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### Rheology and Stability of Acidified Food Emulsions Treated with High Pressure

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The stability and rheology of acidified model oil-in-water emulsions (pH  $3.6 \pm 0.1$ ) were evaluated before and after high-pressure treatments. Varying concentrations of canola oil (0–50% w/w), whey protein isolate, polysorbate 60, soy lecithin (0.1–1.5% w/w each), and xanthan (0.0–0.2% w/w) were chosen. Exposure to high pressures (up to 800 MPa for 5 min at 30 °C) did not significantly affect the equivalent surface mean diameter D[3,2], flow behavior, and viscoelasticity of the whey protein isolate and polysorbate 60-stabilized emulsions. Pressure treatments had negligible effects on emulsion stability in these systems, except when xanthan (0.2% w/w) was present in which pressure improved the stability of polysorbate 60-stabilized emulsions. Soy lecithin-stabilized emulsions had larger mean particles sizes and lower emulsion volume indices than the others, indicating potential instability, and application of pressure further destabilized these emulsions.

## KEYWORDS: High-pressure processing; oil-in-water emulsions; rheology; particle size distribution; emulsion volume index

#### INTRODUCTION

High-pressure processing (HPP) offers great potential for producing food with natural attributes that are otherwise lost during traditional thermal processes. The commercial application of HPP to high-acid foods such as orange juice, guacamole, etc. has been very successful in making high-quality products (1, 2). Recent studies have shown that HPP up to 500 MPa for 1 min at 25 °C can eliminate spoilage microorganisms from commercial salad dressings, a high-acid food product, without significantly altering their rheology (3). However, because of compositional differences, these salad dressings exhibited different viscosity and physical stability after high-pressure treatment. French dressing, stabilized mainly by the presence of egg yolk, was the least stable while Caesar dressing, with whey proteins and xanthan, showed maximum stability toward highpressure treatment. It is therefore important to pursue studies detailing the effects of pressure treatment on the rheology as well as the physical stability of acidic food emulsions.

Few studies, mostly at neutral pH, have dealt with effects of high pressure on food emulsions. Model oil-in-water emulsions of pH 7.0, containing sodium caseinate (50 g/kg) and peanut oil (300 g/kg), did not show a change in particle size distribution or emulsion viscosity after pressure treatment at 450 MPa for 30 min (4). However, replacement of sodium caseinate by  $\beta$ -lactoglobulin caused an increase in viscosity, without affecting the particle size, after pressurization at 450 MPa and 40 °C for 30 min. Dickinson and James (5) reported that high-pressure treatment up to 800 MPa for 60 min increased average droplet diameter D[4,3] and storage modulus G' of the emulsions (pH 7.0) stabilized by  $\beta$ -lactoglobulin. Dickinson and James (6) also found that following pressure treatment (200-800 MPa for 30 min) the average droplet diameter D[4,3] as well as complex modulus G\* of oil-in-water emulsions (20% soybean oil, 0.5%  $\beta$ -lactoglobulin) increased with an increase in pH from 2.0 to 5.0, followed by a decrease at pH 7.0. Similar studies on emulsions at neutral pH have concluded that high pressure does not adversely affect particle size, flow behavior, or viscoelasticity of the model food emulsions.

The objective of this study was to examine the effects of surfactant type and concentration, dispersed phase concentration, and the presence of a stabilizer on the rheological properties as well as physical stability of pressure-treated acidic emulsions, using materials and conditions found in commercial salad dressings. As a result, conclusions may be drawn regarding the compositional factors that impart sufficient physical stability for acidified emulsions to withstand high pressure.

#### MATERIALS AND METHODS

**Reagents and Chemicals.** Whey protein isolate (WPI) (BiPRO, Lot No. JE 135-1-420, 97.6% protein on dry matter basis) obtained from Davisco Foods International, Inc., Eden Prairie, MN, oil-free soy lecithin (SL) (Centrolex F, Lot No. 01155110) obtained from Central Soya Company Inc., Fort Wayne, IN, and polysorbate 60 (P-60) (Uniquema, Wilmington, DE) were used as emulsifiers. Canola oil (The Kroger Co., Cincinnati, OH) was used as the dispersed phase, while xanthan (CP Kelco, Wilmington, DE) was added as a stabilizer. HCl (5 N) (Lab Chem Inc., Pittsburgh, PA) was used as the acidifying medium.

