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## Antimicrobial Peptides: A New Alternative for Food Preservation

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### Review Article

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#### ABSTRACT

Food contamination is a serious problem related to public health as it causes great morbidity. For that reason, there is a need to develop alternative preservation methods able to provide a better and safer quality of food from a microbiological and toxicology point of view. Thus natural conservatives will have an important role in the near future. Faced with this reality the investigation of antimicrobial peptides which have activity against important pathogenic microorganisms in food remains promising and is the focus of ongoing research. Although there is some knowledge about these peptides, many aspects related with structure and function, biosynthesis and mode of action remain unknown.

### INTRODUCTION

The foodborne diseases (FBDs) are one of the most common problems of public health in the contemporary world. The FBDs are caused by etiological agents, mainly microorganisms and/or their toxins through ingestion of contaminated water and food, resulting in disorders to the health and well-being of affected individuals, as well as serious economic consequences for the country.

Thus, the microbiological safety of foods is becoming increasingly relevant and governments around the world are giving great importance in actions to ensure safe food production in response to the increasing number of FBDs <sup>[1]</sup>.

The epidemiological profile of diseases transmitted by food have changed in the last decade due to: expansion of consumer markets; economic globalization; changes in dietary habits; increased consumption of processed or manufactured foods outside the home; intensification of production, which makes the control of FBDs by public health authorities and the intensive rearing of livestock. According to the Ministry of Health in 2013 called mixed food prevailed as the main cause of the occurrence of FBDs. There were eight cases with this type of food, totaling 1,529 outbreaks. This is probably due to the increase of manipulation and transport of food, which offers new doors for bacteriological contamination and the spread and proliferation of bacteria from the slaughter and marketing process.

Examples of microorganism that cause Foodborne infections in humans are bacteria of the genus *Salmonella* which remains a major problem for the poultry industry. Food intake and water containing viable cells of the bacterium is the most common route of infection for the host <sup>[2]</sup>. Other bacteria frequently involved in human food infections are members of the genus *Listeria* spp. *Listeria monocytogenes* is the only species of the genus *Listeria* that is pathogenic to humans. Listeriosis leads the post of diseases associated with foods that cause more deaths and hospitalizations (91%), mainly involving pregnant women, new born and immunocompromised persons <sup>[3]</sup>. Although listeriosis causes more complications, the main microorganisms responsible for FBDs, between the years 2000-2013, continue to be *Salmonella* spp, *Staphylococcus aureus* and *Escherichia coli* <sup>[4]</sup>.

FBD outbreaks result in a large number of sick people and the "recall" of food products (withdrawal and replacement batches of contaminated food products) can reduce consumer confidence, decreasing the demand for these products resulting in significant economic losses for all parts of the supply chain. Therefore, it is crucial to protect the food and consumer health by adopting good food preservation techniques. Thus, it is of vital importance, the search for natural substances that exhibit specific antimicrobial activities and, above all, that carry alternative mechanisms of action available to the chemical preservatives. Among these substances, antimicrobial peptides (AMPs) emerged as an interesting alternative, since they act by mechanisms whereby pathogens may hardly develop resistance. Also, the AMPs have high specificity to their target cell and can be used in combination with other antimicrobial agents. The AMPs are considered an integral part of innate immunity and the first line of defense against invading microorganisms<sup>[5]</sup>. Most of these AMPs are cationic and hydrophobic and act on the cytoplasmic membrane of target cells, causing them death. However, some of these AMPs have other target sites of action, such as cell wall, hydrolyzing it.

The bacteriocin, a class of AMPs, are being employed in bio preservation of food with the aim of controlling the growth of pathogenic and spoilage microorganisms. For this reason, there is growing interest in the food industry for the potential use thereof. Being described as a natural preservative, its use in food is very promising, since the chemical additives have been seen as villains in the food industry, because of poisoning risks that constant intake of these substances can cause. Also, chemical preservatives, has reduced its inhibitory action due to the continuous emergence of multidrug-resistant microbial strains and are potentially toxic to the human body<sup>[6]</sup>. Despite a variety of compounds as subtilin, cerein, turicina, plantaricin, pediocin, among others, nisin is the only one so far that has FDA GRAS status (United States)<sup>[7]</sup>.

The use of AMPs as an alternative to conventional agents to combat microorganisms of interest for human and animal health has been an interesting alternative. In the unseen world of microbes, the search for nutrients and overpopulation formed by different bodies, leads to several strategies to ensure the survival of a species and its dominion over others. Thus, it is of vital importance, the search for substances that exhibit antimicrobial activity and, above all, that carry through alternative mechanisms of action available to the chemical preservatives.

Bacteriocins are synthesized in ribosome of most bacteria that have the ability to eliminate bacteria closely related<sup>[8]</sup>. They have various chemical structures and their role in innate immunity is essential. These are usually composed of small peptides (3-10 kD), and are inactivated by proteolytic enzymes. It is also known that in a single bacterium, various peptides with these characteristics can be produced. The study of bacteriocins began in 1925 with the discovery of colicins produced by *Escherichia coli*<sup>[9]</sup>. Because of their potential application as a natural preservative, a large number of substances have been identified and characterized, particularly those produced by lactic acid bacteria<sup>[10]</sup>.

