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Original Article

Binary egg white and collagen protein hydrogels: mechanism of formation, physicochemical properties, and digestibility

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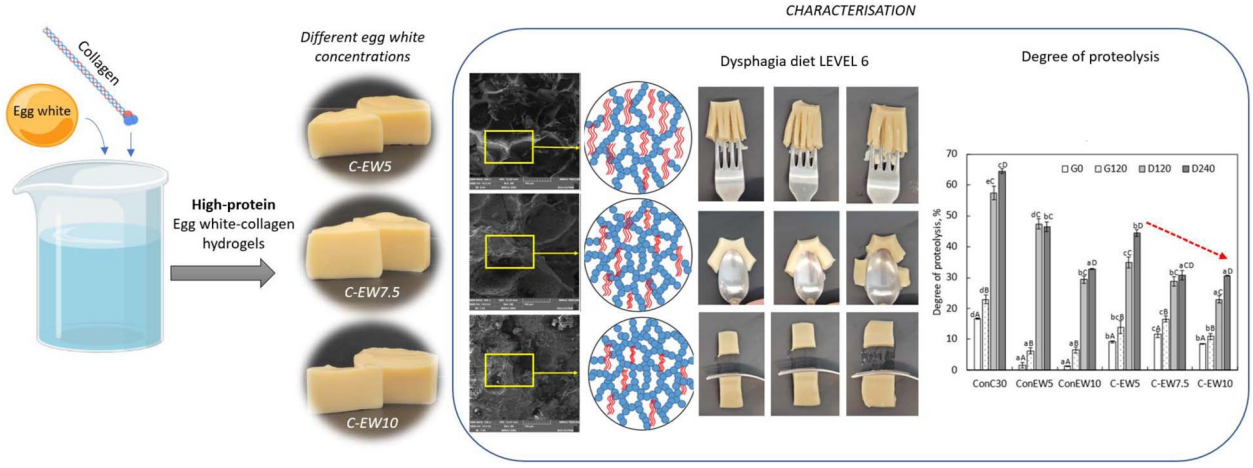
Abstract

Dysphagia is characterized by difficulty in swallowing and chewing. Texture-modified soft foods are intended for patients with dysphagia to facilitate safe swallowing and meet nutritional requirements, with a primary focus on increasing protein intake. This study examined the feasibility of creating high-protein (35%) gel systems suitable for dysphagia patients by adjusting the levels of egg white protein and collagen hydrolysate. It was observed that increasing the concentration of egg white and decreasing the amount of collagen improved the water-binding capacity (from 95.23% to 100%), hardness (from 0.85 N to 5.43 N), elasticity (higher G′ moduli), and network protein content (from 89.59% to 96.33%), while structural recovery declined (from 54.89% to 36.85%). Based on the International Dysphagia Diet Standardisation Initiative test, the hydrogels were classified as Level 6, fulfilling texture safety standards for dysphagia management. This classification makes them more appropriate for patients with mild dysphagia. Fourier-transform infrared spectroscopy, differential scanning calorimetry, and scanning electron microscopy analysis confirmed that the main network in the hydrogel composition is formed by egg white protein, with collagen serving more as an inactive filler. Hydrophobic interactions were dominant and primarily contributed to the stabilisation of the gel network. The protein hydrolysis rate in the hydrogels gradually declined as the egg white concentration increased, resulting in a denser, firmer, and less porous structure that may reduce enzyme interactions. These findings offer a viable approach for designing tailored high-protein food formulations for dysphagia.

Keywords dysphagia, high protein, food design, hydrogel

Graphical Abstract



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Introduction

Dysphagia is an increasing health problem among an ageing population, negatively affecting quality of life. Nutritionally and texturally modified foods are an essential strategy for developing foods for people with dysphagia (Liu et al., 2024). In this regard, soft gels could be used as manageable transitional foods for safe swallowing. While most of the studies focused on the composite gels made of the polysaccharide protein blends, (Lin et al., 2025a, 2025b; Li et al., 2025a, 2025b). We hypothesized that by using protein mixture gels, we could not only achieve high protein content, but also soft-textured gels that are safe to swallow with controlled digestive behaviour of proteins. Protein is an essential nutrient in the design of dysphagia food. Ageing reduces muscle synthetic capacity and protein absorption efficiency; therefore, it is recommended to consume foods high in protein (Gallego et al., 2022).

Egg white protein is widely known for its high biological availability and excellent amino acid score, with high levels of essential amino acids that stimulate muscle protein synthesis (Lee et al., 2024). This macrocomponent also exhibits excellent functional properties, such as foaming, emulsification, and gelation, the latter of which makes egg white essential for creating texture-modified products (Jalili-Firoozinezhad et al., 2020; Dong and Zhang, 2021). Previous studies utilized egg white protein in the formulation of potential dysphagia food in the form of 3D printed soy protein emulsion gels with egg white supplementation, (Zhao et al., 2025) hydrogel foam (Zhang et al., 2025) and cod protein composite gels prepared by using egg white-based microgels-high internal phase emulsions (Yu et al., 2024). All of these studies share the common feature that the total protein content in the systems was relatively low, with the exception of the study by Zhao et al. (2025), which reported 22%. In many cases, it may not be possible to significantly increase the protein content of the gel system, as this may result in gels that are too hard and brittle and no longer suitable for safe chewing and swallowing. For this reason, it was assumed that gels from different protein sources, one of which is non-structuring, could be used to develop tailored soft gels with the maximum possible protein content. Moreover, different protein sources can provide a greater variety of amino acids (Guo et al., 2022).

Collagen is a well-known connective tissue protein that, in the form of hydrolyzed, biologically active peptides, plays an important therapeutic and regenerative role in osteoporosis and osteoarthritis, enhancing immune function and participating in wound-healing and regeneration. It has also been reported that, compared to native collagen, hydrolyzed collagen is more easily digested and absorbed by the human body (Hajj et al., 2024) These benefits of collagen make it well-suited for inclusion in the diets of older adults with dysphagia. Composite gels composed of bovine tendon collagen and cassava starch were previously developed to meet the diverse needs of dysphagia patients, but with relatively low protein content (<10%) (Lin et al., 2025a, 2025b).

Thus, this study focused on exploring the feasibility of preparing functional, high-protein (35%) gel systems suitable for individuals with dysphagia by varying the concentrations of egg white protein (5%, 7.5%, and 10%) and collagen hydrolysate (CH) (30%, 27.5%, and 25%). In addition to stability and water-holding capacity (WHC) tests, textural, rheological, and International Dysphagia Diet Standardisation Initiative (IDDSI) analyses were conducted to assess the ability of the gels obtained to support dysphagia management. Molecular interactions were explored using Fourier-transform infrared (FTIR) spectroscopy, differential scanning calorimetry (DSC), scanning electron microscopy (SEM), and solubility analysis. Furthermore, we investigated how the in vitro protein digestibility of the hydrogels was affected by changes in the protein ratio.