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**Table 1.** Experimental Design of Fractional Factorial Study to Determine the Effects of Independent Variables on the Physical Characteristics of Acidified Emulsions; n = 36

independent variable	level of independent variable			
pressure (MPa) lipid content (% w/w) type of surfactant surfactant concentration (% w/w)	0.1 0 WPI 0.1	500 10 SL 0.3	50 P-60 1.0	

**Table 2.** Full Factorial Design for Acidified Emulsions with 30% Dispersed Phase; n = 54

independent variable	levels of independent variable			
pressure (MPa) surfactant (% w/w) xanthan (% w/w)	0.1 WPI (1.5) 0	500 SL (0.3) 0.2	800 P-60 (0.3)	



**Figure 1.** Pressure (–) and temperature ( $\Box$ ) profiles during a typical HPP run (500 MPa for 5 min).

Emulsion Preparation. Aqueous solutions were prepared by dissolving preweighed quantities of each surfactant (0.1-1.5% w/w) separately in deionized water by gentle magnetic stirring at room temperature. In certain instances, xanthan gum (0.2% w/w) was also added. The specific compositional details are outlined in Tables 1 and 2. These solutions were acidified to pH 3.6  $\pm$  0.1 using 5 N HCl. A coarse emulsion (150 g) was prepared by blending canola oil (10-50% w/w) and surfactant solution with an Ultra-Turrox (T-25 Basic, IKA Labortechnik, Wilmington, NC) high-shear probe for 2 min at 10 000 rpm and room temperature. Fine emulsification was achieved by homogenizing the entire coarse emulsion premix in a valve type homogenizer (H-pump, Foss Electric, Eden Prairie, MN) at an operating pressure of 2000 psi and room temperature. The preparation and homogenization processes achieved oil droplet distributions with mean D[3,2] values ranging from 0.5 to 5  $\mu$ m. Aliquots (100 g) of the fine emulsion were vacuum-packed in Nylon-EVA pouches (Winpak Ltd., Winnipeg, Manitoba, Canada) and subjected to high-pressure treatment, with the exception of nonpressure-treated controls.

**High-Pressure Treatment.** High-pressure treatment was carried out in a QFP-6 Cold Isostatic Press (Flow Pressure Systems, Kent, WA) at pressures ranging from 500 to 800 MPa and temperatures of 25-30°C, for 5 min. A 1:1 solution of distilled water and glycol (Houghton-Safe 620-TY, Houghton International, Valley Forge, PA) was used as the transmitting pressure in the isostatic press. The samples as well as pressure-transmitting fluid were preconditioned at 1 and 10 °C to control adiabatic heating (7) during pressurization at 800 and 500 MPa, respectively. The pressure, product temperature, and temperature of the water-jacketed pressure chamber were monitored and recorded in 3 s intervals using a 21X Micrologger (Campbell Scientific Inc., Logan, UT) connected to a computer running PC208W datalogger support software (Campbell Scientific Inc.). A representative pressure temperature profile is shown in **Figure 1**.

**Particle Size Measurement.** Particle size distributions of the emulsions were characterized by multiangle static light scattering using

a Mastersizer Microplus laser diffractometer (Malvern Instruments Ltd., Malvern, U.K.) with a relative refractive index of 1.10 (ratio of the refractive index of the dispersed phase relative to that of the continuous phase) and zero absorbance. The sample was added slowly in deionized water until an obscuration of 20% was reached. The measurements on the pressurized samples as well as nonpressure-treated controls (of same age) were performed within 2 h after the pressure treatment. Emulsions were diluted as follows to disperse flocculates: 90 g of water containing 2% sodium dodecyl sulfate (SDS) and 10 g of emulsion sample were mixed by gentle magnetic stirring. The mixture was transferred to a 100 mL standing cylinder, and after the phases were separated, the upper emulsified layer was sampled for particle size measurements. It was verified that the use of SDS on unflocculated emulsions did not significantly affect the particle size. Droplet size was characterized by the mean surface-weighted average diameter, D[3,2], defined by

