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Review Article

Bacteriocins: Properties and Potential Applications in Food and Pharmaceutical Industry

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Abstract

Infections caused by antibiotic-resistant pathogenic organisms are significantly rising worldwide. In response, numerous approaches, including the usage of bacteriocins, have lately been investigated to cure them. A class of antimicrobial peptides known as bacteriocins is produced by bacteria and is effective at controlling clinically significant susceptible and drug-resistant pathogens. Bacteriocins are a heterogeneous group of bioactive peptides, displaying antimicrobial activity against similar or closely related bacterial strains. To be able to alter and enhance their physicochemical characteristics, pharmacological effects, and biosafety, bacteriocins have been researched. These antimicrobial peptides have enormous promise as both food preservatives and next-generation antibiotics that can combat infections with multidrug resistance strains. Bacteriocins, not only focuses on being the perfect solution for food spoilage but also strives to become a potential drug candidate for replacing antibiotics in order to treat multiple drug resistance pathogens in the future. There is still a lot to learn about this class of peptide antibiotics, as evidenced by the rise in reports of novel bacteriocins with distinctive characteristics. In this review, we emphasize on properties and applications of bacteriocins including their biosafety. Bacteriocins vary in their biochemical properties, molecular weight, mechanism of action, spectrum of activity, and sequence of amino acids. They are produced both by Gram positive and negative bacteria. Many recent studies have also proven that bacteriocins aim to extend food preservation time, treat disease and cancer therapy, and maintain human health. Researchers and Scientists considered bacteriocins as an effective probiotic and declared confidently about its future-changing role in the field of pharma and food industry which is responsible for improvement in human health.

Keywords: Bacteriocins; Drug Resistance; Antimicrobials; Biosafety; Food Preservation

Introduction

Over the past decade, there has been a growing interest on bioactive peptides of ribosomal origin also known as ribosomally synthesized and post-translationally modified peptides (RiPPs) produced by a wide range of bacterial species. Perhaps one of the most popular members of the diverse group of RiPPs are the family bacteriocins [1]. Bacteriocins are antibacterial peptides synthesized by ribosomes produced by bacteria that inhibit the growth of similar or closely related bacterial strains. Bacteriocins are cationic molecules with hydrophobic or amphiphilic characteristics, which exhibit a narrow to broad inhibitory activity against closely related and non-related species [2]. Bacteriocins can inhibit the growth of bacteria, fungi and viruses [3]. Bacteriocins are also called "Designer Drugs" as they target specific bacterial pathogens [4]. Bacteriocins are mostly like antibiotics but the only major difference between antibiotics and bacteriocins are that bacteriocins restrict their activity to strains of species related to the producing species and particularly to strains of the same species, antibiotics on the other hand have a wider activity spectrum and even if their activity is restricted this does not show any preferential effect on closely related strains. A number of bacteriocins from a wide variety of bacteria are discovered and their various structures are described [5]. Ammonium sulfate precipitation, dialysis membrane, pH-mediated cell adsorption/desorption, solvent extraction, macroporous resin column, and chromatography are always used as purification methods for bacteriocins [6]. Bacteriocins discovery started in the year 1925 when gratia described the antagonism between strains of *E. coli* due to the production of colicin V, a heat stable substance that inhibited other strains of *E. coli* growth [7]. A huge variety of chemical structures, allows bacteriocins to affect various vital functions of a living cell (Transcription, translation, replication, and cell wall biosynthesis [5]. Bacteriocins are found in Gram-positive and Gram-negative bacteria. Bacteriocins are usually produced by lactic acid bacteria (LAB) or other bacteria such as *Bacillus* strain, *Staphylococcus* strains and *Escherichia coli* strains [2]. But not only LAB instead almost all bacterial species have the ability for the production of bacteriocins as a part of the defending molecules [8]. From some studies, LAB were found to have an ability to produce antimicrobial substances, especially which possess bacteriocins that are heat-stable, proteases-sensitive, ribosomally synthesized.

They do not adversely affect the gut microbiota to protect themself from other spoilage bacteria and pathogens. Three years later, Rogers and Whittier discovered an antimicrobial substance produced by LAB in the year 1928. Mattick and Hirsch succeeded in purifying this substance and termed it as "nisin" in the year 1947. Concerning the word 'bacteriocins', it was first invented by Jacob., *et al*. in 1953 [7]. Some investigators prefer separating 'true' bacteriocins such as colicins, first discovered by Gratia in 1925, and colicin-like bacteriocins, from so-called bacteriocin-like inhibitory substances (BLIS). There is a continuing interest in bacteriocins from lactic acid bacteria (LAB) [9]. Bacteriocin-producing bacteria could be extracted from various sources such as water, soil, animal intestines and food products (Unconventional sources) while the gastro-intestinal tract is considered as the conventional source of bacteriocin producers [2]. Bacteriocins are now the focus of increased attention due to i) consumer requirements for minimally processed foods free from chemical additives ii) their potential as natural alternatives to antibiotics due to increasing concerns about the emerging problem of antimicrobial resistance iii) as modulators of the human microbiome and, therefore, potential to address complex metabolic conditions such as diabetes and inflammatory bowel disease and iv) as bacteriocin-producing probiotic cultures for inclusion in animal feed to promote growth, improve animal health and/or reduce infection [10].

