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Barrier and Mechanical Properties of Milk Protein-Based Edible Films Containing Nutraceuticals

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Calcium caseinate (CC) and whey protein isolate (WPI) films were prepared to contain 5 or 10% Gluconal Cal (GC), a mixture of calcium lactate and gluconate, or 0.1 or 0.2% α-tocopheryl acetate (VE), respectively. The pH and viscosity of film-forming solutions and the water vapor permeability and tensile property of the films were determined using standard procedures. CC and WPI films have the capabilities to carry high concentration of GC or VE, but some of the film functionality might be compromised. Adding VE to CC and WPI films increased film elongation at break, whereas incorporating 0.2% VE decreased WVP of CC films and tensile strength of both CC and WPI films. Incorporation of GC reduced the tensile strength of CC films ($P < 0.05$), with 10% GC decreasing both elongation at break and WVP ($P < 0.05$). These types of films may be used for wrapping or coating to enhance the nutritional value of foods. The concentration of GC and VE added to the films must be carefully selected to meet required water barrier and mechanical properties of the films depending on their specific applications.

KEYWORDS: Milk protein films; calcium; vitamin E; water vapor permeability; tensile properties

INTRODUCTION

Milk proteins have become excellent materials to form edible films because of their high nutritional value and numerous functional properties that are important for the film formation (*1*−*4*). Milk protein-based films have been claimed as effective carriers of many functional ingredients, such as antimicrobial agents to improve safety and stability of foods (*5*), antioxidants to prevent lipid oxidation, and flavorings and pigments to improve quality of food (*6*, *7*). The concepts of incorporating nutraceuticals into edible coatings and films to enhance nutritional value of foods have also been discussed, but few studies have been reported.

Nutraceuticals are chemicals found as natural components of foods or other ingestible forms that have been determined to be beneficial to the human body in preventing or treating one or more diseases or improving physiological performance. Calcium and vitamin E are important nutraceuticals as they play significant roles in the human body to prevent certain diseases (*8*, *9*). For adults, dairy products supply 72% of the calcium in the U.S. diet, grain products ∼11%, and vegetables and fruits ∼6% (*10*). With the change of dietary habit, it is difficult for those individuals whose staple food is vegetables and fruits to ingest enough calcium. Many of them have turned to dietary supplements to meet their needs. The design of edible films or coatings containing high concentrations of calcium and/or vitamin E for wrapping or coating foods would provide opportunities to increase the intake of these nutraceuticals, thus helpting to satisfay the dietary needs of these nutrients. Mei et al. (*11*) reported that when fresh peeled baby carrots were coated with xanthan gum coatings containing calcium or vitamin E, a serving (=85 g) of carrots could offer ∼6.6% of the Dietary Reference Intake (DRI) of calcium and ∼70% of the DRI of vitamin E based on the DRI values of 1000 mg/day for calcium and 15 mg/day for vitamin E.

The intended use of edible coatings or films would require a clear understanding on their moisture barrier and mechanical properties. These properties depend strongly on film composition, its formation, and the methods of application to the products (*12*−*16*). When calcium is added to protein-based coatings and films, the calcium ions may induce static cross-linking and the level of ionic strength may affect protein microstructure and interactions, which would in turn impact the mechanical and barrier properties. In a few previous studies low concentrations of calcium (calcium chloride, sulfate, or ascorbate) were added to film solutions for improving film barrier and mechanical properties (*6*, *17*−*19*). However, the results from these studies were very inconsistent. In addition, these calcium salts have no important nutritional function nor do they provide a health benefit to the human body (*20*). Calcium has been one of the most difficult minerals to fortify because of its high DRI value, its low solubility at neutral pH, and the bitter taste of some calcium salts. Mei et al. (*11*) identified that Gluconal Cal (Glucona America Inc., Janesville, WI), a mixture of calcium lactate and calcium gluconate, has high nutritional value, good bioavailablity and water solubility, and neutral taste and

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successfully incorporated it into edible coatings. However, its influence on the film functionality was unknown.

The performance of vitamin E in edible coatings or films has not been evaluated. Lipid was added to assist the carrying of vitamin E in coating and films because of the fat-soluble nature of vitamin E. The addition of lipids may affect the functionality of the coating and films because of their hydrophobic nature.

Our objectives in this study were to formulate calcium caseinate and whey protein isolate based film solutions that can effectively carry high concentrations of calcium and vitamin E, to develop procedures for making films from these solutions, and to investigate the impacts of calcium and vitamin E on the pH and viscosity of film solutions and the water vapor permeability (WVP) and tensile properties of the films.

