Dynamic Rheological Properties and Microstructure of Partially Insolubilized Whey Protein Concentrate and Chicken Breast Salt-Soluble Protein Gels

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Viscoelasticity and microstructure of gels prepared by heating mixtures of chicken breast salt-soluble protein (SSP) and partially insolubilized whey protein concentrate (WPC) with solubilities ranging from 98.1 to 27.5% in 0.1 M NaCl, pH 7.0, were evaluated. Storage (G') and loss (G'') moduli of solutions containing 4% SSP, or combinations of 4% (w/w protein) SSP and 12% (w/w protein) WPC, were determined while heating from 30 to 95 °C or isothermally at 65 or 90 °C. The WPC altered the temperature and magnitude of G' and G'' transitions of SSP during heating. At 65 °C, insolubilized WPCs increased elasticity of SSP gels. Highly soluble WPC enhanced elasticity of combination gels more effectively at 90 °C. Microstructure of combination gels containing highly soluble WPCs was composed of a fibrous network of SSP at 65 °C and globules of WPC at 90 °C. Large denatured whey protein aggregates distorted the ordered SSP fibrous matrix of combination gels containing highly insoluble WPCs.

INTRODUCTION

The use of whey protein concentrates (WPCs) by the meat industry has not reached its full potential, due in part to variations in protein functionality (Harper, 1984) which result from wide differences in whey composition and processing conditions (de Wit and Klarenbeek, 1984; Morr and Foegeding, 1990). The capacities to bind water and form heat-induced gels are some of the important functional properties of whey proteins in processed meat formulations (Mulvihill and Kinsella, 1988).

Nonmeat proteins are added to processed meat formulations to improve yields, modify textural properties, and control costs. Nonmeat proteins are often selected on the basis of cost with little consideration of the specific functional properties they may impart. To optimize the use of nonmeat proteins in meat products and obtain desirable functional properties, it is necessary to understand the interactions that may occur between meat proteins and the added protein during processing.

During processing, the salt-soluble muscle proteins form a gel which is largely responsible for the yield and textural properties of the meat product (Acton et al., 1983; Smith, 1988). Nonmeat proteins may be dispersed in this muscle protein gel matrix to bind water, or they may gel and thus interact with the muscle proteins (Foegeding and Lanier, 1987) to form one of several types of complex or multicomponent gels (Tolstoguzov and Braduo, 1983; Morris, 1986; Brownsey and Morris, 1988). These multicomponent gels can impart different textural properties to the meat product, depending on the type of gel formed.

Myosin is the protein primarily responsible for gelation in meat systems, whereas β-lactoglobulin is the primary gelling protein in WPC. Chicken muscle myosin is a multidomain protein that denatures in the temperature range between 44 and 71 °C, depending on environmental conditions (Wang, 1993). The transition temperature of β-lactoglobulin as determined by differential scanning calorimetry is about 78 °C and may vary with environmental conditions (de Wit and Klarenbeek, 1984; Foegeding et al., 1992). When these two proteins are mixed, different types of gels can be formed depending on the processing temperature, solubility of the whey proteins, and environmental conditions, such as pH (Beuschel et al., 1992a).

The effects of WPC solubility on chicken salt-soluble protein (SSP) gel properties were determined by Beuschel et al. (1992a) at two different temperatures. Large differences in gel properties were observed due to heating temperature and WPC solubility. When combination gels containing SSP and WPC were heated at 65 °C, expressible moisture (EM) decreased, whereas hardness and deformability increased as WPC solubility decreased. At 90 °C, EM increased and hardness decreased as WPC solubility decreased in the combination gels (Beuschel et al., 1992a). These results suggested that WPC solubility and heating temperature could be manipulated to impart different rheological properties to gel systems.

The present study was conducted to further understand changes in rheological properties during formation of combination gels due to heating temperature and WPC solubility. Four WPCs with solubilities ranging from 27.5 to 98.1% in 0.1 M NaCl, pH 7.0, prepared under different time–temperature processing conditions were used in the experiments. The objectives were to monitor the viscoelastic behavior of solutions containing SSP and combinations of SSP and WPC during heating and to examine the microstructure of the heat-induced gels.

