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Review Article

Preservatives Added to Food Preservation and Their Impact Against the Bacteria

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Abstract

The Meat, milk, fruits, and vegetables are vital nutrients for humans because they include protein, essential amino acids, vitamin B groups, and minerals. Because of its high-water activity and nutrient content, it also promotes deterioration and food-borne disorders. Thus, food preservation is essential for guaranteeing food safety and security. This paper examines the critical significance of animal origin natural preservatives in food preservation systems. This review article explains how these preservatives extend the shelf life of food products in a variety of categories by thoroughly examining their efficacy, methods of action, and diverse applications. Furthermore, it explains the inherent benefits of using animal origin natural preservatives, demonstrating their superiority to synthetic counterparts in terms of safety, sustainability, and environmental effect. Regulatory frameworks and customer opinions are also investigated, providing useful information about the uptake and acceptance of various preservatives in the food business. This analysis emphasises the importance of using natural and animal-derived solutions to promote a safer, more sustainable, and healthier food supply chain.

Key words: preservatives; safety; sustainability; environmental effect; food-borne disorders

Introduction

The Food, including meat, milk, fruits, and vegetables, are crucial nutrient sources for humans due to their high protein content, essential amino acids, vitamin B groups, and minerals. However, its nutrient composition also provides an appropriate environment for spoilage microorganisms or food-borne pathogens due to their high-water activity and nutrient factors. This deterioration is primarily due to microbial activity, enzymatic reactions, and chemical changes that occur during storage and handling. Proteins in food are particularly prone to spoilage by proteolytic bacteria, which break down proteins into smaller peptides and amino acids. This process, known as proteolysis, results in the production of off-flavours, off-odors, and undesirable textures. For instance, the spoilage of milk by lactic acid bacteria leads to souring and curdling, while the spoilage of meat by Clostridium and Pseudomonas species results in putrefaction and the development of foul-smelling compounds such as ammonia and hydrogen sulphide [1.2,3,4,5,6,7 and 8]. The spoilage of food not only compromises its sensory qualities but also poses significant health risks. The Spoiled food can harbor pathogenic microorganisms that cause foodborne illnesses. The Common foodborne pathogens include Salmonella, Escherichia coli, Listeria monocytogenes, and Campylobacter. These pathogens can cause a range of illnesses, from mild gastroenteritis to severe systemic infections. Moreover, the economic impact of food spoilage is substantial, contributing to food waste and financial losses in the food industry. So, the application of preservatives is one way to protect the fatty acids, proteins, and vitamins in food from auto-oxidation and deterioration. The Food preservation involves the application of processes that inhibit microbial growth, retard enzymatic activity, and prevent chemical changes that lead to spoilage. Traditional methods of food preservation include drying, salting, smoking, and fermenting. These methods were employed long before the advent of modern technology and remain in use today for their effectiveness and simplicity. Synthetic preservatives such as butyl-hydroxy-anisole (BHA), sulphur dioxide, butyl-hydroxy-toluene (BHT), tert-butyl-hydroquinone (TBHQ), propyl-gallate (PG), nitrites, nitrates, ascorbates, monosodium glutamate, and liquid smoke can be used to slow the rate of lipid and protein auto-oxidation.This extends the product's shelf-life and prevents deterioration. Despite their effectiveness, these preservatives have been linked tovariety of health problem including food poisoning, liver damage, carcinogenesis, and mutagenesis [9,10,11,12,13,14,15 and 16]. Moreover, the long-term consumption of foods containing synthetic preservatives may also contribute to the development of chronic health conditions such as asthma, hyperactivity in children, and hormonal imbalances. Because of the potential health risks associated with synthetic preservatives, the food industry is looking for animal origin natural preservatives as a safer alternative. Moreover, natural preservatives offer a promising alternative due to their biodegradability, lower toxicity, and potential health benefits. Animal origin natural preservatives including Lyzozymes, lactoferrin, lactoperoxidase system, Nisin, and chitosan are of crucial concern for food safety and security. These natural compounds and possess intrinsic antimicrobial properties that can effectively inhibit the growth of spoilage microorganisms and pathogens. Therefore, the key objectives of this research are to investigate and highlight the use of animal origin natural preservatives in food preservation. This review seeks to provide a complete understanding of how natural preservatives can improve food safety and quality by investigating their sources, mechanisms of action, and applications in various foods. The food preservation, several substances have garnered attention due to their natural origin and effectiveness. These substances

include Lysozyme, Nisin, Lactoferrin, Propolis, Lactoperoxidase System, Ovotransferrin and Chitosan [17,18,19,20,21,22,23 and 24].

The Lysozyme:

