

## Antimicrobial biodegradable packaging with nanotechnology application

Embalagens biodegradáveis antimicrobianas com aplicação de nanotecnologia

Envases biodegradables antimicrobianos con aplicación de nanotecnología

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### Abstract

The production of sustainable food packaging from renewable sources represents a prominent alternative to the use of petrochemical-based plastics. For example, starch remains one of the most studied replacement options due to its wide availability, low cost, and significant advances in improving packaging properties. In this context, nanoparticles with antimicrobial properties as additives play a key role in manufacturing renewable active packaging with superior performance. In this review, a comprehensive summary is provided on the research papers that addresses strategies for using active packaging, using starch as a sustainable polymer, and antimicrobial nanoparticles to extend the lifespan of foods. After a brief introduction to the fundamental concepts related to starch and biodegradable and active packaging, details are presented about the latest advances in nanotechnology, which can minimize the impact on the organoleptic properties of food products, as well as an increase in bioactivity, due to the size in nanometric scale to improve the diffusion of active compounds in the matrix of starch-based active film.

**Keywords:** Green polymer; Food packaging; Nanocomposites; Nanoemulsion; Starch.

### Resumo

A produção de embalagens de alimentos sustentáveis a partir de fontes renováveis representa uma alternativa de destaque ao uso de plásticos de base petroquímica. Por exemplo, o amido continua sendo uma das opções de substituição mais estudadas devido à sua ampla disponibilidade, baixo custo e avanços significativos na melhoria das propriedades das embalagens. Neste contexto, nanopartículas com propriedades antimicrobianas como aditivos desempenham um papel fundamental na fabricação de embalagens ativas renováveis com desempenhos superiores. Nesta revisão, é fornecido um resumo abrangente sobre os trabalhos de pesquisa que abordam estratégias de uso de embalagens ativas, utilizando amido como polímero sustentável e nanopartículas antimicrobianas para estender a vida útil dos alimentos. Após uma breve introdução aos conceitos fundamentais relacionados ao amido e as embalagens biodegradáveis e ativas, são apresentados detalhes sobre os mais recentes avanços da nanotecnologia, podendo minimizar o impacto nas propriedades organolépticas dos produtos alimentícios, bem como um aumento da bioatividade, devido ao tamanho em escala nanométrica melhorar a difusão dos compostos ativos na matriz dos filmes ativos à base de amido.

**Palavras-chave:** Polímero verde; Embalagem de alimentos; Nanocompósitos; Nanoemulsão; Amido.

## Resumen

La producción de envases alimentarios sostenibles a partir de fuentes renovables representa una alternativa destacada al uso de plásticos de origen petroquímico. Por ejemplo, el almidón sigue siendo una de las opciones de reemplazo más estudiadas debido a su amplia disponibilidad, bajo costo y avances significativos en la mejora de las propiedades del empaque. En este contexto, las nanopartículas con propiedades antimicrobianas como aditivos juegan un papel clave en la fabricación de envases activos renovables con prestaciones superiores. En esta revisión, se proporciona un resumen completo de los trabajos de investigación que abordan estrategias para el uso de envases activos, el uso de almidón con polímero sostenible y nanopartículas antimicrobianas para prolongar la vida útil de los alimentos. Tras una breve introducción a los conceptos fundamentales relacionados con el almidón y los envases biodegradables y activos, se detallan los últimos avances en nanotecnología, que pueden minimizar el impacto en las propiedades organolépticas de los productos alimentarios, así como un aumento de la bioactividad, debido a el tamaño en escala nanométrica para mejorar la difusión de compuestos activos en la matriz de películas activas a base de almidón.

**Palabras clave:** Polímero verde; Envasado de alimentos; Nanocompuestos; Nanoemulsión; Almidón.

## 1. Introduction

Global plastic production has grown rapidly since the 1950s, recently accounting for approximately 450 million metric tons of annual production (Wu et al. 2021). Despite the convenience and high quality of this material, environmental concerns about pollution have attracted attention. It was reported that over 340 million tons of plastic waste were generated globally, being around 46% of this coming from the packaging sector (Ahankari et al. 2021).

Inspired by growing concerns about the environment and health safety, several researchers have focused their research on different methods for developing safe and reliable biodegradable packaging. Therefore, biodegradable polymer-based food packaging material has received significant interest from the food industry due to its renewable and environmentally friendly properties.

Among the main raw materials used for the development of biodegradable polymers, natural polysaccharides and proteins are highlighted, due to their characteristics such as high abundance, renewable, biodegradable, biocompatible and for having low toxicity (Ji et al. 2016; Żołek-Tryznowska & Holica 2020).

According to Amiri et al. (2019), films derived from biodegradable polymers proved to be an excellent matrix for the development of food packaging materials incorporating various additives such as antimicrobial and antioxidant agents considered active materials that increase food quality as well as the shelf life of the product limiting microbial proliferation.

