

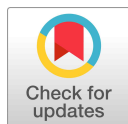
Review Article

Bacteriocins of Lactic Acid Bacteria and Their Industrial Application

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Abstract

Lactic acid bacteria produce different types of inhibitory compound that have a bactericidal effect. The inhibitory compounds produced include metabolic end products, bacteriocins that are antimicrobial peptides with antibiotic effect, hydrogen peroxides and numerous organic acids depending on their fermentation pathway. The inhibitory activity by bacteriocins produced by lactic acid bacteria is diverse, comprising of strains capable of inhibiting different types of Gram-positive microorganisms to those that affect only organism of close general. With the growing concern over the spread of antibiotic-resistant microorganisms and the possibility of current therapies becoming inefficient, it is imperative to find antibiotics alternatives. In food preservation, naturally occurring peptides with antimicrobial activity are favoured over chemical preservatives a more reason why there should be a holistic approach to taking advantage of bacteriocinogenic bacteria in medicine and food industry. This review focuses on the application of bacteriocin in food preservation, food industry, livestock and medicine.

Keywords

bacteriocin, lactic acid bacteria, antimicrobial activity, health, biopreservation

INTRODUCTION

Several inhibitory factors are produced by lactic acid bacteria (LAB). This factor is often from metabolic end-products that act like antibiotics and they are termed bacteriocins since they are bactericidal proteins with antimicrobial activity (Oliveira *et al.*, 2008). Many of these peptides such as lactacin, nisin, pediocins and subtilisin with antimicrobial activity (AMA) have been characterized and found to be potent with an activity similar to that of antibiotics (Chandrakasan *et al.*, 2019). There is growing concern over the rise of antibiotic-resistant microorganisms and the likely inefficiency of current therapies in the near future. These problems highlight the need to search for alternative strategies. On the other hand, interest in the biopreservation of food has considerably increased due to the adverse effects of chemical preservatives on human health. Well-characterized bacterial

peptides with antimicrobial activity, referred to as bacteriocins, have been utilized in various sectors such as the food industry and medicine (Rahmdel *et al.*, 2019).

The inhibitory factors which are made up of protein have been isolated from Gram-negative bacteria with more attention drawing to the same factor being produced by Gram-positive bacteria. The interest is basically due to the killing nature they possess against sensitive strain (Tagg *et al.*, 1976; Mathur *et al.*, 2017). Interestingly, the substances do not have a detrimental effect on the organism producing it. If this were the case, it would be self-limiting, hence, typically considered narrow-spectrum antibiotics, but sometimes they can be relatively broad-spectrum antibiotics. Bacteriocins can be described as a conglomeration of different protein types that deter the growth of other organisms susceptible to it. They are heterogeneous as a group and can be mainly classified as molecular weight differences (Klaenhammer *et al.*, 1993; López-Cuellar *et al.*, 2016; Mahdi *et al.* 2018). They can be either peptide (19-37 amino acids) or polypeptides (of about 90kDa) that are produced during the logarithmic growth phase, as against antibiotics that are produced during the stationary phase with no form of recorded allergy by humans as in the case of antibiotics (Beasley *et al.*, 2004). Specifically, we can separate bacteriocin from antibiotic on two main grounds: (a) bacteriocins are synthesized predominantly by the bacteria ribosomes; and (b) the inhibitory potency of bacteriocins are stronger against close genera, mainly on other bacteria whereas antibiotics are effective on diverse organism including insect and fungi (Cleveland *et al.*, 2001).

Consumption of bacteriocin has been part of our regular diet for several years through fermented foods (Oyewole, 1997) with no documentation of being harmful to humans even though, the knowledge of its importance was not known as at then (De Vuyst and Leroy, 2007). However, with the evolution of science and technology, the agent of fermentation is known (Table 1) (Franz *et al.*, 2014) and bacteriocin discovered to be the active component (Tagg *et al.*, 1976; Caplice and Fitzgerald, 1999). After that, more studies focused on how it can be of commercial importance,

