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REVIEW ARTICLE

Bacteriocin: safest approach to preserve food products

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Abstract Start of the 21st century with its universal call to feed the hungry is an appropriate time to refocus attention on food security and especially the impact of biopatenting on poor communities who are the primary victims of hunger in our world. Antibacterial metabolites of lactic acid bacteria and *Bacillus* spp have potential as natural preservatives to control the growth of spoilage and pathogenic bacteria in food. Among them, bacteriocin is used as a preservative in food due to its heat stability, wider pH tolerance and its proteolytic activity. Due to thermo stability and pH tolerance it can withstand heat and acidity/alkanity of food during storage condition. Bacteriocin are ribosomally synthesized peptides originally defined as proteinaceous compound affecting growth or viability of closely related organisms. Research is going on extensively to explore the nascent field of biopreservation. Scientists all over the world are showing their keen interest to isolate different types of bacteriocin producing strains and characterize bacteriocin produced by them for food preservation.

Keywords Bacteriocin · Biopreservative · Food pathogen · Chemical preservative

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Introduction

Among biopreservatives, bacteriocin has caught the attention of food scientists to be used as a natural food biopreservative due to its antimicrobial activity against food spoilage and pathogenic bacteria. Different bacteriocin producing strains of lactic acid bacteria as well as *Bacillus* spp. have been isolated for this purpose but the keen interest towards bacteriocin of lactic acid bacteria wordwide is due to their essential role in majority of food fermentation, flavor development and preservation of food products alongwith proving safer for health. Preservation action of lactic acid bacteria is due to production of lactic acid, acetic acid, hydrogen peroxide as well as bacteriocin resulting from metabolic activity of organism. Since adequate levels of organic acid, acetic acid, hydrogen peroxide are not desirable in many food products, therefore there is a demand for microbial agent suitable for use in such products [1].

Use of microorganisms in food fermentation is one of the oldest method for producing and preserving food. Much of the world depends upon various fermented food that are staples in the diet. Till the end of 1950, a very few of the food items were processed and packaged. Processed and packaged foods were luxury item in colonial times but after 1960 these food items were in great demand globally due to growing urbanization, breakdown of large families in to nuclear families and increase in the number of working women. Chemical preservatives and other traditional barriers have been used in food products to inhibit microbial growth which lead to serious health disasters, thus challenging the food scientists for providing safer and healthier food. Food preservation has become a major issue because food borne pathogens can cause havoc in preserved/fresh

food items at high temperature, room temperature and even at low temperature [2]. However consumer demand for faster, healthier and ready-to-eat products have strongly demanded the use of more natural preservatives instead of chemical preservative. Microbiologists around the world got interested in bacteriocin-producing microorganism to overcome this problem that fulfill the requirement of food preservation. Although many other type of bacteriocin such as subtilin, cerein, thuricin, plantaricin etc have been isolated and characterized and are still in a process of getting commercial status to be used as food preservatives [3, 4], so far only one bacteriocin, Nisin, has been given the status of preservative to be added in food items commercially. The US food and drug administration had given GRAS status to Nisin [5]. Nisin was first discovered in England in 1928 as a result of difficulty experienced during cheese making. Storage of milk had allowed contaminated organism to grow and produce inhibitor. The potential application of nisin in food preservation was demonstrated in 1951 [6] and later its use as a food preservative were elaborated. Nisin have been concentrated as as Nisaplin which is used as preservative in milk, dairy products, canned foods, cured meats and other segments of fermentation industry [7, 8]. Nisin inhibits virtually all gram-positive bacteria in food. International acceptance of Nisin was given in 1969 [9].

The following requirements should be fulfilled by any bioperservative to be used commercially [10–12].

1. The biopreservative to be used should not be toxic.
2. It should be accepted by recognized authorities.
3. It should be economical to the industries using it.
4. The product in which the biopreservative is being used should not be affected by it, i.e. biopreservative should not show any deleterious effect toward the organoleptic properties of that product.
5. When used at relatively low concentrations it should show effect.
6. The biopreservative should be sufficiently stable if being stored.
7. It should not have any medicinal use.

Bacteriocins fulfill all the above requirements and hence are gaining popularity in the food industry day-by-day. Bacteriocin have been grouped into four main distinict classes [13].

Class-I	Lantibiotics characterized by the presence of unusual thioether amino acid which are generated through post translational modification.
Class-II	Bacteriocin represent small (<10 kD) heat-stable, membrane active peptides.
Class-IIA	Subclass II A represented by *Listeria* active peptides which contain the N terminally located consensus sequence YGNGCXV where X is any amino acid.
Class IIB	Representing portion complexes that require two different peptides for activity.
Subclass-IIC	Peptide whose externalizations into the growth medium of the producing bacterium is dependent on the general secretory pathway.
Class-III	Bacteriocins belonging to class III consist of large (>30 kD) heat labile protein.
Class-IV	Represent complex bacteriocin that contain essential lipid, carbohydrate moieties in addition to a protein compared.

