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Chickpea flour and soy protein isolate interacted with κ-carrageenan via electrostatic interactions to form egg omelets analogue

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ABSTRACT

In recent years, demand for the plant-based egg substitutes has increased significantly, especially in Singapore, a country seeking for innovative food sources imminently. In the current study, chickpea flour, soy protein isolate, shortening, baking powder, mono, diglyceride, transglutaminase, potassium chloride, flour, and hydrocolloids (κ-carrageenan (κ-C) or gellan gum (GG)) were used to develop the eggless omelets. A formulation comprising 0.3% κ-C (0.3κ-C) best matched the physiochemical properties of egg, in terms of hardness (4437 vs. 4614 g), specific volume (1.24 vs. 1.19 cm^3/g), and gel strength (19.3 vs. 17.5 kPa). This could be attributed to the highest synergistic κ-C-protein interactions in 0.3κ-C, along with the most homogeneous gel structure observed under confocal laser scanning microscopy (CLSM). The addition of 0.1% κ-C induced more κ-C-protein interactions than the one without hydrocolloids, but such increase was not as dominant as 0.3κ-C. When the κ-C concentration reached 0.5%, however, the rheological synergism decreased while the electrostatic interactions increased; that signifies the increased κ-C-κ-C interactions. Contrastingly, a segregated GG-protein interaction occurred in all GG systems, as indicated from synergism and CLSM images. These differences in interactions and structures affected the macroscale properties of our plant-based egg products, explaining the different physiochemical properties among them. A schematic diagram was therefore proposed to build connections between physiochemical properties, interactions, and structure.

1. Introduction

The global egg replacement ingredients market has reached USD 1.4 billion in 2021 and is expected to surpass USD 1.6 billion by 2026 (Market Data Forecast, 2021). The growing interest in the development of egg substitutes is driven by various factors, such as consumer preference, reducing allergens, enhancing food safety, improving nutrition profile, reducing price volatility, and promoting environmental sustainability (Grizio & Specht, 2018). In Singapore, the current high egg consumption (388 pieces per capita (Singapore Food Agency, 2020)) relies mostly on importation from Malaysia (73%). The shortage of egg supplies from overseas during avian influenza period, and the more energy required to produce eggs than that to produce milk and raise swine combined (Sabate & Soret, 2014), have driven a rapid shift in egg markets from animal products to plant-based alternative.

The plant-based egg replacement category has already experienced some early harbingers, such as egg-free mayonnaise and dressings (Ali & EL Said, 2020; Armaforte, Hopper, & Stevenson, 2021), eggless cakes (Lin, Tay, Yang, Yang, & Li, 2017a; 2017b), and eggless noodles (Khouryieh, Herald, & Aramouni, 2006). Currently, several companies (e.g., Oggs Aquafaba, Just egg, Beyond egg, etc.) have launched novel plant-based egg products that successfully mimic the taste and appearance of eggs. From all these egg replacement applications, it was noted that the combination of proteins, hydrocolloids, and emulsifiers was promised in developing egg substitutes (Keys & Goldberg, 2018). In the current study, chickpea flour and soy protein isolate were selected as protein sources due to their exceptional emulsifying, foaming, and gelling properties, nutritional value, low cost, and wide availability (Boukid, 2021; Grizio & Specht, 2018; Romagnesi & Sharma, 2021; Söderberg, 2013). In addition, κ-carrageenan (κ-C) and gellan gum (GG) were evaluated to further improve our eggless formulations based on their thickening, gelling, and water binding capacity (Saha & Bhattacharya, 2010) and the successful application in eggless products (Just egg, 2019; Keys & Goldberg, 2018).

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Although several plant-based egg products were commercialized as mentioned above, few of them have explored the physiochemical properties of the plant-based eggs, as well as the mechanism underlying these properties. Protein-polysaccharide interaction is vital and common in novel food development (Lopes-da-Silva & Monteiro, 2019; Wu, Lin, Singh, & Ye, 2020). Mixing of protein and polysaccharide would lead to electrostatic interactions, steric exclusion, hydrophobic interactions, and hydrogen bonding that affect the structure development (McClements, 2006), and the gelation process increases complexity of the structure (Panouillé & Larreta-Garde, 2009). This microstructure could further affect the macroscopic properties of food systems (Sow, Kong, & Yang, 2018). As a result, protein-polysaccharide interactions could be customized to achieve the desirable physicochemical properties via creation of microstructures in foods (Aguilar et al., 2011). To date, mixing of κ-C or GG into proteins has been widely studied in various bi-polymeric systems (e.g., κ-C-soy protein (Zhang et al., 2021), κ-C-fish gelatin (Sow, Chong, Liao, & Yang, 2018), κ-C-casein (Bourriot, Garnier, & Doublier, 2000), GG-milk protein (Picone & da Cunha, 2010), GG-fish gelatin (Sow, Tan, & Yang, 2019), etc.). However, to the best of our knowledge, there are few studies about the behavior of complex structures in the tri-polymeric protein-polysaccharide-starch systems. Agoda-Tandjawa, Le Garnec, Boulenguer, Gilles, and Langendorff (2017) and Matignon et al. (2014) have proposed a composite κ-C-milk protein-starch gel structure where κ-C-milk proteins interactions are formed preferentially with starch granules filled inside. Limited information was reported regarding the κ-C/GG interactions with soy proteins in the presence of starch, which could finally determine the physiochemical properties of our egg omelets analogue.

Consequently, the main objective of our study was to explore the underlying mechanisms that contributed to the different physiochemical properties of plant-based egg samples prepared with different hydrocolloids (κ-C or GG) at the structural and interaction levels. For these purposes, texture profile analysis (TPA), specific volume measurement, rheological tests were performed to characterize the physiochemical properties, confocal laser scanning microscopy (CLSM) analysis was performed to observe the structure, and rheological synergy, molecular interaction test, sulfhydryl measurement, surface hydrophobicity measurement, zeta-potential measurement, and Fourier transform infrared (FTIR) spectroscopy were performed to explore the interactions.

2. Materials and methods

2.1. Materials

The formulations of plant-based liquid egg were developed based on Perret (1974) and Keys and Goldberg (2018) with modifications (Table 1). Chickpea flour (Dr Gram®), soy protein isolate (Myprotein®), shortening (Crisco®), double action baking powder (Redman®), unbleached wheat flour (Prima flour®), and sunflower oil (Naturel®) were obtained from Lazada, Singapore. The liquid egg control (N&N®) was obtained from N&N Agriculture Pte Ltd. Other ingredients used were mono and diglycerides (MDG) (Emulpals 110®), obtained from Palsgaard Asia Pacific Pte Ltd (Singapore); transglutaminase (Ajinomoto®) from Amazon (Seattle, U.S.A); potassium chloride (KCl) from Sigma Aldrich (Singapore); κ-carrageenan (κ-C) from Better 4U Holdings Pte Ltd. (Singapore); and high acyl gellan gum (GG) from CP Kelco (Singapore). Protein-free chickpea flour (CP(−)) was prepared by isolating protein fraction from chickpea flour as described by Sánchez-Vioque, Clemente, Vioque, Bautista, and Millán (1999). Sodium chloride (NaCl), urea, guanidine hydrochloride (GuHCl), dithiothreitol (DTT), phosphate buffer, tris(hydroxymethyl)aminomethane (Tris), glycine, Bradford reagent, ethylenediaminetetraacetic acid (EDTA), sodium dodecyl sulfate (SDS), 5,5′-dithio-bis-(2-nitrobenzoic acid) (DTNB), bromophenol blue (BPB), potassium bromide (KBr), rhodamine B and fluorescein isothiocyanate (FITC) were obtained from Sigma Aldrich (Singapore). Ethanol was obtained from VWR Singapore Pte Ltd. (Singapore). All food components were at food grade and chemicals for analytical purpose were at analytical grade.

2.2. Preparation of plant-based liquid egg, omelet, egg gels, and determination of nutritional compositions of liquid eggs

The plant-based liquid egg samples were prepared by blending all ingredients (Table 1) in the electric blender (Mayer, Singapore) at speed 2 (20,000 rpm) for 6 min. One hundred grams of liquid egg was poured in a frypan (Lamart®, Singapore) pre-heated at 130 °C for 2.5 min with sunflower oil (5 g), heated at 130 °C for 2.5 min without stirring, and then flipped to the other side to continue heating for another 1 min. The omelets were cooled at room temperature for 60 min before analyses. Egg gels were formed by heating the liquid egg samples at 90 °C for 30 min, followed by maturation at 4 °C for 12 h before analyses.

