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Green Nano-Composite Film Coating in Food Preservation

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THE HIGH demand for fresh vegetables, fruits, cheese, meats, etc. has led to advancements in technology, enabling year-round availability. However, these same technologies have increased the risk of microbial contamination. The processing steps of fresh vegetables as well as fruits can compromise their quality and make them susceptible to contamination and deterioration. Contamination could be by pathogenic bacteria, which can be present in soil or irrigation water. Bacterial spore-formers pose a threat to public health only when they have the opportunity to grow and develop vegetative cells. Meanwhile, fungi are the main contributors to the decay of fruits and fruit products, as they can rapidly grow within tissues and adjust their attack payload. The use of nanotechnology in the development of bioactive packaging materials based on natural biopolymers, such as chitosan, introduced promising results in enhancing the barrier properties of the films. Furthermore, the incorporation of nanoparticles in chitosan films has demonstrated moderate antibacterial behavior and biodegradability, making them suitable for antimicrobial packaging and wound dressings. It is proposed that nano-composite films can improve the functional properties and extend the shelf life of fruits and vegetables. Moreover, they are capable of preserving food at nutritional value and at the same time, they are considered safe and sometimes edible.

Keywords: Alginate films, Chitosan films, Food preservation, Microbial contamination, Microbial susceptibility, Nano-composite films.

Introduction

Microbial contamination of food is a major concern for the food industry, considering the waste of rotten products and the public health risks associated with foodborne infections. This is why food quality assurance (QA) systems are essential for manufacturing products free from microbial threats. Fruits and vegetables could be infected by pathogenic microorganisms, at any stage of their growth, from field or orchard growth to harvesting, post-harvest handling, processing, and distribution. Pathogens including fungi, bacteria, viruses, and parasites are harmful to humans and can be present in irrigation water or the soil in which food is grown. However, on a global scale, bacteria pose the greatest threat of dangerous disease and the highest risk of infection. The information is restricted to a short list of pathogens found in fresh foods that have been identified or suspected to be present in human cases of gastroenteritis (Gerba & Smith, 2005).

The use of Food preservation extends from ancient times. Preservation restrains development of microbial growth (Seetaramaiah et al., 2011). There are many goals for the food preservation process. One goal is maintenance of food taste, flavor, quality, texture, nutritional value and quality. A second goal is to minimize wasting the excess of food. The third goal would be elongating the shelf-life of foods even during transport. Moreover, food materials could be easily handled (Devi et al., 2015).

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Innovative techniques of preservation are being developed in order to meet consumer demands. The nanofood sector is emerging as a viable solution to meet the needs of a growing customer base, as traditional food technologies are becoming less effective in meeting the demands of consumers (Chellaram et al., 2014). In recent years, researchers have been increasingly interested in creating sustainable food-preserving products made of renewable and biodegradable materials (Gupta et al., 2022). Efforts are ongoing to enhance the efficacy of edible food-preserving films through the use of functional additives, such as phenolic compounds and essential oils, besides nanoforms (Nair et al., 2020).

Sources of pathogenic microorganisms

Bacteria that produce spores are known as spore-forming pathogens. Spores produced by harmful bacteria often contaminate crops and fruits, such as those found in soil containing harmful bacteria including *Bacillus cereus* (*B. cereus*), *Clostridium botulinum*, or *Clostridium perfringens* (*C. perfringens*). It was found that *B. cereus* and *C. perfringens* existed in approximately a third of the tested hundred vegetable samples, which were mostly in the form of salads, obtained from United Kingdom retail stores. Only when the produce is processed in a manner that permits the growth of spores and the growth of vegetative cells is there a threat to public health posed by these spore-forming bacteria (Bader et al., 2012).

Fungi are able to rapidly expand into tissues, and they have evolved sensing systems that allow them to adjust their attack payload by acidifying and alkalinizing the environment. Fungi, contrary to bacteria, possess the capability of rapidly developing within tissues. As a result, fungi are invariably the primary contributor to the decay of fruits and fruit products (Prusky & Yakoby, 2003).

Post-processing technology can help preserve food quality during its shelf life. Biocidal chemicals can be directly incorporated into food or the environment that surrounds it to provide antimicrobial benefits (Carbone et al., 2016). In this regard, active antimicrobial packages have been developed for a variety of food products, especially those containing active biocides, which may enhance product performance, reduce shelf life, and prevent microbial degradation (Mahdi et al., 2012).

Improving shelf life of fresh foods

Fresh fruits and vegetables

Fruits and vegetables are one of the most widely consumed foods in the world. Where land is available, families can grow fruits and vegetables. In other cases, production is purchased from local farmers and grocery stores for home use or as a part of meals served at restaurants and other eateries (Ouf et al., 2019). Continued advances in agronomy and processing, maintenance, distribution, and marketing technologies have enabled the produce industry to provide nearly all types of high-quality fresh produce year-round to consumers who want it and can afford it. Nevertheless, some of these same technologies have increased the risk of human disease caused by a broad spectrum of harmful microbes (Ramos et al., 2013).

Fruits and vegetables that are whole and intact are protected from microbial contamination and loss of quality due to the presence of natural barriers (cuticles, skins, rinds, etc.) (Baldwin, 1994; Del Rosario & Beuchat, 1995). Fresh cut fruits and vegetables have been in the spotlight lately because people have more awareness of importance of healthy eating and have less time to prepare food (Olivas & Barbosa-Cánovas, 2005). Consumers' preferences change in terms of convenience without any loss in quality (Wong et al., 1994). Fruits and vegetables that are minimally processed go through many steps of preparation, from washing and peeling to cutting, slicing, and coring. These steps remove barriers, cause lesions of the tissues, compromise the quality of the fruit, and inflict wounding stress (Soliva-Fortuny & Martín-Belloso, 2003). This makes the product susceptible to contamination, enzyme browning, unwanted volatiles, and texture changes (Olivas & Barbosa-Cánovas, 2005). Deterioration, spoilage, and pathogenic bacteria can be avoided by improving the natural barriers or by replacing them with artificial barriers around the product (e.g., edible film, coating applications) (Watada & Qi, 1999; Yahia, 2009).

