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Whey Protein Films and Coatings: A Review

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Abstract: Whey is a by-product of the cheese-making industry. The most important constituent of whey is Whey Protein (WP) generally used in infant formulas and sports food. Nowadays, great efforts are being made to find out new WPs applications, e.g. production of edible films. Edible or biodegradable films constitute a convenient means to prolong the shelf life of foods and increase their quality without contributing to environmental pollution. Apart from acting as selective barriers for moisture and gas migration, these films may operate as carriers of many functional ingredients. Such ingredients may include antioxidants, antimicrobial agents, flavors, spices and colorants which improve the functionality of the packaging materials by adding novel or extra functions. In this article, the functional properties of edible films made with WPs and their applications in food industry will be reviewed.

Key words: Whey protein, edible film, coating, functionality, permeability

INTRODUCTION

Edible films and coatings have been involved in food preservation for centuries. Since the twelfth century, civilizations have been using wax coatings as a method to lengthen shelf life of foods (Debeaufort *et al.*, 1998). The main purpose was to prevent the loss of moisture and to maintain quality and texture during storage. Today, waxes are still commonly used to preserve fruits, vegetables and meats for extended shipping and shelf life, but there are now more materials available for edible films and coatings. These alternative sources, along with improved processing techniques, have extended the use of edible films and coatings beyond simple moisture barriers. Since the early 1900s, food-grade shellac resins have been used for improving the appearance of foods (Beckett, 2000; Valencia-Chamorro *et al.*, 2009). While these particular edible coatings have provided nominal protection or improvement to the physical appearance of foods, their uses are limited. Research in the area of edible films has improved the technology and created more options. Carbohydrates and proteins are biopolymers that can form films like waxes, lipids, or resins. Corn zein has been employed commercially as a food coating since World War II to enhance food appearance (Lawton, 2002). The diverse structures and chemistries of carbohydrates and proteins offer a wide array of films and coating properties. Protein-and carbohydrate-based edible films are generally more cohesive and flexible than wax films and possess better gas-barrier properties at certain conditions. Biopolymers also have the advantage of having more possibilities for adjusting film properties for specific application development through chemistry and processing.

Many publications have been written reviewing the properties of edible films and coatings formed from biopolymers such as wheat gluten, soy protein, starch, cellulose and casein (Gennadios and Weller, 1990; Brandenberg *et al.*, 1993; Krochta, 2002; Gennadios, 2002; Zhang and Mittal, 2010). While many biopolymers have been studied as edible films and coatings, this article focuses on Whey Protein (WP).

Edible films and coatings: Specifically, an edible film is a thin continuous sheet formed from a biopolymer matrix that is cohesive enough and has the physical integrity to stand alone. The thickness of an edible film is typically 2-10 mils (0.050-0.250 mm). Depending on their thermal properties and surface chemistry, edible films can be formed into pouches or laminated onto other packaging substrates. The main purpose for edible films from biopolymers is to control mass transfer of multiple compounds including gas, aroma, oil and water vapor into or out of a food, preserving food quality. Edible films must also be both strong and flexible to withstand forces experienced during handling and processing.

Edible coatings are edible films formed directly on the surface of a food or material. They are typically thinner than stand-alone edible films. While edible coatings themselves can improve the physical integrity of a coated product, they do not necessarily need to be as tough and resilient as a stand-alone film because of the underlying support of food. Additionally, edible coatings can improve appearance of a product by adding color or gloss, making it more appealing to consumers. Edible films are the form used to study mechanical, barrier and surface properties. Coatings are studied as one type of application of an edible film.

