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Bacteriocins as promising antimicrobial peptides, definition, classification, and their potential applications in cheeses

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Abstract

There is increased importance to finding alternative solutions to antibiotic resistance which require more research, bacteriocins are promising antimicrobial peptides with inhibitory and bactericidal activities that might be one of these solutions. Bacteriocins are small antimicrobial peptides synthesized by bacterial ribosomes, active against the bacterial pathogen, multidrug-resistant bacteria, and cancer therapy. Lactic acid bacteria (LAB) are one of the most used bacteria to produce bacteriocins and dairy products (i.e. cheeses) consider rich sources of LAB isolates. *Enterococcus faecalis*, *Lactobacillus fermentum*, *L. plantarum*, *L. helveticus*, *L. pentosus*, *L. paracasei subsp. paracase*i, *L. rhamnosus I*, and *L. delbrueckii subsp. lactis* are strong strains in bacteriocins production. Several applications were applied to control bacterial pathogens spread in cheeses, one of them is using bacteriocins and bacteriocins-like inhibitory substances (BLIS). To reduce foodborne pathogens and spoilage bacteria in cheese, bacteriocins can be applied in several means such as inoculating cheese with bacteriocin-producer strain and adding purified or semi-purified bacteriocin as a food additive. This review is focused on bacteriocins and BLIS classification, mechanism, and applications in dairy products i.e. cheeses.

Keywords: bacteriocins; antimicrobials; *Lactic acid bacteria*; foodborne pathogens; cheese.

Practical Application: Bacteriocins obtained from LAB have strong antimicrobial activity against bacterial pathogens, which makes them a promising alternative to antibiotics.

1 Introduction

The antimicrobial activity of bacteriocins has promising potential in inhibiting and killing bacterial pathogens (Pato et al., 2022a). Bacteriocins are antimicrobial peptides synthesized by bacterial ribosomes, with bacteriostatic or bactericidal effects against pathogens (Simons et al., 2020). It could be produced by gram-positive and gram-negative bacteria. They are mostly used in the food industry as natural bio preservatives instead of chemical preservatives to protect products from spoilage and pathogenic bacteria (Todorov et al., 2019; Pato et al., 2022b; Barboza et al., 2022). Bacteriocins-like inhibitory substances (BLIS) are also ribosomally synthesized peptides that possess abilities like bacteriocin but have not been yet characterized for their amino acid sequence (Caulier et al., 2019). Nisin is the first approved bacteriocin used as a food preservative (Khelissa et al., 2021). Lactic acid bacteria (LAB) are one of the most used bacteria to produce bacteriocins and BLIS (Tankoano et al., 2019; Jawan et al., 2021; Tang et al., 2022).

Foodborne diseases are a real threat to consumer health, especially in sensitive highly perishable dairy products such as cheeses (Cheng et al., 2022). Since cheeses are rich in various nutrient values which provide an optimum environment for pathogenic bacteria that escape during the manufacturing process or survive in retail stores (Kaur & Kaur, 2021). As a result of the widespread of antibiotic-resistant bacteria due to the overuse of antibiotics, there is a requirement for effective therapeutic approaches to control the spread of resistance (Soltani et al., 2021).

LAB and their bacteriocins are considered safe and useful food additives to control the growth of pathogens and spoilage microorganisms in foods (Mokoena et al., 2021). Nisin, Pediocin PA-1, and Micocin are the only FDA-approved bacteriocins to use as food preservatives since they inhibit the growth of many gram-positive bacteria and food-borne pathogens (Naskar & Kim, 2021). Bacteriocin of *lactobacillus plantarum* isolated from a Tulum cheese reduced the growth of *Staphylococcus aureus* during production and ripening of white-brined cheeses (Taban et al., 2019). *L. Plantarum* ST71KS isolated from Bulgarian goat milk feta cheese was able to produce class IIa bacteriocin that had a bactericidal effect on *Listeria monocytogenes* (Martinez et al., 2013). Pediocin PA-1 gene was detected in *pediococcus pentosaceus* isolated from ripened Minas cheese, an artisanal cheese made with raw cow's milk (Gutiérrez-Cortés et al., 2018). Several applications were applied to control bacterial pathogens spread in cheeses, one of them is using bacteriocins and bacteriocins-like inhibitory substances (BLIS). This review is focus on bacteriocins and BLIS classification, mechanism, and applications in dairy products i.e. cheeses.