MATERIALS AND METHODS

Whey Protein Concentrate Preparation. Whey protein concentrates (WPCs) were prepared in triplicate and characterized by Beuschel et al. (1992b). Liquid whey protein concentrate from ultrafiltration of Parmesan cheese whey was obtained from Foremost Whey Products (Clayton, WI) and used to prepare four WPCs using different time–temperature processing conditions. The extent of protein insolubilization was measured and represented by percentage protein solubility in 0.1 M NaCl, pH 7.0 (Morr et al., 1985). One portion of the WPC was not heat treated and served as a control with a solubility of 98.1%. Whey protein concentrates with solubilities of 41.0 and 27.5% were obtained by heat treatments of 92.2 °C/30 s and

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0021-8561/93/1441-1372$04.00/0

126.7 °C/30 min, respectively. A mixture of the 98.1% soluble WPC and 41.0% soluble WPC was used to prepare a treatment with a solubility of 80.0%. Whey protein concentrates were freeze-dried and stored in low-density polyethylene freezer bags at −12 °C. The average composition of the WPC was 62.4% protein, 27.8% lactose, 5.7% fat, 5.4% moisture, 4.7% nonprotein nitrogen, 3.8% ash, and 0.47% calcium (Beuschel et al., 1992b).

Extraction of Chicken Breast Salt-Soluble Protein. Salt-soluble protein (SSP) was extracted from fresh chicken breast muscle purchased from a local grocery store as described by Wang et al. (1990) using two low-salt (0.1 M NaCl/0.05 M sodium phosphate buffer, pH 7.0) and one high-salt (0.6 M NaCl/0.05 M sodium phosphate buffer, pH 7.0) extraction. The supernatant from the high-salt extraction was diluted with 5 volumes of deionized water, followed by centrifugation at 21500*g* for 30 min. Protein concentration of the pellet was determined by Kjeldahl (AOAC, 1990). The SSP pellet was adjusted to 0.6 M NaCl by dissolving in one-third volume of 2.4 M NaCl/0.2 M sodium phosphate buffer, pH 7.0. This solution was diluted to contain 40 or 80 mg of protein/mL with 0.6 M NaCl/0.05 M sodium phosphate buffer, pH 7.0.

Solution Preparation. Whey protein concentrate (24% w/w protein) solutions were prepared as described by Beuschel et al. (1992a) in 0.6 M NaCl/0.05 M sodium phosphate buffer, pH 7.0. Solutions composed of 12% WPC protein (w/w protein) and 4% SSP (w/w protein) were prepared by mixing 24% (w/w protein) WPC solution with 8% (w/w protein) SSP at a 1:1 ratio. The protein solutions were blended with a Polytron homogenizer (Model PT 10/35, Brinkman, Westbury, CT) with a Model PTA 10 TS generator for three 3-s periods at a setting of 4 and allowed to stand overnight at 4 °C before using.

Dynamic Testing. Protein solutions containing 4% SSP or a combination of 4% SSP and 12% WPC in 0.6 M NaCl/0.05 M, pH 7.0, buffer were heated isothermally at 65 or 90 °C for 15 min or from 30 to 95 °C at a rate of 2 °C/min, and oscillatory dynamic measurements were recorded using a Rheometrics fluid spectrometer (RFS-8400, Rheometrics, Inc., Piscataway, NJ) equipped with a 25-mm-diameter parallel plate and 100-g transducer. Temperature was controlled with a programmable circulating oil bath (MTP-6 microprocessor, Neslab Instruments, Inc., Newington, NH). About 7 mg of protein solution was loaded in the sample cup, which was preheated to the desired temperature. The gap between the upper and lower plates was set at 1.5 mm. A few drops of corn oil (Mazola, Best Foods, CPC International Inc., Engelwood Cliffs, NJ) were added to cover the top of the protein solution to inhibit evaporation during testing. Limits of constant viscoelasticity were determined in preliminary experiments by conducting frequency (0.1–50 rad/s) and strain sweeps (0.001–0.5) at 65 and 90 °C using solutions of 4% SSP or a combination of 4% SSP and 12% WPC (98.1 and 27.5% soluble WPC) after heating for 15 min at 65 or 90 °C. Data parameters [storage modulus (G') and loss modulus (G'')] were recorded every 0.3 min at a fixed frequency of 10 rad/s and strain of 0.02.

Dynamic measurements of each treatment combination were replicated three times using a different batch of WPC and SSP. Transition temperatures in thermal scanning experiments were defined by the intersection of two regression lines determined from the slope on each side of each rheological transition.

Scanning Electron Microscopy. Gel specimens of 4% SSP or combination gels of 4% SSP and 12% WPC heated isothermally at 65 or 90 °C were prepared for scanning electron microscopy using glutaraldehyde fixation, ethanol dehydration, and critical point drying as described by Nuckles et al. (1991). Microstructure was observed with a JEOL scanning microscope (Model JSM-35CF, Osaka, Japan) at a voltage of 10–15 kV.

Statistical Analysis. Triplicate batches of each WPC and SSP in a completely randomized block design were used for all experiments. MSTAT (1990, Michigan State University, East Lansing, MI) was used to determine a two-way analysis of variance (replication × treatment) and separate means by Tukey's test using the mean square error term at the 5% level of probability.

