Rheological Properties of Cholesterol-Reduced, Yolk-Stabilized Mayonnaise

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ABSTRACT: The effects of cholesterol on droplet size distribution and rheological properties of cholesterol-reduced egg yolk-stabilized emulsions were determined. Oil-in-water emulsions were prepared by using spray-dried eggs submitted to different levels of cholesterol reduction (14–81 wt% of cholesterol removed). Cholesterol was extracted in a modified Jennings apparatus with subcritical CO_2 (9°C and $4.66 \cdot 10^6$ Pa). Oscillatory and steady flow tests, as well as droplet size distribution measurements, were carried out on the cholesterol-reduced egg yolk-stabilized emulsions. The rheological parameters increased with the level of cholesterol reduction up to 40–80 wt% for emulsions having the same total amount of egg yolk. Opposite results, however, were obtained with some emulsions stabilized by a highly cholesterol-reduced (≈80 wt%) egg yolk. These results were explained by taking into account two opposite phenomena: an increase in the concentration of surface-active agents as cholesterol content decreased, and a lipoprotein structural rearrangement induced by cholesterol removal.

Paper no. J10062 in *JAOCS 79*, 837–843 (August 2002).

KEY WORDS: Droplet size distribution, egg yolk, emulsifier, low-in-cholesterol emulsion, mayonnaise, rheology.

Many food products are oil-in-water (o/w) emulsions, where the oil droplets are usually stabilized by protein molecules that, either alone or together with other surface-active components, form strong and cohesive interfacial films that may resist tensile or shearing stresses and protect the oil droplets from coalescence (1). Egg yolk is often used in foods as an emulsifier because it imparts desirable flavor, mouthfeel, and color (2,3). The emulsifying capacity of egg yolk is mainly a result of phospholipids, lipoproteins (LDL and HDL), and nonassociated proteins (livetin and phosvitin), with LDL being the most important contributor to these emulsifying properties (4). The good emulsifying properties of egg yolk lipoproteins are attributed to their highly flexible structures, allowing great affinity and adsorption at oil–water interfaces, followed by their ability to form viscoelastic films (4).

A problem associated with egg yolk is its high cholesterol content, which has been implicated as a causative agent for heart disease. Extraction with organic solvents has been proposed as an effective way of removing cholesterol from egg yolk (5,6). Paraskevopoulou and Kiosseoglou (5) observed that extracting yolk with petroleum ether resulted in a yolk-protein concentrate with emulsifying properties comparable to those of native egg yolk. A significant deterioration, however, was observed when the yolk was extracted with a mixture of petroleum ether and ethanol. In general, organic solvents such as hexane/isopropanol (6), ethanol (7), or petroleum ether (8,9) remove all the lipids, including cholesterol, as well as the phospholipids responsible for the functional properties. On the other hand, treatment with subcritical or supercritical CO_2 offers a more attractive way of selectively extracting cholesterol from egg (10). The advantages of CO_2 extraction are product safety, low toxicity, low cost, lack of flammability, lipid extraction selectivity, and higher maintenance of yolk functionality in food products (8–10). Unfortunately, spray-drying, in which relatively high temperatures are used, is usually the first step prior to cholesterol extraction (3). This prior treatment may denature the lipoproteins, thus destroying the noncovalently stabilized lipid–protein complexes that impart high functionality (11).

The relationship between rheology and textural properties and its importance to consumer acceptance is well known (12). The effect of spray-drying on the rheology of egg yolk-stabilized emulsions has been reported (13). Some research was carried out (7,8,14) on the rheological properties of emulsions stabilized with low-in-cholesterol egg products, but contradictory results were obtained. No systematic studies concerning the influence of the extent of cholesterol removal on the resulting egg yolk-stabilized emulsion have been reported. The objective of this investigation was to evaluate the rheological properties of model mayonnaise stabilized by processed egg products with different cholesterol contents.