The Lyzozyme (LZ) is a GRAS (generally recognized as safe) antimicrobial protein found in mammalian milk and avian eggs. It has been used in food processing since the 1970s, including in cheese, sushi, and Chinese noodles. The Source, The Lysozyme is commonly extracted from egg whites, particularly hen egg whites, where it is abundantly found. The Chemical Structures, The Lysosomal enzymes are proteins; thus, their chemical structure is composed of long chains of amino acids. Each enzyme has a unique primary structure (amino acid sequence), secondary structure (local folded structures like alpha-helices and beta-sheets), tertiary structure (three-dimensional shape), and sometimes a quaternary structure (assembly of multiple polypeptide chains). The Procedure of Lysosome extraction, the extraction of lysosomal enzymes involves several steps, Purification, The Lysosomal enzymes are further purified using techniques such as chromatography (e.g., ion-exchange, affinity, and size-exclusion chromatography) to obtain highly pure enzyme preparations. Specific inhibitors or substrates can be used to aid in the purification of individual enzymes. The Characterization, the purity and activity of the enzymes are characterized using various biochemical assays and analytical techniques like SDS-PAGE and mass spectrometry. The Mechanism of Action, The Lysozyme efficiency can be challenged in its ability to control bacterial growth. The generally recognized mechanism adapted by this protein is the enzymatic degradation of the glycosidic β-linkage in the cell wall to kill the sensitive bacteria. However, increasing evidence suggests that lysozyme has additional bactericidal mechanisms towards bacteria beyond those related to the catalytic action [25,26,27,28,29,30,31,32 and 33]. The Catalytic Mode of Antibacterial Action, The Lysozyme functions by attacking, hydrolyzing, and breaking the muco polysaccharide part of the PG in the bacterial cell wall. Similarly, this enzyme can also break glycosidic bonds in chitin. The lysozyme molecule generally employs a compact, globular structure with hydrophilic groups of residues exposed on the surface and hydrophobic ones clustering internally. To accommodate the long chain substrate, there is a deep groove on the surface of lysozyme. This groove is the active site involved in binding to the bacterial carbohydrate chain and subsequently cleaving it. The binding substrate is a polysaccharide of six amino sugars long and is positioned along the active site by hydrogen bonding and hydrophobic interactions. During this fitting process, the strain on the glycosidic bond between 4th and 5th sugar unit increases, and thus the carbon-oxygen bond between them will be broken by a general-acid catalyst residue, glutamic acid (Glu) and a general- base catalyst residue, aspartic acid (Asp) or cysteine (Cys) in the active site of lysozyme. In this reaction, glutamic acid acts as a proton donor through the free carbonyl group of its side chain, whereas aspartic acid acts as a nucleophile to produce a glycosyl-enzyme intermediate. This intermediate product immediately reacts with a water molecule and generates the hydrolysis product. Besides the Glu and Asp residues, a third catalytically important residue—threonine (Thr) or serine (Ser), serving as a catalytic water positioning residue (in the sequence of Glu-8aa-Asp/Cys-5aa-Thr catalytic triad), was previously demonstrated for lysozymes of coliphages T4 and P21. As an exception, goose egg-white lysozyme (GEWL) has only a single catalytic residue-Glu, suggesting that a second acidic residue is not essential for the catalytic activity of goose lysozyme. This phenomenon has also Lysozyme functions by attacking, hydrolyzing, and breaking the muco polysaccharide part of the PG in the bacterial cell wall. Similarly, this enzyme can also break glycosidic bonds in chitin. The lysozyme molecule generally employs a compact, globular structure with hydrophilic groups of residues exposed on the surface and hydrophobic ones clustering internally [34,35,36,37,38,39,40 and 41]. To accommodate the long chain substrate, there is a deep groove on the surface of lysozyme. This groove is the active site involved in binding to the bacterial carbohydrate chain and subsequently cleaving it. The binding substrate is a polysaccharide of six amino sugars long and is positioned along the active site by hydrogen bonding and hydrophobic interactions. 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As an exception, goose egg-white lysozyme (GEWL) has only a single catalytic residue-Glu, suggesting that a second acidic residue is not essential for the catalytic activity of goose lysozyme. This phenomenon has also been observed in a lytic transglycosylase, i.e., phage lambda endolysin and an endolysin of Burkholderia AP3 phage with lysozyme-like catalytic subunit. The Non-Catalytic Mode of Antibacterial Action, The Researchers had long believed that lysozyme only exerts its antimicrobial activity by functioning as a hydrolytic enzyme. However, there is emerging evidence showing that non-enzymatic microbicidal activity rather than hydrolytic activity of lysozyme contributes substantially to the killing of microbes. A direct proof is that the lysozyme with less or no enzymatic activity had maintained, or even improved, bactericidal activity against both Gram-positive and Gram-negative bacteria. It was shown that heat denaturation of HEWL, resulting in an enzymatically inactive, more cationic, and hydrophobic dimeric form, enhanced the bactericidal activity against Gram-negative bacteria [42,43,44,45,46,47,48 and 49].

