Physical and Mechanical Properties of Pea-Protein-based Edible Films

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ABSTRACT: Edible films produced from denatured pea protein concentrate (PPC) solution possessed the strength and elasticity to resist handling. Increasing the concentration of the plasticizer (glycerol) in the film decreased tensile strength and elastic modulus, and increased elongation and water vapor permeability (WVP). Very strong and stretchable films were obtained from 70/30 and 60/40 of PPC/glycerol composition, respectively. The low WVP value was maintained over a range of glycerol concentration from 20% to 40%, in the dry film. Film solubility was not affected significantly by the amount of the plasticizer. The physical and mechanical properties of the PPC films were comparable with those of soy protein and whey protein films.

Key words: yellow pea (*Pisum sativum* L. Miranda), pea protein concentrate, edible film, tensile property, water vapor permeability

Introduction

PEAS HAVE RECEIVED SPECIAL ATTENTION FOR THE UTILIZATION of legume protein because they are already consumed as a human dietary resource, have a high nutritional value and possess various functionalities. Increased legume protein utilization by the food industry has led to research on the functionality of legume and seed protein in foods (Kinsella 1979; Won and others 2000; Kim and others 1990). Although soybean products are still dominant, efforts have been made to explore the potential uses of other legumes for their functional properties including field peas, horsebeans, faba beans, Great Northern beans, chick-peas and cowpeas (Vose 1980; Sosulski and McCurdy 1987; Sathe and others 1981). Currently, the protein sources of edible films and coatings include a variety of protein-rich legumes. For example, peanut protein has recently been studied for use in edible film materials. The mechanical and physical properties of the peanut protein films were determined by Jangchud and Chinnan (1999).

Peas, generally various cultivars of *Pisum sativum* L., belong to the family *Leguminosae* (Pomeranz 1985). Dried Peas are generally composed of carbohydrates (35 %), proteins (27 %), fibers (27 %) and a very small amount of lipids (Boulter 1983). Compared to other pulse products, peas are high in carbohydrates and fibers, and low in lipids. While the fiber and starch fraction of peas are highly utilized by bakery and meat industry, the protein fraction has been underutilized with only a limited use as animal feed (Betker 1990). Pea protein has experimentally been used as a meat extender in sausage and as a protein fortifier for bread (Delaquis 1983; Grant 1983). Pea protein consists primarily of globulins mainly and a small fraction of albumins. The globulins (> 80% of total proteins) are storage proteins located in the cotyledons and consist of legumin, vicilin and convicilin (Pate 1977; Boulter 1983). Legumin is usually the major, and vicilin is the second major globulin fraction (Gatehouse and others 1981). Albumins, which compose 13 – 14% of the total proteins, are cytoplasmic proteins consisting of many kinds of subunits and contain more sulfur amino acid residues than the globulins (Mosse and Pernollet 1983; Pate 1977; Grant and others 1976).

Pea protein concentrate and isolate possess good nutritional quality (that is, protein efficiency ratio, essential amino acid content) and has potential as a dietary protein fortifier (Linder 1985). However, the relatively poor functionality has been reported for protein solubility, emulsion stability and gelation property (Jackman and Yada 1989; Patel and others 1981; Sumner and others 1981; Johnson and Brekke 1983; Naczk and others 1986; Klein and Raidl 1986). To increase the utilization of pea protein concentrate or isolate, the functional properties of pea protein have to be developed and modified to meet the requirements for use as a functional ingredient, and other applications need to be explored.

Edible polymer films have been studied for food applications because of their potential for providing a barrier against hazards, carrying active ingredients, and improving the mechanical integrity of coated foods (Perez-Gago and others 1999). Edible films can also reduce and simplify the packaging materials required for the protection of food products (Han and Krochta 1999; Perez-Gago and others 1999). Edible films and coatings afford numerous advantages over conventional nonedible polymeric packaging because of their biodegradability, and have been used to protect pharmaceutical ingredients and extend the shelf-life of food products (Gontard and others 1993). Soy protein, whey protein, wheat glutens, corn zein protein, collagen and gelatin have been studied intensively for edible film applications.