EXPERIMENTAL PROCEDURES

Materials. Different o/w emulsions having 70 and 77.5% by weight (wt) sunflower oil (Koipe S.A., Jaén, Spain) were stabilized by using spray-dried (temperature, 80–85ºC; residence time, 2.5 min) egg yolks from Ovosec (Valladolid, Spain). This product was leached with subcritical CO_2, achieving various levels of cholesterol contents. Reduced-cholesterol egg yolk concentrations ranged between 3 and 8 wt% in the emulsion. This product was reconstituted with distilled water prior to emulsification. The reconstitution process

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*To whom correspondence should be addressed at Dept. de Ingeniería Química, Universidad de Huelva, 21819, Palos de la Frontera, Huelva, Spain. E-mail: franco@uhu.es was carried out by keeping the solid egg/water weight ratio at 1:1.2. Other ingredients added to the emulsions were 3 wt% vinegar, 0.4 wt% salt, and the corresponding amount of distilled water.

Extraction of cholesterol. Cholesterol was extracted from egg yolk in a modified Jennings extraction apparatus, with subcritical CO_2 as the solvent. Subsamples of spray-dried yolk (8 g) were inserted in a Soxhlet device and extracted at 9ºC and 4.66×10^6 Pa. CO_2 gas was put in contact with a refrigerated metallic plate to induce condensation at the operating conditions. Batch extractions were conducted for different operating times (20, 65, 115, and 200 h) in order to obtain different cholesterol contents in the final product. The term α_{chol} was calculated as the relative amount of cholesterol removed (Table 1). Different samples of leached yolk submitted to the same operation time were blended to obtain a homogeneous reduced-cholesterol product.

Emulsion preparation. Emulsions were prepared by using a rotor-stator turbine, Ultra Turrax T-50 (IKA Instruments, Staufen, Germany) equipped with a S50G-45F dispersing tool. Vegetable oil was added to a dispersion of reduced-cholesterol spray-dried egg yolks and the rest of ingredients, all at room temperature. The mixture was stirred at 6,000 rpm for 3.5 min. Once prepared, emulsions were stored at 5ºC in a refrigerator. Selected emulsions were prepared five times in order to establish the variability of the emulsification technique.

Compositional analysis. The compositions of the different egg products used as emulsifiers are shown in Table 1 as a function of α_{chol}. A significant amount of nonpolar lipids also was extracted from fresh egg yolk along with the cholesterol, so the more reduction of cholesterol in the egg yolk, the greater the concentrations of proteins and phospholipids. The total lipid content was determined by extracting with chloroform/methanol (AOAC 17014-17016) (15). This extract was then used to determine cholesterol concentration according to Beyer *et al.* (16), using a Shimadzu Model GC-14A gas chromatograph with an SP-2380 column. Protein content was determined by applying the Kjeldahl method (AOAC 17008) (15). Finally, moisture was determined by vacuum oven drying (AOAC 16002) (15).

Droplet size distribution (DSD). DSD measurements were determined by laser light scattering by using a Malvern Mastersizer X-analyzer (Malvern Instruments Ltd., Malvern, United Kingdom) in a range of droplet sizes between 0.1 and 300 µm. Values of the Sauter diameter ($d_{3,2}$), which is inversely proportional to the specific surface area of the droplets, was obtained as follows:

$$d_{3,2} = \frac{\Sigma n_i d_i^3}{\Sigma n_i d_i^2} \qquad [1]$$

where n_i is the number of droplets having a diameter d_i.

Rheological tests. Oscillatory shear experiments, inside the linear viscoelastic region (up to 2–15 Pa, depending on the emulsion), and steady-state flow tests were performed in a RS-100 controlled-stress rheometer (Haake, Karlsruhe, Germany) in a range of 10^{-2}–10^2 rad/s and 10^{-5}–50 s^{-1}, respectively. A cone–plate geometry (4° angle, 60 mm diameter) was used for dynamic shear tests, whereas steady-state flow measurements were done by using a serrated plate–plate geometry (35 mm, 1-mm gap) to prevent slip effects. In addition, a CV100 controlled-shear-rate rheometer (Haake) was used to obtain additional data of steady-state viscosity in a range of 0.02–50 s^{-1} with a serrated plate–plate geometry (20 mm, 1-mm gap). All samples were submitted to the same thermomechanical history. The measuring device was covered with a thin layer of low-viscosity vaseline oil to prevent evaporation. All measurements were done 2 d after manufacturing emulsions at 25°C. At least three replicates of each test were performed on fresh samples.