The delay in respiration and physiological processes is necessary for extending the shelf life of vegetables and fruits (Parreidt et al., 2018). In this way, the coatings and films that modify the gas transport can be used for fresh produce coating (Perez-Gago et al., 2003). In particular, the polysaccharide-based coatings are used to reduce respiration in fruits and vegetables because of their selective permeability to the O_2 & CO_2 gases. The swelling ratio along with the solubility of the alginate films in terms of water are very important properties. Alginate films resist being dissolved in water, so they can be used to coat high moisture fresh cut fruits (Parreidt et al., 2018).

Poultry, meats, and seafood

There are a number of problems that can occur with meats, poultry, and seafood products over their shelf life. These problems include moisture loss and its impact on texture, color, and taste, product juice dripping (clean-up losses), lipid oxidation and brown discoloring, spoilage due to microorganisms, loss of volatile flavor, and/or foreign odor gathering (Pringle et al., 1996; Ambardekar, 2007).

Earle (1968) designed a patent formulation of an alginate-based coating, which is known commercially as Flavor-Tex®. Meat and seafood as well as poultry products were submerged in a water-soluble dispersion of algin and carbohydrate (mono- and/or disaccharide sugar). They were then gelled with a solution of $CaCl_2$-CMC (addition of CMC to calcium crosslinking bath decreases gelling time and $CaCl_2$ concentration requirement) (Earle & Snyder, 1966; Earle, 1968). Gelatinized alginate was first patented in 1956 in Norway for the block freezing of fishery products. In this way, the harmful effects of direct contact with these products have been removed (Oyvind & Alf, 1956).

Cheese

The main loss of quality occurs during the storage period. This can be broken down as follows: (1) microbial contamination, (2) loss of moisture, and (3) the development of an undesirable off-flavor and other organoleptic characteristics. It is important to emphasize that adding an extra calcium for crosslinking purposes is not a necessary step in the cheese coating process due to the presence of calcium itself (Costa et al., 2018).

Edible coating and film applications have been used and studied in various cheese types as a packaging system to avoid loss of quality. Zhong et al. (2014) examined the performance of 3 coating materials (sodium alginate (SA), chitosan, and soy protein isolate) with 4 different application methods on mozzarella cheese. Alginate coated samples had better overall quality due to higher wettability of the product surface. Lucera et al. (2014) showed similar antimicrobial properties for potassium sorbate and sodium benzoate, as well as lactate and ascorbate salts of calcium, which were used as alginate coatings for mozzarella cheese. The main difference was sensory. Artiga-Artigas et al. (2017) added ginger essential oil (EO) to the composite coating of the alginate-whey protein, resulting in kashar cheese that is lower in acidity and higher in fat.

Nanotechnology in food preservation

Nanotechnology is an emerging, interdisciplinary field of modern research that deals with materials and interactions at the molecular (or even atomic) scale, which ranges from 0.1 nm to 100 nm along the boundaries of physical, chemical, and biological sciences. Due to their small size, these materials are characterized by different chemical and physical properties (magnetic, behavior, optical, chemical, etc.) compared to larger-scale materials that give permission for novel applications (Nikolova et al., 2020).

Historical background on nanotechnology

The origin of modern science and technology lies largely in human creativity and visualization. The emergence of nanotechnology comes out of these ambitions. Although human beings have encountered nanoparticles throughout history, nanotechnology came into being significant during the industrial revolution (Leon et al., 2020). The term nanoparticle examination is not new. In fact, it was not until 1925 that the term was coined by a Nobel laureate in chemistry, Siggmondy, who coined the phrase “nanometer” and was one of the first to utilize a microscope to measure the size and shape of particles like gold colloids and to define the size of particles (Nikolova et al., 2020).

Richard Feynman—the father of nanotechnology today—was awarded the Nobel Prize in Physics in 1965. He gave a speech at Caltech’s American Physical Society meeting in 1959 titled “There’s Plenty of Room at the Bottom.” He introduced the idea of manipulating something at the atomic level. Feynman’s theories were recently proven correct. This revolutionary intellect paved the way for the new ways of thinking. Because of these factors, he is viewed the founder of recent nanotechnology (Fanfair et al., 2007).

Firstly, a Japanese scientist developed carbon nanotube technology, then the field of nanotechnology advanced dramatically (Hulla et al., 2015). At the beginning of the 21st century, there was a lot of interest in nanosciences and nanotechnology. It was because of Feynman's influence in the United States and his idea of handling things at atomic levels that national science programs got started. Many United States presidents wanted to fund research into this new technology, and three years later, President Bush enacted what is known as "Nanotechnology Research and Development Act." The Act created the NTI (National Technology Initiative) making nanotechnology a national priority (Tolochko, 2009).

Types of nanomaterials

Different classification methods have been used to classify nanomaterial compounds based on their sizes, shapes, phase compositions, and other parameters as shown in Fig. 1 (Nikalje, 2015). Each nanoparticle has a distinct physical structure (shape, volume, surface, permeability, and magnetism), which has a remarkable impact on its properties and distinguishes it from materials with large surface areas (Ventola, 2012).

