

## Bacteriocin Producing Probiotic Lactic acid Bacteria

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### Abstract

Human intestinal tract is filled with an enormous number of helpful bacteria called probiotic bacteria. Human bodies are actually designed to have symbiotic relationships with these probiotic bacteria. They help in digestion of food, killing harmful microorganisms and keep the body functioning properly in a number of ways. Lactic acid bacteria including *Lactobacillus leuconostoc*, *lactococcus*, *pediococcus* and *Bifidobacterium* are found throughout the gastrointestinal tract. The beneficial effects of Lactic acid bacteria as probiotic and production of bacteriocin is discussed in this article.

**Keywords:** LAB (Lactic acid Bacteria); Bacteriocins; Nisin; Food preservation

### Introduction

Lactic acid bacteria (LAB) and *bifidobacteria* are the most common types of microbes used as probiotics [1]. Probiotics are commonly consumed as part of fermented foods with specially added active live cultures; such as in yogurt [2], soy yogurt, or as dietary supplements.

Lactic acid bacteria are Gram-positive bacteria, usually non-motile, non-spore-forming rods and cocci which are able to grow both in the presence and absence of oxygen. They have a long history of application in fermented foods because of their beneficial influence on nutritional, organoleptic, and shelf-life characteristics [3]. Lactic acid bacteria cause rapid acidification of the raw material through the production of organic acids, mainly lactic acid [4]. In addition, their production of acetic acid, ethanol [5], aroma compounds, bacteriocins, exopolysaccharides, and several enzymes is of importance (Figure 1). Whereas a food fermentation process with LAB is traditionally based on spontaneous fermentation or backslopping, industrial food fermentation is nowadays performed by the deliberate addition of LAB as starter cultures to the food matrix. This has been a breakthrough in the processing of fermented foods [6], resulting in a high degree of control over the fermentation process and standardization of the end products.

The genus *Lactobacillus* currently consists of over 125 species and encompasses a wide variety of organisms. The species which have been therapeutically used are:

- *L. sporogenes*
- *L. acidophilus*
- *L. plantarum*
- *L. casei*
- *L. brevis*
- *L. delbruckii*
- *L. lactis*

Lactic acid bacteria produce antibacterial compounds that are known as bacteriocins [7]. Bacteriocins act by punching holes through the membrane that surrounds the bacteria. Thus, bacteriocins

activity is usually lethal to the bacteria. Examples of bacteriocins are nisin and leucocin. Nisin inhibits the growth of most gram-positive bacteria, particularly spore-formers (e.g., *Clostridium botulinum*). This bacteriocin has been approved for use as a food preservative in the United States since 1989. Lactic acid bacteria are also of economic importance in the preservation of agricultural crops and in the biodegradation [8,9]. A popular method of crop preservation utilizes what is termed silage. Silage is essentially the exposure of crops (e.g., grasses, corn, and alfalfa) to lactic acid bacteria [10]. The resulting fermentation activity lowers the pH on the surface of the crop, preventing colonization of the crop by unwanted microorganisms.

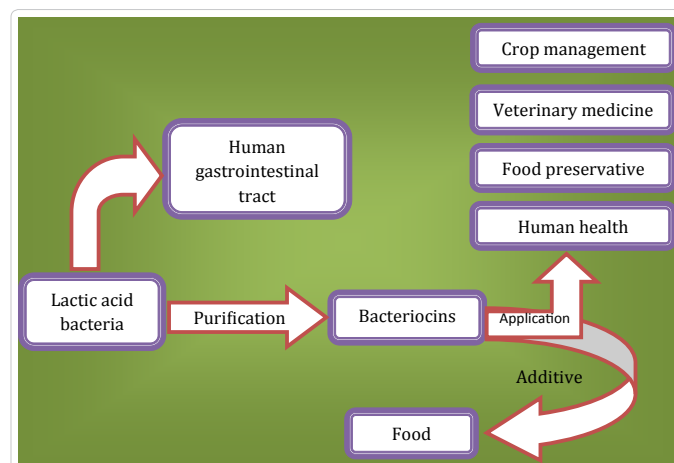


Figure 1: Over view of the applications of bacteriocin produced by LAB.

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Bacteriocins are proteinaceous toxins ribosomally synthesized single polypeptides produced by bacteria to inhibit the growth of similar or closely related bacterial strain(s).