$$D[3,2] = \sum_{i} n_i d_i^3 / \sum_{i} n_i d_i^2$$
<sup>(2)</sup>

with  $n_i$  being the number of droplets with a diameter  $d_i$ . Plots of size distribution were reported as volume% vs droplet diameter in the range of 0.05–1000  $\mu$ m. In addition, the dispersion index of the emulsions was characterized using "span", which is defined as (D[90] – D[10])/D[50] where D[x] is the average droplet size in volume under which [x]% of total sample weight remains (8). Span describes the width of the particle size distribution, independent of the median size. Measurements were repeated at least three times for each observation.

**Determination of Emulsion Volume Index (EVI).** Capillary tubes containing 1 mL of emulsion sample, dyed with  $5 \mu L$  each of Methylene Blue and Oil Red "O", were centrifuged in a Damon/IEC microhematocrit centrifuge (model MB) at 14 000g (gravity) for 30 min. The EVI was determined by calculating the ratio of height of upper emulsion length to the total sample length and normalizing for the sample's lipid concentration using the following relation (9):

$$EVI = \frac{(mm \text{ emulsion layer/total volume}) \times 100}{(weight\% \text{ lipid/0.9})}$$
(3)

where 0.9 is the specific gravity of the lipid phase.

**Rheological Measurements.** Rheological analysis of the samples was performed using an AR 1000-N controlled stress rheometer (TA instruments, New Castle, DE) with an acrylic cone and plate geometry (diameter 60 mm, cone angle 4°) at 20 °C. Flow behavior and viscoelastic measurements on pressurized emulsions and untreated controls (of same age) were performed immediately after pressurization to minimize time effects. Flow curves indicate the shearing behavior of a material in response to applied stress during or after processing. Flow measurements were performed by ramping shear rate from 100 to 450 s<sup>-1</sup> (0.2–2.0 Pa shear stress) within 2 min. The Rheology Advantage data analysis software (TA Instruments) was used to fit the Ellis model (4) to the stress sweep data in order to characterize the flow behavior of the dressings.

$$\frac{\eta - \eta_{\infty}}{\eta_0 - \eta_{\infty}} = \frac{1}{1 + (K \cdot \sigma)^n} \tag{4}$$

where  $\eta$  = viscosity (Pa s),  $\eta_0$  = zero stress viscosity (Pa s),  $\eta_{\infty}$  = infinite stress viscosity (Pa s), K = Ellis constant (1/Pa s), and n = stress index. Zero stress viscosity characterizes the state of emulsion at rest or restructuring after shearing while infinite stress viscosity characterizes the viscous state of the emulsion at very high shear stresses. The Ellis constant and stress index are power law parameters somewhat similar to parameters of other power law models, such as Herschel-Bulkley and Van Wazer (10).

The viscoelastic properties of the emulsions were characterized by dynamic oscillation experiments. Oscillatory testing is the most common dynamic method to study the viscoelastic behavior of food. It describes material properties in terms of storage modulus G' (elastic behavior) and loss modulus G'' (viscous behavior). Initially, stress sweeps at an angular frequency of 1 Hz were performed to establish a maximum oscillatory stress within the linear viscoelastic range of the emulsions.