Most edible films are formed by removing the solvent, called solvent casting. For years, solvent casting has been the main method of forming WP films for research. Two possible solvent choices to keep the films safe for Food-grade are the water and ethanol consumption. Solvent casting starts with a dilute solution of biopolymer. The solution is spread in a thin layer on a level surface and the solvent evaporates to form a film. Under ambient conditions, edible film drying can occur with hot air, infrared energy, or microwave energy. Method of drying can significantly affects the physical properties of the final film. There are two periods during drying of the films: The Constant Rate Period (CRP) and the Falling Rate Period (FRP) (Kozempel *et al*., 2003). During the CRP, the major phenomenon is mass transfer between the surface of the film and air. Once the surface comes into equilibrium with air conditions, the FRP begins. During this period of drying, the mass transfer of water from the film to the air is limited by diffusion of water from the inside of the film to the surface. Some factors are important in air drying are (1) exposed surface area, (2) drying air temperature, (3) drying air Relative Humidity (RH), (4) drying air velocity and (5) drying period (Alcantara *et al*., 1998). Microwave energy can penetrate more quickly into films, leading to faster diffusion of water from the center of the film to the film surface. Different drying conditions can change edible film morphology, which affects appearance, barrier and mechanical properties (Perez-Gago and Krochta, 2000).

Whey Protein (WP) film and coating formation

Whey Proteins (WPs): There are several individual proteins within the mixture of WP, with β-lactoglobulin, α-lactalbumin, Bovine Serum Albumin (BSA) and immunoglobins being the main proteins (deWit and Klarenbeek, 1983; Kinsella, 1984). Among them, the most abundant and important protein for film formation is β-lactoglobulin and the second most abundant WP is α-Lactalbumin.

Solvent casting of WP films: Solvent-cast WP films, for which the solvent is water, can form from native proteins through the electrostatic interactions, hydrogen bonding and van der Waals forces that occur between the protein chains as the water evaporates. Native films are cohesive, but the protein film network can be improved and the resulting solvent-cast film tensile and barrier properties improved through heat denaturation and cross-linking of the WP chains (Perez-Gago and Krochta, 1999). Thus, most research on WP films has involved heat denaturing of the WP in aqueous solvent and then casting the solution to form a film with cross-linked WP upon evaporation of the water. Polymerization is not the only chemical reaction involved in WP film network formation. Noncovalent aggregation also occurs through new hydrophobic, ionic and van der Waals interactions that occur between newly exposed groups of the heat-denatured whey protein. These interactions increase as pH decreases toward the isoelectric point of WP (Kinsella, 1984; Kinsella and Whitehead, 1989).

There are other methods for inducing protein chain cross-linking besides heat denaturation. Irradiation has been successfully used to cross-link casein proteins, as well as soy proteins (Brault *et al*., 1997; Lacroix *et al*., 2002). A hypothesized mechanism is radical polymerization through tyrosine and the formation of bityrosine linkages between protein chains. However, WPs are low in tyrosine residues and irradiation alone does not produce a significant increase in molecular weight of WP (Vachon *et al*., 2000).

Properties of solvent-cast WP films: The most important characteristics of edible films are mechanical, barrier and appearance properties, because they determine under what conditions they can be applied and used. As with traditional plastic film packaging, the most significant mechanical properties of interest are tensile strength, elastic modulus and percent elongation. The most important barrier properties are determined as film oxygen permeability and water vapor permeability (Zhang and Mittal, 2010). Carbon dioxide, oil and aroma permeability properties are also of interest, but the information is of value for more specific applications. The most important appearance properties are transparency, color and gloss.

Mechanical properties: Tensile properties-tensile strength, elastic modulus, percent elongation and resiliency-are indicators of protein-protein interactions in WP film matrices. Tensile strength is the maximum amount of force applied to a film per unit original cross-sectional area before film breakage. Elongation is the distance the film will stretch before breaking divided by the original film length. Resiliency is the film's overall toughness. It can be estimated by multiplying tensile strength by percent elongation. The tensile properties can be adjusted to make more flexible, stretchable, resilient films by changing the state of the protein or by the addition of plasticizers. Increased cross-linking that occurs during denaturation leads to stronger and stiffer films with greater elongation (Perez-Gago and Krochta, 1999) compared to films made with WP in the native form. The cross-linking of WP chains produces stronger films, but also allows for greater deformation of the films. The amount and type of plasticizer in a WP film also affects tensile properties. Plasticizer efficiency, or how well a plasticizer adjusts tensile properties, is dependent on the size, shape and compatibility of the plasticizer with the protein.