Objective

Objective of this review is to summarize important characteristics of bacteriocin of lactic acid bacteria and *Bacillus* sp.

Availability of bacteriocin-producing strain

Bacteriocin producing microflora commonly isolated from food and food products. Lactic acid bacteria play essential role in food fermentation and are employed as starter culture in manufacture of dairy, meat, vegetable and bakery products. In a mixed natural population and under adequate circumstance the incidence of bacteriocin producer strains among fresh isolates of any gram-positive species may approach 100% [14]. Bacteriocin production seems to be aimed to compete against other bacteria which are closely related or present in same ecological niche while antibiotic is a microbial product or its derivative that kills susceptible microorganism or inhibit their growth [15]. Bacteriocin productions provide producer strains with a selective advantage over other non producing bacteria.

Isolation of bacteriocin-producing strain

Bacteriocin-producing biotas have been isolated from different sources enlisted in Table 1.

But there is a considerable interest to isolate bacteriocin producing strains from lactic acid bacteria because preservation of fermented food items is due to the metabolic activity of lactic acid bacteria [19]. Bacterial strain isolated from different food samples by pour plate method and isolated strains were screened on the basis of their inhibitory activity against

Table 1 List of different bacteriocin-producing isolates and their sources

Sources	Name of isolate	References
Sour dough	*L. reuteri, L. bavaricus, L. curvatus, L. plantarum*	[16, 17]
Ripened soft cheese	*C. piscicola*	[18]
Chicken meat	*L. mesenteroides, L. plantarum*	[19, 79]
Cheese	*B. linens* ATCC 917520	[20]
Vacuum packed meat	*L. sake, L. sake*	[21, 22]
Whey	*L. helviticus* G51, *B. mycoides*	[23, 24]
Sausages	*L. brevis* SB27, *L. curvatus* SB	[25, 26]
Goat meat	*E. faecium*	[27]
Gajani sikhe	*Lactobacillus SPY21*	[28]
Raw barley	*Lactobacillus* sp.	[29]
Barley beer	*L. paracasei, L. pentosus, L. plantarum, Lactococcus lactis* subsp. *lactis*	[30]
Chilli pickle	*Pediococcus acidilactici*	[31]
Commercial salad	*L. casei, L. plantarum, Pediococcus* sp.	[32]
Tempeh	*Lactobacillus* sp.	[33]
Soil	*B. thuringiensis*	[34, 35]
Human hand	*Bacillus* sp. strain 8a	[36]
Vari kandal	*L. brevis*	[37]
Khameera yoghurt	*B. lentus, L. plantarum*	[2, 80]

selected pathogenic or spoilage causing bacteria. Indicators selected for screening are those which cause spoilage in food or pathogenic for human consumption. Many spoilage causing/pathogenic bacteria which have been used by different microbiologists as a indicator strains to check of activity of bacteriocin of different strains have been listed in Table 2.

Composition of growth media for bacteriocin-producing strain

Different media such as de man Ragosa sharpe (MRS) [46–49] Brain heart infusion [50], Todd-Hewitt broth [51], nutrient broth [2, 24, 37] skimmed milk [52] yeast extract medium [53] Lactose broth [51] litmus milk [54, 55], tryptic soya broth [56, 57] are best for bacteriocin production [54]. Bacteriocin-producing strains depends upon nutrients supplied by growth media. [52, 55] Amino acid, small peptide voucher trypson, glucose, sucrose, B vitamin, yeast extract contribute in growth of microorganism [58]. In addition, minor amount of Tween 80, Mg, Mn, salt acetate, phosphate chloride facilitate bacteriocin-producing capacity of strain. A general liquid media containing 1% each of tryptone, glucose, yeast extract together with 0.05% $MnSO_4$, $MgSO_4$ and 0.2% Tween 80 (TGE broth) was found to facilitate growth and high level of bacteriocin production by lactic acid bacteria. Frequency of bacteriocin producing strain in a food sample can be achieved by incubating sample at 30°C for 16 to 18 h in a nutritional broth containing 1% glucose with pH adjusted to 4.9 to 5.0.

Effect of pH on bacteriocin production

Influence of pH on bacteriocin production has also been studied. Optimum pH for bacteriocin production is between 6.0 to 7.0 [59]. Some of the bacterial strains were found to produce active bacteriocin at pH range 5.8 to 7.9 [60, 61] while strain *Leuconostoc* MF215B was found to produce bacteriocin at pH 6.0 but not at pH 7.5 [46]. Strain *L. gelidin* showed best production at 6.5 [62]. Similarly, amylovorin L471 produced at pH 6.5 [47].While *C. piscicola* produced bacteriocin at pH 7.0 [18]. None of the author in the literature so far reported bacteriocin production in the alkaline range, i.e. between pH 9.0 and 12.0. So it could be concluded that optimum bacteriocin production is exhibited between the range of pH 5.0 and 8.0 and production declined beyond these limits.