RESULTS AND DISCUSSION

Thermal Scanning from 30 to 95 °C. Storage modulus (G'), which is an indication of the elastic character of a viscoelastic material, changed when SSP and SSP/WPC combination gels in 0.6 M NaCl/0.05 M phosphate buffer, pH 7.0, were heated from 30 to 95 °C (Figure 1). Storage modulus of SSP gels reached a maximum at 63.2 °C but on further heating decreased to a minimum at 69.1 °C. The G' increased again until 78.4 °C and then did not change, suggesting network formation by SSP was complete. The first G'' transition (Figure 2) occurred at a lower temperature than the first G' transition, probably because proteins unfolded and increased solution viscosity before subsequent network formation (Wang et al., 1990). A similar dynamic rheogram of SSP was reported by Wang et al. (1990); however, transition temperatures were about 10 °C higher in the present study, which may be attributed to a faster heating rate (Wu et al., 1991).

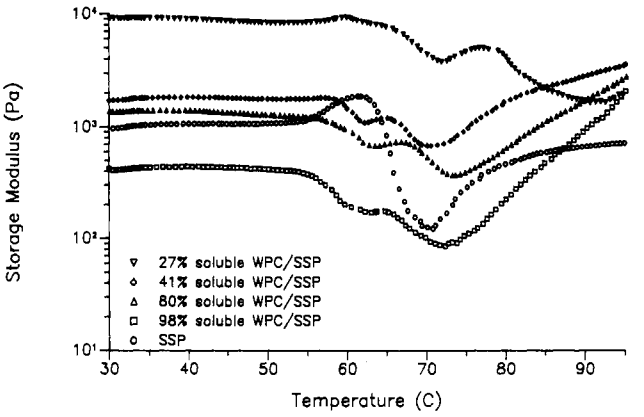

Figure 1. Representative rheogram illustrating the storage moduli (G') of 4% salt-soluble protein (SSP) and combinations of 4% SSP and 12% whey protein concentrate (WPC) heated from 30 to 95 °C.

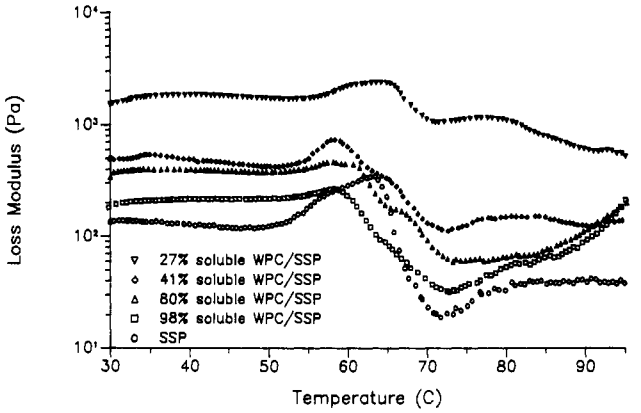

Figure 2. Representative rheogram illustrating the loss moduli (G'') of 4% salt-soluble protein (SSP) and combinations of 4% SSP and 12% whey protein concentrate (WPC) heated from 30 to 95 °C.

The SSP/WPC combination gels had different dynamic rheograms from SSP gels when heated from 30 to 95 °C. Addition of WPCs generally shifted the first G' and G'' transition to a higher temperature. The G' of combination gels containing 41.0, 80.0, or 98.1% soluble WPC did not change when heated from 30 to 58 °C and then decreased in the temperature range 58–72 °C. Whey proteins appeared to mask or interfere with gel network formation by SSP as the transition peak at 63.2 °C was not observed.

Storage moduli increased at a faster rate than G'' in 42, 80.0, and 98.1% soluble combination gels heated above 70 °C. At the end point of 95 °C, G' of all combination gels was significantly greater than that of SSP gels (Table I), suggesting the importance of whey proteins to final gel properties. The ratio of G'' to G' or tangent delta of all

Table I. Dynamic Moduli of Protein Solutions Containing 4% Salt-Soluble Protein (SSP) or Combinations of 4% Salt-Soluble Protein and 12% Whey Protein Concentrate (SSP/WPC) Heated from 30 to 95 °C in 0.6 M NaCl/0.05 M Sodium Phosphate Buffer

	dynamic moduli, Pa	
treatment[a]	initial (30 °C)	final (95 °C)
	Storage Modulus	
SSP	852.2[c,d]	940.5[d]
SSP/WPC 98	533.6[d]	2043.3[c]
SSP/WPC 80	1205.9[c]	2705.0[b]
SSP/WPC 41	1915.3[c]	3123.0[b]
SSP/WPC 27	10537.7[b]	2664.0[b]
	Loss Modulus	
SSP	119.9[c]	44.4[c]
SSP/WPC 98	207.9[c]	256.1[c]
SSP/WPC 80	336.4[c]	277.8[c]
SSP/WPC 41	456.1[c]	148.8[c]
SSP/WPC 27	2229.0[b]	577.2[b]

[a] Treatments indicated by percentage whey protein solubility in 0.1 M NaCl, pH 7.0. [b-d] Within each modulus means in columns with a different letter are significantly different ($P < 0.05$).