MATERIALS AND METHODS

Materials. The materials used for film-forming solution include calcium caseinate (CC; Alanate 380, NZMP, Santa Rosa, CA) with 92.4% protein and 1.4% calcium; whey protein isolate (WPI; Bipro, Davisco, Le Sueur, MN) with 93.6% protein; Gluconal Cal (GC; Glucona America Inc.), a mixture of calcium lactate and calcium gluconate with a water solubility up to 40 g/100 mL and neutral taste; α-tocopheryl acetate (VE; Sigma, St. Louis, MO), a very stable form of vitamin E that exhibits vitamin E activity and in vivo antioxidant effects as the result of enzymatic cleavage of the acetate ester; glycerol (GLY; Fisher Scientific Inc., Fair Lawn, NJ), serving as plasticizer in all film-forming solutions; and acetylated monoglyceride (AM; Danisco, New Century, KS), acting as lipid to incorporate vitamin E into the film-forming solutions.

Film Formation. A complete 2 × 5 factorial design was deployed in the manufacture of films. The experiment included two proteins (CC and WPI) and five nutraceutical incorporations (0 as control, 5% GC, 10% GC, 0.1% VE, and 0.2% VE). Aqueous solutions of 10% (w/w) CC or WPI were made according to the method of Banerjee and Chen (*21*). A measured quantity of CC or WPI was added into an ice−water (1:1, w/w) mixture that had been previously ground for 3 s in a blender (model 31BL41, Waring Products Division, New Hartford, CT). The new mixture was ground for 15 s, then manually mixed for 30 s, and ground for another 30 s in the same blender. Glycerol was added at 1:2 (w/w; GLY/protein) as a plasticizer.

CC/VE or WPI/VE film-forming solutions were prepared by first dissolving 0.1 or 0.2% (w/w of solution) VE into melted AM at a ratio of 1:4 (w/w) and then adding dissolved VE into CC and WPI solutions that were previously heated to 60 °C in a shaking water bath (Precision Scientific, Winchester, VA). The solutions were then homogenized using a homogenizer (Brinkman, model PT10/35, Westbury, NY) for 1 min at the speed setting of 5 before making films. CC/GC or WPI/GC solutions were prepared by directly dissolving 5 or 10% (w/w of solution) GC into CC or WPI solutions that had been kept in a refrigerator overnight.

pH and Viscosity of Film Solutions. The pH of the film solutions at room temperature was measured using a pH-meter (model IQ240, IQ Scientific Instruments, Inc., San Diego, CA). The viscosity was determined using a Brookfield digital rheometer (model DV-III+, Brookfield Engineering Laboratories, Middleboro, MA) with a spindle set at RV5 and a rotation speed set at 110 rpm. Twenty-five milliliters of solution at room temperature immediately after incorporating GC or VE was used for the measurement. For CC + GC solutions, two sets of solutions were used: (1) solution immediately after the addition of GC at room temperature; and (2) solution stored at 10 °C overnight before GC or VE was incorporated and then recovered to room temperature for viscosity measurement. The percent torque and viscosity (equal to shear stress divided by shear rate with a unit of centipoises, cP) were recorded.

Film Casting. Film-forming solutions were cast on disposable polystyrene Petri dishes (Krackeler Scientific, Albany, NY) of 9.1 or 14.4 cm diameter. The small-diameter films were used for the WVP measurement and large ones for mechanical properties. For minimizing variation of film thickness, a pipet was used to carefully drip a controlled amount of solution on each dish. The weight of solution in each dish was maintained at 1.2 ± 0.05 g for 9.1 mm diameter dishes and at 2.7 ± 0.05 g for 14.4 mm diameter dishes using an electronic balance (Denver Instrument, model XP-1500, Arvada, CO). The dishes were placed on a leveled plate to achieve uniform films. Cast solutions were allowed to dry for 24−48 h at room conditions of 23 ± 2 °C and 40 ± 3% relative humidity and then peeled intact from the dishes for further tests.

Films used for each property measurement were manufactured and stored under the same temperature and relative humidity conditions, but at different times. Despite careful control during film manufacture, it appeared that the film thickness might vary. The potential influence of film thickness variation on the determination of WVP and tensile strength is discussed under Results and Discussion.

