Revista Ciencia en Desarrollo. Vol. 16 Num. 1. pp. 149 - 159
enero - junio 2025

DOI: 10.19053/01217488.v16.n1.2025.16130

Artículo de revisión

Modified starches and emerging technologies for biofilms production: a review

Almidones modificados y tecnologías emergentes para obtención de biopelículas: una revisión

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Recepción: 23-junio-2023 **Aceptado:** 27-julio-2024 **Publicado:** 20-mayo-2025
Cómo citar: Joya Cardenas, L. P., Usgame Fagua, K. G., & García Torres, A. M. (2025). Modified starches and emerging technologies for biofilms production: a review. *Ciencia en Desarrollo*, 16(1). doi: 10.19053/01217488.v16.n1.2025.16130

Abstract

The optimization of processes for the preparation of biodegradable materials from renewable sources has been a constant challenge in the field of biotechnology. Polymers, such as starch, have been improved and applied as raw materials in both food and non-food industries. Particularly in non-food industries, including the production of biodegradable biofilms, native starch possesses unfavorable characteristics, like high hydrophilicity, water vapor permeability, low stability in acidic/basic environments, and high sensitivity to environmental changes, specially humidity, temperature, and pH. Therefore, the incorporation of modification methods, emerging technologies, and reinforcement systems capable of counteracting these adverse characteristics has become necessary. This review focuses on research conducted over the past five years, highlighting outstanding methods of chemical modifications in starch, including cross-linking, esterification, and oxidation. It also includes dual methods and emerging modification technologies, like cold plasma treatment, ozonization, and the implementation of reinforcement materials with nanoparticles. Finally, the scope of the main characterization techniques employed in modified starches is detailed.

Keywords: biofilms, emerging technologies, modified starch, nanotechnology.

Resumen

La optimización de procesos para la preparación de materiales biodegradables a partir de fuentes renovables ha sido un desafío constante en el área de la biotecnología. Polímeros como el almidón, han sido mejorados y aplicados como materia prima en industrias alimentarias y no alimentarias. Particularmente, en industrias no alimentarias como las dedicadas a la producción de biopelículas biodegradables, el almidón nativo posee características poco favorables, tales como: alta hidrofilicidad, permeabilidad al vapor de agua, baja estabilidad en medios ácidos/básicos y alta sensibilidad a los cambios ambientales, especialmente humedad, temperatura, y pH. En consecuencia, por lo que ha surgido como necesidad la incorporación de métodos de modificación, tecnologías emergentes y sistemas de refuerzo, capaces de contrarrestar las características adversas mencionadas. La presente revisión se centra en las investigaciones que durante los últimos cinco años se han dirigido hacia métodos sobresalientes de modificaciones químicas en almidones como lo son el entrecruzamiento, la esterificación y la oxidación. Se incluyen los llamados métodos duales y tecnologías emergentes de modificación como el tratamiento con plasma frío, la ozonización y la implementación de materiales de refuerzo con nanopartículas. Finalmente, se detalla el alcance de las principales técnicas de caracterización empleadas en los almidones modificados.

Palabras Clave: biopelículas, tecnologías emergentes, almidón modificado, nanotecnología.

E-ISSN: 2462-7658

1 Introduction

Starch is one of the naturally occurring polysaccharides abundantly found in seeds, leaves, roots, flowers, stems, and tubers. It consists of two polymeric conformations: amylose and amylopectin, organized in a granular structure with varying sizes and shapes depending on its source and variety [1]. Starch has been referred to as a biopolymer of high commercial and research interest due to its abundance and biodegradability, aspects that make it a favorable material in both food and non-food industries [2-5].

The functionality of starch in the food industry has been guided by various modification processes that this polymer has undergone, which have allowed for enhancing and optimizing its gelling and thickening capacity in foods such as pasta, cookies, bread, cakes, among others [6]. Within the field, some of the most relevant methods of starch modification include physical, enzymatic, and chemical methods [7, 8].

Regarding physical modification methods, some of the techniques used include pre-gelatinization, heat-moisture treatment, retrogradation, dry heat treatment, osmotic pressure treatment, ultrasonication, milling, high-pressure treatment, pulsed electric field treatment, among others. These methods typically involve thermal or non-thermal methodologies that can induce significant changes in the granular structure of starch as well as its physicochemical properties, such as swelling, solubility index, water absorption, retrogradation, and gelatinization [9, 10, 11].