For the use of plastic materials in ready-to-eat foods, traditional methods of controlling the growth of microorganisms include the application of baths or antimicrobial sprays on the surface of products (Ferreira et al. 2019; Kocharunchitt et al. 2019). However, the direct application of these compounds on the surface of food can interfere with the sensory characteristics of these products, generating free radicals or interfering with food safety (toxicity). Therefore, recent research has focused on the study of the antimicrobial effects of active packaging in food. Such systems should be able to interact with food or packaging media to reduce or inhibit the growth of pathogenic and/or deteriorating microorganisms on the food surface without compromising its sensory characteristics (Chawla et al. 2021). These systems should also act to prevent the diffusion of molecules into/out of the package or control the release of the antimicrobial compound into the medium.

Several organic and inorganic compounds have antimicrobial activity, e.g., metal ions and nanoparticles (Jung et al. 2018; Ortega et al. 2017), metal oxides (Jayakumar et al. 2019), synthetic agents (Sullivan et al. 2020), natural agents of microbial origin (Settier-Ramírez et al. 2021) or plant source (essential oils) (Kong et al. 2020) and biopolymers (chitosan) (Kongkaoroptham et al. 2021). Several essential oils are classified by the Food and Drug Administration (FDA) as Generally Recognized as Safe (GRAS) and exhibit biocide, antimicrobial, and medicinal activities, and can be used in air purifiers and aromatherapy (Klangmuang & Sothornvit 2016; Raut & Karuppaiyl 2014).

When the goal is to promote the release of antimicrobial film agents to the food surface, the packing characteristics and release profile are crucial. The concentration of the antimicrobial agent should allow the activity of the compound to be

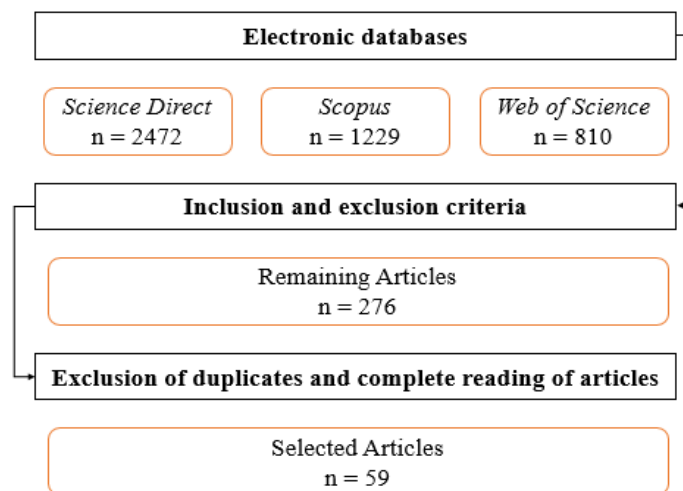
maintained for long periods, so that the release does not occur very quickly. However, if the release is too slow, an adequate concentration of the antimicrobial agent at the early stage may not be achieved and food deterioration is not controlled (Van Long et al. 2016).

The combination of active polymer systems with nanoparticles is an interesting approach to reduce the permeability of molecules (such as O<sub>2</sub>, CO<sub>2</sub>, and water vapor) that can cause oxidation, changes in moisture and loss of texture or aroma in food (Brandolt et al. 2019), and to control the release of antimicrobial agents (Hendessi et al. 2016). In addition, the mechanical properties as well as the thermal and degradation properties of the films can be affected according to the morphology, degree of dispersion, and interaction with the polymer matrix. Despite the great scientific and industrial challenges in this area, studies have contributed to the elucidation of the potential of nanomaterials as a synonym for advancement in the area of biodegradable packaging. Therefore, the aim of this review is to provide a comprehensive overview of research work addressing strategies for using active packaging, using starch as a sustainable polymer and antimicrobial nanoparticles to extend food life.

## 2. Methodology

The present study is a narrative review about the use of nanotechnology applied to food packaging. The review was performed by searching bibliographic references in different indexing databases, on topics related to the use of nanotechnology applied to biodegradable packaging. The scientific articles were searched by adding the keywords "starch-based biodegradable packaging", "nanotechnology in starch-based biodegradable packaging", "metal nanoparticles as antimicrobial", "essential oil nanoemulsion as antimicrobial". The information on the subject was taken from electronic databases such as Scopus, Science Direct (Elsevier) and Web of Science. Initially, 2472 were selected from Science Direct, 1229 from Scopus, and 810 from Web of Science. After applying the inclusion and exclusion criteria and eliminating duplicates, the final sample was 59 articles, as shown in the flowchart in Figure 1. The search period was from 2002 to 2021 and the criteria adopted for the selection of articles included publications in the English language.

**Figure 1** – Flowchart regarding the data search.



Source: From the authors.

### 3. Biodegradable Packaging

Packaging is a key element for the success of food in the market, in terms of sales, and it is one of the responsible for communication between the brand and the final consumer. More than a mere involucres for food products, packaging also plays an important role in protecting and better conserving food and nutrients, protecting them from adversity and climatic phenomena that reduce their shelf life.

According to data from the Brazilian Packaging Association – ABRE (2021), packaging is used by a wide variety of industrial sectors, and the worldwide packaging industry moves more than US\$ 500 billion, and the economic market of the Brazilian packaging industry currently moves more than R\$ 90 billion with the largest share in the value of production represented by plastic materials.