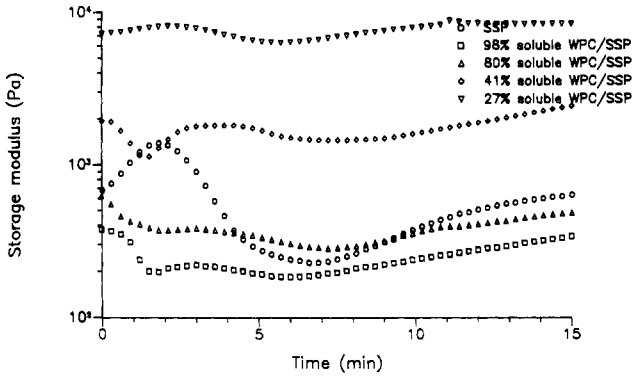

Figure 3. Representative rheogram illustrating the loss moduli (G') of 4% salt-soluble protein (SSP) and combinations of 4% SSP and 12% whey protein concentrate (WPC) heated at 65 °C for 15 min.

gels except the combination gel containing 27.5% soluble WPC was lower at 90 °C than at 30 °C, indicating the formation of an elastic cross-linked gel matrix (Beveridge et al., 1984).

The combination gel containing 27.5% soluble WPC had the highest G'' at 30 °C. This was probably due to the high water holding capacity of this WPC (Beuschel et al., 1992b). The G' and G'' of the combination gel containing 27.5% soluble WPC gradually decreased at temperatures above 77.7 °C. Dynamic moduli of the 27.5% soluble WPC combination gel were significantly higher than those of the SSP gel throughout heating. The ratio of G'' to G' was the same at 30 and 90 °C, suggesting little change in the viscoelastic properties of the gel matrix on heating. Denatured whey protein aggregates might have interferred with SSP cross-linking during network formation, distorting the SSP gel structure, which led to structural incompatibility and lack of gel matrix formation (Tolstoguzov and Braudo, 1983).

Isothermal Heating at 65 °C for 15 min. When SSP was heated isothermally at 65 °C, G' reached a maximum after 1.6 min of heating, decreased to a minimum of 195.3 Pa after 6.2 min, and then increased to 600.8 Pa after 15 min (Figure 3). The G'' of SSP followed a similar pattern during the first 5 min of heating but decreased to 33.1 Pa after 15 min (Figure 4). Addition of whey proteins masked the G' SSP transition peak, although small transitions in G'' were noted during the first minute of heating in 41.0, 80.0, and 98.1% soluble WPC combination gels.

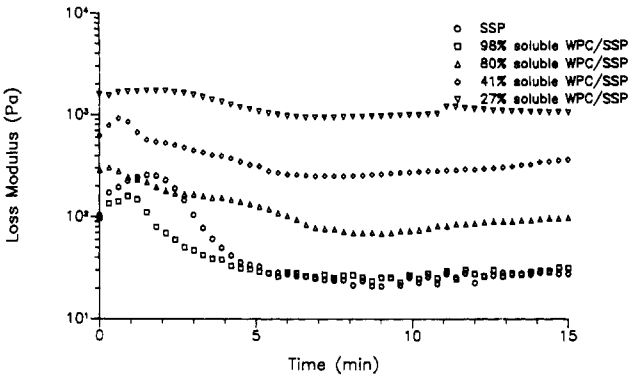

Figure 4. Representative rheogram illustrating the loss moduli (G'') of 4% salt-soluble protein (SSP) and combinations of 4% SSP and 12% whey protein concentrate (WPC) heated at 65 °C for 15 min.

Table II. Dynamic Moduli of Protein Solutions Containing 4% Salt-Soluble Protein (SSP) or Combinations of 4% Salt-Soluble Protein and 12% Whey Protein Concentrate (SSP/WPC) Heated Isothermally at 65 °C for 15 min in 0.6 M NaCl/0.05 M Sodium Phosphate, pH 7.0

	dynamic moduli, Pa	
treatment[a]	initial	final
	Storage Modulus	
SSP	633.4[d]	600.8[d]
SSP/WPC 98	416.5[d]	320.3[d]
SSP/WPC 80	693.6[d]	574.7[d]
SSP/WPC 41	1693.7[c]	1839.7[c]
SSP/WPC 27	8725.3[b]	8023.7[b]
	Loss Modulus	
SSP	112.9[d]	33.1[c]
SSP/WPC 98	167.7[d]	36.3[c]
SSP/WPC 80	243.1[d]	116.0[c]
SSP/WPC 41	606.2[c]	236.7[c]
SSP/WPC 27	1623.7[b]	811.6[b]

[a] Treatments indicated by percentage whey protein solubility in 0.1 M NaCl, pH 7.0. [b-d] Within each modulus means in columns with a different letter are significantly different ($P < 0.05$).