Film Thickness and Moisture Content (MC). The thickness of the film was measured using a caliper micrometer (no. 293-766-30, Mitutoyo Manufacturing Co. Ltd.) at eight random positions on each type of test film. The average value was taken to calculate the WVP and tensile properties.

The moisture content of each film was determined according to the method of Anker et al. (*22*). Small test specimens were cut, placed into aluminum Petri dishes, and then dried inside a laboratory oven (Single-Wall, Blue M) at 105 ± 2 °C for 24 h. After drying, Petri dishes were placed in a desiccator to cool at room condition. Weight was taken before and after drying using a balance with an accuracy of 0.0001 g (Denver Instrument, model TL-204, Arvada, CO). The moisture content was calculated as

$$\mathrm{MC} = \frac{M_i - M_f}{M_i - M_d} \times 100\%$$

where M_i is the weight of Petri dish and film specimen before drying, M_d is the weight of Petri dish, and M_f is the weight of Petri dish and film specimen after drying. Eight replicates of each film type were measured for moisture content, and the average is reported.

Water Vapor Permeability. Films used for WVP measurement were first conditioned at 53% relative humidity inside a desiccator containing a saturated magnesium nitrate (Fisher Scientific Inc.) solution at room temperature (23 ± 2 °C) for >48 h. A modification of the ASTM E96-92 (*23*) gravimetric method was used to measure the WVP of the films (*21*, *24*). Twenty milliliters of distilled deionized water was placed in the test cups made of Plexiglas and having an inside diameter of 5.6 cm and inner depth of 1.5 cm. Films were sealed onto the cup base with silicon sealant (high-vacuum grease, Dow Corning, Midland, MI) and a ring with four screws symmetrically located around the circumference. To mount the composite films in the same orientation, the smooth and shiny surface of films faced down. The mounted films were then put into a desiccating cabinet (Fisher Scientific Inc.) with an installed motor, speed controller, and fan (*24*). Saturated magnesium nitrate and sodium bromide solutions (1:1) were placed at the bottom of the cabinet to adjust the relative humidity to 55 ± 3%. Weights of the cups were recorded every 4 h at an accuracy of 0.0001 g to measure the moisture loss across the film. Changes in the weight of the cups over time were calculated by linear regression. Three replicates of each film type were tested, and the result was reported as the mean value in g·mm/(m^2·h·kP).

Tensile Properties Determination. Dried films were conditioned at 58% realtive humidity for one week inside a desiccator containing a saturated sodium bromide (Fisher Scientific Inc.) solution to stabilize and simplify handling and cutting of the films. Tested films were cut using a striking die (The Right Image, Sacramento, CA) with 15 mm width × 100 mm length and flaring to 25 mm × 25 mm square grips on each end. All film strips were equilibrated for 36 h at 53% relative humidity inside a desiccator at room temperature (23 ± 2 °C).

Tensile measurement was performed following ASTM method D882-91 (*25*). Film strip ends were mounted and clamped with rubber-lined aluminum grips on a Universal testing machine (Instron Corp., model 4301, Canton, MA) with a 500 kg load cell. The initial gauge length was set to 100 mm, and the speed of the crosshead was 50 mm/min. Tests were conducted at room conditions of 50 ± 5% relative humidity

Table 1. pH and Viscosity of WPI and CC Film-Forming Solutions[a]

film type[b]		temp		torque	viscosity	viscosity[c]
protein	nutrients	(°C)	pH	(%)	(cP)	(cP)
CC	0	22.9	6.41a	0.4a	14.5a	18.2a
	5% GC	22.7	6.02b	0.6a	21.8a	21.8a
	10% GC	23.1	5.97b	3.0b	109.1b	61.8b
	0.1% VE	22.8	6.36a	0.4a	14.5a	18.2a
	0.2% VE	22.8	6.37a	0.5a	18.2a	18.2a
WPI	0	22.8	7.41c	3.7b	134.5cb	
	5% GC	21.6	6.66c	>96c	>3000d	
	10% GC	21.6	6.40a	>96c	>3000d	
	0.1% VE	22.5	7.17d	3.8b	138.2c	
	0.2% VE	22.3	7.26d	4.0b	149.1c	

[a] Mean values with different letters in the same column differ ($P < 0.05$). [b] CC, calcium caseinate; WPI, whey protein isolate; GC, Gluconal Cal; VE, α-tocopheryl acetate. [c] Solutions were stored in a refrigerator (~10 °C) overnight before GC or VE was added.

and 23 ± 2 °C. Tensile properties were reported as maximum tensile strength (MPa) and elongation at break (%). Six replicates of each film type were tested, and an average value is reported.