Permeability properties

Oxygen permeability: Good oxygen-barrier properties of packaging materials are critical for maintaining the initial high quality of the packaged product. The oxidation of fats, oils and other food components produces off-flavors, off-colors and nutrient loss. Thus, the protection of the content against oxygen is one of the most important requirements in packaging of food products. Despite the availability of a variety of excellent synthetic oxygen barriers, the disadvantages of any such composite polymeric structures are the difficulties entailed in their recycling. Existing composite films containing layers of different plastic materials may not be recycled, because typically only single component plastics are recyclable. Therefore, there is an increasing interest in the development of biodegradable polymers (i.e., biopolymers) for packaging materials that have suitable application properties and can be disposed of after use in an economically and ecologically acceptable way. Properties, as well as potential practical uses of biopolymer films and coatings based on polysaccharides, proteins and lipids from numerous plant and animal sources, have been well reviewed (Debeaufort *et al*., 1998; Gennadios, 2002). Particularly among biopolymers, the extremely low oxygen permeability of WP films, in addition to good gloss and mechanical properties, makes WP potentially useful as a transparent coating material for improving the oxygen-barrier property of food packaging (Miller *et al*., 1998). The compositions of WPI and WPC products also differ in the levels of the various constituents, especially lactose that acts as a plasticizer. These composition differences may influence markedly the barrier and mechanical properties of WP films. Polyethylene (PE) and Polypropylene (PP) have been widely used in diverse packaging applications due to abundant supply, low cost, good processability, low energy demand for processing and resistance to chemicals and harsh environments. Common polyolefin films such as PE and PP are also excellent moisture barriers, but they must be coated or laminated with synthetic polymer layers including EVOH (ethylene vinyl alcohol) copolymers, PVDC (poly-vinylidene chloride) and nylon to provide an oxygen barrier (Hernandez *et al*., 2000). The resulting structures are quite expensive and non-recyclable. Replacing these synthetic oxygen-barrier layers with WP coatings could provide a new path for use of WP sand perhaps improve recyclability of the base plastic film (Hong and Krochta, 2003, 2004).

Water vapor permeability: Since whey is hydrophilic protein, these films are only moderate barriers to moisture at best. The water vapor permeability of WP films is high, but the barrier properties of the films have been improved through addition of hydrophobic materials like waxes and lipids. While addition of lipids and waxes can greatly improve the water vapor permeability of WP films, their affect on tensile properties must be considered. At high levels, especially for brittle waxes, tensile strength and elongation decrease and films become brittle and hard to handle without breaking (Shellhammer and Krochta, 1997). However, there is a positive effect on tensile properties of decreasing particle size of insoluble additives in protein films (Dangaran *et al*., 2006). Perez-Gago and Krochta (2001) found tensile strength and elongation significantly increased when particle size of beeswax in WP films decreased. Compare to synthetics, WP films are only moderate moisture barriers. Even with the inclusion of lipids, WP films still have higher water vapor permeabilities than Low-Density Polyethylene (LDPE), High-Density Polyethylene (HDPE) and nylon. In terms of applications, WP films may be best for food products needing a low to moderate moisture barrier to avoid condensation from forming on the surface. Moreover, the appearance needs to be considered because including lipids and waxes confers some opaqueness. They may have some short-term use inside food as protective layers between high and low water activity layers like cookies and cream fillings or piecrusts and fruit fillings.

Aroma and oil permeability: WP films have been found to be excellent barriers to aroma compounds and oil (Miller *et al*., 1998). This is consistent with the findings that WP limits the flavor perception of benzaldehyde, citral and D-limonene (Hansen and Heinis, 1992) and that β-lactoglobulin has been found to be a binder of aromatic compounds (Farrell *et al*., 1987). Miller *et al*. (1998) developed a method for determining permeability of aroma compounds through films. They used the method to determine that WP films had better barrier to D-limonene than vinylidene chloride copolymer (co-VDC) by 250-15,000 times, depending on relative humidity, but not as good a barrier as Ethylene Vinyl Alcohol Copolymer (EVOH). By monitoring the penetration of dyed vegetable oil on whey protein-coated paper over time, Lin and Krochta (2003) compared WP coatings with various plasticizers as barriers to oil. They found WP plasticized with glycerol (1.3 M) prevented the penetration of oil into the paper for at least 16 h.

