

## Research Article

# New View to Obtain Dryer Food Foams with Different Polysaccharides and Soy Protein by High Ultrasound

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The objective of this work was to determine the effects of high intensity ultrasound application on the foaming properties of soy protein-polysaccharides mixed solutions. To this end, foaming parameters during foam formation were analyzed. The samples were sonicated for 20 min using ultrasonic processor Vibra Cell Sonics, and model VCX 750 at a frequency of 20 kHz and an amplitude of 20%. The foams were produced by a Foamscan instrument. The evolution of the bubble size change in the foam was also determined by a second CCD camera. For all foamed systems, at two pHs 3 and 7, *Foam expansion* and *Relative Foam Conductivity* showed a great increase after ultrasonic treatment. Other parameters studied did not show difference. On the other hand, *Final Time of Foaming* and the *Total Gas Volume* incorporation for foams formation were correlated with the *Relative Foam Conductivity* decrease and the *Foam Expansion* increase when HIUS were applied in every system. Comparative bubble size and shape during the foam formation according to the treatments and pH used confirmed the parameters results.

## 1. Introduction

Proteins are a particular class of biopolymers with tension-activity character; as a result, they are fundamental in dispersed systems formations such as foams and emulsions. The use of soy proteins as functional ingredients in food manufacturing is increasing because of their role in human nutrition and health. Native soy protein, because of its quaternary and compact tertiary structure, has limited foaming [1–3] and emulsifying [4] properties. However, structural modifications by chemical methods such as deamidation, succinylation, reductioning, or denaturation allow greater conformational flexibility of this protein, improving its surface behavior and functionality [5–8]. One way to increase soy proteins functionality would be the high intensity ultrasound (HIUS) application. The effect of ultrasound is related to cavitation, heating, dynamic agitation, shear stresses, and turbulence [9]. It may cause physical changes producing protein aggregates through covalent and noncovalent bonds.

Polysaccharides are used in admixture to proteins mainly to enhance stability of dispersed systems. They can strongly enhance the stability of protein foams by acting as thickening or gelling agents [10]. In addition, the surface-activity difference along the structure of polysaccharides influences the foam formation parameters of proteins as well as the protein state and concentration.

In the present work, various polysaccharides solutions were added to soluble soy protein solution and two pH conditions were considered. Ultrasound was applied to the mixed solutions in order to analyze the potential modifications in foaming properties in the process of foam formation.

## 2. Materials and Methods

*2.1. Soy Protein Isolate Characterization and Sample Preparation.* Soy protein isolate (SPI) was provided by Instituto de la Grasa, Seville, Spain, with the following chemical composition (%): protein:  $88.41 \pm 0.22$ ; lipids:  $1.32 \pm 0.00$ ;

moisture:  $3.98 \pm 0.37$ ; ashes:  $4.41 \pm 0.08$ ; fiber:  $0.00 \pm 0.00$ ; polyphenols:  $0.15 \pm 0.002$ ; and soluble sugar:  $0.46 \pm 0.006$ .

Differential scanning calorimetry was used to determine the percentage of native protein from soy isolate with a Mettler TA4000 Thermal Analysis System equipped with TA72 software (Schwerzenbach, Switzerland). The instrument was calibrated with indium ( $156.6^\circ\text{C}$ ), lead ( $327.5^\circ\text{C}$ ), and zinc ( $419.6^\circ\text{C}$ ). The thermal parameters were determined by heating 15–20 mg of a protein solution at 12% w/w of sample from 0 to  $95^\circ\text{C}$  at  $10^\circ\text{C}/\text{min}$ . An empty pan was used as reference. The average value of at least three replicates is reported. The calorimetric thermogram obtained showed that SPI was 80% denatured.

Soluble SPI (SSPI) at different pHs (7 and 3) were used as starting material for the current work. Protein solutions, 4% w/w, at the corresponding pH were centrifuged for 1 hour at room temperature at 10,000 g. The protein content was determined in the soluble fraction by the Kjeldahl method ( $N \times 6.25$ ), resulting in 1.73 and 0.43 w/w% of soy protein for pHs 7 and 3, respectively.

