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PECTIN – CANDELILLA WAX: AN ALTERNATIVE MIXTURE FOR EDIBLE FILMS

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ABSTRACT

Edible films and coatings have received special attention in recent years due to the advantages that represent their use as edible packaging over synthetics materials. This contributes in high degree to reduce the environmental pollution with non-biodegradable materials. By functioning as barriers, such edible films and coatings can improve the recycling and/or substitute some synthetic packaging materials. New packaging materials have been developed and characterized by some scientists from natural sources (biomaterials); however, it is necessary the manufacture tailor-made to every food. The main objective of this review is to provide basic and applied information and benefits that can be generate the use of two products with low cost (candelilla wax and pectin), but with great importance in the food industry and that can be used to manufacture edible films and coatings.

Keywords: Pectin, candelilla wax, glycerol, edible films, foods

INTRODUCTION

Two of the major problems in the food industry are the waste disposal from fruit industry and the short shelf life of fresh products. However, at present there are few but important alternative, like eco-solutions, for these problems, such as composts, the use as supports for solid-state fermentation, recovery of bioactive compounds, etc. Among these, we can find the recovery of pectin for use as an additive in the food industry (jams, preserves, jams, jellies, cheeses, ice creams, sauces, etc.). The main sources of pectin are apples, citrus, pineapple, guava, tomato, passion fruit and beetroot. It is worth mentioning that the quality of pectin depends on the source and the extraction process (physical-chemical or enzymatic). Also used in other industries such as pharmaceuticals, to modify the viscosity of their products and in the plastics industry and in the production of sparkling wines, as fining agent and binders (**Gomez, 1998**). However, there are some studies that point to the possibility of generating biomaterials (biofilms groceries) to coat and extend the shelf life of these products fruits.

PECTIN

Pectin is a complex hetero-polysaccharide, which is present in nature (fruits and vegetables in particular) as a structural element of the cellular system of the plants (cell wall) with important roles in their physiology (**Contreras-Esquivel *et al.*, 2006; Voragen *et al.*, 2009; Qiu *et al.*, 2009; Jamsazzadeh-Kermani *et al.*, 2015**). Chemically are linear chains of D-galacturonic acid joined by α-1,4-glycosidic linkages, which is partially esterified with methanol (**Albertsheim *et al.*, 1996; Berardini *et al.*, 2005; Lazar, 2005; Xie *et al.*, 2008; Fishman *et al.*, 2015**) (Fig 1.).

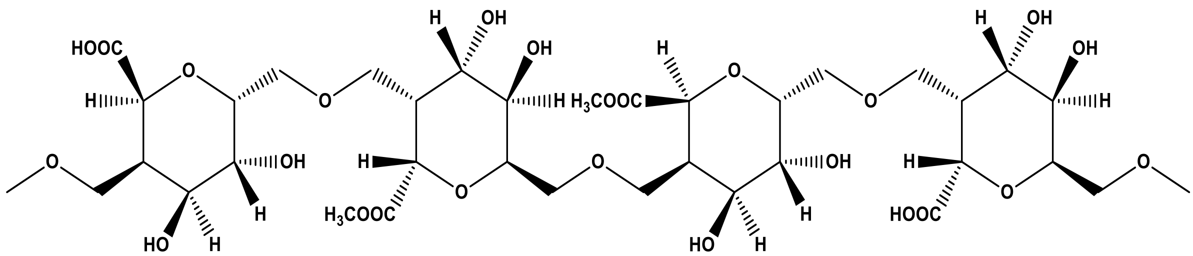

Figure 1 Chemical structure of pectin

The amount of esterified units influences the properties and functional classification. Commercially are divided into pectin with a degree of methylation above 50% called high methoxyl (HMP), which gels very easily after heating in sugar solutions at a concentration above 55% and a pH of less than 3.5. Furthermore, the gel formation with low methoxyl pectin (LMP; DM <50%) requires the presence of calcium ions, this can be used as a gelling agent in low-sugar products such as jams and jellies low calorie (**Oakenful and Scott, 1984; Pszczola, 1991; Thakur *et al.*, 1997; Contreras-Esquivel *et al.*, 2006; Jamsazzadeh-Kermani *et al.*, 2014**). Usually, low methoxyl pectins are produced by high methoxyl pectins demethylation methods in which various agents can be used: acid, alkali, enzymes, and ammonia in alcohol (**Iglesias and Lozano, 2004**). Traditionally, the pectin is obtained from raw materials at acidic conditions with elevated temperatures (**Koubala *et al.*, 2008; Wang *et al.*, 2014**), followed by precipitation using alcohols such as isopropanol (IPA), methanol or ethanol. (**May, 1990; Fishman and Cooke, 2009; Zhang *et al.*, 2013; Xu *et al.*, 2014**).

