



Review on Application of Antibacterial Peptides in Food Preservation

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Received: 10-06-2023; Revised: 20-08-2023; Accepted: 26-08-2023; Published on: 15-09-2023.

ABSTRACT

Scope and Approach: This review will outline the characteristics, functions, qualities, kinds, and mechanisms of action of AMPs and how they are used in food preservation.

Insight: With evolution of several mutant strains of microorganisms, a rise in the microbial resistance to effective treatments have been observed. Antimicrobial proteins and peptides are generally found in animals, plants, and microorganisms and are of great interest to medicine, pharmacology, and the food industry. These peptides are capable of inhibiting pathogenic microorganisms. They possess the ability to attack parasites, while causing little or no damage to the host cells. Antimicrobial peptides (AMPs) are a class of small peptides that widely exist in nature and they are an important part of the innate immune system of different organisms. AMPs have a wide range of inhibitory effects against bacteria, fungi, parasites and viruses. To synthesise anti-microbial peptides - chemical, enzymatic and recombinant techniques are used. Bacteriocins are particularly grouped as proteins or peptides produced by bacteria. This paper will systematically represent the anti-microbial peptides having the ability to act as natural preservatives and their application in food preservation (i.e. bio- preservation).

Keywords: Antimicrobial peptide, food safety, spores, bacteriocin, food preservation.

INTRODUCTION

A very important aspect of the food industry is the preservation technique which involves certain processes which increases the shelf life of the food. Approximately 1.3 billion tons of food is wasted every year. This represents around 1/3 of all food generated for human consumption is lost due to spoilage which results in a huge economic loss. Contamination can occur at any point during processing, storage, transport or distribution. To avoid spoilage in most cases chemicals are used for preservation. Chemical use can bring about deterioration of human health and lower down the nutritive value of food. Chemicals like sodium nitrate and potassium bromate have shown carcinogenic properties. That's where antimicrobial peptides come into play with an edge over the chemical preservatives due to their ability to preserve food without degrading its quality and minimizing the toxic effects. It is crucial to find microbes for bio preservation to enhance food safety and standardization with the aim to reduce the rising levels of food spoilage.

History

Alexander Fleming is believed to have discovered the first antimicrobial peptide. In the late 1920s, Fleming identified Lysozyme as an antimicrobial peptide and laid the foundation of modern innate immunity. Bactericidal activities were observed by Fleming when he treated bacterial culture plates of a person having acute coryza. Therefore he termed it as lysozyme activity because of its capacity to 'lyse' bacterial cells. At first the structures of two AMPs named cecropins A and B were characterized

from the haemolymph of *Hyalophora cecropia* (cecropia silk moth). Tyrocidine, an antimicrobial peptide, was discovered to have activity against both Gram-positive and Gram-negative bacteria in 1941. Defensin, an antibacterial peptide, was first isolated from rabbit leukocytes in 1956. *Xenopus laevis* produced the first anionic antimicrobial peptide in 1990.

Nisin

One example of an antibacterial protein involved in food preservation is called Nisin. Nisin is a natural peptide produced by certain strains of the bacteria *Lactococcus lactis*. It has been used as a food preservative for over 50 years and is recognized as safe by regulatory agencies such as the U.S. Food and Drug Administration (FDA).

Nisin has strong antibacterial properties and is effective against a wide of harmful bacteria such as *Listeria*, *Staphylococcus* and *Streptococcus*. It works by disrupting the cell membranes of these bacteria, preventing their growth and multiplication.

Nisin is commonly added to a variety of foods including cheese, meat, canned foods, and beverages. Its use as a preservative allows for longer shelf-life and helps to prevent spoilage and foodborne diseases.

