusion, after the collection of a purified bio-product, the approaches required for identification can be employed. These tests are performed for the sake of comprehending the structural component of the bacteriocin (Mass Spectroscopy or IR) and understanding its stability (Enzymatic sensitivity tests, stability in pH gradient, and thermostability, among other tests) (Ye et al., 2021; Li et al., 2021).

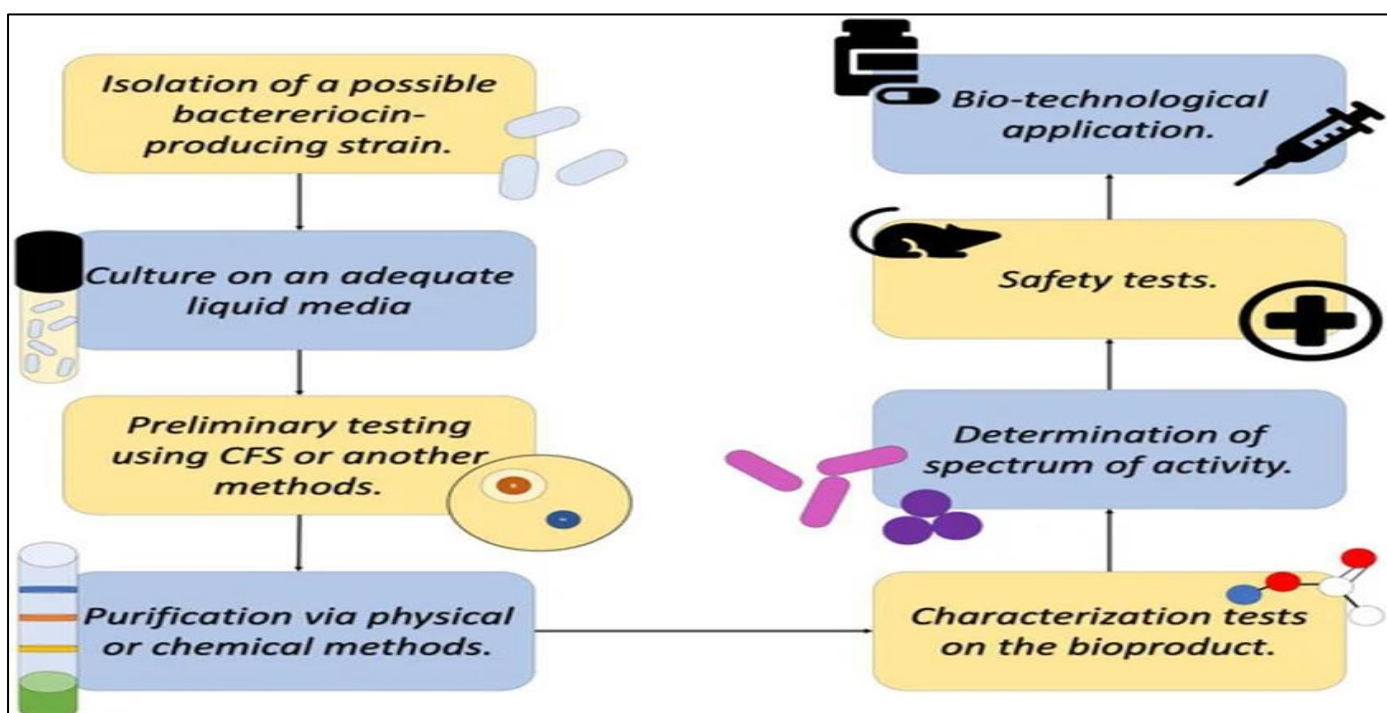


Fig 3 Diagram Showing a General Procedure of Detecting, Isolating, and Characterizing Bacteriocin. Adapted from (Twomey et al., 2021).

V. FACTORS AFFECTING THE EFFECTIVENESS OF BACTERIOCINS

The action that is performed by different bacteriocins produced by different LAB is irregular and not consistent and relies on the composition of chemicals and physical parameters of food; its dependence is solely on pH and is shortened by bacteriocin that attaches to food components, adsorbing to cell wall, mechanism of action of proteases and other enzymes (Schillenger et al., 1996). At specific concentrations, NaCl can slow the development of LAB and alternatively the release of bacteriocins, aside from defending the bacteria of interest such as *Listeria monocytogenes* from their action (Hugas et al., 2002). Sarantinopoulos et al. (2002) noticed the setbacks in the activities of bacteriocin and *E. faecium* FAIR-E 198 growth rate following the addition of 2% NaCl to MRS broth. Nilsen, Nes, and Holo, 1998 as well assigned this protocol to the effectiveness of NaCl in the production attributes joining the receptor and inductor.