Chemical Composition

Chemically, pectin's are macromolecules composed of a complex group of heteropolysaccharides. Pectin molecules consist of three different sequences known as homogalacturonan (HGA), rhamnogalacturonan I (RG-I) and rhamnogalacturonan-II (RG-II), which constitute the two main regions of pectin, i.e. the linear region (HGA) and branched region (RG-I,-II). Homogalacturonan sequences consist α acid residues D-galacturonic α -4 linked together with partially methylated substitutions and / or acetylated, which are interrupted by L-rhamnose residues in adjacent positions and/or staggered. RG-I consists of a disaccharide repeat units [2] - α-L-rhamnose-acid-α-1-4-D-galacturonic-(1], plus units also contain L-arabinose and D- mainly galactose, while the RG-II is a low molecular weight region, which is composed of a backbone of units of D-galacturonic acid α -4 linked together and some other rare sugars such as apiose, 2 -O-methyl-fructose, 2-O-methyl-xylose, 2-keto-3-deoxy-D-mannan-octulosonico (KDO), 3-C-carboxy-5-deoxy-L-xylosyl, among others; in both cases the content of such sugars vary depending upon the source, location, extraction method and other environmental factors (**Berardini *et al*., 2005; Contreras-Esquivel *et al*., 2006; Yuliarti *et al*., 2015**)

CANDELILLA

The plant (*Euphorbia antisyphilitica Zucc*) is a perennial shrub with densely compact, erect, cylindrical stems coated in wax (serves as protection for moisture retention), with the appearance of small candles, leafless and that becomes full of small flowers in the rainy season (**Scora *et al*., 1995; Saucedo-Pompa, 2007**). It belongs to the *Euphorbiaceae* family (**Steinmann, 2002**) and is considered an endemic species of the semiarid regions of the states of Chihuahua, Coahuila, Durango, Hidalgo, Nuevo Leon, Oaxaca, Puebla, Queretaro, San Luis Potosi, Tamaulipas and Zacatecas, México. In the United States of America, is distributed in the states of New Mexico and Texas. This plant promotes the growth of some other plants such as Lechuguilla and Sotol. Its uses date back to the Indians of northern Mexico that used for ornamental or plant used as candles, to bend a bow, tanning hides and traditional medicine (**Romahn, 1992; Kowalczyk and Baraniak, 2014**).

Candelilla wax

Candelilla wax is a complex material, hard, shiny and easy spray that is derived from plant with the same name through a "traditional" extraction of candelilla wax using a boiling solution of sulfuric acid (scrap fertilizer industry) in which the plant is submerged and the wax is recovered as a foam on the boiling water surface. However, this process is highly polluting, generates low quality wax and high health damage. Its color varies from light brown to yellow depending on the degree of refining. This material serum is used to harden other waxes, manufacturing synthetic polishes, polishes, for transport and storage of products as well as in various industries such as food, cosmetics, electrical, mechanical and plastic production (**Cervantes-Ramirez, 2005; Canales-Gutiérrez *et al*., 2005; Rojas-Molina *et al*., 2011; Rojas-Molina *et al*., 2013**).

Chemical Composition

Chemically, are esters of fatty acids and long chain fatty acids. Characterized by a high content of hydrocarbons (about 50%) and a relatively low amount of volatile esters. Its resin content can reach 40% of its weight, which gives it a sticky consistency. Is insoluble in water, but soluble in acetone, chloroform, benzene and other organic solvents. (**CENAMEX, 2006; Ochoa *et al*., 2011; Rojas-Molina *et al*., 2013**). The following tables describe the composition and physicochemical properties of candelilla wax and because make it one of the most versatile products for industry (Tables 1 and 2).