Effect of temperature on bacteriocin production

Incubation temperature for obtaining high yield of bacteriocin also vary from strain to strain. Most of the strains which

Table 2 Name of some bacteriocin-producing strains which have been tested against several pathogenic spoilage-causing bacteria by various authors

S. No.	Name of bacteriocin producing strain	Test indicator	Reference
1.	*Leuconostoc carnosum*	*L. monocytogens*	[38, 39]
2.	*L. bavaricus*	*L. monocytogens*	[17]
3.	*L. casei, L. plantarum*	*A. hydrophila, L. monocytogens, S. typhimurium, S. aureus*	[32, 79]
4.	*L. brevis* SB27	*P. pentosus, L. plantarum, L. sake, L. acidophilus*	[25]
5	*L. brevis* MTCC 7539	*S. aureus, L. monocytogens*	[36]
6.	*Lactobacillus* sp.	*L. innocua*	[29]
7.	*L. curvatus* LTH 1174	*L. innocua*	
8.	*L. Plantarum, L. brevis* OG1	*E. coli E. feacalis*	[4]
9.	*L. paracasei, L. pentosus, L. plantarum, L. lactis*	*L. casei*	[30]
10.	*L. lactis*	*Enterococcus, Lactobacilli, Leuconostoc, Pediococcus, Cornobacteria*	[41]
11.	*L. lactis* subsp *cremoris R*	*Clostridium, Staphylococcus, Listeria, Bacillus, Micrococcus, Enterococcus, Lactobacillus, Leuconostoc, Streptococcus*	[42]
12.	*E. faecium*	*L. plantarum L. mesenteroids*	[27]
13.	*Enterococus faeciumL1*	Strains of *Listeria*	[43]
14.	*P. acidilactici*	*E. faecalis*	[44]
15.	*Cornobacterium piscicola CP526*	*Cornobacterium, Enterococcus, Listeria* sp	[18]
16.	*S. warneri*	*L. luteus*	[45]
17.	*B. lentus* NG 121	*L. monocytogens, S. aureus*	[2]
18.	*B. mycoides* MTCC 7538	*L. monocytogens, L. mesenteroides*	[24]
19.	*Brevibacterium linens 9175*	*L. monocytogenes*	[20]

have been isolated so far showed bacteriocin production at temperature 30°C to 37°C. However, strain D53 isolated from vegetables has been found to produce bacteriocin from 10°C to 37°C [41]. *Brevibacterium linens* produce bacteriocin at 25°C than at 30°C and no significant growth or production was observed at 37°C. Similarly high amount of bacteriocin of *L. sake* was produced at 25–30°C while decline in production was observed at 33.5°C followed by nil production at 34.5°C or higher [63]. Bacteriocin production of *L. plantarum* Y21 was obtained at 30°C. In milk, bacteriocin was produced during incubation at 37°C or under temperature gradient reproducing at first 24 h of soft cheese manufacture [52]. So it could be concluded from the study of different authors that the best temperature range for bacteriocin production is 30°C to 37°C.

Purification and characterization of bacteriocin

Released from producer cells, bacteriocins either are adsorbed to the cell wall or remain free in the medium depending upon the pH. In the purification process removal of cells by centrifugation and precipitating bacteriocins from the supernatant liquid using a saturated solution of ammonium sulfate. Instead of ammonium sulfate, trichloroacetic acid, ethanol, methanol and acetone has also been used for extraction of bacteriocin but most of the scientist have precipitated bacteriocin with ammonium sulphate. During this process co-precipitation of other protein in supernatant can also occur therefore to remove contaminant proteins from the supernatant. Initially 25% to 30% ammonium sulfate precipitation is suggested followed by saturated ammonium sulfate precipitation of bacteriocins. In addition ethanol can also be used to precipitate bacteriocins. Regardless of the method used for precipitation precipitate collected by centrifugation and extensive dialyzed against deionized water using 1000 mol wt cut-off dialysis tubing. Centrifugal speed to extract bacteriocin vary from strain to strain. Different workers working on bacteriocin extracted bacteriocins at different centrifugal force. Recovery of different bacteriocin at different centrifugal speed given in Table 3. Activity of bacteriocin-producing strain is determined by well diffusion assay/bit disc method widely but at times the results may be misleading because of different factors viz, aggregation, non-diffusible bacteriocin, protease inactivation and concentration effects. Some bacteriocin producing strains which showed positive activity with agar spot test/bit disk method/cross streak method have shown negative results with well diffusion assay [62].