Table 3. ANOVA Probability Values from Fractional Factorial Study (Table 1)

	D[3,2]	EVI	$\eta_0$	$\eta_{\infty}$	К	п	W	X	y	Ζ
pressure	0.39	0.47	0.50	0.38	0.98	0.52	0.14	0.90	0.60	0.83
surfactant	0.55	0.10	0.32	0.64	0.26	0.90	0.20	0.94	0.01	0.03
concentration lipid level	0.09 < <b>0.01</b>	<0.01 <0.01	0.28 <b>0.01</b>	0.50 0.68	0.68 0.51	0.16 < <b>0.01</b>	0.47 <b>0.047</b>	0.02 0.01	0.26 < <b>0.01</b>	0.63 <b>0.01</b>

Well within the linear viscoelastic region, an oscillatory stress of 0.5 Pa was chosen and samples were subjected to an angular frequency sweep from 0.1 to 70 rad/s. A power law equation (5 and 6) was used to model the dependency of the storage and loss moduli, G' and G'' (Pa), on the angular frequency  $\omega$  (rad/s) (10).

$$G' = w \cdot \omega^x \tag{5}$$

$$G'' = \mathbf{y} \cdot \boldsymbol{\omega}^{\boldsymbol{z}} \tag{6}$$

where w, x, y, and z are fitted constants.

**Experimental Design and Statistical Analyses.** A four level fractional factorial study (**Table 1**) was designed with Minitab statistical software (Release 13.31) based on a Taguchi design. Canola oil concentration (0-50% w/w), type of surfactant (WPI, SL, and P-60), surfactant concentration (0-1% w/w), and level of pressure (0-500 MPa) were chosen as four independent factors. Thirty-six randomized runs were carried out based on this design.

A full factorial study, in three replicates (**Table 2**), was designed, using same software, to observe the effects of high pressure (0-800 MPa) and xanthan (0 and 0.2% w/w) on the stability of model emulsions. The lipid level in these emulsions was held constant at 30% w/w while using a single level of each surfactant, 1.5 wt% whey protein isolate, 0.3 wt% soy lecithin, and polysorbate 60. For each experimental observation in both studies, the mean of 2-3 repeated measurements was used. Analysis of variance (ANOVA) was performed using a general linear model (GLM) procedure in Minitab to establish the influence of independent variables on the droplet size distribution and EVI.

#### **RESULTS AND DISCUSSION**

The results of the fractional factorial study indicate that pressure did not have an effect on the rheological properties or the emulsion particle size and stability (**Table 3**). The lipid level, however, had a statistically significant influence on the emulsion stability and rheological properties. This may have been in part due to the extreme ranges of lipid levels chosen, from 0 to 50%. The rationale for the "no lipid" cell in the design was to examine the pressure effects on the continuous phase vs the entire emulsion. Furthermore, the study included a range of surfactant levels that extended above and below that needed to stabilize the emulsion.

A full factorial experiment was implemented using a constant lipid level (30% w/w) that was intermediate to the levels used in the fractional factorial study, along with constant surfactant levels commonly used in salad dressings (11, 12). The overall results of the full factorial study indicated no significant effects of pressure on all measured variables. Significant differences were attributable to the different surfactant types. On the basis of this result, the data were reanalyzed examining effects of pressure, xanthan, and their interaction within each surfactant type (**Table 4**).

**Flow Curves.** The flow curves of different emulsions were compared using the Ellis model to determine zero stress viscosity ( $\eta_0$ ), infinite stress viscosity ( $\eta_\infty$ ), Ellis constant (*K*), and stress index (*n*). Attempts at fitting the Herschel–Bulkley model to shear stress vs shear rate data to determine a "fitted" yield stress produced variable, and in some cases negative, results. These occasional negative results are an artifact of fitting the nonlinear

Table 4. ANOVA Probability Values from the Full Factorial Study, Analyzed by Surfactant Type

surfactant	factor	D[3,2]	span	EVI
whey protein (1.5% w/w)	pressure xanthan pressure × xanthan	0.50 0.94 0.60	0.52 0.42 0.41	0.54 0.19 0.17
soy lecithin (0.3% w/w)	pressure xanthan pressure × xanthan	0.99 0.14 0.80	0.01 <0.01 <0.01	0.26 0.20 0.29
polysorbate-60 (0.3% w/w)	pressure xanthan pressure × xanthan	0.05 <0.01 0.03	0.58 < <b>0.01</b> 0.11	0.05 <0.01 0.03