Compared to the price of whey protein isolate ($13.5 to 27/Kg), soy protein isolate ($3 to 3.8/Kg) and corn zein ($23 to 35/Kg) (Krochta and De Mulder-Johnston 1997), pea protein concentrate ($2.5 to 2.8/Kg) provides an excellent opportunity for use of pea protein in food and other industries.

Like most synthetic plastic polymers, edible coatings and film materials require property modifiers to improve the physical and mechanical properties of the film and to prevent chemical changes. As with synthetic plastics, plasticizers are incorporated into the edible coating/film materials, which overcomes the brittleness caused by extensive intermolecular forces. The plasticizers reduced these forces and increase the flexibility and extensibility of the film (Guilbert and Biquet 1989). On the other hand, the addition of plasticizers generally increases the permeability of both plastic

and edible films to gas, water vapor and solute (Krochta and De Mulder-Johnston 1997; Han and Krochta 1999) and can decrease elasticity and cohesion (Delporte 1981). The most common edible plasticizers are polyols, mono/di- or oligosaccharides, lipids, and their derivatives.

The objectives of this research are: (1) to produce edible protein films for food coating and packaging from pea protein concentrate; and (2) to determine their tensile properties, water solubility and water vapor permeability and to evaluate the potential of pea protein for use as a coating/film material. The ultimate goal of this study is to simplify the total packaging structure by substituting synthetic packaging materials with a pea protein coating/film to reduce packaging requirements, cost and contribution to solid wastes.

Materials and Methods

Materials

Pea protein concentrate (PPC) extracted from yellow field peas (*Pisum sativum* L. Miranda), was supplied by Parrheim Foods Co. (Portage-La-Prairie, MB, Canada) as a liquid. This PPC was produced by acidified protein curds from the soluble fractions of ground yellow peas. The PPC contained 83% protein, 3% fat, 4% ash, and 10% non-protein soluble fractions by total solid content. The pH of the liquid PPC was increased to 9 by the addition of 2N NaOH solution which increased the solubility of the PPC. The PPC solution was freeze-dried and stored at 4 °C. Glycerol (Sigma Chemical Co., St. Louis, MO) was used as a plasticizer. The pH-adjusted liquid PPC are generally spray-dried for the commercial products. However, to maintain the high water-solubility of PPC powder, we used a lab-scale freeze-dryer instead of a commercial-scale spray-dryer.

Film Preparation

Aqueous solutions of 10%(w/w) PPC were prepared and glycerol (Gly) was added at a ratio of 80/20 to 50/50 of PPC/Gly. The film-forming solutions were degassed under vacuum and heat-denatured at 90 °C for 25min in a water bath. The heat-denatured solutions were cooled to room temperature. Before film casting the solution were degassed under vacuum again to remove any dissolved air. Native film solutions were prepared by the same process with the exception of heat denaturating treatments.

Edible PPC films were cast by transferring 5 - 6g of the film-forming solution onto poly styrene petri-dishes (10cm dia) resting on a well-leveled wood slab. The film-forming solution was allowed to dry at room temperature over 24h. The films were then peeled off from the casting dishes and their thickness were measured with a caliper micrometer (B.C. Ames Co., Waltham, MA) at 5 random positions of the film. The average thickness was used for the proceeding mechanical test and water vapor permeability determination.

Tensile Test

Film specimens, 1 cm wide and 8 cm long, were cut after conditioning in a desiccator for 3 days. Tensile strength (TS), elongation-at-break (E) and elastic modulus were determined from a stress-strain curve using texture analyzing instrument (Texture Analyser, TA-XT2, Texture Technologies, Corp., NY) following the procedure outlined in ASTM method D882-91 (ASTM, 1991). Initial grip distance and cross-head speed were 5 cm and 100 mm/min, respectively. Tensile strength (TS) was calculated by dividing the peak load by the cross-sectional area (thickness $\times$ 1 cm) of the initial specimen. Elongation was expressed as the percentage of change in the length of the specimen to the original length between the grip (5 cm). Each tensile strength (TS), elongation-at-break (E) and elastic modulus was obtained from 6 replications of samples taken from the same film.