Table 3 Recommended centrifugal speed for bacteriocin extraction

Name of strain	Centrifugal speed	References
L. brevis	20000xg 30 min at 4°C	[37]
B. mycoides	20000xg30 min at 4°C	[24]
L. amylovorous DCE4712	5500g 30 min at °C	[47]
Leuconostoc MF215 B	10000xg 15 min at 4^0C	[46]
C. piscicola 213	15000xg 30 min at 4^0C	[64]
P. acidilactici	13000 rpm 30 min at 4°C	[31]
L. sakelb674	7000xg 20 min4°C	[21]
*P. acidilavtici*TTV26	5000xg 15 min at 4°C	[61]
B. lentus NG121	20000xg 60 min at 4°C	[2]

Production of bacteriocin during growth cycle

Maximum bacteriocin production can be obtained during late exponential and early stationary growth phase [65]. Production has also been reported at mid log phase in case of *B. thuringiensis* [57] while *B. lentus*, [2] *B. mycoides* [24] and *L. brevis* [37] showed maximum production at end of the logarithmic phase.

Mode of action

Most of the bacteriocins are bactericidal with some exceptions Leucocin A UAL 187 being bcteriostatic [66]. Inhibitory activity of bacteriocin producing strains are mostly confined to gram-positive bacteria. Bacteriocins are bactericidal to sensitive cells. Death of indicator occur rapidly at a very low concentration. Sensitivity of gram-positive and sensitivity of gram-negative bacteria towards bacteriocins has been demonstrated on the basis of cell wall composition. It has been observed that gram-negative bacteria become sensitive towards bacteriocin action if it is destabilized by physical or chemical stresses [58]. Gram-negative bacteria possess an additional layer so called outer layer which is composed of phospholipids proteins and lipopolysacharides and this layer is impermeable to most molecules. Presence of porin in this layer allow free diffusion of molecular with a molecular mass below 600 Da. The smallest bacteriocin produced LAB are approximately 3 kD and too large to reach their target cytoplasmic membrane [67]. It has been proposed that Nisin act on cytoplasmic membrane of gram-positive bacteria to cause lesions [68]. Following nisin treatment whole or intact sensitive cells and membrane vesicles exhibit efflux of amino acid and cations. Lose of these substances depletes proton motive force, which ultimately interferes with cellular biosynthesis. These events result in collapse of membrane potential and ultimately cause cellular death [69]. Similarly, other bacteriocins such as Lactococin A, Pediocin J D, etc. have also been reported to cause dissipation of the membrane potential and increase in membrane permeability to ions leading to collapse of proton motive force [70, 71].

Use of bacteriocin as food biopreservatives

Recent interest to isolate bacteriocin producing strains is due to its effectiveness against food spoilage/pathogenic bacteria and also due to its proteinaceous nature which made it safer for human consumption. It is assumed to be degraded by protease in gastrointestinal track [1]. Digestive enzymes rapidly inactivate bacteriocin and consequently it can not alter bacterial microflora in the intestinal track. Many regulatory agencies have advocated the use of hurdle concept, i.e. combining several physical and chemical methods for preservation in sub optimal levels to control microbial growth. *Pediocin* when combined with low dose irradiation showed a greater inhibitory effect on growth of *L. mesenteroides* [72]. Till today many food scientists have advanced packaging techniques to incorporate antimicrobial agents into food products to control microbial growth and enhance safety and shelf life of food products.

Different methods of applications of bacteriocin in different food items

Bacteriocin have been incorporated into different food items by following various techniques such as

1. Direct soaking food items into bacteriocin solution [72].
2. Using polyethylene-based plastic films and edible cellulosic films [73, 81].
3. Adsorption of bacteriocin on different surfaces such as polyethylene, ethylene vinyl acetate, polypropylene, polyanile, polyester, acrylic, polyvinyl chloride and salanized silica etc. [68].
4. Antimicrobial casing containing bacteriocin preparation also aid preservative effect [56].
5. Bacteriocin producing LAB cultures can be used in hurdle technology strategies to reduce food borne disease [74].

Combination of two different bacteriocin can also be used to increase shelf life of food eg two bacteriocins named nisin and pediocin PA-1/ACH were used in combination to prevent spoilage in dairy, meat and fish foods. Instead of using combination of two bacteriocins some workers have successfully combined nisin with lysozyme or nisin with some lactate against food spoilage bacteria. Bacteriocin preparation can be used at 1–2% level and bacteriocin activity should kept below 5000 AU/g or ml of food [56].