Statistical Analysis. Data were determined by analysis of variance (ANOVA) using SAS (*26*). General linear model (GLM) procedures were performed ($P < 0.05$) for all treatments at different levels. Duncan's multiple-range test was used to determine any significant difference among all of the treatments at $P \leq 0.05$.

RESULTS AND DISCUSSION

Rheology Properties of Film Solutions and Film Formation Capability. The pH values of film solutions are reported in **Table 1**. Incorporation of GC reduced the pH value of both CC and WPI solutions due to the presence of lactic acids in GC ($P < 0.05$). Incorporation of VE slightly decreased the pH value of the WPI solution, but not that of the CC solution. The WPI solution had a higher pH value than the CC solution.

The WPI solution had ~9 times higher percent torque and viscosity than the CC solution (**Table 1**), probably due to the high protein concentration of WPI. Incorporation of GC increased the percent torque and viscosity in both WPI and CC solutions ($P < 0.05$), especially in the WPI solutions, where the percent torque and viscosity were >96% and >3000 cP, respectively. Changes in the rheology property of WPI and CC solutions might contribute to the gelation caused by the interactions between protein and calcium ions, typically in the case of high concentration of calcium added to the solutions. It is well-known that calcium enhances gel formation of proteins. VE incorporation did not affect the viscosity of both CC and WPI solutions. Measured rheology properties were consistent with the observations on the solutions. The CC solution became thicker immediately after the incorporation of calcium, but turned thinner, whiter, and opaque after manual stirring, whereas the WPI solution became very viscous and formed strong gels after the addition of 5 or 10% GC. It turned out that the CC solution was able to form films when high concentrations of GC or VE were incorporated, but the WPI solution could not form films when 5 or 10% GC was added.

The different film-forming capabilities of CC and WPI solutions may be explained by their different film-forming mechanisms. Caseinate film formation is extensive hydrogen bonding, electrostatic interaction, and hydrophobic bonding (*3*, *7*, *27*), whereas WPI film formation is protein penetration and exposure of internal sulfhydryl groups, promoting intermolecular disulfide bond formation after heat denaturation (*28*). Gel formation is generally affected by protein concentration, quantity and state of water, ionic type and strength, heating time and temperature, pH, and interactions with other components (*29*). WPI is a stronger ionic type protein than CC and thus has greater interaction with calcium cations, which leads to the formation of strong gel as reflected by the higher torque and viscosity values.

Storing CC solutions in a refrigeration temperature of ~10 °C before incorporation with calcium is an important step to prevent strong gel formation and obtain more uniform solutions. According to Damodaran (*30*), cooling the CC solution decreased the thermal kinetic energy and thus facilitated formation of stable non-covalent bonds among exposed functional groups of the various molecules and constituted reversible gels. The gelation may make many exposed functional groups less available to calcium cations, thus resulting in weak gels. This was confirmed by reduced viscosity in a CC + 10% GC solution previously subjected to cold storage (**Table 1**). However, the same pretreatment (cold storage) did not prohibit the strong gel formation of a WPI solution, because the ionic strength is much stronger than that of reversible non-covalent bonds.

Table 2. Moisture Content and Thickness of WPI and CC Films[a,b]

film type[c]			
protein	nutrients	moisture content (%)	thickness (mm)
CC	0	24.50d	0.1495d
	5% GC	27.57abcd	0.1402e
	10% GC	30.73a	0.1462de
	0.1% VE	27.24bcd	0.1904a
	0.2%VE	27.29abc	0.1860a
WPI	0	27.28bcd	0.1590c
	0.1% VE	28.91ab	0.1690b
	0.2% VE	25.07dc	0.1704b

[a] Mean values with different letters in the same column differ ($P < 0.05$). [b] As means of eight film samples from each film type. Standard deviation of the thickness ranged from 0.002 to 0.01 mm for each film type. [c] CC, calcium caseinate; WPI, whey protein isolate; GC, Gluconal Cal; VE, α-tocopheryl acetate.

Film Thickness and Moisture Content. The thickness of all the film types is reported in **Table 2**. The simple CC film was thinner than the simple WPI film because the cross-linking of the caseinate caused by calcium might tighten the molecular structure of the calcium caseinate (*17*). The thickness of films containing VE was significantly greater than those of simple CC films and WPI and CC films containing GC due to the lower density of VE and AM (*21*). The concentration of GC and VE did not affect the film thickness.