Materials and methods

Materials

Collagen hydrolysate (bovine, I and III type mixture; 90% of protein; hydrolysis degree—20%) and egg white powder (EP) (78% of protein, 5.3% of carbohydrate, and 3.0% of salt) were obtained from MyProtein (Manchester, UK). Sodium azide was used as a bacteriostatic agent to prevent microbiological deterioration (Sigma-Aldrich, UK). KCl, KH_2PO_4, NaCl, $MgCl_2{\cdot}6H_2O$, $(NH_4)_2CO_3$, $CaCl_2{\cdot}2H_2O$, bile extract porcine (B8631), pepsin A (600–1,200 U/mg, 77,160), and pancreatin (P1625) were purchased from Sigma-Aldrich (Steinheim, Germany). All chemicals were of analytical grade.

Preparation of hydrogels

Three different solutions of egg white protein and CH at various concentrations were prepared (EW 5% and CH 30%; EW 7.5% and CH 27.5%; and EW 10% and CH 25%). The calculated amounts of CH, egg white proteins, and distilled water were weighed into a beaker using an analytical balance, then stirred with a magnetic stirrer for 60 min. The beakers were placed in a water bath and heated for 30 min at 85 °C to form gels, then cooled to room temperature and stored overnight in a refrigerator at 4 °C to allow complete structure formation. It was previously reported that after exposure to the heating of 60–90 °C, the secondary structure of the egg white protein was extensively denatured (Farjami et al., 2021). Therefore, we concluded that maintaining the gels at the specified temperature for 30 min allows the gel structure to fully develop.

Sodium azide (0.1% v/v) was used as a bacteriostatic agent during hydrogel preparation. The three different binary hydrogels were named C-EW5, C-EW7.5, and C-EW10. Increasing the egg white concentration further produced a very hard, brittle hydrogel that was not used for testing because it was visibly unsuitable for a dysphagia diet. Hydrogel samples were prepared with the same total protein content in the system (35% w/w).

For the protein hydrolysis experiment, control hydrogels were prepared under the same conditions as previously described, but with a different total protein content that fully matches the protein content of one of the proteins in the complex system (pure CH 30% w/w—ConC30%; pure egg white 5% w/w—ConEW5%; and pure egg white 10% w/w—ConEW10%).

Water-holding capacity

The WHC of the composite gels was measured according to the procedure outlined by Wang et al. (2024), with minor modifications. Weighed samples (W_i) were placed into the 30 ml centrifuge tubes and centrifuged (Labofuge 200, Thermo Scientific, Bracknell, UK) at 4,000× g for 20 min at room temperature. After centrifugation, the hydrogel was weighed again and recorded as W_f. The WHC was calculated

according to the following equation:

$$\text{WHC } (\%) = \frac{W_f}{W_i} \times 100 \quad (1)$$

where WHC is the water-holding capacity, %; W_i is the weight of the sample before centrifugation, g; and W_f is the weight of the sample after centrifugation, g.

Texture analysis

The textural properties, including hardness and cohesiveness, were measured by using a TA-XT plus texture analyser (Stable Micro Systems, TA.XT Plus, Godalming, England) with a P/05S probe. A hydrogel sample (cylindrical, 3 × 3 cm) was placed on a platform, and a cylindrical probe was inserted into the sample to a depth of 10 mm, at a compression speed of 1.0 mm/s, and a trigger force of 5 g.

Rheological properties

The rheological behaviour was evaluated using a controlled-stress rotational rheometer, MCR92 (Anton Paar, Graz, Austria), fitted with a 25 mm parallel plate (PP25) and a gap size of 1 mm. All measurements were carried out at a temperature of 25 °C.

Frequency sweep: the samples underwent a frequency sweep ranging from 0.1 to 10 Hz with a constant strain (1.0%).

Amplitude sweep: a strain sweep test was performed at a fixed frequency (1 Hz) and a strain range of 0.01%–1,000%.

The structural recovery: a strain-variation scanning mode was applied, with small-strain intervals (0.1%) for 600 s and large-strain intervals (50%) for 600 s, repeated three times (total duration: 1,800 s). The recovery rate was calculated by using the equation below (Equation 2):

$$\text{RR } (\%) = \frac{G'_{\text{at } 0.1}}{G'_{\text{at } 50}} \times 100 \quad (2)$$

where RR is the structural recovery rate, %; $G'_{\text{at } 0.1}$ is the initial storage modulus (G′) at 0.1% of the strain, Pa; and $G'_{\text{at } 50}$ is the storage modulus at 50% of the strain, Pa.

To evaluate rheological synergism, we conducted thermal cycle tests on hydrogels following the protocols outlined by Tanger et al. (2022). Silicone oil and solvent traps were used to prevent sample dehydration. The thermal cycles involved heating from 25 to 85 °C, holding at 85 °C for 10 min, cooling back to 25 °C, and holding at this temperature for another 10 min. The strain was set to 1%, with heating and cooling rates of 2 °C/min. The rheological synergism (R) among the egg white solution, collagen solution, and the obtained hydrogel systems was then calculated using the method described by Lu et al. (2022b):

$$\text{RS} = \frac{G'_{\text{mixture}} - \sum G'_{\text{ingredients}}}{\sum G'_{\text{ingredients}}} \quad (3)$$

where G′mixture represents the storage modulus of the composite system, and G′ ingredients refers to the storage modulus of each individual component measured at 25 °C after completing the thermal cycle (2).

IDDSI tests

The International Dysphagia Diet Standardisation Initiative framework is used for classifying dysphagia diets. Based on the appearance and texture properties of the formulated hydrogels, it was expected to be a grade 6–7 food; therefore, spoon-pressure, fork-pressure, and break-apart tests (fork and spoon cut tests) were performed. For testing, hydrogel samples were cut to dimensions of 1.5 × 1.5 cm.

Network and non-network proteins

A total of 2 g of gel was poured into 20 ml of distilled water and incubated for 48 hr in a water bath at 25 °C (GFL 1092, Thermolab, Hanover, Germany). The protein content in the soluble fraction released from the hydrogel was measured using the Bradford assay. Non-network proteins are calculated as the ratio of the soluble protein concentration to the initial protein concentration in the hydrogel. The remaining proteins are considered network proteins and are expressed as a percentage of the total protein content.

SEM analysis

For sample lyophilisation, an Alpha 1–4 LSC freeze dryer (Martin Christ, Germany) was used. Freeze-drying was carried out for 18 hr at 1 mbar, with the condenser temperature set to −55 °C. The microstructure of the samples was examined using a Tescan Mira3 XMU Scanning Electron Microscope (Czech Republic). The samples were mounted on SEM aluminium stubs and coated with a 5 nm gold layer using a Quorum Q150 sputter coater (Quorum Tech, UK). Imaging was performed at an accelerating voltage of 10 kV using a backscattered electron (BSE) detector at a working distance of 10 mm. Representative images were obtained at various magnifications 500× and 5,000×.