The Nisin: The Nisin is also known by its trade names such as Nisaplin and Nisatin. It comes from the N-inhibitory substance, as a product of 'lactic streptococci' or lactococci (later known as nisin) which can inhibit several types of pathogenic bacteria. In further developments, nisin is known as bacteriocin, a bio preservative compound produced by Lactococcus lactis subsp. Lactis is used commercially in food products. The Bacteriocin is a toxic protein produced by bacteria. It can inhibit other bacteria (of the same species, different strains for a narrow spectrum, or other bacteria of different species for a broad spectrum). These bacteriocins are categorized into Class I, Class IIa/b/c, and Class III. More specifically, nisin belongs to the Bacteriocin Class I group as a type A antibiotic in the form of a long non-globular molecule. Other food products, such as fisheries goods, use nisin. Nisin can decrease the amount of L. monocytogenes in crab meat or smoked salmon. Smoked salmon has also been demonstrated to lower C. botulinum toxin type E production at 10 and 26°C. Nisin can be added to fish sausage to extend its shelf life. The shelf life of fish sausages maintained at room temperature can be increased by 50 ppm, while that of fish sausages stored at low temperatures (6°C) can be increased by 30 to 150 days [50,51,52,53,54,55,56,57,58 and 59]. The pathogenic bacterium Carnobacterium piscicola (CECT 4020) was found to be successfully inhibited using nisin in turbot fish (Scophthalmus maximus L.). While enteric bacterial populations of Podamasys jubelini and Arius heudelotii were able to be reduced by 4 and 2 logs, respectively, when starter culture Lactococcus lactis subsp. lactis was added to ferment fish filets. Nisin is approved as a food preservative in the United Kingdom in the 1950s and is now widely used in over 50 countries. Since 1969, the World Food Agency (FAO/WHO) has approved the use of nisin as a natural preservative, and it is designated as food additive. The FDA authorized the use of nisin in canned foods in 1988 to prevent the formation of Clostridium botulinum. The only pure bacteriocin now permitted for use in the US for food preservation is nisin, which has also received approval for use in more than 50 other nations. Nisin is a bacteriocin, and compared to other bacteriocins, it has a substantially broader inhibitory spectrum. Bacteriocins typically only affect Gram-positive bacteria. However, nisin has also been shown to inhibit several Gram-negative bacteria, typically when combined with other preservatives, such as surfactants, chelating agents, and adjuvants. The Source, The Nisin is primarily obtained from the fermentation of Lactococcus lactis, a gram-positive bacterium commonly found in dairy products 60,61,62,63,64,65,66 and 67. The Chemical Structure, The Nisin is a polycyclic peptide composed of 34 amino acid residues, including the unusual amino acids lanthionine, β-methyllanthionine, didehydroalanine, and didehydroaminobutyric acid. These residues form multiple intramolecular thioether bridges, resulting in a characteristic three-

dimensional structure. The chemical structure of nisin consists of a linear peptide chain with five amino acid rings and three bridges formed by thioether linkages. The Procedure of Nisin Extraction, the extraction of nisin involves several steps, The Fermentation, The Lactococcus lactis bacteria are cultured in a suitable medium under controlled conditions to produce nisin. The Purification, the fermented broth is centrifuged to separate bacterial cells from the supernatant containing nisin. Nisin is then purified using techniques such as ultrafiltration, ion exchange chromatography, and reverse-phase high-performance liquid chromatography. The Mechanism of Action, The Nisin acts by disrupting bacterial cell membranes, leading to leakage of intracellular contents and ultimately cell death. It binds to lipid II, a precursor molecule involved in bacterial cell wall synthesis, and forms pores in the membrane, causing depolarization and loss of membrane integrity. Nisin's mechanism of action is primarily bactericidal, targeting a wide range of gram-positive bacteria, including foodborne pathogens like Listeria monocytogenes and Staphylococcus aureus. The Applications, The Nisin has a wide range of applications in the food industry due to its antimicrobial properties [68,69,70,71,72,73,74,75 and 76].

The Lactoferrin: The Lactoferrin is also known by its abbreviated form LF, and in some contexts, it is referred to as Lactotransferrin. The Lactoferrin, belonging to the transferrin family of proteins, is multifunctional and participates in the regulation of free iron levels in body fluids, making the protein bacteriostatic and beneficial to health. It is the most important iron-binding protein in milk. The Lactoferrin is present in higher quantities in saliva, tears, seminal fluid, white blood cells, and milk of mammals. It is naturally found in cow's milk at an average level of about 0.2 g/L. The maximum lactoferrin content is about 50 to 100 g/L in cow mammary secretions. In human milk, 2–4 g/L, and colostrum, 6 to 8 g/L of LF has been found. It is revealed that lactoferrin has bacteriostatic properties against gram-negative bacteria requiring high iron like coliforms. It also acts against bacteria like Staphylococcus aureus, Listeria monocytogenes, and Bacillus species. The Lipopolysaccharides from the external membrane of bacteria are released leading to changes in the permeability due to lactoferrin interaction. It holds iron at a lower pH and has a basic nature because of a 300 times higher affinity constant than transferrin. Lactoferrins are polypeptides having a single chain with a molecular weight of 80 kDa. It comprises 1–4 glycans but it depends upon the species [77.78,79,80,81,82,83 and 84]. The Lactoferrin of Bovine and human origin have the same sequence identity. The Bovine Lactoferrin contains sugars including N-acetylglucosamine, acetyllactosamine, galactose, fucose, mannose, and neuraminic acid. It has been reported that antibacterial activity is due to the N-terminus of bovine Lactoferrin which is cationic. The Lactoferrin functions as an antioxidant by binding the free ferric iron with high affinity. It is helpful for the regulation of iron transport and transcriptional regulation. It possesses proteolytic and enzymatic activities. This protein has amino acids with the highest contents including alanine, leucine, glycine, tryptophan, histidine, and methionine with 10%, 9%, 7%, 1.5%, 1.3%, and 0.6% respectively. The Current uses of bovine lactoferrin include dietary iron supplements, infant formulas, beverages, chewing gums, fermented milk and milk products. It inhibits bacterial adhesion on abiotic surfaces. LF has also been discovered to be effective against the formation of biofilms. The Food and Drug Administration certified this protein as GRAS prestige. The Source, The Lactoferrin is primarily derived from bovine milk, although it is also found in human milk and the milk of other mammals (85,86,87,88,89 90 and 91). The Chemical Structure, The Lactoferrin is a single-chain polypeptide consisting of approximately 691 amino acid residues, with a molecular weight of about 80 kDa. It has a unique structure characterized by two symmetrical lobes, each containing an iron-binding site. The chemical structure of lactoferrin includes a high proportion of basic amino acids, such as lysine and arginine, which contribute to its cationic nature and antimicrobial activity. The Procedure of Lactoferrin Extraction, The Lactoferrin can be extracted from milk using various methods as the Fractionation. The Milk is subjected to fractionation techniques such as ultrafiltration or chromatography to isolate lactoferrin from other milk proteins. The Purification, the lactoferrin-rich fraction obtained from milk is further purified using methods such as ion-exchange chromatography, affinity chromatography, or size-exclusion chromatography to achieve high purity (92,93,94,95,96, 97,98 and 99). The Mechanism of Action, The Lactoferrin exerts its biological activities through multiple mechanisms as The Antimicrobial Activity, The Lactoferrin binds to bacterial cell surfaces, disrupting membrane integrity and inhibiting bacterial growth. It also sequesters iron, limiting its availability to pathogens and depriving them of an essential nutrient. The Antioxidant Properties, The Lactoferrin scavenges free radicals and inhibits oxidative damage, thereby protecting cells and tissues from oxidative stress-induced injury. The Application of Lactorferrin in food industry is Important [100,101,102,103,104,105 and 106].