2 Bacteriocin and Bacteriocins Like Inhibitory Substances (BLIS)

Bacteriocin is a small proteinaceous compound with low-molecular-mass, consists of 30 to 60 amino acids synthesized

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by bacterial ribosomes, heat-stable at 100 °C for 10 min, and extracellularly excreted to inhibit or kill other bacterial strains (Mokoena, 2017; Raval et al., 2020; Moradi et al., 2021). Normally not termed antibiotics to avoid confusion with therapeutic antibiotics (Sen & Ray, 2019) rapidly digested by proteases in the human digestive tract which is the difference between them and most therapeutic antibiotics (Tkhruni et al., 2020).

Bacteriocins have a narrow or wide spectrum via inhibiting taxonomically close bacteria, or a wide variety of bacteria (Silva et al., 2018). Bacteriocins have a different spectrum of activity, mode of action, molecular weight (MW), genetic origin, and biochemical properties (Darbandi et al., 2022) BLIS are antimicrobial peptides produced by LAB that possess bacteriocin ability, but that have not yet been characterized for their amino acid sequence (El-Gendy et al., 2021) Recently, BLIS gained interest due to their potential use as natural antimicrobial peptides that inhibit the growth of pathogenic and contaminant bacteria in several applications such as food, clinical, veterinary, and others (Agriopoulou et al., 2020; Hefzy et al., 2021; Hu et al., 2018).

BLIS activity can also be measured by various methods such as agar well diffusion assay, spot-on-lawn assay, turbidimetric assay, ELISA, radiometry, conductance measurements, and bioassays based on self-induction skills (Sidek et al., 2018; Zou et al., 2018; Arakawa, 2019). The most widely used assay is the determination method that exhibits the growth inhibition potential of a bacteriocin (Sidek et al., 2018). The physical and chemical properties of BLIS are important, especially in the food industry because of the complexity of food processing (Yi et al., 2022).

3 Bacteriocins of Lactic Acid Bacteria (LAB)

LAB are non-spore-forming bacteria, gram-positive, anaerobic but aerotolerant rods or cocci, catalase-negative, and fastidious bacteria, with a high tolerance for low pH, ferment carbohydrates to produce energy and lactic acid as the primary product of fermentation (Gupta et al., 2018; Miranda et al., 2021; Wang et al., 2021). Moreover, it is produced antimicrobial substances such as bacteriocins, BLIS, hydrogen peroxide, organic acids, diacetyl, acetoin, and antifungal peptides (Egan et al., 2016). LAB produce a large number of bacteriocins from strains like *Lactobacillus*, *Pediococcus*, *Lactococcus*, and *Enterococcus* (Trejo-González et al., 2021).

LAB bacteriocins are often active at various pH values, resistant to high temperatures, and active against plenty of food pathogenic and spoilage bacteria (Ahmad et al., 2017). Furthermore, bacteriocins produced by LAB are sensitive to digestive enzymes such as pancreatin complex, trypsin, and chymotrypsin, and for this reason, has no impact negatively on the gut microbiota (Egan et al., 2016) *Lactobacillus* and their bacteriocins have been used in food preservation, and to control human pathogens (Mokoena et al., 2021).

4 Bacteriocins classification

Based on the biosynthesis mechanism and biological activity of bacteriocins, it's classified into three major classes: Class I – small post-translationally modified peptides with molecular mass < 5 kDa, class II – unmodified peptides with molecular mass 6-10 kDa and including or not stabilizing disulfide bridges and class III – larger peptides [> 10 kDa, thermo-labile; Negash & Tsehai (2020); Zimina et al. (2020)].

Class I Bacteriocins/Lantibiotics are heat-stable small peptides (< 5 kDa) and are subdivided into two types based on charge difference (Negash & Tsehai, 2020). It is highly posttranslational modified and contains characteristic polycyclic thioether amino acids such as lanthionine, methyl-lanthionine (Veettil & Chitra, 2022), and unsaturated amino acids such as dehydroalanine and 2-amino isobutyric acid (Roy et al., 2018). Type A-lantibiotics are flexible screw-shaped molecules with a positive charge of 2–4 kDa. Effect on the cell membrane by forming pores which lead to depolarization of the target species cytoplasmic membranes such as nisin and lacticin 3147 (Negash & Tsehai, 2020). Type B-lantibiotics are globular peptides 2-3 kDa, without net charge or negative charge, that interfere with cellular enzymatic reactions of sensitive bacteria cell wall synthesis, such as Mersacidin (Moravej et al., 2018)