Overall design and statistical analyses. The independent variable evaluated in this study was the relative amount of cholesterol removed from egg yolk (α_{chol}). This variable was modified in emulsions stabilized with either different total egg yolk concentrations or different concentrations of the surface-active components (proteins and phospholipids). Also, the effect of α_{chol} was evaluated in emulsions with different oil-weight percentages (70 and 77.5 wt%). The dependent variables considered were the Sauter diameter, the zero-shear rate-limiting viscosity, and the plateau modulus, all of them defined below. A statistical analysis, ANOVA (17), was done in order to establish the influence of α_{chol} on the dependent variables. The significance level was set at 95%.

RESULTS AND DISCUSSION

Rate of extraction. The experimental evolution of the relative amount of cholesterol removed with the operating time during extraction showed a linear variation of this parameter, which implies a constant rate of extraction in the experimen-

TABLE 1
Composition (total wt%) of the Cholesterol-Reduced Egg Yolk Samples Studied as a Function of Cholesterol Reduction Level (α_{chol})

Components	α_{chol} = 0%	α_{chol} = 14%	α_{chol} = 24%	α_{chol} = 43%	α_{chol} = 81%
Total lipids	47.0	40.1	36.0	28.0	14.9
Proteins	43.4	49.8	53.5	61.1	73.0
Cholesterol	2.1	1.8	1.6	1.2	0.4
Moisture	3.1	3.5	3.7	4.0	5.0
Ash	4.4	4.8	5.2	5.7	6.7

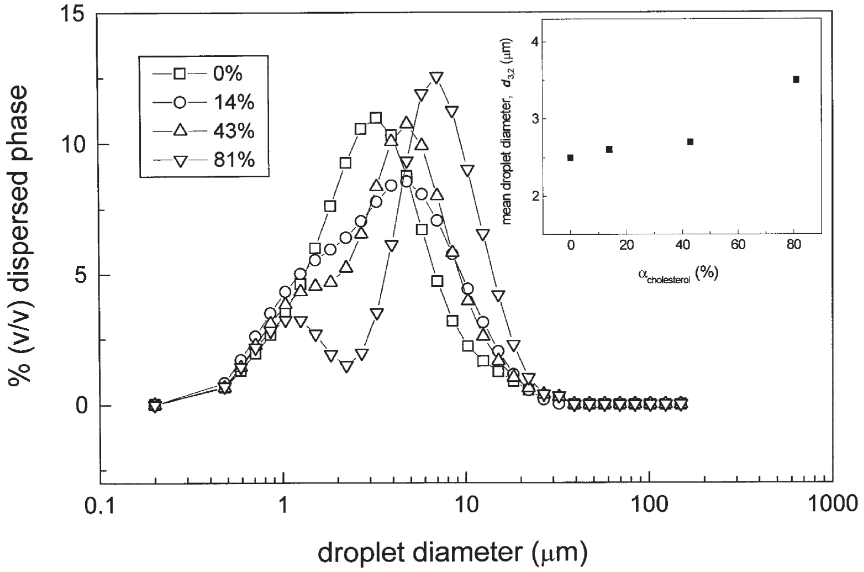

FIG. 1. Influence of the level of cholesterol extraction on the droplet size distribution curves and Sauter's diameter, $d_{3,2}$, for emulsions containing 70 wt% oil and 3 wt% protein.

tal range of the operating times studied (data not shown). The following empirical correlation describes this evolution ($R^2 > 0.99$):

$$\alpha_{chol} = 0.40\, t \qquad [2]$$

where t is the operating time in hours. Similar results were obtained by Roselló *et al.* (18) concerning the kinetics of cholesterol extraction with subcritical CO_2 of different densities as the solvent.