Besides having with to interact with some food particles, bacteriocins can be affected by processing and storage factors such as pH and temperature of the food produce. According to Drosinos, Mataragas, Nasis, Galiotou, and Metaxopoulos (2005), the standard pH for bacteriocin production is 5.5 and this cannot be compared to the microbial growth which is 6.5. As a result of their extreme stability in the presence of acidic conditions, the activity is more effective when applied in acidic food products. Therefore, to use nisin more effectively, it is required that the pH of food should not be up to 7 so that satisfactory solubility can be derived, stability during the processing and the period of storage (Drosinos et al., 2005). Leroy and De Vuyst (1999) report shows that bacteriocin activity is slowed down when the temperature range increases caused by the increase in the action of proteases. The inhibitory effectiveness of bacteriocins can also be connected to the degree of food contaminants by the organism of interest. If the first contamination is on a high rate, mechanism of action of bacteriocin is reduced as such and will be difficult to inhibit the growth of the unwanted organisms (Hernandez et al., 1993).

VI. POTENTIAL APPLICATIONS OF BACTERIOCINS IN THE FOOD INDUSTRY AS FOOD PRESERVATIVES

As regards to the report given by Lahiri et al. (2022) some of the qualities bacteriocins should have is that it should not be harmful to humans when it is consumed and should not have any bad effect in the human gut microbiota, It must show broad spectrum of antimicrobial function against foodborne microorganism, should have the ability to resist enzymes in the food matrix, be stable at high temperatures under broad range of pH values and several concentrations of salt content. LAB-bacteriocins can display a broad or narrow spectrum of activity, enabling them in some distinct times to specifically target harmful bacteria, such as *Listeria monocytogenes* (Todorov et al., 2022). Several preservation techniques though, have been employed so as to avoid contamination

and spoilage of food. These procedures can involve the use of thermal treatment (pasteurization, heating sterilization), pH and reduction of water content (acidification, dehydration) and application of preservatives (antibiotics, organic compounds such as propionate, sorbate, benzoate, lactate, and acetate).

One of the disadvantages of incorporating bacteriocin producing strain is because of the insufficient compatibility between the strains that produces bacteriocin produces and some other cultures that require fermentation (Paul Ross et al., 2002). However, it has been discovered that the usage of bacteriocin alone in a food may not guarantee complete safety; especially when it involves the use of Gram negative bacteria. Therefore, bacteriocins is best used when it is applied together with other techniques that showcase the potentials to destroy the cellular membrane so bacteriocins can kill the pathogenic bacteria (Daw and Falkner 1996).

➤ *Application of Lactic acid Bacteria and Bacteriocins in food as Natural Preservatives Against the Growth of Listeria Monocytogenes*

Bacteriocins has been the most commonly used antimicrobial compounds which has been widely employed in various food. This is because it can inhibit and kill several food contaminants effectively. In addition, bacteriocins are also capable of being produced. They are very active at low temperatures. They can work in association with treatments that work effectively at low storage temperature to enhance the safety of foods against pathogens, especially organisms like *Listeria monocytogenes* that adapt well at low temperatures. Another beneficial aspect of bacteriocin is that it provides an antimicrobial effect without altering the taste and appearance of the resulting product, therefore its application possibilities is numerous. Some of these bacteriocins produced by lactic acid bacteria from food have been isolated and proven to have antimicrobial effects against *L. monocytogenes*, including nisin produced by *Lactococcus lactis*, plantaricin UG1 from *Lactobacillus plantarum* derived from sausage, plantaricin D from *Lactobacillus plantarum* strain BFE 905 isolated from salad vegetables, thermophilin 347 produced by *Streptococcus thermophilus* 347 isolated from yogurt, amongst others. Research on applying lactic acid bacteria and the compounds produced as anti-*Listeria* to food products has been experimented on fresh food products (Mahankumar and Murugalatha, 2011).

