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Review

Application of bio-preservation to enhance food safety: A review

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Abstract Consumers and industry experts frequently have negative perceptions of most chemical preservatives. Although most people concede that they cannot resolve global food waste issues without preservatives, they prefer products without chemical preservatives. Numerous emerging technologies is now surpassing conventional methods for mitigating microbial food deterioration in response to consumer demand and fundamental health and safety considerations, including biological antimicrobial systems such as using food-grade microorganisms and their metabolites primarily originating from microorganisms, plants, and animals. Microbial compounds, including bacteriocins, bacteriophages, and anti-fungal agents, plant extracts such as flavonoids and essential oils; and animal-originated compounds, such as lysozyme, chitosan, and lactoferrin, are considered some of the major bio-preservatives. These natural compounds can be used alone or with other preservatives to improve food safety. Hence, the use of microbes or their metabolic byproducts to extend the shelf life of foods while maintaining safety standards is known as bio-preservation. To manufacture and consume foods in a safe condition, this review primarily aims to broaden knowledge amongst industry professionals and consumers regarding bio-preservation techniques, bio-preservatives, their classifications, and distinctive mechanisms to enhance food safety.

Keywords bio-preservation, bio-preservatives, food safety, food quality, spoilage

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1. Introduction

Sustainable development, which seeks to protect the health and well-being of people, animals, and the environment, places high priority on ensuring food safety (FAO, 2022). There is an increasing demand for high-quality food products (Garvey, 2022). Food preparation, processing, and preservation in a manner that diminishes the chances of humans contracting foodborne illnesses are referred to as “food safety.” (Australian Institute of Food Safety, 2019). This is an essential aspect of food handling and production because it ensures consumers’ well-being and safety from food-related complications. The wide-scale development of industries, proliferation of food trades, and dramatic changes in food manufacturing patterns are some of the major issues related to food safety in the 21st century

(Fung et al., 2018). In addition, the emergence of novel microorganisms and diseases, unfavorable environmental changes, bacterial and fungal toxins, high amounts of chemical preservatives, and antibiotic resistance among foodborne pathogens are major challenges that must be overcome (Rajanikar et al., 2021).

Foodborne pathogens, including bacteria, viruses, fungi, yeasts, and parasites, are primarily responsible for these illnesses. Foodborne pathogens are the microorganisms that cause outbreaks of foodborne illnesses (Center for Food Safety and Applied Nutrition, 2019). Foodborne disease occurs when a toxigenic microbe proliferates itself in a food product and produces a toxin that is then consumed by the human host or when disease carrying microbes are ingested along with food and settle (and often multiply) in the human host (Bintsis, 2017). *Clostridium botulinum,* Norovirus, *Toxoplasma gondii, Escherichia coli O157:H7, Salmonella, Campylobacter, Listeria monocytogenes*, *Staphylococcus aureus, Vibrio vulnificus,* and *Shigella* are the most common pathogens in the food industry (Bintsis, 2017). Chemical compounds, heavy metals, and the excessive addition of chemicals also create acute food safety problems (Gizaw, 2019).

Consequently, in accordance with the most recent information provided by the World Health Organization, food safety has become an issue that needs to be emphasized even more. According to their estimates, after consuming unhygienic foodstuffs, at least 600 million individuals worldwide are infected with foodborne illnesses (WHO, 2020) and at least 420,000 people die annually. More than 200 diseases, ranging from cancer to diarrhea, are caused by unsafe foods containing pathogenic bacteria, viruses, parasites, or artificial chemicals (Gizaw, 2019). This fosters a vicious cycle of disease and malnutrition that disproportionately affects the elderly, sick, and young children (WHO, 2019).