DSD. Figure 1 shows DSD curves for selected emulsions containing 70 wt% oil and the same concentration of the main surface-active components, proteins and phospholipids (i.e., 3 wt% proteins), as a function of the level of cholesterol reduction. The protein fraction and the phospholipids are not extracted from egg yolk by CO_2 and, consequently, if the concentration of these components is held constant, the total amount of solid egg yolk becomes lower as the level of cholesterol reduction increases. As can be observed in Figure 1, droplet size and polydispersity increased with the level of cholesterol reduction, and the maximum of the distribution appeared at larger diameters. A bimodal distribution was found at the highest level of extraction studied (i.e., 81 wt% reduction of cholesterol), showing a secondary maximum located at low values of the droplet diameter and the maximum of the distribution at relatively large sizes. In addition, as shown in Figure 1, the Sauter diameter continuously increased with the level of cholesterol reduction, changes also found at different oil and protein concentrations (70–77.5% oil, 1.3–3.5 wt% protein, data not shown).

Tables 2 and 3 show the evolution of the Sauter diameter with the level of cholesterol reduction for emulsions containing different egg yolk and oil concentrations. A higher level

TABLE 2
Influence of the Level of Cholesterol Extraction (α_{chol}) on Sauter's Diameter, Zero-Shear Rate-Limiting Viscosity, and Plateau Modulus[a]

Egg yolk concentration (wt%)	α_{chol} (%)	d_{sv} (μm)	η_0 (× 10^5) (Pa·s)	G_N^0 (Pa)
3	0	4.7^{a}	0.86^{a}	58^{a}
	14	4.3^{b}	0.72^{b}	58^{a}
	24	3.9^{c}	0.95^{a}	60^{a}
	43	3.9^{c}	0.71^{b}	65^{b}
	81	4.6^{a}	0.68^{b}	55^{a}
4	0	4.0^{a}	1.31^{a}	117^{a}
	14	3.5^{b}	1.05^{b}	102^{a}
	24	3.5^{b}	1.04^{b}	109^{a}
	43	3.2^{b}	1.63^{c}	143^{b}
	81	3.5^{b}	1.81^{c}	155^{b}
5	0	3.1^{a}	1.62^{a}	177^{a}
	14	3.0^{a}	2.01^{a}	151^{b}
	24	2.9^{a}	2.05^{a}	179^{a}
	43	2.7^{b}	2.60^{b}	200^{c}
	81	3.0^{a}	4.02^{c}	231^{c}
6	0	2.8^{a}	2.60^{a}	230^{a}
	14	2.6^{b}	3.89^{b}	261^{b}
	24	2.6^{b}	4.67^{b}	285^{b}
	43	2.5^{b}	5.02^{c}	315^{c}
	81	2.8^{a}	9.49^{d}	429^{d}
7	0	2.5^{a}	3.76^{a}	350^{a}
	14	$2.4^{a,b}$	6.54^{b}	394^{b}
	24	2.3^{b}	7.96^{c}	413^{b}
	43	2.2^{b}	8.76^{c}	471^{c}
	81	2.6^{a}	14.10^{d}	651^{d}
8	0	2.4^{a}	10.24^{a}	700^{a}
	14	2.3^{b}	14.26^{b}	833^{b}
	24	2.2^{b}	15.77^{b}	846^{b}
	43	2.1^{b}	16.17^{b}	932^{c}
	81	2.5^{a}	17.49^{b}	990^{c}

[a] For emulsions containing 70 wt% oil and different cholesterol-reduced egg yolk concentrations. Different roman superscripts (a–d) within the same column for each egg yolk concentration indicate that the values differ significantly ($P < 0.05$).