Herschel-Bulkley model to the flow data and are not uncommon when trying to approximate yield stress values of low magnitude (C. R. Daubert, personal communication, August 15, 2001). The Ellis model alleviated this problem. This model incorporates power law behavior at high-shear stresses while allowing a plateau of Newtonian behavior at low-shear stresses. The zero stress viscosity serves as a close approximation of the yield stress since it describes the viscosity of the fluid at very low stresses. Flow behavior was unaffected by pressure and dependent upon lipid level. Measured viscosities across the shear stresses (0.2-2.0 Pa) were low, with a maximum of 0.005 Pa·s. The viscosities of the 50% lipid emulsions were significantly greater (p < 0.05) than the 10% w/w lipid emulsions, irrespective of surfactant type or pressure treatment (Table 5). Nearly all emulsions displayed a slight shear thickening behavior, the exception being the whey protein-stabilized, 50% lipid emulsion (Figure 2). Anton et al. (13) observed a similar trend for pressure emulsions, and they attributed the increase in viscosity to enhanced flocculation of oil droplets. However, in the case of whey protein-stabilized emulsions, electrostatic or steric repulsion between adsorbed proteins at the interface can hinder flocculation of oil droplets (14), which can explain lower viscosity in the later case. A minor, but statistically significant, increase in the zero stress viscosity was observed with increasing levels of lipid but was unaffected by pressure.

Oscillatory Curves. The oscillatory data indicated no significant affects of pressure treatment on emulsion viscoelasticity (**Table 3**). Variation in G' and G'' were largely a function of lipid level and secondarily the surfactant type and amount. Fitted power law constants ( $r^2 > 0.9$ ), w, x, y, and z, were used to quantify variation in the frequency dependency of G' and G'' for different emulsions. Statistical analysis of these power law constants indicated that the storage modulus was affected by lipid level and surfactant concentration, while the loss modulus was affected by lipid level and surfactant type. The addition of a small amount of lipid (10%) caused little variation in both moduli from the lipid-free continuous phase. Furthermore, the lipid-free continuous phase was unaffected by pressure treatments. A significant increase in the loss modulus and decrease in the storage modulus occurred with the addition of 50% lipid, as seen with the polysorbate 60-stabilized emulsions (Figure 3). Whey protein provided the greatest structure to emulsions, yielding a significantly higher loss modulus than

 Table 5.
 Particle Size, Emulsion Stability, and Flow Properties of Emulsions from Fractional Factorial Study, Averaged Across All Pressures,

 Surfactant Types, and Levels
 Surfactant Types, and Levels

lipid level (%)	D[3,2] (µm)	EVI	$\eta_0$ (Pa s)	$\eta_{\infty}$ (Pa s)	<i>K</i> (1/Pa s)	п
0 10 50	0.452 (0.034) 1.060 (0.181)	0.661 (0.210) 0.868 (0.112)	0.001 <sup>a</sup> (0.0002) 0.002 (0.001) 0.020 (0.014)	0.033 (0.039) 0.014 (0.003) 0.024 (0.011)	0.363 (0.226) 0.236 (0.163) 0.336 (0.103)	0.715 (0.135) 0.848 (0.321) 2.086 (0.368)

<sup>a</sup> Mean (standard deviation).



**Figure 2.** Fitted flow curves for 10% (open symbols) and 50% (closed symbols) emulsions (averaged across all treatment pressures and surfactant concentrations) stabilized by polysorbate 60,  $\blacksquare \square$ ; whey protein isolate,  $\blacktriangle \triangle$ ; and soy lecithin,  $\bullet \bigcirc$ .



**Figure 3.** Storage modulus *G'* (closed symbols) and loss modulus *G''* (open symbols) as a function of frequency for polysorbate 60 (0.3%)-stabilized emulsions (pH 3.6, 20 °C) not treated with pressure containing 0% lipid,  $\blacksquare$   $\square$ ; 10% lipid,  $\blacktriangle$   $\triangle$ ; and 50% lipid,  $\blacklozenge$   $\bigcirc$ .

those emulsions stabilized by either soy lecithin or polysorbate (**Figure 4**). Dickinson and James (5) demonstrated that  $\beta$ -lactoglobulin can undergo unfolding after pressure treatment, thus allowing protein-coated oil droplets to join together to form a network structure following protein—protein or protein—oil interactions at the exposed hydrophobic sites.