Table 1 Typical composition of refined candelilla wax

Compound	% Weight
Hydrocarbons	**50-57**
Nonacosane	2.5
Hentriacontane	46-46.5
Tritriacontane	2.5
Esters	**28-29**
Simple esters and lactones	20-21
Hydroxilated esters	6-8%
Alcohols, Esterols y Resines	**12-14**
Miricílic alcohol	ND
Sitosterol and other esterols	7-8
Beta-amirine acetate	5-6
Free acdis	**7-9**
Lineal chain	6-7
Cycles	ND
Moisture	**0.5-1**
Inorganic residues	**0.7**

ND: Not detected

Tabla 2 Physico-chemical composition of candelilla wax

Properties	Refined
Acidity value	12-22
Iodine index	14-27
Saponification number	35-87
Melting point	67-79 °C
Refraction index	1.4545-1.462 @ 85 °C
Unsaponificable material	67-77
Specific gravity	0.885
Flame point	-----

SOURCE: Candelilla institute (http://www.candelilla.org/es/propiedades.htm)

EDIBLE FILMS

Conservation through coatings is an ancient practice that was developed to mimic natural edible covers of vegetable products. There are reports dating from the twelfth and thirteenth centuries in which it is mentioned that in China, oranges and limes were dipping in wax to retard water loss (**Kaplan, 1986; Greener and Fennema, 1994**). During the sixteenth century in England was practiced the "buttered", that is the fat use as a coating of food products to prevent moisture loss also of these. In the nineteenth century were used films based on gelatin for preservation of meats and other foods, around the 30's, paraffin waxes were already commercially available, that melted with heat, was used for coating citrus (**Nussinovich and Lurie, 1995; Park *et al*., 2014**), and in the early 50's were developed oil-water emulsions of carnauba wax for coating fresh fruits and vegetables (**Kaplan, 1986; Kester and Fennema, 1986**). From the mid 50's to mid 80's a lot of work was focused on the use of films and coatings to extend shelf life and improve the quality of fresh, frozen and processed foods, which has been reported in both the scientific literature as patent. Unfortunately, most of this work is of limited value because of the lack of quantitative data on the characteristics of barrier coatings. It has been reported that the waxes were first used in edible coatings on fruits like apples and pears. In recent years, it has been reported that it is possible to achieve similar effects barrier to water vapor and gases in diverse products using different mixtures of oils (**Shojaee-Aliabadi *et al*., 2014; Wang *et al*., 2014a**; **Sanchez-Aldana *et al*., 2015**), waxes (**Fagundes *et al*., 2014; Pérez-Gago and Rhim, 2014; Rodrigues *et al*., 2014**; **De León-Zapata *et al*., 2015**), proteins (**Vonasek *et al*., 2014; Khanzadi *et al*., 2015; Hopkins *et al*., 2015**) and polysaccharides (**Costa et al., 2015; Wan *et al*., 2015; Razavi et al., 2015; Gutierrez *et al*., 2015; Dang and Yoksan, 2015;)**

The growing demand for food maintain maximum organoleptic properties, has fostered the continuous improvement of the processes used in the food industry in order to ensure its preservation and without affecting the quality and shelf life (**Soliva-Fortuny and Martín-Belloso, 2001; Ochoa *et al*., 2011; De León Zapata *et al*., 2015**). The development of edible films and coatings and biodegradable coatings has received the most attention because it leads to the extension of the shelf life of food. Edible films are defined as one or more thin layers of a material that can be consumed by living organisms and which in turn can act as a barrier to the transfer of water, solutes and gases of food (**Guilbert *et al*., 2006**). **Krotcha *et al*. (1994)** defined edible films as thin continuous layers of edible material formed on (as a cover) or placed between the food components, and provide a means to carry food ingredients or additives and improve the handling of the thereof.