Thus, the consumption of plastic products over the years has produced a large amount of waste that accumulates in landfills, generating considerable environmental problems (Jaramillo et al. 2016). Conventional plastics or non-biodegradable polymers contribute to these problems, as they take 100 to 500 years to decompose in nature (Silva & Rabelo 2017).

Numerous alternatives to minimize the environmental impacts caused by improper disposal of products manufactured from conventional plastics have been carried out, such as reuse and recycling of packaging. Moreover, the production and use of biodegradable polymers has been an alternative for the preservation of the environment, and the main advantage in relation to the use of biopolymers under conventional polymers is the degradation time since they can be consumed in weeks or months under favorable conditions of biodegradation.

Biodegradable polymers are degradable materials whose decomposition results primarily from the action of naturally occurring microorganisms, generating CO<sub>2</sub>, CH<sub>4</sub>, cellular components, and other products. The principle of biodegradability is also related to the molecular weight and chemical structure of the molecules (Jaramillo et al. 2016).

Biopolymers can be subdivided into four groups: polymers obtained from biomass, polymers resulting from the metabolism of microorganisms, polymers from biotechnological processes, and polymers derived from petrochemical products. The development of polymeric products derived from biomass is one of the most important, due to its affordable cost, abundance and because they come from renewable sources. These include proteins and polysaccharides.

Among the polysaccharides, starch is one of the most promising materials used due to its ease of processing (Pelissari et al. 2013; Sanches et al. 2020). Considered a low-cost, widely available renewable resource, it can be obtained from different by-products of harvesting and industrialization of plant raw materials (Fitch-Vargas et al. 2016).

#### 3.1 Starch as a natural biodegradable polymer

Starch is considered one of the most abundant biopolymers on the planet and it is widely studied by the scientific community. As good alternatives to conventional polymers, starch-based materials provide intrinsic biodegradability and biocompatibility properties and are even used as edible polymers in the food packaging industry (Sanches et al. 2020; Żołek-Tryznowska & Holica 2020).

Produced by most green plants to store energy, starch consists of glucose units linked together in the form of two macromolecules: amylose, which is represented by a helical D-glucose unit linked to  $\alpha$ -(1 $\rightarrow$ 4) ), and amylopectin, which is a highly branched macromolecule of  $\alpha$ -(1 $\rightarrow$ 4) linked D-glucose units joined by several  $\alpha$ -(1 $\rightarrow$ 6) branch points (Ji et al. 2016; Farahnaky et al. 2013). Joints comprise 98-99% dry mass of starch and are packed into concentric rings forming semicrystalline and amorphous layers (Ojogbo et al. 2020).

Amylopectin (highly branched structure) predominantly contributes to the crystalline organization of starch granules and amylose, although linear presents a conformation that makes it difficult to associate it regularly with other chains (Farahnaky et al. 2013).

Starch can be isolated from numerous botanical sources with different proportions and molecular weights of amylose and amylopectin, which makes their physical and chemical properties different and therefore, also differ in processing and possible applications (Araujo- Farro et al. 2010).

The semicrystalline structures of native starch granules allow that when heated in the presence of water, these granules can undergo a process called gelatinization in which water enters the starch granule, then the starch granules swell, the crystalline amylopectin melts and the native molecular architectural order is lost. The temperature at which this process occurs is called the gelatinization temperature, which is often determined using differential scanning calorimetry, Rapid Visco Analyzer techniques, or heated stage polarized light microscopy (Bemiller & Whistler 2009).

Once the starch suspension is heated and reaches its gelatinization temperature, as the chains of the molecules extend and separate, the viscosity increases and consequently results in a three-dimensional gel structure (Jaramillo et al. 2016) which is important for product quality and process optimization.

Starch films are thin and flexible materials produced from biopolymers, which can act as coatings, when applied directly to the surface of food, and as films, when they have the ability to form their own structures independent of the packaged product (Ojogbo et al. 2020).

The unit chain length distribution pattern in the internal structure of amylose and amylopectin affects the thermal properties and the retrogradation profile of starch (Thakur et al. 2018). In this regard, the composition of these components can influence the properties of the starch-based film. Films with higher amounts of amylose typically have better film-forming characteristics, including elongation and gas barrier properties (Maniglia et al. 2019).

However, starch-based packaging has restrictions due to the nature of the films for application in food packaging, such as low barrier to water resistance and low mechanical strength, they are delicate structures and easy to dissolve (Ji et al. 2016; Maniglia et al. 2019). These shortcomings limit their application particularly for food packaging purposes. Therefore, the improvement of mechanical and water barrier properties using nanotechnology can be a good alternative in film development. Table 1 shows examples of films made from different types of starch, especially mango seed starch, which proved to be a natural and safe alternative treatment to increase the postharvest life of tomatoes and other climacteric fruits.