➤ *Application of Bacteriocins in Poultry Meat and Livestock*

Bacteriocins produced by lactic acid bacteria have been widely studied to provide antagonistic effects against *Listeria monocytogenes* when applied to meat products aside from being added in the form of bacterial culture. The antimicrobial impact of commercial nisin has been discovered by Udhayashree et al. (2012) to stop the growth of *Listeria monocytogenes* in turkey meat. The turkey is marinated in hot water infused with nisin. *Leuconostoc monocytogenes* can be employed by two methods: by soaking the turkey in hot water or applying

directly to the skin of the turkey skin. The application of nisin can mitigate the amount of *L. monocytogenes* by 1 log CFU/ml when cells were rubbed directly to turkey skin. At cold temperature the reduction is continuous. The heat that is released from the hot water used for soaking can indicate a synergistic effect with nisin, revealed by a more significant decrease of log 2 after immersion when *L. monocytogenes* cells were given to the washing water. After 48 hours of being refrigerated, the *L. monocytogenes* cells were all destroyed as a result of the treatment. When nisin is used in combination with EDTA, it was also shown to have an antagonistic effect against *L. monocytogenes* in fresh beef cut into cubes (2.5 cm³). After the inoculation of the meat with about 7 log CFU/ml *L. monocytogenes* Scott A and immersion into a solution of nisin or nisin that is added to EDTA for 10 minutes (Azhar et al., 2017; Udhayashree et al., 2012). After draining the samples for 15 minutes, they were vacuum packed and refrigerated at 4°C for 30 days. The results revealed the immersion in the nisin solution made the population of *L. monocytogenes* to decrease by 2.01 CFU/cm². Also, Cleveland et al. (2001) made use of bacteriocins in its powered state that were derived from milk-based media and applied to packaging films.

➤ *Application of Bacteriocins to Fish and other Aquatic Inhabitants*

Majorly bacteriocins are the major compounds that are effective against *L. monocytogenes* in fish products or foodstuffs of aquatic origin. Nisin when used in combination with dehydroacetic acid and applied to the catfish cut (*Ictalurus punctatus*) can prevent the growth and also kill *L. monocytogenes*, which has been introduced in the product as much as 105 CFU/ml. The fish meat products are processed and stored at 2°C for six days. If nisin is used alone at a concentration of 0.1% or when used together with 0.1% dehydroacetic acid, it can reduce the growth of *L. monocytogenes* by 2.2 and 3.1 log CFU/ml, respectively. Using nisin with NaCl treatment, CO₂ regulation and storage at low temperatures also revealed antagonistic activity against the replication of *L. monocytogenes* inoculated in smoked salmon that is at a cold temperature (Arquest et al., 2015). According to the investigations of Bharal and Sohpal (2013), employing the use of nisin together with mild heat increased the antimicrobial attributes of *L. monocytogenes* in coldpacked canned lobster. Combination of nisin with salt solution when introduced into a basin filled with lobster as high as 25 mg/kg of the contents of the cans, together with continuous heating process until the internal temperature gets to 60°C for 5 min or 65°C for 2 min, reduces the potency of pathogenic bacteria, *L. monocytogenes*, at 3-5 log CFU/g. The inhibitory listeria nature of lactic acid bacterial cultures was researched by Cleveland et al. (2001). They revealed the characteristics of *Carnobacterium piscicola* to prevent the growth of *L. monocytogenes* in smoked salmon that is cold and kept in vacuum packaging kept at 5 °C without altering their sensory properties. Contrary to this investigation, Cintas et al. (2006) investigated the inhibition of *L. monocytogenes* by strains. *Carnobacterium* and crude extract of bacteriocin were released in cold smoked salmon, which was air tight

packed and kept at cold temperature. *Carnobacterium piscicola* V1 exhibited bacteriocidal effect against *L. monocytogenes* at 2°C, while *Carnobacterium divergens* V41 had a bacteriostatic impact. *Carnobacterium piscicola* SF668 at a temperature of 8 °C extended the lag phase, and at 4°C showed bacteriostatic effect on *L. monocytogenes*. The replication of listeria was independent and showed no effect by non-bacteriocin compounds produced by *C. piscicola*.

➤ *Application of Bacteriocins on Eggs*

Bacteriocins that is released by lactic acid bacteria also has antagonistic effects against *L. monocytogenes* in liquid egg products. Arabestani et al.(2014) investigated the D and Z figures of *L. monocytogenes* Scott A in liquid whole eggs after being immersed with the combination of 90 or 10 mg/mL nisin and NaCl (0 or 10%). When nisin is added to unsalted liquid eggs can result in the rapid reduction of the presence of bacteria by 4 logs in an hour. Nisin was revealed to reduce the D value in liquid whole eggs, with the presence of salt, and pasteurized at a temperature of less than 58°C. Also, for liquid eggs which were pasteurized at the standard pasteurization temperature in Canada and the US (60°C with absence of salt or 63°C with presence of salt), whenever nisin is added 2 hours before undergoing the treatment of pasteurization, treatment showed a significant reduction in the value of D due to the wounded situation the bacteria has been subjected to. Due to the presence of bacteriocin, they appear to be sensitive to heat treatment. Nisin functions synergistically with the process of heating, so it has the capability to be employed for use in order to regulate *L. monocytogenes* in pasteurized liquid egg products.