The protein and moisture contents were determined by the Kjeldahl method and the oven-drying method, respectively. The fat content was gravimetrically determined after Soxhlet extraction. The nutritional composition of liquid egg was 77.05 ± 0.57 wt% of water, 10.56 ± 0.03 wt% of protein, and 13.79 ± 0.12 wt% of lipid. The nutritional composition of plant-based liquid egg was 72.11 ± 0.29 wt% of water, 4.99 ± 0.08 wt% of protein, and 9.11 ± 0.21 wt% of lipid.

2.3. Physiochemical properties of plant-based omelets

Before determining the physiochemical properties, the cooled omelets were cut into pie shape (diameter = 20 mm) using a circular ring mold. Thickness of five stacking omelet pieces was measured in

Table 1
Formulations of different plant-based liquid egg samples.

Ingredients	Composition (%)						
	0gum	0.1κ-C	0.3κ-C	0.5κ-C	0.1 GG	0.3 GG	0.5 GG
CF	8.82	8.82	8.82	8.82	8.82	8.82	8.82
SPI	3.78	3.78	3.78	3.78	3.78	3.78	3.78
Shortening	9.60	9.60	9.60	9.60	9.60	9.60	9.60
Baking powder	1.00	1.00	1.00	1.00	1.00	1.00	1.00
MDG	0.50	0.50	0.50	0.50	0.50	0.50	0.50
TGA	0.025	0.025	0.025	0.025	0.025	0.025	0.025
KCl	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Flour	2.00	2.00	2.00	2.00	2.00	2.00	2.00
κ-C	–	0.10	0.30	0.50	–	–	–
GG	–	–	–	–	0.10	0.30	0.50
Water	74.175	74.075	73.875	73.675	74.075	73.875	73.675

*CF – chickpea flour; SPI – soy protein isolate; MDG – mono, diglycerides; TGA – transglutaminase; KCl – potassium chloride; κ-C – κ-carrageenan; GG – gellan gum.
*LE, 0gum, 0.1κ-C, 0.3κ-C, 0.5κ-C, 0.1 GG, 0.3 GG, and 0.5 GG refer to liquid egg, plant-based liquid egg with κ-C/GG addition of 0.0, 0.1, 0.3, 0.5% (w/w), respectively.

millimeter using a vernier caliper. The volume of the omelet pieces was estimated using the following formula (Julina & Thyagaraj, 2019):

$$Volume = \pi\left(\frac{D}{2}\right)^2 T \quad (1)$$

where, D and T are the diameter and thickness of the omelet pieces, respectively.

The specific volume of the omelet pieces was then calculated as below (Lin et al., 2017a):

$$Specific\ volume = \frac{Volume}{Mass} \quad (2)$$

The textural profile of the omelet pieces was analyzed using TA-XT2i texture analyzer (Stable Micro System Ltd., Surrey, UK) equipped with the P/35 probe according to Zhang et al. (2019). Double-compression test was conducted under the following setting: crosshead speed = 2 mm/s, compression deformation = 50% of the initial sample height, and time interval between the two compressions = 5 s. Hardness and springiness were determined as published previously (Mao et al., 2017; Yang, Wu, Ng, & Wang, 2017).

2.4. Rheological properties

Rheological properties of the plant-based liquid egg samples were characterized using a rotational stress-controlled rheometer (MCR 102, Anton Paar, Graz, Austria) fitted with a Peltier temperature controller. A stainless-steel parallel plate (25 mm in diameter) with 0.5 mm gap was chosen. All liquid egg samples were equilibrated at room temperature for 30 min before testing. Then, about 1 mL of the sample was loaded on the rheometer plate (T = 20 °C), with silicone oil covering the edge to avoid evaporation. Before measurements, linear viscoelastic region (LVR) was determined from strain sweep. The samples were subject to the following steps:

(1) A temperature sweep was conducted from 20 to 90 °C, kept constant at 90 °C for 30 min, and declined from 90 to 20 °C (rate = 1.5 °C/min, frequency = 1 Hz, strain = 0.1%) (Huang, Mao, Li, & Yang, 2021);
(2) The gel from (1) was matured at 20 °C (frequency = 1 Hz, strain = 0.1%) for 30 min;
(3) A frequency sweep from 100 to 0.1 rad/s was carried out at the gel state from (2) (strain = 0.1%, temperature = 20 °C) (Yang, Gao, & Yang, 2020). The powder law model was applied to describe the relationship between the angular frequency (ω) and storage modulus (G'):

$$G' = A\omega^n \quad (3)$$

where, A is the indicator of gel strength and n is the relaxation exponent showing the gel network correlation;

(4) The rheological synergistic effect of the plant-based egg gels prepared with/without proteins was quantified as followed (Agoda-Tandjawa et al., 2017):

$$R = \frac{G'_{mixture} - \sum G'_{ingredients}}{\sum G'_{ingredients}} \quad (4)$$

where, $G'_{mixture}$ is the storage modulus of the mixed system, and $G'_{ingredients}$ is the storage modulus of each ingredient at 20 °C after step (2).

2.5. Molecular interactions analysis

To evaluate the molecular interactions involved in the gelation of the different plant-based egg systems, different treatments were applied according to Zhang et al. (2019) with modifications. Sodium chloride (NaCl, 1 mol/L) was applied to investigate the electrostatic interactions, urea (2 mol/L) was applied to disrupt the H bonding, guanidine hydrochloride (GuHCl, 2 mol/L) was applied to disrupt the H bonding and hydrophobic interactions, and dithiothreitol (DTT, 0.5% (w/w)) was applied to dissociate the disulfide bonds. The loss of gel strength (storage modulus G' at 20 °C) was calculated to show the effect of each molecular interaction:

$$Loss\ of\ gel\ strength\ (\%) = \left(1 - \frac{G'_1}{G'_0}\right) \times 100 \quad (5)$$

where, G'_1 is the gel strength of the gel samples under different treatment, and G'_0 is the gel strength of the non-treated control samples.

2.5.1. Total sulfhydryl groups and free sulfhydryl groups

Total and free sulfhydryl groups were measured as described by Beveridge, Toma, and Nakai (1974) with modifications. Three grams of plant-based egg gel sample was homogenized in 27 mL of phosphate buffer (100 mmol/L, pH 8.0) at 12,000 rpm for 2 min using a Wiggens high-speed homogenizer (Bio Laboratories Pte Ltd., Singapore). The mixture was then centrifuged at 8,000 *g* for 20 min, followed by collection of supernatants. The Bradford method was applied to determine the protein concentration in the supernatants.

For free sulfhydryl measurement, 2.8 mL of Tris-Gly buffer (0.1 mol/L Tris, 0.1 mol/L glycine, 4 mmol/L EDTA, pH 8.0) and 20 μL of Ellman's reagent (4 mg/mL DTNB in Tris-Gly buffer) were added into 200 μL of the supernatant collected above. For total sulfhydryl measurement, 2.8 mL of 0.5% SDS in urea-Tris-Gly buffer (8 mol/L urea, 0.1 mol/L Tris, 0.1 mol/L glycine, 4 mmol/L EDTA, pH 8.0) and 20 μL of Ellman's reagent (4 mg/mL DTNB in Tris-Gly buffer) were added into 200 μL of the supernatant collected above. After that, the mixtures were incubated under 40 °C for 15 min, followed by cooling to room temperature. The absorbance of the solutions at 412 nm was measured using a UV-1800 spectrophotometer (Shimadzu Corporation, Kyoto, Japan). The blank control consisted of phosphate buffer only.

The content of sulfhydryl groups was quantified by:

$$SH\ (\mu M / g\ protein) = \frac{73.53 \times A_{412} \times D}{C} \quad (6)$$

where, 73.53 is result from unit conversion divided by molar extinction coefficient ($10^6/1.36 \times 10^4\ M^{-1}cm^{-1}$) (Ellman, 1959), A_{412} is the photometric absorbance at 412 nm, D is the dilution factor (15.01 in the current study), and C is the protein concentration (mg/mL) in the supernatant.

The content of disulfide bonds was further calculated according to Zhao et al. (2013):

$$S - S\ (\mu M / g\ protein) = \frac{SH_{total} - SH_{free}}{2} \quad (7)$$

where, SH_{total} and SH_{free} are the content of total and free sulfhydryl groups, respectively.

2.5.2. Protein surface hydrophobicity

Protein surface hydrophobicity was assayed as described by Bertsch, Mayburd, and Kassner (2003) with modifications. To 1 mL of the supernatant collected above, 200 μL of bromophenol blue solution (BPB, 1 mg/mL in deionized water) was added. The control sample was prepared by adding 200 μL of BPB solution into 1 mL of phosphate buffer. The mixtures were agitated under room temperature for 10 min, and then centrifuged at 20,000 *g* for 15 min. The absorbance of 200 μL supernatant was determined at 595 nm against a blank (phosphate buffer only) using a PowerWave XS2 Microplate Spectrophotometer (Biotek, Winooski, VT, U.S.A.). The content of BPB bound was quantified by:

$$BPB\ bound\ (\mu g\ /\ g\ protein) = 200 \times \frac{A_{595}\ from\ control - A_{595}\ from\ sample}{A_{595}\ from\ control \times C \times 10^{-3}} \quad (8)$$

where, A595 is the absorbance at 595 nm, and C is the protein concentration (mg/mL) in the supernatant.