Classification

Bacteriocin can be categorized into mainly three classes:

Class I bacteriocins which are also considered as L antibiotics - these are made up of peptides that are small in size which is less than 5kDa (kilodalton). It includes a group of compounds-thiopeptides that have multiple biological activity (antibacterial, antiviral, antiparasitic, and immunosuppressive). Antibacterial thiopeptides interfere with protein synthesis by binding to the 50S ribosome subunit or elongation factors [5]. They contain specific post-translationally modified residues, which include lanthionine and β-methyllanthionine such as Nisin Z, A and Q, enterocin W and nukacin ISK-1 [11]. It is reported that some lantibiotics can act also inhibit the germination of *Bacillus* spores [12,13].

Class II bacteriocins which are also considered as Non-Lantibiotics - these are resistant to heat, non-modified and hydrophobic peptides with size less than 10 kDa. Usually, they can be grouped into two sub-classes, which are class IIa and class IIb. Class IIa bacteriocin, such as leucocin A and pediocin PA1, are widely used in food preservation due to its pediocin-like *Listeria*. Class IIb bacteriocins exert or improve the antimicrobial effects *via* synergistic activity of two complementary peptides, such as plantaricin A and enterocin X. They contain amphiphilic and hydrophobic regions, and are mostly cationic and active in the range of nanomolar to picomolar concentrations [11]. Class II bacteriocins are unmodified membrane-active peptides that act over a narrow spectrum of target bacteria. They bind a specific receptor protein on the membrane to form a pore, leading to membrane permeabilization and cell death. As these constructs display a lethal effect when they are heterologously expressed, they are called "suicide probes" [14].

Class III bacteriocins which are also considered as Heat labile proteins as these exert or improve the antimicrobial effects, and contain amphiphilic and hydrophobic regions. These are made of large proteins with sizes of more than 30 kDa and are likely to be layered or degraded when subjected to heat such as Lysostaphin, enterolysin A and helveticin J [11]. Further it is classified into three groups, which is - bacteriolysins, non-lytic bacteriocins, and tailocins. Bacteriolysins are large lytic polypeptides that target the peptidoglycan layer such as lysostaphin, zoocin A, millericin B, and enterolisin. Non-lytic bacteriocins are large non-lytic polypeptides, their mechanism of action is not based on cell wall lysis, it is believed that blocking the absorption of glucose and its inclusion in cellular macromolecules leads to carbohydrate starvation that kills the target cell such as helveticin J and casecin 80. The last one are tailocins which is multiprotein complex, with structure like a phage tail that target the lipopolysaccharides such as diffocin and monocin [15].

Classification	Features	Subcategories	Example
Class I bacteriocins (Lantibiotics)	Lanthionine or peptides containing beta-lanthionine	Type-A (linear molecules) Type-B (globular molecules)	Nisin, subtilin, epidermine Mersacidin
Class II bacteriocins (Non-Lantibiotics)	Heterogeneous class of small thermostable peptides	Subclass IIa (antilisterial pediocine bacteriocins type) Subclass IIb (composed of two peptides) Subclass of IIc (other bacteriocins)	Pediocin, enterocin, sakacin Plantaricin, lactacin F Lactococcin
Class III bacteriocins (Heat-liable Proteins)	Large thermostable peptides	-	Helveticin J. millericin B

Table 1: Classification of bacteriocins based on its features [4].

Mode of action of bacteriocin

Bacteria themselves produce more complex antibacterial agents, termed bacteriocins, which specialize in killing closely related bacterial strains [16]. Bacteriocins act by interacting and destroying cells with specific surface receptors. It is widely hypothesised that the interaction of bacteriocin with the target cell occurs in two stages. The first stage, which is probably reversible, corresponds to the physical adsorption of bacteriocin in the cell using a receptor. Removing the bacteriocin at this stage appears to leave the cell intact, since there is no permanent physiological damage. The second stage leads to irreversible pathological changes as a result of specific biochemical damage to the cell [5]. Bacteriocin induces cytoplasmic membrane permeability, inhibiting cell wall synthesis, interfering with metabolic pathways [2]. The mechanistic action of bacteriocin includes promotion of bactericidal and bacteriostatic effect with or without cell lysis [17]. They inhibit the growth of target organisms by various mechanisms. These can be subdivided into:

- Function on cell envelope - The mechanism of the antimicrobial action of bacteriocins is based on its ability to directly interact with the bacterial cell membrane that would cause its permeation resulting to the leakage of ions, dissipation of membrane potential, and eventually, cell death [1].
- Affects gene expression and protein production - Colicins are bacteriocins that inhibit protein synthesis at various stages by their explicit 16S rRNase activity. RNase domain of colicin E3 has been reported to bind to the A site in the 70S ribosome, triggering the cleavage of 16S rRNA. Bacteriocins, such as E3-E6 colicins and DF13 cloacin, demonstrate development of rRNase at 16S [18].