Table 1. Some traditional fermented food and implicated lactic acid bacteria

S/N	Fermented food	Region/country consumed	Isolated lactic acid bacteria
1.	Ogi	Nigeria/ West Africa	<i>Lactobacillus fermentum</i> <i>Lactobacillus plantarum</i>
2.	Abreh	Sudan	<i>Lactobacillus plantarum</i>
3.	Uji	Kenya/East Africa	<i>Leuconostoc mesenteroides</i>
4.	Kenkey	Ghana	<i>Lactobacillus</i> spp.
5.	Mahewu	South Africa	<i>Lactobacillus bulgaricus</i>
6.	Mawe	Benin/West Africa	<i>Pediococcus pentosaceus</i>
7.	Zabadi	Egypt/North Africa	<i>Lactobacillus bulgaricus</i>
8.	Kawal	Egypt	<i>Lactobacillus plantarum</i>
9.	Kaffir	North Africa	<i>Lactobacillus delbrueckii</i>
10.	Burukutu	West Africa	<i>Lactobacillus</i> spp.
11.	Malawa	Zambia, Uganda	<i>Lactobacillus</i> spp.
12.	Enjera	Ethiopia	<i>Leuconostoc mesenteroides</i>
13.	Palm wine	West Africa	<i>Lactobacillus plantarum</i> <i>Leuconostoc mesenteroides</i>
14.	Gari	Africa	<i>Lactobacillus plantarum</i>
15.	Fufu	West Africa	<i>Lactobacillus brevis</i>
16.	Nono	West Africa	<i>Lactobacillus acidophilus</i> <i>Lactococcus cremoris</i>

Oyewole, 1997; Franz *et al.*, 2014.

its application in food preservation, and many ways to be utilized (Ghanbari *et al.*, 2013; Mahdi *et al.*, 2018). Alongside bacteriocin, there are other types of inhibitory substances such as diacetyl, organic acids and hydrogen peroxide produced by lactic acid bacteria though dependent on their fermentation profile that could be homofermentative or heterofermentative. The genera of lactic acid bacteria are *Lactobacillus*, *Streptococcus*, *Lactococcus*, *Leuconostoc*, *Pediococcus* and *Enterococcus* having varying degrees of inhibitory substances (Jamuna and Jeevaratnam, 2006) produced by them that limit the growth of different types of microorganism as well (Mathur *et al.*, 2017).

HISTORICAL BACKGROUND

Research on bacteriocins stemmed from investigations on colicins - the bacteriocin of Enterobacteriaceae family (Jack *et al.*, 1995), which led to increased knowledge of the genetic basis; study on how the substances are formed, the biochemical structure and the methods by which the molecules exact their antimicrobial properties (Pugsley, 1984). Nevertheless, the drive to know more about the antibiotic-like activities of Gram-positive bacteria, mainly from fermented food, never ceased. The study on these various food-grade organisms, which is now referred to as Generally Regarded as Safe (GRAS) organism, focused on their desirable attribute in the food industry as they are now applicable in food preservation to inhibit bacterial pathogens and spoilage microorganisms (Chandrakasan *et al.*, 2019).

It has been reported that Pasteur, together with Joubert were first to systematically record the ability of some bacteria to inhibit another in an experiment where two different bacteria were co-inoculated (Jack *et al.*, 1995). The experiment was carried out using urine as a culture medium and an animal infected with *Anthrax bacilli* with some level of inhibitory action detected. From the findings, it can be adjudged that the presence of a particular organism can interfere with the normal growth of the other organism when they are co-inoculated in the same medium, but the mechanism of action was unknown (Mallesha *et al.*, 2010). In order to know what inhibitory factor that brought about such an antagonistic relationship, a whole field of research opened up, which has been active from then till now.

Gratia was the first scientist that recorded his findings from an experiment conducted in 1925 about the nature of an unknown substance produced by *E. coli* having an activity like an antibiotic (Jack *et al.*, 1995). In his experiment, Gratia used a virulent strain V that produces a heat-stable element in broth culture that can be dialyzed. This element which was later on termed colicin V has an inhibitory activity against *E. coli* and when tested at different dilution it was still potent at higher dilution

(Gratia, 2000).

Subsequently, different types of colicins were discovered over time, while Jacob *et al.* (1953) used the word bacteriocin to give all the different substances a common name. The definition of bacteriocins considers that they are proteinaceous and act like antibiotics though of a different type (Jack *et al.*, 1995). Further to this, they are molecules that, after biosynthesis, become leather to mostly close general microorganisms by adsorbing to their cell surface (Jack *et al.*, 1995). Tagg *et al.* (1976) had opined in their review on the bacteriocins of Gram-positive bacteria that “most of the definitive investigations in the field of bacteriocins had centered on those of Gram-negative bacteria, but an increase in research emphasis on bacteriocins of Gram-positive lactic acid bacteria is needed”. Lactic acid bacteria have now turned to the most sought-out bacteria for bioprospecting for novel bacteriocin as they are considered a safe organism. The renewed interest cannot be separated from the fact that bacteriocin has much practical application when it comes to food preservation, industrial usage, or medicine to manage other bacterial that might cause diseases (Rahmdel *et al.*, 2019).