Storage moduli of combination gels containing 27.5 and 41.0% soluble WPC were higher ($P < 0.05$) than those of 80.0 and 98.1% soluble WPC combination gels and the SSP gel after isothermal heating at 65 °C for 15 min (Table II). Results of dynamic tests coincided with that of failure compression tests on the same material reported by Beuschel et al. (1992b). When heated to 65 °C, combination gels prepared with highly soluble WPC were less firm than gels prepared with more insoluble WPC.

At 65 °C whey proteins do not form a gel (Beuschel et al., 1992b; Hung, 1992); thus, SSP was probably solely responsible for the protein network of combination gels. This type of combination gel matches the model of a "filled gel" (Tolstoguzov and Braudo, 1983; Oakenfull, 1987; Ziegler and Foegeding, 1990; Lanier, 1991) with whey proteins acting as the filler.

Combination gels containing highly solubilized WPCs (80.0 or 98.1% soluble WPCs) might form a type I filled gel, in which the filler (WPC) remains soluble in the interstitial fluid of the SSP gel matrix (Ziegler and Foegeding, 1990; Lanier, 1991). The addition of highly solubilized WPCs seemed to "dilute" the SSP or interfere with SSP gelation, resulting in gels with a lower G' than an additive relationship (due to increased protein concentration) would predict (Burgarella et al., 1985). Lanier (1991) suggested that soluble fillers might increase the viscosity of the interstitial fluid and increase the G'' value; however, Hermansson and Akesson (1975) reported that highly soluble whey proteins do not swell and impart low

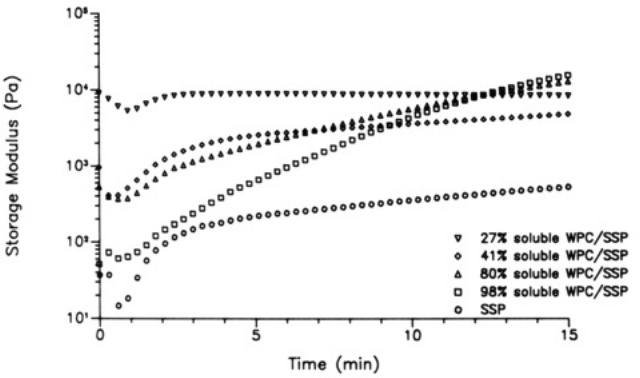

Figure 5. Representative rheogram illustrating the storage moduli (G') of 4% salt-soluble protein (SSP) and combinations of 4% SSP and 12% whey protein concentrate (WPC) heated at 90 °C for 15 min.

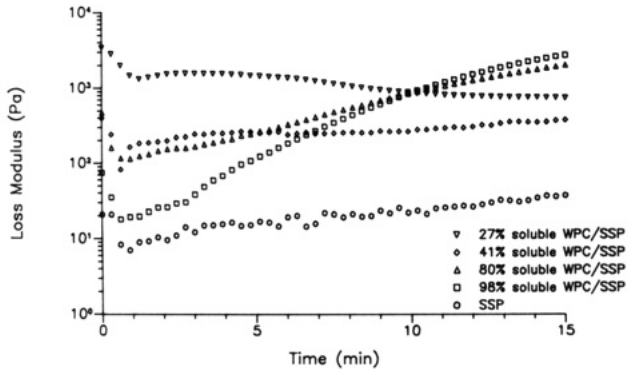

Figure 6. Representative rheogram illustrating the loss moduli (G'') of 4% salt-soluble protein (SSP) and combinations of 4% SSP and 12% whey protein concentrate (WPC) heated at 90 °C for 15 min.

viscosities to solutions. This could explain the similar G'' values observed in the SSP and highly soluble WPC combination gels.

Combination gels containing highly insolubilized WPCs (27.5 or 41.0% soluble WPC) might act like a type II filled gel, in which the filler (insolubilized WPC aggregates) exists as dispersed particles within the SSP gel matrix (Ziegler and Foegeding, 1990; Lanier, 1991). The insolubilized whey proteins absorbed water and concentrated the continuous protein phase (SSP). In addition, whey proteins occupied the interstitial spaces within the gel matrix and reinforced the gel, causing an increase in G' and G'' as compared to those of SSP alone.