Polysaccharides (PS) used were two tension-active polysaccharides, hydroxypropylmethylcelluloses called E4M and E50lv, and a nontensionactive polysaccharide,  $\lambda$ -Carrageenan (only at pH 7, it precipitates at pH 3), all in powder form, provided by Sanofi Bioindustries. The PSs were added to soluble soy protein solutions in order to reach a final concentration of 0.25%, wt/wt of PSs in each mixed system.

**2.2. Foam Formation.** The foaming properties of solutions coming from sonication treatment were characterized through their formation measured by the commercial instrument Foamscan instrument (Teclis-It Concept, Longessaigne, France) as described elsewhere [11]. Briefly, the foam is generated by blowing nitrogen gas at a flow of 45 mL/min through a porous glass filter ( $0.2 \mu\text{m}$  pores) at the bottom of a glass tube where 20 mL of the foaming aqueous solutions ( $25 \pm 1^\circ\text{C}$ ) is placed. In all experiment, the foam was allowed to reach a volume of 120 mL. The bubbling was then stopped, while the volume is followed by image analysis using a CCD camera.

The following parameters were determined. *Foam Expansion* (FE), as the inverse of the *Foam Maximum Density* (MD) determined by (1), is a measure of the liquid retention in the foam; the *Overall Foaming Capacity* (OFC, mL/s) was determined from the slope of the foam volume curve up to the end of the bubbling. The *Foam Capacity* (FC), a measure of gas retention in the foam, was determined by (2) and the *Relative Foam Conductivity* ( $C_f\%$ ) was measured to study the foam density and was determined by (3).

Consider

$$\text{FE} = \frac{V_{\text{foam}(f)}}{(V_{\text{liq}(i)} - V_{\text{liq}(f)})}, \quad (1)$$

$$\text{FC} = \frac{V_{\text{foam}(f)}}{V_{\text{gas}(f)}}, \quad (2)$$

$$C_f = \frac{C_{\text{foam}(f)}}{C_{\text{liq}(f)}} \times 100, \quad (3)$$

where  $V_{\text{liq}(i)}$  and  $V_{\text{liq}(f)}$  are the initial and the final liquid volumes;  $V_{\text{foam}(f)}$  is the final foam volume and  $V_{\text{gas}(f)}$  is the final gas volume injected;  $C_{\text{foam}(f)}$  and  $C_{\text{liq}(f)}$  are the final foam and liquid conductivity values, respectively.

Moreover, the evolution of the bubble size change during the first 40 s of foam formation was determined by a second CCD camera set with a macroobjective which allows capturing the variation of the gas bubble size every 5 s.

**2.3. High Intensity Ultrasound (HIUS) Treatment.** SSPI solutions at different pH were sonicated for 20 min using an ultrasonic processor Vibra Cell Sonics, model VCX 750 (maximum net power output: 750 W) at a frequency of 20 kHz and an amplitude of 20% (maximum amplitude 40%,  $228 \mu\text{m}$ ), which were constant. A 13 mm (1/2 inch) high grade titanium alloy probe threaded to a 3 mm tapered microtip was used to sonicate 10 mL of the solutions. Samples contained in glass test tubes were, in turn, immersed into a glycerine-jacketed circulating constant temperature cooling bath at  $0.5^\circ\text{C}$  to dissipate most of the heat produced during sonication treatments (Polystat, Cole-Parmer).

**2.4. Statistical Analysis.** All the experiments were performed in triplicate.

The model goodness-of-fit was evaluated by the coefficient of determination ( $R^2$ ) and the analysis of variance (ANOVA), using Statgraphics Plus 3.0. software.

### 3. Results

**3.1. Effect of HIUS Treatment on the Foaming Properties.** Foaming parameters for HIUS SSPI- $\lambda\text{C}$ ; SSPI-E4M and SSPI-E50lv treated solutions at pH 7 on mixed systems are compared in Figure 1 for FE (*Foam Expansion*); FC (*Foam Capacity*); OFC (*Overall Foam Capacity*); and  $C_f$  (*Conductivity of Foam*). The results showed a statistically significant difference change after treatment for FE and  $C_f$  parameters. Other parameters, FC and OFC, did not show any difference in any case. It can be said that a better foam is possible to form after HIUS treatment (FE increase), but less water is incorporated at the foam ( $C_f$  decrease), which would imply further changes in their corresponding foam stability. In Figures 2(a)–2(d) SSPI-E4M and SSPI-E50lv systems are shown at pH 3. The same trend was obtained with only 0.43 w/w% of SSPI in this case, indicating that the performance of these systems after treatment was the same at other protein-polysaccharide concentration relations. Another analysis was made by comparing the effect of HIUS on components alone. On the basis of these results (not shown), it is concluded that this behavior after treatment, where FE increased and  $C_f$  decreased, happened only in the mixed systems. This means that when SSPI or PS were HIUS treated alone and finally mixed to make the foams, no difference was observed for any foam formation parameters. Probably, the ultrasound may cause more structural changes when viscosity medium