2.5.3. Zeta-potential

Zeta-potential was measured according to Sow et al. (2019) with modifications. Plant-based liquid eggs were diluted to 0.25% (w/w) with deionized water, followed by shaking for 2 h under room temperature. Measurement was conducted in the NanoBrook Omni Particle Size and Zeta Potential analyzer (Brookhaven Instruments, NY, U.S.A) under the phase analysis light scattering (PALS) mode.

2.6. Fourier transform infrared (FTIR) spectroscopy

Lyophilized egg gels were ground with KBr (3 mg sample/97 mg KBr) for pellet preparation (Sow & Yang, 2015). The FTIR spectra (4000-450 cm^{-1}) were obtained using a Spectrum One FTIR spectrometer (PerkinElmer, Waltham, MA, U.S.A.), at 4 cm^{-1} resolution and scan number of 32. Background was corrected before every sample spectrum. Fourier self-deconvolution was performed at the amide I region (1700-1600 cm^{-1}) using the Omnic software 8.2 (Thermo Fisher Scientific Inc. Waltham, MA, U.S.A) with the settings of 30 cm^{-1} bandwidth and 1.3 enhancement factor, followed by curve normalization and gaussian peak fitting using OriginPro 9.0 software (OriginLab, Northampton, MA, U.S.A). The percentages of the secondary structures were determined by integrating the areas of the fitted peaks.

2.7. Confocal laser scanning microscopy (CLSM) analysis

Egg gels were fluorescently labelled as described previously (Huang et al., 2021). Rhodamine B (0.1 g/L in ethanol) and FITC (0.1 g/L in ethanol) were chosen to stain the proteins and polysaccharides, respectively, with a ratio of 1:2 (v/v). The solid egg gels were cut into slices, followed by soaking into the diluted dye mixture (1.0% in ethanol) for staining. After setting at room temperature for 15 min, the solids were rinsed by ethanol, and then transferred onto a glass slide. The microstructure was observed using CLSM with an Olympus Fluoview FV1000 confocal scanning unit (Tokyo, Japan) embedded with argon ion and helium–neon (HeNe) lasers. The excitation/emission wavelength of rhodamine B and FITC was 540/625 and 490/525 nm, respectively. Images were captured at 10× magnification.

2.8. Statistical analysis

All measurements were repeated at least in triplicate independently. For CLSM, at least 10 parallel images were captured for each sample to achieve reliable microstructure observation. Results were presented in mean ± standard deviations. Significant differences among groups were evaluated using one-way ANOVA with post-hoc Bonferroni test in the SPSS Statistics 20 software (IBM, Chicago, IL, U.S.A.) with $P < 0.05$ (two-tailed) considered as statistically significant.

3. Results and discussion

3.1. Physiochemical properties and gel strength

The physiochemical properties of omelet made from liquid egg were set as the target reference for the development of plant-based omelets. Specific volume of omelets with different recipes is shown in Fig. 1. The substitution of eggs resulted in a significant decrease in specific volume compared with that of the control (1.24 cm^3/g vs. 1.00 cm^3/g). Addition of κ-C increased the specific volume of the eggless omelets significantly to a level comparable to that of the control omelet, while addition of GG

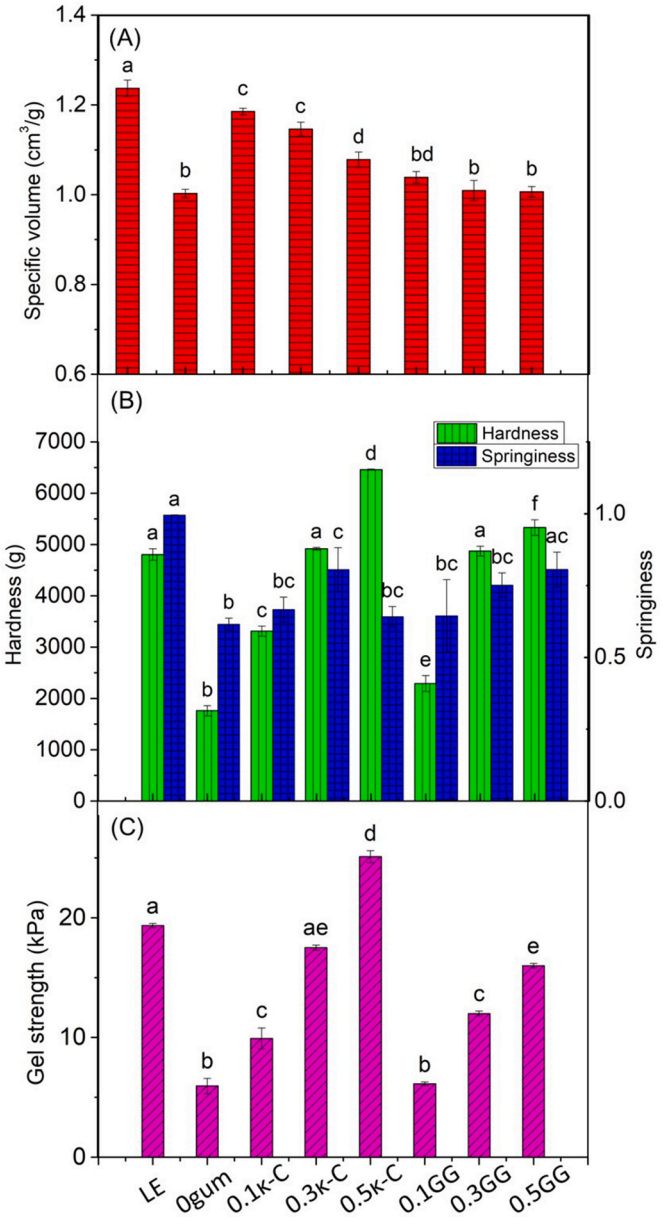

Fig. 1. Effect of the concentration of κ-carrageenan (κ-C) and gellan gum (GG) on the specific volume (A), texture (hardness and springiness) (B), and gel strength (C) of plant-based eggs compared with commercialized liquid egg (LE). *Groups with different letters indicate significant difference ($P < 0.05$). *0gum, 0.1κ-C, 0.3κ-C, 0.5κ-C, 0.1 GG, 0.3 GG, and 0.5 GG refer to plant-based liquid egg with κ-C/GG addition of 0.0, 0.1, 0.3, 0.5% (w/w), respectively.

cannot improve the specific volume of the products. Analogy with the bakery products, specific volume is dependent on the specific gravity and viscosity of the batter (Ashwini, Jyotsna, & Indrani, 2009; Gómez, Ronda, Caballero, Blanco, & Rosell, 2007; Sahi & Alava, 2003) as well as the gel strength of the proteins (Kiosseoglou, 2003). Although previous evidence has shown that chickpea flour exhibited similar foaming capacity to egg white (Mustafa, He, Shim, & Reaney, 2018), a hybrid of chickpea flour and soy protein isolate (SPI) cannot guarantee the specific volume in the eggless omelet (Fig. 1A), which may be attributed to the limited foam stability and gel-forming properties of SPI (Xie & Hettiarachchy, 1998).

The κ-C addition increased the specific volume significantly ($P < 0.05$) compared to those with GG addition or without hydrocolloids. However, as the concentration of κ-C increased, the specific volume

decreased. The balance between gelling properties and thickening effects of hydrocolloids was responsible for this phenomenon. Our κ-C contained liquid egg samples had significantly higher gel strength than the GG samples (Fig. 1C), which contributed to their higher specific volumes. However, when the concentration of κ-C reached 0.5%, the viscosity of liquid egg batter was too high (data not shown), and the aeration was impeded during mixing (Lin, Tay, Yang, Yang, & Li, 2017b), thus resulting in a smaller omelet.

Textural profiles of omelets with different recipes are presented in Fig. 1B. The plant-based sample without hydrocolloid addition (0gum) displayed significantly lower gel strength and hardness compared with egg omelet ($P < 0.05$). As the concentration of two hydrocolloids increased, gel strength and hardness of the plant-based samples increased (Fig. 1B and C). Although κ-C and GG showed similar impacts on the hardness of the plant-based system at the same concentration, it was found that κ-C provided more pronounced increase in gel strength than GG (Fig. 1C). In particular, the κ-C contained gels matched the gel strength of egg gel at the concentration of 0.3%, while that of GG contained gels cannot reach the benchmark. This finding was inconsistent with the gel strength behavior of the fish gelatin-polysaccharide gels reported earlier (Sow, Kong, & Yang, 2018). In our κ-C system, interactions between κ-C and protein are more pronounced, while in the GG system, GG and proteins tend to form separative double network structure (Picone & da Cunha, 2010). Such difference in gel structure may contribute to the different mechanical properties of our gel samples.

