Effect of Processing Temperature on Detergent-Solubilized Protein in Extrusion-Cooked Cornstarch/Soy Protein Subunit Blends

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The effects of extrusion processing temperatures (50, 100, 150 °C) on protein solubilized by phosphate buffers (with and without sodium dodecyl sulfate) were examined. The study was done utilizing cornstarch/soy protein fraction model food systems. All samples containing β-conglycinin had the highest levels of protein solubilized by phosphate buffer at all processing temperatures. There was about a 5-fold increase in the level of phosphate buffer insoluble protein solubilized by SDS–phosphate buffer when unprocessed samples were compared with those processed at 50 °C. Virtually all added protein (more than 0.9 g of protein/g of added protein) was solubilized by SDS–phosphate buffer in the samples containing β-conglycinin. Samples containing soy protein isolate or glycinin had lower levels of protein solubilized by both phosphate buffers.

INTRODUCTION

Soy protein is very often used in food systems to achieve various functional characteristics. Studies on thermal processing effects on soy proteins have been reported by various researchers (Bikbov et al., 1981; Schmidt, 1981; Hermansson, 1985; German et al., 1982; Damodaran and Kinsella, 1982) and reviewed by Kilara and Sharkasi (1986). They all show that, following denaturation, the proteins will often interact either with themselves or with other molecules.

Tanteeratarm et al. (1990) found that the binding capacities of the two major soy globulins were significantly different. They showed that β-conglycinin's binding capacity was superior compared to that of glycinin. Binding capacity is likely to be due to the chemical interaction nature of the individual proteins. One of the protein–protein or protein–polymer interactions that commonly occurs is noncovalent binding. Sodium dodecyl sulfate is frequently used for the determination of these noncovalent interactions (Hager, 1984; Sureschchandra et al., 1987; Chandra et al., 1984; Shimada and Cheftel, 1988; Burgess and Stanley, 1976).

Therefore, the purpose of this study was to determine the influence of extrusion processing temperature on SDS-solubilized protein in cornstarch/soy protein subfraction model food systems.

MATERIALS AND METHODS

Materials. The ingredients consisted of commercial cornstarch (Pure-dent B700 (a gift from Grain Processing Corp., Muscatine, IA)) and commercial soy protein isolate (Ardex R (a gift from Archer Daniels Midland Co. (ADM), Decatur, IL)). Soy protein subfractions were isolated from soy protein isolated according to the method of Saio et al. (1973). A 1:1 β-conglycinin/glycinin blend was obtained by mixing equal parts of β-conglycinin and glycinin samples.

Cornstarch Preparation. A 2-kg starch sample was mixed with 188 mL of 0.08 N sodium hydroxide solution for 10 min using a Hobart Model D-300 mixer (The Hobart Manufacturing Co., Troy, OH) set at the lowest speed. The prepared sample had 25% w/w moisture and a measured pH of 7.0. Samples were sealed in plastic bags and left at room temperature for extrusion processing the following day.

Protein Pellet Preparation. A total of 2 mL of 0.3 N sodium hydroxide was mixed with 2.5 g of soy protein isolate, and a round pellet was manually made. The prepared pellet had a measured pH of 7.0. These pellets were stored overnight at room temperature in sealed plastic containers to prevent moisture loss for extrusion processing the following day.

Extrusion Processing. Extrusion thermal processing was performed in a single-screw Brabender Plasticorder extruder, Model PL-V500 (C. W. Brabender Instruments, Inc., South Hackensack, NJ), equipped with a variable D-C drive unit, a tachometer, and a torque meter. The extruder barrel had a diameter of 19.05 mm with a 20:1 length:diameter ratio and eight 0.79 × 3.18 mm longitudinal grooves. In addition to a tapered screw with a 3:1 compression ratio, a die with a diameter of 4.8 mm was used.