**Table 1** – Properties of biodegradable films from different starch sources.

Source	Plasticizer	Results	Reference
<b>Jabuticaba peel</b>	Glycerol	The influence of the concentration of jabuticaba peel flour (JPF) and the concentration of glycerol (GC) on the starch films was evaluate. The increase in JPF and GC concentrations increased the thickness and water vapor permeability and reduced the tensile strength of the films. The film containing 15.80% JPF and 15.80% GC obtained the best mechanical and water barrier properties.	Sanches et al. (2021)
<b>Potato</b>	Glycerol	Potato starch films containing 20%, 40%, 60% and 80% glycerol were produced. The contact angle values with respect to water decreased and the optical density increased with increasing glycerol content. The printing performance of the starch film was of inferior quality compared to the polylactide (PLA) film and the biodegradable paper. Nevertheless, the results of this study show strong potential of using starch as a raw material to produce biodegradable and eco-friendly packaging.	Zołek-Tryznowska & Holica (2020)
<b>Babassu</b>	Glycerol, sorbitol, urea, glucose	The presence of the plasticizers glycerol and urea produced films that were more soluble and more permeable to water vapor. The films plasticized with sorbitol and glycerol were more mechanically resistant. All babassu starch films show good antioxidant activity due to the presence of phenolic compounds.	Maniglia et al. (2019)
<b>Mango kernel</b>	Glycerol, sorbitol	The coating was promising in delaying the ripening process of tomatoes by up to 20 days at 20°C. The formulations containing sorbitol obtained the best results, followed by the combined plasticizers (glycerol:sorbitol) and glycerol.	Nawab et al. (2017)
<b>Amaranth</b>	Glycerol	Transparent films, with minimum thickness, low water vapor permeability and moderate mechanical resistance.	Chandla et al. (2017)
<b>Corn</b>	Glycerol, sorbitol	Sorbitol plasticized film showed higher deformation, while slightly increasing the sorbitol content in the plasticizer mixture produced stiffer films. On the other hand, glycerol played an improvement in flexibility. The films plasticized with glycerol and sorbitol showed low WVP.	Fitch-Vargas et al. (2016)
<b>Cassava</b>	Glycerol, yerba mate extract	Yerba mate extract acted as a plasticizer when it was incorporating as an antioxidant into starch-glycerol based films. The melting temperature, enthalpy of crystallization, and amorphous/crystalline ratio decreased with increasing yerba mate extract content. The extract led to faster degradation of the films in plant compound before two weeks, preserving the stability of the films in acidic and neutral media, and higher stability in alkaline media.	Jaramillo et al. (2016)
<b>Wheat</b>	Glycerol	Increasing the concentration of glycerol promoted the formation of more soluble, transparent, moist and crystalline films. The mechanical properties (Young's modulus and tensile strength) increased with increasing plasticizer concentration.	Farahnaky et al. (2013)
<b>Banana</b>	Glycerol	The banana starch film showed low water vapor solubility. The X-ray pattern showed that the banana starch film is type C and the glass transition temperature corresponds to 46.4 °C.	Pelissari et al. (2013)
<b>Quinoa</b>	Glycerol	The influence of glycerol, pH and drying temperature and time on the properties of quinoa starch films was evaluating by employing response surface methodology. Films produced under optimum process conditions exhibited high mechanical properties, low solubility (15,9%), low water vapor permeability (0,204±0,012 g mm.m <sup>-2</sup> .h <sup>-1</sup> .kPa <sup>-1</sup> ) and high oxygen permeability (4,34±1,03 cm <sup>3</sup> µm.m <sup>-2</sup> .d <sup>-1</sup> .kPa).	Araujo-Farro et al. (2010)

Source: From the authors.

Numerous research have been aimed at improving biopolymers for application in food packaging systems due to the various advantages offered, along with ecological safety and sustainability. The development of active food packaging systems can provide greater system efficiency and better food safety, as they can act as a carrier of bioactive compounds that keep products fresh, prolong product life, and reduce the risk of pathogen growth.

### 3.2 Active packaging

In the conventional sense, a food package is intended to contain, protect, transport, identify, and preserve the product from the industry to the consumer's table. For Żolek-Tryznowska & Holica (2020), old qualities of conventional packaging remain essential, but are now complemented by other important functions. In this way, active packaging arises as a strategy that aims, through the interaction of the packaging with the food, to inhibit the development of pathogenic microorganisms, increase the shelf life of the product, improve sensory characteristics, avoid chemical, and microbiological deterioration, and ensure food safety. (Mousavi et al. 2021). Furthermore, changes in customer lifestyles have increased the demand for clean, high-quality, fresh, minimally processed, and ready-to-eat products with an extended shelf life, which subsequently creates the urgency and need for a technology of modernized packaging (Firouz et al. 2021).

The systems used in active packaging can be divided into two main groups: a) emitter systems, which add or release compounds into the free space of the packaging or into the food (examples: carbon dioxide emitters, antioxidants, antimicrobials and preservatives) and; b) absorber systems, which remove undesirable compounds from the internal atmosphere of the package that could cause food spoilage (examples: absorbers of oxygen, carbon dioxide, ethylene, moisture and impurities) (Chawla et al. 2021). The compounds used in these systems are responsible for the active function and can be incorporated directly into the matrix or using a separate material inside the package.

Regarding emitting systems, antimicrobial and antioxidant packaging are the ones with the greatest application. These systems are effective in releasing active compounds, emitting only the amount necessary to prevent microbial growth or stop oxidation reactions inside the package.