Thermal analysis

To determine the thermal properties of the samples, a Hitachi DSC 7020 (Japan) DSC was used. An empty aluminium pan was used as the reference. Nitrogen gas was employed as the purge gas at a flow rate of 40 cm^3 min^{-1}. Samples (~2 mg) were weighed into hermetically sealed aluminium pans, equilibrated at −30 °C, and heated from −30 to 300 °C at 10 °C min^{-1}.

FTIR spectroscopy

Lyophilized samples were diluted with KBr at a 1:10 ratio and homogenized. KBr pellets were prepared using a hydraulic press, and the hydrogel samples were analysed using a Bruker Tensor II FTIR spectrophotometer (USA). Spectra were recorded over 400–4,000 cm^{-1} with a resolution of 4 cm^{-1}. Measurements were carried out in percent transmittance mode.

Molecular interaction forces

Protein solubility (PS) mapping was performed to investigate the intermolecular interaction mechanisms in hydrogels following the description of Ge et al. (2022) with some minor modifications. A total of 2 g of hydrogel samples were soaked in 20 ml of different solutions (potassium phosphate buffer [PBS] (100 mM, pH 7.5) or PBS containing 50 mM dithiothreitol [DTT], 2 M thiourea, 8 M urea, or 8 Murea and DTT). The samples were shaken at a speed of 80 rpm (GFL 1092, Thermolab, Hanover, Germany) for 48 hr (at room temperature) and then centrifuged at 5,000 rpm (Labo-fuge 200, Thermo Scientific, USA) for 10 min. Bradford assay was used to determine protein content in the collected and filtered (through a 100-μm filter) supernatants. Protein solubility in different solvents was calculated using the following

equations (Equations 4 and 5):

$$\mathrm{PS}_{\text{in PBS solvent (\%)}} = \frac{C_1 \times V_{\text{solvent}}}{C_2 \times m_{\text{gel}}} \times 100 \quad (4)$$

$$\mathrm{PS}_{\text{in different solvents(exept PBS)}}\,(\%) = \frac{C_1 \times V_{\text{solvent}}}{C_2 \times m_{\text{gel}}} \times 100 - \mathrm{PS}_{\text{inPBS}}\,(\%) \quad (5)$$

where C_1 is the protein concentration (mg/ml) in the supernatant; C_2 is the protein content (mg/ml) in the hydrogel; m is the weight of the hydrogel (g); and V is the volume of solvent (ml).

In vitro digestion

Digestibility of hydrogel samples was evaluated using the INFOGEST static in vitro digestion model adapted to the older adult population, following the international consensus protocol described by Ménard et al. (2023). Control solutions of collagen (15% and 30% w/w) and egg white (5% w/w) were prepared, subjected to the same processing and digestion conditions as the hydrogel samples, and used for comparative analysis.

Digestions were conducted in a shaking water bath at 37 °C and 80 rpm (GFL 1092, Thermolab, Germany). Simulated digestive fluids were prepared to represent oral, gastric, and intestinal environments. Simulated salivary fluid (SSF) was prepared without amylase, while simulated gastric fluid (SGF) and simulated intestinal fluid (SIF) were used in combination with digestive enzymes. Pepsin was added at a concentration of 2,000 U ml^{-1} of digest during the gastric phase, while pancreatin (100 U ml^{-1} trypsin activity) and bile salts (10 mM) were introduced in the intestinal phase.

For the oral phase, 5 g of the sample was mixed with 5 g of SSF, and five glass beads were added to simulate oral processing. The mixture was incubated for 3 min. To initiate the gastric phase, 10 ml of SGF containing pepsin was added, the pH was adjusted to 3, and the mixture was incubated for 2 hr. For the intestinal phase, 20 ml of SIF containing pancreatin was added to the gastric digesta, the pH was adjusted to 7.0, and the mixtures were incubated for 2 hr.

Independent digestion vessels were prepared for each sampling time point. Samples were collected at different time points corresponding to the start and end of the gastric phase (G0 and G120) and during the intestinal phase (D120 and D240). To terminate pepsin activity, the pH of gastric samples was adjusted to 7.0, followed by immediate cooling in an ice-water bath. To terminate pancreatin activity, intestinal samples were directly cooled in an ice-water bath. Samples with inactivated enzymes were subsequently centrifuged at 6,000 rpm for 20 min at 4 °C (MPW-260RH, MPW Med. Instruments, Warsaw, Poland). The resulting supernatants were filtered, and the soluble fractions were stored at −18 °C until analysis.

The extent of protein digestion was assessed by determining proteolysis, calculated as the proportion of free α-amino groups released during in vitro digestion relative to the total α-amino groups measured after complete acid hydrolysis (6 M HCl, 24 hr). Free α-amino groups were quantified as leucine equivalents using an external standard curve (Jansson et al., 2014).

Statistical analysis

All data processing was carried out in Excel and SPSS 12.0 (StatSoft, Inc., USA), and the data were analysed using analysis of variance. The significance level was set at 5%. To ensure methodological reproducibility, three independent replicates were performed for each test, unless specified otherwise. Data are presented as mean values ± standard deviation.

Results and discussion

WHC and texture analysis

Water-holding capacity and texture are essential quality parameters that define the mouthfeel of food. Higher values of WHC ensure a greater sensation of moistness and juiciness, which is so important for patients with dysphagia. After increasing egg white concentration, a statistically significant ($p < .05$) increase from 95.23% to 100% in WHC was observed (Table 1). The increased WHC achieved by increasing the egg white concentration is associated with the formation of a denser, self-supporting network that is less sensitive to deformation under centrifugal force (Min et al., 2023a). However, increasing the proportion of egg white also resulted in a higher gel hardness (more than five times), due to the thermal denaturation temperature of egg ovalbumen (84 °C), which constitutes the largest portion of egg protein (Iesel et al., 2006). Generally, high hardness and a compact network structure correlate with higher WHC values in gels, (Min et al., 2023b) a finding confirmed by our study. However, while a higher WHC is associated with better lubrication of the food bolus, facilitating its passage through the esophagus and stomach, (Xie et al., 2021) higher hardness is associated with a greater force required to masticate semisolid foods into a swallowable bolus (Cai et al., 2026). High hardness is undesirable, as crushing such a hydrogel in the mouth requires greater tongue force and additional chewing, making it difficult to use for people with weakened masticatory muscles. Compared to the data of other authors, the hardness of the prepared hydrogels was very similar to dysphagia-oriented starch−protein−oil composite gels, (Cai et al., 2026) yam whole powder-based food, (Li et al., 2025a, 2025b) and much lower than in Alaska pollock surimi gels, (Liu et al., 2026) as well as in high-protein 3D printable whey protein−lotus root composite gels (Wang et al., 2025). Regarding cohesion, it was very low in all samples tested (−0.12 and −0.20), which is desirable because it facilitates the passage of the food bolus through the pharynx, but on the other hand, low cohesion reduces the internal binding necessary for proper food bolus formation, facilitating the breakdown of the hydrogel into small pieces, thus increasing the risk of aspiration (Zhang et al., 2024).