On the other hand, enzymatic and chemical modifications lead to alterations in the molecular structure of starch by depolymerizing and substituting functional groups [12, 13]. Among enzymatic modifications, hydrolytic processes stand out, in which progressive degradation of starch occurs due to the action of enzymes such as glucoamylase, α-amylase, β-amylase, among others. These enzymes have the ability to reduce the content of amylose or amylopectin and cause significant changes in the solubility, viscosity, gelation, and retrogradation of this polymer [14, 15]. In contrast, chemical modification methods, including acetylation, succinylation, and oxidation, primarily involve the substitution of the three available hydroxyl groups in the units of anhydroglucose with acetyl, succinyl, carbonyl, carboxyl or acyl groups [16, 17], which are directly related to changes in physicochemical characteristics including morphology, crystallinity pattern, amylose and amylopectin contents, moisture, solubility, among others [18, 19].

Due to the feasibility of modifying this biopolymer and its effect on its physicochemical properties, the different physical, enzymatic, and chemical processes for transforming starch have been extended to non-food applications, for example: pharmaceutical and cosmetic production, textiles, adhesives, and the production of biodegradable biofilms [20].

Particularly, the production of biofilms from starch modified by chemical methods, for example: esterification, cross-linking, and oxidation, has allowed advancements towards the optimization and improvement of some physicochemical and functional characteristics of the material. Additionally, other modification methods have been incorporated, namely dual approaches, emerging technologies involving cold plasma treatment or ozonation and, even, the use of nanoparticles as reinforcing systems. All of these methods aim to reduce the use of contaminating chemical agents and provide techniques that do not generate adverse effects on the environment. Some chemical modifications have been combined to form dual processes like oxidation and cross-linking, acetylation and hydroxypropylation or acetylation and cross-linking, among others [21, 22, 23]; resulting in strong and functional materials suitable to use in food packaging [24, 25]. On the other hand, emerging technologies, including cold plasma treatment, which involves the reordering of the double helices of the amylopectin component of the polymer chain through electronic bombardment on the surface of the biofilm, have allowed significant structural changes impacting its physicochemical and mechanical properties. In the case of ozonation, the hydroxyl groups of the anhydroglucose units are oxidized, allowing the formation of carbonyl or carboxyl groups, resulting in intramolecular stability when combined with plasticizers such as glycerol [26, 27].

During the last decade, the production of starch biofilms has incorporated the use of both organic and inorganic nanoparticles as a system for structural and functional reinforcement. These nanoparticles are biocompatible with the components of amylose and amylopectin. This has conferred the material the designation of active biofilms, as it has been highlighted, for example, for their high capacity to reduce pathogenic microorganisms when used in foods with high moisture content and water activity [28].

In general, regardless of the type of starch modification, it has been characterized and evaluated using different instrumental techniques. Among them, Infrared Spectroscopy (IR), X-Ray Diffraction (XRD), Thermogravimetric Analysis (TGA), Scanning Electron Microscopy (SEM), Atomic Force Microscopy (AFM), and Optical Microscopy (OM) stand out. These techniques have allowed the determination of physical and chemical characteristics, namely the elucidation of formed functional groups, possible changes in crystallinity patterns and phases, decomposition, thermal stability, microstructural changes, among others.

This review provides details on the methods of chemical modification of starch for its subsequent use as a raw material in the production of biodegradable biofilms, such as cross-linking, esterification, and oxidation. It also includes dual methods, emerging technologies, like cold plasma treatment and ozonation as well as reinforcement systems including the use of nanoparticles. Finally, it encompasses the most relevant characterization techniques for this type of material.

2 Structure of starch and chemical modifications

Starch is a homopolysaccharide composed of glucose units and is found in granules of various sizes. It consists of two types of polymeric conformations: amylose and amylopectin. Amylose is the amorphous component with a linear chain structure that forms spatial arrangements in helices through α-1,4 linkages between glucopyranose units. On the contrary, amylopectin is the crystalline component composed of linear chains connected by α-1,4 linkages and branching points with α-1,6 linkages, as shown in Figure 1.

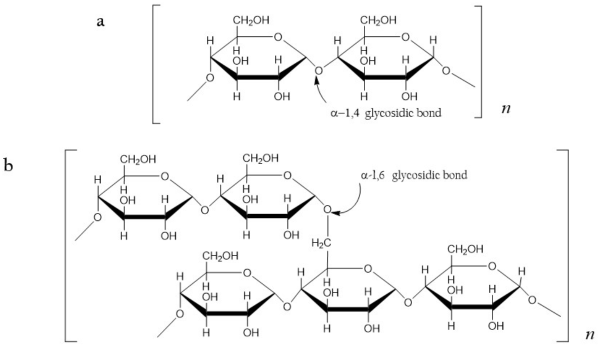

Figure 1: Starch structure: a) Amylose and b) Amylopectin. From author.