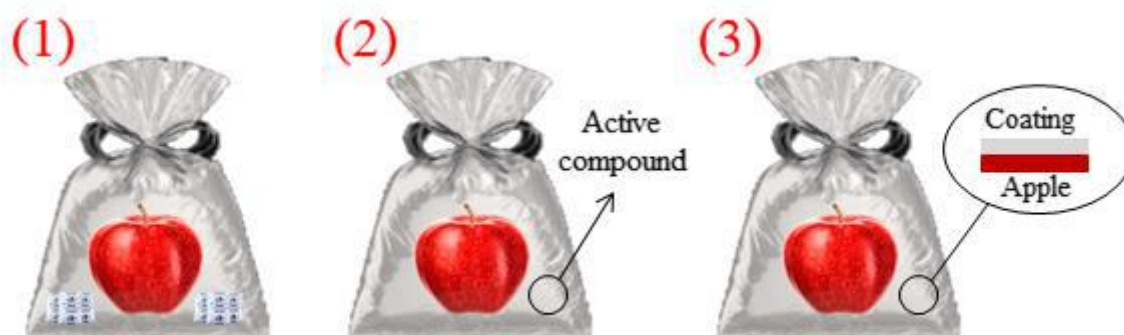
Some of the most studied active agents incorporated into biodegradable coatings and films are antimicrobial agents. Antimicrobial agents consist of different classes of molecules that suppress the multiplication and growth of microorganisms or can even eliminate organisms such as bacteria, fungi, and viruses (Khaneghah et al. 2018), these classes comprise organic agents, bacteriocins, essential oils, extracts from plants, enzymes, organic acids (e.g., lauric acid) and inorganic/metallic nanoparticles (Chawla et al. 2021).

According to Varghese et al. (2020), the inorganic agents commonly used in antimicrobial food packaging are acids and their salts, sulfites, chlorides, phosphates, epoxides, hydrogen peroxide, antibiotics and bacteriocins. Although these inorganic agents are available, natural antimicrobial agents are increasingly gaining consumer interest. By-products from the fruit and vegetable industries that are often discarded have demonstrated ingredients such as vitamins, minerals, antioxidants, and antimicrobial compounds for food preservation. Therefore, the judicious selection and use of such materials can reduce the use of synthetic antimicrobials and, at the same time, increase the value of the by-products. Phenolics, xylans, and essential oils are plant products that are being commonly used as antimicrobial agents in the food packaging industry (Varghese et al. 2020).

Appendini and Hotchkiss (2002) mention the existence of some basic ways used to carry out the incorporation of antimicrobial compounds in packaging. They are: (1) a sachet containing antimicrobial substances connected to the food packaging (the sachet is responsible for the release of the bioactive substance during storing); (2) a film with direct incorporation of antimicrobial agents; and (3) covers that act as carriers for the antimicrobial agent (Figure 2). In addition to

the forms of incorporation of these compounds, the authors mention that one of the alternatives to obtain these active packages would be using polymers that are already inherently antimicrobial such as chitosan and polyamide.

**Figure 2** – Ways of incorporating active agents into packaging: (1) sachet containing antimicrobial substances connected to the food packaging (the sachet is responsible for releasing the bioactive substance during storage); (2) film with direct incorporation of the antimicrobial agent and (3) coverings that act as carriers of the antimicrobial agent.



Source: From the authors.

Research carried out in this field shows that antimicrobial films are more effective compared to the direct addition of antimicrobial agents to foods because the combination of antimicrobial agents is slowly released from the surface of the packaging film for food products at the concentration required to prevent microbial growth. The direct addition of antimicrobials can cause some loss of activity due to leaching into the food matrix. The reaction of some antimicrobials with food components such as lipids and proteins would be inevitable (Appendini & Hotchkiss 2002).

Active packaging with a sustainable concept has been developed since the 20th century. However, research is still carried out and new techniques are applied to further improve its functions and applications. Recent advances in nanoscience and surface engineering hold great promise for solving some of the significant problems facing the food industry in general, particularly in the packaging sector. Nanotechnology allows scientists to design the nanostructure of packaging materials to improve their functions, achieve desirable mechanical and barrier properties, and facilitate the transport of bioactive compounds. Furthermore, this new technology proves to be a great alternative for being cheap, relatively safe, clean, and for presenting a considerably high financial return. The use of micro-or nanotechnology emerges as one of the new trends in relation to the incorporation of active compounds in films and coatings. This technique increases the surface area of the antimicrobial so that smaller amounts of compounds are needed to achieve the same functionality in the active package.

#### 4. Potential Application of Nanotechnology in Sustainable Antimicrobial Polymers

In response to the growing demand for sustainability and ecological safety, recently many investigations have focused on the development of easily degradable and biocompatible food packaging materials. Furthermore, with further improvements in terms of extended shelf life and higher overall food quality, phytochemicals, or bioactive compounds are the most considered for the development of active materials, but unfortunately their efficiency as preservative ingredients are limited by the low solubility, high instability, and degradability (Khaneghah et al. 2018). To overcome these limitations, research has focused on the implementation of nanotechnological concepts for antimicrobial packaging, offering advantages in the field of active packaging (Mousavi et al. 2021).