Specifically, Lanitbiotics inhibit by eliminating the synthesis of peptidoglycan present on the cell membrane. Other bacteriocins form pores. They can get inserted into membranes and form pores that lead to rapid cell death. Gram - bacteria control their target bacteria by interfering with DNA, RNA and protein metabolism. Some can bind to lipid-II, the main transporter of peptidoglycan subunits from the cytoplasm to the cell wall. Lipid II is also known to be the target of antibiotics of the glycopeptide family [19]. Non-lantibiotics bind to the pore-forming receptors (MAN-PTS) [4]. The main function of this four-component system (MAN PTS) is the uptake of mannose and glucose with their concurrent phosphorylation [20]. The mechanism of bacteriocins' synthesis can often be induced by stress conditions such as population increase and nutrient shortage, as well as can be affected by the type of carbon, nitrogen, and phosphate sources present in the media, or even by cation surfactants and other inhibitors [21].

Applications of bacteriocin

Recently, studies on bacteriocin or bacteriocin-like inhibitory substances (BLIS) have been focused on finding out an alternative

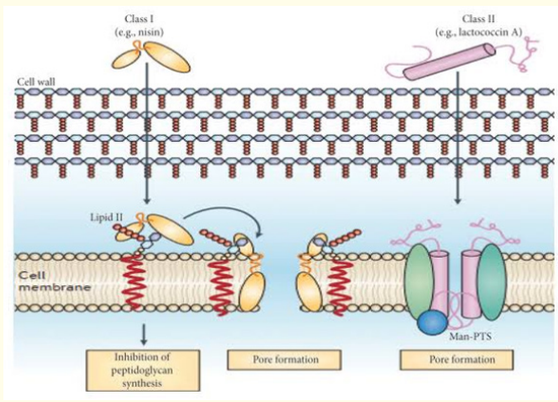

Figure 1: Mechanism of action of bacteriocins on bacteria [4].

therapeutic option for bacterial infections. Bacteriocin has been chosen as a potential drug candidate to replace chemicals and antibiotics in future due to its lower toxicity and proteinaceous nature. Living environments, which include soil, sea, river and air, are occupied with microorganisms. Some of them may cause food and beverage contamination, which leads to food and beverage spoilage. Food and beverage spoilage is always a concern in the food industry, as it may destroy the taste of the food and beverage, as well as cause some foodborne illnesses in human beings. For instance, *Staphylococcus aureus* can cause inflammation of the small intestine in humans after consuming food contaminated with enterotoxin produced by it. Chemical additives have been used widely to preserve food, but they can cause a lot of human health problems due to the toxicity of the chemical additives. This concern has led to the high demand of natural and chemical-free products used to preserve food in the market to avoid health problems [11]. Since 1965, one hundred and eight patents relating to manufacturers of Gram-positive bacteriocins have been filed, 57% of which are related bacteriocins derived from Lactococcus, Lactobacillus, Streptococcus, and Pediococcus strains. Bacteriocins are usually active against phylogenetically related bacteria, providing competitive advantage to their producers in the natural bacterial environment [22]. Surprisingly, patents for the production of heterologous bacteriocins have been presented mainly only in the last decade. Bacteriocins function in bacterial ecosystems and energetic costs associated with their production [23]. Although the main application of bacteriocins is in the food industry to combat spoilage and foodborne bacteria, in recent years the use of bacteriocin has been shifted to the diagnosis and treatment of cancer, as well as resistance to plant diseases and growth stimulation [5]. With the rise in antimicrobial resistance (AMR) and lack of antibiotic development, so we find our alternative option in Bacteriocin. Drug resistance has become a major threat due to the frequent use of commercial antibiotics and there is an urgent need to combat this problem. Hence, bacterocin has come into action with its remarkable properties that suggests that it could be used in various pharmaceutical and food industries [24]. Therefore, advances in the identification of bacteriocins and their characterization have prompted an interest in the

use of these molecules as either new food additives or therapeutic agents. Moreover, the efficacy of many bacteriocins with the potential to treat human and animal infections has been described [25].

The fact that bacteriocins cause much less collateral damage to the host microbiome makes them highly desirable therapeutic [26]. Bacteriocins exhibit antibacterial activity and a specific immunity mechanism toward strains closely related to the producer bacteria. Most bacteriocin-producing strains have an immunity mechanism involving an immunity protein (Bacteriocin operon). Bacteriocin encoding genes are organized into this operons located in the chromosome, plasmids, or other mobile genetic elements. In general, these operons are inducible and require secretion and extracellular accumulation of bacterial peptides for induction [27]. However, these immunity mechanisms vary from one bacteriocin to another [2].