Class II Bacteriocins are small heat-stable peptides (< 10 kDa) with amphiphilic helical structures that allow them to insert into the membrane of the target cell which leads to cell depolarization and death (Negash & Tsehai, 2020). In addition, it is non-lanthionine-containing peptides that are not modified after translation beyond the elimination of a leader peptide and the formation of a conserved N-terminal disulfide bridge (Kozic et al., 2018). Subclass IIa peptides are monomers and have an N-terminal consensus sequence Tyr-Gly-Asn-Gly-Val-Xaa-Cys. They are active against *L. monocytogenes* such as pediocin PA-1 and sakacin A. Subclass IIb with two-component bacteriocins in which two distinct peptides act synergistically to generate an antimicrobial effect such as lactation F and lactococci G (Negash & Tsehai, 2020). Subclass IIc has circular bacteriocins carrying two transmembrane segments that facilitate pore formation in the target cell such as gassericin A, circularin A, and carnocyclin A (Yaghoubi et al., 2020).

Class III Bacteriocins are peptides with high molecular weight (> 30 kDa), and heat-labile proteins (Yaghoubi et al., 2020). In addition, it includes three groups 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 A. 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 (Zimina et al., 2020).

5 Bacteriocins: mechanisms of action

Bacteriocins inhibit the growth of target organisms in different mechanisms. There is a mechanism that can function primarily on the cell envelope and other mechanisms are primarily active in the cell by affecting gene expression and protein production

(Negash & Tsehai, 2020). Bacteriocins have bactericidal effects that may be accompanied with or without cell lysis (Qiao et al., 2021). It produced from LAB mostly inhibits gram-positive bacteria and exerts its antibacterial effect by targeting the cell envelope-associated mechanisms (Rahmeh et al., 2020). Lantibiotics and some class II of bacteriocins target lipid II and eliminate the synthesis of peptidoglycan (Negash & Tsehai, 2020).

Other bacteriocins use lipid II as a docking molecule to start pore formation which resulted in variation of the cytoplasm membrane potential and finally, cell death (Du et al., 2018). Nisin, the most studied bacteriocin is capable of both mechanisms (Kumariya et al., 2019). Lactococcin A is a class II bacteriocin that targets the cell by binding to the pore-forming receptor mannose phosphotransferase system [Man-PTS; Daba et al. (2018)].

Some bacteriocins show antimicrobial activity by their enzymatic activities, such as colicin E2 showing DNase activity, colicin E3 showing RNase activity, and megalin A-216 showing phospholipase activity against the target organism (Negash & Tsehai, 2020). Class II of peptides due to their structure can be inserted into the membrane of the target cell, causing depolarization and death. On the other hand, class III bacteriocins directly function on the cell wall of gram-positive resulting in cell death-like lysostaphin (Baindara et al., 2018; Negash & Tsehai, 2020).

The bactericidal mode of action of recombinant bacteriocin BMP32r against gram-positive and gram-negative bacteria was similar with slightly different. Cell damage was observed such as cell membrane disruption, intracellular material outflow, and even cell lysis in *Escherichia coli* after being treated with BMP32r (Qiao et al., 2020). Several bacteriocins inhibit gram-negative bacteria by interfering with their nucleic acids, and protein metabolism. For example, MccJ25 inhibits RNA polymerase, microcin B17 (MccB17) inhibits DNA gyrase, and MccC7-C51 inhibits aspartyl-tRNA synthetase. In addition, MccE492 as an exception works by pore formation (Cotter et al., 2013).

6 Application of bacteriocins

Bacteriocins are promising antimicrobial peptides with potential applications in different sectors such as food, clinical health, and others.

6.1 Food industry

Bacteriocins got the attention as natural antimicrobial agents to use in food as preservatives instead of chemical preservatives (Gokoglu, 2019). Usually, their application includes the use of bacteriocin-producer strain as inoculation of food, adding purified or semi-purified bacteriocin as a food additive, and using a previously fermented product with a bacteriocin-producing strain during food processing as an ingredient (Barcenilla et al., 2022).

Nisin, Pediocin PA-1, and Micocin are the only FDA-approved bacteriocins to use as food preservatives, by inhibiting the growth of many gram-positive bacteria and food-borne pathogens such as *L. monocytogenes* (Naskar & Kim, 2021). Applied mostly in dairy products and canned food, especially in processed cheese protect the product from heat-resistant spore-forming bacteria such as Bacillus and Clostridium (Anumudu et al., 2021).