Isothermal Heating at 90 °C for 15 min. When SSP was heated isothermally at 90 °C, G' decreased in the first minute of heating and then increased during the remainder of the heating period (Figure 5). Typical G' and G'' transitions between 55 and 69 °C in the thermal scanning experiment did not appear when SSP was heated isothermally at 90 °C (Figure 6). The G' of combination gels containing 41.0, 80.0, or 98.1% soluble WPC decreased during the first 30 s of heating and then increased throughout the heating period. Similar trends were observed for G'' under the same conditions, but the effect upon G'' was smaller. There was a 1-min lag time between the time sample was put in the sample cup and data collection; thus, only the last part of the transition was detected. The G' and G'' of 27.5% soluble WPC/SSP combination gel decreased during isothermal heating at 90 °C.

The combination gel containing 98.1% soluble protein had the highest G' of 15490.0 Pa after 15 min at 90 °C. The G' of combination gels was about 10–23 times higher than

Table III. Dynamic Moduli of Protein Solutions Containing 4% Salt-Soluble Protein (SSP) or Combinations of 4% Salt-Soluble Protein and 12% Whey Protein Concentrate (SSP/WPC) Heated Isothermally at 90 °C for 15 min in 0.6 M NaCl/0.05 M Sodium Phosphate, pH 7.0

	dynamic moduli, Pa	
treatment[a]	initial	final
	Storage Modulus	
SSP	41.4[d]	660.7[e]
SSP/WPC 98	124.9[d]	15490.0[b]
SSP/WPC 80	350.2[d]	9976.0[c]
SSP/WPC 41	1056.0[c]	6688.0[d]
SSP/WPC 27	8701.0[b]	8553.1[c]
	Loss Modulus	
SSP	16.9[c]	41.9[e]
SSP/WPC 98	145.5[c]	2645.0[b]
SSP/WPC 80	338.8[c]	1455.0[c]
SSP/WPC 41	318.7[c]	766.3[d]
SSP/WPC 27	2303.0[b]	805.7[d]

[a] Treatments indicated by percentage whey protein solubility in 0.1 M NaCl, pH 7.0. [b–e] Within each modulus means in columns with a different letter are significantly different ($P < 0.05$).

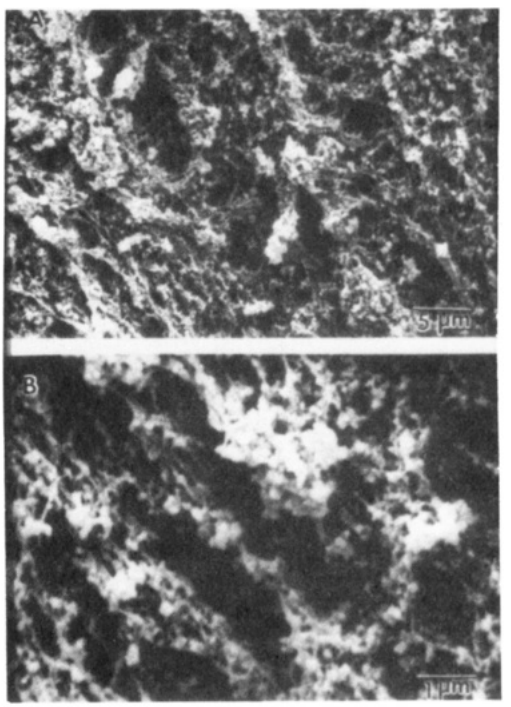

Figure 7. Scanning electron micrographs of 4% salt-soluble protein (SSP) gels prepared in 0.6 M NaCl/0.05 M phosphate buffer, pH 7.0, heated at 65 °C for 15 min. (A) Bar length equals 5 μm; (b) bar length equals 1 μm.

that of the SSP gel (Table III). Combination gels containing WPC had greater G' and G'' than an additive relationship would predict when compared to WPC and SSP heated individually at 90 °C (Hung, 1992). The higher the solubility of WPC in the combination gels, the greater the dynamic moduli increased during the 15-min heating period, suggesting soluble whey proteins were responsible for the increased dynamic moduli of combination gels.

Heat had different effects on the dynamic moduli of combination gels prepared with solubilized and insolubilized whey proteins. The dynamic moduli of combination gels containing more solubilized whey proteins were greatly increased by heating, suggesting participation of the WPC in the gel matrix. In contrast, the combination gels containing more insolubilized whey proteins had