Thus, there is an urgent need to discover potential solutions to counteract the harmful consequences of food safety issues. To ensure safer food manufacturing processes, it is crucial to stop the growth of pathogenic and spoilage microorganisms and reduce the need for chemical preservatives (Amit et al., 2017). In light of this, preservation techniques play an important role in the food industry as the inclination of consumers toward minimally processed and chemically free food products is constantly increasing (Rajanikar et al., 2021). Diversified preservation techniques are used to store food products and reduce food waste globally, in contrast to chemical preservation (Yu et al., 2021). Food preservation is the process of treating foods in a way that stops and considerably slows down food deterioration and prevents possible diseases, while preserving nutritive value and sensory attributes (Sridhar et al., 2021). Common compounds used to preserve food include salt, sugar, vinegar, butylated hydroxyl anisole, nitrites, citric acid, tert-butylhydroquinone, calcium propionate and butylated hydroxy toluene. In contrast, food microorganisms are more difficult to remove with chemical preservatives, and health concerns related to chemical substances are proliferating simultaneously (Yu et al., 2021). Therefore, there is an immediate need to develop more appropriate preservation techniques that can safeguard food production and consumer health.

The use of bio preservative methods has proven to be an effective solution to several food-related problems (Yusuf, 2018) and can help meet customer demands for “healthier” and more “natural” foods. In addition, bio-preservation can lower the amount

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of salt, sugar, and chemicals in foods by utilizing microorganisms or their metabolic by-products (Borges et al., 2022). Extending the shelf life of foods by using controlled or natural microbiota or antimicrobials is known as bio-preservation (Singh, 2018). Different bio-preservatives have unique mechanisms for targeting organisms in food. Therefore, it has been successfully applied to various foods, including seafood, dairy products, vegetables and fruits, raw and processed meat, cereal products, and non-fermented plant products (Mohammed et al., 2020). In addition to being economically advantageous, bio-preservation methods can improve the sensory and nutritional attributes of food and its microbiological stability and safety. One of the main obstacles to the widespread application of bio-preservation is the complexity and diversity of food microbial populations and food matrices (Borges et al., 2022). The aim of this review was to narrow the knowledge gap concerning how food preservation techniques can improve food safety and reduce risk of food borne illnesses. In particular, it provided reasons for the use of bio-preservation methods over other techniques to improve food safety during production and summarized the types of potential bio-preservatives of different origins that can be used and how they may ensure safer food production with their unique mechanisms in food systems.

2. Bio-preservatives and their classification based on origin

Numerous microorganisms, plants, animals, and other natural resources have antimicrobial characteristics; hence, they can be used as bio-preservatives to eliminate unnecessary physical and chemical processing of foods, while ensuring food safety (Saeed et al., 2019). Several types of foods contain natural preservatives, including lysozyme in egg whites, the lactoperoxidase system in raw milk, chitin and chitosan in arthropod shells, plant-origin antimicrobial compounds in spices and herb extracts, secondary metabolites in microorganisms, such as bacteriocins, antifungal compounds, and antiviral metabolites (Teshome et al., 2022). These bio-preservatives work together to inhibit and eradicate harmful microbial colonies as part of the host's defense system against microbial diversity problems (Quinto et al., 2019). According to Vaishali et al. (2019) and Mani-López et al. (2018), there are three basic categories of bio-preservatives based on their origin: microorganisms, animals, and plants.

2.1. Bio-preservatives derived from microorganisms

When considering bio-preservatives that have originated from microorganisms, bacteriophages including myoviridae and siphoviridae; bacteriocins such as nisin, reuterin, pediocins, lacticins, and endolysin; and antifungal compounds like natamysin stand out (Singh, 2018). These can be applied to food systems as an additive, a direct ingredient, or a protective culture.

Bacteriocins function as inhibitors or toxins against other related or unrelated microorganisms. Recently, bacteriocins have gained interest for their potential use as safe food preservatives, as they are readily digested by the human gastrointestinal tract (Huang et al., 2021).

According to Yusuf (2018), four groups of bacteriocins are derived from gram positive bacteria, viz., class Ⅰ (sub classes Ⅰa and Ⅰb), class Ⅱ (sub classes Ⅱa, Ⅱb, Ⅱc, Ⅱd), class Ⅲ, and class Ⅳ. Class Ⅰ bacteriocins (also called Lantibiotics) have an unusual thioether amino acid present in their structure as a result of post-translational changes composed of the cyclization and dehydration of certain amino acid sequences. Nisin, epidermin, and gallidermin are examples of bacteriocins belonging to subclass Ia. Subclass Ib is composed of negatively charged, globular, and inflexible protein structures. Lacticin 481, cytolysin, and salivaricin belong to the Ib subclass (Simons et al., 2020; Zarrilli et al., 2023).