All nanoparticles are, by definition, in the range between 0.1 to 100 nanometers. Because of their small size, they can pass through biological membranes from one side to the other. This allows for better medication delivery, better solubility, better bioavailability, and longer circulation inside the living organism (Tolochko, 2009). It also increases the total surface area and conjugation of nanoparticles that have electrostatic surface or larger molecules. However, the higher permeability & bioavailability of nanoscale materials increases the risk of permeation & accumulation in specific tissues & cells, and the presence of a gene that in some cases can turn a cell into a tumor cell (Nikolova et al., 2020).

Nanotechnology in food

About 1.3 billion tons of foods produced for the human consumption go to waste every year due to rotting and inadequate management of the world's food supply. On the other hand, according to World Food Program estimates, the waste of edible food costs the Australian economy over $8 billion a year (Nile et al., 2020). There are now four main food safety concerns: food-borne infections, food adulteration, genetically modified foods, and the presence of a significant number of chemicals and pollutants in the food supply (Yuan et al., 2023). Accordingly, as the world continues to experience a shift in food consumption patterns, an increasing number of individuals are beginning to question the safety and quality of the food they are consuming. This is due to a variety of factors, such as the mass production and mass manufacturing of agricultural products, as well as changes in consumer lifestyles. Furthermore, there has been a surge in the importation of food goods. To save both money and time, many people are now turning to pre-packaged meals and quick-service dining establishments. Additionally, the introduction of new types of transmission vehicles (such as raw and fast food) has led to the appearance of new food-borne diseases and the resurgence of previously identified infections (Ball et al., 2019). In addition, new pathogens are introduced into our food every year. The United States government estimates that the annual costs of medical treatment, loss of productivity, and mortality due to disease are estimated to amount to $55.5 billion in 2020 (Kearney, 2010). Agri-food companies are beginning to recognize the importance of nanotechnology to extend product shelf life, enhance food safety and quality, regulate transportation and storage processes, and create nanosensors to detect food contamination (Ranjan et al., 2016). For instance, essential oils contain bioactive chemicals with antibacterial properties (Chellaram et al., 2014). Essential oils have been proved capable of prolonging the shelf life of foods as well as other products (Duncan, 2011). On the other hand, when the bioactive compounds are present in the form of nanoemulsions, they have higher antibacterial activity than their conventional forms. The reason for this is that the size of the nanoemulsions droplets is significantly smaller than the size of conventional droplets. Nanoemulsions containing carvacrol and cinnamaldehyde as well as limonene were tested against three different species of microorganisms: *Saccharomyces cerevisiae*, *Lactobacillus delbrueckii*, and *Escherichia coli* (*E. coli*). The antibacterial properties of these nanoemulsions were relatively strong against all three species. Silver, zinc, selenium, copper, cerium, and gold nanoparticles have antibacterial properties and are used in a wide variety of applications (Yuan et al., 2023).

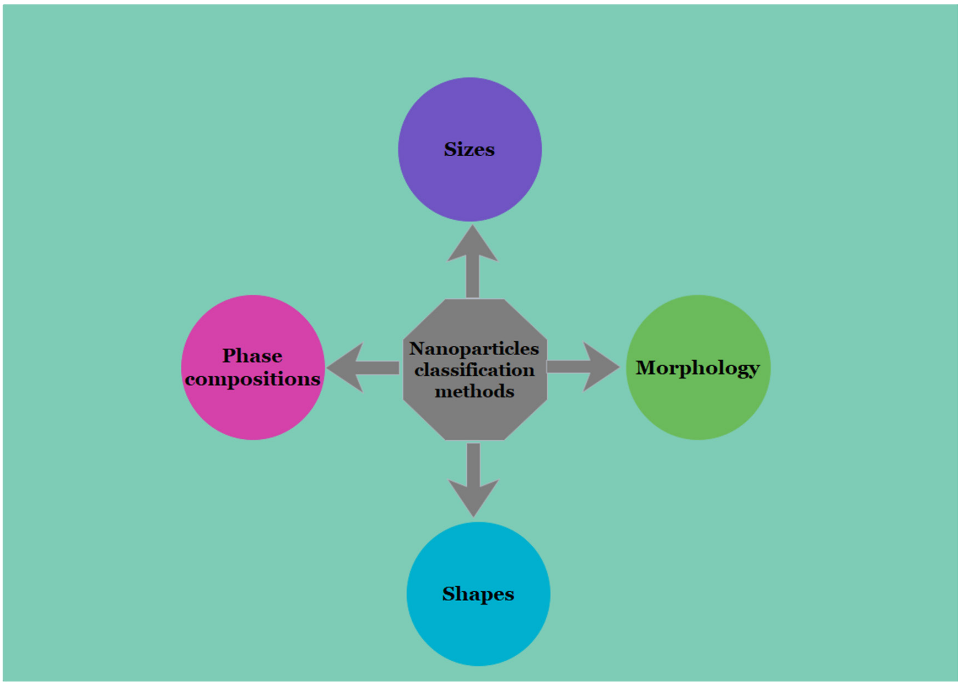

Fig. 1. Classification methods of nanoparticles

Attributes of some nanoparticles

A wide range of infectious microbes, including viruses, fungi, yeasts, and bacteria, are effectively inhibited by silver nanoparticles (AgNPs). In addition, AgNPs have low melting temperatures and are easily processed (Abd El-Ghany et al., 2023), which makes them a good choice for food packaging (Nakamura et al., 2019). According to Botha et al. (2019), the cytotoxicity effects of AgNPs synthesized from the plant extract on H4IIE-luc (rat hepatoma) cells and HuTu-80 (human intestine) cells were determined at a dose of 50µg/mL. While AgNPs migrate relatively slowly to food products and stimulants, their toxicity becomes an issue at higher concentrations when employed in food packaging. Even more concerning is the fact that environmental levels of AgNPs have risen due to the widespread use of these particles in many industrial processes. According to Yu et al. (2013), these AgNPs can readily move into soil and liquids as Ag^+. Once there, they may penetrate microorganisms and animals, leading to harmful impacts on the entire ecosystem.