More advanced genetic methods provide an excellent advantage for improving shelf life of cheese. Inhibition of spoilage, pathogenic bacteria was attained by introducing bacteriocin genes into cheese starter culture by conjugation [75]. In one study hot dogs were inoculated with two bacteriocin based on preservatives BPI ie, (mixture of Pediocin ACH and nisin A) and BP2 ie, (BP1+bacterial metabolites (organic acid) and then challenged before vacuum packaging with two common processed meat spoilage bacteria, *L. mesenteroides* and *L. viridesans*. During 9 weeks of storage at refrigeration temperature, both bacterial strains grew beyond spoilage detection levels I in the control samples but not in BP treated samples. In other studies, viability and growth of three gram-negative pathogens especially spores of *C. botulinum* in beef products at 25°C were reduced by BP preparation. Similar studies were also conducted by preparing processed meat products using BP as an ingredient in the raw material and inoculating the finished products with spoilage bacteria *L. mesenteroides.* During storage the growth of *L. monocytogenes* was controlled in the product prepared with bacteriocin preparation. These results clearly indicate that bacteriocin based products have the potential to control spoilage and pathogenic bacteria and ensure safety to enhance shelf life of food products [58].

Effect of bacteriocin in comparison to chemical preservative has been studied in milk, cheese, apple juice. Bacteriocin of *L. brevis, B. mycoides* showed encouraging results in all treated food items [76].

In one of the study immersion solutions of enterocin AS-48 alone or in combination with chemical preservatives was applied on fresh green asparagus, alfalfa and soyabean sprouts to inhibit growth of *L. monocytogenes* CECT 4032. [77].

The combination of nisin (25µg/ml) with 1%H_2O_2, 1% sodium lactate and 0.5% citric acid and was found to reduce the contamination of *L. monocytogenes* and *E. coli*. This mixture has been recommended as a washing treatment to decontaminate melon surfaces and hence to improve the microbial safety and quality of fresh cut melon [78].

Though bacteriocins play a major role in food industry, these preparations have been also applied in sugar processing, seed treatment, antibacterial cream, cosmetics, mouth wash and toothpaste etc. [56].

Thus we may conclude from the study of various authors that, that day is not far off when chemical preservative will be replaced by biopreservative completely or partially in the food ultimately providing safer and healthier food to the consumer and leading to a revolution in the food industry.

References

1. Cleveland J, Chiknids M and Montiville TJ (2002) Multimethod assessment of commercial nisin preparations. J Industrial Microbiol Biotech 29:228–232
2. Sharma N, Kappor G and Neopaney B (2006) Characterization of a new bacteriocin produced from a novel isolated strain of *Bacillus lentus* NG121. Antonie Van Leeuwenhoek 89:337–343
3. Cherif A, Quzari H, Daffonchio D, Cherif H, Bem SK, Hassen A, Jaaua S and Boudabous A (2001) Thuricin 7: a novel bacteriocin produced by *Bacillus* thuringiensis BMG-17, a new strain isolated from soil. Lett Appl Microbiol 32: 243–247
4. Ogunbanwo ST, Sanni AI and Onilude AA (2003) Characterication of bacteriocin produced by *Lactobacillus plantarum* F1 and *Lactobacillus brevis* OG1. African J Biotech 2(8): 219–227
5. Federal R (1988) Nisin preparation affirmation of GRAS status as a direct human ingredient. Food Registration 54: 11247–11251
6. Hirch A, Griensted E, Chapman HR and Mattick A (1951) A note on the inhibition of an anaerobic spore former in Swiss type cheese by a nisin producing *Streptococcus*. J Dairy Res 18:205
7. Delves BJ (1990) Nisin and its use as food preservative. Food Technol 44:110–117
8. Thomas LV and Wimpenny JWT (1996) Investigation of the effect of combined variation in temperature, pH, NaCl concentration on nisin inhibition of *L. monocytogenes* and *S. aureus.* Appl Environ Microbiol 62:2006–2012
9. Chen H and Hoover DG (2003) Bacteriocins and their food application. Comprehensive Rev Food Sci Food Safety 2: 82–100
10. Holo H, Fye T, Brede DA, Nilsen T, Odegard I, Langsrud T, Brendehaug J and Nes I F (2002) Bacteriocin of propionic acid bacteria. INRA, EDP Sciences 82:59–68
11. Kim and Bhowmik SR (1990) Survival of lactic acid bacteria during spray drying of plain yoghurt. J Food Sci 55: 1008–1010
12. Mauriello G, Aponte M, Andolfi R, Moschetti and Villani F (1999) Spray drying of bacteriocin producing lactic acid bacteria. J Food Protection 62 (7):773–777
13. Klaenhammer TR (1993) Genetics of bacteriocin produced by lactic acid bacteria. FEMS Microbiol Rev 12:39–86
14. Rodriguez JM, Cintas LM, Casaus P, Suarez A and Hernandez PE (1995) PCR detection of the Lacatocin S. structural Gene in bacteriocin-Producing *Lactobacilli* from Meat. Appl Environ Microbiol 61(7):2802–2805
15. Dugas B, Mercenier A and Lenoir-Wijnkoop I (1999) Immunity and probiotics. Immunol. Today 20:387–390