The Propolis: The Propolis is a natural resinous substance collected by honeybees from various plant sources and used to seal and protect their hives. It has been used for centuries in traditional medicine for its various health benefits. Propolis (bee glue) is a sticky dark-coloured material that honeybees collect from living plants, mix with wax and usein construction and adaptation of their nests. The term 'propolis' was used in Ancient Greece: pro (for, in front of, e.g., at the entrance to) and polis (city or community); a substance that is for or in defence of the city or hive. Bees apply propolis in a thin layer on the internal walls of their hive or other cavity they inhabit. It issued to block holes and cracks, to repair combs, to strengthen the thin borders of the comb, and for making the entrance of the hive weathertight or easier to defend. Propolis also is used as an "embalming" substance to cover hive invaders which bees have killed but cannot transport out of the hive [107,108,109,110,111,112 and 113]. Bees make use of the mechanical properties of propolis and of its biological action: bee glue contains the putrefaction of the" embalmed" intruders, it is responsible for the lower incidence of bacteria and molds within the hive than in the atmosphere out-side. The action against microorganisms is an essential characteristic of propolis and it has been used by human beings since ancient times for its pharmaceutical properties. The Propolis possesses antibacterial, antifungal and antiviral properties and many other beneficial biological activities: anti-inflammatory, antiulcer, local anesthetic, hepatoprotective, antitumor, immuno stimulating, etc. For this reason, propolis is widely used as a popular remedy in folk medicine, in apitherapy as a constituent of "biocosmetics", "health food" and for numerous further purposes. Propolis is also known by various other names, including bee glue, hive dross, and bee propolis [114,115,116,117,118,119 and 120]. The Source, The Propolis is primarily derived from tree buds, sap flows, and other botanical sources. Honeybees collect these resins, mix them with saliva, beeswax, and enzymes, and use them to coat and seal the interior of their hives. The Chemical Structure, the chemical composition of propolis varies depending on its botanical source, geographical location, and season of collection. However, it typically contains a complex mixture of polyphenols, flavonoids, terpenes, and other organic compounds. The chemical structure of propolis is characterized by its high content of phenolic compounds, such as caffeic acid, cinnamic acid, and their derivatives, which contribute to its antioxidant and antimicrobial properties. The Procedure of Propolis Extraction, The Propolis extraction methods vary, but commonly involve the following steps as The Collection, The Propolis is harvested from beehives using special traps or screens that allow beekeepers to collect it without disturbing the hive structure. The Cleaning, the collected propolis may contain impurities such as beeswax, bee parts, and debris. It is cleaned by washing or filtering to remove these impurities [121,122,123,124,125,126,127 and 128]. The Extraction, the cleaned propolis is then extracted using solvents such as ethanol or water. The choice of solvent and extraction method may affect the composition and properties of the resulting propolis extract. The Mechanism of Action, the biological activities of propolis are attributed to its complex chemical composition as The Antimicrobial Activity, The Propolis contains various compounds, such as flavonoids and phenolic acids, that exhibit broad-spectrum antimicrobial activity against bacteria, fungi, and viruses. The Antioxidant Properties, The Propolis scavenges free radicals and inhibits oxidative stress, protecting cells and tissues from damage caused by reactive oxygen species (ROS) (129,130,131,132,133,134,135 and 136). The Applications of Propolis in food industry, the probable function of propolis as a food additive has yet to be established, but it has an unpleasant taste that can alter food texture and taste. However, propolis has shown promise in various food-related applications. For instance, propolis has been proposed as a potential natural preservative due to its antimicrobial properties and ability to extend the shelf life of fresh fish products, such as Shibuta fillets. Studies have also explored