Class II bacteriocins are membrane-active peptides smaller than 10 kDa. They maintain their morphology, structure, and functionality even under extreme heat. Helveticin is an example of a class Ⅲ bacteriocin and is a heat-sensitive protein of size >30 kDa. Lacticin, reuterin, sakacins, and enterocins are class Ⅳ bacteriocins that have complex structures that contain non-protein components, such as carbohydrates and essential lipids (Simons et al., 2020).

A bacteriophage, or simply a "phage," is a type of virus that can infect and eradicate bacteria. They can be employed to eliminate foodborne pathogens, thereby preventing food spoiling. Their use has increased because they are host-specific (Singh, 2021). According to Zinke et al. (2022), there are three groups of bacteriophages: Myoviridae (with a contractive tail), Siphoviridae (with a noncontractile tail), and Podoviridae (with a very short tail). They can selectively control bacterial species and are, therefore, effectively used to ensure food safety (Endersen and Coffey, 2020). Endolysins are also called phage lysins and contain peptidoglycan hydrolases, which are responsible for the degradation of the peptidoglycan layer by the enzymatic action of the target bacterium during food preservation (Harper et al., 2021).

2.2. Bio-preservatives derived from animals

Antimicrobial proteins and peptides are components of the natural defense mechanisms of all living organisms, including mammals, insects, and amphibians (Huan et al., 2020). Lysozyme, pleurocidin, chitosan, defencins, protamine, and ovotransferrin are some of the main animal-derived bio-preservatives that can be utilized in the food industry (Gokoglu, 2018). Thanatin, diptericin, ceratotoxin, stomoxyn, drosocin, spinigerenin, sapecin, defensin A, gallerimycin, termicin, royalisin, apidaecin, formaecin, pyrrhocoricintin, drosomycin, heliomicin, attacins, coleoptericin, smD1, metchnikowin, and lebocin are some potential insect derived bio-preservatives (Buonocore et al., 2021).

2.3. Bio-preservatives derived from plants

Herbs and spices contain a range of active ingredients with antibacterial, antifungal, antiparasitic, and antiviral properties (Parham et al., 2020). According to Quinto et al. (2019), herbs, including lemongrass, parsley, oregano, coriander, sage, rosemary, garlic, and spices (clove and cinnamon); organic compounds such as vanillin; and oils such as citral, have different antimicrobial compounds and have the potential to be used as food bio-preservatives. Plant-derived bio-preservatives are substances that give an organism a selective advantage; however, they are not necessary for growth or reproduction. These compounds are defined as secondary metabolites and include tannins, alkaloids, terpenoids, phenols, and their oxygen-substituted derivatives (Othman et al., 2019). Table 1 provides a synopsis of bio-preservatives.

3. Modes of action of bio-preservatives

The different inhibitory mechanisms of bacteriocins impede the development of disease-causing agents and spoilage microorganisms in foods (Darbandi et al., 2022). They prevent the growth of other bacteria

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Table 1. Classification of bio-preservatives

Bio-preservatives derived from microbial sources	Bio-preservatives derived from animal sources	Bio-preservatives derived from plants	References
Bacteriocins Bacteriophages Endolysin	Lysozyme Pleurocidin Defensins Lactoferrin Ovotransferrin Protamine Chitosan	Plant extracts and essential oils from herbs, spices	Baindara and Mandal, 2022; Batiha et al., 2021; Beya et al., 2021; Chang, 2020; Kell et al., 2020; Martínez-Graciá et al., 2015; Meena et al., 2021; Singh, 2018; Yusuf, 2018

by piercing target cells, limiting the production of peptidoglycans, preventing the synthesis of proteins by interacting with ribosomes or tRNA, and directly degrading the DNA of target cells (Wu et al., 2022). Subclass Ⅰa bio-preservatives are cationic and hydrophobic. Therefore, they can act as membrane permeabilizers of pathogenic microorganisms in foods, which can cause death by pore formation. Nisin is one of the best examples exhibiting this mode of action (Alhanout and Duval, 2020). Many food products, including dairy and meat products, use nisin as a natural preservative to inhibit *L. monocytogenes* and other gram-positive food spoilage microbes (Gharsallaoui et al., 2015). Subclass Ib bacteriocins inhibit specific essential enzymes in target bacteria, and maintain their bio-preservative functionality in foods (Simons et al., 2020).