CLASSIFICATION OF BACTERIOCIN

The producing strains of bacteriocin are found in both Gram-positive and Gram-negative bacteria with the name generally derived from the producing genus or species (Rahmdel *et al.*, 2019). Bacteriocins are classified based on criteria such as, production method - ribosomal and non-ribosomal (De Vuyst and Leroy, 2007), genetics - which could be chromosomal or the size of the plasmid, the type of sugar and protein present, molecular weight of the substance and chemistry reaction it undertakes (López-Cuellar *et al.*, 2016) and the killing mechanism such as nuclease, murein production inhibition and pore formation (Rahmdel *et al.*, 2019).

With all these different factors considered based on physiochemical properties, bacteriocins are classified into three major classes (Table 2) (Silva *et al.*, 2018; Kumariya

Table 2. Classification of bacteriocin

Class	Sub-class	Example	Producing strain
I	Ia	Nisin	<i>Lactococcus lactis</i>
	Ib	Labyrinthopeptin A1	<i>Actinomadura namibiensis</i>
	Ic	Thuricin CD	<i>Bacillus thuringiensis</i>
II	IIa	Pediocin PA-1	<i>Pediococcus pentosaceus</i>
		Sakacins A and P	<i>Lactobacillus sakei</i>
		Leucocin A	<i>Leuconostoc gelidum</i>
	IIb	Lactococcins G	<i>Lactococcus lactis</i> subsp. <i>cremoris</i>
		Plantaricin EF	<i>Lactobacillus plantarum</i>
		Plantaricin JK	<i>Lactobacillus plantarum</i>
	IIc	Gassericin A	<i>Lactobacillus gasseri</i>
		Enterocin AS-48	<i>Enterococcus faecalis</i>
		Garvicin ML	<i>Lactococcus garvieae</i>
	IId	Bactofencin A	<i>Lactobacillus salivarius</i>
		LsbB	<i>Lactococcus lactis</i> subsp. <i>lactis</i>
	III	Helveticin M	<i>Lactobacillus crispatus</i>
		Helveticin J	<i>Lactobacillus helveticus</i>
		Enterolysin A	<i>Enterococcus faecalis</i>

López-Cuellar *et al.*, 2016; Mahdi *et al.* 2018; Kumariya *et al.*, 2019.

et al., 2019). On sound scientific basis, Savadogo *et al.* (2006) reported the establishment of three types of LAB bacteriocins: the lantibiotics (class I); the non-lantibiotics (class II) and the heat sensitive (class III) bacteriocins. Class IV was reported by Caplice and Fitzgerald (1999) as complex bacteriocins with glycol- and/or lipid moieties but it is no longer considered in the classification (Kumariya *et al.*, 2019).

Class I bacteriocins are referred to as lantibiotics. Basically, bacteriocins which comprise modified amino acids and lanthionine in their structure are known as lantibiotics. They are inhibitors that target bacterial peptides with a weight of

about 10 kDa. Class I bacteriocin are heat stable and post-translationally modified (Prada *et al.*, 2007). Nisin is an example of Class I bacteriocin and most common among the class (Lay *et al.*, 2016). They are further classified into Class Ia, Ib, Ic corresponding to lantibiotics, labyrinthopeptins and sanctibiotics, respectively (Kumariya *et al.*, 2019).