the use of propolis in nanoemulsions, which could prevent the degradation of propolis while masking its off flavour, thus enhancing its acceptability in food products. Furthermore, propolis extracts have demonstrated efficacy in inhibiting yeast contamination in industrial foods and reducing anthracnose growth on fruits like Kent mangoes. The use of propolis as a preservative offers a natural alternative to synthetic chemicals, aligning with the growing consumer demand for clean-label and sustainable food options [137,138,139,140,141,142,143 and 144]. The Lactoperoxidase System, the lactoperoxidase system may also be referred to as the milk peroxidase system or the thiocyanate peroxidase system. The lactoperoxidase system is a natural antimicrobial system found in various secretions, including milk, saliva, and tears, in mammals. The LPS is an enzymatic and antibacterial system which consists of the enzyme LP, SCN– resulting from hepatic metabolism, and H2O2 from cellular metabolis. The system is activated through an oxidation reaction of SCN– by H2O2 and catalysed by LP. The products deriving from this process are hypothiocyanic acid (HOSCN) and the hypothiocyanite ion (OSCN–), which has a broad spectrum of antimicrobial effects against bacteria, fungi and viruses. LP is an oxidoreductase and catalyses the oxidation of SCN– at the expense of H2O2 to generate the antimicrobial product of OSCN–. Thiocyanate ion. SCN– is widely distributed in animal tissues and secretions, being present in the mammary, thyroid and salivary glands, in organs such as stomach and kidneys, and in biological fluids such as plasma and brain fluid. The concentration of SCN– in bovine milk reflects serum levels in blood, varying according to the breed, species, udder health, type of diet, season and geographic regions. The concentration of SCN– present in the animal's metabolism partly depends on the food supplied, with there being two important dietary sources that give rise to SCN–, glucosinolates and cyanogenic glycosides. It has been reported that fresh cow's milk contains 1 to 35 mg of SCN– per litre, which is not always sufficient to activate the LPS. About 10 mg of SCN– per litre is added to raw milk for exogenous activation of the system. The Chemical Structure, The Lactoperoxidase is a heme-containing glycoprotein enzyme belonging to the peroxidase family. It contains a prosthetic group known as heme, which consists of an iron atom bound to a porphyrin ring [145,146,147,148,149,150 and 151]. The chemical structure of lactoperoxidase includes a catalytic site where hydrogen peroxide and a halide ion (usually thiocyanate or iodide) bind, allowing the enzyme to catalyze oxidation of various substrates. The Procedure of Lactoperoxidase System Extraction, The Lactoperoxidase can be extracted from milk using various methods, The Fractionation, The Milk is subjected to fractionation techniques such as ultrafiltration or chromatography to isolate the lactoperoxidase from other milk proteins. The Purification, the lactoperoxidase-rich fraction obtained from milk is further purified using methods such as ion-exchange chromatography, affinity chromatography, or size-exclusion chromatography to achieve high purity [152,153,154,155,156,157 and 158]. The Mechanism of Action, the lactoperoxidase system exerts its antimicrobial effects through a mechanism involving the generation of reactive oxygen species (ROS) and the oxidation of various substrates. The Hydrogen Peroxide Production, The Lactoperoxidase catalyzes the oxidation of halide ions (e.g., thiocyanate or iodide) by hydrogen peroxide, resulting in the production of reactive oxygen species such as hypochlorous acid and hypothiocyanite ions. The Oxidative Damage, these reactive oxygen species can react with microbial cell components, including proteins, lipids, and nucleic acids, leading to oxidative damage and cell death. The Applications of The Lactoperoxidase system in food industry, The lactoperoxidase system is used as a natural preservative in dairy products, such as milk and cheese, to inhibit the growth of spoilage and pathogenic bacteria.The antimicrobial agents of the LPS inhibit milk deterioration, thus preserving the microbiological quality of the milk. The method can be applied to the raw milk of several species, although the system's effectiveness depends on the type of microbiological contamination, the number of microorganisms and the milk temperature during its use. The most recommended industrial application of the LPS in food production is in the dairy industry, more specifically for preserving raw milk during the storage and transport of milk to processing [159,160,161,162,163,164,165 and 166].

The Ovotransferrin: The Ovotransferrin is also known as conalbumin due to its ability to bind to heavy metals such as cadmium and nickel. Ovotransferrin (Otrf) or conalbumin belongs to the family of transferrin iron-binding glycoproteins. In mammals, two different soluble iron-binding glycoproteins are present, serum transferrin, involved in iron transport and delivery to cells and, lactoferrin, involved in the so-called natural immunity. Differently, Otrf is the only soluble glycoprotein of the transferrin protein family present in avian. Otrf is present both in avian plasma and egg white and possesses both iron-transfer and protective properties. Otrf represents about 12%–13% of total egg white proteins and contributes to promoting the growth and development of the chicken embryo mainly preventing the growth of micro-organisms together with other proteins such as lysozyme, cystatin, ovomacroglobulin and avidin. Galliformes (chicken, Gallus gallus and turkey, Meleagris gallopavo) appear to possess albumens with greater antimicrobial activity than those of the anseriformes (duck, Anas platyrhynchos), possibly due to higher concentrations of ovotransferrin and of the broad active c-type lysozyme. However, recent evidence indicates that Otrf is endowed not only with the antibacterial activity related to iron withholding, but also with other roles related to the protection of the growing embryo, including regulation of iron absorption; immune response; and anti-bacterial, anti-viral and anti-inflammatory properties. Some of these properties are shared by both the human protein homologues and peptides deriving from its partial enzymatic hydrolysis, being in this latter case also increased. The Source, The Ovotransferrin is derived from avian eggs, primarily chicken eggs, and is present in the egg white alongside other proteins such as ovalbumin and lysozyme. The Chemical Structure, the trf is a monomeric glycoprotein containing 686 amino acids, with a molecular weight of 77.9 kDa and an isoelectric point of 6.0. Like the mammalian transferrins, the single chain of Otrf consists of two globular lobes (N- and C-lobes), interconnected by a α-helix of nine amino acidic residues (residues 333–341) that can be released by tryptic digestion. Each lobe contains an iron binding site and is divided in two domains, (domains N1 and N2 in the N-lobe and domains C1 and C2 in the C-lobe, respectively). The Procedure of Ovotransferrin the Extraction Methods, the extraction of ovotransferrin from egg white involves several steps as Separation, The Egg white is separated from the yolk and filtered to remove any impurities or eggshell fragments. The Precipitation, The Ovotransferrin is precipitated from the egg white by adjusting the pH or adding salt solutions such as ammonium sulfate. The Purification, the precipitated ovotransferrin is further purified using techniques such as chromatography to remove other proteins and contaminants. The Mechanism of Action, the antimicrobial activity of ovotransferrin is attributed to its ability to sequester iron and deprive pathogens of an essential nutrient as Iron Binding, The Ovotransferrin binds to iron ions with high affinity, forming a complex that is unable to interact with microbial cells. The Microbial Inhibition, by sequestering iron, ovotransferrin inhibits the growth and proliferation of pathogenic microorganisms, including bacteria and fungi, which require iron for their metabolic processes. The Applications of Ovotransferrin in food industry is important [167,168,169,170,171,172,173 and 174].