Food preservation

Nowadays, Consumers are becoming more concerned about the consequences to health from food additives and are now demanding foods that are processed without any addition of chemical preservatives. One of the alternatives to satisfy this desire is bacteriocins [28]. The use of biopreservatives in the food industry has been an alternative to increasing the safety, quality, and replacement of chemical preservatives in food. Bacteriocins are biologically active, having bacteriostatic or bactericidal action, including pathogenic bacteria. Commercial bacteriocins permitted by the Food and Drug Administration (FDA) for use as biopreservatives are nisin and pediocin PA-1 [29]. Bacteriocins extensively investigated in dairy products, meat products, eggs, vegetables and fruits. The bacteriocins can be incorporated into the food by three ways,

- The use of a purified/semi purified bacteriocins preparation as an ingredient in a food
- By incorporating an ingredient previously fermented with a bacteriocins-producing strain.
- Using bacteriocins-producing culture to replace all or part of starter culture in fermented foods to produce bacteriocins *in situ*.

Direct incorporation of antimicrobials into food tends to impair their biological activity against microorganisms due to their diffusion through the food matrix [30]. Bacteriocins can also be used to improve food quality and sensory properties, for example, increasing the rate of proteolysis or in the prevention of gas blowing defects in cheese. Another application of bacteriocins is bioactive packaging, a process that can protect the food from external contaminations, which improves food safety and shelf life [4]. There are three conventional strategies for using bacteriocins in the food industry: pure bacteriocins, bacteriocin-containing fermentates, and bacteriocin-producing live cells [31]. Food pathogenic bacteria are present in both planktonic and biofilm forms as foodborne pathogens. In addition to being able to counteract with drug-resistant bacteria, bacteriocins may also be used as food and dairy products preservatives. According to studies, LAB-derived bacteriocins such as nisin, pediocin PA-1, pediocin, mersacidin, mutacin, and lacticin are mostly used in the food processing industry as preservatives that are capable of preventing the growth of *C. botulinum*, *E. faecalis*, VRE, *L. monocytogenes*, *S. aureus*, and other foodborne pathogens [2]. Based on the characterization, the wide inhibitory spectrum and mode of action determined so far, enterocin TJUQ1 also is a potential preservative for the food industry [32]. One interesting fact about bacteriocins is that the antimicrobial activity can be improved when combined with other barriers (e.g., chemical additives, high pressure, and heating treatments) to foodborne pathogens [33]

Nisin

Nisin is one of the bacteriocins that has been approved by the US Food and Drug Administration (FDA) and World Health Organization (WHO) to be applied in food factories. It was the first bacteriocin used in the food as the research advances novel bacteriocins introduced which are being used in food industry for the preservation of food [34]. The first identification of Nisin was done in 1928 from fermented milk cultures, and sold in England in 1953 as a biopreservative due to its ability to inhibit the growth of pathogens, which is beneficial to food preservation. Nisin is a bacteriocin with a molecular size of 3354 kDa, and is made up of 34 amino acids. It is produced by Gram-positive bacteria, including *Lactococcus* and *Streptococcus* species. Nisin belongs to class I (lantibiotic) bacteriocin, which contains five lanthionine rings. Nisin is found to exert inhibition activities against many groups of Gram-positive foodborne bacteria, such as *Bacillus cereus*, *Clostridium botulinum*, *L. monocytogenes* and *S. aureus*. Nisin plays a vital role in preventing food spoilage, as it can kill or inhibit many foodborne pathogens in a wide range of foods, either in liquid form or solid form. Nisins are reported to act by pore formation using lipid II as docking molecule, leading to increased membrane permeability of the targeted cell, and thereby its death [35]. Besides that, a previous study demonstrated that the prevention of milk and milk product spoilage can be done by adding nisin to the food matrix to inhibit the growth of *B. cereus*, *C. botulinum* and *Clostridium perfringens*. Besides that, nisin Z was found to exert inhibitory activity against methicillin-resistant *Staphylococcus aureus* (MRSA), a pathogen that might cause skin and lung infection in humans with MIC (Minimum Inhibitory Concentration) of 4.17 µg mL−1. Nisin is also used in meat and meat products to inhibits the growth of *C. botulinum* and *L. monocytogenes* [11]. However, the use of nisin as a food preservative is limited by its low production during fermentation [36].

Pediocin

Pediocin is a class II bacteriocin with a molecular weight of 2.7–17 kDa, which comprises a hydrophilic N-terminal and a hydrophobic C-terminal variable. It was first described in 1990. It is made of 44 non-posttranslationally modified peptides, which comprises aliphatic and aromatic amino acids. PLBs (Pediocin-like bacteriocins) can be synthesized by components of symbiotic mi-

croflora and participate in the maintenance of homeostasis in various compartments of the digestive tract and on the surface of epithelial tissues contacting the external environment [37]. Pediocins are sensitive to most protease enzymes such as papain, pepsin, and trypsin [38]. However pediocin, which is produced by *Pediococcus* strains, has high stability towards heat, a wide range of pH values, and some protease enzymes. Pediocin was found to have ability in the inhibition of pathogens that may cause food spoilage, such as *C. perfringens* and *L. monocytogenes*, by absorbing the amino acids at the phospholipid layer of the cytoplasmic membrane of the targeted cells. *L. monocytogenes* is a major concern for meat producers due to its ubiquitous nature and its survival capacity under adverse conditions [39]. Pediocin can be applied in food *via* two ways, either through the *in situ* method by inoculating the food matrix with *Pediococcus*, *Enterococcus* or *Lactobacillus* strains with the optimal control to produce pediocin for inhibiting the growth of pathogens in food, or the direct use of pediocin to the food matrix with optimal concentration. However, adding pediocin directly to food has some disadvantages, such as changes in its solubility and amphiphilic nature.