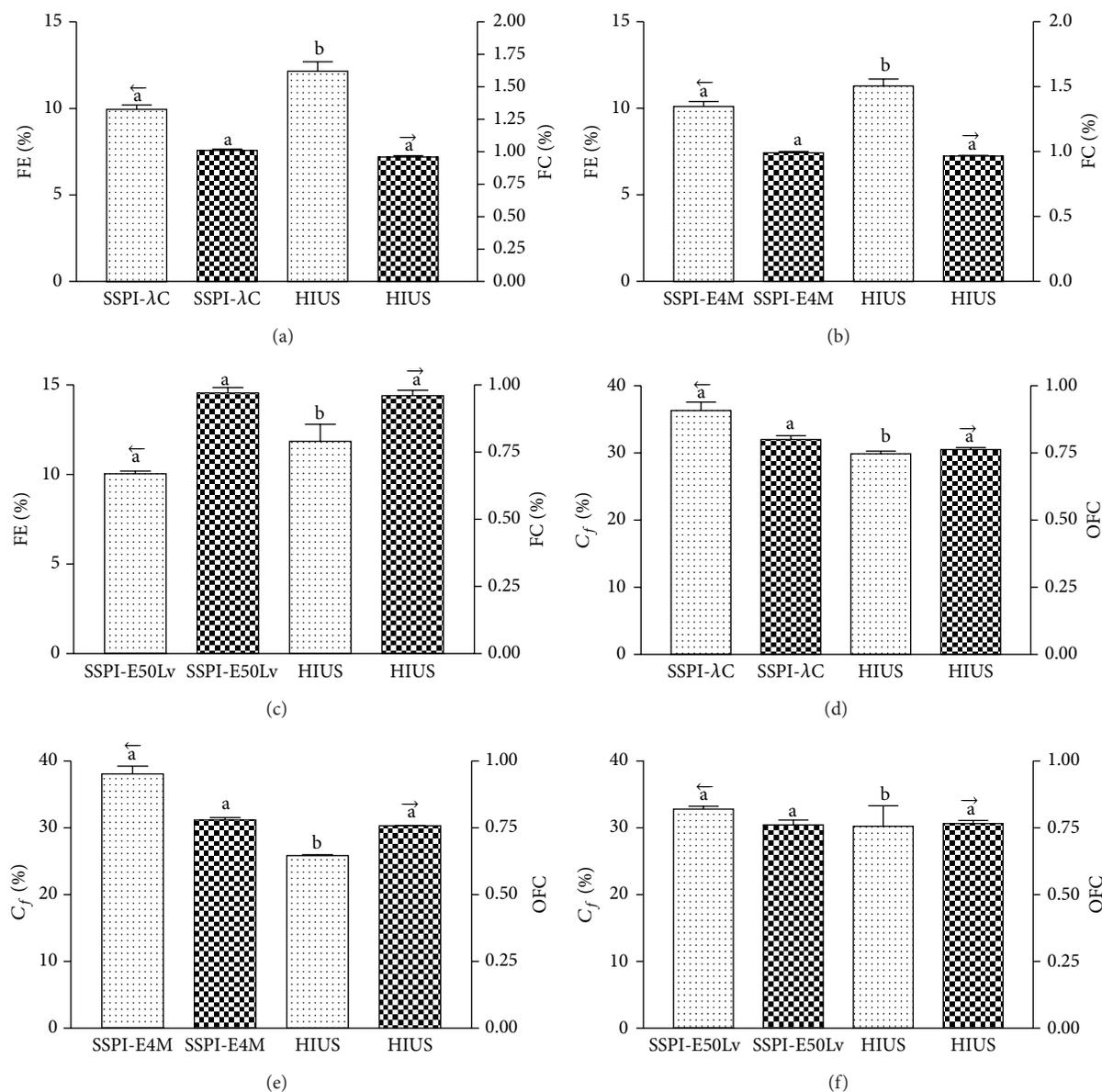


FIGURE 1: HIUS effect on mixed SSPI- $\lambda$ C; SSPI-E4M and SSPI-E50lv systems at pH 7 for ((a)–(c)) FE and FC; ((d)–(f)) OFC; and C<sub>f</sub> parameters. Same pattern columns correspond to the same sample, made at least in triplicate. Different letters for the same protein indicate a significant difference at  $P < 0.05$ . HIUS conditions: power output:  $4.27 \pm 0.71$  W; frequency: 20 kHz; amplitude: 20%; time of treatment: 20 min.

exceeds a certain value during the treatment. In a previous work, we explored the impact of high intensity ultrasound on the functionality of some food proteins: whey protein concentrate (WPC), soy protein isolate (500E), and egg white protein (EW) [12]. In spite of the structural difference between proteins, we clearly observed that 500E with an appreciable viscosity comparatively with WPC and EW was highly susceptible when HIUS was applied in most of the properties studied at that work.

In another previous publication [13], we observed that these same systems of soy protein and polysaccharides would be affected by thermodynamic incompatibility between protein and polysaccharides that would increase the apparent

concentration. There would be also another possible influence on the structural state when sonication is applied.

According to these results, the *Final Time of Foaming* until 120 mL and the *Total Gas Volume* incorporation for its formation were compared with the studied foaming parameters. Results obtained for the mixed solutions after sonication at the considered pHs are shown in Table 1. The results pointed out that the time required and gas quantity incorporated were correlated with the C<sub>f</sub> decrease, where more time and gas volume were required when HIUS was applied at each mixed system, independent of the pH media. This means that it is likely that structural modification by noncovalent linkages as surface hydrophobicity change of