Water Vapor Permeability (WVP)

The modified ASTM E96-92 gravimetric method of McHugh and others (1993) was used to determine the WVP and relative humidity (RH) at the film underside. Distilled water (6mL) was dispensed into flat-bottom acrylic (Plexiglas®) cups with wide rims. The cup was covered with a film, sealed with a ring and silicon sealant (High Vacuum Grease, Dow Corning, Midland, MI), and placed in a desiccator cabinet containing fans to be held at 0% RH using anhydrous calcium sulfate (W.A. Hammond Drierite Co., Xenia, OH) at 25 °C. Weight changes were taken periodically after the steady state of weight loss was achieved and used to calculate the %RH at the film underside and the resulting water vapor transmission rate (WVTR). The WVP was calculated by multiplying WVTR by the thickness of the film.

Soluble Protein Content (%SP) and Total Soluble Matter (%SM)

Film specimens (1.5 cm $\times$ 1.5 cm) were dried for 3 days in a desiccator over anhydrous calcium sulfate. The specimens (approximately 15 mg) were accurately weighed and immersed in test tubes containing 5 mL distilled water (DW). The tubes were covered with aluminum foil and incubated at ambient temperature for 24 h with occasional gentle agitation. After dissolving water-soluble components which may include soluble proteins, soluble carbohydrates and other soluble ingredients, the soluble protein contents (%SP) of the films in the DW were determined by measuring absorbance at 280 nm using a spectrophotometer (Ultrospec2000, Biochrom Ltd., Cambridge, England) and quantified with a standard concentration curve ($R^2 = 0.9933$) constructed from pea protein isolate.

The remaining pieces of film after soluble protein tests were dried in the oven at 70 °C for 24h to obtain the final dry weight of the film (total solid matter). The percentage of total soluble matter (%SM) of the films was calculated using the following equation:

$$\%SM = [(\text{Initial dry wt} - \text{Final dry wt}) / \text{Initial dry wt}] \times 100$$

Results and Discussions

Film Formation

After drying the film-forming solutions, free peeled transparent films were obtained. All film forming processes were conducted under the %RH lower than 20. The films were slightly yellowish, but the color was negligible due to their transparency. The films were strong and flexible enough to be handled, except for the 80/20 (PPC/Gly) films which were very brittle and fragile. This brittleness was increased by the lower humidity level. Because the physical and mechanical properties of PPC films are related to the storage humidity, like most other protein films, the film specimens had been stored in a desiccator until examinations to maintain the same dry condition. It is also recommended to store PPC film specimens at constant humidity in order to compare the physical and mechanical characteristics between different films.

Tensile Properties

Table 1 shows the tensile properties of heat-denatured

and native PPC films. Increasing the concentration of a plasticizer (glycerol) decreased tensile strength (TS) and elastic modulus, while increasing the elongation-at-break (E), for native PPC films (Table 1b). Like most other protein films, the addition of glycerol made the film more ductile, which indicates that glycerol takes a place between protein molecules and interferes with the intra-molecular forces. In the case of heat-denatured films, the 80/20 film was so brittle that the TS of the film was weaker than that of a 70/30 film. The 70/30 film showed the largest value of TS among tested specimens. In terms of elongation properties, the 60/40 film was the most stretchable of both heat-denatured and native films. No data for the tensile test was obtained for the native 80/20 film due to its extreme brittleness.