The Chitosan: The Chitosan is also known by its chemical name β-(1→4)-2-amino-2-deoxy-Dglucose, as well as by trade names such as Kytex, Kytosan, and Chitopharm. Chitosan is a polysaccharide derived from chitin, a natural polymer found in the exoskeletons of crustaceans such as shrimp, crabs, and lobsters. It is known for its versatile properties and has various applications in industries ranging from pharmaceuticals to agriculture. Chitosan, a partially deacetylated derivative of chitin, is a hetero-polysaccharide composed of 2-amino-deoxy-β-D-glucopyranose and 2-acetamido-deoxy-β-D-glucopyranose (chitin) residue. The major property of chitosan is dictated by the presence of three different functional groups (primary —OH, secondary —OH and —NH2) and its water solubility in acidic pH. Due to the presence of reactive groups, it inhibits the growth of a wide variety of bacteria and fungi. Chitosan has many different applications and can be utilized for developing various formulations. Chitosan origin edible coatings can also be used as carriers of food ingredients (antimicrobials, texture enhancers and nutraceuticals) to improve the safety, quality and functionality of the F&V. Edible coating without disturbing sensory quality and nutritional value of the F&V needs further scientific research. Chitosan/chitin refers to one of the most abundant natural polysaccharides in nature. It can be obtained from several different sources, but the main source of chitosan is usually marine crustacean's shells. Atlantic Canada, with its long coastline, offers a great source of different marine crustaceans, i.e., shrimp, lobster,

crab, etc. that can be utilized for the extraction of chitosan. Aquaculture industries are growing fast to meet current demand. However, disposal of crustacian shells is an environmental concern for aquaculture industries and recovery of these shells from various sources can leverage industries for producing chitosan and its derivatives at competitive costs. The Source, The Chitosan is primarily derived from the deacetylation of chitin, a process that involves the removal of acetyl groups from chitin molecules using alkaline hydrolysis or enzymatic treatment. The Chemical Structure, The Chitosan is a linear polysaccharide composed of repeating units of β-(1→4)-linked D-glucosamine and N-acetyl-D-glucosamine residues. The Procedure of Chitosan extraction, The Chitin Isolation, The Chitin is extracted from crustacean shells by demineralization and deproteinization, typically using acidic or alkaline solutions. The Chitosan is derived by modifying chitin structure through removal of the acetyl groups, which are bonded to amine radicals in the C2 position on the glucan ring. Chemical hydrolysis in concentrated alkaline solution is performed at elevated temperatures to produce a partially deacetylated form of chitin referred to as chitosan. Chitosan preparations differ by the degree of deacetylation. The Deacetylation, The Chitin is treated with alkali, such as sodium hydroxide, to remove acetyl groups and convert it into chitosan. The Purification, the resulting chitosan is purified by filtration, precipitation, or chromatography to remove impurities and obtain the desired molecular weight and degree of deacetylation. The Mechanism of Action, The Antimicrobial Activity, The Chitosan exhibits antimicrobial activity against a wide range of bacteria, fungi, and viruses by disrupting cell membranes, inhibiting enzyme activity, and chelating metal ions essential for microbial growth. Chitosan is cationic biopolymer, having antimicrobial properties which can be affected by pH, concentration, molecular weight, degree of polymerization and cross-linking. Chitosan solution is highly stable over a long period of time, however its stability in neutral pH is highly important for exhibiting antimicrobial activity against a wide variety of foodborne pathogens. The major mechanism of action in antimicrobial activity involves interaction with bacterial cell wall, cell membrane and cytoplasmic constituents via electrostatic interactions. Chitosan has been found to be effective against both gram-positive and gram-negative bacteria. The outer membrane (OM) of gram-negative bacteria such as, Escherichia coli, is composed of an asymmetric lipidprotein bilayer (lipopolysaccharide, LPS). The divalent cations (i.e., Ca2 +, Mg2 +) present in the OM play an important role in the stabilization of the core anionic charges of the LPS molecules. It can be hypothesized that chitosan replaces the divalent cations from their binding sites and reduces the interaction between the LPS molecules, causing membrane disruption and cell lysis due to penetration (through electrostatic interaction) of positively charged chitosan through cell membrane of gram-negative bacteria. Unlike gram-negative bacteria, the gram-positive bacteria do not have an outer membrane. Hence, chitosan as a polycationic long chain molecule can adhere better with gram-positive bacterial members such as Staphylococcus aureus. For this reason, the inhibition effort from chitosan is more effective against gram-positive bacteria than gram-negative bacteria. In the literature, it was reported that gram-positive bacteria containing teichoic acid and lipoteichoic acid that are poly-anionic surface polymers, interact with intracellular substances, so that the vital bacterial activities are impaired. The Applications of Chitosan in food industry, The Chitosan is used as a natural preservative, antimicrobial agent, and food coating material to extend the shelf life of perishable food products and prevent microbial contamination. The use of chitosan is widely investigated as an edible coating, which is defined as the formation of a thin film directly on the surface of the product they are intended to protect. Edible coatings/films form a protective barrier around the F&V and can be consumed along with the coated product. In the F&V preservative applications, the creation of a moisture and gas barrier may lead to weight loss and respiration rate reductions with a consequent general delay in spoilage, which will extend the shelf-life of the product. Water vapor permeability (WVP) and oxygen permeability (OP) are the barrier properties commonly studied to determine the ability to protect foods from the environment. Edible chitosan films are extremely good barriers for permeation of oxygen, while exhibiting relatively low water vapor barrier characteristics. Films and coatings develop selective permeability characteristics, especially to O2, CO2 and ethylene and allow some control of fruits respiration and reduce growth of microorganisms. Coating practice has long been followed for preservation of citrus, apples (shellac and carnauba wax), tomatoes (mineral oil) and cucumbers (various waxes). Different types of irradiated chitosan coatings were studied for enhancing the shelf life and improving quality of mangos. The effect of coating with irradiated crab and shrimp chitosan (MW = 5.14 × 104) and un-irradiated crab chitosan (MW = 2.61 × 105) on postharvest preservation of mangos (Mangifera indica L.) was studied and results showed effectiveness at an appreciable level. The effect of both control and irradiated chitosan was observed on the fruits-spoiling fungi (Colletotrichum gleosporioides). The percentages of spoiled fruits were 13.3% and 6.9% respectively, for untreated and treated mangoes after 14 d of storage. At the end of storage, the control fruits were fully spoiled. However, 75% of irradiated chitosan coated fruits were not attacked by diseases. In another study, it was reported that the application of irradiated chitosan was effective on preservation of fresh fruits, as well as limiting the growth of fungi without affecting ripening characteristics of fruits. In a recent study, the use of a 150ku MW chitosan for coating over papaya was compared to chitosan of 300ku MW and an increased shelf life of papayas at ambient storage temperatures was found and the count of mesophilic bacteria, yeasts and molds were substantially decreased. They explained that chitosan of 150 ku has less organized structure with lower crystallinity and a rough surface with protuberances and cavities which improved its solubility in acid solvent, forming a more homogeneous solution and consequently a more homogeneous coating was obtained. Chitosan is a non-toxic, biologically compatible polymer. Its use for dietary applications is well known in many different countries and it has been approved by the Food and Drug Administration for use in wound dressings.