**Particle Size Distribution.** The mean particle size was significantly affected by lipid concentration. In the fractional factorial study, the 50% lipid emulsions had significantly higher (p < 0.01) D[3,2] values than the 10% lipid counterparts (**Table 5**). Desrumaux (8) showed that as the fat content increases, emulsifier availability decreases, which favors oil droplet coalescence and, thus, higher mean droplet diameters. In the full factorial study, soy lecithin emulsions were characterized by considerably larger D[3,2] values, indicative of potential instability. Treatment with pressure, 500 or 800 MPa, lead to significant increases in mean particle size for soy lecithin-stabilized emulsions (**Figure 5**). Pressure treatments did not have the same destabilizing effect on the whey protein or polysorbate 60 systems.



**Figure 4.** Storage modulus *G'* (closed symbols) and loss modulus *G''* (open symbols) as a function of frequency for pressure-treated (500 MPa, 5 min) emulsions (pH 3.6, 20 °C) containing 50% lipid and stabilized by polysorbate 60 (1.0%),  $\blacksquare$   $\Box$ ; whey protein isolate (1.0%),  $\blacktriangle$   $\triangle$ ; and soy lecithin (0.3%),  $\blacksquare$   $\bigcirc$ .



**Figure 5.** Oil droplet size distributions in 30% lipid emulsions stabilized by 1.5% whey protein and 0.3% soy lecithin, treated with pressure (thick lines) and nonpressurized controls (thin lines).

One hypothesis for the breakdown of the soy lecithinstabilized emulsions is linked to the relative volume reductions of the continuous and dispersed phases under high pressure. Earlier work of Cheftel (15) confirmed that structural components of foods, such as water, lipids, proteins, etc., undergo volume contraction to varying degrees under high-pressure conditions. Lipids have a higher adiabatic heat of compression (7) and a corresponding greater volume reduction than those of water. Under pressure, the lipid phase would have a relatively reduced particle size due to its greater compressibility than the aqueous continuous phase. Presumably in a pressurized state, excess surface coverage of surfactant may diffuse from the lipid surface. Desorption of smaller molecules, like soy lecithin, might be favored over comparatively larger protein molecules from the oil surface. Upon rapid decompression, the lipid phase experiences a relative increase in volume due to the system's expansion leading to insufficient lipid surface coverage and in turn coalescence and destabilization. This effect would be more pronounced in weakly stabilized systems, such as the lecithin case here.

Rheology and Stability of Pressure-Treated Emulsions



**Figure 6.** D[3,2] values for polysorbate 60 (0.3%)-stabilized emulsions at 30% lipid level with ( $\blacksquare$ ) and without ( $\Box$ ) xanthan.

The D[3,2] values of polysorbate 60- (**Figure 6**) and soy lecithin-stabilized emulsions were higher in the presence of xanthan. This could be due to delayed diffusion of surfactants at the oil-water interface due to xanthan-thickened continuous phase, resulting in depletion flocculation (*16*, *17*). The span for polysorbate 60- and soy lecithin-stabilized emulsions increased significantly with the presence of xanthan across all pressures. There was a significant pressure  $\times$  xanthan interaction in the span term of the soy lecithin emulsions. That is, in the presence of xanthan, increases in pressure resulted in increases in the span value, while the opposite was observed in emulsion without xanthan. However, the decrease in the span term in the later case was accompanied by a significant increase in D[3,2].