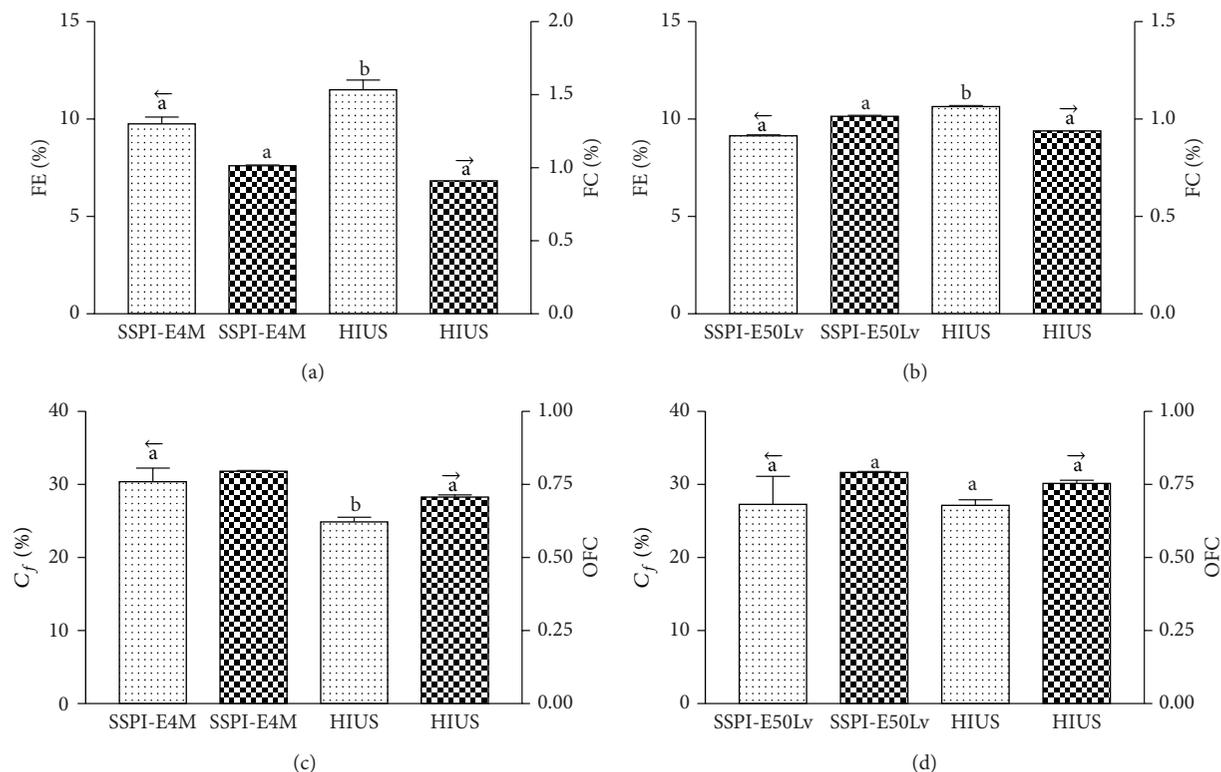


FIGURE 2: HIUS effect on mixed SSPI-E4M and SSPI-E50lv systems at pH 3 for ((a)–(b)) FE and FC; ((c)–(d)) OFC; and Cf parameters. The same pattern columns correspond to the same sample, made at least in triplicate. Different letters for the same protein indicate a significant difference at  $P < 0.05$ . HIUS conditions: power output:  $4.27 \pm 0.71$  W; frequency: 20 kHz; amplitude: 20%; time of treatment: 20 min.

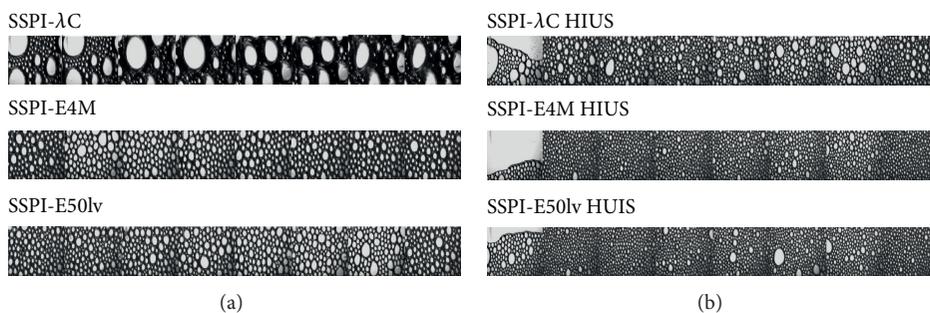


FIGURE 3: Gas bubbles structure in mixed foam during their formation. Images were taken for (a) SSPI-kappaC, SSPI-E4M, and SSPI-E50lv systems and (b) foams obtained