The use of natural preservatives in food preservation: The use of natural preservatives in food preservation is an emerging field driven by the growing demand for natural, sustainable, and clean-label food products. These bio-preservatives offer an alternative to synthetic additives, addressing concerns about their safety and environmental impact. The Natural Preservation Methods, Historically, various natural methods have been employed to preserve food, including drying, salting, smoking, and fermentation. These techniques have been used for centuries by different cultures worldwide to extend the shelf life of perishable foods and prevent spoilage. For example, fermentation not only preserves food but also enhances its flavour, texture, and nutritional value by promoting the growth of beneficial microorganisms. The Animals produce a wide array of bioactive molecules with potential preservative properties, including antimicrobial peptides, proteins, and lipid origin compounds. These compounds often serve defensive functions in the animal kingdom, protecting against microbial infections and environmental stressors. Additionally, some animals exhibit innate behaviours that aid in food preservation, such as burying food to prevent spoilage or secreting antimicrobial substances onto food items. Many animal origin compounds exhibit significant antimicrobial activity against a broad spectrum of microorganisms, including bacteria, fungi, viruses, and parasites. This antimicrobial efficacy makes them attractive candidates for use as natural preservatives in food products [175,176,177,178,179,180 and 181]. Furthermore, some animal origin compounds possess antifungal properties, which can inhibit the growth of Molds and yeasts commonly associated with food spoilage. Despite their potential benefits, the utilization of animal origin bio-preservatives in the food industry faces regulatory constraints and ethical considerations. Regulatory approval for new preservatives can be a complex process, requiring extensive safety assessments and efficacy studies. Additionally, there are concerns about the potential allergenicity of animal-derived compounds and their impact on consumers with specific dietary restrictions or preferences. Recent advancements in biotechnology and analytical techniques have contributed to the exploration of animal origin preservatives. These advances enable researchers to identify, isolate, and characterize bioactive compounds from animal sources more effectively. High-throughput screening methods and bioinformatics tools facilitate the discovery of novel antimicrobial peptides and proteins, accelerating the development of natural preservatives for food applications. Changing consumer preferences towards natural and sustainable products have also fuelled interest in animal-derived bio-preservatives [182,183,184,185,186,187 and 188]. The Consumers are increasingly seeking food products with cleaner labels, free from synthetic additives and