➤ *Application of Bacteriocins on Sausages*

The minimum infectious dose for listeriosis in humans still remains vague, so it is very necessary to control the presence of *L. monocytogenes* in food products, mostly low-Aw fermented foods like sausages, which can adequately support the growth of *L. monocytogenes*. Joerger (2003) investigated the Italian salami sausage for the antimicrobial potential of bacteriocin against *L. monocytogenes*. The lactic acid bacteria used were *Lactobacillus plantarum* MCS1 strain-producing bacteriocin, the mutant strain that cannot produce bacteriocin, and two commercially produced starter cultures. One of the commercial starter cultures has a combination of *Lactobacillus curvatus* and *Pediococcus pentosaceus*, while the other contained *Lactobacillus* sp that is unidentified. These sausages that were to be tested contained 2.5% NaCl, 250 mg/kg NO₂, and 0.3% sucrose. In addition, *L. monocytogenes* were inoculated from SSICA strains 38 and 150, as much as 103 to 104 CFU/g. The Italian salami sausage that lack lactic acid bacteria, the development *monocytogenes* was spotted after storing for about 7-14 days. In the ones that contain lactic acid bacteria, the number of *L. monocytogenes* was revealed to decrease in the process of the salami ripening, but some of the bacteria survived until the ripening process was completed. Only the sausage that was supplemented with *Lactobacillus plantarum* MCS1-producing bacteriocin totally eliminated

all *L. monocytogenes* cells in the sausage after completion of ripening.

➤ *Application of Bacteriocins to Vegetables*

Bacteriocins or lactic acid bacteria compounds have also been employed as antimicrobial agent against *L. monocytogenes* in plant products. Alvarez-Cisneros et al. (2011) revealed the ability of the bacteriocin released by

Enterococcus mundtii, mundticin, to inhibit and kill *L. monocytogenes* in minimally processed vegetable products. Bacteriocins can be added at the washing step or by coating procedures, and is discovered to inhibit the growth of *L. monocytogenes* on bean sprouts stored at modified atmospheric conditions (1.5% of O₂; 20% of CO₂; 78.5% of N₂) at a cold temperature of 8°C (Lopez et al., 2023; Martin et al., 2023).