Pediocin PA-1 has been proved that it can help in preserving the fish fillets by inhibiting the growth of *L. monocytogenes*. For sous vide products, *Bacillus subtilis* and *B. licheniformis* can be inhibited by pediocin PA-1 to extend its shelf life. In the production of fermented soymilk, pediocin PA-1 from *E. faecium* NCIM 5423 or *Lactobacillus plantarum* Acr was claimed to exert the antimicrobial effect against *L. monocytogenes* to prolong the storage period. Besides that, pediocin 34 extracted from *Pediococcus pentosaceous* 34 was proved to have an inhibitory effect on *L. monocytogenes* in milk products and meat [11].

Producing strain	Types of bacteriocins	Food application	Targeted pathogens
L. lactis spp	Nisin	Cheddar cheese	*L. monocytogenes* and *S. aureus*
		Milk and milk products	*B. cereus, C. botulinum* and *C. perfringens*
		Dairy, culinary, bakery products and beverages	*L. monocytogenes, B.cereus* and *C. botulinum*
		Meat and sausages	*C. botulinum* and *L. monocytogenes*
L. lactis MG1614	Enterocin A	Cottage chees	*L. monocytogenes*
E. faecium CTC492 and *L. sakei* CTC494	Enterocin A and B, and sakacin K	Cooked pork	*L. sakei* CTC746
E. faecium WHE 81	Enterocins A and B	Munster Cheese	*L. monocytogenes*
E. faecium CTC492	Enterocins A and B	Dry fermented sausages	*L. innocua*
		Cooked ham	*L. monocytogenes*
		Cooked ham blended with distilled water	*L. monocytogenes* and *L. sakei*
E. faecium F58	Enterocins L50A and B9	Goat's milk and goat milk's cheese	*L. monocytogenes*
E. casseliflavus IM 416K1	Enterocin 416K1	Italian sausages and cottage cheese	*L. monocytogenes*
E. faecalis A-48-32	Enterocin AS-48	Non-fat hard cheese	*B. cereus*
		Skimmed milk and non-fat unripened soft cheese	*S. aureus*
		Infant rice-based food	*B. cereus*
		Fruit juice	*A. acidoterrestris*
		Apple cider	*B. licheniformis*
		Vegetable soups and puree	*B. cereus, B. macroides* and *Paenibacillus* spp.
		Vegetable sauces	*S.aureus*
		Cooked ham	*L. monocytogenes*
		Canned fruits and vegetable foods	*B. coagulans*

E. faecium CCM 4231, *E. faecium,* RZS C13 and *L. sakei* CTC 494 *E. faecalis* BFE 1071 *E. faecium* CRL35 *E. faecalis* EJ97 *E. faecalis* N1-33 *E. faecium* L50 *E. faecium* CCM 4231	Enterocin CCM 4231, enterocin 13 and sakacin K Enterocin 1071A and B Enterocin CRL 35 Enterocin EJ97 Enterocin MR-10A Enterocin L50A and B Enterocin CCM 4231	Spanish style dry fermented sausages Fish spread Goat cheese Vegetable (Zucchini) puree Custard cream Alcoholic and non-alcoholic beer Skimmed milk and yoghurt Bryndza Saint-Paulin cheese Dry fermented Hornad salami	*L. monocytogenes* and *L. innocua* *L. innocua, S. epidermis* and *P. vulgaris* *L. monocytogenes* *B. macroides* and *B. macroccanus* *B. cereus* *L. brevis* and *P. damnosus* *S. aureus* and *L. monocytogenes* *L. innocua, E. coli and S. aureus* *L. monocytogenes* *L. monocytogenes*
P. acidilactici MCH14	Pediocin PA-1	Dried sausages adn fermented meat products Salad dressings Fresh beef, vacuum packed beef, cottage cheese, ice cream mix Fish fillets, chicken meat Sous vide products	*L. monocytogenes and C. perfringens* *Lactobacillus biofermentans* *Ln. mesenteroides* *L. monocytogenes* *B. subtilis, B. licheniformis*
E. faecium NCIM 5423 and *L. plantarum* Acr2	Pediocin PA-1	Fermented soymilk products	*L. monocytogenes*
P. pentosaceous 34	Pediocin 34	Milk products and meat	*L. monocytogenes*
Ln. gelidum UAL187	Leucocin A	Milk product, fresh meat and sausage Meat	*L. monocytogenes* *C. divergens* UAL9
Ln. mensenteroides K7	Leucocin K7	Milk	*L. monocytogenes*
L. lactis spp. (*lactis* BZ)	Lactococcin BZ	Both skim and full-fat milk	*L. monocytogenes*

Table 2: Application of Bacteriocin from Lactic acid bacteria (LAB) in food preservation [11].