Bacteriocins are colorless, odorless, and tasteless so, it is a perfect solution for food products without changing their properties (Negash & Tsehai, 2020). Moreover, they are stable at high temperatures, low pH, and a wide range of salt concentrations (Yang et al., 2018; Costa et al., 2019).

There are several advantages of using bacteriocins as food preservatives included increase food shelf life, reducing the transmission of risky foodborne pathogens via food, offering more protection during thermal conditions, lowering the economic losses due to food spoilage and outbreaks, and applying less severe treatment to food during processing to keep nutrients values and properties of food product without change (Reinseth et al., 2020).

Despite these advantages, we should keep in mind the addition of specific bacteriocins to food products could be limited by a narrow, limited spectrum of activity (Reinseth et al., 2020) or to the hydrophobic nature of used bacteriocin which may lead to the separation of organic fat within the food matrix (Khelissa et al., 2021). In addition, uneven distribution or poor solubility of bacteriocin may affect the antimicrobial activity of these molecules. Using bacteriocins in combination with other preservation methods may increase their antibacterial activity (Soltani et al., 2021).

Due to the interaction with food components or inactivation by proteolytic enzymes, nisin loses its activity easily, and to overcome this problem encapsulation of nisin Z in liposomes improves the inhibitory action and stability in the cheddar cheese matrix (Kaur & Kaur, 2021). Also, encapsulation of nisin with alginate/resistant starch increased its efficiency in cheddar cheese, *Clostridium tyrobutyricum* count was reduced after one week and inhibited completely after four weeks (Hassan et al., 2020).

7 Bacteriocins produced by LAB isolated from cheeses

Bacteriocin production can be obtained by any type of bacteria. Table 1 shows examples of several bacteriocins produced from LAB isolated from cheeses and their activity against foodborne pathogens. Five strains of LAB i.e. *Lactococcus lactis* IMAU32258, *L. garvieae* JB2826472, *Enterococcus durans* FMA8, *E. faecium* L3-23, *E. faecium* IMAU9421 were isolated from Carpathian cheese as cell-free supernatant (CFS) without any addition of hydrogen peroxide or adjusted pH value. The antagonistic activity of the bacteriocins LAB against pathogenic was studied by Musiy et al. (2020). They found that *E. durans* FMA8 had the highest activity against *S. typhimurium* PCM 2182, followed by *L. monocytogenes* PCM 2191, *E. coli* PCM 2208, and *S. aureus* PCM 458. Similarly, *L. lactis* IMAU32258 bacteriocins had the highest activity against *S. typhimurium* PCM 2182, followed by *E. coli* PCM 2208, *L. monocytogenes* PCM 2191, and *S. aureus* PCM 458. However, a bacteriocin from *L. garvieae* showed the lowest antagonistic activity against studied pathogenic bacteria. Surprisingly, the LAB cultures of the isolates demonstrated higher antagonistic activity towards these tested pathogens as compared to their bacteriocins.

Table 1. Activity of bacteriocins isolated from lactic acid bacteria of different types of cheeses.