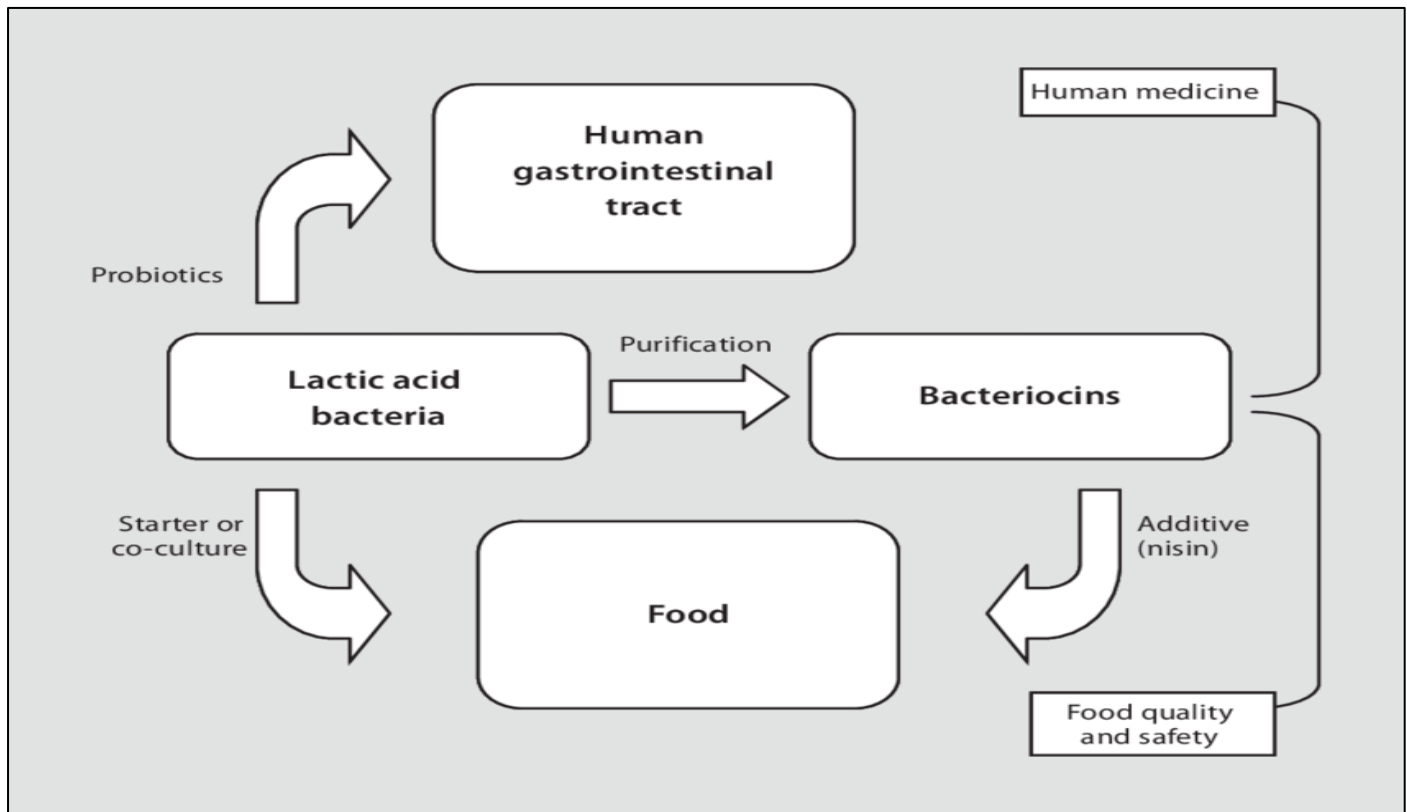


Fig 4 Overview of the Importance of the Application of Bacteriocin Production by Lactic acid Bacteria in food Quality and Safety and in Medicine, while Focusing their role as food Supplement and in the Gastrointestinal tract of Humans, Respectively (De Vuyst et al., 2004).

VII. LIMITATIONS OF USAGE OF BACTERIOCINS PRODUCERS

By applying bacteriocin producers when they are used as starter catalyst should be identified a key component for the process of biopreservation, that should be implemented by different authors to suggest that specific lactic acid bacteria can produce technological (for the conversion of natural products to a fermented finished product) characteristics, in combination to biopreservation properties, producing several bioactive compounds and adding to ensure the quality of the finished product (Favaro et al., 2017; Garca-Cono et al., 2019; Brown et al., 2017). Nonetheless, just few of them are usually employed in practical industry practices. The main challenge might be due to lack of collaboration between academia and industries. One of the disadvantages of applying lactic acid bacteria as raw cultures with bio-preservative characteristics can be attached to the fact some releasers of bacteriocin lack the ability to fulfil the requirements to meet the standard criteria. The standard regulatory governmental organizations such as WHO, EFSA, FDA adjusts and regulates the utilization of live and adjunct cultures in

order to ensure the god quality of food products both locally and globally. Different lactic acid bacteria produce bacteriocin (Cotter et al., 2005). However, not all LAB are safe for use. A perfect example is seen in species that belong to the genera *Streptococcus*, in which the GRAS standard is applied majorly for *Streptococcus thermophilus*. While the rest of the *Streptococcus* spp. are regarded as being unsafe, and some of them have been proven to be potentially toxic to human and animals and are associated to specific examples of cancers, associated with some treptococcus bovis strains (Corredoira et al., 2008; Boleji et al., 2011; Jans et al., 2013). A perfect example is those found in genera *Enterococcaceae*. Few species are known to be used for long periods as starter cultures, majorly in the Mediterranean region, and they have been used in the manufacture of several types of cheese products and salami (Todorov et al., 2022). Another issue is that several researches are not correctly organized and utilized, as a result of the obvious disagreement between optimal standard conditions that support the growth of the organisms, the release of bacteriocins, and the requirements that are necessary when undergoing model evaluation.

VIII. CONCLUSION

In the history of human, a key role has been played by the presence of food. Some religion, family and traditional practices will always be connected to food. From the onset, producing and preserving food products have been an ancient method, passing across the years. The bacteriocins that lactic acid bacteria release play key role in the food processing companies, mostly in fermented food products. Their potential can impede the growth of several microorganisms that cause spoilage and food-borne illness. The advantage of adding bacteriocins in food is not limited to prevent spoilage only but also to prolong the quality of food and suppress the growth or kill pathogenic bacteria. It is very crucial to be aware that the future use of bacteriocins should be implemented carefully to avoid, as seen in antibiotics, them from going into extinct and their utilization for the preventions and control of pathogens.

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