On the other hand, the plant-based omelet displayed poor springiness with respect to the control omelet (91.2% vs. 62.2%) (Fig. 1B). This could be attributed to the variations in protein source of our plant-based formulations (soy protein, chickpea protein) and the control (egg proteins) (Wilderjans, Pareyt, Goesaert, Brijs, & Delcour, 2008). The value of springiness increased with the increase in polysaccharide concentration, except for the 0.5% κ-C sample. Previous research has shown that the hydrocolloids' ability of binding water can lead to an increase in

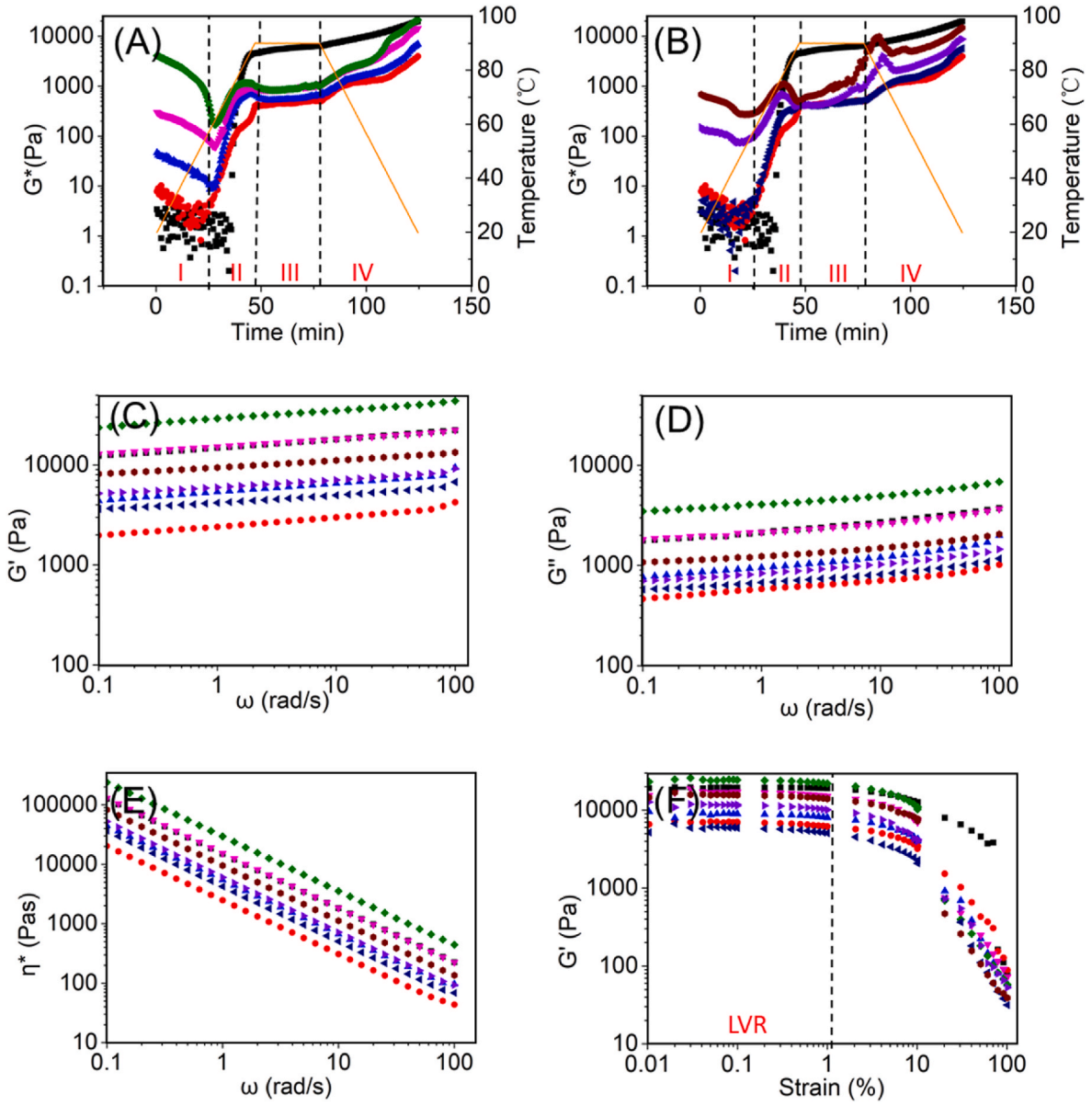

Fig. 2. Temperature sweep results of complex modulus G* for (A) κ-C systems and (B) GG systems; Frequency sweep results of (C) storage modulus G', (D) loss modulus G", and (E) complex viscosity η*; Strain sweep results of (F) storage modulus G'.

*■, liquid egg control; ●, 0 gum sample; ▲, 0.1κ-C sample; ▼, 0.3κ-C sample; ◆, 0.5κ-C sample; ◀, 0.1 GG sample; ▶, 0.3 GG sample; ⬢, 0.5 GG sample. *0gum, 0.1κ-C, 0.3κ-C, 0.5κ-C, 0.1 GG, 0.3 GG, and 0.5 GG refer to plant-based liquid egg with κ-C/GG addition of 0.0, 0.1, 0.3, 0.5% (w/w), respectively.

springiness (Azmoon et al., 2021). However, when the κ-C concentration reached 0.5%, the syneresis effect was responsible for the lower springiness value. Therefore, the concentration of hydrocolloids applied in the products should be limited.

3.2. Viscoelastic behavior of plant-based liquid egg systems

The thermal behavior of liquid egg samples at pH 7.5 is illustrated by the temperature sweep curves (Fig. 2A and B). Four stages were identified for the control sample over the thermal cycle (Aguilar, Cordobés, Raymundo, & Guerrero, 2017; Huang et al., 2021; Zhang et al., 2019), representing the gel development process involving egg proteins. As for our plant-based egg samples, this four-stage thermal behavior was still apparent (Fig. 2A and B). It was suggested that our plant-based egg system exhibited similar gelling performance to the commercial liquid egg, mainly due to the gelatinization of starch and gel network formation of proteins. For both plant-based egg/κ-C and plant-based egg/GG systems, the complex modulus (G*) increased with increased hydrocolloid concentration, which corresponded to the entanglement of these hydrocolloids with starch and proteins. In the cooling stage, only the eggless samples with 0.3% and 0.5% κ-C presented comparable G* values with the liquid egg control, indicating their similar gel strength.

In addition, the loss factors tan δ (tan δ = G″/G′) that reflect the changes in liquid-like characters of the liquid eggs over the heating-cooling cycle are presented in Fig. S1. The values of tan δ > 1 denote a predominance of loss modulus (G″) over storage modulus (G′) (liquid-like behavior), while tan δ < 1 denote a predominance of G′ over G″ (solid-like behavior). For the control liquid egg and the 0gum, 0.1 GG plant-based liquid eggs, the values of tan δ decreased from >1 to <1 as temperature increased, with the crossover point (tan δ = 1) representing the liquid-to-solid transformation (Zhang et al., 2019). However, for the other plant-based liquid eggs, the tan δ values were always lower than 1, suggesting the predominant solid-like behaviors throughout the temperature sweep. As discussed earlier, the addition of κ-C can induce electrostatic interactions with proteins, which resulted in the dominant solid-like behavior of these structured fluids. For the GG addition, GG-GG network formation crosslinked by K^+ was preferential, therefore the 0.3 GG and 0.5 GG liquid eggs delivered a more predominant solid-like behavior even before the heat-induced protein denaturation. Despite the more predominant solid-like behaviors in the plant-based egg samples, they can still be pourable before heating, which is quite important for a plant-based liquid egg product.

To further explore the effects of hydrocolloids, the gelling points at which G' started to rapidly increase are summarized (Table 2) (Gunasekaran & Ak, 2000; Zhang et al., 2019). Overall, the addition of hydrocolloids increased the gelling point significantly, with κ-C delivered more pronounced increase than GG. This could be explained by the high water-binding capacity of hydrocolloids (Tan, Tan, & Easa, 2018), allowing them to compete with starch for the free water and thus delaying the starch gelatinization process (Wilderjans, Luyts, Goesaert, Brijs, & Delcour, 2010). However, another study (Huang et al., 2021) reported a decrease in gelling point in κ-C/yolk system, possibly resulted from the more exposure of hydrophobic groups due to the protein-polysaccharide interactions, which promoted protein aggregation and gel formation. The overall increase in gelling points found in the current study indicated that starch gelatinization is the more pronounced process involved in the gelation of our plant-based egg systems.