According to Wu et al. (2022), enterocins exert different actions on target microorganisms, such as suppression of genetic expression and destruction of the cell envelope. Most enterocins target the lipid component, which can act as an intermediate in the peptidoglycan biosynthesis mechanism in pathogenic organisms and inhibit its synthesis in target pathogens (Wu et al., 2022). The cell membrane becomes permeable upon interaction with the cell envelope-associated mannose phosphotransferase system. In addition, target cells undergo cellular leakage at the first instance after enterocin initiates membrane permeabilization (Wu et al., 2022).

Lacticin is another bacteriocin widely used in the dairy industry for food safety and spoilage prevention. Its preservation mechanisms are similar to those of nisin in food systems (Todorov et al., 2022). Pediocins are bacteriocins produced by *Pediococcus*. They are generally active against *Clostridium, Listeria*, *Streptococcus, Enterococcus, Leuconostoc, Lactobacillus*, and *Carnobacterium* (Khorshidian et al., 2021). Pediocins are used as bio-preservatives for meat, vegetables, and fruit products. Enterocins are often used to preserve meat because they are highly efficient in preventing the growth of *L. monocytogenes* and other pathogens in food systems (Kasimin et al., 2022).

Bacteriocins derived from gram-negative bacteria mostly maintain their bio-preservative functionality by forming pores in bacterial cell wall and degrading RNA, DNA, or tRNase-like nucleic acid structures; by disrupting essential enzymatic reactions in the ATP synthase complex, RNA polymerase, and DNA gyrase: or by perforating bacterial cell membranes (Simons et al., 2020).

Bacteriophages can coexist symbiotically with fermentation microorganisms during bio-preservation (Ramos-Vivas et al., 2021) and eradicate harmful organisms. A few industrial applications of bacteriophage-based bio-preservation include the inhibition of *Salmonella* in cheddar, *S. aureus* growth in curd, *L. monocytogenes* in cheese, *Salmonella typhimurium* in chicken frankfurters, and *Enterobacter sakazakii* in reconstituted infant formula milk (Moye et al., 2018). Bacteriophages use endolysins in the latter part of cell replication in target bacteria to impair the peptidoglycan layer of the host (Singh, 2018). In food systems, breaking the peptidoglycan layer causes the target bacteria to undergo osmotic lysis and death (Harper et al., 2021).

Natamycin has little impact on bacteria or viruses; however, it is effective against almost all foodborne yeasts and molds (Meena et al., 2021). Owing to its strong affinity for ergosterol, natamycin attaches to fungal cell membranes in an irreversible manner. This results in membrane hyperpermeability, which causes rapid leakage of vital ions and peptides, ultimately leading to cell lysis (Szomek et al., 2022). Plant secondary metabolites such as phenols, quinones, phenolic acids, tannins, coumarins, flavonoids, terpenoids, saponins, and alkaloids possess different antimicrobial effects and vary in structure and chemical composition (Yeshi et al., 2022). Hydroxyl groups in phenolic compounds inhibit the activity of target organisms because they can interact with bacterial cell membranes to alter membrane structures, resulting in the leakage of cellular components (Takó et al., 2020). Essential oils cause membrane disruption via lipophilic compounds that inhibit protein translocation, phosphorylation, electron transport, and other enzymatic activities, ultimately destroying target microorganisms by interrupting cell membrane integrity (Mukarram et al., 2021). They can be added to food directly or as a surface spray for bio-preservation (Mani-López et al., 2018). Cinnamon-like spices have antibacterial effects against both gram-negative and gram-positive bacteria such as *S. aureus, Salmonella anatum, Escherichia coli, L. monocytogenes,* and *Bacillus cereus* (Liu et al., 2017).