The temperature of the extruder barrel was controlled by two electrically heated zones. The first heating zone was in the compression section and the second one in the metering section just next to the die. The dough temperature was maintained by compressed-air-cooled barrel collars. The compressed air was controlled by thermostats found inside the barrel wall. The preconditioned starch was fed manually into the feed zone of the extruder while the screw speed was brought to the processing speed of 120 rpm. After a steady-state flow, as shown by a torque variation of ±2.5 in.-lb, had been maintained and the dough temperature just before the die (either 50, 100, or 150 °C) had been obtained, an experimental protein pellet was introduced in the feed zone. Immediately after a pellet was introduced, the extrudate strand was cut off and discarded. The subsequent exiting extrudate strand, which contained the protein sample, was cut off and saved. The presence of protein was determined by observing the difference in color of the exited strand.

Protein Solubilization by Phosphate Buffer. To determine the amount of protein that was not involved in either protein–protein or protein–starch interactions, 0.35 M phosphate buffer at pH 7.0 (phosphate buffer) was used in the solubility study. The phosphate buffer extracted the free (unbound) protein from the experimental samples.

A 0.100-g portion of unprocessed or ground extruded sample was added to 10 mL of phosphate buffer in a 15-mL beaker and covered with parafilm. The sample was stirred using a magnetic stirrer for 60 min, then transferred into a 15-mL centrifuge tube and centrifuged in a Dynac centrifuge (Clay Adams, Parsippany, NJ) at 1000g for 10 min. The supernate was removed and the protein content determined according to the Mitchell (1972) method.

To calculate the amount of solubilized protein per gram of added protein (S), the equation

$$S = (P_{SB} - P_{SS})/(P_{TP} - P_{TS})$$

was used, where P_{SB} is the solubilized protein (grams) per gram of thermally processed cornstarch/soy protein blend, P_{SS} is the solubilized protein (grams) per gram of thermally processed cornstarch, P_{TB} is the amount of protein (grams) per gram of

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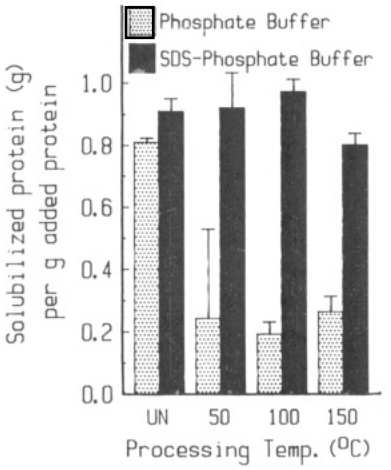

Figure 1. Effect of processing temperature on buffer-solubilized protein in cornstarch/β-conglycinin extrudates. Samples were extruded at 120 rpm screw speed and 25% w/w feed moisture. "UN" signifies unprocessed samples.

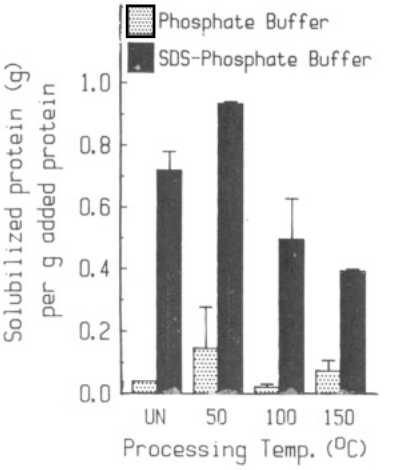

Figure 2. Effect of processing temperature on buffer-solubilized protein in cornstarch/glycinin extrudates. Samples were extruded at 120 rpm screw speed and 25% w/w feed moisture. "UN" signifies unprocessed samples.

thermally processed cornstarch/soy protein blend, and P_{TS} is the amount of protein (grams) per gram of thermally processed cornstarch sample.

Protein Solubilization by Detergent–Phosphate Buffer. Sodium dodecyl sulfate (SDS) at a level of 0.01 M in the same phosphate buffer as above was used in this solubility study. SDS was used to disrupt protein–protein or protein–starch complexes produced by noncovalent interactions. The solubility methodology described above was also used here.

Statistical Analysis. The data obtained from all experimental results were statistically evaluated using analysis of variance utilizing the SPSS statistical package.