16. Ganzle MG (2004) Reutericyclin: biological activity, mode of action and potential application. Appl Microbiol Biotech 64:326–332
17. Larsen AG, Vogensen FK and Josephen J (1993) Antimicrobial activity of Lactic acid bacteria isolated from sour doughs: purification and characterization of bavaricin A, a bacteriocin produced by *Lactobacillus bavaricus* M1401. J Appl Bacteriol 75:113–122
18. Herbin S, Mathien F, Brule F, Branlant C, Lefebvre G and Lebrihi A (1997) Characteristics and genetic determinants of bacteriocin activities produced by *C. piscicola* CP5 isolated from cheese. Curr Microbiol 35:319–326
19. Martinez R and Martinis E (2006) Effect of *Leuconostoc mesenteroides* 11 bacteriocin in the multiplication control of *Listeria monocytogenes* 4b. Ciênc Tecnol Aliment 26 (1): 52–55
20. Motta AS and B Nandelli A (2003) Influence of growth conditions on bacteriocin production by *Brevibacterium linens*. Appl Microbiol Biotech 62:163–167
21. Holck AL, Axelsson L and Huhne K (1994) Purification and characterization of Sakacin 674 A-A bacteriocin from *Lactobacillus sake* Lb 674. FEMS Microbiol Letters 115: 143–150
22. De martinez ECP, Pubilo MRP, Santarosa PR and Freitas FZ (2001) Antilisterial activity of lactic acid bacteria isolated from vacuum packaged Brazillian meat and meat products. Brazilian J Microbiol 32 (1):32–37
23. Bonade A, Murelli F, Vescovo M and Scolari G (2001) Partial characterization of a bacteriocin produced by *Lactobacillus helviticus*. Lett Appl Microbiol 33 (2):153–158
24. Sharma N and Gautam N (2008) Antibacterial activity and characterization of bacteriocin of *B. mycoides* isolated from whey. Indian J Biotech 7:117–121
25. Benoit V, Lebrihi A, Bernard Milliere J and Lefebvre G (1997) Purification and partial amino acid sequence of breivicin 27, a bacteriocion produced by *Lactobacillus brevis* SB 27. Curr Microbiol 34:173–179
26. Sudirman IM, Michael M and Lafebvre G (1993) Detection and properties of curvaticin 13, a bacteriocin like substance produced by *L. curvatus* SB13. Curr Microbiol 27:35–40
27. Dutta S, Chakraborty P and Ray B (1999) Production, purification and characterization of enterocin Cal 1, a bacteriocin of *Enterococcus faecium* Cal 1, isolated from goat meat. Indian J Microbiol 39:15–22
28. Chin HS, Shim JS, Kim JM, Yang R and Yaan SS (2001) Detection and antibacterial activity of bacteriocin produced by *Lactobacillus plantarum*. Food Sci Biotech 10 (4):335–342
29. Hartnett DJ, Vaughan Aand Van Sindereu D (2002) Antimicrobial producing lactic acid bacteria isolated from raw barley and sorghum. J Institute Brewing 108 (2):169–177
30. Todorov S, Vaz-Velho M and Dicks LMT (2003) Isolation and partial characterization of bacteriocins produced by four lactic acid bacteria isolated from traditional South African beer. Electronic J Environ/Agric Food Chem 2 (4):525–530
31. Kaur B and Balgir PP (2004) Purification and characterization of an antimicrobial range of bacteriocin obtained from an isolate of *Pediococcus* species. Punjab Academy of Sci 1(2):139–144