As an animal-originated bio-preservative, chitosan is a polycationic biopolymer naturally present in the exoskeletons of crustaceans and arthropods (Batiha et al., 2021). It is industrially manufactured from chitin, a byproduct of shellfish processing, and can minimize the growth of foodborne bacteria, yeasts, and molds, including *Salmonella, Saccharomyces cerevisiae, Botrytis cinerea, Aspergillus flavus, Zygosaccharomyces bailii, Byssochlamys spp., Mucor racemosus,* and *Rhizopus stolonifera* (Pellis et al., 2022). Lactoperoxidase is an enzyme that occurs in colostrum, saliva, raw milk, and other biological secretions in animals, and this enzyme forms antimicrobial compounds after reactions with thiocyanate in the presence of hydrogen peroxide (Magacz et al., 2019). Lysozyme is a bacteriolytic enzyme found in different animal-based sources such as avian eggs, insects, fish, mammalian milk, tears, and other secretions. They lyse microorganisms in food systems and have preservative functions (Singh, 2018). A summary of the importance of bio-preservatives is presented in Table 2.

4. Summary and future aspects of bio-preservatives

The use of bio-preservatives is a realistic strategy, given that microbes are resistant to antibiotics and consumers prefer healthier food products. For bio-preservatives to reach their highest potential in food preservation, further research into their physical and chemical characteristics, modes of action, amount added to food systems, and structure-function interactions is required. Different problems are associated with the use of bio-preservatives in food systems and need to be

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Table 2. Summary; bio-preservatives and their importance to the food safety

Bio-preservative	Mode of action of the bio preservative in foods	Contribution to the food safety	References
Nisin	Make pores in target bacterial membranes and destroy them. Therefore, act as a membrane permeabilizer.	Prevent the growth of pathogenic bacteria in canned vegetables and fruit juices. Shelf-life extension of brined shrimp. Inhibit the activity of *L. monocytogenes* on cold-smoked rainbow trout fish. Natural preservative for processed and ripened cheese (*C. botulinum*), puddings (semolina or tapioca), clotted cream, mascarpone, salads, pasteurized liquid egg, soups, dressings, sauces. Inhibition of *L. monocytogenes* in ready to eat meat products (it is more successful to use Nisin in combination with little amount of nitrite). Disrupt the life cycle of *Clostridium* spp. and *L. monocytogens* in vacuum packed fish products.	Simons et al., 2020 Szomek et al., 2022
Enterocin	Catalyzing cell-wall hydrolysis, bind with the bacterial membranes of the target cells which are negatively charged and destroyed them.	Reduce the effect of *E. coli O157:H7, B. cereus, Alicyclobacillus acidoterrestris* and *S. aureus* in rice, vegetables & fruit juices, *L. monocytogenes* in meat products, *S. aureus, L. monocytogenes, B. cereus,* in cottage cheese, cheddar and yogurt. For preservation of Soya milk.	Wu et al., 2022
Lacticin	Inhibit the essential enzymes in target bacteria.	Beneficial in dairy products for preserving storage life and ensuring safety measures in foods.	Simons et al., 2020
Natamycin	Bind with ergosterol component in plasma membrane and inhibit the ergosterol-dependent fusion of vacuoles.	Surfaces treatments in salami type sausages and hard cheese in order to inhibit the spoilage microorganisms. Direct addition to tomato paste, fruit juices, wine, and yoghurt mixes to minimize the spoilage and pathogenic fungal species. Prevention of the fungal growth on bread.	Szomek et al., 2022
Pediocin	Permeabilization and weakening of bacterial membranes, or the generation of membrane pores.	Growth inhibition of *B. cereus, L. monocytogenes, E. coli O157:H7, S. aureus* in milk, and *L. monocytogenes* in meat products.	Simons et al., 2020
Lactoperoxidase	Start oxidation reaction involving thiocyanate and hydrogen peroxide.	Preservation of cheeses, ice cream, whole eggs and infant formula.	Al-Baarri et al., 2019
Pleurocidin	Target the membrane and the DNA of the bacteria and damage the cell integrity.	Preservation against gram (-) bacteria such as *L. monocytogenes, V. parahemolyticus, S. cerevisiae, E. coli O157:H7,* and *P. expansum.*	Amiri et al., 2021 Ko et al., 2019
Essential oils from basil, clove, thyme, and oregano	Disrupt the transport of nutrients and ions, through the membrane and affect to the overall permeability of the cell.	Show the antibacterial effects against *B. cereus* in rice-based foods.	Mahmud and Khan, 2018
Eugenol in cinnamon		Prevent the mold growth in cheese.	Amiri et al., 2021
Citric acid, maleic acid, propionic acid, and lactic acid		Used against to *S. typhimurium,* and *L. monocytogenes, E. coli O157:H7* in lettuce and red apples.	Amiri et al., 2021
Lysozyme	Lysing the peptidoglycan in microbial cell walls, damage the bacterial cellular membranes, activate cell wall autolytic enzymes in bacteria.	Reduce the gas formation in cheeses such as Gouda and Edam (by *C. Tyrobutyricum*). Preservative effects on salads, vegetables, seafood, wines and pasta.	Magacz et al., 2019
Chitosan	Bind to the negatively charged bacterial cell wall and cause cell disruption It alters the membrane permeability.	Inhibit the growth of foodborne bacteria yeasts, and molds (*M. racemosus, S. cerevisiae, Salmonella, A. flavus, Z. bailii,* etc.).	Yilmaz Atay, 2019