Cheese	Technique	Bacteriocins LAB strain	Pathogenic strain	Inhibition zone (mm)	Control	Inhibition zone (mm)	p value	References
Traditional Kargı Tulum cheese	Agar well diffusion assay	*Enterococcus faecalis* KT11	*Klebsiella pneumoniae* ATCC 13883	14.0 ± 0.8	CSF	N.M.	p < 0.05	Abanoz & Kunduhoglu, 2018
			Serratia marcescens NRRL 2544	18.0 ± 0.6		N.M.		
			Enterobacter aerogenes ATCC 13048	15.0 ± 0.8		N.M.		
			Pseudomonas aeruginosa ATCC 2783	14.0 ± 1.0		18.0 ± 1.0		
			Listeria monocytogenes LMG 13305	15.0 ± 0.5		N.M.		
			Bacillus subtilis NRRL NRS 744	15.0 ± 0.8		N.M.		
			M. luteus NRRL 1018	16.0 ± 1.6		15.0 ± 0.8		
			Staphylococcus aureus ATCC 25923	16.0 ± 1.8		15.0 ± 0.5		
Mongolian Fermented Cheese	Oxford cup assay	*L. plantarum KLDS 1.0344*	*Escherichia coli ATCC 43889*	(+++)*	Sterile water	N.D.	p < 0.05	Muhammad et al., 2019
			Salmonella typhimurium ATCC 14028	(+++)*				
			Staphylococcus aureus ATCC 25923	(+++)*				
			Listeria monocytogenes ATCC 19115	(++)*				
Traditional cheese samples	Agar well diffusion assay	*Lactiplantibacillus plantarum*	*Listeria monocytogenes* ATCC 7644	86.60%	N.D.	N.D.	N.M.	Afshari et al., 2022
			Staphylococcus aureus PTCC 1431	80%				
			Salmonella typhimurium ATCC 14028	93.30%				
			Pseudomonas aeruginosa ATCC 9027	73.30%				
			Escherichia coli PTCC 1338	66.60%				
		Lactobacillus rhamnosus	*Listeria monocytogenes* ATCC 7644	80%				
			Staphylococcus aureus PTCC 1431	93.30%				
			Salmonella typhimurium ATCC 14028	86.60%				
			Pseudomonas aeruginosa ATCC 9027	100%				
			Escherichia coli PTCC 1338	93.30%				
		Limosilactobacillus fermentum	*Listeria monocytogenes* ATCC 7644	100%				
			Staphylococcus aureus PTCC 1431	93.30%				
			Salmonella typhimurium ATCC 14028	86.60%				
			Pseudomonas aeruginosa ATCC 9027	93.30%				
			Escherichia coli PTCC 1338	86.60%				

*+++ (> 10 mm), ++ (5-10 mm); [1]+ (0.5-10 mm), ++ (11-19 mm); +++ (> 20 mm); CSF = Cell-free supernatant; N.D. = Not detected; N.M. = Not mentioned.

Table 1. Continued...

Cheese	Technique	Bacteriocins LAB strain	Pathogenic strain	Inhibition zone (mm)	Control	Inhibition zone (mm)	p value	References
Carpathian cheese	Agar well diffusion assay	*L. lactis* IMAU32258	*S. aureus* PCM 458	8.4 ± 1.3	Bacteriocins producing culture	20.2 ± 2.8	$p < 0.05$	Musiy et al., 2020
			L. monocytogenes PCM 2191	17.1 ± 1.2		18.5 ± 1.6		
			S. typhimurium PCM 2182	18.9 ± 0.6		24.1 ± 1.9		
			E. coli PCM 2208	17.3 ± 2.1		18.7 ± 1.6		
		L. garvieae JB282647 2	*S. aureus* PCM 458	6.3 ± 0.9		8.4 ± 1.1		
			L. monocytogenes PCM 2191	6.2 ± 0.4		10.2 ± 0.9		
			S. typhimurium PCM 2182	7.4 ± 1.3		9.5 ± 1.4		
			E. coli PCM 2208	7.1 ± 1.3		8.4 ± 0.7		
		E. faecium IMAU9421	*S. aureus* PCM 458	6.1 ± 0.6		18.1 ± 1.4		
			L. monocytogenes PCM 2191	14.3 ± 2.1		14.4 ± 1.3		
			S. typhimurium PCM 2182	15.7 ± 1.7		20.3 ± 1.7		
			E. coli PCM 2208	16.5 ± 1.5		23.6 ± 2.4		
		E. faecium L3-23	*S. aureus* PCM 458	10.2 ± 1.7		19.5 ± 1.6		
			L. monocytogenes PCM 2191	18.5 ± 1.7		21.4 ± 0.8		
			S. typhimurium PCM 2182	20.1 ± 2.5		25.2 ± 1.2		
			E. coli PCM 2208	18.4 ± 2.7		20.1 ± 1.2		
		E. durans FMA8	*S. aureus* PCM 458	12.5 ± 2.9		20.6 ± 2.1		
			L. monocytogenes PCM 2191	22.6 ± 1.8		23.6 ± 1.3		
			S. typhimurium PCM 2182	23.8 ± 1.2		24.0 ± 0.9		
			E. coli PCM 2208	20.4 ± 1.4		20.3 ± 1.9		

*+++ (> 10 mm), ++ (5-10 mm); ᵗ+ (0.5-10 mm), ++ (11-19 mm); +++ (> 20 mm); CSF = Cell-free supernatant; N.D. = Not detected; N.M. = Not mentioned.