from the respective HIUS treated solutions, at pH 7. HIUS conditions: power output:  $4.27 \pm 0.71$  W; frequency: 20 kHz; amplitude: 20%; time of treatment: 20 min.

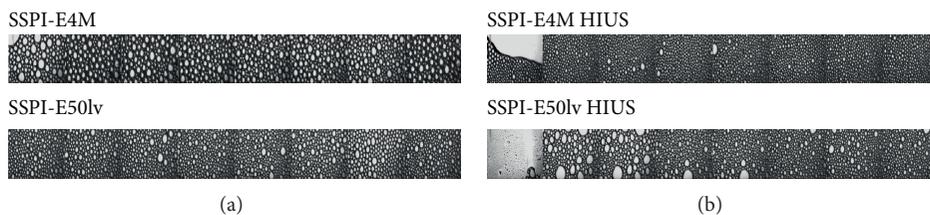


FIGURE 4: Gas bubbles structure in mixed foam during their formation. Images were taken for (a) SSPI-E4M and SSPI-E50lv systems and (b) foams obtained from the respective HIUS treated solutions, at pH 3. HIUS conditions: power output:  $4.27 \pm 0.71$  W; frequency: 20 kHz; amplitude: 20%; time of treatment: 20 min.

TABLE 1: Final Time of Foaming and Total Gas Volume incorporated in the foams of the mixed soluble soy protein and polysaccharides systems.