RESULTS AND DISCUSSION

Extrusion thermal processing, even at the relatively low temperature of 50 °C, influenced the amount of protein solubilized by phosphate buffer. Samples containing β-conglycinin had the highest level of protein solubilized by phosphate buffer for unprocessed samples (Figure 1). At 50 °C, the level of protein solubilized by phosphate buffer increased for samples containing glycinin (Figure 2), but there was a reduction for those containing β-conglycinin (Figure 1) or 1:1 β-conglycinin/glycinin (Figure 3) fractions. No observable change was found with samples containing soy protein isolate fraction (Figure 4). At 100 °C, the level of protein solubilized by phosphate buffer was reduced for all samples in the study (Figures 1–4). There

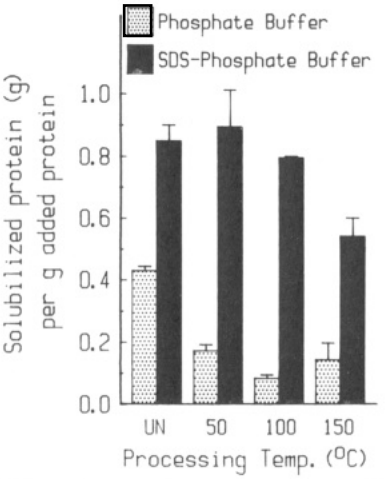

Figure 3. Effect of processing temperature on buffer-solubilized protein in cornstarch/1:1 β-conglycinin:glycinin extrudates. Samples were extruded at 120 rpm screw speed and 25% w/w feed moisture. "UN" signifies unprocessed samples.

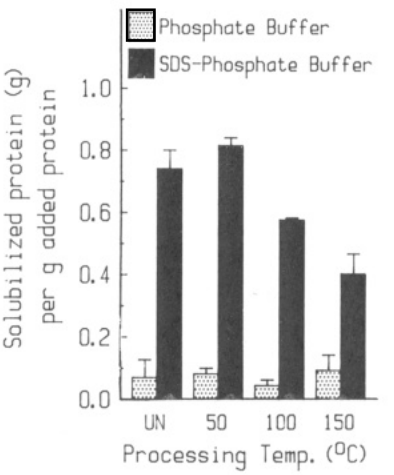

Figure 4. Effect of processing temperature on buffer-solubilized protein in cornstarch/soy protein isolate extrudates. Analyzed samples were extruded at 120 rpm screw speed and 25% w/w feed moisture. "UN" signifies unprocessed samples.

was again an increase in the level of phosphate buffer solubilized protein for all samples at 150 °C. Increased protein solubility with phosphate buffer might have been due to the thermal cleavage of certain susceptible peptide polypeptides at this extrusion temperature.

The amount of phosphate buffer insoluble protein, which was solubilized by SDS–phosphate buffer, increased greatly for samples containing β-conglycinin (Figure 1). There was an increase from about 0.1 to 0.7 g of solubilized protein/g of added protein when levels of phosphate buffer insoluble protein, solubilized by SDS–phosphate buffer of unprocessed sample, were compared with that of samples that had been processed by extrusion at 50 °C. As the temperature was increased, the level of protein solubilized by SDS–phosphate buffer was reduced.

Only the temperature linear main effect was shown to be statistically significant at a p value of 0.000, while the soy protein–temperature interactions were not significant ($p > 0.05$).

The results also showed that β-conglycinin was capable of mainly forming noncovalent bonds, since more than 90% of sample protein was solubilized by phosphate buffer containing SDS at all processing temperatures. β-Conglycinin has been shown to be deficient in cysteine or cystine side chains (Hermansson, 1985; Koshiyama, 1971). Therefore, the major interactions were likely to be non-

covalent in nature. The thermally induced noncovalent interactions within the cornstarch/β-conglycinin model food system were not conclusively identified as either protein–protein or protein–starch interactions. It was safe to assume that both kinds of interactions took place in this particular model food system. At temperatures above 100 °C, the formation of some covalent bonds may take place, thereby reducing the level of protein solubilized by SDS–phosphate buffer but not necessarily reducing noncovalent interactions. This may explain why there was a reduction in the level of protein solubilized by SDS–phosphate buffer at these temperatures.