The chemical modification of starch aims to optimize its physicochemical characteristics depending on the industrial functionality. Particularly for the production of biofilms, modified starch provides the material with improved properties related to low solubility, impermeability to water vapor and oils, high thermal stability, resistance to breakage, among others [29]. These properties will also depend on the botanical origin of starch, its variety, contents of amylose and amylopectin, morphology, and crystallinity pattern: type A in cereals, type B in tubers, and type C in fruits [30, 31].

Among the main methods of chemical modification of starch used for the production of biofilms, cross-linking, esterification, and oxidation are prominent. In the case of cross-linking, the formation of random intermolecular bonds occurs using bifunctional reagents capable of stabilizing the interaction and enabling the generation of ether or ester linkages with available hydroxyl groups in the polymer chain. In terms of esterification, the substitution of the three hydroxyl groups of the anhydroglucose unit with succinyl, acetyl or acyl groups takes place in a basic medium, sometimes with the presence of catalysts. Finally, oxidation involves the substitution of hydroxyl groups of the anhydroglucose unit with carbonyl or carboxyl groups derived from oxidizing agents in an acidic medium. Each of the mentioned methods is further described in detail below.

2.1 Cross-linking (CL)

Cross-linking results from the reaction between the hydroxyl groups of the anhydroglucose units of starch and functional groups derived from bifunctional cross-linking agents such as sodium trimetaphosphate/sodium tripolyphosphate (STMP/STPP), phosphorus oxychloride, boric acid, and epichlorohydrin [32, 33, 34]. During CL, starch is mixed with specific amounts of the cross-linking agent, which acts as a precursor in the formation of intramolecular covalent bonds favored under basic aqueous conditions, constant stirring and temperatures ranging between 25-30 °C [35]. In Figure 2, an example of the CL reaction for potato starch (*Solanum tuberosum*) using epichlorohydrin as the cross-linking agent is presented.

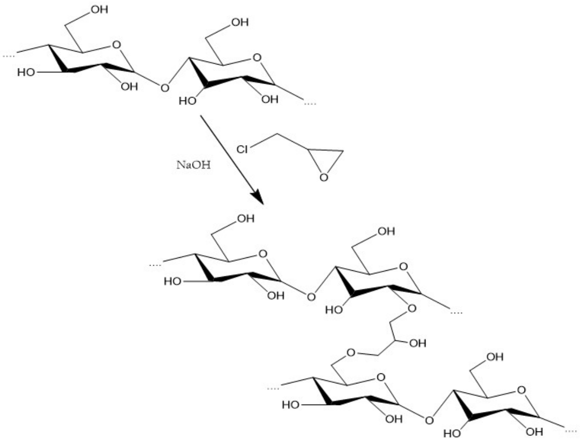

Figure 2: Cross-linking of starch with epichlorohydrin. From Bendoraitiene et al. [36].

Cross-linked starch has contributed to the improvement of functional properties in biofilms or thermoplastic materials, among which water vapor permeability stands out. This property has been studied using STMP (1-5 %) as the cross-linking agent in starches, like corn (Zea mays) and Pearl (*Pearl millet grains*), resulting in a decrease in water vapor permeability, moisture, and solubility of the material. This is attributed to the cross-linking effect, which restricts the mobility of the amylose content chains, thereby preventing water absorption [24, 37].

Another functional aspect of biofilms is related to their mechanical properties, which determine the physical behavior under external forces exerted on the material. With cross-linked starch, it has been reported that the material exhibits high tensile strength due to the limitation of macromolecular movements and swelling of the granules caused by reinforcements between the hydrogen bonds that interconnect the polymer chain, plasticizer and cross-linking agent. As a result, there is an increase in the Young's modulus, thermal resistance, tensile strength, material elasticity, and thickness [38, 39].

2.2 Esterification

Esterification is based on the substitution of hydroxyl groups in starch with ester groups under reaction conditions, which can be either basic or acidic [3, 12]. Among the substances that donate these groups include anhydrides, like acetic, succinic, dodecenyl succinic, and octenyl succinic [22, 40]. Additionally, the use of catalysts, for instance: pyridine, acid chlorides, dioxane, toluene, dimethyl sulfoxide (DMSO) or N,N-dimethylformamide (DMF), is necessary to achieve degrees of substitution (DS) between 1.0 > DS > 2.3 [17, 41]. As an example, Figure 3 represents the esterification reaction of starch in an acidic medium, using DMSO as a catalyst and acetic anhydride as the acetyl group donor.