Nanotechnology is a growing interdisciplinary field that involves the engineering of nanosized materials (1-100 nm), whose physicochemical properties can differ greatly from their raw-size forms (Oleyaei et al. 2016; Ortega et al. 2017). Nanoparticles (NPs) exhibit significantly different physical and chemical properties compared to macroscale ones of the same substance. The properties of nanomaterials depend on size, particle size distribution, surface area, surface roughness, porosity, hydrophobic/hydrophilic properties which generally influence the properties of food packaging (Jildeh & Matouq 2020).

Research shows advantages of the application of nanotechnology in the food packaging industry such as improvements in physical characteristics (increased mechanical strength, barrier properties, flexibility, and stability) using nanoparticles with reinforcing properties and in biochemical properties (increased biocompatibility, biodegradability, eco-sustainability, edibility and low waste) using bio-based nanoparticles (Kuswandi & Moradi 2018). Nanoparticles can not only conceal flavors and prevent chemical breakdown of functional compounds or decomposition due to the action of microorganisms but may also increase the solubility of bioactive compounds along with controlled release. When antimicrobial NPs are incorporated into traditional polymers, bioactive nanocomposites can be obtained. These prepared nanocomposites can be exploited for the preparation of active packaging much more efficient in increasing the shelf life and quality of the food storage product.

Nanoparticles with their modified properties have great potential to be used as antimicrobial agents as mentioned above. There are two types of antimicrobial agents, organic (organic acids, bacteriocins, essential oils, etc.) and inorganic (mainly metal oxides). Organic antimicrobial agents have some disadvantages as they are very sensitive to processing conditions such as high pressure and temperature. In contrast, inorganic antimicrobial agents have good stability at high pressures and temperatures, long shelf life, and show strong antimicrobial activity even at low concentrations (Kumar et al. 2020). Inorganic nanomaterials used in food packaging and storage are transition metals (silver and iron); alkaline earth metals (calcium and magnesium) and nonmetals (selenium and silicates). Metal nanoparticles have gained attention as powerful antimicrobial agents due to their broad spectrum of activity, durability, strength, selectivity, and specificity (Jayakumar et al. 2019; Jung et al. 2018; Ortega et al. 2017; Cano et al. 2016; Oleyaei et al. 2016). Table 2 reports the effects of applying metallic nanoparticles in food-active biodegradable packaging, we can highlight silver nanoparticles (AgNPs) for applications in food packaging due to their improved physicochemical, optical, thermal, and antimicrobial properties.

**Table 2** – Effects of applying metallic nanoparticles to biodegradable active food packaging.

Starch used	Metal nanoparticle	Nanoparticle size (nm)	Application type	Results	Reference
<b>Soluble</b>	Graphitic carbon nitride (gC <sub>3</sub> N <sub>4</sub> )	None	Film	Cu deposited gC <sub>3</sub> N <sub>4</sub> nanoparticles were using as reinforcing agents in starch/NaAlg films. The presence of Cu-gC <sub>3</sub> N <sub>4</sub> can greatly increase the tensile strength and barrier properties of the film. In addition, Cu5% -gC <sub>3</sub> N <sub>4</sub> can effectively prevent the growth of gram-positive and gram-negative bacteria.	Mousavi et al. (2021)
<b>Jackfruit seed</b>	Zinc oxide (ZnO)	None	Film	Starch-PVA based films incorporated with ZnO nanoparticles and nutmeg oil demonstrated improved water barrier and mechanical properties, as well as showing promising antibacterial activity against <i>S. typhimurium</i> .	Jayakumar et al. (2019)
<b>Soluble</b>	Silver (Ag)	10	Paper Overlay	Starch-silver coating solutions (ST-AgNPs) containing a mixture of starch, silver nitrate and distilled water were prepared by ultrasonication. The starch-coated paper with AgNPs showed not only highly enhanced oil resistance, but also excellent antibacterial activity against gram-negative <i>E. coli</i> and gram-positive <i>S. aureus</i> .	Jung et al. (2018)
<b>Corn</b>	Silver (Ag)	5–20	Film	Nanocomposite films containing concentrations of AgNPs higher than 71.5 ppm inhibited the growth of <i>E. coli</i> and <i>Salmonella</i> spp. The AgNPs content caused a slight increase in film thickness and opacity while maintaining the UV barrier capability of the material. A decrease in WVP was observed with increasing AgNPs concentration.	Ortega et al. (2017)
<b>Pea</b>	Silver (Ag)	40	Film	Starch-PVA-based films incorporating silver nanoparticles (AgNPs) exhibited remarkable antibacterial activity against <i>Listeria innocua</i> and <i>Escherichia coli</i> and antifungal activity against <i>Aspergillus niger</i> and <i>Penicillium expansum</i> .	Cano et al. (2016)
<b>Corn</b>	Silver (Ag)	2–6	Films	The nanocomposites showed increased thermal stability through the incorporation of AgNPs. The composite films exhibited strong antibacterial activity against <i>Escherichia coli</i> and <i>Staphylococcus aureus</i> , with the films being most effective against gram-negative bacteria ( <i>E. coli</i> ).	Ji et al. (2016)
<b>Potato</b>	Titanium dioxide (TiO <sub>2</sub> )	21	Film	Bionanocomposite films containing two types of nanoparticles: sodium montmorillonite (MMT) and TiO <sub>2</sub> affected some properties. Tensile strength and thermal stability increased with the incorporation of NPs and water vapor permeability and the transmittance of visible UVA, UVB and UVC lights decreased with increasing TiO <sub>2</sub> and MMT.	Oleyaei et al. (2016)
<b>Quinoa</b>	Gold (Au)	10	Films	The nanocomposites showed improved mechanical, optical and morphological properties, while maintaining thermal and barrier properties unchanged when compared to the standard film. The nanocomposites exhibited strong antibacterial activity against foodborne pathogens with inhibition percentages of 99% against <i>E. coli</i> and 98% against <i>S. aureus</i> .	Pagno et al. (2015)