Table 1. Continued...

Cheese	Technique	Bacteriocins LAB strain	Pathogenic strain	Inhibition zone (mm)	Control	Inhibition zone (mm)	p value	References
Minas artisanal cheeses	Agar well diffusion assay	*Lb. plantarum* 1QB77	*S. aureus* FRI S6	inhibited	CSF	N.D.	$p < 0.05$	Margalho et al., 2020
			S. aureus FRI 361	no zone was observed				
			L. monocytogenes ATCC 3968	inhibited				
			L. monocytogenes ATCC 3973	inhibited				
Manteiga		*lactobacillus sp.* 3QB167	*S. aureus* FRI S6	inhibited				
			S. aureus FRI 361	no zone was observed				
			L. monocytogenes ATCC 3968	inhibited				
			L. monocytogenes ATCC 3973	inhibited				
Manteiga		*L.lactis* 1QB167	*S. aureus* FRI S6	inhibited				
			S. aureus FRI 361	no zone was observed				
			L. monocytogenes ATCC 3968	inhibited				
			L. monocytogenes ATCC 3973	inhibited				
Caipira		*lactobacillus sp.* 1QB459	*S. aureus* FRI S6	inhibited				
			S. aureus FRI 361	no zone was observed				
			L. monocytogenes ATCC 3968	inhibited				
			L. monocytogenes ATCC 3973	inhibited				
Colonial		*Lb. plantarum* 1QB314	*S. aureus* FRI S6	inhibited				
			S. aureus FRI 361	inhibited				
			L. monocytogenes ATCC 3968	inhibited				
			L. monocytogenes ATCC 3973	inhibited				
white cheese	Agar spot and well diffusion assay	*E. faecium* S1113	*E. coli* ATCC 25922	(++)[ǂ]	Bacteriocins producing culture	(+++)[ǂ]	$p < 0.05$	Mohammed & Çon, 2021
			B. cereus NRRL B-3711	(+++)[ǂ]		(+++)[ǂ]		
		E. durans S092	*E. coli* ATCC 25922	(++)[ǂ]		(++)[ǂ]		
			B. cereus NRRL B-3711	(+++)[ǂ]		(+++)[ǂ]		
		E. gallinarum S142	*E. coli* ATCC 25922	(++)[ǂ]		(+++)[ǂ]		
			B. cereus NRRL B-3711	(++)[ǂ]		(+++)[ǂ]		
		L. pentosus S056	*E. coli* ATCC 25922	(+)[ǂ]		(+++)[ǂ]		
			B. cereus NRRL B-3711	(+)[ǂ]		(+++)[ǂ]		
		E. durans S1121	*E. coli* ATCC 25922	(+++)[ǂ]		(+++)[ǂ]		
			B. cereus NRRL B-3711	(++)[ǂ]		(+++)[ǂ]		

*+++ (> 10 mm), ++ (5-10 mm); [ǂ]+ (0.5-10 mm), ++ (11-19 mm); +++ (> 20 mm); CSF = Cell-free supernatant; N.D. = Not detected; N.M. = Not mentioned.

Moreover, Margalho et al. (2020) used different types of cheese to isolate LAB bacteriocins. Among 27 isolates, five isolates had a strong inhibitory effect (Table 1). *L. plantarum* (1QB314) bacteriocin isolated from Colonial cheese showed an ability to inhibit the growth of all tested pathogens (*S. aureus* FRI S6, *S. aureus* FRI 361, *L. monocytogenes* ATCC 3968, and *L. monocytogenes* ATCC 3973). In addition, several bacteriocins were also produced from *Lb. plantarum* 1QB77 (Minas artisanal cheeses), *lactobacillus sp.* 3QB167 (Manteiga cheese), *L.lactis* 1QB167 (Manteiga cheese), and *lactobacillus sp.* 1QB459 (Caipira cheese) which showed varied in their inhibition activity. The treated bacteriocins with enzymes such as protease, α-chymotrypsin, proteinase K, and trypsin showed strong inhibition against tested pathogens with no zone of inhibition were observed. *E. faecalis* KT11 bacteriocin obtained from traditional Kargı Tulum cheese was tested against several pathogens and had the highest inhibition activity against the growth of *Serratia marcescens* NRRL 2544, *S. aureus* ATCC 25923, *M. luteus* NRRL 1018, *Bacillus subtilis* NRRL NRS 744, *Enterobacter aerogenes* ATCC 13048, *L. monocytogenes* LMG 13305 and *Klebsiella pneumoniae* ATCC 13883, respectively. Moreover, bacteriocin treated with pepsin and α-amylase had a significant ($p< 0.05$) inhibition activity against *Pseudomonas aeruginosa* ATCC 2783, *M. luteus* NRRL 1018, and *S. aureus* ATCC 25923. In contrast, *P. aeruginosa* ATCC 2783 and *M. luteus* NRRL 1018 bacteriocins had similar inhibition activity (16.0 ± 0.5 mm) when treated with catalase (Abanoz & Kunduhoglu, 2018).