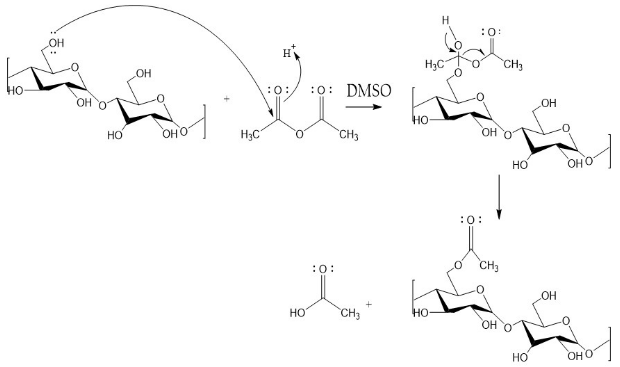

Figure 3: Esterification of starch through acetylation. From Carvalho et al. [42].

However, the use of the mentioned catalysts generates an adverse environmental effect. Therefore, over the last decade, the use of biological catalysts, specifically immobilized lipases, from *Candida antarctica* sp and *Thermomyces lanuginosus* sp, has been proposed as an alternative [43, 44]. In the production of biofilm, esterified starch has significantly improved the physicochemical properties of the material. Specifically, using starches derived from sweet potato (*Ipomoea batatas*) and Sago (*Metroxylon sagu*) esterified with 3 % 2-octenyl succinic anhydride and 3-5 % acetic anhydride, there has been a significant decrease in water solubility, water vapor permeability, moisture content and, consequently, an increase in elongation levels, transparency, tensile strength, and tear resistance has been achieved [45, 46, 47].

Based on the above, several studies related to esterified starches in the production of biofilms have reported similarities in the physicochemical properties of the material when using reagents such as succinic anhydride, 2-octenyl succinic anhydride, acetic anhydride, etc. Therefore, the possible variations in the reported values will mainly depend on the botanical origin of the starch [22, 40, 48].

2.3 Oxidation

Oxidation involves the conversion of hydroxyl groups of starch at positions C_2, C_3, and C_6 to carbonyl or carboxyl groups, followed by depolymerization of chains through the cleavage of α-1,4 linkages between glucopyranose units [49]. They are commonly used as oxidizing agents under controlled acidic conditions, like sodium hypochlorite, sodium hyposulfite, sodium persulfate, hydrogen peroxide, nitrogen dioxide, chrysamic acid, among others [50]. Figure 4 illustrates the reaction of starch oxidation by sodium hypochlorite.

Biofilm, obtained from oxidized starch as a raw material, have shown significant changes in the physicochemical and functional properties of the material. For instance, it has been found that using oxidized starches derived from potato (*Solanum tuberosum*) and cassava (*Manihot esculenta*) with sodium hypochlorite ranging from 0.5-1.5 % and potassium periodate at 0.01 %, the biofilm exhibits reduced water solubility and absorption, decreased water vapor permeability and moisture content as well as increased thermal stability and paste clarity. Regarding morphology, the polysaccharide exhibits highly homogeneous and continuous surfaces and cross-sections, attributed to the depolymerization effect of starch resulting from oxidation. This leads to enhanced interaction between the biopolymer and the plasticizer employed [18, 52].

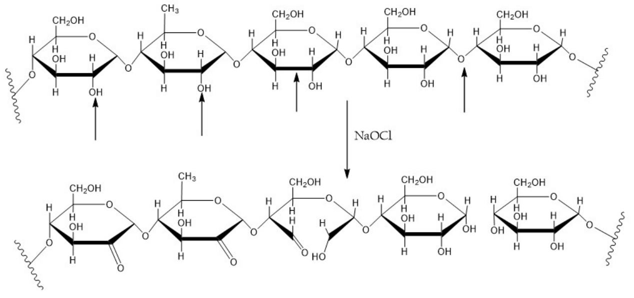

Figure 4: Oxidation of starch using sodium hypochlorite. From Vanier et al. [51].