Class II bacteriocins have different mode of action and are subdivided into IIa, IIb and IIc. Although, Kumariya *et al.* (2019) reported subclass IId and Mahdi *et al.* (2018) reported subclass IIE. They are small proteins and are heat stable as well. Class IIa – with N-terminal forms the largest subclass of Class II. It is a bacteriocin with pediocin-like nature. They also contain certain sequence in the group that recognizes specific species having the activity of targeting the bacteria cell wall affecting its permeability, thereby causing leakage on the cell (Oppegård *et al.*, 2007). It is also an anti-*Listeria* bacteriocin that is found in this class only (Drider *et al.*, 2006). Class IIb is a two peptide bacteriocin. For its activity, it requires two different peptides for optimum inhibitory activity. Class IIc bacteriocin is a circular and thiol activated peptide (López-Cuellar *et al.*, 2016) that affects the pheromone as well as the permeability of the cell membrane of target cells, including the disruption of the formation of the cell wall (Klaenhammer *et al.*, 1976). Class IId, a linear, non-pediocin-like, unmodified bacteriocins was described by Kumariya *et al.* (2019) while Class IIE contains three or four nonpediocin-like peptides (Mahdi *et al.*, 2018).

Class III is a class of bacteriocins with a molecular weight greater than 30kDa and made up of proteins that are very sensitive to heat (Mahdi *et al.*, 2018). The LAB of genus *Lactobacillus* have been the major producer (Klaenhammer *et al.*, 1976) and from several studies they have shown to have a wide range of inhibitory effects on several common pathogenic organisms (Kumariya *et al.*, 2019). It should be noted that Class IV has been reclassified as bacteriolysins that are made up of leuconocin S and lactocin (Kumariya *et al.*, 2019). It is a complex bacteriocin containing different chemical compounds that include lipids (Klaenhammer *et al.*, 1976).



NATURE AND MODE OF ACTION OF BACTERIOCCIN

Bacteriocins are bactericidal protein that, when produced by some bacteria genera are toxic and affect other bacteria stain growth. Usually, bacteriocin are classified as narrow-spectrum antibiotics, but such classification has been a subject of unending debate (López-Cuellar *et al.*, 2016; Rahmdel *et al.*, 2019). After the first discovery of bacteriocin termed colicin by Andre Gratia in 1925 as a result of its inhibitory effect on *E. coli*, they have been classified in several ways by their mode of inhibitory action with consideration on the type of strain producing them and mechanism by which they exert resistance (Gratia, 2000). Bacteriocins are very diverse, having several categories that are phenomenologically related. It has been discovered in Gram-negative bacteria, such as colicins (Cascales *et al.*, 2007) and from Archaea that are termed microcins. Specifically, colicins are bacteriocins from *E. coli*, which are the most studied bacteriocins. Warnericin is another bacteriocin produced by *Staphylococcus warneri*, while Colicin V is part of the oldest type of colicins discovered from Archea, which has been renamed as microcin V (Prema *et al.*, 2006).

The mode of action of bacteriocin is often by attacking the bacterial membrane. They bind to specific sites on the cells and often affect cell membrane integrity and function, resulting in a bacteriostatic effect on bacterial species and bacteriocidal influence on other species (Arsi *et al.*, 2015). The absorption of bacteriocins may have different biochemical effects on a cell, such as interference with specific cleavage of 16S ribosomal RNA, degradation of cellular DNA, and peptidoglycan synthesis inhibition. The bacteriocins of lactic acid bacteria have been studied extensively and numerous reports show that their inhibitory effect is wide and effective against Gram-negative and Gram-positive bacteria. Some bacteriocins allow surfactant-like activities on cell membranes, thereby disrupting cellular functions (Muriana *et al.*, 1990; Jamuna and Jeevaratnam, 2006), whereas as reported by Tagg *et al.* (1976) some have very specific bactericidal effect only

on some particular Gram-positive bacteria. Nisin is a good example of such inhibiting *Staphylococci*, *Streptococci*, *Bacilli*, *Clostridia*, and *Mycobacteria* (Cleveland *et al.*, 2001; López-Cuellar *et al.*, 2016).

One question that might linger on how minds is, how does a particular strain not get affected by its own bacteriocin? This is where specific immunity comes into play, whereby a strain producing a specific bacteriocin is not affected by it. This mechanism is believed not to be the same by which some Gram-negative and Gram-positive bacteria evade the killing effect of bacteriocin (Mathur *et al.*, 2017). The most likely explanation to how this evading mechanism is possible would be the suggestion by Stevens *et al.* (1991) that there are some specific barriers on the outer membrane of a bacteria cell wall. Through the aide of this membrane, some Gram-positive and Gram-negative bacteria are able to develop an iota of a higher level of resistance to the inhibitory effect of bacteriocin (Kumariya *et al.*, 2019).