Bacteriocins were first discovered by A. Gratia in 1925. He was involved in the process of searching for ways to kill bacteria, which also resulted in the development of antibiotics [11] and the discovery of bacteriophage [12], all within a span of a few years. He called his first discovery a colicine because it killed *E. coli*. *Escherichia coli* (*E. coli*) belong to a group of pathogenic bacteria called gram-negative enterobacteria that also includes *Salmonella* and *Pseudomonas* [13,14]. *E. coli* and *Salmonella* found in contaminated foods can cause diarrhea, and even death in severe cases [15,16]. Bacteriocins are categorized in several ways, including producing strain, common resistance mechanisms, and mechanism of killing. There are several large categories of bacteriocin which are only phenomenologically related (Table 1). These include the bacteriocins from gram positive bacteria- the colicins, microcins, bacteriocins from Archaea. Besides the production of bacteriocins, some LAB are able to synthesize other antimicrobial peptides that may also contribute to food preservation and safety [17-19].

### Classification of bacteriocins

According to Klaen hammer [20], bacteriocins can be classified into four groups on the basis of to their molecular mass, thermo-stability, enzymatic sensitivity, presence of postrationally modified amino acids, and mode of action.

**Class I bacteriocins:** The class I bacteriocins are small peptide inhibitors and include nisin and other lantibiotics. They are further

divided into two subgroups on the basis of structure and charge of the compound: Group Ia, which consists of screw-shaped, amphipathic, small cationic peptides that produce voltage-dependent pores by unspecific interaction with the membrane of the target cell; and group Ib, which consists of anionic or neutral peptides having a globular shape.

**Class II bacteriocins:** This group comprises heat-stable peptides with molecular masses smaller than 10 kDa and with no modified amino acids. Members of this class can be further sub-classified into four groups: Group IIa consists of anti-listerial peptides showing the consensus sequence YGNGV at their N-terminal sequence. Class IIa bacteriocins have a large potential for use in food preservation as well medical applications, due to their strong antilisterial activity, and broad range of activity. The class IIb bacteriocins (two-peptide bacteriocins) require two different peptides for activity. Other bacteriocins can be grouped together as Class IIc (circular bacteriocins). These have a wide range of effects on membrane permeability, cell wall formation and pheromone actions of target cells.

**Class III bacteriocins:** This group consists of peptidic antibiotics that are heat-labile proteins with a molecular mass larger than 30 kDa.

**Class IV bacteriocins:** This group consists of either glycoproteins [21,22] or lipoproteins [23,24] that require non-protein moieties for their activity.

### Probiotics

Probiotics are live microorganisms thought to be healthy for the host organism. According to the currently adopted definition by WHO, probiotics are: "Live microorganisms which when administered in adequate amounts confer a health benefit on the host" [25,26].

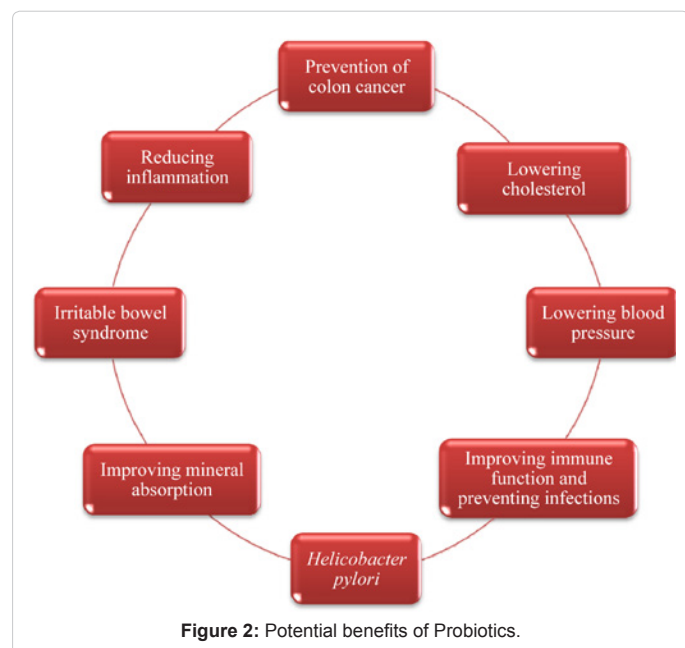
The term "probiotics" was first introduced in 1953 by Kollath [27]. Lactic acid bacteria were referred to as probiotics in scientific literature by Lilley and Stillwell (1965). However probiotic took on a different terminology when Sperti in 1971 used the term "probiotic" to describe tissue extracts that stimulated microbial growth. Parker in 1974 redefined it as organisms and substances that contribute to the intestinal microbial balance [28]. The most recent and accurate description of probiotics was undertaken by Fuller who redefined it as "a live microbial feed supplement beneficial to the host by improving the microbial balance within its body" [29]. Probiotics also appear to enhance the host response towards disease, and improve the quality of the rearing environment [30,31] Benefits of probiotic LAB.