Interest in the edible films have grown considerably, because have several advantages, they can be ingested by the consumer, the cost is usually low, their use reduces waste and pollution, can improve the organoleptic, mechanical and nutritional properties in food, provide personal protection to small pieces or portions of food and can be used in heterogeneous foods as barriers between components. Edible films should be good moisture barrier to protect the food completely coated, have good barrier properties against oxygen, and have good mechanical and organoleptic properties (**Guilbert *et al*., 2006; Sánchez-Aldana *et al*., 2014**). However, for the development of these films is necessary to consider the mechanical factors involved in chemical and physical storage of the fruit. They can provide nutritional and organoleptic properties to foods when added antioxidants (**Saucedo-Pompa *et al*., 2009; Ochoa *et al*., 2013; Ferreira *et al*., 2014; De León Zapata *et al*., 2015**), artificial colors or flavors (**Perez and Gaonkar, 2014; Kim *et al*., 2014**) and others additives (**Arrieta *et al*., 2014; Chiumarelli *et al*., 2014; Jouki *et al*., 2014; Kulkarni *et al*., 2015**).

Lipid-polysaccharide Interactions

The feasibility of the formulation of edible coatings based on candelilla wax and pectin is based on pectin-lipid interaction. Due to candelilla wax are long chain fatty acids esters, the interaction may occur mainly by hydrogen bonds involving carboxylic moieties of pectin. An example is the occurrence of hydrogen bonds between methoxycarbonyl groups with oleic acid as the main bonding force. This was demonstrated by **Falk and Nagyvary, (1982)** in their study exploratory of pectin-lipid interactions by using C13 NMR. However, the galacturonic acid sequences is the more suitable than glucuronic acid sequences to entrap cholesterol or linoleic acid molecules probably due to the difference of conformation of the two molecules (**Pau-Roblot *et al*., 2010**). This is because the long-chain lipid (in this case, candelilla wax) may interact with most of the carbons in a "pocket apolar" in the galacturonan and with the helix structure of glucuronan that provides only external carbon to interact with the lipid. Furthermore it is known that pectin under optimal conditions (in a physiological system) can bind four times its weight in lipids (**Falk and Nagyvary, 1982**).