Effects of bacteriocin on several pathogenic organisms cut across organisms responsible for food spoilage and also microorganisms that are food-borne pathogens (Schillinger *et al.*, 1996). Nisin has this type of attribute and was also the first bacteriocin that was used as a food preservative on a commercial scale. However, a study on lactic acid bacteria bacteriocins has dramatically changed over the years (López-Cuellar *et al.*, 2016) to bio-prospecting from different sources such as traditionally fermented foods and plant for new strains with the ability to produce bacteriocin which as lead to the discovery of many more bacteriocins (Cleveland *et al.*, 2001; Chandrakasan *et al.*, 2019).

FACTORS THAT CAN AFFECT BACTERIOCCIN PRODUCTION

Temperature, pH, salinity and media components, which are environmentally driven factors, are important external factors for a higher amount of bacteriocin and biomass production by lactic acid bacteria (Kanmani *et al.*, 2011, Sahar *et al.*, 2017). Moreover, bacteriocin production by LAB strains is growth associated and is, therefore, induced

by growth conditions, including temperature and medium pH. The temperature is one of the important culture parameters which can significantly enhance the growth of bacterial strains by improving the fermentation conditions. The major consequence of temperature during the growth of LAB is in the production of bacteriocin with little or no effect on their inactivation (Kim *et al.*, 2006; Holtsmark *et al.*, 2008).

The pH of the medium is another important cultural requirement for bacteriocin and biomass production by LAB strains (Kanmani *et al.*, 2010). During the fermentation period of LAB strains, most of the by-products are organic acids that reduced the pH of the growth medium. The low pH of the medium can either slow down the growth rate or completely stop the growth of the bacterial cells. Thus, controlled pH fermentation can help improve the growth of bacterial cells and increase bacteriocin production (Anthony *et al.*, 2009, Sahar *et al.*, 2017).

Growth media and its components are other key factors for a higher bacteriocin production and a good fermentation process. Bacteriocin-producing lactic acid bacteria needed complex nutritious media to grow and bacteriocin production as the cell growth and metabolite production are greatly influenced by various carbon and nitrogen sources, growth factors and organic salts (De Vuyst and Leroy, 2007; Sahar *et al.*, 2017).

APPLICATION OF BACTERIOCIN

Bacteriocins from lactic acid bacteria have been demonstrated as food preservatives (Rihakova *et al.*, 2009) and as therapeutic agents for veterinary and medical uses (Tagg *et al.*, 1976) also as phytosanitary for the protection of plants (Holtsmark *et al.*, 2008). The growing interest in bacteriocin usage in medicine cannot be unconnected because the source is often from a non-pathogenic organism commonly found in the gastrointestinal tract and is mainly used as a probiotic (Bamgbose, 2014).

Their usage has been predominantly in the food industry for the preservation of food products. Often, bacteriocin

can be used directly or alongside other treatment methods determined by reason of usage and nature of the food (Lade *et al.*, 2006). Lactic acid bacteria bacteriocins are applied either in the form of protective culture or as an additive and they are known to satisfy industrial and consumer demands for minimally processed and fresh foods (Todorov, 2008). In essence, the bacteriocins produced by LABs are very desirable as they are known to be naturally produced by an organism that falls under GRAS. Their consumption is also safe due to the ease by which they can be digested (Cleveland *et al.*, 2001).

BACTERIOCINS AS FOOD BIOPRE-SERVATIVES

Naturally, bacteriocin is produced by the LAB strain within the food matrix, this knowledge is important to its application as a food additive. One of the most successful applications in food industry is that of nisin that has been purified to a certain degree, mixed with pediocin and available in the market (Silva *et al.*, 2018). To substitute adding bacteriocin directly into foods, a starter culture that can produce the same bacteriocin can be used or alongside a co-culture. A couple of studies have shown that lactic acid bacteria used as starter culture can directly produce their bacteriocins in food, thereby inhibiting food spoilage and pathogenic bacteria. For example, bacteriocin extraction has been demonstrated in Cheddar cheese, fermented sausages, sourdough (Foulquié *et al.*, 2003) and milk (Bamgbose, 2014). The effectiveness of bacteriocin as food preservatives have been well documented by Cleveland *et al.* (2001) and the advances in the past 20 years in food and pharmaceuticals (Chandrakasan *et al.*, 2019).