TABLE 3
Influence of the Level of Cholesterol Extraction (α_{chol}) on Sauter's Diameter, Zero-Shear Rate-Limiting Viscosity, and Plateau Modulus[a]

Egg yolk concentration (wt%)	α_{chol} (%)	d_{sv} (µm)	η_0 (× 10^5) (Pa·s)	G_N^0 (Pa)
3	0	3.1^a	5.65^a	290^a
	14	3.0^b	5.42^a	260^b
	24	2.7^b	7.21^b	307^a
	43	2.6^b	7.19^b	337^a
	81	3.2^a	3.14^c	243^b
4	0	2.8^a	6.96^a	440^a
	14	2.8^a	9.82^b	480^a
	24	2.5^b	10.97^b	591^b
	43	2.5^b	13.80^c	671^c
	81	2.9^a	10.52^b	601^b
5	0	2.4^a	11.66^a	750^a
	14	2.6^b	20.45^b	105^b
	24	2.5^b	26.93^c	1235^c
	43	2.2^c	27.00^c	1370^c
	81	2.8^d	21.74^b	1100^b

[a] For emulsions containing 77.5 wt% oil and different cholesterol-reduced egg yolk concentrations. Different roman superscripts (a–d) within the same column for each egg yolk concentration indicate that the values differ significantly ($P < 0.05$).

of cholesterol reduction initially produced an important decrease in the Sauter diameter, generally yielding a minimum for the emulsions prepared with a 43 wt% cholesterol-reduced egg yolk. Increased concentration of surface-active agents reduce the surface tension and, consequently, make the disruption of droplets easier (19). In contrast, emulsions stabilized by highly cholesterol-reduced egg yolk (81 wt% of cholesterol reduction) show higher values of the Sauter diameter, similar to those found in emulsions stabilized with egg yolk containing the highest cholesterol concentration. Concerning these higher values of droplet size, similar results were found in emulsions stabilized by cholesterol-reduced yolk when organic solvents (5) or supercritical CO_2 (10) was used for the extraction. These results were attributed to a lower capacity of egg yolk lipoproteins to stabilize the emulsion, owing to altered conformation when the associated lipids were removed. In addition, the slightly higher water content (Table 1) may reduce the stabilizing effect at the interface.

Flow behavior. All emulsions showed a qualitatively similar flow behavior (Fig. 2). Thus, a shear-thinning viscous flow curve with a tendency to a constant zero-shear-rate-limiting viscosity, η_0, always was observed. As has been reported for similar food emulsions (20), this behavior is a consequence of a dramatic shear-induced structural breakdown, related to a mechanism of oil droplet deflocculation. As was pointed out for spray-dried egg yolk-stabilized emulsions (13), the Carreau model satisfactorily describes this behavior:

$$\frac{\eta - \eta_\infty}{\eta_0 - \eta_\infty} = \frac{1}{[1 + (\dot{\gamma}/\dot{\gamma}_c)^2]^s} \qquad [3]$$

where $\dot{\gamma}_c$ is the critical shear rate for the onset of the shear-thinning region, s is a parameter related to the slope of this region, and η_∞ is the high-shear rate-limiting viscosity, included to fit the slight tendency to reach a high shear rate plateau region. In all cases, s remained almost constant (0.45 ± 0.01).

Figure 2 shows steady-state flow curves for emulsions, stabilized by egg yolk submitted to different levels of cholesterol reduction, containing 3 wt% proteins. Values of more significant fitting parameters (η_0 and $\dot{\gamma}_c$) are also shown in