Pediocins

Pediocins are classified as small unmodified peptides belonging to subclass IIa of bacteriocin. They have low molecular weight (2.7-17 kDa) and are produced by some *Pediococcus* bacteria. Pediocins show a wide range of antimicrobial activity against Gram- positive bacteria

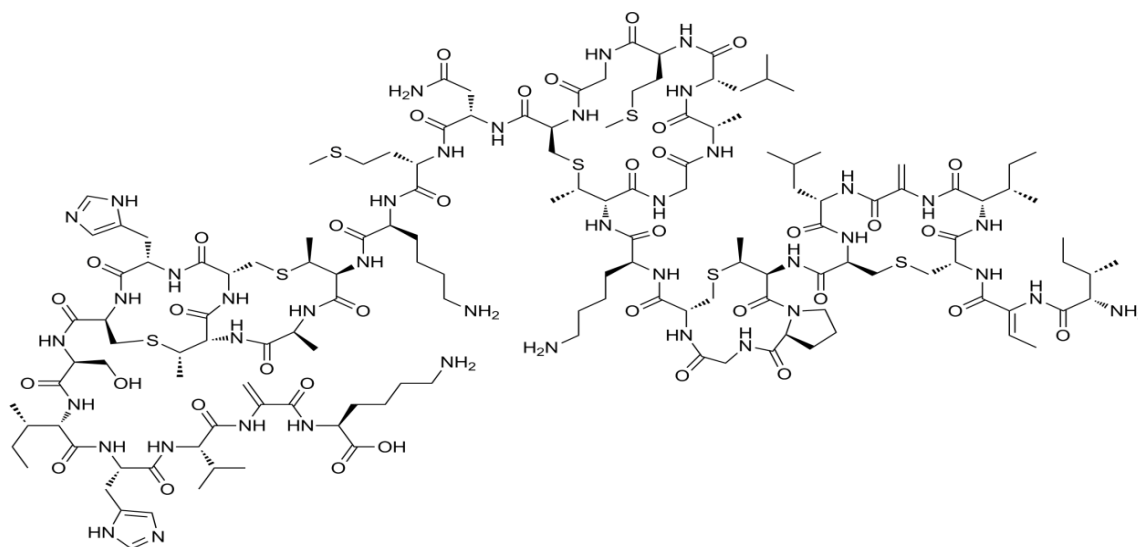


which are particularly pathogenic in nature such as *Listeria monocytogenes* by forming pores in the membrane of the cytoplasm. It is capable of exercising its antimicrobial activity at very low temperatures (-70 C) and are resistant to heat. They can withstand the action of

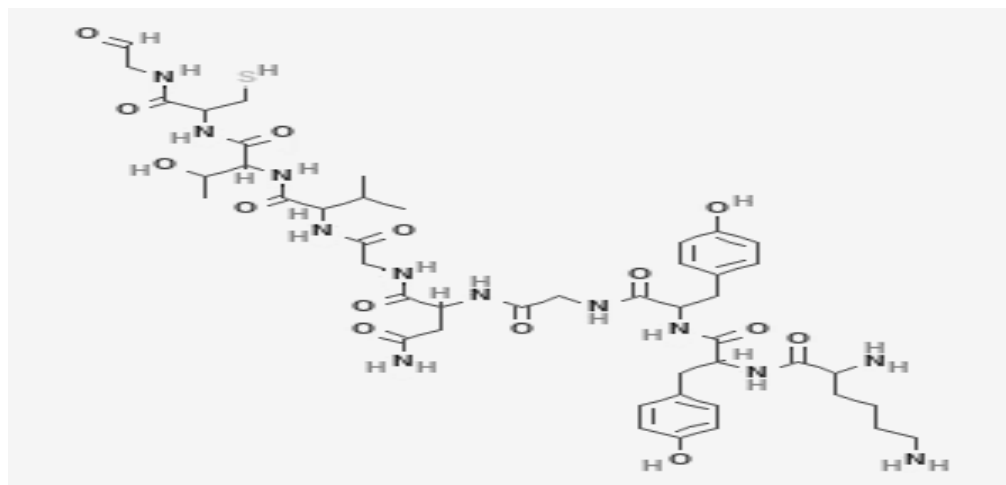
RNase, DNase, lipase and lysozyme. The most common pediocins which are hydrophobic and heat resistant are obtained from the strains of *P. acidilactici*, *P. damnosus*, and *P.pentosaceus*.