Experiments into the benefits of probiotic therapies suggest a range of potentially beneficial medicinal uses for probiotics [32]. Recent research on the molecular biology and genomics of *Lactobacillus* has focused on the interaction with the immune system [33,34], anti-cancer potential, and potential as a biotherapeutic agent in cases of antibiotic-associated diarrhoea, travellers' diarrhoea, pediatric diarrhoea, inflammatory bowel disease [35] and irritable bowel syndrome. *Lactobacilli* provide the enzyme  $\beta$ -galactosidase which hydrolyzes lactose into lactic acid; it helps in maintaining lactose intolerance (Figure 2).

Most human trials have found that the LAB strains tested may exert anti-carcinogenic effects by decreasing the activity of an enzyme called  $\beta$ -glucuronidase which can generate carcinogens (Heterocyclic amines) in the digestive system [36,37]. *Lactobacilli* deconjugate the bile salts in the intestine to form bile acids and thereby inhibit micelle

Substance	Producing Organism
Curvalicin	<i>Lactobacillus curvatus</i>
Acidocin J1132 $\beta$	<i>Lactobacillus acidophilus</i>
Plantaricin S $\beta$	<i>Lactobacillus plantarum</i>
Bacteriocin J46	<i>Lactococcus lactis</i>
Lacticin 481 (Lactococcin DR)	<i>Lactococcus lactis</i> subsp (Streptococcus lactis)
Lactocin-705	<i>Lactobacillus paracasei</i>
Nisin	<i>Lactococcus lactis</i> subsp (Streptococcus lactis)
Plantaricin C19	<i>Lactobacillus plantarum</i>
Lactocin-S	<i>Lactobacillus sakei</i>
Lactococcin MMFII	<i>Lactococcus lactis</i> subsp (Streptococcus lactis)
Curvaticin FS47	<i>Lactobacillus curvatus</i>
Bavaricin	<i>Lactobacillus sakei</i>
Curvacin-A	<i>Lactobacillus curvatus</i>
Sakacin-A, Sakacin-P	<i>Lactobacillus sakei</i>
Lactococcin-B	<i>Lactococcus lactis</i> subsp (Streptococcus cremoris)
Lactobin-A (Amylovorin-L471)	<i>Lactobacillus amylovorus</i>
Lactacin-F (lafA)	<i>Lactobacillus johnsonii</i>
Plantaricin W $\alpha$	<i>Lactobacillus plantarum</i>
Plantaricin 1.25 $\beta$	<i>Lactobacillus plantarum</i>
Acidocin B (AcdB)	<i>Lactobacillus acidophilus</i>
Reuterin	<i>Lactobacillus reuteri</i>

**Table 1:** Bacteriocins produced by different *Lactobacillus* species.



formation. This leads to decreased absorption of cholesterol [38]. Several small clinical trials have indicated that consumption of milk fermented with various strains of LAB may result in modest reductions in blood pressure. It is thought that this is due to the ACE inhibitor-like peptides produced during fermentation.

Lactic acid bacteria have several beneficial effects on immune function such as protecting against pathogens by competitive inhibition and also improve immune function by increasing IgA producing plasma cells [39], increasing or improving phagocytosis as well as increasing the proportion of T lymphocytes and natural killer cells [40,41]. Probiotics are also known to reduce dental carries in children as well as aid in the treatment of *Helicobacter pylori* infection [42,43].

### Mechanism by which probiotics influence human health

Studies have shown that probiotics can down regulate inflammation and responses to allergens, and interfere with pathogen infections either directly or through enhancing gut barrier function [44]. Probiotics can also serve a role for stabilizing intestinal microflora [45]. Many of the mechanisms are similar to the roles that our normal colonizing microfloras play. Probiotics can down regulate:

- Inflammation [46,47]
- Responses to allergens
- Pathogen infections( interference); and
- Alteration in the gut

### Conclusion

The potential application of bacteriocins as consumer friendly biopreservatives either in the form of protective cultures or as additives is significant. LABs are typically involved in a large number of spontaneous food fermentations. Food fermentations have a great economic value and it has been accepted that these products contribute in improving human health. LABs have contributed in the increased volume of fermented foods worldwide especially in foods containing

probiotics or health promoting bacteria. Bacteriocins produced by LAB are the subject of intense research because of their antibacterial activity against food borne bacteria.

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