32. Vescova M, Torriani S, Orsi C, Macchiorolo F and Solari G (1996) Application of antimicrobial producing lactic acid bacteria to control pathogens in ready to use vegetables. J Appl Biotech 81 (2):113–119
33. Kormin S (1998) Isolation and screening of bacteriocin production LAB from temph In: Digitized by web Admin team FSMB, LAB or their metabolic end products as biopreservatives (Radu S, Ali GRR and Jainudesy M R eds.) Putra University, Malaysia
34. Jurez Perez VM, Ferranadis MD and Frutus R (1997) PCR-based approach for detection of novel *B. thuringiensis* genes. Appl Environ Microbiol 63:2997–3002
35. Dffonchio D, Cherif A and Borin S (2000) Homoduplex and heteroduplex transcribed spacers describe relationship in the *Bacillus cereus* groups. Appl Environ Microbiol 66: 5460–5468
36. Bizani D and Brandelli A (2002) Characterization of a bacteriocin produced by a newly isolated *Bacillus* sp. strain 8A. J Appl Microbiol 93:512–519
37. Gautam N and Sharma N (2007) Purification and characterization of a bacteriocin produced by *Lactobacillus brevis* MTCC 7539 isolated from traditional fermented food of Himachal Pradesh. Submitted for publication in Indian J of Biochem Biophy
38. Madigan, M (1997) Brock biology of microorganism. Simon and Scluster: New Jerssey, 5p
39. Balow A (1992) Prokaryotes:Handbook on the biology of bacteria Springer-Verlag, New York 5p
40. Verluyten J, Messens W and Vuyst LD (2004) Sodium chloride reduces production of curvacin A, a Bacteriocin produced by *Lactobacillus curvatus* strain LTH 1174, originating from fermented sausage. Appl Environ Microbiol 70(4): 2271–2278
41. Uhlman L, Schrillinger U, Rupnew J and Holzapfel WH (1992) Identification and characterization of two bacteriocin producing strains of *Lactococcus lactis* isolated from vegetables. Int J Food Microbiol 2:141–151
42. Yildrim Z and Johnson MG (1998) Detection and characterization of a bacteriocin produced by *Lactococcus lactis* subsp. *cremoris* R. isolated from radish. Lett Appl Microbiol 24 (4):297–304
43. Lyon WJ, Olson DG and Murano E A (1995) Isolation and purification of enterocin EL1, a bacteriocin produced by a strain of *Ehterococcus faecium*. J Food Protec 58(8):890–898
44. Calderon M, Mendoza PG, Garcia MA and Escuderu B1 (2001) Effect of physical factors on the production of bacteriocin from *Pediococcus acidilactici* ITV26. J Indus Microbiol Biotechnol 26:191–195
45. Kurniasih D and Ray B (2000) Isolation and characterization of warnerin 20, a bacteriocin produced by *Staphyloccous warneri* FM20. Indian J Microbiol 40:41–47
46. Blom H, Katla T, Holck A, Sletten K, Axelsson L and Holo H (1999) Characterization, production and purification of leucocin H, a two-peptide bacteriocin from leuconostoc MF 215 B. Curr Microbiol 39:43–48
47. Callewaert R, Holo H, Devresse B, Beeumen JV, Nes Iand Vuyst LD (1999) Characterization and production of amylovorin L471, a bacteriocin purified from *Lactobacillus amylovorus* DCE 471 by a novel three-step method. Microbiol 145: 2559–2568
48. Himelbloom B, Nilsson L and Gram L (2001) Factors affecting production of an antilisterial bacteriocin by *C. piscicola*