The methods that are commonly used in the bio-preservation of foods using bacteriocins include:

- (1) The direct inclusion during food processing of lactic acid bacteria that can produce bacteriocin (Silva *et al.*, 2018). Due to the inherent potential of the LAB to use the food matrix as a growth medium and secret, the



- desired metabolite is the main factor for utilizing this method (Schillinger *et al.*, 1996; Cleveland *et al.*, 2001).
- (2) Another method is to extract the desired bacteriocin and apply it into food (Schillinger *et al.*, 1996), which can be done either by using a purified bacteriocin extract or partially purified in a mixture of other substances (Silva *et al.*, 2018) while several methods of nano-encapsulation is also being embraced (Chandrasekaran *et al.*, 2019).
 - (3) Likewise, products that have been formerly produced using the desired LAB strain can be incorporated as part of the constituents in processing other foods (Schillinger *et al.*, 1996).

BIO-PRESERVATION OF MEAT PRODUCTS

One of the most rigid policies regarding *Listeria monocytogenes* is that of The United States government that does not permit the existence of the slightest number of *L. monocytogenes* in any type of food (Mohamed *et al.*, 2012). This food pathogen has been discovered as an agent responsible for food poisoning in meat (Ming *et al.*, 1997) and are commonly found within abattoir, meat processing industry, cooked and even refrigerated meat (Cleveland *et al.*, 2001). In order to curtail and control the incidences of *L. monocytogenes* in meat products, there have to be more aggressive research towards utilizing natural and safer methods with bacteriocin as a good metabolite to be considered. Moreover, there needs to be more intensive study to consolidate on the finding of Hugas *et al.* (1998) that reported the efficacy of bacteriocin produced by *Lactobacillus sakei* CTC494 that was deterrent to the growth of *Listeria innocua*, which he called sakacin. Vignolo *et al.* (2000) also reported that *Lactobacillus casei* CRL 705 secretes lactocin 705 that controls the growth of *L. monocytogenes*. Other studies have shown the potential benefit of using bacteriocin as a preservative agent in raw meat, cooked pork, and packaged

poultry products (Schillinger and Lucke, 1989; Rhamdel *et al.*, 2019). Different bacteriocins used have been documented by Cleveland *et al.* (2001), giving us insight on different ones used for various purposes. All the identified bacteriocin needs in-depth study to have a broader knowledge of their activity, the active compounds in them, and the fermentation condition for their optimum production.

BIOPRESERVATION OF DAIRY PRODUCTS

Listeria monocytogenes incidences are not limited to only meat; outbreaks associated with dairy products like cheese, pasteurized milk have also been documented (Schillinger *et al.*, 1996; Mohamed *et al.*, 2012). Nisin has also been applied in the dairy industry since it can control *L. monocytogenes* (Vignolo *et al.*, 2000). Its usage includes adding it to the cheese during pasteurization to stop the germination of clostridial spores, causing *Clostridium*-associated butyric acid fermentation a problem in cheese production (Schillinger *et al.*, 1996). Another useful example is the bacteriocin of *Lactobacillus lactis* subsp. *lactis* DPC 3147 (Lacticin 3147) has a two-component bacteriocin having a broad-spectrum activity as diverse microorganisms are sensitive to it. It was found to be useful in improving the quality of cheddar cheese by limiting the growth of other LAB that were not part of the starter culture, especially during the ripening stage (Ross *et al.* 2002). It has also been shown to have a potential in milk preservation (Bamgbose, 2014) and also in keeping dairy products in good condition for an extended time protecting it from spoilage organism thereby maintaining its safety (Table 3) (Silva *et al.*, 2018). Nisin, pediocins, lacticins and enterocins have all been suitable except for the drawback of using enterocins due to the pathogenic strain of the producing genera (Silva *et al.*, 2018).

Table 3. Some lactic acid bacteria in dairy industry

S/N	LAB strain	Application
1.	<i>Lactobacillus plantarum</i> TF711	Cheese production from cow milk as a co-starter culture to reduce <i>Clostridium</i> spores.
2.	<i>Lactobacillus reutri</i> INIA P572	Added to starter culture in cow milk cheese in order to produce reuterin for <i>Clostridium</i> control.
3.	<i>Lactobacillus gasseri</i> K7	In soft cheese production to elongate the time for cheese blowing.
4.	<i>Lactococcus lactis</i> IFPL3593	Control of <i>Clostridium</i> spores in soft cheese production.
5.	<i>Lactococcus lactis</i> subsp. <i>lactis</i> IPLA 729	For Vidiago cheese production to inhibit <i>Clostridium tyrobutyricum</i> growth.
6.	<i>Lactococcus lactis</i> subsp. <i>lactis</i> INIA 415	For organoleptic improvement in cheese production from ewes' milk and control <i>Clostridium</i> overgrowth.
7.	<i>Streptococcus macedonicus</i> ACA-DC 198	To control the growth of <i>Clostridium tyrobutyricum</i> during cheese production from unpasteurized sheep milk.