Pharmaceutical treatment

In addition to adverse effects of some antibiotics, the emergence of antibiotic-resistant, MDR (multidrug-resistant), and XDR (extensively drug-resistant) strains has recently become a major concern. Multidrug resistance and toxicity associated with antimicrobial agents among pathogenic bacteria leading to a surge in morbidity and mortality in humans need bold proclamation in the area of research and development of new biological agents [40]. It is estimated that by 2060, at least 20 new antibiotics are needed to overcome the problem of antibiotic resistance, while the design of new antibiotics is a time-consuming and slow procedure. Therefore, it is necessary to develop new treatment strategies that could eliminate antibiotic-resistant microorganisms. One of the strategies is to use antimicrobial peptides to achieve this goal. Bacteriocin is considered as a suitable antimicrobial peptide due to its thermal stability and high efficacy with nano-molecular size. In addition to the immune system, bacteriocins are able to affect other bacteria through competition in colonisation. Bacteriocins have been shown to possess advantages over antibiotics. These antimicrobial peptides are considered to provide more protection with no side effects compared to antibiotics. According to the International Nosocomial Infection Control Consortium 2010, gram-positive (*Staphylococcus* spp.) and gram-negative (*Escherichia coli*) bacteria have been shown to acquire antibiotic resistance [41]. A study examining the differences between bacteriocins and antibiotics found that oral administration of pediocin PA-1 had no side effects on the gastrointestinal tract, while under the same conditions, the use of antibiotics such as penicillin and tetracycline exhibited different results [2]. Many research studies have been done to investigate bacteriocin, which can be used to solve human health's

problems, such as urinary tract infection, skin infection, diarrhoea, dental caries, lung infection, bloodstream infection, mastitis, respiratory tract infection and cancer. The bacteriocins applied in the infectious disease treatment for humans, comprises nisin, lacticin, salivaricin, subtilosin, mersacidin, enterocin, gallidermin, epidermin and fermencin [11].

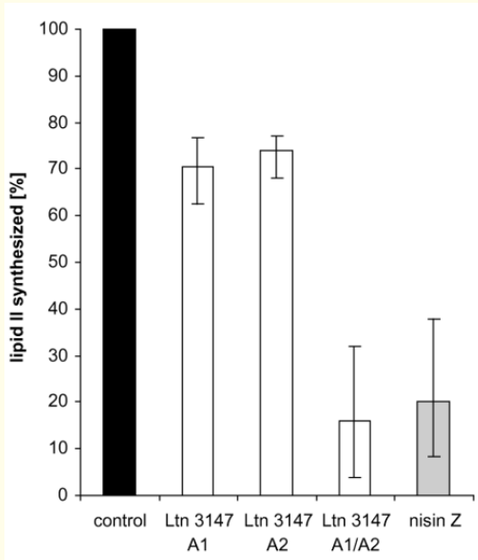

Figure 2: Inhibition of the *in vitro* lipid II synthesis by the individual lacticin A1/A2 and nisin Z (grey column) [42].

Salivaricin

Salivaricin is a bacteriocin that is produced by *Streptococcus salivarius* and belongs to class II lantibiotics. Many research studies proved that salivaricin has contributed to the health sustainability in humans. Salivaricin was shown that it not only can be used to maintain oral health, but also can be used in the treatment or prevention of skin or lung infections. Furthermore, salivaricin B was also shown to have inhibition activity against *Corynebacterium* spp GH17.112 *Corynebacterium* spp was found in triggering diseases, such as pharyngitis, endocarditis, gastrointestinal tract infection and skin infection.113 The MIC for the inhibition of growth of *Corynebacterium* spp GH17 by salivaricin B was 2690 nM.112 Besides that, salivaricin D from *S. salivarius* 5M6c was proved to exert inhibitory ability against *Clostridium bifermentans*. Salivaricin D was also demonstrated as an antimicrobial agent in the inhibition of the growth of *S. pneumoniae* D39, TIGR4 and R6 for the prevention of pneumonia [11].

Subtilosin

Subtilosin is a bacteriocin that can be produced by *Bacillus* strains. *Bacillus* spp is an aerobic, Gram-positive bacteria with rod shape, which can be usually found in soil, water or natural flora in the intestines. Subtilosin A is a bacteriocin with a molecular weight of 3398.9, which consists of 32 usual amino acids and some non-amino acid residues. Subtilosin A, produced by *B. subtilis* 168, was claimed to exert antimicrobial effects against many Gram-positive and Gram-negative pathogens. Over the last seven decades, applications using members of the *Bacillus subtilis* group have emerged in both food processes and crop protection industries [43]. Antimicrobial substances produced by Bacillus species have more diverse characteristics and a broader range of activities than those of substances produced by lactic acid bacteria [44]. Subtilosin A was proved to have an inhibitory effect against *E. faecalis*. *E. faecalis* is a Gram-positive bacterium that may cause infections in humans, especially infection of the urinary tract. For instance, bacteriocin obtained from *E. faecalis* KT11 had a broad antimicrobial spectrum, and inhibited foodborne pathogens and vancomycin- and/or methicillin-resistant bacteria [45]. Besides that, subtilosin A was proved to be used as an antimicrobial agent in the treatment of shigellosis. Shigellosis is the infection that may be caused by *Shigella sonnei*, leading to diarrhoea, fever and stomach cramps. Although being a causative agent, *Shigella* along with other gram negative bacteria like *Salmonella*, Vibrio, *Listeria*, and *E. coli*, have been utilized as test organisms to examine the antagonistic action of recently identified antimicrobial peptides [46]. In addition, subtilosin A was proven to exert inhibitory effect against *S. gordonii* Challis, which was the pathogen found in the oral cavity to cause dental plaques [11].