Source: From the authors.

Despite the advantages, the fact of the toxicological risks related to the migration of heavy particles and nanoparticles must be analyzed. It is essential to investigate how these nanoparticles migrate within the human body as well as their toxic and immunogenic effects (Bajpai et al. 2018). According to the European Food Safety Authority (EFSA), the upper limit for silver migration in food packaging cannot go beyond 0.05 mg/L in water and 0.05 mg/kg in food (Chawla et al. 2021). Therefore, the developed nanobiocomposites and their disposition must be carefully studied considering the migration phenomenon.

The most studied organic antimicrobial agents today are essential oils (EO). The use of essential oil has gained considerable attention in recent years not only because it is a natural food additive, but also because of its beneficial effects on human health and the concern about the negative consumer perception of chemical preservatives. Essential oils have antimicrobial and antioxidant properties, which makes them interesting for application in packaging in the food industry.

However, the high reactivity and hydrophobicity of EO represent a challenge for their incorporation into food packaging (Prakash et al. 2018). These challenges can be overcome using encapsulated EO delivery systems, which can include emulsions, liposomes, solid lipid nanoparticles, nanostructured materials, micelles, among others (Amiri et al. 2019). Among the aforementioned, emulsions are emerging on the market as they are able to guarantee a uniform distribution of partially or fully hydrophobic compounds in a hydrophilic matrix (Xiong et al. 2020). Usually, the two fluids are oil and water, but other immiscible liquids can also be emulsified. There are two common types of emulsions: O/W which consist of small droplets of oil dispersed in water, while W/O emulsions consist of small droplets of water dispersed in oil (McClements 2021).

Important elements of food packaging are evaluations of the film's mechanical, optical, and barrier characteristics. The mechanical properties of biodegradable active packaging incorporated with EO depend on several characteristics such as the composition of the EO, the amount added, and the material used for packaging (Shojaee-Aliabadi et al. 2013). Several researchers have reported in the past that OEs could be incorporated into the coating matrix. However, the immediate inclusion of EOs in the films or coatings significantly reduces the mechanical characteristics, reduces the carrying capacity of EOs, and increases the risk of oily packaging (Sánchez-González et al. 2011). On the other hand, the use of nanoemulsions in solutions formed from a coating offers numerous advantages, such as reduced mass transmission of the compounds through the coating, greater stability of the compound in stress situations, decreased communication with other components of the food matrix, reduced compound dosage, better film homogeneity, and better antimicrobial and antioxidant activity (Al-Tayyar et al. 2020). Table 3 depicts the effect of the application of EO nanoemulsions in active food packaging, using starch as a polymeric matrix, among the EO, carvacrol stands out, approved by the Food and Drug Administration (USFDA) for use in foods and beverages, it does not contain significant toxic effects in commonly used amounts and has broad biological properties, including antioxidant and antimicrobial properties (Kong et al. 2020).

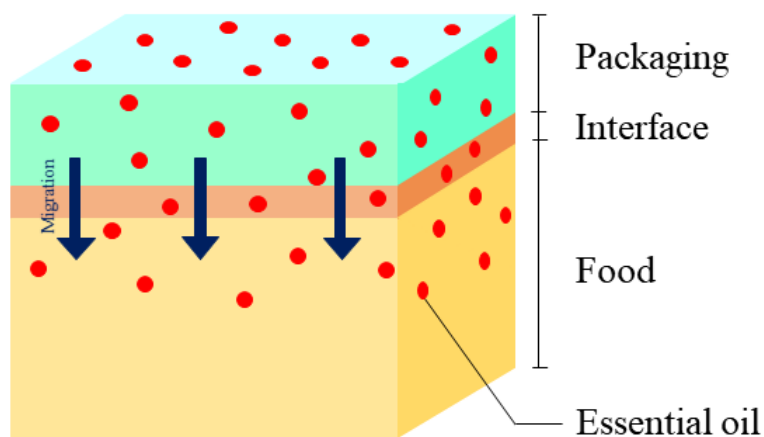
**Table 3** – Effects of the application of EO nanoemulsions in active food packaging.