Optical properties (color, gloss, haze and transparency): WP films have the two characteristics-transparent and highly glossy-that are very important to coating applications. Trezza and Krochta (2000) found whey protein-glycerol films had gloss value 90.8. The gloss of WP films can be affected by plasticizer choice. Lee *et al*. (2002) found sucrose-plasticized WP coatings had the highest gloss compared to glycerol-or propylene glycol-plasticized coatings. They hypothesized that the Refractive Index (RI) of the plasticizer affected the gloss of the final film. Amorphous sucrose has an RI higher than other commonly used plasticizers. The amount of light a surface reflects is related to the RI-the higher the RI, the more the light is reflected. Dangaran and Krochta (2003) showed that as sucrose content increased, the gloss of WP films and coatings significantly increased. However, crystallization of the sucrose that occurred over time gave the WP films a hazy appearance and lowered the gloss. To be acceptable coatings, crystallization of the plasticizer needed to be controlled. Dangaran and Krochta (2006a) found that sucrose crystallization in WP films could be hindered by the addition of inhibitors. Raffinose and modified starch prevented crystal growth in whey protein-sucrose films for at least 1,800 h of storage at 53% relative humidity. WP films without inhibitors had noticeable crystallization after 50 h of storage. When applied as coatings to chocolates for a glossy finish, the whey protein-sucrose coatings with raffinose inhibitors maintained gloss longer than WP coatings containing only sucrose (Dangaran and Krochta 2006b).

Applications of whey coatings: Based on their inherent properties, some specific applications of WP films formed as coatings have been researched and developed. By taking advantage of the passive gas barrier, glossy appearance properties or active film capabilities, WP films and coatings have been designed to be coatings that lengthen shelf life, improve consumer acceptability, or raise the level of food safety for products like nuts, eggs, confectionary, meats and fruits and vegetables.

Since nuts and peanuts are susceptible to lipid oxidation and quickly oxidized when exposed to oxygen and forms rancid off-notes, which make the product unacceptable to consumers and shortens shelf life, an excellent oxygen-barrier coating is needed. A WP coating can apply directly to the nut surface and allow the protective layer to remain with the food, also reducing the high-performance barrier requirement for the outside product packaging. In a study by Mate *et al.* (1996), peanuts coated with WP isolate had lower peroxide and hexanal formation during storage as compared to uncoated peanuts. Krochta (2002) also found that the WP coatings could extend the shelf life of peanuts to 273 days at 25°C compared to 136 days for uncoated nuts. Lin and Krochta (2003) found that incorporation of surfactant in the WP coating significantly increased its coating efficiency.

Whey films also improve shelf life of eggs by reducing loss of water and carbon dioxide through the shell during storage. In a study by Caner (2005), the shelf life of whey protein-coated eggs was 1 week longer and color, yolk index (yolk height and yolk width) and pH changed slower and remained at higher quality levels than uncoated eggs.

As edible films are so glossy and transparent, they can be used to impart a smooth and glossy appearance in finished dried or confectionery products. Lee *et al.* (2002) applied the WPI films as coatings to panned chocolate candies and found sucrose-plasticized WPI coatings to be the glossiest. However, in a consumer study, the lower level of WPI gloss coatings was preferred overall.