Among the simple films, CC film had a lower MC than WPI film, which is the same as previous findings (*17*, *21*). It was suggested that ionic cross-linking in the simple CC film reduced protein polymer segmental mobility (*17*), resulting in lower MC. Adding GC or VE into film-forming solutions did not significantly change the film MC except for CC + 10% GC and WPI + 0.2% VE films (**Table 2**). The MC of the CC + 10% GC film was significantly higher, whereas that of the WPI + 0.2% VE film was significantly lower than those of other films. Calcium salt is a humectant and could absorb and trap moisture inside (*31*), thus increasing the film MC when added in a high concentration. VE and AM usually stayed in the upper part of the film as a result of film orientation, thus preventing moisture escaping from the film. Addition of lipid to the film-forming solutions made the films more hydrophobic; therefore, the films containing VE and AM should contain less moisture (*21*). In this study, the impacts of GC and VE on the film MC occurred only when they were added in high concentrations (10% GC or 0.2% VE).

Water Vapor Permeability. In general, WPI films had higher WVP than CC films. Incorporating a high concentration of GC (10%) or VE (0.2%) decreased the WVP of the CC films, but adding VE to WPI films did not significantly change their WVP (**Table 3**).

Table 3. Water Vapor Permeability (WVP) of WPI and CC Films[a,b]

film type[c]		thickness	WVP	corrected
protein	nutrients	(mm)	(g·mm/m²·h·kPa)	RH (%)
CC	0	0.1335	12.602ab	78.82
	5% GC	0.1280	11.206bc	78.84
	10% GC	0.1268	9.600c	79.85
	0.1% VE	0.1200	11.242bc	81.87
	0.2% VE	0.1132	8.576c	83.89
WPI	0	0.1549	15.013a	77.81
	0.1% VE	0.1536	14.522a	78.81
	0.2% VE	0.1450	13.526ab	80.84

[a] Mean values with different letters in the same column differ ($P < 0.05$). [b] As means of triplicate determination. [c] CC, calcium caseinate; WPI, whey protein isolate; GC, Gluconal Cal; VE, α-tocopheryl acetate.

Table 4. Tensile Strength (TS) and Elongation at Break (EL) of WPI and CC Films[a,b]

film type[c]		thickness	cross area		
protein	nutrients	(mm)	(mm²)	TS (MPa)	EL (%)
CC	0	0.1749	2.5130	3.2178c	34.73cd
	5% GC	0.1515	2.2720	1.7921d	40.63bcd
	10% GC	0.1714	2.4880	1.7652d	13.34e
	0.1% VE	0.1716	2.4938	2.7880c	61.69a
	0.2% VE	0.2050	3.1200	0.8246e	50.37abc
WPI	0	0.1661	2.4921	4.5553a	12.14e
	0.1% VE	0.1744	2.6166	4.4527ab	29.42d
	0.2% VE	0.1749	2.6235	4.0504b	52.40ab

[a] Mean values with different letters in the same column differ ($P < 0.05$). [b] As means of six determinations. [c] CC, calcium caseinate; WPI, whey protein isolate; GC, Gluconal Cal; VE, α-tocopheryl acetate.

The WVP of edible films depends on many factors, including the integrity of the film, the ratio between crystalline and amorphous zones, the hydrophilic−hydrophobic ratio, and the polymeric chain mobility (*32*). Ionic calcium is known to form strong molecular cross-linking in caseinate films, which would improve protein network formation and stability (*18*), reducing the moisture diffusion rate. Ionic calcium cross-linking also reduced protein solubility in water and led to decreased WVP through the protein matrix (*17*). Park et al. (*18*) indicated that calcium sulfate improved water barrier properties of soy protein isolate (SPI) film by reacting with protein constituents to yield insoluble proteinates and altered film consistency. From this study associated with a high concentration of calcium incorporation into film-forming solutions, it might be concluded that calcium improves the WVP of CC films due to its capacity to promote aggregation, thus reducing the solubility of protein. The mechanism was similar to those of low concentration calcium additions.

Incorporating AM and 0.2% VE mixture into CC films reduced their WVP; however, similar results were not found in the WPI films (**Table 3**). This may be related to the hydrophobic nature of AM and VE and the structure of proteins. In the protein−lipid emulsion films, protein can act as a cohesive matrix, whereas lipid can distribute uniformly as discrete crystalline particles to provide resistance to moisture movement (*17*). Homogenization used in forming emulsion could reduce lipid droplet sizes, thus another potential for reducing the WVP (*7*, *33*). Increased concentration of VE and AM might increase the ratio of crystalline and amorphous zones and decrease the ratio of hydrophilic−hydrophobic properties of the protein system.