Frequency sweeps of all egg gels showed the greater G' values than the G" (Fig. 2C and D), indicating the strong gel systems (Mohtar, Perera, Quek, & Hemar, 2013). The linear decrease in complex viscosity (η*) along with frequency suggested the shear thinning behavior of all gels (Fig. 2E). The similar η* values of 0.3κ-C and LE samples pointed out their similar mouth feel (Piorkowski & McClements, 2014), while the κ-C addition at 0.5% achieved the greatest thickening effect. The power law fitting parameters are summarized in Table 2, with $R^2 > 0.99$ for all samples. As the pre-exponential factor A was regarded as the indicator of gel strength (Yang et al., 2020), the stronger gel was observed at the higher hydrocolloid concentrations. No significant difference ($P < 0.05$) in A was found between 0.3κ-C and LE, confirming the similar gel strength between them. In addition, the relaxation exponent n was found to associate with the structure rigidity positively (Wu et al., 2018), with n = 1 corresponding to a completely elastic structure. The low n values (7.07–9.78%) of the plant-based egg gels indicated that they mimicked the flexible structure of the control egg gel (n = 8.85%).

Table 2
Gelling points and power law model fitting of different egg systems.

Sample	Gelling points (°C)	Power law parameters			
		A ($\times 10^4$ Pa $s^{(1-n)}$)	n ($\times 10^{-2}$)	R^2	RMSE
LE	71.99 ± 1.40^a	1.39 ± 0.13^a	8.85 ± 0.28	0.999	0.405
0gum	43.41 ± 0.76^b	0.24 ± 0.01^b	9.54 ± 0.15	0.994	0.399
0.1κ-C	61.45 ± 0.53^c	0.51 ± 0.04^{b,d}	8.66 ± 0.02	0.998	0.102
0.3κ-C	63.31 ± 1.06^c	1.39 ± 0.19^a	7.80 ± 0.26	0.999	0.532
0.5κ-C	62.30 ± 0.69^c	2.90 ± 0.00^c	8.25 ± 0.13	0.999	0.913
0.1 GG	52.34 ± 0.77^d	0.45 ± 0.05^b	7.65 ± 0.11	0.995	0.202
0.3 GG	52.14 ± 1.05^d	0.54 ± 0.01^{b,d}	7.30 ± 0.05	0.997	0.415
0.5 GG	51.88 ± 0.69^d	0.91 ± 0.05^d	7.07 ± 0.02	0.996	0.409

*Means with different lowercase letters within each column are significantly different ($P < 0.05$) among the different groups.
*LE, 0gum, 0.1κ-C, 0.3κ-C, 0.5κ-C, 0.1 GG, 0.3 GG, and 0.5 GG refer to liquid egg control, and plant-based liquid egg with κ-C/GG addition of 0.0, 0.1, 0.3, 0.5% (w/w), respectively.

3.3. Rheological synergism of plant-based egg mixture in the presence or not of proteins

Fig. 3 shows the rheological synergism (R(−)) of the plant-based egg

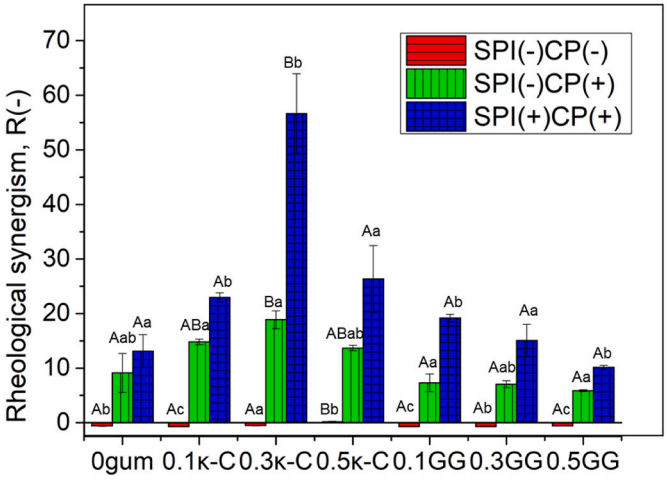

Fig. 3. Rheological synergism (R) of different plant-based eggs prepared with chickpea protein-free chickpea flour (CP(−))/chickpea flour (CP(+)) in the presence (SPI(+))/absence (SPI(−)) of soy protein isolate.
*Within each formulation, groups with different capital letters indicate significant difference ($P < 0.05$) among samples; For the same sample, groups with different lowercase letters indicate significant difference ($P < 0.05$) among different formulations. *0gum, 0.1κ-C, 0.3κ-C, 0.5κ-C, 0.1 GG, 0.3 GG, and 0.5 GG refer to plant-based liquid egg with κ-C/GG addition of 0.0, 0.1, 0.3, 0.5% (w/w), respectively.

samples prepared with chickpea flour (CP(+)) or protein-free chickpea flour (CP(−)), and with soy protein isolate (SPI(+)) or without (SPI(−)). For the chickpea protein and soy protein-free systems (CP(−)SPI(−)), the R(−) values are closed to 0 regardless of the hydrocolloid addition; that signifies the absence of synergistic effect between hydrocolloids and the protein-free systems. In the systems with chickpea protein but without SPI (CP(+)SPI(−)); however, synergistic effects occurred in all systems (R(−) > 0), corresponding to the important role of chickpea protein in reinforcing the network structure of our plant-based egg gels.

On the other hand, the κ-C-based mixed systems displayed greater rheological synergy than that of the GG-based systems and the no hydrocolloid system. This behavior could explain the above observation about the greater gel strength of κ-C samples in comparison to GG samples as κ-C tends to interact with proteins and improve the gel strength. It was also clearly shown that the κ-C contained systems presented the highest synergistic effect at the concentration of 0.3%, where a particular composite gel network may be formed. The addition of SPI (CP(+)SPI(+)) seemed not to change the concentration tendency towards the highest synergistic effect, but strengthened the rheological synergy, especially for the 0.3κ-C system. These increase in synergism values evidenced the contribution of soy protein in further modifying the composite gel network, leading to an improvement in gel strength. Also, the greatest synergy observed at the κ-C concentration of 0.3% rather than 0.5% indicated a predominant κ-C-protein network in the 0.3κ-C gel. Previous studies have proposed that carrageenan chains may bind to the casein micelles preferentially with starch granules acting as the "filler" in the starch/carrageenan/milk protein gel system (Agoda--Tandjawa et al., 2017; Matignon et al., 2014). In the current study, similar interaction preference was observed as the synergistic effect increased with increasing κ-C concentration (<0.5%) in both protein contained systems (CP(+)SPI(−), CP(+)SPI(+)). However, upon increasing κ-C concentration to 0.5%, the κ-C-protein interactions may be progressively reduced, replaced by the κ-C-κ-C interactions.

Contrarily to the predominant synergistic effects of the κ-C systems, no statistically evident synergism was witnessed in the GG systems compared with the 0gum system, whether the presence of proteins or not. This suggested that GG-GG formed the relatively independent network, which interacted less with proteins or starch. Picone and da Cunha (2010) also observed such "phase separation" in whey protein-gellan gum bi-polymeric systems at the concentrations of 3 and 0.3% (w/w) respectively. The authors pointed out that the high polymer concentrations and increasing molar mass due to protein aggregation were responsible for this phenomenon (Picone & da Cunha, 2010). As a result, it could be suspected that interpenetrating networks were formed in our GG-based mixed systems due to the high concentrations and molecular mass of our biopolymers. Heterogeneous structure thus occurred in these samples; that may help to explain their higher fragility and lower deformability than the κ-C samples as discussed earlier.

3.4. Molecular interactions involved in the gelation of plant-based liquid egg

The molecular interactions involved in the gelation process of our plant-based egg systems were explored by the addition of different chemical reagents. Losses of gel strength (%) in comparison with non-treated gels are presented in Fig. 4. As can be seen, all plant-based egg gels showed a high loss of gel strength with 2 mol/L GuHCl addition, associating with their high hydrophobic interactions. Consistent results were achieved from protein surface hydrophobicity analysis (Table 3), in which surface hydrophobicity was found to correlate positively with the loss of gel strength for samples treated with 2 mol/L GuHCl ($r = 0.85$, $P = 0.02$). Hence, we can conclude that hydrophobic interactions contributed to the gel formation significantly in all plant-based egg samples.