Starch used	Essential oil	Nanoemulsion formulation	Nanoemulsion size (nm)	Application Type	Results	Reference
Cassava	Lemongrass	Lemongrass essential oil, pectin, glycerol, Tween 80	1280–301	Edible Films	Emulsification improved the mechanical properties of the film by increasing extensibility, strength, and stiffness. The films showed adequate degradation in vegetal compost, ensuring their complete biodegradation in a short period.	Mendes et al. (2020)
Arrowroot	Carnauba wax	Carnauba essential oil and water	40	Edible Films	The presence of carnauba wax in the films increased their hydrophobic characteristics, reduced water vapor permeability and light barrier properties, as well as improved tensile strength and a smoother microstructure.	Oliveira Filho et al. (2020)
Corn	Carvacrol	Carvacrol essential oil, Tween 80	None	Food Packaging	The incorporation of carvacrol nanoemulsion (CNE) significantly increased tensile strength, Young's modulus, elongation at break, barrier (water vapor and ultraviolet), antioxidant and antifungal activity. Films with 25% CNE exhibited excellent antifungal activity against <i>Trichoderma</i> sp.	Kong et al. (2020)
Corn	<i>Zataria multi flora</i>	<i>Zataria multi flora</i> essential oil, tween 80 and water	93,2	Film on fresh ground beef	It showed antioxidant activity during 20 days stored under refrigeration conditions ( $4 \pm 1$ °C), in addition to chemical and sensory quality superior to the control film. The results of the mechanical evaluation exhibited lower tensile strength ( $\rho \leq 0.05$ ) and higher elongation at break ( $\rho \geq 0.05$ ) than the control samples.	Amiri (2019)

Source: From the authors.

In the packaging, the antibacterial substances of the EO gradually diffuse through the micropores of the polymer matrix to the surface of the film, where contact with the food occurs (Figure 3).

**Figure 3** – Representation of the migration process of essential oils from the packaging matrix to the food.



Source: From the authors.

In active systems with nanoemulsions, these transporters will also improve the transport of antimicrobial agents through the cell membrane of the target microorganisms, allowing the compound to enter the cell easily and promoting its interaction mainly with the microorganisms' protein synthesis mechanisms. According to Khaneghah et al. (2018), close or direct contact with the microorganism is an essential demand for the mechanisms of action of antimicrobial agents.

The release of the substance in the packaged food can be activated by chemical, biochemical, or biological transformation, in addition to extrinsic factors. The design of an active antimicrobial package needs to incorporate technologies for controlled release and kinetics of microbial growth to allow for optimal system action. For this to occur, some factors need to be considered such as the mechanism of diffusion of the active compound through the coating and the volatilization or solubility of the substance in the food.

By having high solubility in the food product, the active agents will diffuse freely so that the concentration of antibacterial in the package will decrease as the highly soluble antibacterial migrate into the food. On the other hand, in active packages with compounds of low solubility, the concentration of antibacterial agents on the surface of the food decreases along with the concentration of the package and may run out before the end of the expected storage period. Therefore, it is important to adjust the release rate of antimicrobial agents from the packaging material to the food to match the growth kinetics of the target microorganisms (Ju et al. 2019).

According to Prakash et al. (2018), EOs are considered safer than synthetic preservatives because they are secondary metabolites of plants; however, being aware of the complexity of the components of these aromatic oils and the conclusions of toxicological assessments, it is important that comprehensive safety assessments are carried out before applying to foods.

The adequacy and safety of EUs as antimicrobial agents and their toxicological effects when encapsulated in nanoemulsions are the main concerns related to the application of nanoemulsions in food systems. Experimental evidence of its toxicity is rare in the literature, except for some studies based on cell culture; therefore, it is crucial that more research be conducted to obtain an in-depth overview of the safety of OE nanoemulsions (Prakash et al. 2018).

The food industry's concern is evident due to the growing consumer demand for natural products and safe foods as well as the implementation of increasingly stringent standards to prevent foodborne pathological diseases. It reinforces the

hypothesis that the development of studies and the application of active packaging with the potential to improve food safety seem to be essential for the future. The design of optimized and effective delivery systems for EO is a challenge to reduce the dose to be incorporated into foods by reducing the possible biological effects associated with the use of these compounds (Salvia-Trujillo et al. 2015).

## 5. Final Considerations

With the increasing demand for efficient and innovative packaging technologies associated with the expansion of the market for minimally processed food products, research using biodegradable and antimicrobial biopolymers as an alternative to conventional sources due to their ecological and functional nature has prevailed. Although commercialization on an industrial scale is still hampered by the high cost of production, functional and technical limitations have also been barriers to the development and application of biodegradable and antimicrobial packaging materials in the food industry as there is no internationally established standard for testing the performance of food packaging with such functionality.

In this review, we present a comprehensive overview of methods of starch-based biopolymers associated with antimicrobials for use in packaging. Such activities have been shown to depend on several factors including method of preparation (coating, film, etc.), type of antimicrobial (organic, inorganic), size, and shape of the antimicrobial material. In addition, each packaging material requires not only different testing, but also method modification to meet the requirement of a specific active packaging to ensure consistent and reliable results. Recently, biodegradable and antimicrobial food packaging has gained the attention of researchers, therefore it is necessary to develop international norms and regulations in this area of research. As a suggestion for future work, that other biopolymers used in the food industry that show improved properties with the use of nanotechnology be addressed.

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