During the production of biofilm, the starch granules are fragmented due to high temperatures and shear forces, resulting in an amorphous conformation of the polymer. This induces interaction between the plasticizer and the oxidized starch through hydrogen bonding, leading to intramolecular weakening that promotes molecular rearrangement and stability. As a result, the resulting biofilms exhibits improved mechanical properties like tensile strength, elongation at break, and Young's modulus. Additionally, it has been concluded that the mechanical property results are directly related to the proportions between the modification and the plasticizer. With a higher degree of modification and a lower amount of plasticizer employed, the tensile strength increases while elongation and breakage decrease [2],[18],[53, 54].

3 Dual modifications

Until now, the functionality of chemically modified starches has been highlighted when used as raw material in the production of biofilms, resulting in desirable material characteristics. However, the combination of chemical and physical methods, known as dual modifications, has been implemented to optimize the material by improving properties, including swelling power, viscosity, gelatinization temperatures, paste clarity, and solubility. This aims to expand their functions in both food and non-food industries, including the preparation on foods and the production of biofilms that require heat and shear-resistant starches [55, 56].

In general, dual modifications allow, for substitutions of functional groups, molecular rearrangements and granular transformations. In the particular case of functional group substitutions, oxidized and acetylated starches have been prepared using sodium hypochlorite and acetic anhydride, crosslinked-acetylated starches using STMP or STMP/STPP and acetic anhydride, crosslinked-oxidized starches using STMP and sodium hypochlorite and oxidized and crosslinked starches using hydrogen peroxide and boric acid [19],[23],[57]. As an example, Figure 5 shows the dual modification (oxidation and crosslinking) of cassava starch (*Manihot esculenta*), which begins with the oxidation of hydroxyl groups to carbonyl and carboxyl groups using hydrogen peroxide, simultaneously causing the cleavage of the polymer chain and the formation of short chains with low molecular weight. The remaining non-oxidized hydroxyl groups can then be cross-linked by using boric acid as a crosslinking agent.

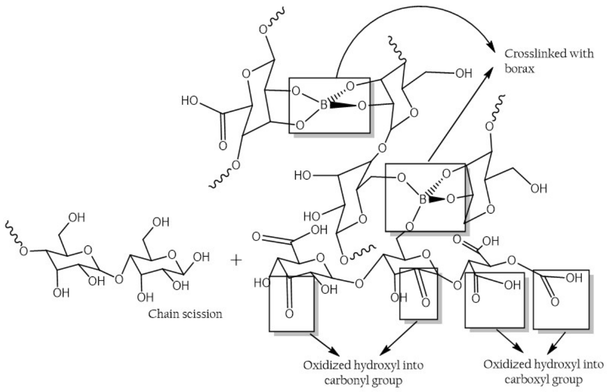

Figure 5: Dual modification in starch: oxidation and crosslinking. From Tanetrungroj and Prachayawarakorn. [57].

Dual modification in starches, including succinylation and annealing, enables both the substitution of hydroxyl groups with succinyl groups (derived from succinic anhydride) and their molecular rearrangement without altering the crystallinity pattern and morphology. This way, starch undergoes changes in its physicochemical properties by increasing the swelling power, apparent viscosity, and gelatinization enthalpy [55]. In contrast, dual modification, like acetylation and microwave irradiation, involve a pre-treatment by dielectric heating followed by the substitution of hydroxyl groups present in the anhydroglucose units with acetyl groups (derived from acetic anhydride in glacial acetic acid). This modification has improved the starch's stability against retrogradation, plasticity and hydrophobicity [58]. Additionally, succinylation has also been combined with non-thermal methods, like pulsed electric field, allowing the starch to be exposed to electric discharges after the esterification process, resulting in an increase in the degree of substitution (> 0.01) within short reaction times [59].

Recently, in the production of starch-based biofilms, dual methods have been employed that combine chemical modifications with thermoplastic synthetic polymers, specially polyethylenes, polybutylene succinates, polyamides, polylactic acid, and polyurethanes. These obtained materials have demonstrated improved thermal stability and tensile or rupture strength, and also moderate changes in crystallinity and morphology [45].

4 Emerging Technologies

Over the last five years, the methods used for the modification of starches, a raw material in the production of biofilms, have undergone significant development due to the need to reduce waste with adverse effects on the environment. Thus, the use of technologies, for example: cold plasma treatment and ozonation, has enabled both surface and structural changes to the already prepared material, along with the substitution and incorporation of functional groups into the polymer, eliminating the use of chemical reagents.