Mersacidin

Mersacidin bacteriocin was produced from the *Bacillus* species (strain-HIL-Y85), it is considered as a lantibiotic type B class, which consists of 20 amino acids and a lanthionine group in its structure [47]. It is proved to have inhibition activities against Gram-positive pathogens. *S. aureus* is a Gram-positive bacterium with rod shape, which may cause infectious diseases in humans, such as abscesses, furuncle, bloodstream infection and pneumonia. Mersacidin produced by the *Bacillus* sp. strain HIL Y-85, 54728 was claimed to exert antimicrobial effects on a few *S. aureus* strains. Furthermore, mersacidin was demonstrated to have inhibition activity against *Micrococcus luteus*. *M. luteus* is a Gram-positive and non-motile bacterium reported as an opportunistic pathogen that may cause pneumonia and meningitis. In addition, mersacidin was proved to have an inhibitory effect on the *E. faecium* and *E. faecalis* strains, which might cause infections in the abdomen, skin, bloodstream and urinary tract [11].

Epidermin

Epidermin is a tetracyclic bacteriocin that belongs to type A lantibiotic. It comprises 21 amino acids with *meso*-lanthionine, 3-methyl lanthionine, and *S*-(2-aminovinyl)-d-cysteine in the structure. Epidermin produced by *Staphylococcus epidermidis* exhibits antimicrobial activities against a wide range of Gram-positive bacteria. Epidermin was claimed to exhibit inhibition activities against pathogens *S. aureus* SG 511 and E 88, which might cause respiratory tract, skin and surgical site infections. S. aureus is responsible for various infections such as skin abscesses, wound infections, deep tissue abscesses, osteomyelitis, endocarditis, toxic shock syndrome, sepsis, and bacteremia [48]. Besides that, epidermin was also applied in the treatment of infection caused by *S. epidermidis*.

S. epidermidis is a Gram-positive and facultative anaerobe that can be usually found in normal human flora. In addition, epidermin was proved to have inhibition activity against *Corynebacterium xerosis*. *C. xerosis* is a Gram-positive aerobe that can be found in the normal flora of the nasopharynx and skin, and may cause endocarditis, cerebrospinal fluid shunt infection in an infant, mediastinitis and spontaneous bacterial peritonitis. *Peptostreptococcus anaerobicus* is a Gram-positive and non-spore forming anaerobe that may lead to infection of the brain, liver, breast, and lung abscesses in humans. It was claimed to be inhibited by epidermin with MIC of 0.25 μg mL−1 [11]. In Table 3 down below lists the most famous species, *Lactococcus lactis* shows a good technological property (acidifying activity, proteolytic ability, and diacetyl production, autolytic activity) and high antibacterial activity [49-52].

Producing Strain	Types of Bacteriocin	Application In Medicine	Targeted Pathogens or Cells
L. lactis spp.	Nisin	Skin infections	MRSA
	Nisin Z	Lung infections	*P. aeruginosa*
	Nisin A	Stomach ulcers	*H. pylori*
		Mucosal and bloodstream infections	*C. albicans*
L. lactis spp. *lactis* DPC3147	Nisin A (combined with doxorubicin)	Cancer	Reduce HNSCC tumorigenesis by inducing preferntial apoptosis
	Nisin F	Cancer	Reduce tumour in skin carcinogenesis
	Nisin A and Z	Respiratory tract infection	*S. aureus*
	Lacticin 3147	Diarrhoea and inflammation of colon	*C. difficile*
		Skin and surgical site and prosthetic joint infections	MRSA and *C. acnes*
		Dental carriers	*S. mutans*
S. salivarius K12	Salivaricin A2	Pneumonia, sinus infection, ear infection, bacteremia and meningitis	*S. pneumoniae*
		Scarlet fever, rheumatic fever, pharyngitis, tonsillitis, cellulitis, erysipelas and necrotizing fasciitis	*S. pyogenes*
	Salivaricin B	Pharyngitis, endocarditis, gastrointestinal tract infection and skin infection	*Corynebacterium* spp.
S. salivarius 5M6c	Salivaricin D	Empyema and pneumonia	*C. bifermentans*
		Infections in immunocompromised humans	*Ln. lactis*
		Pneumonia, sinus infection, ear infection, bacteremia and meningtis	*S. pneumoniae*
		Scarlet fever, pharyngitis, tonsillitis, cellylitis, erysipelas and necrotizing fasciitis	*S. pyogenes*
B. subtilis 168	Subtilosin A	Urinary tract infection	*E. faecalis*
		Periodontitis and tooth loss	*P. gingivalis*
		Methylmalonic aciduria in immunocompromised patients	*K. rhizophila*
		Shigellosis	*S. sonnei*
		Dental plaque formation	*S. gordonii*