The industrial sector makes heavy use of copper nanoparticles (CuNPs) as antimicrobials, catalysts, surfactants, sensors, and antifouling coatings. However, as their use increased, CuNPs became harmful, particularly to aquatic creatures (Wu et al., 2020). This toxicity impact is the fundamental reason why CuNPs have not been extensively studied in food packaging. Also the migration studies on CuNPs are few (Ahari & Lahijani, 2021).

The inert and harmless nature of gold nanoparticles (AuNPs) along with their medicinal, oxidative catalytic, and antibacterial capabilities makes them ideal for use in food packaging (Paidari & Ibrahim, 2021). Due to the high expense, very little study has been conducted on AuNPs.

To prevent cell damage, control thyroid function, and aid in immune system regulation, Se is a necessary mineral that proteins must include (Srivastava & Mukhopadhyay, 2015). The daily recommended intake of selenium for males is 56.0mg, but the average male body contains 82.0mg (Mosallam et al., 2018). For the simple reason that selenium is an essential mineral for healthy bodily functions, in the next paragraph, we will discuss selenium in more detail.

Selenium nanoparticles (SeNPs) have the potential to repair other nanoparticles flaws because they are nontoxic, highly photoconductive, and able to catalyze the hydration and oxidation reactions. SeNPs are produced through a range of physical processes, such as direct physical vapor degradation, hydrothermal treatment, and ultrasonic irradiation. Chemical processes involve reduction and oxidation of selenium dioxide, selenates, or selenites as shown in Fig. 2. Reduction is done by reducing agents including, for instance, hydrazine, sodium ascorbate, or glycol. The green method involves the synthesis of SeNPs with the use of plants, fungi, or bacteria (Langi et al., 2010).

SeNPs have been shown to have anticancer and anti-inflammatory properties, as well as anti-diabetes and antioxidant properties. They have also been shown to possess antimicrobial properties against pathogens such as bacteria, fungi, and yeasts, according to numerous studies published over the past few years (Vahdati & Moghadam, 2020).

SeNPs are widely used for a variety of medicinal purposes, including antioxidant activity, drug delivery system, and anticancer activity. The anticancer activity is mainly due to its prooxidative activity, which causes the formation of different reactive oxygen species in cells, resulting in damage to mitochondria and endoplasmic reticulum, resulting in DNA damage. SeNPs can scavenge free radicals, in vitro, by size-dependent mechanisms (5 nm to 200 nm). A study showed that SeNPs with smaller sizes (5 to 15 nm) had a greater ability for scavenging free radicals and prevent DNA damage (Shnoudeh et al., 2019).

Because of their unique characteristics, SeNPs are widely used in consumer products, the healthcare sector, food storage, the environment, and biomedical applications. Several aspects of SeNPs use have been discussed in numerous books and reviews.

Antimicrobial effect of selenium nanoparticles

SeNPs are nontoxic, highly bioavailable, and biocompatible (Solaiman et al., 2017). SeNPs are antimicrobial compounds, meaning they inhibit bacterial, fungal, and viral growth. SeNPs inhibit Gram-positive as well as Gram-negative bacteria equally. SeNPs also have anti-fungal properties, as they inhibit spore germination. Furthermore, SeNPs have the capability to break down microbial biofilm (Wadhwani et al., 2016). In a study on biologically synthesized SeNPs, it was demonstrated that SeNPs stopped the growth of the bacteria *Pseudomonas aeruginosa (P. aeruginosa)*, *Streptococcus pyogenes*, *Staphylococcus aureus (S. aureus)*, and *E. coli* at the respective doses 100, 100, 100, and 250µg/mL (Jolly et al., 2020).

SeNPs are chemically synthesized using vitamin C reducing SeO_3^{2+}. SeNPs demonstrated antibacterial activity against ten commonly transmissible food-borne bacteria. Colony-forming unit (CFU) assays showed SeNPs inhibit growth of *Staphylococcus epidermidis* and *Listeria monocytogens* (*L. monocytogenes*) at a starting dose of 0.05 µg/ mL. Higher concentrations were needed to inhibit the growth of *Vibrio alginolyticus*, *S. aureus*, and *Salmonella enterica* (Yuan et al., 2023).

SeNPs also inhibit the growth of the fungal pathogen *Aspergillus clavatus* when administered at a 500 µg/mL dose (Jolly et al., 2020). In other studies, selenium nanoparticles were extracted from the leaves of *Amphipterygium glaucum* and the flowers of *Calendula officinalis*. SeNPs demonstrated antifungal activity against two commercially important plant pathogenic fungi: *Fusarium oxysporum* and *Colletotrichum gloeosporioides* (Lazcano-Ramírez et al., 2023).

It was previously shown that SeNPs had high antifungal activity through inhibiting spore germination (Joshi et al., 2019). Troni et al. (2021) reported changes in *Fusarium proliferatum* morphology and coloration with the application of various forms of selenium, including Na_2SeO_3, Na_2SeO_4, SeMet, and SeCys. Meanwhile, during evaluating *Pleurotus ostreatus*' mycelial growth, there was a strong odor in the culture media containing SeNPs and this could be caused by the formation of volatile compounds (dimethyl selenide, $(CH_3)_2Se$). Dimethyl selenide is considered one of the fungal mechanisms invoked to attenuate the toxicity of SeNPs, according to Silva et al. (2019). The presence of the genus *Fusarium* mycotoxins zearalenone, trichothecenes, deoxynivalenol, and fumonisins could also be a sign of the secondary defense mechanism exerted by the fungus to protect itself against the reactive oxygen species elicited by the SeNPs (Hu et al., 2019).