Rheological behaviour analysis

A comprehensive rheological analysis of hydrogel behaviour is essential for creating foods suitable for patients with dysphagia. Rheological techniques, such as frequency-sweep, amplitude-sweep, and structure recovery, help assess the structural properties of hydrogels and provide insights into how they change during chewing and swallowing.

Frequency-sweep tests were conducted over 0.1–10 Hz to better understand the structure of the obtained hydrogels (Figure 1A). The results showed that the G′ values for all tested gels were higher than the G″ values within the tested frequency range, indicating a dominant elastic behaviour. A clear concentration-dependent behaviour was observed as G′ and G″ moduli increased, suggesting a stronger gel network, consistent with other studies (Zhang et al., 2019). Both G′ and G″ showed an increasing trend with increasing frequency, consistent

Table 1 The water-holding capacity (WHC), hardness, and cohesiveness in hydrogels with different egg white and collagen concentrations.

	C-EW5	C-EW7.5	C-EW10
Image			
WHC, %	95.23 ± 1.43^{a}	98.04 ± 1.00^{b}	100.00 ± 0.00^{c}
Hardness, *N*	0.85 ± 0.14^{a}	2.58 ± 0.22^{b}	5.43 ± 0.32^{c}
Cohesiveness	-0.12 ± 0.01^{a}	-0.12 ± 0.01^{a}	-0.20 ± 0.03^{b}
Rheological synergism	−0.95	−0.93	−0.87

Values are reported as means ± *SD*; lowercase letters indicate significant ($p < .05$) differences in characteristics such as WHC, hardness, and cohesiveness between hydrogels with different egg white and collagen concentrations.

with findings in dysphagia-oriented surimi soft gels produced by enzymatic hydrolysis (Ding et al., 2025). The frequency dependence is mainly attributed to physical entanglement and non-covalent interactions within the gel structure (Qi et al., 2024).

An amplitude sweep analysis was performed to evaluate the mechanical behaviour of the hydrogel under large-amplitude oscillatory shear conditions, which closely mimic the sample's squeezing behaviour after tongue pressure (Ren et al., 2024). It was observed that G′ increased with higher egg white concentrations, indicating the formation of a denser, more rigid gel network. As shown in Figure 1B, the G′ and G″ modulus remained stable within the linear viscoelastic region (LVR) but gradually declined after reaching the critical strain, signalling the disruption of the network. It was previously reported that natural biopolymer gels exhibit enhanced structural stability and greater resistance to mechanical impact, with broad LVRs and critical strains ≥1%, (Hesarinejad et al., 2014) which was also confirmed by the results of our study. This is considered a positive outcome, as a critical force that is too low (not a stable hydrogel, even at low deformation) may cause the hydrogel to disintegrate before a food bolus forms in the mouth, leading to separate gel fragments that may enter the pharynx and cause choking (Cai et al., 2026). Furthermore, the LVR G′/G″ ratio was below 10, with values of 4.0, 4.5, and 5.0 for C-EW5, C-EW7.5, and C-EW10, respectively, aligning with the texture profile analysis results. This indicates that hydrogels can be considered weak gel systems that may be easier to consume even with reduced tongue strength.

To assess the hydrogels' capacity to regain their original properties under intense mechanical stress, repetitive amplitude fluctuations were applied (Figure 1C). Results showed that the hydrogels deform when subjected to external force, indicated by decreases in G′ and G″ after higher strain levels. This behaviour is favourable for their potential use in applications involving consumption, crushing, and bolus formation in the mouth. However, when the applied force is removed, the hydrogel gradually recovers some elasticity but does not fully return to its original shape. This indicates that formulated hydrogels undergo irreversible deformation and lose energy under higher stress. The observed recovery of structure decreased as egg white concentration increased, reaching 54.89%, 51.31%, and 36.85%, respectively. Improved hydrogel structural recovery ensures that it retains its shape after being chewed or compressed by the tongue, reducing the formation of food residues as it moves through the throat.

Network and non-network protein

Since hydrogels are composed of protein mixtures, it is important to assess their interactions and roles in structure formation. Understanding how these structures develop is crucial because it enables us to tailor hydrogel properties for specific applications. As expected, the protein content in the network increased with higher egg white content, while collagen content decreased accordingly (Figure 2), a pattern linked to the formation of egg white aggregates due to thermal denaturation (Zhang et al., 2025). In this context, it is probable that collagen functions more as a filler than as a component that provides structure, which could also be confirmed by the negative rheological synergism values (Table 1) that signify the absence of a synergistic effect between hydrocolloids (Lu et al., 2022a). Authors studying collagen-cassava starch composite gels found that collagen can act as a barrier, blocking water and preventing interactions between water and other biopolymers in the hydrogel composition (Lin et al., 2025b). The same phenomenon probably occurs in our study, which is seen as a positive aspect regarding texture and rheological properties.

The partial hydrolysis of collagen increases its effectiveness as a filler because, in this form, collagen molecules cannot form a self-supporting elastic gel, unlike pure collagen protein (Lin et al., 2025a, 2025b).

The results suggest a possible structure for hydrogels in samples with varying compositions (see Figure 2). The visualisation indicates that higher EWP concentrations lead to a denser, more uniform network with embedded collagen molecules. This is also confirmed by the significant ($p < .05$) increase in network protein content from 89.59% to 96.33%.

As collagen levels decline, its ability to interact with water and disrupt the egg protein network diminishes, thereby increasing hydrogel hardness and elasticity. This is further supported by the reduction of non-network proteins from 10.41% to 3.67%.

Microstructure analysis

To confirm our hypothesis about the formation of the protein structure, we examined the surface morphology of egg white protein CH hydrogel formulations using SEM images taken at different magnifications (500× and 5,000×). The SEM images revealed that the collagen-to-egg white ratio in the formulations clearly and consistently affected the microstructure (Figure 3). All samples exhibited an interconnected, sponge-like, porous structure similar to what was