L. plantarum KLDS 1.0344 bacteriocins isolated from Mongolian fermented cheese was tested against four pathogens, three of them had an inhibition zone larger than 10 mm for *E. coli* ATCC 43889, *Salmonella typhimurium* ATCC 14028, and *S. aureus* ATCC 25923 (Muhammad et al., 2019). However, *L. monocytogenes* ATCC 19115 had an inhibition zone between 5-10 mm. In addition, *L. plantarum* KLDS 1.0344 bacteriocins were treated with several factors such as pH, temperature, and enzymes (pepsin and protease). Bacteriocins treated with pepsin and protease have antagonistic activity against *S. aureus* ATCC 25923 with inhibition zone 18.5 ± 0.8 mm and 20.4 ± 0.8 mm, respectively whereas *L. monocytogenes* ATCC 19115 had the lowest activity [12.7 ± 0.5 and 14.5 ± 0.5 mm, respectively; Muhammad et al. (2019)]. All tested pathogens had the same inhibition zone of 100.0 ± 0.0 mm when treated with Mongolian fermented cheese bacteriocin at different temperatures (80 °C and 100 °C) and times (20, 30, and 40 min). In addition, the authors reported that antagonistic activity of *L. monocytogenes* was significantly ($p < 0.05$) enhanced when the temperature increased to 120 °C for 20 and 40 min (85.0 ± 0.7 and 61.2 ± 0.1 mm, respectively). On the other hand, lower activity towards *E. coli* was noticed at 120 °C for 20 and 40 min (81.0 ± 0.2 and 40.5 ± 0.0 mm; respectively). Furthermore, bacteriocin's treated at different pH values had the highest inhibition zone ($p < 0.05$) at pH 2 and the lowest at pH 6 against all tested pathogens (Muhammad et al., 2019). Recently, Lactiplantibacillus *plantarum, L. rhamnosus,* and *Limosilactobacillus fermentum* bacteriocins isolated from traditional cheese samples were found to have the highest inhibition zone activity against *S. typhimurium* ATCC 14028 (93.30%), *P. aeruginosa* ATCC 9027 (100%), and *L. monocytogenes* ATCC 7644 (100%); respectively (Afshari et al., 2022). Mohammed & Çon (2021) have investigated five bacteriocins isolated from LAB of white cheese (*E. faecium* S1113, *E. durans* S092, *E. gallinarum* S142, *L. pentosus* S056, and *E. durans* S1121). Both *E. faecium* S1113 and *E. durans* S092 had a positive effect on *B. cereus* NRRL B-3711 (> 20 mm) whereas *E. durans* S1121 affected significantly ($p < 0.05$) on *E. coli* ATCC 25922. In addition, the antagonistic activity against these two pathogens significantly improved when pH ranged between 3 – 9.5 with an inhibition zone (0.5-10 mm). All bacteriocins produced from the five isolates inhibited the growth of the tested pathogens with zones ranging from 0.5 to 10 mm (Mohammed & Çon, 2021).

8 Conclusion

Bacteriocins obtained from LAB have strong antimicrobial activity against bacterial pathogens, which makes them a promising alternative to antibiotics. Based on previously bacteriocins research, several strains of LAB-producing bacteriocins isolated from various types of cheese showed high inhibitory activity towards foodborne bacteria such as *L. monocytogenes*, *S. aureus*, *E. coli*, and others. *L. plantarum* bacteriocins were among the most studied bacteriocins against both gram-positive and gram-negative bacteria. More studies must be conducted to determine the best type of cheese that produce the most effective bacteriocin. Furthermore, bacteriocins differ in their inhibitory effect, so there is a need to find more bacteriocins with a broad spectrum against foodborne bacteria to be used as a food preservative to eliminate the growth of undesirable bacteria in cheese.

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