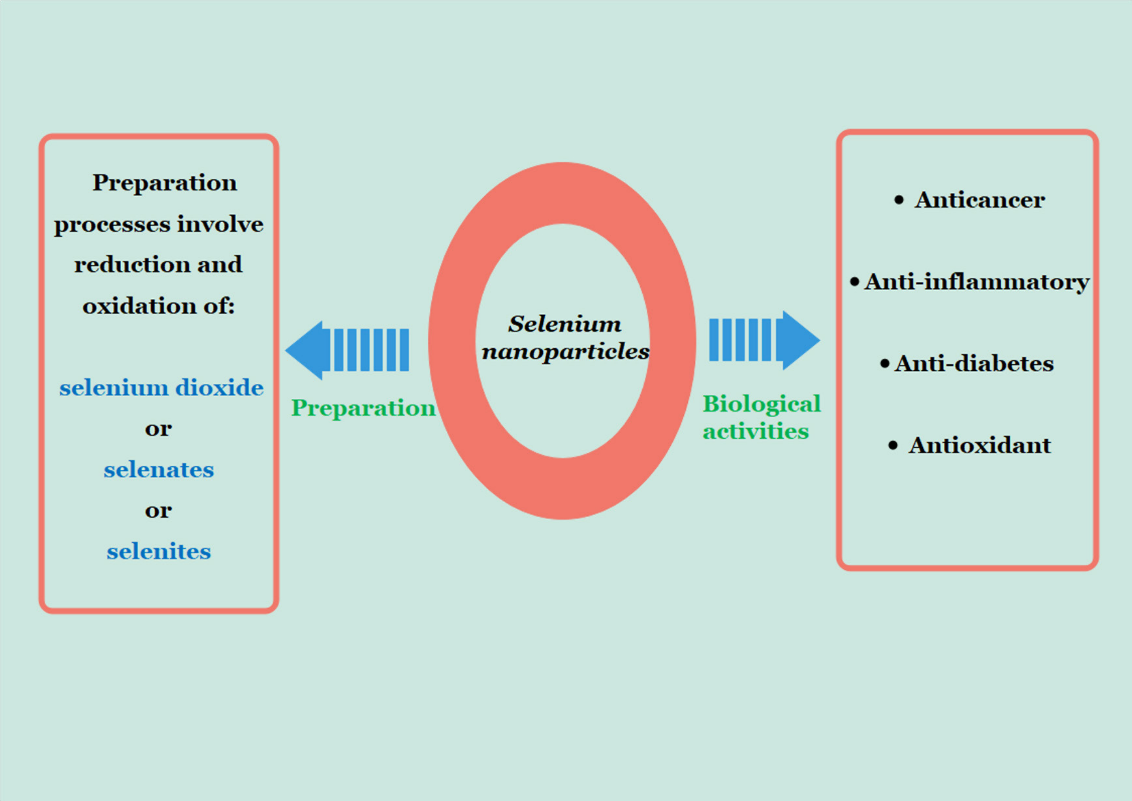

Fig. 2. Preparation and biological activities of selenium nanoparticles

Natural polymers and their application in food packaging

Biodegradable or natural polymers could be obtained by physical or chemical extraction from their natural environment as appears in Fig. 3. Biodegradable polymers are those that are naturally derived from a polymerase enzyme and have a natural biodegradability. In some cases, the breakdown of natural polymers may be catalyzed by depolymerases to maintain a natural evenness (Gupta et al., 2022). One of the most frequent ways to extend the shelf life of food after harvest is by using biopolymers such as chitosan, alginate, gelatin, soy protein, cellulose, pectin, starch, dextran, and gums to make films and coatings. Compared to petroleum-based and non-biodegradable polymers, such as polyvinylchloride, polyamide, polystyrene, polyethylene, and polypropylene, these materials are significantly better (Basumatary et al., 2022). A variety of studies have demonstrated the potential of edible film/coatings based on chitosan or alginate to maintain the quality characteristics of fruits and vegetables as shown in Fig. 3. The findings indicate that these films/coatings are acting as a protective layer on surface of fruits and vegetables, resulting in increased moisture and water retention. Additionally, they create favorable microenvironmental conditions by optimizing the concentration of gases and delaying ripening (Nair et al., 2020).

The emergence of functional additives has opened up a new area of application for improving the functional properties of films based on alginate or chitosan. These compounds are now becoming increasingly popular as an ingredient in edible films/coats for the purpose of extending shelf life (Kumar et al., 2020). Several reviews provide an in-depth analysis of recent studies on the mechanisms of action of functional additives, as well as their ability to prolong the quality and shelf life of a variety of fruits, such as pear, guava, and blueberries and vegetables such as capsicum, cucumber, and mushroom. Additionally, the principles behind additives' antimicrobial and antioxidant activities in preventing the spoilage of food are also discussed. The efficacy of phenolic compounds, essential oils, and nanoform substances in extending the shelf life without compromizing the nutritional and safety properties of fruits and vegetables remains to be studied further (Aristizabal-Gil et al., 2019). Films of chitosan or alginate will be discussed in the following sections.