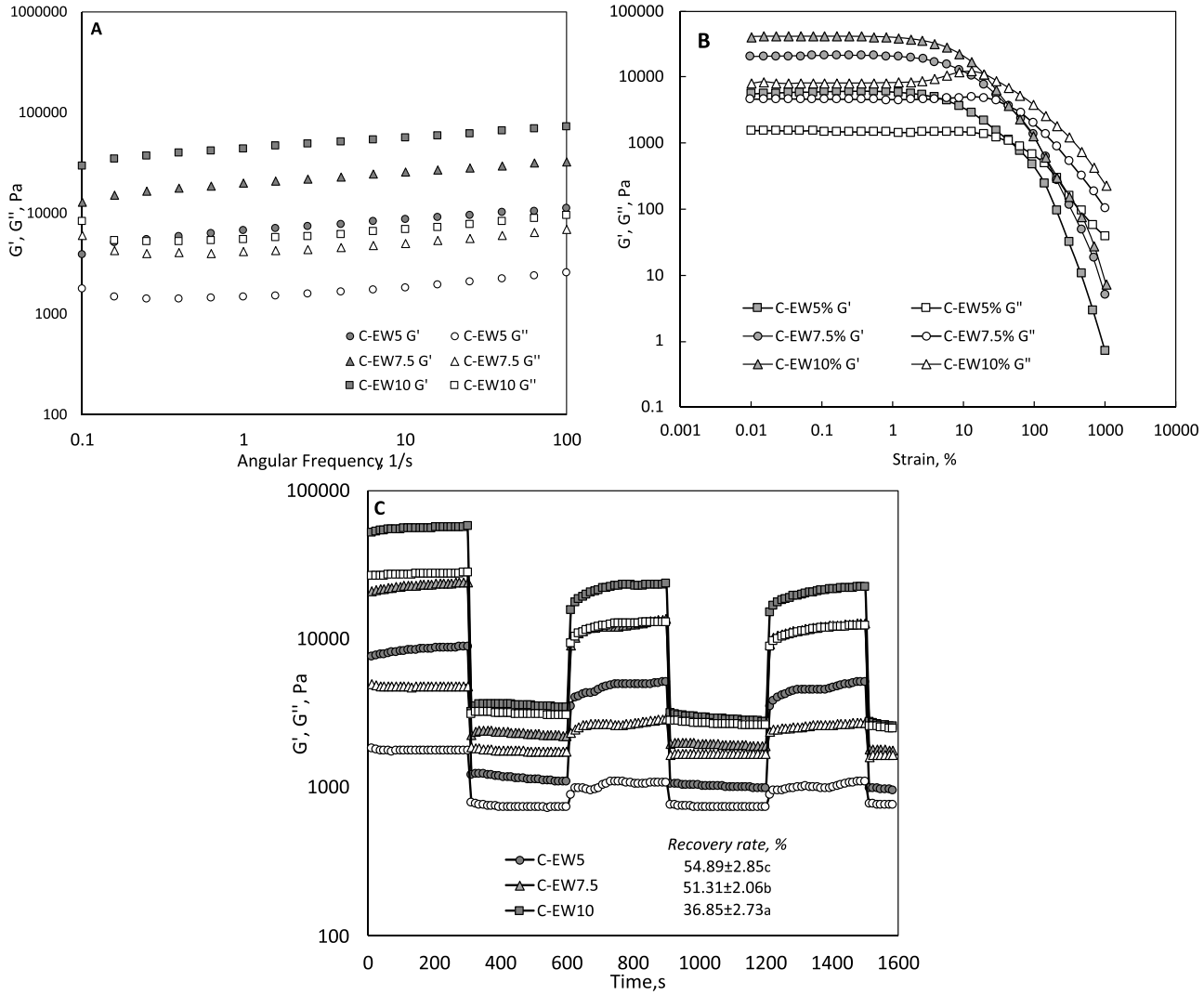


Figure 1 Frequency sweeps (A), amplitude sweeps (B), and structure recovery (C) of hydrogels formulated with different egg white and collagen hydrolysate concentrations.

previously reported by other authors (Yao et al., 2024). It is evident that samples with higher egg white concentrations have denser structures with fewer pores (easily visible in both low- and high-magnification images), which suggests increased cross-linking in the network, as well as in the previously reported study (Hu et al., 2026). This correlates with the greater hardness of these hydrogels,their higher WHC, and their lower structural recovery rate. Meanwhile, higher collagen content resulted in a fibril-dense network. These findings align with existing research indicating that increasing collagen concentration decreases pore size, while higher egg white protein levels lead to smoother, more compact pore walls (Duan et al., 2022; Martínez-García et al., 2022; Negrescu and Cimpean, 2025). These observations align with the frequency- and amplitude-sweep analyses, indicating that higher egg white concentration correlates with denser structural features.

DSC analysis

Differential scanning calorimetry analyses showed the thermal behaviour of collagen–egg white protein hydrogels (Figure 4). The thermograms displayed characteristic endothermic transitions from 50 to 90 °C, related to protein thermal events. These transitions correspond to the denaturation of egg white proteins, especially ovalbumen, and collagen (Figure 4). Thermal denaturation of collagen is usually reported between 55 and 70 °C (Friess, 1998), while egg white proteins often have multiple denaturation transitions between 70 and 85 °C (Donovan, 1977). The endothermic peaks observed in this study aligned with these literature values.

With increasing egg white protein content, a more pronounced endothermic peak intensity and an increase in peak enthalpy were observed in the DSC curves. This behaviour indicates that higher egg white protein concentrations promote more extensive protein–protein and protein–water interactions within the gel network, thereby enhancing overall thermal stability. Similarly, previous studies have reported that increased protein density in polysaccharide-containing protein hydrogels stabilizes the gel structure and raises denaturation temperatures (Nagasawa et al., 2004). Overall, the DSC findings demonstrate that the thermal stability of collagen–egg white protein hydrogels increases with egg white protein concentration, while the denaturation behaviour remains characteristic of collagen-

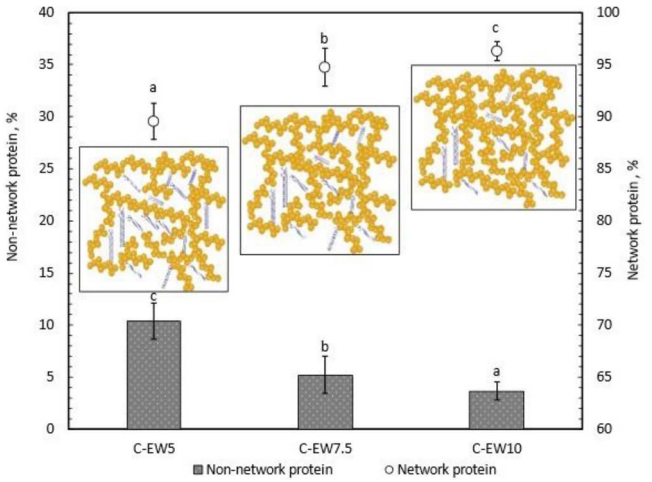


Figure 2 Network and non-network proteins of hydrogels formulated with different egg white and collagen hydrolysate concentrations. Values are reported as means ± *SD*; lowercase letters indicate significant ($p < .05$) differences in network and non-network protein content between hydrogels with different egg white and collagen concentrations.— —egg protein network; —collagen molecules. Structure images created by BioGDP.com.

and ovalbumin-related endothermic transitions. These results are consistent with earlier studies on the thermal properties of protein–polysaccharide-based hydrogels (She et al., 2024; Zuniga et al., 2021).