**EVI.** Emulsion stability was characterized using an EVI. A high EVI value indicates greater physical stability (18). Lipid content and surfactant concentration had a significant effect on the EVI of emulsions analyzed in the fractional factorial study. In the case of higher lipid levels (50% w/w) and low surfactant concentration (0.1% w/w), the formation of free fat was observed after centrifugation, which indicated emulsion destabilization. Inadequate coverage of oil droplets or the displacement of surfactant from oil—water interface under pressure could lead to flocculation of lipid molecules at exposed surfaces, thus resulting in weak emulsions. Emulsions having lower lipid levels (10% w/w) did not exhibit formation of free fat and had high (stable) EVI values, the exception being 0.1% soy lecithin-stabilized emulsions.

Among the factors studied, surfactant type constituted the greatest variation in emulsion stability. For the whey protein isolate-stabilized emulsions, none of the experimental factors contributed to variation in EVI. Pressure had a stabilizing effect on polysorbate 60-stabilized emulsions (Figure 7) as indicated by increasing EVI values following pressure treatment, both with and without xanthan. The addition of xanthan significantly increased (p < 0.01) the emulsion stability. Emulsions prepared with soy lecithin (0.3%) produced unstable emulsions regardless of pressure treatment (Table 6). Increases in pressure magnitude further reduced soy lecithin emulsion EVI values and, as mentioned earlier, increased both the dispersed phase droplet size and the droplet distribution uniformity (span). These results suggest that emulsions formed using soy lecithin were inherently weak and pressure treatment resulted in further destabilization of these emulsions.

The variation in stabilizing behavior of different surfactants is a reflection of the differences in their molecular structure, diffusion rates at interfaces, and affinity toward aqueous or lipid phase. Soy lecithin is lipophilic in nature and produces weak



Figure 7. EVI values for polysorbate 60 (0.3%)-stabilized emulsions with  $(\blacksquare)$  and without  $(\Box)$  xanthan.

surfactant	D[3,2] (µm)	span	EVI
whey protein (1.5% w/w)	0.49 <sup>a</sup> (0.04)	3.20 (0.85)	1.12 (0.04)
soy lecithin (0.3 w/w)	2.15 (1.56)	10.45 (10.17)	0.16 (0.18)
polysorbate-60 (0.3% w/w)	0.70 (0.10)	1.99 (0.48)	1.16 (0.10)

<sup>a</sup> Mean (standard deviation).

oil-in-water emulsions while polysorbate-60 and whey protein isolate, being hydrophilic surfactants, work effectively in oilin-water emulsions even under higher magnitudes of pressure. The significant increase in the EVI following pressurization of polysorbate 60-stabilized emulsions is particularly interesting. The reason for this phenomenon remains unclear but is likely due to altered interactions between the emulsifier and the dispersed phase. Large biopolymers can undergo significant structural rearrangement under pressure resulting in disruption of hydrophobic interactions, gel formation, and in some cases, polymer precipitation (19, 20). For instance, ovalbuminstabilized oil-in-water emulsions were destabilized by pressure (>400 MPa) in the presence of >0.02 M NaCl due to pressureinduced polymerization of ovalbumin (21). However, polysorbate 60 is a relatively small molecule (MW  $\sim$  1300) and thus not susceptible to significant or permanent restructuring as a result of pressure treatment. Furthermore, there are no published reports of pressure-induced structural rearrangements in any biopolymer that resulted in improved emulsification properties. High pressure will enhance the hydrophobic interactions between the emulsifier and the lipid phase, particularly when the system is at pressure. Whether this enhancement persists following depressurization is only speculative.

#### CONCLUSIONS

This study confirmed that pressure treatment had no significant detrimental effects on the rheological behavior as well as physical stability of acidified emulsions stabilized by whey protein isolate and polysorbate-60. Differences in the flow behavior, viscoelasticity (G' and G''), particle size distribution, and physical stability (EVI) of emulsions were influenced largely by the lipid content and the type of surfactant. Emulsions stabilized by soy lecithin were inherently unstable, and this instability was further aggravated under pressure. Pressure stable oil-in-water emulsions can be formed using hydrophilic surfactants such as polysorbate 60 or whey protein. The addition of xanthan improved stability in systems emulsified with polysorbate 60, and this stability was further improved by the application of pressure treatment.

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