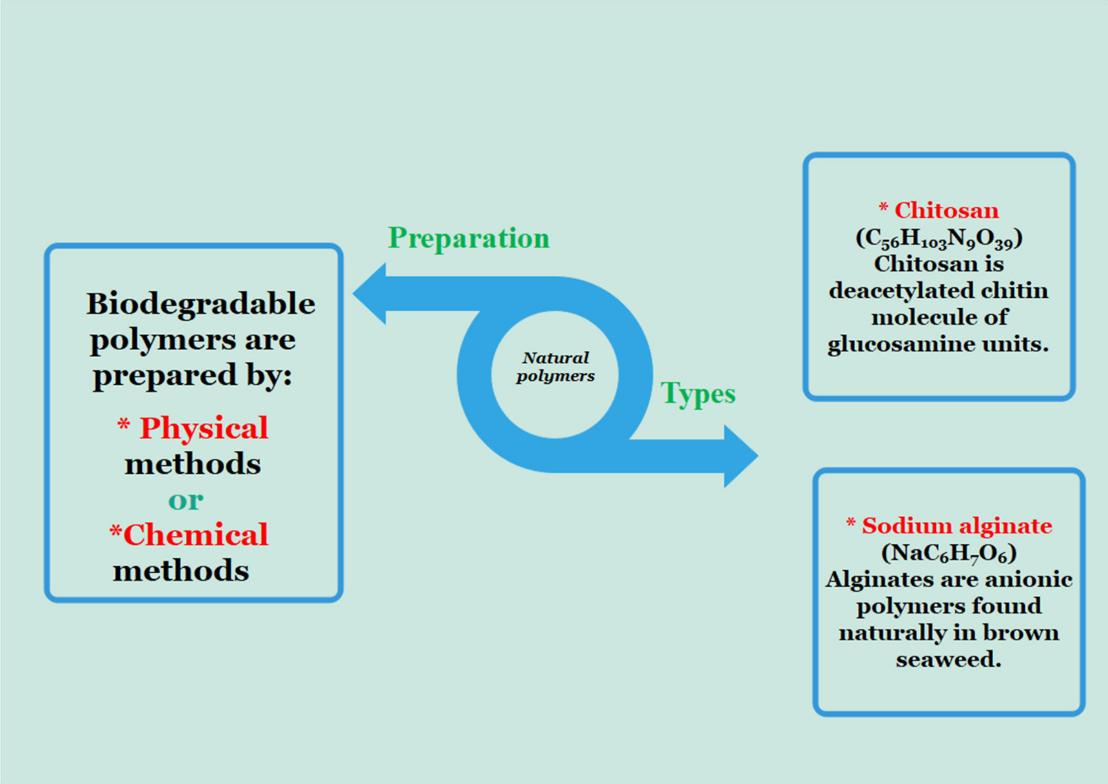

Fig. 3. Preparation and types of biodegradable or natural polymers

Chitosan

Chitosan is a deacetylated chitin molecule; in this deacetylated process, certain N-acetyl glucosamine moieties are transformed into glucosamine units. Chitosans are the only polycations in nature. The charge density of the polymers depends on the deacetylated state of the chitin molecule and on the pH of the medium. Solubility depends on the state of the acetylated state and on molecular weight of the polymer. The oligomers of chitosans are soluble in a broad range of pHs (acidic to basic) whereas higher molecular weight (MW) samples are soluble only in acidic water (acidic media) even at high degrees of deacetylating state (Aranaz et al., 2021).

Therapeutic polymers may be derived from the biological activities of Chitosan or its derivatives, which possess a variety of anti-tumor, antimicrobial, antioxidant, anti-inflammatory, and other properties. It is noteworthy that, as of the present time, Chitosan is only accepted as an excipient by regulatory agencies, rather than as a drug to treat diseases (Aranaz et al., 2021).

Chitosan is used for food packaging practices either as packaging films or as coatings directly applied to food material. Various researchers around the world have explored both approaches. In this section, we will discuss the fabrication methods of chitosan films and chitosan coatings. Chitosan film fabrication techniques have been developed depending on the intended use of the material in food packaging. Current fabrication methods include direct cast, coating, layering, and extrusion and can be applied either to neat films without any polymers or to films blended with polymers/biopolymers like polysaccharides and proteins as well as synthetic plastics (Priyadarshi & Rhim, 2020).

Direct casting is the most commonly used method because it is the simplest method for manufacturing a biopolymer film. Solution cast films have a lot of potential as packaging materials for food. They are used in various food products and have been found to maintain food quality and increase shelf life. While casted films have significant benefits and extraordinary properties, they also have some drawbacks. In addition, small variations in formulation can have a remarkable impact on film properties. Therefore, formulation design while using a direct casting method should be carefully considered. Furthermore, solution casting is currently not being used on an industrial scale, as it is uneconomic and time consuming (Shankar & Rhim, 2018; Mujtaba et al., 2019; Zhang et al., 2019).

As for coating, Chitosan thin films are often applied directly to surfaces of food materials (fruits, vegetables, and meat products) or on surfaces of packaging materials (González-Saucedo et al., 2019; Ortiz-Duarte et al., 2019). When direct coating is applied, the objective is to immobilize the polymer containing active substances directly on surface of foods in order to protect it from environmental factors such as microbes. Direct coatings can also change gas permeability coefficient of fruits and vegetables, which can affect respiration rate, thus affecting shelf life. Direct coatings are eco-friendly, biodegradable, and in most cases edible. The process includes five steps: first, preparing chitosan formulations containing active ingredients such as antioxidants, antimicrobial, or strengthening agents, second, preparing food material for coating through treatments such as washing, slicing, pasteurization, irradiation, or heating, third, using a sterile tool to evenly distribute chitosan on the food material surface, fourth, drying in specified conditions in a sterile environment such as a laminar airflow cabinet, and fifth, packing and storing in a refrigerator or a vacuum chamber. Indirect coating on the packaging material's surface is designed to create multi-layer functional packaging material that has better properties compared to single-layer packaging. The chitosan covering on the paper's surface not only gives the packaging material antimicrobial activity but also reduces its moisture permeability, without significantly affecting its mechanical properties (Khwaldia et al., 2014).