Film thickness often affects the WVP determination of hydrophilic films. McHugh et al. (*24*) suggested that as film thickness increased, the film provided an increased resistance to mass transfer across it. In this study, film thickness varied among different types of films used for WVP determination (**Table 3**) and thus might affect the accuracy of WVP calculation. Future research must have good control of film thickness to fully understand the WVP of films associated with the incorporation of calcium or vitamin E.

Tensile Properties. The tensile strength and elongation at break (EL) of CC and WPI films are shown in **Table 4**. Adding GC to CC films or 0.2% VE to CC and WPI films reduced their tensile strength ($P < 0.05$). For EL, 10% GC incorporation significantly reduced EL of CC films, whereas adding VE to film-forming solutions greatly improved the EL values of both CC and WPI films.

As cross-linking agents, calcium ions have been reported to enhance the mechanical properties of materials when used at a very low concentration (*18*, *19*). It was suggested that calcium cations bind strongly with polar groups of protein to form denser three-dimensional networks in CC films (*18*). In this study, a significantly high concentration of calcium was used in film-forming solutions. An increase in calcium concentration could affect the association/dissociation equilibrium of Ca^{2+} binding to the proteins, probably adversely causing further aggregation of the proteins (*34*). It is very important to identify the maximum concentration of calcium that could be incorporated into different film formation solutions without loss of film mechanical properties. It is also critical to understand how different types of calcium salts may function differently in respect to film functionality.

Whereas binding calcium to the carboxyl groups of proteins decreased the protein solubility and increased the mechanical properties, a decrease in pH aggregated proteins by weakening the electrostatic repulsion and liberated the hydrated water of protein, thus increasing the mechanical properties (*35*). The pH improvement on the mechanical property of CC films did not occur in this study, although the pH value of the CC solution decreased from 6.41 to 6.02 or 5.97 when 5 or 10% GC, respectively, was incorporated (**Table 1**). Many other factors contribute to the mechanical property of films. For example, the presence of air bubbles in the solution could reduce the mechanical properties. McHugh et al. (*33*) indicated that the high viscosity of the weak gel materials could result in the inability to remove air bubbles prior to film formation. In this study, high-concentration calcium incorporation resulted in high-viscosity heterogeneous structure of the solutions, thus making it difficul to completely remove air bubbles. The tiny air bubbles were observed on the films containing high concentrations of GC. In a future study, examination of the microstructure of the film must be conducted to understand fully the presence of air bubbles and their effect on the mechanical property of the film. In addition, film thickness played a significant role in the mechanical property and needs to be well controlled.

When lipids are incorporated into film-forming solutions, crystalline lipid particles dispersing into the protein matrix would bring about heterogeneous structure and decrease the tensile strength of edible films, which explained the decreased tensile strength in CC and WPI films containing VE and AM. On the other hand, lipophilic compounds acting as emulsifiers and plasticizers increase film flexibility (*32*) by reducing the excessive intermolecular forces and increasing the mobility of polymer chains, thus improving the elongation at break of CC and WPI films incorporating VE and AM.

Conclusion. Along with increased market demands on nutritionally fortified foods, edible coatings and films containing high concentrations of nutraceuticals would provide alternative ways to fortify foods that otherwise cannot be, such as fresh

fruits, vegetables, and other unprocessed food items. Products could be either coated or wrapped with nutritionally fortified coatings or films. Our previous study has confirmed the feasibility of this type of coating to enhance the nutritional value of fresh vegetables. The intended use of the coating or films would dictate the requirements on water barrier and mechanical properties. This study demonstrated that the WVP and tensile property of milk protein-based edible films, such as CC and WPI, may be compromised when high concentrations of calcium and vitamin E are incorporated. High concentrations of calcium and vitamin E reduced the WVP and tensile strength of CC films, probably due to the heterogeneous structure and decreased protein solubility. Vitamin E incorporation had no effect on the WVP of WPI films but improved the elongation at break and reduced the tensile strength of the films because of its hydrophobic nature, the effect of plasticizer, and the heterogeneous structure of the emulsion films. To understand their effects fully, future research must study the thermal property, gas permeability, and microstructure of the films at different levels of nutraceutical incorporation.

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