There is great interest in the study of these peptides due to its therapeutic potential as antimicrobial and antitumor agents. Because of natural production of bacteriocins by lactic bacteria in fermented foods the addition of antimicrobial peptides as food additives will be more easily accepted by health authorities and consumers. GÁLVEZ et al.<sup>[11]</sup> emphasize that the bacteriocin can be used in the development of bioactive food packaging. Moreover, GÁLVEZ et al.<sup>[11]</sup> emphasized that many bacteriocins showed synergistic effects in combination with other antimicrobial agents, including chemical preservatives, natural phenolic compounds and other antimicrobial proteins, which is relevant when microbial resistance is emphasized. Consequently, the possibility of application of these antimicrobial peptides becomes of great interest to the scientific community, because they are considered safe for either human animal health<sup>[12]</sup>.

The activity of bacteriocins varies according to the bacterial species and the environment, in which they are being divided into different classifications<sup>[13]</sup>.

- **Bacteriocins Class I:** Are usually cationic molecules amphipathic, consisting of 12-45 amino acid residues. Basically, the mode of action of this peptide class is common: they dissipate the proton-motive force to changes in membrane potential and the H<sup>+</sup> concentration gradient (pH), causing the formation of pores in the cytoplasmic membrane<sup>[14]</sup>.

- **Bacteriocins class II:** Have limited spectrum of activity and seem to have its action mediated by some type of receiver<sup>[15]</sup>. According to Moll et al.<sup>[16]</sup> the nature of these "receptors" has not yet been elucidated and could be intracellular enzymes. It is known, however, that the bacteriocins class II also depends of anionic phospholipids for initial interaction with the membrane<sup>[17]</sup>.

- **Bacteriocins of classes III and IV:** Are complex peptides with high molecular weight, thermolabile, further containing glucídicas or lipid moieties that have bacteriolitic activity via mechanisms still poorly understood<sup>[18]</sup>. According to Jack et al.<sup>[19]</sup>, high molecular weight thermolabile bacteriocins include many extracellular bacteriolytic enzymes (muramidases and hemolysin) that can mimic the physiological activity of bacteriocins.

The use of preservatives in food is a recurring topic in public discussion and many consumers associate them to the presence of harmful chemicals in food. However preservatives became necessary for the manufacturing of foods because of the search for food with a longer shelf life boosting studies on the use of natural preservatives. Maciel and collaborators<sup>[20]</sup> studied the intensity of inhibitory activity (IINIB) and bacterial inactivation (IINAB) *in vitro* in two alcoholic extracts obtained from chalice and fruits with seeds of different hibiscus accesses on the bacteria: *Enterococcus faecalis*, *E. coli*, *Salmonella enteritidis* and *S. aureus*. *Salmonella enteritidis* and *E. coli* were the most sensitive bacteria, respectively, the alcoholic extracts of cups of hibiscus and fruits with seeds. *S. aureus* (5.2 and 0.1) was the most resistant to both extracts.

Another example of use of natural compounds for preservation of food was demonstrated by Silva et al. [21], which assessed the antimicrobial activity of essential oil of oregano, with different concentrations, in coating chicken patties during storage. They found that the essential oil of *Origanum vulgare* shown an alternative technique for food preservation systems, as it was effective against coliforms.

Sobral et al. [22] evaluated the effect of addition of different concentrations of nisin on the behavior of two groups of lactic acid bacteria, *Lactococcus* and *Lactobacillus* in cheese artisan Minas Araxá region during the 60 days of ripening. The research result was satisfactory, because it was found that nisin did not interfere in the multiplication of these microorganisms along the maturation of artisanal Minas cheese from the Araxá region. Another research using nisin was done by Giroldo et al. [23], which analyzed the effect of nisin in the inactivation of *Alicyclobacillus acidoterrestris*, microorganism found in unpasteurized juices industrialized and concluded that the use of antimicrobial inhibited bacterial growth and yet their use can be very promising in the processing of fruit juices.

Costa et al. [24] evaluated the antagonism of 30 lactic acid bacteria (LAB) isolated from artisanal rennet cheeses of Pernambuco. 40% inhibited the growth of *Listeria innocua*, 89% were effective in *Enterococcus faecalis* and 100% inhibited the growth of *Bacillus cereus* [25].

## CONCLUSION

Although there are already available various storage technologies, none of them completely ensure the microbiological quality of food. Thus, it becomes evident the need to develop new food preservatives combined with existing technologies, to provide microbiological and toxicological quality for the food and its consumers. In this context, PAMs naturally produced by microorganisms have proven to be of great importance to contribute to the development of new and more efficient natural biopreservatives since these PAMs have high specificity of action mode, distinct mechanism from those of conventional chemical preservatives and probably represents low toxicity comparing to other preservatives.

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