In the dip-coating process, the food material is submerged in chitosan-based solutions, resulting in a uniform film forming on surfaces of foods. The film formulation is highly dependent on the surface wetting capacity, processing time, and draining time. This process includes (1) preparing chitosan solutions of definite pH; (2) selecting and processing the food sample that will be coated; (3) submerging the food sample into the solution for specified time (30 s to 30min), depending on the food type; (4) emersion of the food sample, draining of the excess solution, and drying; (5) placing food in trays/plastic support; (6) packaging and storing. The dip-coated food samples were found to be less likely to suffer from chilling injury and had less color variations and decay resulting in better quality and longer shelf life. In general, the dip-coating process is simple, time-efficient, and not requiring large equipment while maintaining high levels of food preservation and quality (Zhang et al., 2015).

Sodium alginate

Alginates are anionic polymers found naturally in brown seaweed. Alginate has been extensively studied and used in a variety of biomedical applications due to its bioavailability, low toxicity, low cost, and gentle gelation by the add-on of divalent cations like Ca^{2+} (Gombotz & Wee, 2012). D-mannuronate used to be the main ingredient in SA, but it was not until L-guluronate residue was discovered. Fractional precipitation using manganese (Mn) and calcium (Ca) salts showed at a later date that alginate is actually a block copolymer and that the proportion of guluronate to mannuronate depends on the source (Haug et al., 1959). Alginate is currently known as a whole linear copolymers family which contains (1,4)-linked β-D-mannuronate (M) blocks and residues of α-L-guluronate (G). The blocks constitute consecutive G residues and consecutive M residues, besides alternating residues of G and M (Lee & Mooney, 2012).

Antimicrobial activity of chitosan and alginate-based edible films

Antibiotic resistance is considered a major threat to public health, which is why there is a pressing need to develop new antimicrobial alternatives to antibiotics. Chitosan, its derivatives, and chitooligosaccharides act as antimicrobials against various microorganisms, including bacteria, yeasts, and fungi (Raafat & Sahl, 2009). Examples for these susceptible microorganisms are *Aeromonas hydrophila*, *Flavobacterium columnare*, *Edwardsiella ictaluri*, *Candida albicans (C. albicans)*, *Bacillus cereus*, *S. aureus*, *Salmonella typhimurium*, *E. coli*, and *Vibrio cholerae* (Aranaz et al., 2021). Chitosan appears to inhibit the growth of bacteria because bacteria can grow after removing the polymer from the medium. This is important as resistant populations may develop if the cells can adapt to the chitosan (Raafat et al., 2008).

A study was conducted to investigate the properties of four distinct types of nano-composite films based on chitosans. The film properties, as well as antimicrobial functions, of these films were tested. The compositing of nanoparticles (e.g., unmodified montmorillonite, organically modified nanosilver, or silver zeolite) was used to improve the mechanical and water vapor barrier properties of the films. The mechanical and vapor barrier properties were significantly enhanced ($P < 0.05$) when compared to those of control films. The antimicrobial ability of nanoparticles was also altered depending on the type of nanoparticles used. The results of this study suggest that nanotechnology may be applicable to the development of bioactive packaging materials based on natural biopolymers with additional biodegradability (Rhim et al., 2006).

The solution combustion method was successful in the synthesis of the NiONPs. The NiONPs were incorporated into chitosan films and the physicochemical and functional properties of the nano-composite films were examined. The doping of the NiONPs on chitosan macromolecules, as well as the fabrication of a genuine nano-composite, was confirmed by the results of XRD and FT-IR analyses. The uniform distribution of the NiONPs at chitosan W/W concentrations of 6% and 3% was also confirmed by observations from the FE-SEM test. When the NiONPs content reached 9%, the moisture absorption, water volatile potential (WVP), and surface wettability of the chitosan film were reduced, resulting in a decrease in mechanical, water barrier, and thermal characteristics of the films. This research examined the effects of aggregated nanoparticles at higher concentrations on all of the characteristics of chitosan films. The addition of nanoparticles had an adverse impact on the transparency of the films, with more opaque films being produced by the incorporation of nanoparticles. The primary functional characteristics of the films produced were the photocatalytic activity and the antimicrobial properties. The produced films exhibited a strong pigment photodegradation capability. Furthermore, the films demonstrated good antibacterial activity against Gram-positive as well as Gram-negative bacteria. All previous studies of the photocatalytic and antimicrobial capabilities of NiONPs have been conducted using the powdered form. However, in this study, chitosan was used as a biopolymer support for immobilization of NiONPs. In summary, this research has demonstrated that a chitosan-NiONPs nano-composite film is multifunctional, suitable for use in active food packaging, and also applicable for photodegradation applications, including waste water treatment (Marand et al., 2021).

The chitosan films containing silver nanoparticles showed moderate antibacterial behavior against *S. aureus* and *E. coli*. The chitosan films were biodegradable when put in soil, supporting their eco-friendliness after use. These chitosan films can be used in antimicrobial packaging and in wound dressings (Batubara et al., 2023).

Citrus essential oils were coated with chitosan and methylcellulose in another investigation. According to research by Randazzo et al. (2016), this substance reduced the ability of *L. monocytogenes* to infect several fruits.

Sodium alginate-based films with high antibacterial and antioxidant activity were prepared and modified with polyphenol rich kiwi peel extract-bioreduced silver nanoparticles. The tested NPs demonstrated excellent antibacterial activity and a characteristic hypotoxicity effect. The antibacterial test showed that treatment of silver NPs can cause disruption of cell membrane leading to leakage bacterial contents and finally resulting in bacterial death. In addition, silver NPs were incorporated into the SA film-forming matrix (Sun et al., 2021).

The nano-composite films of alginate/SiO_2 were developed with glycerol as a plasticizer by blending nano-SiO_2 solution. By increasing SiO_2 content in UV radiation, transmittance decreased. This is desirable in food packaging (Yang & Xia, 2017).