strain A 96 in laboratory media and model fish system. J Appl Microbiol 91:506–513
49. Bromberg R, Moreno I, Zaganini CL Delboni RR and Oliveira JDE (2004) Isolation of bacteriocin producing lactic acid bacteria from meat and meat products and its spectrum of inhibititory activity. Brazillian J Microbiol l35 (1–2):1–13
50. Geis A, Singh J and Teuber M (1983) Potential of lactic Streptococci to produce bacteriocin. Appl Environ Microbiol 45(1):205–211
51. Laukovo A and Czikkova S (1999) The use of enterocin CCM 4231 in soy milk to control the growth of *Listeria monocytogens* and *Staphylococcus aureus*. J Appl Microbiol 87:182–186
52. Tarelli TG, Carminati D and Giraffa G (1994) Production of bacteriocin active against *Listeria monocytogenes* and *Listeria innocua* from dairy enterococci. Food Microbiol 11(3):243–252
53. Suzuki M, Yamamoto T, Kawai Y, Inove, N and Yamazakk (2005) Mode of action of Piscicocin CS526 produced by *C. piscicola* CS526. J Appl Microbiol 98:1146–1151
54. Sarkar S and Misra AK (2001) Preservation of milk and milk products. Indian Food Industry 20 (4):73–75
55. Elliker PR, Anderson A and Hammersion G (1956) An culture medium for lactic acid Streptococci and Lactobacilli. J Dairy Sci 39:1611–1612
56. Gravesen A, Axelsen A M J, Da Silva JM, Hansen TB and Knochal S (2002) Frequency of bacteriocin resistance development and associated fitness costs in *Listeria monocytogenes*. Appl Environ Microbiol 68 (2):756–764
57. Paik HD, Bae SS, Park SH and Pan JG (1997) Identification and partial characterization of tochicin, a bacteriocin produced by *Bacillus thurieingensis* subsp. *tochigiensis*. J Indus Microbiol and Biotech 19:294–298
58. Ray B, Miller KW and Jain MK (2001) Bacteriocins of lactic acid bacteria: current prospectives. Indian J Microbiol 41: 1–21
59. Skytta E, Haikara A and Mattila STC (1993) Production and characterization of antibacterial compounds produced by *Pediococcus damnosus* and *Pediococcus pentosaceous*. J Appl Bacterio 74:134–142
60. Passos FV, Fleming HP, Ollis DF, Hassan HM, Felder RM (1993) Modeling the specific growth rate of *Lactobacillus plantarum* in cucumber extract. Appl Microbiol Biotech 40: 143–150
61. Santoyo C, Mendoza Garcia PG, Garcia Alvarado MA and Escudero Abarca BI (2001) Effect of physical factors on the production of bacteriocoin from *Pediococcus acidilactici* ITV 26. J Indus Microbiol Biotech 26:191–195
62. Stiles ME and Hasting JW (1991) Bacteriocin production by lactic acid bacteria: potential for use in meat preservation. Trends Food Sci Technol 2: 247–251
63. Diep BD, Axelsson, L, Grefsli, Camilla and Nes, IF (2000) The synthesis of bacteriocin sakasin A is a temperature sensitive process regulated by a pheromone peptide through a three component regulated system. Microbiol 146:2155–2160
64. Khouiti Z and Simon JP (1997) Detection and partial characterization of a bacteriocin produced by *Carnobacterium piscicola* 213. J Industrial Microbiol
65. Schillinger U and Weke FK (1989) Antibacterial activity of *Lactobacillus sake* isolated from meat. Appl Environ Microbiol 55:1901–1906
66. Jeevaratnam K, Jamuna M and Bawa AS (2005) Biological preservation of foods-Bacteriocin of lactic acid bacteria. Indian J Microbiol 4:446–454
67. Abee T, Krockel L and Hill C (1995) Bacteriocin: Modes of action and potential in food poisoning. Int J Food Microbiol 28 (2):169–185
68. Bower CK, Mcgure J and Deschel MA (1995) Influence on the antimicrobial activity of surface adsorbed nisin. J Industrial Microbiol 15:227–233
69. Nettles CG and Barefoot SF (1993) Biochemical and genetic characteristics of bacteriocins of food associated lactic acid bacteria. J Food Production 56:358–366
70. Christenson DP and Hutkins RW (1992) Collapse of the proton motive force in *L. monocytogenes* caused by a bacteriocin produced by *Pediococcus acidilactici*. Appl Environ Microbiology 58:3312–3315
71. Bruno MEC and Montville TJ (1993) Common mechanistic action of bacteriocin from lactic acid bacteria. Appl Environ Biotech 40:143–150
72. Appendini P and Hotchkiss JH (2002) Rev of antimicrobial food packaging. Innov Food Sci Emerg Technol 3:113–126
73. Siragusa GR, Cutter CN and Willett JL (1999) Incorporation of bacteriocin in plastic retains activity and inhibited surface growth of bacteria on meat. Food Microbiol 16:229–235
74. Deegan LH, Cotter PD, Hill C and Ross P (2006) Bacteriocins: biological tools for biopreservation and shelf life extension. Int Dairy J 16:1058–1071
75. Ross P and Hill C (1998) Use of bacteriocin to improve cheese quality and safety. Irish Agri Food Development 3: 1–11
76. Sharma N and Gautam N (2007) Use of bacteriocin as potential biopreservative in milk, cheese and apple juice. Beverage and Food World 34 (7):44–47
77. Mollinos AC, Abrioul H, Ben O N, Valvidia E, Lucas L R, Maqueda M, Martinez CM and Galvez A(2005) Effect of immersion solutions containing enterocin AS-48 on *L. monocytogenes* in vegetables food. Appl Env Microbiol 71: 7781–7787
78. Ukuku DO, Bari ML, Kawamoto S and Isshiki K (2005) Use of hydrogen peroxide in combination with nisin, sodium lactate and citric acid for reducing transfer of bacterial pathogens from whole melon surfaces to fresh cut pieces. Int J Food Microbiol 104:225–233
79. Enan G (2006) Inhibition of *C. perfringenes* LMG11264 in meat samples of chicken turkey and beef by the bacteriocin plantarum UG1. Int J Poultry Sci 5(2):195–200
80. Aly E and Abo A (2007) Characterization of a bacteriocin like inhibitory substance produced by *Lactobacillus plantarum* isolated from Egyptian home-made yogurt. Sci Asia 33: 313–319
81. Settani L, Massittio O, Vansinderen D and Corsetti A (2005). In situ activity of bacteriocin producing *Lactococcus lactis* strain. Influence on the interaction s between lactic acid bacteria during sourdough fermentation. J Appl Microbiol 99:670–681