Cold plasma treatment in biodegradable bioplastic films refers to the use of partially or fully ionized gas, which is generated by the action of ions, electrons, molecules or atoms in their excited or ground states. Gases such as oxygen (O_2), argon (Ar), nitrogen (N_2), among others, have been used for this purpose [26],[60, 61, 62]. The application of cold plasma to biodegradable biofilms is performed using a dielectric bar placed on one of the metallic electrodes. The bar acts as a stabilizer, providing control over the multiple discharges and ensuring a high homogeneity on the material's surface, as observed in Figure 6.

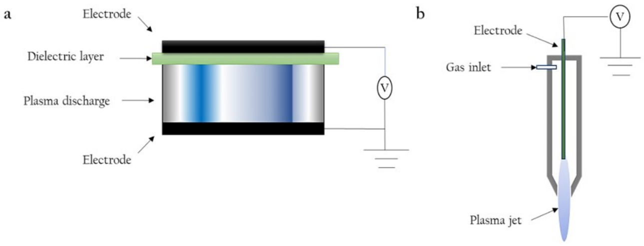

Figure 6: Treatment with cold plasma. a) Dielectric barrier discharge and b) Plasma impregnation. From Pankaj et al. [63].

It has been demonstrated that ionized gases of O_2 or Ar allow an increase in both surface roughness and tensile strength as like hydrophilicity. This is due to the formation of polar functional groups that generate crosslinking reactions along the polymeric conformations [60, 61]. However, when N_2 gas is used, surface changes in the material have been revealed, leading to an increase in water vapor permeability, Young's modulus, and tensile strength [26]. Additionally, the efficiency of plasma treatment depends on various factors, among which the botanical origin of starch, plasma composition, material exposure time to plasma and energy supply are highlighted. [64]. In Figure 7, a sequential diagram is presented illustrating the effects caused by the chemical interaction between plasma and biodegradable biofilms through electrode action, using energy sources such as electric fields, radiation, X-rays, among others.

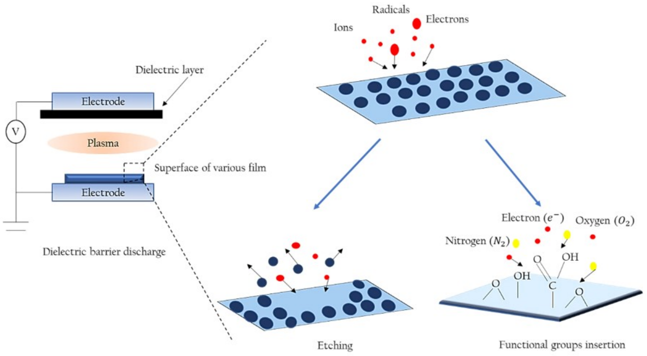

Figure 7: Chemical interaction of cold plasma treatment on starch biofilms. From Bahrami et al. [62].

Another technique is ozonation, a non-thermal method that involves the oxidation of starch with ozone (O_3). This technology is widely employed in the food industry for the use of starch as a thickener or gelling agent [65]. The functionality of this technique can be extended to the production of biodegradable biofilms, as it significantly enhances the physical and chemical properties of the material, following the principles of green chemistry. One of the most commonly used methods for ozone generation is the so called "corona discharge," which involves the use of a high voltage electrical energy source to transform O_2 into O_3 [66].

Initially, a stream of O_2 flows into an O_3 generator and the resulting product is directed into a reactor where dispersion occurs (between 0.1-2.0 ppm) into the starch, while maintaining constant agitation for 15-30 minutes. Subsequently, the suspensions are decanted and the sample is dried at 35°C until reaching a moisture content of 12 %. Finally, any excess O_3 is decomposed, releasing O_2 into the environment. This process is illustrated in Figure 8.

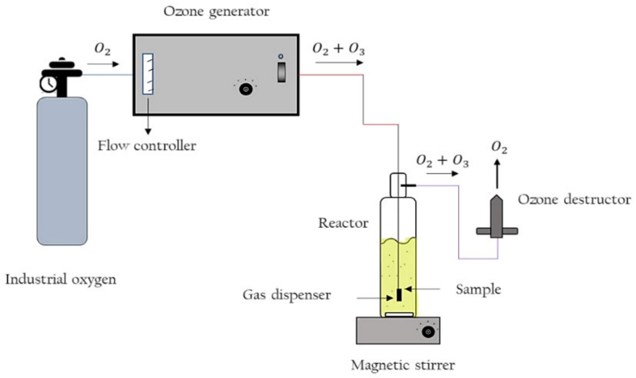

Figure 8: Starch processing system with ozone. From Castanha et al. [67].