Bacillus spp. strain HIL Y-85, 54728	Mersacidin	Abscesses, furuncle, bloodstream infection and pneumonia Pneumonia and meningitis Infections of abdomen, skin, bloodstream and urinary tract	MRSA *M. luteus* *E. faecium* and *E. faecalis*
L. lactis MG1614	Enterocin A	Listeriosis Infection of bladder	*L. monocytogenes* *E. coli*
E. hirae LD3	Enterocin LD3	Pneumonia and meningitis	*M. luteus*
E. faecalis	Enterocin AS-48	Diarrhoea and infections of respiration tract and wounds	*B. cereus*
E. hirae 20C	Enterocin E20C	Gastroenteritis, bacteremia adn enteric fever	*S. enterica*
S. epidermidis	Epidermin	Endocarditis, cerebrospinal fluid shunt infection in an infant, mediastintis and spontaneous bacterial peritonitis Pneumonia and meningitis Infection of brain, liver, breast and lung abscesses Acnes Infections of urinary tract, wound and soft tissue	*C. xerosis* *M. luteus* *P. anaerobicus* *P. acnes* *S. faecalis*

Table 3: Application of Bacteriocin from Lactic acid bacteria (LAB) in infectious disease treatment for humans [11].

Future Perspectives

The strengthening of the view of bacteriocins being versatile antimicrobials with considerable potential for use as bio preservatives, antibiotic alternatives, health-promoting gut modulators and animal growth promoters is its significant characteristic. It is expected that for industrial applications of bacteriocins in the near future those and other natural bacteriocins-producing strains are the most suitable. It's possible that advancements in bacteriocins could lead to improved antimicrobial actions and bridge the gap between *in-vitro* (Study performed "outside the living"-test tubes, labs) and *in-vivo* (study performed "within the living" organisms) applications in the coming years, potentially contributing to the improvement in medical field. The combined use of *in vitro* and *in silico* plays a vital role in carrying a broader investigation of the bacteriocinogenic potential of the athlete gut which needs to be yet developed. However, there are still less bacteriocins to be marketed and approved for use in food preservation and treatment for infectious diseases by the FDA and WHO, as compared to antibiotics. This may be due to the lack of clinical tests done for bacteriocins to be used in humans or animals. In the future, more research studies should be done to completely characterize potent bacteriocins and clinically test them. However, further studies and a continuous monitorization effort are necessary for the safe application of LAB in animal food products and in the treatment of pathogenic microorganisms, providing a thorough assessment of the possible risks associated with the dissemination of antimicrobial resistance genes and, thereby, protecting public health. Despite their advantages as alternative therapeutics over existing strategies, several limitations of bacteriocins, such as the high cost of isolation and purification, narrow spectrum of activity, low stability and solubility, and easy enzymatic degradation, need to be improved.

Conclusion

Several studies are carried out to determine different bacteriocin-producing strains with a broad spectrum of antibacterial action. However, the application of bacteriocins in foods is limited due to the low physicochemical stability of the molecule. From previous studies, there are pros and cons of each bacteriocin species based on their expertise. However, especially during the last decade, a number of reports have appeared about unanticipated extensions to the generally rather narrow anti-bacterial activity spectrum of some of the LAB bacteriocins and novel applications have been proposed for bacteriocins ranging from controlling the growth of an increasingly-heterogeneous variety of pathogens, including Gram-negative multidrug resistant bacteria, viruses, yeasts, and in particular, difficult to control *Mycobacterium* spp, to their potential application as anticancer agents. The bacteriocins are produced as a competitive strategy, which acts against closely related bacteria. This might be a reason the good inhibitory activity of bacteriocin was observed against gram-positive bacteria than gram-negative ones. Recently, the investment in bacteriocin research has shown a clear upward trend in response to the potential applications of

these antimicrobial peptides in the field of food, livestock and medicines. The patent development for bacteriocins has been increasing since the last decades. According to the present study results, the correct selection of bacteriocin-producing strains suitable for use in the food industry is very important. Thus, this emphasises the necessity of investigating LAB bacteriocins to prove their beneficial and nutritional properties as well as inhibitory activity against the growth of functional pathogens, as they are potentially crucial for the final preservation of functional foods and for medicinal applications. Although bacteriocins have offered a way of solving the problems of food infections and food spoilage in the food industry, to date, only a few commercially bacteriocins are available. Thus, to further improve the bacteriocin arsenal against these unwanted spoilage microorganisms and pathogens, it is important to promote the study on the mode of antimicrobial action and their biosynthetic mechanisms of known bacteriocins to look for more new bacteriocins with promising properties.

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