Edible films are becoming more popular due to their ability to extend the shelf life and minimize the use of conventional packaging materials. New eco-friendly edible films that contain graphene glycerin, SA, and nanoplatelets have been developed (Taşkın Çakıcı et al., 2023).

Arroyo et al. (2020) found that coating zinc oxide nanoparticles with edible alginate and chitosan extended the storage life of guava fruits. Table 1 displays researches on the use of chitosan and alginate that incorporates natural compounds and nanoparticles as antimicrobial biopolymers.

TABLE 1. Antimicrobial activities of chitosan and alginate-based edible films

Source of natural polymers	Susceptible microorganisms	References
Chitosan with nanosilver and silver zeolite	*S. aureus ATCC-14458, Leuconostoc monocytogenes ATCC-19111, Salmonella typhimurium ATCC-14028,* and *E. coli O157:H7*	(Rhim et al., 2006)
Chitosan	*S. aureus SG511*	(Raafat et al., 2008)
Chitosan-AgNPs	*Colletotrichum gloeosporioides*	(Chowdappa et al., 2014)
Chitosan with *Citrus* essential oils and methylcellulose	*L. monocytogenes*	(Randazzo et al., 2016)
Chitosan-TiO_2	*E. coli, S. aureus, C. albicans*, *and Aspergillus niger*	(Zhang et al., 2017)
Chitosan-SiNPs	*Botrytis cinerea*	(Youssef et al., 2019)
Chitosan-$CuSiO_2$NPs	*Botrytis cinerea*	(Hashim et al., 2019)
Chitosan with *Byrsonima crassifolia* extract	*Alternaria alternata*	(González-Saucedo et al., 2019)
Chitosan-Fe_2O_3	*Rhizopus stolonifer*	(Saqib et al., 2020)
Chitosan-NiONPs	*S. aureus* and *Salmonella typhimurium*	(Marand et al., 2021)
Chitosan with cinnamaldehyde, *Nigella sativa* or blackseed oil, and AgNPs (CH/CIN/BSO/AgNP)	*S. aureus* (ATCC 29213) and *E. coli* (ATCC 25922)	(Batubara et al., 2023)
SA-AgNPs	*E. coli* and *L. monocytogenes*	(Shankar et al., 2016)
SA with bacteriophages (EC4 and φ135 phages) and cinnamaldehyde (CNMA)	*E. coli* and *Salmonella enteritidis*	(Alves et al., 2020)
SA-SNPs	*E. coli* and *L. monocytogenes*	(Priyadarshi et al., 2021)
SA with (Ag@PE NPs)	S. *aureus* and *E. coli*	(Sun et al., 2021)
SA with AgNPs and gelatin (GL) (SA/GL/Ag film)	S. *aureus* and *E. coli*	(Li et al., 2022)
SA with graphene nanoplatelet and glycerin	*E. coli* ATCC 25922 and *S. aureus* ATCC25923	(Taşkın Çakıcı et al., 2023)
SA-SeNPs	*Neopestalotiopsis rosae* and *Fusarium oxysporum*	(Tran et al., 2023)
SA-AgNPs	*E. coli, P. aeruginosa, Vibrio vulnificus, S. aureus, B. cereus*, and *C. albicans*	(Kang et al., 2023)

Conclusion

Sodium alginate and chitosan composite films loaded with SeNPs have emerged as a promising solution for preserving packaged food. These eco-friendly edible films have gained attention because of their antibacterial and antifungal properties making them effective against common plant pathogens. The incorporation of SeNPs into the chitosan and alginate matrix enhances the film's ability for extending the shelf life of food materials and enhancing the antioxidant properties of packaging films. These green nano-composite films offer a potential alternative for food preservation and packaging.

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الأفلام النانوية المركبة كأغشية خضراء فى حفظ الأغذية

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أدى ارتفاع الطلب على الخضروات الطازجة والفواكه والجبن واللحوم وما إلى ذلك إلى التقدم في التكنولوجيا، مما أتاح توافرها على مدار العام. ومع ذلك، فإن هذه التقنيات نفسها زادت من خطر التلوث الميكروبي. إن خطوات معالجة الخضروات الطازجة وكذلك الفواكه يمكن أن تؤثر على جودتها وتجعلها عرضة للتلوث والتدهور. يمكن أن يكون التلوث عن طريق البكتيريا المسببة للأمراض، والتي يمكن أن تكون موجودة في التربة أو مياه الري. تشكل الجراثيم البكتيرية تهديدًا للصحة العامة فقط عندما تتاح لها الفرصة للنمو وتطوير الخلايا الخضرية. وفي الوقت نفسه، تعد الفطريات هي المساهم الرئيسي في تعفن الفواكه ومنتجات الفاكهة، حيث يمكنها النمو بسرعة داخل الأنسجة وضبط قدرتها الهجومية. إن استخدام تكنولوجيا النانو في تطوير مواد التعبئة والتغليف النشطة بيولوجيا القائمة على البوليمرات الحيوية الطبيعية، مثل الشيتوزان، قدم نتائج واعدة في تعزيز الخصائص الحاجزة للأفلام. علاوة على ذلك، أظهر دمج الجسيمات النانوية في أفلام الشيتوزان سلوكًا معتدلًا مضادًا للبكتيريا وقابلية للتحلل البيولوجي، مما يجعلها مناسبة للتغليف المضاد للميكروبات وتضميد الجروح. يقترح أن الأفلام النانوية المركبة يمكنها تحسين الخصائص الوظيفية وإطالة العمر الافتراضي للفواكه والخضروات. علاوة على ذلك، فهي قادرة على حفظ الطعام بقيمة غذائية وفي نفس الوقت تعتبر آمنة وصالحة للأكل في بعض الأحيان.