System	Final Time of Foaming (s)*	Total Gas Volume (cm <sup>3</sup> )*
SSPI- $\lambda$ C pH7	164.5 $\pm$ 3	120 $\pm$ 2
SSPI- $\lambda$ C pH7 HIUS	172.5 $\pm$ 3	126 $\pm$ 2
SSPI-E4M pH7	166.5 $\pm$ 3	121.5 $\pm$ 3
SSPI-E4M pH7 HIUS	172 $\pm$ 0	125 $\pm$ 0
SSPI-E50lv pH7	170 $\pm$ 2	186.5 $\pm$ 6
SSPI-E50lv pH7 HIUS	172.5 $\pm$ 7	125.5 $\pm$ 5
SSPI-E4M pH3	163.5 $\pm$ 3	119 $\pm$ 3
SSPI-E4M HIUS	181 $\pm$ 0	132 $\pm$ 0
SSPI-E50lv pH3	163.5 $\pm$ 1.5	119 $\pm$ 0
SSPI-E50lv pH3 HIUS	175 $\pm$ 0	128 $\pm$ 0

\* Mean  $\pm$  SD of at least three replicates, independently foamed. HIUS conditions: time of treatment: 20 minutes; frequency: 20 kHz; amplitude: 20%.

proteins in the mixed systems caused by HIUS application would be detrimental to the ability for liquid incorporation into the foam when SSPI and PS are together. In the mentioned work [12], we also found that HIUS increased surface hydrophobicity upon HIUS application for the three proteins studied: soy protein isolate, egg white protein, and whey protein. Those results suggested that the HIUS treatment induced a certain degree of molecular unfolding of the protein molecules causing more or less regions to be exposed to the environment promoted by the polysaccharide presence.

Thus, in food manufacturing, when good Foam Expansion but less water incorporation is desired, the use of HIUS would be a good tool to produce foams with mixed polysaccharides and ensure further foam stabilization.

**3.2. Effect of HIUS on Foams Air Bubbles Formation.** Bubble sizes could not be determined because the system of the foam itself is not transparent [9]. Therefore, qualitative comparison of bubble size and shape during the first 40 s of foam formation was analyzed according to the treatments and pH used.

Figures 3 and 4 show comparatively the images obtained for the air bubbles throughout the foams formation of SSPI-PS without (a) and HIUS treated (b) mixed solutions at pHs 7 and 3, respectively.

A direct relation can be seen between previous parameters analyzed with size and shape of bubbles produced at different conditions. In Figure 3, it is observed that the quantity of liquid incorporated was clearly seen by the thickness of its bubbles; lighter and smaller bubbles could be observed when HIUS was applied. At pH 3 (Figure 4), the same tendency was also observed. HIUS treated mixtures (b) showed lower bubble density, indicating a decrease of water incorporation in the first 40 s of foaming process when HIUS was applied.

For both pHs, bubble images of the single components were also studied (not shown), and with the carrageenan exception, which does not foam by its own, the others did not show any influence or a perceptible change after HIUS treatment.

Therefore, the analysis of gas bubble behavior throughout foams formation during the first 40 s would be a good tool to characterize foams quickly as an early approximation.

## 4. Conclusions

Through all analyzed parameters for HIUS treatment on mixed SSPI and PS systems at pHs 7 and 3, FE and Cf showed a significant change after treatment. Other parameters such as FC and OFC did not show a difference between the systems at any pH studied. The results exhibited an increase for FE, which was obtained after HIUS application, even when less liquid was incorporated, denoted by the Cf decrease. On the other hand, it could be said that this behavior can be observed only for mixed systems (it did not happen with the components being alone) and at each concentrations ratio, at both pHs studied (where SSPI contents were different for each condition; 1.73 and 0.43 w/w% resp.).

The time required and gas quantity were correlated with the Cf decrease, where more time and gas volume were required when HIUS was applied in each system.

Comparative bubble size and shape during the foam formation were analyzed according to the treatments and pH studied. A lower quantity of liquid was observed for both pHs incorporation during the foam formation when HIUS was applied in mixed systems.

The polysaccharide addition in this framework has dual function: drier foam production and further foam stabilization [10]. In addition, at these conditions, surface-activity character of polysaccharides of studied pHs of soy protein was not influencing factors of importance when foam parameters of formation were studied, which allow combining them in a broad spectrum of soluble soy protein-polysaccharides compositions concerning the food context requirements and more favorable economically.

For this reason, it is important to consider the HIUS application in this mixed system. Although more experimental trials should be performed to investigate other protein sources, it can be said that, in industrial processing of soy protein, when good Foam Expansion and relatively dry foams are necessary (e.g., angel food cake like products), its application should be in the mixed system to reach the desired food foams characteristics.

## Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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