Moreover, samples treated with 2 mol/L urea displayed comparable lost gel strength proportion, except for 0.3κ-C sample which presented a significant lower loss of gel strength. The least H-bonding involved in the 0.3κ-C gel development provided extra evidence to the "κ-C/protein/starch" model proposed above as the most predominant κ-C-protein network filled with starch granules may hinder the H bond formation between starch and water molecules.

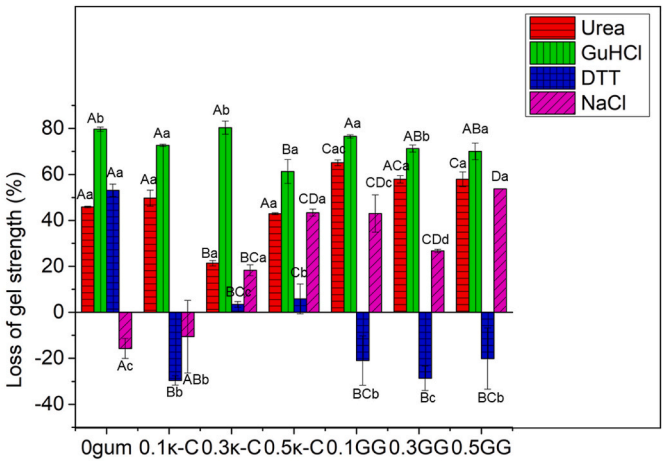

Fig. 4. Loss of gel strength (%) of different plant-based egg gels treated with different dissociation reagents.
*Within each treatment, groups with different capital letters indicate significant difference ($P < 0.05$) among samples; For the same sample, groups with different lowercase letters indicate significant difference ($P < 0.05$) among different treatments. *0gum, 0.1κ-C, 0.3κ-C, 0.5κ-C, 0.1 GG, 0.3 GG, and 0.5 GG refer to plant-based liquid egg with κ-C/GG addition of 0.0, 0.1, 0.3, 0.5% (w/w), respectively.

Table 3
Zeta-potential, disulfide bonding, and BPB bound of different plant-based egg gels.

Sample	Zeta-potential (mV)	Disulfide bonding (μM/g)	BPB bound (μg/g)
0gum	−22.49 ± 0.56^{a,b}	34.40 ± 2.15^{a}	14.73 ± 0.06
0.1κ-C	−21.55 ± 0.53^{a}	5.92 ± 1.78^{b,d}	14.77 ± 3.21
0.3κ-C	−32.89 ± 0.84^{c,d}	3.54 ± 1.18^{b}	18.56 ± 3.08
0.5κ-C	−33.68 ± 0.86^{c}	23.39 ± 1.23^{c}	9.20 ± 0.29
0.1 GG	−25.61 ± 0.30^{a,b}	11.56 ± 2.92^{d}	13.35 ± 0.06
0.3 GG	−27.85 ± 0.25^{b,d}	12.21 ± 3.14^{d}	11.64 ± 0.17
0.5 GG	−33.64 ± 3.06^{c}	10.33 ± 0.62^{b,d}	13.51 ± 0.29

*Means with different lowercase letters within each column are significantly different ($P < 0.05$) among the different groups.
*BPB bound is the indicator of the hydrophobic sites on the protein surface.
*0gum, 0.1κ-C, 0.3κ-C, 0.5κ-C, 0.1 GG, 0.3 GG, and 0.5 GG refer to plant-based liquid egg with κ-C/GG addition of 0.0, 0.1, 0.3, 0.5% (w/w), respectively.

Adding 1 mol/L NaCl to the 0gum and 0.1κ-C samples increased their gel strength, while at higher κ-C concentrations or GG added groups, losses of gel strength were positive. Reasons for such phenomenon could be the case that when the concentration of hydrocolloids was low, the electrostatic interactions within the system were so low that NaCl may promote the hydrophobic interactions instead (Dihort-García et al., 2016), which contributed to a stronger gel. As κ-C amount increased, the effect of NaCl on destructing electrostatic interactions was more pronounced than improving hydrophobic interactions, therefore, a higher loss of gel strength value indicates more electrostatic interactions. In GG systems, loss of gel strength was always observed at any concentrations, being coherent with what has been discussed earlier since the pronounced GG-GG interactions in all GG samples were electrostatically driven (Sow, Kong, & Yang, 2018).

As for the 0.5% DTT treatment, samples with hydrocolloid (0.1–0.5% κ-C and 0.1–0.5% GG) presented significant lower loss of gel strength compared to the one without (0gum). This clearly indicates that disulfide bonds may not be critical to network formation for our hydrocolloid

contained systems, which is quite different from the behavior of the 0gum sample. Our finding agreed with the very weak disulfide bonds found in the canola protein-κ-C systems (Uruakpa & Arntfield, 2006). Interestingly, the structural changes due to DTT inclusion may improve the gel strength, as seen in the 0.1κ-C and GG systems, implying the presence of other interactions. Furthermore, decreased disulfide bonds were also observed in hydrocolloid/plant-based egg systems from –SH measurement (Table 3). This further supported that these systems do not require S–S covalent bonds to form a strong network.

3.4.1. Zeta-potential

The particle charge of the plant-based egg samples is shown in Table 3. Since the pH of all our eggless systems (6.9–7.3) was higher than the isoelectric point of soy protein and chickpea protein (4.0–6.0) (Boukid, 2021; Freitas, Albano, & Telis, 2017), the zeta-potential of all the plant-based egg samples was negative. κ-C and GG carry the zeta-potential of −53.84 and −40.14 mV, respectively. In the samples consisting of hydrocolloids, all the zeta-potential values fell between the 0gum sample and pure hydrocolloid sample, confirming the existence of electrostatic interactions.

Previous studies have pointed out that the sulphated polysaccharides (e.g., carrageenan) can interact with proteins through electrostatic interactions ($-OSO_3^-$ & NH_3^+) even above the isoelectric points (Huang et al., 2021; Samant, Singhal, Kulkarni, & Rege, 1993). However, an increased electrostatic repulsion between GG and proteins at pH > pKa (3.5 for GG) was reported by Picone and da Cunha (2010), which weakened the electrostatic interactions ($-COO^-$ & NH_3^+) between molecules. These different electrostatic behaviors may be due to the relatively low charge density (0.25 negative charges/mol of monosaccharide in average) of GG (de Jong & van de Velde, 2007) and the stronger electrostatic interactions between sulfate group and proteins than those between carboxylic group and proteins (Doublier, Garnier, Renard, & Sanchez, 2000). Thus, one can suspect that the electrostatic interactions in GG systems mainly come from GG-GG interactions crosslinked by K^+ rather than polysaccharide-protein interactions predominant in κ-C systems.