Sources of Antimicrobial Peptides

Sr. No	Source of Antimicrobial Peptide	Name of Antimicrobial Peptide	Function
1.	Insect	Cecropin A	It has promising action against fungus.
		Attacins	They constitute an active form of inducible immune protein P5.
		Pyrrhocoricin	It promotes ATPase activity.
		Defensins	Bactericidal activity against multi-drug resistant strains of <i>Pseudomonas</i> .
2.	Milk	β -casein	Production of extracellular enzymes to kill pathogens
3.	Amphibians	Magainins	Cidal activity against gram- positive and gram-negative bacteria, fungi and protozoa.
4.	Bacteria	Pediocin, Nisin	Antibacterial activity against pathogenic bacteria such as <i>Listeria monocytogenes</i>



Structure of Nisin



Structure of Pediocin

Mechanism of Antimicrobial Peptides

The mode of action in antimicrobial peptides generally follows the interactions which happens between the peptides and the cellular membranes, constituents and its composition. Membrane permeability plays a major role in these processes because they can lyse the cells on entering. Few models have been shown for better understanding of the mechanism of antimicrobial peptides.

Barrel-Stave model

Arrangement of antimicrobial peptides around the bacterial membrane in the form of a bundle occurs in this model. The first step of this model involves peptide binding with the membrane as it happens in a monomer. The phospholipid layer of bacterial membrane which consists of hydrophobic chains of fatty acids attaches with the positive terminal of the peptide. Clustering of peptides lead to the pore formation. In the final stage, the bacterial cell dies.

Carpet model

It is also known as detergent model. In this model, a parallel accumulation of peptides around the microbial membrane is observed which leads to membrane destabilization. When the clustered peptides gain maximum intensity, the lipid membrane of bacteria gets penetrated by the molecules. This results in the disruption of microbial membrane.

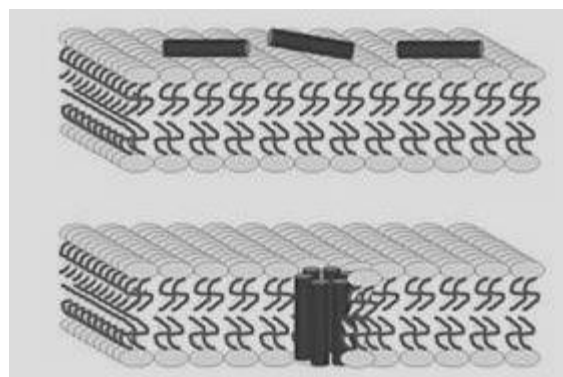
Aggregate model

As a result of electrostatic interaction between the hydrophilic regions of the peptides and the phospholipid layers of the membrane, the peptides are observed to get attached in this model. Aggregates with sphere like structure contains water molecule which release fluids resulting in membrane disruption. Antimicrobial peptides inhibit translational and transcriptional processes by binding onto the cell wall, nucleic acids, etc. They interact with lipid bilayers and form pores.