Silva *et al.*, 2018.

BIO-PRESERVATION OF SEAFOOD PRODUCTS

At a lower temperature, when carbon dioxide is combined with nisin, it becomes more active in controlling *Listeria monocytogenes* as they found it difficult to survive in such conditions. This method has been successfully used in extending the shelf life of salmon (Nilsson *et al.*, 1997). Some lactic acid bacteria were isolated from sea creatures such as salmon and shrimp (Noordiana *et al.*, 2012), with their metabolite being a good source of bio-preservation in seafood and seafood products (Ghanbari *et al.*, 2013). Also, from the bacteriocin study secreted by *Lactobacillus plantarum* F12, it was found to have a broad inhibitory spectrum against many indicator strains for food spoilage bacteria (Mohamed *et al.*, 2012). Thus, in this era where

many reports claim how unhealthy red meats are and the need to switch to eating more seafood, there must be more efforts to treat and extend the shelf life of seafood and its product. Considering how tender seafood are which make them not suitable for rigorous chemical or physical treatment; to elude nutrient loss and taste, industries need to embrace the use of natural bio-preservative agents in which bacteriocin is a good agent for controlling spoilage microorganism while retaining the integrity of the seafood and/or its product (Ghanbari *et al.*, 2013).

BACTERIOCINS IN HUMAN MEDICINE

In the human gut, there exists a continuous interaction between good and bad bacteria. Lactic acid bacteria fall under the category of good bacteria and they are the major source of bacteriocins which brought about the interest in medicine. The depletion of good bacteria - natural gut microbes by overuse of antibiotics might increase the population of undesirable pathogenic bacteria invading the gut, thereby causing diseases. A number of studies have focused on the possibility of bacteriocin being an effective anticancer agent (Kumar *et al.*, 2010; Kaur and Kaur, 2015; Prosekov *et al.*, 2015) or diagnostic agent in some type of cancerous growth (Farkas-Himsley *et al.*, 1995). Though, this has not been the main focus by an oncologist as it has only been on an experimental scale. The fact that no strong link has been established between anti-bacteria agents and inhibition of mammalian tumor cells is a major factor for the shift in focus. As part of looking at its application in medicine, bacteriocins have also been studied as a potential agent in ameliorating AIDS (Farkas-Himsley *et al.*, 1995; Martin *et al.*, 2010). While working on *Lactobacillus* from various dairy products in America, Ali *et al.* (1996) from their study reported that *L. acidophilus* has an inhibitory activity against food-borne microbial pathogens thus, concluding that their presence in yogurt and acidophilus milk has a positive impact on human health. A number of studies have also shown how different bacteriocin are important in medicine as documented by Mathur *et al.* (2017).

BACTERIOCINS APPLICATION IN LIVESTOCK

In several studies, probiotic bacteria have been tested in livestock to see how it can control food-borne pathogens (Mathur *et al.*, 2017). One of the easily transmissible pathogens to humans from the consumption of poultry products is *Campylobacter* spp., which was effectively controlled in experimental chicken by the presence of lactic acid bacteria in the gut that reduced enteric *Campylobacter* count (Arsi *et al.*, 2015). The most probable mechanism would be similar to *Paenibacillus polymyxa* and *Bacillus circulans* producing bacteriocin that has an inhibitory effect in broiler chicken by interfering with the growth of *Campylobacter jejuni* in the bird's colon (Cole *et al.*, 2006). This was possible as a result of the antimicrobial effect bacteriocin has on some pathogens and also their ability to disrupt the site used for establishment in the gut as was the case in the study by Cole *et al.* (2006) where bacteriocin fed to Turkey poult limited the adherence of *Campylobacter coli* to their gastrointestinal tract. As discussed earlier, bacteriocins are effective against other food pathogens. It has been shown to have anti-fungal properties especially in poultry feeds (Deepthi *et al.*, 2016) and helps in nutrient assimilability and health status of weaned piglets (Dowarah *et al.*, 2018). Another application involves incorporating bacteriocin in the teat seal of the livestock to prevent staphylococcal infection of the teats (Paul *et al.*, 2005).