chemicals. Moreover, there is a growing awareness of the environmental impact of food production and the desire to support ethical and eco-friendly practices in the food industry. Animal origin bio-preservatives offer potential benefits for food preservation, challenges remain in their commercialization and widespread adoption. These challenges include regulatory hurdles, scalability, and cost-effectiveness. Additionally, there is a need for further research to optimize production methods, enhance efficacy, and ensure the safety of animal origin preservatives in various food matrices. Despite the challenges, the integration of animal-derived bio-preservatives into modern food preservation strategies holds promise for enhancing food safety and sustainability. Continued research and innovation in this field are essential to unlock the full potential of these natural compounds. Collaboration between academia, industry, and regulatory agencies is crucial to address existing barriers and facilitate the successful adoption of animal-derived bio-preservatives in the global food supply chain. In reviewing the significant background on animal-derived preservatives in food, several key findings have emerged, shedding light on their importance and potential in food preservation, The exploration of animal origin preservatives has revealed a diverse array of natural compounds sourced from various animal sources, including egg whites, milk, bee propolis, and crustacean exoskeletons. This diversity underscores the richness of natural resources available for food preservation. Each preservative possesses a unique chemical structure and mechanism of action that influences its effectiveness in inhibiting microbial growth. From enzymes like lysozyme to peptides like nisin and glycoproteins like lactoferrin, these compounds exert antimicrobial effects through mechanisms such as enzymatic hydrolysis, membrane disruption, and oxidative stress. The methods used to extract and purify these compounds play a crucial role in determining their efficacy and safety as food preservatives. Techniques such as chromatography, ultrafiltration, and enzymatic hydrolysis are employed to isolate the active ingredients and ensure their purity and stability [189,190, 191, 192,193,194 and 195].

The Applications and Effectiveness, the Animal origin preservatives find wide-ranging applications in food preservation, contributing to the extension of shelf life, inhibition of spoilage organisms, and maintenance of food quality and safety. These preservatives are used in various food products, including dairy, meat, beverages, and confectionery, where they serve as natural alternatives to synthetic additives. The natural origin of these preservatives aligns with consumer preferences for clean label products, driving the demand for natural and minimally processed foods. This trend has led to increased interest in the animal origin preservatives as effective and sustainable alternatives to synthetic additives. As the food industry continues to evolve and consumers demand cleaner, more sustainable food options, the utilization of animal origin bio-preservatives presents a promising opportunity for enhancing food safety, extending shelf life, and meeting regulatory requirements [196,197,198,199,200,201 and 202]. The preservation, the following recommendations are proposed for the effective integration of animal origin preservatives in food products, Prioritize safety and regulatory compliance throughout the development and commercialization process of animal-derived bio-preservatives. Conduct comprehensive safety assessments, including toxicity studies, allergenicity testing, and microbial risk assessments, to ensure the safety of bio-preservatives for consumer consumption. Work closely with regulatory agencies to navigate the regulatory landscape and obtain approvals for the use of animal origin bio-preservatives in food products [203,204,205,206,207,208 and 209].

The Consumer Education and The Transparency: Educate consumers about the benefits of animal origin bio-preservatives and the science behind their efficacy in food preservation. Emphasize the natural origin of bio-preservatives and their role in minimizing the use of synthetic additives and preservatives in food products. Provide transparent labelling and information about the source, composition, and safety of the animal origin bio-preservatives to build consumer trust and confidence in the products. Establish platforms for sharing best practices, research findings, and technological innovations related to the animal origin bio-preservatives. Encourage open dialogue and collaboration to address common challenges and accelerate the adoption of bio-preservation techniques in the food supply chain. Promote sustainable practices in the sourcing and production of the animal origin bio-preservatives to minimize environmental impact and support the ethical animal welfare standards [210,211,212,213,214,215 and 216]. Explore opportunities for utilizing by-products from food production processes, such as the animal skins, the bones, and organs, to extract bioactive compounds for use as natural preservatives. Implement sustainable sourcing practices and supply chain traceability to ensure the integrity and sustainability of bio-preservatives derived from the animal sources. The Continuous Improvement and Adaptation, embrace a culture of continuous improvement and adaptation to meet evolving consumer preferences, technological advancements, and regulatory requirements. Stay abreast of emerging trends and developments in the field of bio-preservation, including advances in biotechnology, food science, and sustainability. Continuously innovate and iterate on product formulations, production methods, and marketing strategies to remain competitive in the dynamic food industry landscape. In conclusion, the effective utilization of the animal origin bio-preservatives in food products requires a multidisciplinary approach, encompassing research and development, innovation, regulatory compliance, consumer education, collaboration, sustainability, and continuous improvement. By embracing these recommendations and working together towards common goals, the food industry can harness the potential of the animal origin bio-preservatives to create safer, healthier, and more sustainable food products for consumers worldwide [217,218,219,220 and 221].

Conclusion:

The exploration of the animal origin preservatives underscores the rich potential of natural compounds sourced from a variety of the animal origin substances. The substances like Lysozyme, Nisin, Lactoferrin, Propolis, Lactoperoxidase System, the Ovotransferrin, and the Chitosan, each offer unique mechanisms and properties that contribute to the preservation and safety of the food products. From the enzymatic action of lysozyme to the antimicrobial activity of nisin and lactoferrin, these preservatives serve as effective barriers against spoilage microorganisms, extending the shelf life of the food and maintaining its quality. Additionally, substances like the propolis and the chitosan provide antioxidant and antimicrobial benefits, further enhancing the food preservation efforts. The use of the animal origin preservatives aligns with the growing consumer demand for clean-label and natural food products. By harnessing these natural compounds, the food manufacturers can meet these preferences while ensuring the safety and integrity of their products. However, the integration of the animal origin preservatives into the food industry requires careful consideration of factors such as regulatory approval, the scalability, and ethical considerations. Further research and development are needed to optimize the production and application of these preservatives, ensuring their efficacy and safety in a wide range of food products. Overall, the study of the animal origin preservatives represents a promising avenue for enhancing food preservation, sustainability, and safety. By leveraging the unique properties of these natural compounds, we can address the challenges of food waste and foodborne illness while providing consumers with safe, nutritious, and flavourful food options for years to come.

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Conflicts of Interest

The authors declare no conflicts of interest.

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