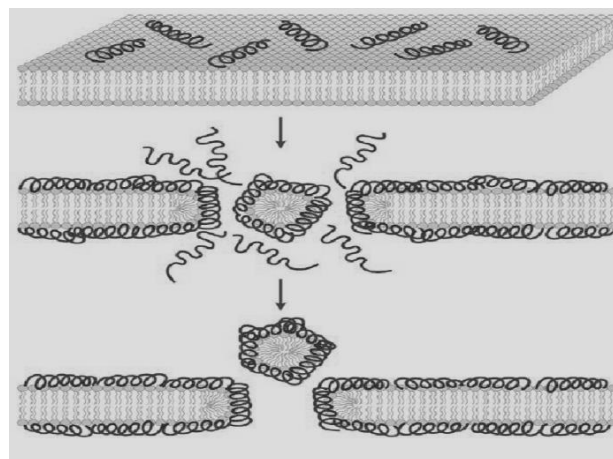
Bacterial Spores inactivated by AMPs

Bacterial spores are widely distributed in the environment and can contaminate food at various stages of manufacturing. Due to their adaptability, spores and spore-forming bacteria must be controlled in food since their survival in processed foods and subsequent germination under favourable conditions would reduce shelf life, result in food deterioration, and cause food poisoning. Foodborne infections caused by toxin production, particularly in canned goods, in addition to food deterioration. For example, the emetic, exo, and neurotoxins produced by *Bacillus* and *Clostridium* species are known to cause certain symptoms brought on by eating contaminated foods. After the germination of the spores and throughout growth, vegetative cells typically produce bacterial toxins in *Clostridium* and *Bacillus* sp. Therefore, it is sufficient to prevent the spores of certain bacteria genera from germinating in foods rather than

completely eradicate them in order to ensure food safety. Food products can be heated to a certain temperature to eliminate the spores but that is always not beneficial. Some food products are heat-sensitive which means if they are exposed to high temperatures, their quality may be degraded and the nutritious value can come down. So that's where antimicrobial peptides help us. AMPs are added directly onto the food so that they can synthesize certain enzymes or generate some processes which would inhibit the pores without actually reducing the nutritious value.



Barrel-stave model



Carpet model

Benefits of AMPs

Increase in health problems depicts that food preservation is a serious issue which needs to be resolved. The bacteriocin is a very safe option due to the following reasons –

- They do not actively interact on coming in contact with protease enzyme.
- They are resistant to heat and can maintain pH.
- They do not have eukaryotic cells.
- They are capable of destroying a wide range of food spoiling pathogens,
- They are very susceptible to be used for their dual nature of being protease sensitive in the gastrointestinal tract but very efficient in pathogen handling.

Limitations and drawbacks

Isolation of several number of AMPs have taken place from various natural sources still date, but very few among them have proven to be suitable for commercial use. The reason behind the successes of only few naturally occurring peptides when used commercially is due to the – (a) toxic effects experienced by the host cell, (b) incapable to remain active at physiological concentrations of salt, (c) very sensitive to the action of protease degradation due to its short half-life, (d) appropriate techniques are not available for focusing on the AMPs delivery at the exact site, (e) change in pH.

CONCLUSION

With time the microbial population is gaining resistance against the pre-existing antibiotics. The AMPs act as remedial agents due to their ability to slow down the rate at which microbes build resistance against the existing antibiotics. The AMPs can be used alone or they can be combined with selected conventional antibiotics against the specific microbial strains which are known to be drug resistant. With proper knowledge about the mode of action of AMPs, new synthetic AMPs can be designed which might be used to target particular strains of microbes. Further development may lead to the creation of certain AMPs to minimize the cytotoxic effects and resist protease activity.

Future Aspects

Huge amounts of food are lost globally due to the spoilage by pathogenic microorganisms. The existing techniques to control the food wastage has become a challenge that needs to be resolved. The AMPs can have a great impact on this global issue. To witness the absolute ability of the AMPs, further studies need to be conducted relating to the development of AMPs to have a better understanding about their activities and interactions. Various biological and chemical procedures can be used to assess the structures and functions of the AMPs. Solid Phase Peptide Synthesis can be very effective. AMPs can be used as therapeutic agents for their ability to prevent microbial growth in food, but they can have cytotoxic effects which may be harmful for humans. Therefore, there is a need to isolate and design few particular AMPs in which the toxins would be absent and some desirable characteristics can be incorporated. Nisin and Pediocin are the only two antimicrobial peptides that have been used in food preservation, so there is a need to isolate and characterize other AMPs which might have similar influence in the field of food preservation.

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Source of Support: The author(s) received no financial support for the research, authorship, and/or publication of this article.

Conflict of Interest: The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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