With the same composition of PPC/Gly in the film, denatured PPC films were stiffer, stronger and more extensible than native films due to the formation of intra-molecular chemical bonds between protein molecules after denaturation. Heat-denaturation of film-forming solutions affects remarkably the physical properties of wheat gluten films (Rangavajhyala and others 1997). Films prepared from heat-denatured soy protein solutions had lower WVP than those prepared from unheated film-forming solution (Stuchell and Krochta 1994). Heating soy protein solution at 80 °C or 95 °C for various period of time results in the films with increased TS, more dark yellow color and lowered E and decreased WVP values (Gennadios and others 1996). As with other edible protein films, heat-denaturation at 90 °C for 25 min enhances the strength of PPC edible films.

Table 1. Tensile properties of pea protein concentrate films.

a. Heat-denatured film.

PPC/Gly	TS (MPa)	Modulus (MPa)	E (%)
50/50	0.692 ± 0.073	3.32 ± 0.53	92.0 ± 21.5
60/40	2.896 ± 0.244	8.15 ± 2.35	147.9 ± 13.6
70/30	7.317 ± 0.436	204.86 ± 66.48	46.8 ± 5.8
80/20	4.938 ± 1.024	49,749 ± 21,916	0.6 ± 0.2

b. Native film

PPC/Gly	TS (MPa)	Modulus (MPa)	E (%)
50/50	0.317 ± 0.024	1.39 ± 0.29	95.3 ± 20.3
60/40	1.105 ± 0.092	7.44 ± 1.79	93.7 ± 11.2
70/30	4.109 ± 1.732	1,015.2 ± 187.4	7.6 ± 3.0
80/20	ND*	ND	ND

* Not Detectable

Table 2. Water vapor permeability (WVP) of heat-denatured pea protein concentrate films at 25°C.

PPC/Gly	Thickness (mm)	WVP (g mm m^{-2} hr^{-1} kPa^{-1})	RH (%)
50/50	5.83 ± 0.85	7.42 ± 0.69	64.52 ± 1.92
60/40	4.45 ± 0.60	4.98 ± 0.50	67.48 ± 1.42
70/30	4.65 ± 0.98	4.10 ± 0.58	72.55 ± 1.57
80/20	5.60 ± 0.14	4.30 ± 1.16	75.70 ± 4.72

Table 3. Protein solubility (%SP) and total soluble matter (%SM) of heat-denatured pea protein concentrate (PPC) film with different levels of glycerol (Gly).

PPC/Gly	Protein solubility (%SP)	Total soluble matter (%SM)
50/50	37.8 ± 4.5	43.5 ± 4.0
60/40	34.1 ± 3.7	41.3 ± 7.9
70/30	32.2 ± 9.8	38.7 ± 4.0
80/20	33.8 ± 9.1	44.6 ± 7.0

Water Vapor Permeability

The water vapor permeability of PPC film was not affected by the amount of plasticizer (Gly) added to the dry film over the range of 20% to 40% (Table 2). Only the 50/50 film has a significantly higher value for WVP compared to the other PPC/Gly combinations. The independence of WVP values on the amount of plasticizer, within the range of 20% to 40% of glycerol, may be of value in applying PPC/Gly film for use with food coating or packaging applications because the film flexibility can be modified by addition of a plasticizer, without significant changes in the moisture barrier properties. However, the 80/20 PPC film which contains less plasticizer possesses a similar WVP value to that of the 70/30 film in spite of the more humid testing condition (higher %RH) at the underside of the film. Therefore, it can be assumed that with the same conditions of relative humidity, the 80/20 film may have a lower WVP value than that of the 70/30 film, as the WVP of edible films generally increases with increasing RH. Like other edible films, increasing the amount of the plasticizer in the film reduces the moisture barrier properties of the PPC film. Overall, the WVP of the PPC film is affected by the relative humidity and the amount of plasticizers.