Though, the result of most of this study has not elucidated the mechanism this is achieved. We know that bacteriocin production is a significant factor in the inhibition of the pathogenic organism, which has been achieved by directly feeding the livestock with strains that produce bacteriocin (Francisco, 2015).

BACTERIOCINS IN PACKAGING FILMS

Considerably, the usage of packaging films is applicable in safeguarding most of the other food items. Hence, the mechanism for impregnating the bacteriocin into the

package films is being explored (Deshmukh and Thorat, 2013). When a packaging film that has been incorporated with antimicrobial peptides is in close proximity with the food surface, the bacteriocin percolates into the food matrix (Appendini and Hotchkiss 2002). Due to the contact, a bacteriocin from the packaging film will gradually diffuse into the food and this might be of an advantage over spraying or dipping the food into bacteriocin. In some cases, the antimicrobial potency may be lost due to the inactivation of the bacteriocins by components in the food or due to weakening of activity as the concentration lowers while being mixed with the food matrix (Appendini and Hotchkiss, 2002).

In preparing packaging films with bacteriocins, the two methods mainly used are the incorporation of nisin into biodegradable protein films and straightly into polymers. In the two-scenario growth, *Lactobacillus plantarum* was inhibited by the resulting film formed. An example is the packaging film for beef using nisin-impregnated polyethylene; the effect was the control of *Bacillus thermosphacta* and *Lactobacillus helveticus* present on the tissue of the beef (Coma *et al.*, 2001). Though, in the case of *L. innocua* and *Staphylococcus aureus*, the effectiveness was reduced because of the stearic acid treatment on the film showing that other factors can interfere with the efficacy of this method (Coma *et al.*, 2001). The process can also be carried out by adsorbing bacteriocin to a polymer surface or incorporating it into biodegradable material like methylcellulose used successfully in poultry (Appendini and Hotchkiss, 2002). It has also been demonstrated how effective bacteriocin coatings can inhibit pathogens. In the study of pediocin coated on cellulose casings and plastic bags, the outcome was the complete inhibition of *Listeria monocytogenes* that was inoculated on meats and poultry refrigerated for 12 weeks (Ming *et al.*, 1997). It also inhibited common food contaminants when used as antibacterial packages in food industries (Damania *et al.*, 2016).

FUTURE PERSPECTIVES FOR BACTERIOCIN

To ascertain food safety and sustenance of its production industries needs to fund more research that would help focus on engineering microorganisms for enhanced anti-microbial production and improve the nutritional aspect. Taking advantage of the era of genomics that we are in, there must be an improvement in the quality of food produced and the content therein without neglecting the safety concerns.

Furthermore, numerous studies focus on enhancing bacteriocins that can be used in place of conventional antibiotics. Take, for example, pyocins produced by *Pseudomonas aeruginosa* is being studied for the possibility of modifying its specificity and, more recently nano-encapsulation of bacteriocin (Chandrakasan *et al.*, 2019). It is possible that in the future, probiotics will be used for different gastrointestinal diseases (López-Cuellar *et al.*, 2016), including *Helicobacter pylori* (Mahdi *et al.*, 2018), common pathogens of the oral cavity, vaginosis (Mathur *et al.*, 2017) or as delivery systems for vaccines, immunoglobulins, and other therapies (Ngwa and Pradel, 2015; Chandrakasan *et al.*, 2019).

CONCLUSION

Bacteriocins are useful in the fields of human and veterinary medicine, as well as meat and dairy preservation. Nevertheless, there are some limiting factors against the potency of their usage, such as enzymes within the food matrix, the possibility of a particular strain to lose its ability to secrete the desired compound, degradation of the bacteriocin by other extracellular enzymes and the adverse effect industrial process can have on the organism and/or its product. Notwithstanding, this is why further study is always advised to navigate these potential downsides to develop a better product. Besides the establishment of how important and potent bacteriocins are, considering their usefulness in medicine, food industry and livestock production; there is the need for more research elucidating their mechanism of action and also on the expansion of their spectrum of action against pathogens.

Author contribution

The final manuscript was read by all authors and approved.

Conflict of Interest

Authors have no conflict of interest to declare.

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