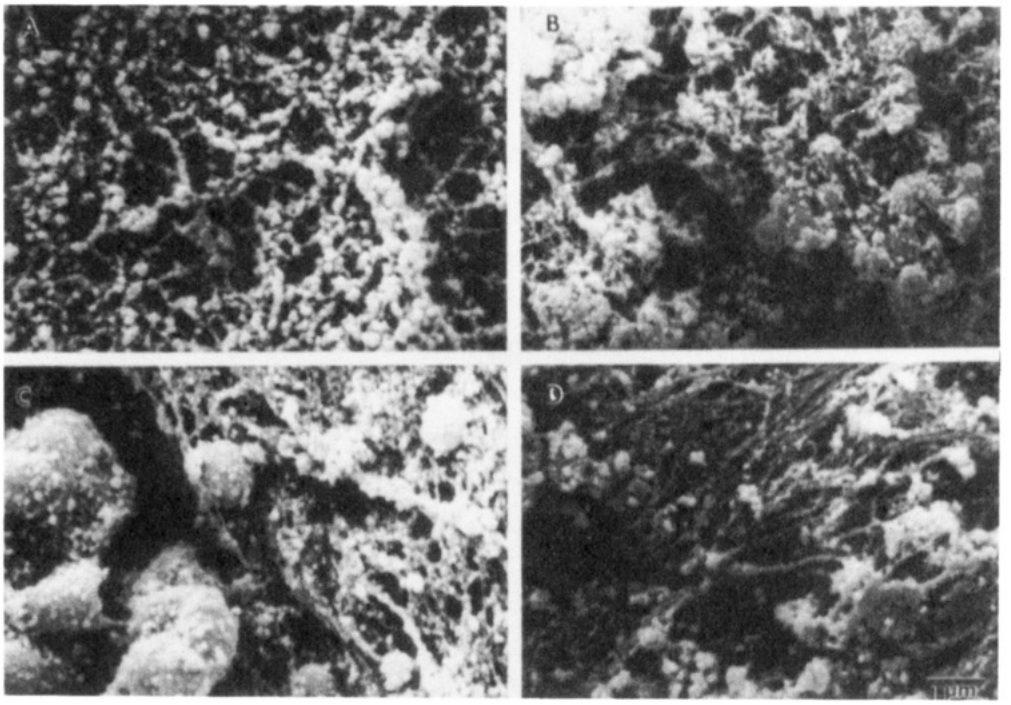

Figure 8. Scanning electron micrographs of 4% salt-soluble protein (SSP) and 12% whey protein concentrate (WPC) combination gels prepared in 0.6 M NaCl/0.05 M phosphate buffer, pH 7.0, heated at 65 °C for 15 min. (A) 98.1% soluble WPC/SSP combination gel; (B) 80.0% soluble WPC/SSP combination gel; (C) 41.0% soluble WPC/SSP combination gel; (D) 27.5% soluble WPC/SSP combination gel. Bar length equals 1 μm.

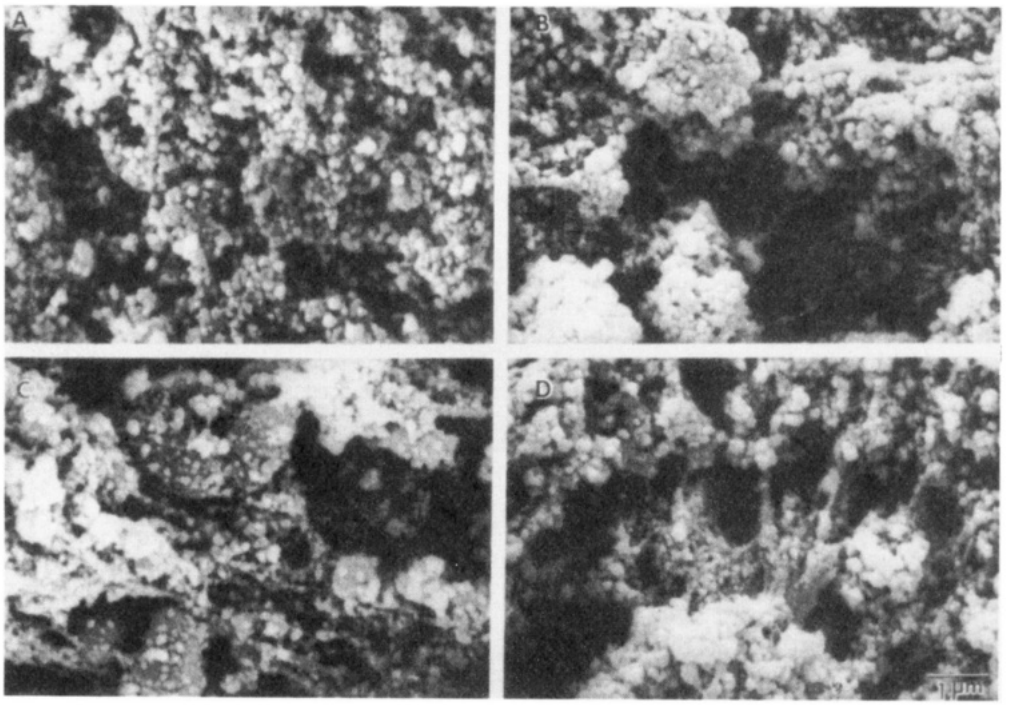

Figure 9. Scanning electron micrographs of 4% salt-soluble protein (SSP) and 12% whey protein concentrate (WPC) combination gels prepared in 0.6 M NaCl/0.05 M phosphate buffer, pH 7.0, heated at 90 °C for 15 min. (A) 98.1% soluble WPC/SSP combination gel; (B) 80.0% soluble WPC/SSP combination gel; (C) 41.0% soluble WPC/SSP combination gel; (D) 27.5% soluble WPC/SSP combination gel. Bar length equals 1 μm.

higher initial dynamic moduli but exhibited smaller increases during heating.