During this process, starch undergoes a chemical structure transformation by replacing hydroxyl groups of anhydrous glucose units with carbonyl or carboxyl groups, resulting in changes in both its morphology and properties of adhesion, gel texture, and paste clarity [68]. In biofilms obtained from starch oxidized by ozonation, the repulsive forces of the formed carbonyl and carboxyl groups generate a structural spacing that allows the migration of polar groups to the material's surface. This leads to an increase in characteristics, such a surface homogeneity, tensile strength, Young's modulus, transparency, crystallinity, and water vapor permeability. These properties are useful in the protection of low water activity and moisture-sensitive food products [27]. Table 1 summarizes the general relationship between the modification method of the raw material and its physical and chemical effects on biodegradable starch biofilms.

During the last decade, the incorporation of nanoparticles as reinforcement systems has been utilized in the formulation of starch-based biofilms. This involves the impregnation and subsequent dispersion of nanoparticles onto starch under specific reaction conditions. Depending on their chemical nature, both organic nanoparticles, like: phenolic compounds, ammonium salts, chitosan, chitin, and cellulose, and inorganic nanoparticles, including titanium oxide (TiO_2), zinc oxide (ZnO), magnesium oxide (MgO), calcium oxide (CaO), silicon oxide (SiO) as well as silver cations (Ag^+), gold cations (Au^+), and copper cations ($Cu^{(2+)}$), can be used [76-79].

Specifically, starch-based biofilms reinforced with MgO and Ag^+ nanoparticles allow a decrease in moisture content, solubility, and water vapor permeability, while increasing their tensile strength. Additionally, it has been demonstrated that these films partially or completely inhibit the growth of pathogenic microorganisms, specially *Escherichia coli* sp, *Staphylococcus aureus* sp, *Pseudomonas aeruginosa* sp, and *Salmonella* spp. In addition, TiO_2 nanoparticles have been incorporated at concentrations ranging from 0.5 % to 4.0 % w/w, resulting in reduced moisture content and solubility. Furthermore, the homogeneity of the film-forming solution is improved due to the optimal surface dispersion of these nanoparticles [28, 80].

Food biofilms reinforced with nanoparticles have been termed active biofilms. These films are defined as having the ability to preserve both the organoleptic properties and the shelf life of the food, while also counteracting potential damage caused by external factors or substances that lead to oxidative or microbial processes. However, one of the limitations of incorporating nanoparticles into biofilms is the potential migration of these particles into the food, which poses a potential risk to consumer health. In this regard, some authors have indicated that this risk can be mitigated by considering polymers with a high surface availability, allowing the nanoparticles to remain within the polymeric matrix and preventing migration into the food [81-84].

5 Characterization techniques for modified starches

The physical and chemical characterization of modified starches used in the production of biofilms represents the necessary research support to analyze, determine and compare variables, parameters, methods, stages, and behaviors that justify the functionality of the material. Among the instrumental analysis techniques, Infrared Spectroscopy (IR), X-ray Diffraction (XRD), Thermogravimetric Analysis (TGA), Scanning Electron Microscopy (SEM), Atomic Force Microscopy (AFM), and Optical Microscopy stand out. Table 2 provides an overview of the main capabilities these techniques offer in the characterization of modified starches as raw materials in the production of biodegradable biofilms.

6 Conclusions

Modified starches through physical, enzymatic, chemical or dual methods represent great importance in industries dedicated to the production of biopackaging films, considering the current environmental impact caused by the accumulation of petroleum-derived wrapping materials. Particularly, chemical modifications, including cross-linking, esterification, and oxidation have become essential processes for obtaining biodegradable biofilms, as they positively influence the material's functionality. These modifications contribute to the improvement of its physicochemical properties (morphological structure, moisture, solubility, water absorption index, water vapor permeability, thermal stability, among others) as well as its mechanical characteristics (tensile strength, maximum elongation at break, Young's modulus, among others).

Table 1: Effects caused in biodegradable biofilm obtained from modified starches.