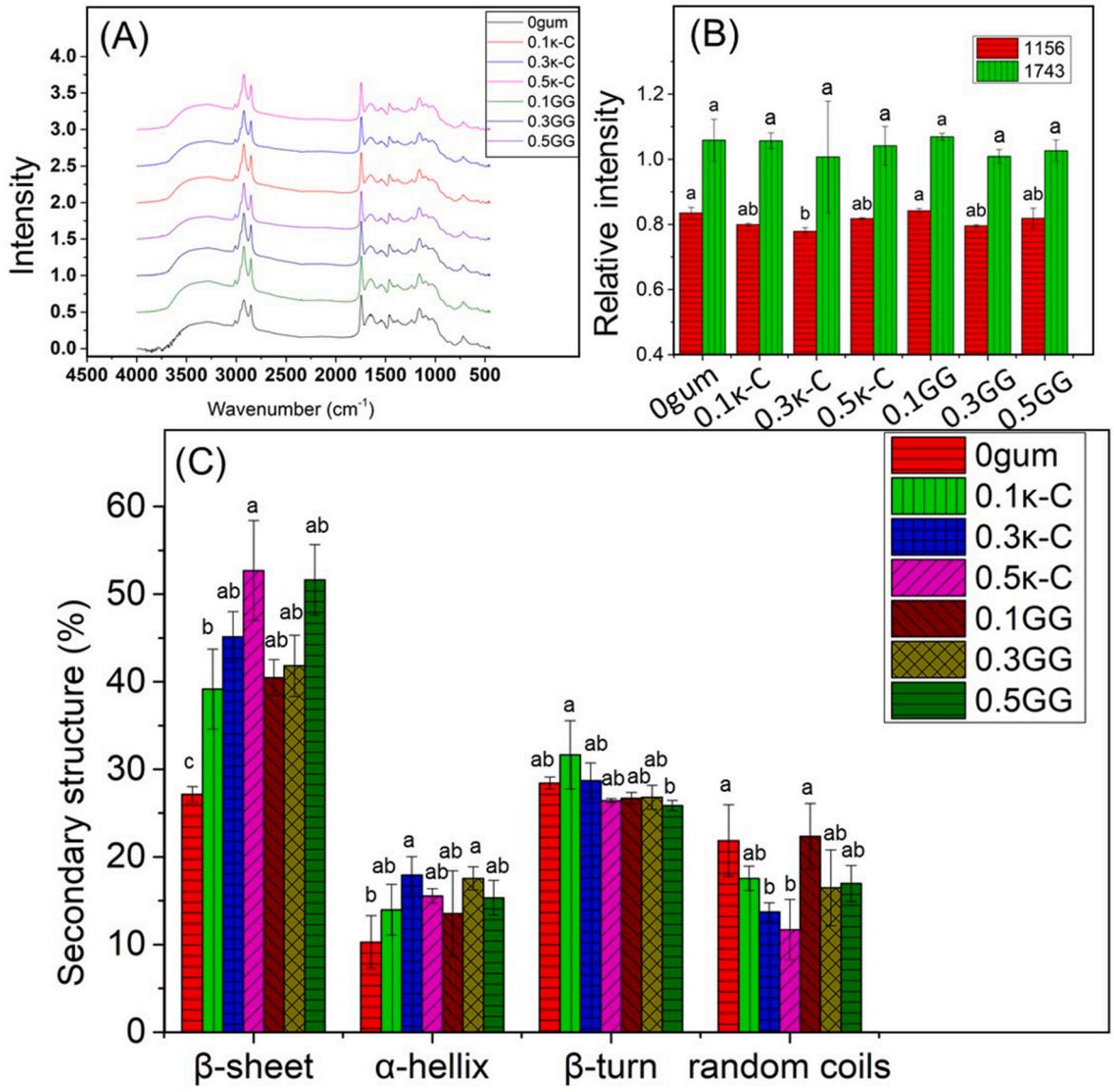

Fig. 5. (A) Fourier transform infrared spectroscopy (FTIR) spectra of different plant-based eggs; (B) Relative intensities of peaks at 1743 and 1156 cm^{-1}; (C) Secondary structure distribution obtained from amide I deconvolution and curve fitting.
*Groups with different letters indicate significant difference ($P < 0.05$). *0gum, 0.1κ-C, 0.3κ-C, 0.5κ-C, 0.1 GG, 0.3 GG, and 0.5 GG refer to plant-based liquid egg with κ-C/GG addition of 0.0, 0.1, 0.3, 0.5% (w/w), respectively.

3.4.2. FTIR

FTIR analysis was conducted to further verify the molecular interactions involved. Fig. 5A shows the spectra of the plant-based egg samples. There was no significant shift on the peak positions and no band occurring/disappearing among all the groups, indicating that the addition of different types or different amount of hydrocolloids in the plant-based egg systems might not significantly alter the skeletal conformations. Bands at ~1659 cm^{-1} and 1536 cm^{-1} were assigned to υ(C═O) stretching (amide I) and δ(N–H) bending (amide II) of amides from proteins, respectively, while bands at ~1380 cm^{-1} and 1260 cm^{-1} were assigned to $\delta_s(CH_2)$ and $\delta_s(CH_3)$ bending of methyl, υ_s(C–O) stretching of COO– groups, and υ_{as}(>P═O) stretching of phosphorus compounds, respectively (Dean, Sigee, Estrada, & Pittman, 2010). In addition, the band at 1743 cm^{-1} was attributed to υ(C═O) of ester groups, and the doublet sharp peaks in the region of 2800–3000 cm^{-1} were associated with υ(C–H) on carbohydrate, lipid, or protein side chains (Kizil & Irudayaraj, 2018). The region of 1200–950 cm^{-1}, corresponding to υ(C–O–C) stretching of polysaccharides, was also notable to illustrate the binding behaviors of starch within the plant-based systems (Ji et al., 2015).

According to Ji et al. (2015), the peaks at 1021, 1083, and 1156 cm^{-1}, resulted from the anhydroglycose ring C–O stretching vibration, can reflect the amount of inter-chain H bonds in starch molecules. Therefore, the lowest relative intensity of the 1156 cm^{-1} peak in the 0.3κ-C system (Fig. 5B) could be attributed to reduced H bonds between the starch chains, which was coherent with the hydrogen bonding measurement result using urea treatment (Fig. 4). Additionally, no significant difference was observed in the relative intensity of the 1743 cm^{-1} peak (Fig. 5B), indicating that the addition of hydrocolloids might not compete with the hydroxyl groups on starch for the carboxylic groups on proteins or other components (Lin et al., 2017a).

Furthermore, the changes in protein secondary structure in response to hydrocolloid addition was characterized from amide I (1600-1700 cm^{-1}) deconvolution and gaussian peak fitting ($R^2 > 0.99$) as described by Lin et al. (2017a) (Fig. S2, Fig. 5C). An increase in α-helix (1645-1662 cm^{-1}) and β-sheets (1615-1638 cm^{-1}) and a decrease in random coils (1638-1645 cm^{-1}) were reported in the κ-C or GG modified pant-based samples compared to the 0gum sample. Hydrocolloid addition might modify the structure of proteins by increasing the more ordered structure (α-helix and β-sheets) and reducing random coils, a phenomenon similar to the fish gelatin-κ-C complex demonstrated by Sow, Chong, et al. (2018). Also, such structural modification could improve the strength of the network, leading to the stronger gels after hydrocolloid modification. However, when the concentration of κ-C reaches 0.5%, the helix structure reduces to the level of the 0gum sample, which further ascertains the predominant κ-C-κ-C network in this system.

3.5. CLSM

Microstructures of different plant-based egg systems were observed through CLSM (Fig. 6). In overlapped channel (Fig. 6A–G), the green domain should be the dyed polysaccharides (κ-C/GG and starch) while the yellow domain is regarded as the overlap of proteins and polysaccharides. After heating, the plant-based eggs formed a gel with many holes, a phenomenon similar to the real egg yolk gels (Huang et al., 2021). The more holes in 0.3κ-C gel (Fig. 6C) corresponded to its higher air retention and greater specific volume in omelet products.

In rhodamine B channel (Fig. 6A'-G'), the protein network structure in our plant-based egg systems was observed. For κ-C samples, 0.3κ-C showed a relatively uniform structure (Fig. 6C'), while 0gum, 0.1κ-C, and 0.5κ-C contained some over-dense signals, signifying the denser protein aggregates. The most homogeneous network structure in 0.3κ-C gel could be correlated with the most κ-C-protein interactions indicated

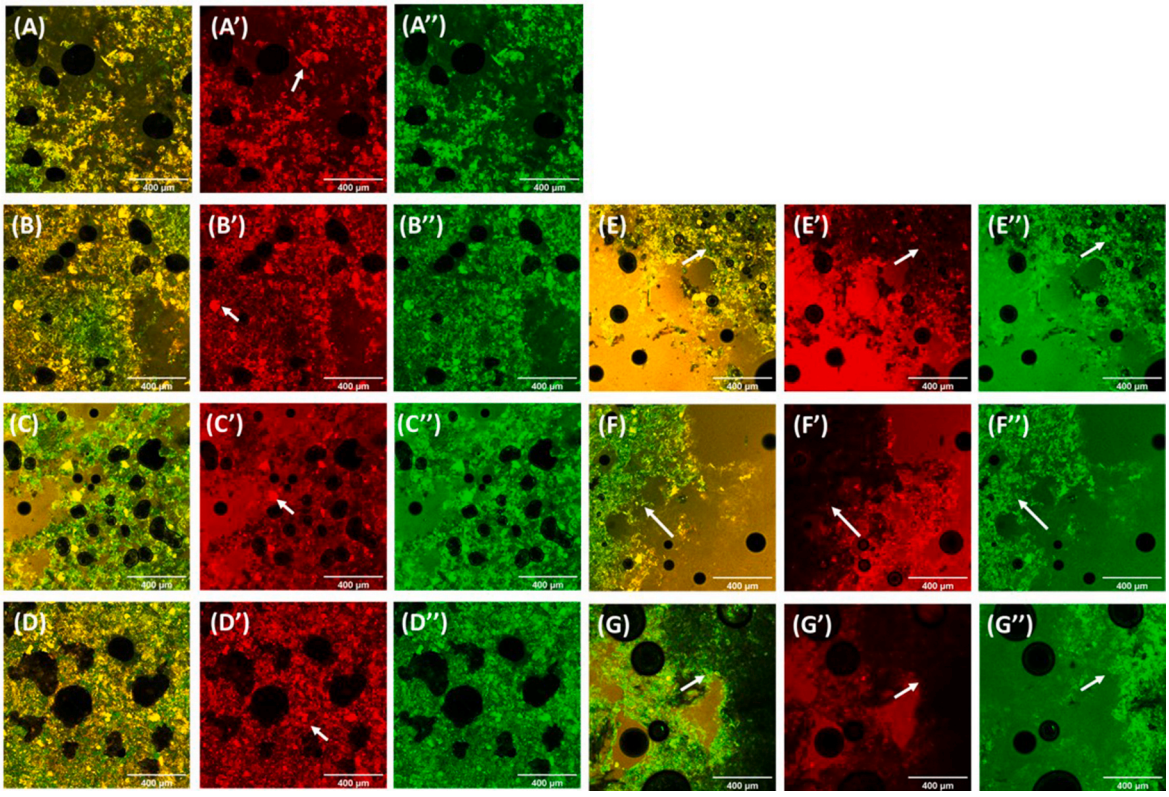

Fig. 6. Microstructure of the plant-based egg gels prepared without hydrocolloid addition (A; A'; A''), with κ-C at 0.1% (B; B'; B''), 0.3% (C; C'; C''), 0.5% (D; D'; D''), or with GG at 0.1% (E; E'; E''), 0.3% (F; F'; F''), 0.5% (G; G'; G'').
*Figures labelled with ' were from Rhodamine B channel, while those labelled with '' were from fluorescein isothiocyanate (FITC) channel.