FTIR analysis

The FTIR spectra of binary egg white collagen hydrogel samples were analysed to evaluate their structural characteristics. Characteristic protein bands corresponding to amide I, amide II, and amide III regions were clearly observed. The amide I band (~1,650 cm^{-1}), associated with C=O stretching vibrations, reflects the secondary structure of collagen chains. Its presence in all samples indicates successful integration of collagen and egg white proteins within the hydrogel matrix. The amide II band (~1,540 cm^{-1}), corresponding to N–H bending and C–N stretching vibrations, intensified with increasing egg white protein content, suggesting stronger hydrogen bonding and enhanced interactions within the collagen matrix. The amide III band (~1,230–1,245 cm^{-1}) became more pronounced in the C-EW10 sample, indicating stabilisation of the collagen triple-helix structure by higher egg white protein content (Figure 5).

A broad band in the 3,300–3,400 cm^{-1} region was observed in all samples, attributed to O–H and N–H stretching vibrations and indicative of hydrogen bond formation. The increase in band intensity with increasing egg white protein content suggests enhanced hydrogen bonding and improved WHC. The C–H stretching bands at 2,800–2,950 cm^{-1}, associated with aliphatic groups, also increased with egg white protein concentration. Overall, the increase in amide band intensity and broadening of O–H/N–H bands from C-EW5 to C-EW10 indicate strengthened interactions between egg white proteins, consistent with the more compact morphology observed in SEM analyses.

Therefore, FTIR analyses confirm that increasing egg white protein concentration enhances the structural integrity of the hydrogel and improves the stability of the network structure. Similar findings have been reported in the literature. It has been previously demonstrated that changes in the position and intensity of amide bands in collagen–protein-based hydrogels are closely associated with network density and intermolecular chain interactions (Li et al., 2020). The pronounced appearance of amide bands in the FTIR spectra of egg white proteins reflects the active involvement of protein functional groups in the hydrogel network (Liu et al., 2018). Furthermore, it has been previously emphasized that the broadening of O–H and N–H bands in protein–polysaccharide hydrogels is directly related to the strengthening of hydrogen bonding (Yang et al., 2021b).

Intermolecular forces

To deepen the understanding of how the structure of the hydrogels forms, the intermolecular interaction forces within the samples were examined, as shown in Figure 6. The following order of dominant intermolecular forces was observed: hydrophobic interactions > hydrogen bonds > disulphide bonds. As was previously reported, heating denatures egg protein, causing its structure to unfold. This process exposes chemical groups, such as sulfhydryl, hydrophobic, and hydrogen-bonding groups, within the protein, (Wang et al., 2020) which may interact and form characteristic intermolecular bonds. Previous researchers have also observed that the dominant intermolecular forces vary with the type of protein gel. For example, the dominant intermolecular forces in egg white-dextran sulphate gels and thermally induced egg white gels were hydrophobic interactions, (Croguennec et al., 2002; Zhang et al., 2022) while ionic and disulphide bonds dominated in preserved egg white gel and marinated egg white gel (Xue et al., 2020; Zhao et al., 2016). As previously reported, collagen addition promotes hydrogen bonds and hydrophobic interactions between molecules, such as collagen-collagen and collagen-egg white (Yang et al., 2021b).

A substantial increase in the proportion of all dominated forces has been observed after the concentration of egg white has been increased with subsequent collagen reduction (Figure 6), which indicates the predominance of egg white proteins in the formation of the structure. Previously, a correlation between hydrophobic interactions and gel strength has also been reported, and intermolecular disulphide bonds have been associated with increased hardness, findings consistent with our results (Lin et al., 2025a, 2025b).

IDDSI tests

Predicting the suitability of hydrogel for dysphagia diets is difficult when evaluating each hydrogel's textural or rheological property separately. Therefore, standardized IDDSI tests (fork pressure, spoon pressure, and fork separation) were used to classify foods and assess swallowing safety. Additionally, the ability to pick up the hydrogel with fingers was tested to evaluate its structural integrity and disintegration during handling. It was found that all hydrogels required greater force to compress, causing the thumb to turn white, and that the gels did not return to their original shape after the pressure was released. This behaviour matches the level 6 definition (soft and bite-sized), indicating that tongue and palate force are not enough to crush the sample, and it should be slightly chewed before swallowing. This makes them more suitable for patients with mild dysphagia (Min et al., 2023a, 2023b). The C-EW5 sample exhibited the highest deformation, indicating that it is currently the softest (Figure 7). This was further confirmed by the fact that it crumbled between the fingers when picked up.

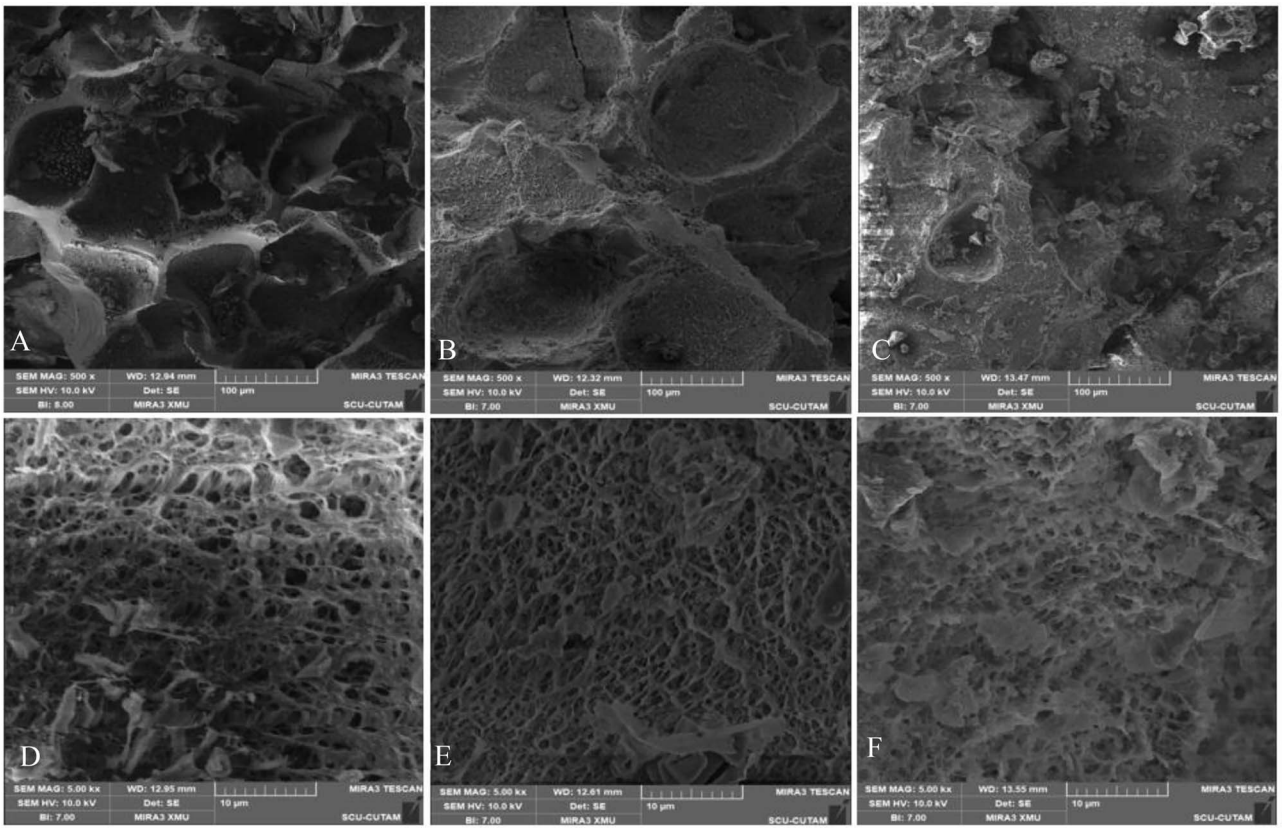


Figure 3 Scanning electron microscopy images of hydrogels formulated with different egg white and collagen hydrolysate concentrations at 500× magnification (A), (B), (C) and 5,000× magnification (D), (E), (F) in different formulations (A, D) C-EW5, (B, E) C-EW7.5, and (C, F) C-EW10.