addressed to find sustainable solutions. First is the increased cost and unfavorable aroma of plant-based bio-preservatives. Second is the lack of information on the complexity of food systems and bio-preservatives. Third, there are insufficient industry manufacturers of bio-preservatives; therefore, producers are unable to use these strategies in mass production. Lastly, there is inadequate awareness regarding bio-preservation, including preservative technology and its importance. There is great potential for the development of active and intelligent packaging materials incorporating different bio-preservatives. However, the interaction of bio-preservatives with packing materials, the release of various bio-preservative compounds into food items, and the stability of the preservatives need to be studied prior to their practical application. Studies on the shelf-life-extending abilities and sensory acceptance of bio-preservatives according to their origin is another potential research area that can be beneficial to the industry. There are no regulatory systems for bio-preservatives like there are for other preservatives or food additives. Therefore, it is necessary to further investigate the health effects of these compounds in animal and human trials. The combination of microbially oriented bio-preservatives with gene technology and enhanced functionality is another potential research topic.

The synergistic effects of these natural preservatives, when combined with cutting-edge technologies such as nanotechnology and gene technology, may allow for the replacement of chemical preservatives or less invasive processing procedures while maintaining adequate microbiological safety and food quality. Moreover, it will help minimize unnecessary costs in the food preservation process. Bio-preservative approaches are sustainable solutions in the food industry because they enhance the quality of food while prolonging its shelf life. A large number of bio-preservatives of microbial, plant, or animal origin can be effectively used in the bio-preservation process. Bacteriocins, bacteriophages, antifungal compounds, lysozymes, chitosan, lacticins, pediocins, and plant extracts are some of the most commonly used bio-preservatives in the industry.

In conclusion, producers can select the most appropriate and cost-effective bio-preservatives for food preservation as they can be acquired from numerous sources. Bio-preservation can be used alone or in combination with other preservatives to achieve optimal results for preventing microbiological growth and ensuring food safety. Based on the results of recent scientific studies, bio-preservation has the potential to enhance the quality and safety of food production without altering the nutritional or sensory food attributes.

Conflict of interests

The authors declare no potential conflicts of interest.

Author contributions

Conceptualization: Wijayasekara KN, Abeyrathne EDNS. Data curation: Ranathunga NS. Formal analysis: Ranathunga NS. Methodology: Ranathunga NS, Wijesekara KN, Abeyrathne EDNS. Software: Ranathunga NS. Validation: Wijesekara KN, Abeyrathne EDNS. Investigation: Ranathunga NS. Writing - original draft: Ranathunga NS. Writing - review & editing: Wijayasekara KN, Abeyrathne EDNS.

Ethics approval

This article does not require IRB/IACUC approval because there are no human and animal participants.

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