WP films incorporating organic acids and various bioactive peptides have been tested against both spoilage and pathogenic organisms. WP films containing antimicrobials provide another layer of protection while potentially reducing the amount of antimicrobials needed for efficacy. The two issues for active edible films containing antimicrobials are the minimum inhibitory concentrations against different levels of contamination from specific microorganisms and diffusion constants of the antimicrobials in the film and in the food. Both have been investigated for WP films. Perhaps the greatest potential for active edible films concerns food safety. Chemical antimicrobial compounds have been and continue to be used. However, there is a growing concern about the use of synthetic pesticides and chemicals with foods (Sloan, 2001). Natural antimicrobials have been researched as effective and socially acceptable alternatives. The bioactive proteins lysozyme, lactoferrin and lactoperoxidase have been extensively investigated as antimicrobials in WP films. As an added layer of protection, WP films can be used in ready-to-eat meat products such as roasted turkey, smoked salmon and sausage products. These products are susceptible to contamination during slicing and packaging (Cagri *et al.*, 2002; Min *et al.*, 2005a). WP coatings with active antimicrobials have been shown to be effective at inhibiting growth of *Escherichia coli* O157: H7, *Salmonella enterica* or *Listeria monocytogenes*, thus increasing food safety and extending product shelf life (Min *et al.*, 2005a,b). Enzymes may be also used for changes in functionality and flavour modification (Jooyandeh *et al.*, 2009a).

Nowadays Fresh cut fruits and vegetables are growing in popularity; however, once cut, the produce becomes highly perishable. The respiration rate of fresh cut fruits can be 1.2-7 times higher than unprocessed fruit, according to Lee *et al.* (2002). Modified atmosphere packaging could slow oxidation, but if oxygen levels are reduced too low, anaerobic conditions could be created and this creates the risk of anaerobic bacterial growth. Edible coatings that have moderate oxygen, carbon dioxide and water vapor permeability can be applied to the surface of fresh cut product to extend shelf life by delaying ripening and browning, reducing water and aroma loss, carrying antioxidants, or/and carrying texture enhancer (Olivas and Barbosa-Canovas, 2005).

Because of their inherent characteristics, WP films are excellent oxygen, aroma and oil barriers without adjustment. They can be passive barriers and add a layer of protection to foods by being incorporated into the product as a film layer or a coating. They serve parallel functions to traditional packaging materials. They can be applied to traditional packaging materials like paper and plastic films to impart a new functional property. Paper is the most widely used packaging material because of its versatility, printability and easy recyclability. However, since it is made from cellulose, which is hydrophilic, paper is a poor water vapor barrier. Moreover, paper loses its strength and integrity when wet. It is often coated with wax or polyethylene to improve the moisture-barrier properties. A next step in both edible film and traditional packaging technology is the incorporation of functional compounds that confer another protective action to the system creating what is known as active packaging. By definition, active packaging interacts directly with the food or headspace of the product (Han, 2000). In traditional packaging systems, the active compounds may be toxic and therefore cannot touch the food directly. To prevent contamination, the active compounds may be incorporated into complex multilayer packaging. As stated previously, layered packaging is difficult to recycle and most often ends up as waste in landfills. Edible films and coatings can also be active layers, but have the benefit of being nontoxic concerning contact with food. WP films can carry such bioactive compounds as flavors, natural oxygen scavengers and antimicrobials without the concern of toxicity.

Future trends: Perhaps the biggest story in the dairy industry in the past couple of decades has been the rise of new applications for whey and whey proteins. Once considered a waste product in the cheese manufacturing process, whey and WP products today are used for a wide range of functional and nutritional properties (Jooyandeh, 2009). Applications of WPs in bread (Jooyandeh *et al.*, 2009b) and cheese (Jooyandeh and Minhas, 2009) to enhance product quality are examples of these properties. An active area of current researches is utilizing WPs to formulate edible and biodegradable films. There is an increasing need for packaging materials that are alternatives to petroleum-based sources. As oil prices continue to go up, so do packaging costs. Renewable sources of materials for packaging will create a steady, reliable supply. Edible films from WP are "green" alternatives to traditional plastics. Based on its excellent oxygen-barrier properties, WP films can be competitive biodegradable materials replacing EVOH, nylon, or polyesters, which are typically used as oxygen barriers. Biodegradable packaging is estimated to grow 20% over the next few years, taking up a larger share of the packaging market. Protein-based films and coatings offer alternative properties to the carbohydrate-based packaging materials that have already been successfully accepted into the market. Moreover, proteins provide more opportunities for change in chemical structure and thus, future property improvement than carbohydrates. More and more companies are seeking out biodegradable agriculturally based packaging and seeing where it can fit into their packaging needs. The future of WP films depends to a great extent on the consumer's demand and his knowledge.

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