On the contrary, thermal processing caused a reduction in the level of protein solubilized by SDS for samples containing other experimental proteins (glycinin, β-conglycinin/glycinin, and soy protein isolate) (Figures 2–4). The highest reduction was observed in blends containing glycinin (Figure 2), followed by those containing soy protein isolate (Figure 4). It appears that other interactions, which could not be disrupted by SDS, took place in addition to noncovalent interactions. Some of these interactions might have included disulfide bonds as suggested by (Nakai and Li-Chan, 1988; Sheard et al., 1986).

Statistical analysis by ANOVA tests showed that the temperature quadratic main effect was significant with a p value of 0.02. The soy protein–temperature interactions were not significant ($p > 0.05$).

LITERATURE CITED

Bikbov, T. M.; Grinberg, V. Y.; Schmandke, H.; Chaika, T. S.; Vaintraub, I. A.; Tolstoguzov, V. B. Study on gelation of soybean globulin solutions, viscoelastic properties and the structure of thermotropic gels of soy bean globulins. *Colloid Polym. Sci.* **1981**, *259*, 536–547.

Burgess, L. D.; Stanley, D. W. A possible mechanism for thermal texturization of soybean protein. *Can. Inst. Food Sci. Technol. J.* **1976**, *9*, 228–231.

Chandra, B. R. S.; Rao, A. G. A.; Rao, M. S. N. Effect of temperature on the conformation of soybean glycinin in 8 M urea or 6 M guanidine hydrochloride solution. *J. Agric. Food Chem.* **1984**, *32*, 1402–1405.

Damodaran, S.; Kinsella, J. E. Effect of conglycinin on the thermal aggregation of glycinin. *J. Agric. Food Chem.* **1982**, *30*, 812–817.

German, B.; Damodaran, S.; Kinsella, J. E. Thermal dissociation and association behavior of soy proteins. *J. Agric. Food Chem.* **1982**, *30*, 807–811.

Hager, D. F. Effects of extrusion upon soy concentrate solubility. *J. Agric. Food Chem.* **1984**, *32*, 293–296.

Hermansson, A. M. Structure of soy, glycinin and conglycinin gels. *J. Sci. Food Agric.* **1985**, *36*, 822–832.

Kilara, A.; Sharkasi, T. Y. Effects of temperature on food proteins and its implications on functional properties. *CRC Crit. Rev. Food Sci. Nutr.* **1986**, *23*, 323–395.

Koshiyama, I. Subunit structure of a 7S protein in soybean globulins. *Agric. Biol. Chem.* **1971**, *35*, 385–392.

Mitchell, H. L. Microdetermination of nitrogen in plant tissues. *J. Assoc. Off. Anal. Chem.* **1972**, *55*, 1–3.

Nakai, S., Li-Chan, E., Eds. In *Hydrophobic Interactions in Food Systems*; CRC Press: Boca Raton, FL, 1988; pp 129–177.

Saio, K.; Watanabe, T.; Kaji, M. Food use of soybean 7S and 11S proteins. Extraction and functional properties of their fractions. *J. Food Sci.* **1973**, *38*, 1139–1144.

Schmidt, R. H. Gelation and coagulation. In *Protein Functionality in Foods*; Cherry, J. P., Ed.; ACS Symposium Series 147; American Chemical Society: Washington, DC, 1981; pp 131–147.

Sheard, P. R.; Fellows, A.; Ledward, D. A.; Mitchell, J. R. Macromolecular changes associated with the heat treatment of soya isolate. *J. Food Technol.* **1986**, *21*, 55–60.

Shimada, K.; Cheftel, J. C. Texture characteristics, protein solubility, and sulfhydryl group/disulfide bond contents of heat-induced gels of whey protein isolate. *J. Agric. Food Chem.* **1988**, *36*, 1018–1025.

Sureshchandra, B. R.; Rao, A. G. A.; Rao, N. S. M. Effect of sodium dodecyl sulfate on the conformation of soybean glycinin. *J. Agric. Food Chem.* **1987**, *35*, 244–247.

Tanteeratarm, K.; Wei, L. S.; Steinberg, M. P.; Yamashita, N. Bound water associated with 7S and 11S soy protein determined by vapor sorption isotherms and pulsed NMR. *J. Food Sci.* **1990**, *55*, 130–132.

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