Film Protein Solubility (%SP) and Total Soluble Matter (%SM)

Heat-denatured PPC films maintain their integrity during the film soaking treatment. No difference was found in protein concentration (%SP) and total soluble matter (%SM) of the tested specimens over the range of glycerol concentration from 20% to 50% (Table 3). The soaking water also contained soluble pea protein concentrate fractions, which are non-protein soluble fractions such as soluble fibers and sugars. The %SP and %SM of the PPC films were approximately 20% higher than those of whey protein isolate films with the same compositions of protein/glycerol, when they were compared to the data of Perez-Gago and others (1999). This indicates that the PPC film has weaker intra-molecular interactions in the aqueous condition compared to those between whey protein (mainly β–lactoglobulin) molecules in heat denatured whey protein isolate films. Table 4 compares the physical and mechanical properties of pea protein concentrate film, soy protein isolate film and whey protein isolate film. The properties of the PPC film were comparable with those of other protein films. Therefore, the pea protein concentrate may be utilized as a replacement for soy protein and whey protein in forming food coatings or edible films. The relatively high solubility of pea protein films in water, especially native PPC films, could possibly make the films appropriate for hot water soluble pouches, like the cellulose ether-based soluble pouch which is commercially available currently (Kunte and others 1997).

Pea protein isolate and concentrate require a heating period of 45 min to 60 min at 90 °C to form a strong gel (Fleming and Sosulski 1975; Betker 1990). A differential scanning calorimetry study by Murray and others (1985) indicated that the denaturation temperature of pea protein isolate was ranged from 88.9 °C to 94.5 °C. The gelation properties of the pea protein suggest that the heating time and temperature distribution of the denaturation process of the PPC could be important in determining the physical and mechanical properties of the PPC films. On the contrary, our preliminary experiments resulted in identical film strengths with both 10 min and 50 min heat-denatured specimens at 90 °C, while the previous studies revealed that over 45 min was required to complete the gelation process at the same temperature. More studies of the conditions and mechanism

Table 4. Physical and mechanical properties of pea protein concentrate, soy protein isolate and whey protein isolate films.

	Pea protein[1)]	Soy protein[2)]	Whey protein[3)]
Tensile strength (MPa)	7.3 ± 0.4	8.5 ± 0.5	6.9
Elastic modulus (MPa)	204.9 ± 66.5		199
Elongation (%)	46.8 ± 5.8	31.9 ± 2.4	41
WVP[5)] (% RH)	4.1 ± 0.6 (72.6 ± 1.6)	3.76 ± 0.16 (73.26)	4.96 (71)[3)], 4.75 (77)[4)]
Protein solubility (%)	32.2 ± 9.8	6.5 ± 0.3	≈10
Total soluble matter (%)	38.7 ± 4.0	35.1 ± 1.0	≈30

[1)]pH 9, 10% PPC, 70/30 (PPC/Gly)
[2)]pH 10, 5% SPI, 10/3 (SPI/Gly); Kunte and others (1997)
[3)]pH 7, 5% WPI, 70/30 (WPI/Gly); Perez-Gago and others (1999)
[4)]pH 7, 10% WPI, 70/30 (WPI/Gly); Perez-Gago and others (1999)
[5)]Unit of WVP is in (g mm m^{-2} hr^{-1} kPa^{-1})

of PPC denaturation are suggested to develop a clearer model of the film forming process.

Nicols and Cheryan (1982) suggested that blending with dairy proteins, such as casein and whey protein, may improve the functionality of relatively poor functional proteins. The functionalities of whey protein and pea flour blends were examined and potential uses for the blends were suggested by Patel and others (1981) including pasta, meat patties, egg white substitutes, milk substitute and bakery goods. Therefore, blended protein films as well as the denaturation characteristics of PPC films are appropriate subjects for future studies on edible protein films.

Conclusion

POTENTIAL APPLICATION OF EDIBLE PROTEIN FILM MADE BY pea protein concentrate was suggested. The pea protein concentrate film was strong and elastic, and possessed a good moisture barrier property and physical integrity. Addition of glycerol plasticized the tough and brittle film characteristics resulting in flexible and stretchable. The characteristics of the pea protein-based edible films were comparable with other edible protein films, such as soy protein and whey protein, in terms of the tensile strength, elongation, moisture barrier property and water solubility. The new film products may exploit the utilization of pea protein.

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