Films based on lipid-polysaccharides emulsions

In the formulation of films and coatings, it is necessary to use at least one component capable of forming a structural matrix with sufficient cohesiveness. Edible films made by combining several compounds, have been refined to take additional functional properties of each component, thus minimizing their disadvantages. Film forming substances create a continuous structure through interactions between molecules under the action of a physical or chemical treatment (**Bureau and Multon, 1995; Guillbert *et al.*, 1996; Debeaufort *et al*., 1998; Soliva-Fortuny and Martín-Belloso, 2001**). Film forming polysaccharide frequently involves the formation of a gel and/or solvent evaporation. The polysaccharides can be used in the preparation of edible films. Generally produce films with good mechanical properties and are effective barriers against non-polar compounds. However, its hydrophilic nature makes present a low resistance to water loss (**Parra *et al*., 2004**). Its selectivity regarding permeability to oxygen and carbon dioxide conditions creating modified atmosphere inside the food, resulting in an increased product life.
Polysaccharides, including pectin, have been studied as edible film forming materials (**Diab *et al*., 2001; Lizaridou and Biliaderis, 2002; Marianello *et al*., 2003, Lee *et al*., 2004, Parra *et al*., 2004, Thomas *et al*., 2004; Turhan and Sahbaz, 2004; Tapia Blacido *et al*., 2005; de Brito *et al*., 2013; Arancibia *et al*., 2014; Lucenius *et al*., 2014; Mohan *et al*., 2015; Yang *et al*., 2015**). These compounds have the characteristics of being long-chain polymers, soluble in water and produce a strong increase in viscosity when dispersed in it. So the use of a polysaccharide such as pectin for the manufacture of biomaterials for fresh fruit conservation is not a good alternative for its high hydrophilicity and water vapor permeability. For this reason, it is necessary the use of a lipid component in the formulation to improve the properties of the laminate. Edible films made from lipid include natural waxes, like candelilla wax. These materials are typically used with the purpose of providing hydrophobicity to a certain surface and make an effective barrier against moisture (**Soliva-Fortuny and Martín-Belloso, 2001**). However, their use individually, not mixed with other substances, it is limited because most lack sufficient structural integrity and stability. Therefore, generally require the presence of a matrix serving as support. In this regard, studies on edible films and coatings using mixtures of lipid compounds and proteins or carbohydrates (support material) were made in order to exploit the characteristics of each material, improving the final properties of the films (**Aguilar-Mendez, 2005**). For this purpose, is considering the use of a polysaccharide and a lipid component, for the preparation of edible films.
Saucedo Pompa *et al*. (2009) studied the effect of the addition of ellagic acid in a matrix of Candelilla wax on quality and shelf life of avocados. According to their results using ellagic acid as part of an edible film that significantly minimizes changes in appearance, solid content, pH, aw, lightness and weight loss, maintaining the quality of avocados, prolonging its shelf life. Furthermore, films were able to significantly reduce the damage caused by *C. gloeosporioides* major fungi which attack these fruits. It is found that using this new protection system, the negative effects by this fungus can be reduced satisfactorily. **Ochoa *et al*. (2011)** report the effect of the application of an edible coating made from candelilla wax and a potent antioxidant 0.01% (ellagic acid) to extend and improve the quality shelf life of apples "Golden Delicious". Their results show that the coating prevents apparent changes in the fruit and it is an excellent barrier against microorganisms tested fungus *Colletotrichum gloeosporioides, Fusarium oxysporum* and *Penicillium expansum*.
Moalemiyan *et al*. (2011) evaluated the effect of an edible coating based on pectin in the quality and in the extending the shelf life of the mangoes, which included various combinations of pectin, beeswax, sorbitol and monoglyceride. The results shown merely mentioned which is covered decreases breathability and reducing weight by evaporation of water, as well as the respiration rate and prevents or retards the production of ethylene. Besides reducing changes in certain physical and chemical properties. **Galus and Lenard, (2013)** characterized the physical properties of composite films, including thickness, color, and kinetic adsorption isotherms of water vapor, water vapor permeability, tensile strength, elongation at break, and microstructure, that were prepared with sodium alginate and low methoxyl pectin. Homogeneous and transparent films were obtained, and changes in its composition did not affect visual appearance. The results show that the thickness depends on the formation and composition of the solutions. The absorption kinetics and isotherms show a hydrophilic character. A high pectin content tends to change the internal structure of the films, less organized than alginate. In the Food Research Department of the Autonomous University of Coahuila, are working on the development of strategies to formulate edible films based on pectin and candelilla wax, in order to establish new methods for preserving fresh foods.

PERSPECTIVES

In recent years, the trend toward using biodegradable packaging has increased considerably. This is due to the serious problem of pollution generated by the use of non-biodegradable materials and deposition of materials in the food industry. Considering the advantages of the use of waste materials for make them packaging systems edible and/or biodegradable, has increased the interest of national and international researchers to develop packaging alternatives. However, for developing packaging materials other components are required to improve and/or modify its properties. The use of glycerol as a plasticizer is large because of its availability and low cost, however, also used some monosaccharides such as glucose, fructose, etc.
From the functional perspective of packaging materials, commercial application will be driven as well as develop of tailor-made materials. Further characterization is necessary to pack products for assessing the compatibility of the components. From the perspective, the use of specialized techniques for the development and characterization of materials and product packaging to pack will be driven by the development of rapid techniques to determine compatibility, stability and shelf life. Specifically, the development of techniques such as permeability to water vapor, O_2, CO_2, ethylene, etc., thermal stability, behavior at different relative humidity's, solubility and faster methods of application such as spraying. However, it is of utmost importance that these methodologies are economical, this will undoubtedly facilitate their integration into the product to be preserved using experimental setups to evaluate accelerated shelf life scale that allows real time to validate its functionality.
From the engineering perspective, not only requires the development, characterization and functionality of new tailor-made packaging materials. Also, it is necessary to work on the design of equipment capable of producing large-scale continuous such materials, as well as application equipment. However, although not easy, is an important line of work as required to reduce the disadvantages that may arise in the development, low cost, efficient and fast.
Finally, from the perspective of innovation, it is necessary to use low-cost materials, accessible and always available, that is why the pectin for being a byproduct of the food industry is an excellent candidate to be used, however, a mixing with other components is necessary to reduce their hydrophilicity, as candelilla wax. Additionally, glycerol is a good candidate for their plasticizing properties, availability and economy.

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