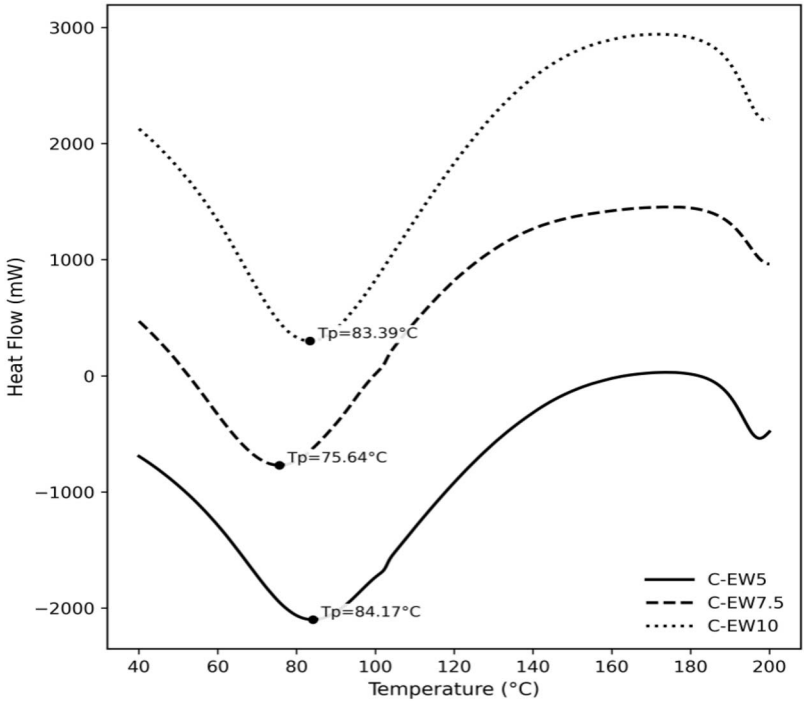


Figure 4 Differential scanning calorimetry thermograms of hydrogels formulated with different egg white and collagen hydrolysate concentrations: (A) C-EW5, (B) C-EW7.5, (C) C-EW10.

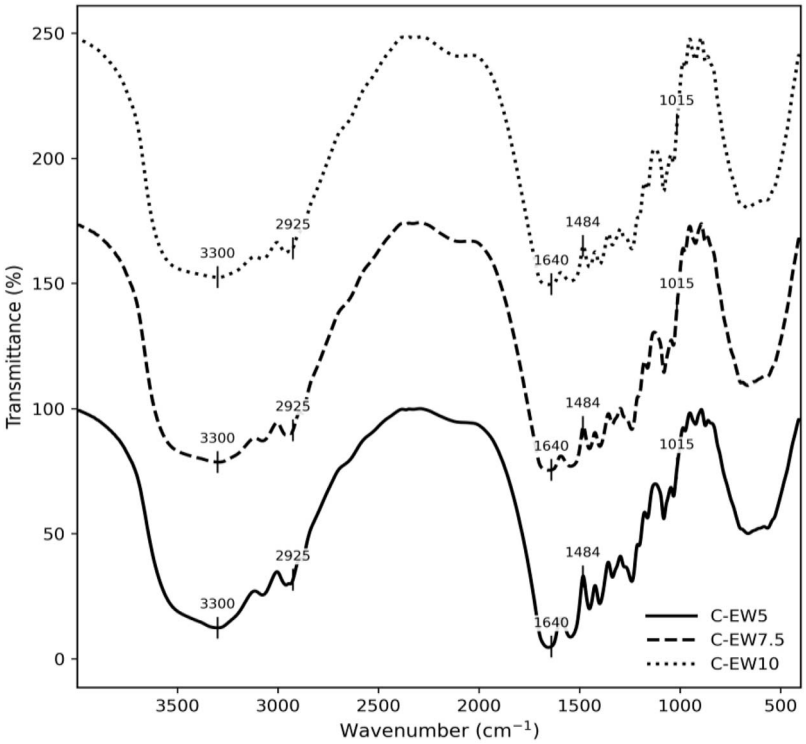


Figure 5 Fourier transform infrared spectra of hydrogels formulated with different egg white and collagen hydrolysate concentrations.

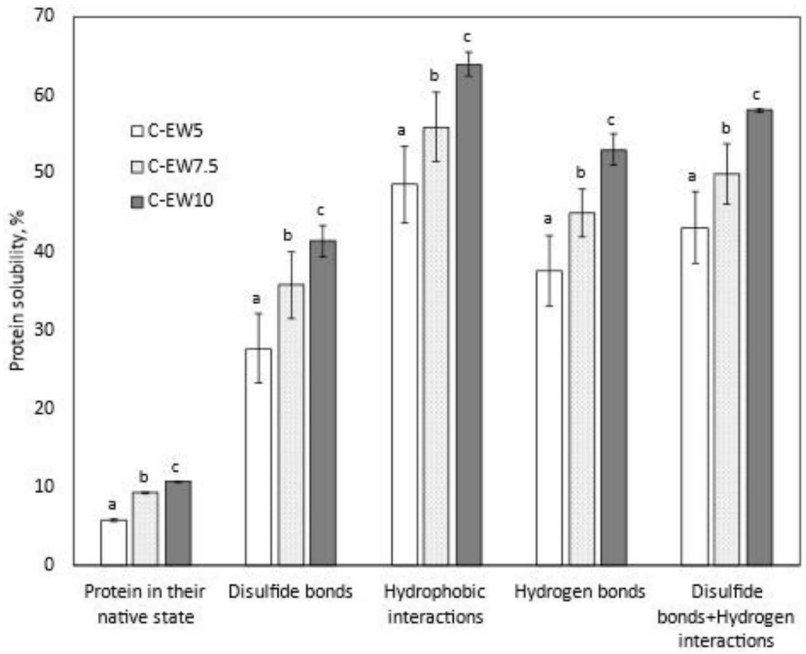


Figure 6 Interaction force analysis of hydrogels formulated with different egg white and collagen hydrolysate concentrations.