	Method of modification	Principal effects	Botanical source	Ref.
Chemicals	**Cross-linking**	Decreased viscosity, water vapor permeability, and crystallinity percentage. Requires high gelatinization temperatures.	Pearl millet starch (Pearl millet grains), corn starch *(Zea mays)*, barnyard millet starch *(Echinochloa esculenta)*.	[37, 38, 69]
	Esterification	Smooth surface. Decrease in humidity, solubility, and water vapor permeability.	Pearl millet starch (Pearl millet grains), sweet potato starch *(Ipomoea batatas)*, sago starch *(Metroxylon sagu)*.	[22, 40, 46]
	Oxidation	Decrease in tensile strength, solubility, and water vapor permeability.	Cassava starch *(Manihot esculenta)*, potato starch *(Solanum tuberosum)*, corn starch *(Zea mays)*.	[18, 52, 70]
Dual	**Oxidation–Crosslinking**	Decrease in crystallinity and water vapor absorption. Improved extensibility.	Cassava starch *(Manihot esculenta)*, corn starch *(Zea mays)*.	[57, 71]
	Esterification–Crosslinking	High elongation percentages. Low solubility and water vapor impermeability.	Potato starch *(Solanum tuberosum)*, Sorghum starch *(Sorghum bicolor)*.	[23, 24]
	Oxidation–Esterification	Increase in transparency, water resistance, water vapor permeability, and tensile strength.	Cassava starch *(Manihot esculenta)*, corn starch *(Zea mays)*.	[72, 73]
Emerging technologies	**Cold plasma (O_2, Ar, N_2)**	Decreased solubility, moisture and water vapor permeation. Improved mechanical properties and high thermal stability.	Corn starch *(Zea mays)*, wheat starch *(Triticum)*, kithul starch *(Caryota urens)*.	[26, 60, 74]
	Ozonation	Low water solubility, high transparency, increased tensile strength, and Young's modulus.	Arracacha starch *(Arracacia xanthorrhiza)*, cassava starch *(Manihot esculenta)*, potato starch *(Solanum tuberosum)*.	[27, 68, 75]

Table 2: Relationship between the scope of the main characterization techniques for starches and the type of modification used in the raw material.

Instrumental technique	Scope	Type of starch modification (examples)	Ref.
Infrared Spectroscopy (IR)	Elucidation of functional groups. –OH and –NH_2 stretching. Plastifier affinity with biopolymer through hydrogen bonding formation.	Crosslinking in potato starch (*Solanum tuberosum*), wheat starch (*Triticum*), and barnyard millet starch (*Echinochloa esculenta*).	[39, 69, 85]
	Formation of carbonyl groups due to the stretching of C=O in the acetyl group and C=O in carboxyl groups of esters.	Esterification in sweet potato starch (*Ipomoea batatas*), and cassava starch (*Manihot esculenta*).	[12, 17, 40]
	Presence of bending vibrations –OH groups (confirmation of intermolecular bonding between oxidized starch and polyvinyl alcohol) and stretching of C=O corresponding to the formed carboxyl group.	Oxidation in bean starch (*Phaseolus lunatus*) and potato starch (*Solanum tuberosum*).	[50, 52, 53]
X-ray Diffraction (XRD)	Detection of ordered structures, changes in three-dimensional networks and crystallinity percentages of the biopolymer.	Cross-linking in oca starch (*Oxalis tuberosa*), esterification in rice starch (*Oryza sativa*), oxidation in cassava starch (*Manihot esculenta*).	[27, 33, 86]
Thermogravimetric Analysis (TGA)	Determination of gelatinization temperatures, decomposition temperatures, mass loss, and thermal stability of the material. Evidence of compatibility with modifying agent.	Cross-linking in porous starches, esterification in corn starch (*Zea mays*), oxidation in cassava starch (*Manihot esculenta*).	[18, 39, 45]
SEM, AFM and Optical Microscopy	Characterization of microstructure through changes in granule surface: vertices, edges, sizes, and shapes.	Cross-linking in porous wheat starch (*Triticum*), esterification in rice bean starch (*Vigna umbellata*).	[10, 39, 87]

With scientific and technological progress, the food industry has focused its research on obtaining active starch biofilms that coat and provide protection against factors, such as: radiation or the growth of pathogenic micro-organisms. The incorporation of modification methods, like cold plasma treatment or ozonation, along with reinforcement techniques using organic and inorganic nanoparticles, represents a promising approach to optimize the material properties, ensure food safety, and extend the shelf life of food products during their distribution and commercialization stages.

Acknowledgements

The authors are grateful to the Research Group in Chemistry and Food Technology (GIQTA) at the Universidad Pedagógica y Tecnológica de Colombia-UPTC for their support in the development of this research. They also thank the anonymous referee for their detailed review, which contributed to improving the presentation of the paper.

Declaration of conflict of interest

The authors declare that they have no conflicts of interest.

Author contributions

All authors contributed equally and collaboratively to all aspects of the research and manuscript preparation, including the conceptual design, data collection and analysis, and drafting and revising the article. All authors approved the final version of the manuscript.

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