from the synergism result. As for the GG systems, predominant phase separation occurred in rhodamine B channel (Fig. 6E'-G'), while FITC channel (Fig. 6E''-G'') showed the stained polysaccharides in the low-rhodamine-B-signal regions. This further supported the bi-continuous network in the GG gels, in line with the rheological synergy results.

3.6. Schematic diagram

From all these results, a schematic diagram was proposed (Fig. 7) based on the physiochemical properties-interaction-structure relationship. This diagram could explain the different gelling mechanism of κ-C and GG in the plant-based egg systems and the success of 0.3κ-C as egg analogue. For the gel systems, the physiochemical properties are determined by the structures, and structures are affected by interactions (Sow, Kong, & Yang, 2018). In the current study, hydrophobic interactions and hydrogen bonding were prominent in all plant-based systems, which was supported by the molecular interaction, surface hydrophobicity, and FTIR results. Addition of hydrocolloids increased electrostatic interactions within the systems (molecular interaction & zeta-potential results), mainly between protein (NH_3^+) and hydrocolloids ($-OSO_3^-$, $-COO^-$) or between K^+ crosslinked hydrocolloid chains. However, disulfide bonding was found to be not critical to the gel formation in both κ-C and GG systems as indicated by the molecular interaction and sulfhydryl measurement results. From the rheological synergism results, low concentration κ-C tends to interact with proteins and present the synergistic effects, followed by a predominant κ-C-κ-C network at high concentration (reduced synergism). While for GG systems, the K^+ induced GG-GG electrostatic interaction was more prominent due to the non-significant synergistic effect between GG and proteins.

Along with the different interactions discussed above, the structures of κ-C systems also differed from GG systems. The evenly distributed green and yellow regions observed under CLSM supported the development of mixed gels in 0.1–0.5κ-C samples, while the segregated color distribution in GG samples suggested the bi-continuous network formation.

Difference in gel structure contributes to the different physiochemical properties of our plant-based products. Although the textural properties of 0.3κ-C and 0.3 GG were both similar to LE, a pronounced difference was noted for gel strength, which could be attributed to the composite gel structures. The predominance of K^+-crosslinked GG-GG network was responsible for the lower gel strength than the predominant κ-C-protein network, where the swollen starch granule filled inside may further strengthen the gel. In addition, the stronger gel strength and less viscosity of the 0.3κ-C sample associated with its higher air holding capacity during frying, thus making 0.3κ-C the best match to LE in terms of specific volume.

4. Conclusion

In conclusion, a plant-based omelet contained 0.3% κ-C matched the gel strength and texture of the egg omelet successfully, with a specific volume closest to it. Such successful matching could be ascribed to the most κ-C-protein electrostatic interactions involved in 0.3κ-C, which eventually resulted in a composite gel network with starch filled in. However, 0.1% or 0.5% κ-C did not induce as predominant synergistic effect with proteins as 0.3% κ-C, mainly due to their inadequate or oversaturated κ-C content. Although 0.3 GG omelet showed comparable hardness with egg omelet, its specific volume and gel strength were significantly lower. The bi-continuous network structure resulted from the predominant GG-GG electrostatic interactions may be responsible for such macroscopic inadequacy. Different behaviors of κ-C and GG in the plant-based egg systems are associated with their different charge density and dynamic to interact with proteins; that in turn affects the microstructures as well as the physiochemical properties of the final omelet products. Although these promising results indicated that eggs

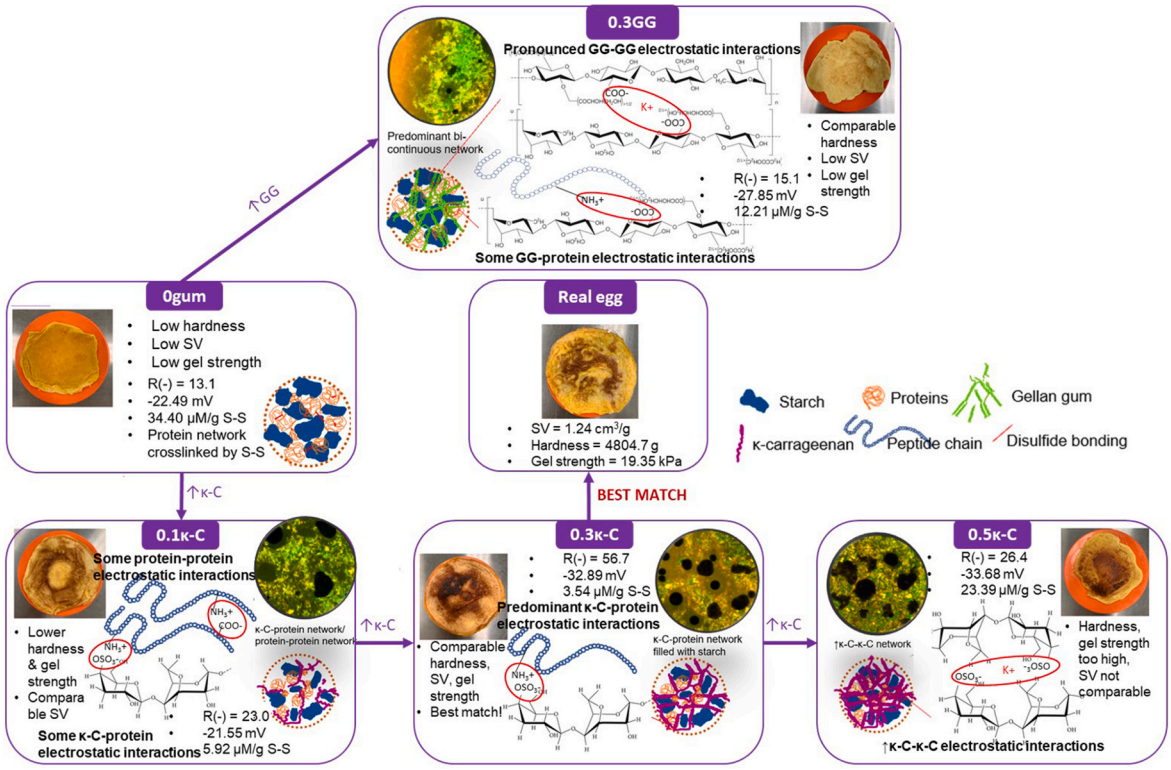

Fig. 7. Schematic diagram depicting the interactions and structure involved in the gelation of plant-based eggs.
*0gum, 0.1κ-C, 0.3κ-C, 0.5κ-C, 0.1 GG, 0.3 GG, and 0.5 GG refer to plant-based liquid egg with κ-C/GG addition of 0.0, 0.1, 0.3, 0.5% (w/w), respectively. *SV, specific volume.

could be replaced in omelet products in terms of gelation functionality, other functionalities of the liquid egg, such as emulsifying and foaming, and their potential applications in other eggless food systems merit further research. In addition, the plant-based liquid egg has less protein, lipid, and water content than the real liquid egg, which merits further modification on the nutritional profile. Despite the limitations in the current plant-based omelet products, this study can provide some insights in developing novel food systems with desirable physiochemical properties with the deepened understanding of protein-polysaccharide-starch interactions and the resulting structure development in the gelling systems.

Author statement

Zhou Lu: Conceptualization, Methodology, Investigation, Software, Visualization, Validation, Writing-original draft, Writing-review & editing. **Pin-Rou Lee:** Methodology. **Hongshun Yang:** Conceptualization, Funding acquisition, Project administration, Supervision, Writing - review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.foodhyd.2022.107691.

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