Compared to other studies that used egg white (Yu et al., 2024) or bovine tendon collagen(Lin et al., 2025a, 2025b) as parts of gel or emulsion systems for dysphagia, Levels of 5 and 6 were achieved, but their overall protein content was much lower. This demonstrates that combining gel-forming and non-gel-forming components can achieve a relatively high protein content while ensuring safe swallowing, aligning with our primary goal.

Degree of proteolysis

All tests indicate that, despite the high protein content of hydrogels, they may be suitable for dysphagia diets. However, an equally important factor is how these systems behave during digestion, especially the hydrolysis of proteins under simulated intestinal conditions. Besides hydrogel digestion, thermally treated collagen (30% w/w) and

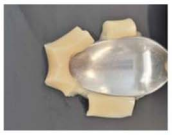
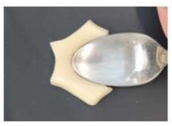
Samples	Fork pressure test	Spoon pressure test	Fork separation test	Disintegrate between fingers	Comment
C-EW5				Yes	Level 6
C-EW7.5				No	Level 6
C-EW10				No	Level 6

Figure 7 Fork pressure test, spoon pressure test, and fork separation test of hydrogels formulated with different egg white and collagen hydrolysate concentrations.

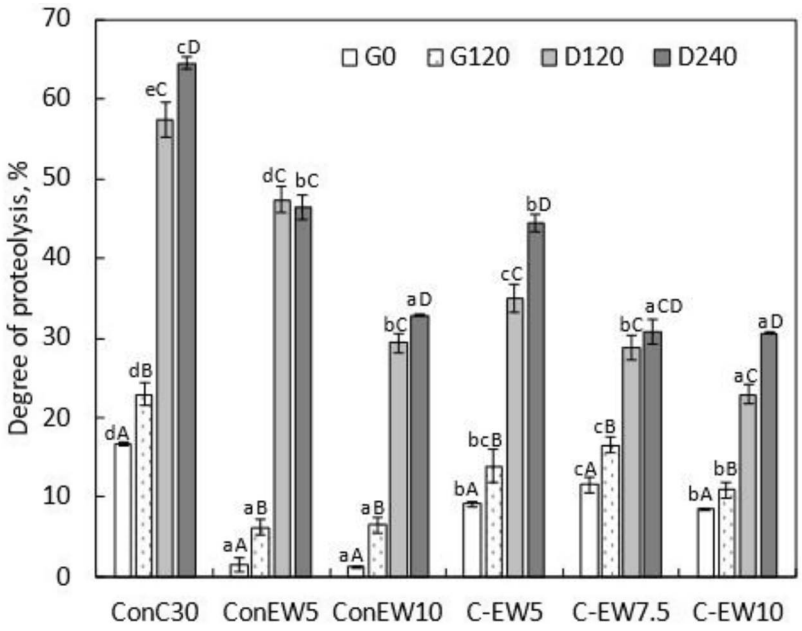


Figure 8 Extent of protein hydrolysis expressed as the degree of proteolysis of hydrogels formulated with different egg white and collagen hydrolysate concentrations. G0—gastric phase 0 min; G120—gastric phase 120 min; D120—intestinal phase 120 min; and D240—intestinal phase 240 min. Data labelled with different lowercase letters (A–D) showed statistically significant differences ($p < .05$) between different hydrogel formulations under the same digestion phase. Data labelled with different uppercase letters (A–B) showed statistically significant differences ($p < .05$) between different digestion phases for individual hydrogel formulation. ConC30—control collagen protein (30.0%w/w) sample; ConEW5—control egg white (5.0%w/w) sample; ConEW10—control egg white (10%w/w) sample.

egg white (5% w/w; 10% w/w) samples were also digested and used as controls. In control samples ConEW5 and ConEW10, the level of proteolysis in the gastric phase (G0−G120) stayed relatively low, while in the intestinal phase (D120-D240), it increased significantly, linked to the breakdown of the main egg protein ovalbumin by trypsin. Additionally, the high temperature used during hydrogel preparation inactivates natural trypsin inhibitors in egg white, which can also block hydrolysis (Bhat et al., 2021).

The study revealed that protein hydrolysis rates in hydrogels decreased gradually as egg white concentration increased, as shown in the digestion curve (Figure 8). Notably, at the end of the intestinal phase (D240), the proteolysis degree was 44.42% in C-EW5, 30.78% in C-EW7.5, and 30.66% in C-EW10. A similar decreasing trend was observed in control egg white samples (ConEW5 and ConEW10). This is attributed to the formation of a denser, less porous structure, confirmed by texture, rheological, PS, and SEM analyses. Such a compact network hampers enzyme diffusion and limits contact between proteins and enzymes, reducing access to hydrolytic sites (Zeng et al., 2023). Collagen, integrated into the hydrogel, might delay hydrolysis by acting as a physical barrier around egg proteins, (Lu et al., 2022b) or by binding enzymes in peptide form and inhibiting their activity (Uraipong & Zhao, 2018). Yet, this is contradicted by digestibility tests that show similar proteolysis in both the hydrogel and control samples, suggesting minimal or no collagen effect. Conversely, collagen molecules likely penetrate the egg white gel network, further reducing enzyme access, which is supported

by the higher digestibility of one collagen sample compared to others.

In summary, the hydrogels' high protein content resulted in 10.7%–15.5% of protein remaining in an available form after digestion, despite the relatively low degree of protein hydrolysis.

Conclusion

This study showed that changing the mixing ratio of egg white protein to CH significantly affects the gelling behaviour of the composite gel, influencing its network structure and texture. Structural analyses using FTIR, DSC, and SEM confirmed the successful formation of a network mainly stabilized by hydrophobic interactions. As the egg white concentration increased, the network became denser and more uniform, suggesting that egg white and collagen do not interact, which was further supported by a network/non-network protein ratio. Instead, collagen acts more like an inactive filler within the structure, which is widely believed to help increase protein content to 35%, maintaining its suitability for dysphagia-friendly diets (verified by the IDDSI test at Level 6). Although the designed networks slightly reduced the proteolysis rate, intestinal-phase digestibility was 44.42% in C-EW5, 30.78% in C-EW7.5, and 30.66% in C-EW10, due to steric hindrance and conformational masking of proteolytic sites. However, this indicates that about 10%–15% of the protein remains bioaccessible at the end of digestion.

This study provided valuable insights into developing protein-rich diets for people with dysphagia. However, further research is needed to examine how compositional differences affect sensory perception and comfort during consumption, as well as the long-term stability of the formulated products.

Data availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Author contributions

Deimante Dagyte (Formal analysis [equal], Investigation [equal]), Ieva Bartkuviene (Data curation [equal], Methodology [equal]), Viktorija Eisinaite (Supervision [equal], Writing—original draft [equal]), Evren Gölge (Formal analysis [equal], Investigation [equal]), Daiva Leskauskaitë (Project administration [equal], Supervision [equal]), and Rimante Vinauskiene (Investigation [equal])

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None declared.

Conflicts of 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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