When the soluble whey proteins formed a gel on isothermal heating at 90 °C, they could interact with SSP to form a "coupled network" as described by Oakenfull (1987) and Brownsey and Morris (1988). Alternatively, whey proteins might gel within the interstitial spaces of the already formed SSP gel network and form a "phase-separated network" as described by Oakenfull (1987) and Brownsey and Morris (1988). The marked increase in dynamic moduli of the combination gel containing 80.0 and 98.1% soluble WPC could be explained as resulting from either of the gel models described above.

The G' values of insolubilized WPCs were higher than those of the more soluble WPCs in combination gels prior to heating. Insolubilized whey proteins may have competed for available water and increased the SSP concentration in the continuous phase (Lanier, 1991). Insolu-

bilized whey proteins could precipitate as part of the entrapped particulate fraction within the SSP gel network to form a "type II filled gel" (Ziegler and Foegeding, 1990; Lanier, 1991).

Microstructure. The microstructure of 4% SSP gels, in 0.6 M NaCl/0.05 M phosphate buffer, at pH 7.0, heated isothermally at 65 °C for 15 min showed a fine three-dimensional filamentous network, consisting primarily of beaded strands with some aggregated areas (Figure 7). A similar SSP filamentous gel matrix has been reported previously (Wang and Smith, 1992; Hermansson et al., 1986). Hung (1992) reported that highly soluble WPC gels (16% w/w protein) heated at 90 °C for 15 min contained grapelike globular clusters 0.6–1-μm diameter as previously observed by Beveridge et al. (1984), Hermansson (1986), and Aguilera and Kessler (1989). As WPC solubility decreased, size and distribution of globular aggregates within gels became larger and more variable in size (Hung, 1992).

When heated to 65 °C, the filamentous beaded strand SSP gel network was evident in the 98.1% soluble WPC combination gel and no whey protein globular aggregates were observed (Figure 8A). Since whey proteins do not gel at 65 °C, they might have remained soluble and been washed out when gels were fixed for scanning electron microscopy. Both the SSP fibrous network and small whey protein globular aggregates were visible in the 80.0% soluble WPC combination gel (Figure 8B). Whey protein aggregates were evenly distributed and were embedded within the SSP gel matrix. The G' values of SSP gel and the 80.0 and 98.1% WPC combination gels after heating for 15 min at 65 °C were similar, which emphasizes the importance of the SSP network in the combination gels made from soluble concentrates. The 41.0% soluble WPC combination gel appeared to contain two different phase-separated protein networks (Figure 8C). In one network, small whey protein aggregates with a diameter of 1.0 μm were embedded within the cross-linked SSP network. In the other network, the microstructure was composed of large aggregated globules of 5.0-μm diameter coated with smaller globules. In the 27.5% soluble WPC combination gel (Figure 8D), SSP fibers were oriented in parallel strands which were interspersed with large whey protein aggregates. Addition of insoluble WPC appeared to act as fillers to reinforce the SSP network as G' of these gels were greater than that of SSP alone. Thus, observations of gel microstructure support conclusions made from the rheological data.

Globular structures, characteristic of whey protein gels, were observed in microstructures of combination gels containing 80.0 or 98.1% soluble WPC (Figure 9A,B) when heated isothermally to 90 °C for 15 min. The typical SSP filamentous network was not observed. Some weblike fibrous threads interspersed throughout the gel structure were observed in the 80.0% soluble WPC combination gel (Figure 9B). These microstructures suggest the importance of whey protein gelation in combination gels heated above the denaturation temperature of β-lactoglobulin. Microstructures of 27.5 and 41.0% soluble WPC combination gels heated at 90 °C contained the SSP fibrous network and aggregated whey protein globules of various sizes (Figure 9C,D). These gels had higher G' values than SSP gels but lower G' values than highly soluble combination gels. Breakdown of the SSP network due to excessive heating and incomplete matrix formation by insolubilized whey proteins might have resulted in less elastic gels.

ACKNOWLEDGMENT

Acknowledgement is made to the National Dairy Promotion and Research Board and the Michigan Agricultural Experiment Station for support of this research.

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Received for review January 21, 1993. Revised manuscript received April 27, 1993. Accepted May 20, 1993.