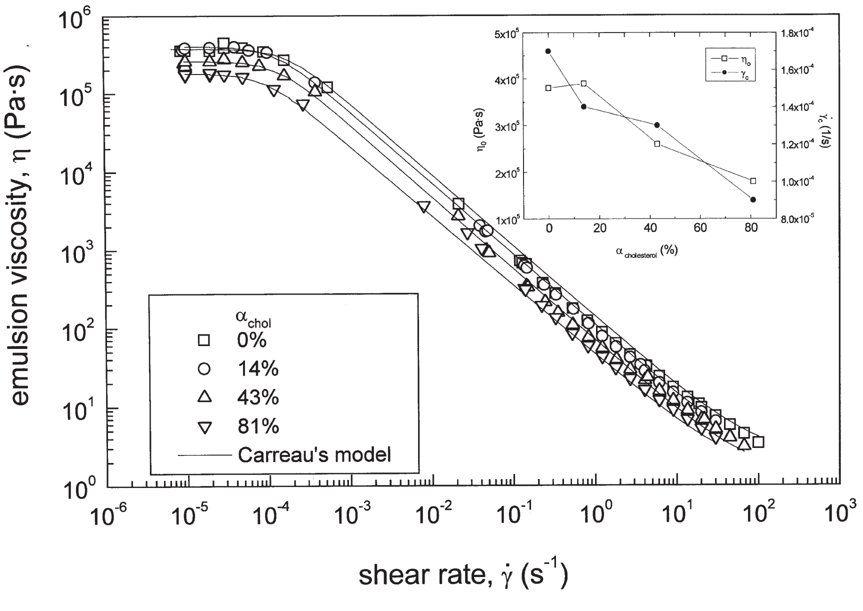

FIG. 2. Steady-state flow behavior and values of the zero-shear rate-limiting viscosity, η_0, and critical shear rate, $\dot{\gamma}_c$, for emulsions containing 70 wt% oil and 3 wt% protein, as a function of the level of cholesterol extraction.

Figure 2. Viscosity significantly ($P < 0.05$) decreased with cholesterol reduction. Thus, for instance, a reduction of around 50% in the values of the zero-shear rate-limiting viscosity, η_0, was seen when 81 wt% cholesterol was removed (Fig. 2). A similar decrease ($P < 0.05$) in the values of the critical shear rate for the onset of the shear-thinning region ($\dot{\gamma}_c$) was found, which indicates a lower resistance of the unperturbed emulsion microstructure to the shear-induced breakdown process.

Nevertheless, an opposite evolution was observed for emulsions stabilized by the same amount of total spray-dried yolk. The zero-shear rate-limiting viscosity, (η_0,) generally increased with the level of cholesterol reduction, in association with a higher content of surface-active agents and lower droplet size (Tables 2 and 3). However, emulsions containing 77.5% oil (Table 3) showed maximal values of η_0 for dispersions stabilized by a 43 wt% cholesterol-reduced yolk, which also showed minimal Sauter's diameter values. A further increase in the level of cholesterol reduction (i.e., 81 wt%) yielded a decrease in viscosity, mainly apparent at the lowest egg yolk concentration. As was previously mentioned, a higher level of cholesterol reduction induced a significant conformational change of the lipoprotein structure, which may affect the rheological properties of the emulsions. Similar results were found by other authors, who have compared some functional properties of food emulsions stabilized with low-in-cholesterol egg yolk ($\alpha_{chol} = 77\%$) with those of native yolk-stabilized emulsions (10). However, the effect of this conformational change on the viscous properties of these emulsions also depends on oil concentration. Thus, emulsion viscosity may continuously increase with the level of cholesterol reduction for systems containing 70 wt% oil. Nevertheless, an initial decrease in viscosity may be found at low egg-yolk concentrations. Emulsions stabilized by 14 wt% cholesterol-reduced egg yolk, at low concentrations (3 and 4 wt%) showed values of η_0 slightly lower than those shown by emulsions stabilized with the same concentration of unleached spray-dried yolk. This fact means that the structural rearrangement induced by cholesterol removal mainly influences the rheological properties of emulsions when the surface-active components are present in the oil–water interface at low concentrations.

Linear viscoelastic behavior. Figure 3 shows the evolution of the storage (G') and loss (G'') moduli with frequency for emulsions containing 70 wt% oil and 3 wt% protein as a function of cholesterol concentration. The storage modulus always was higher than the loss modulus in the frequency range studied. A well-developed plateau region with a minimum in G'' and a slight frequency dependence of G' was observed in all cases. This behavior is characteristic of protein-stabilized emulsions in which an elastic network develops that is due to the occurrence of an extensive bridging flocculation process (12,20). The plateau modulus, G_N^0 defined for polymers as the extrapolation of the entanglement contribution to the viscoelastic functions at high frequencies (21), can be analyzed as a characteristic parameter of this region. This parameter is considered as a measure of the density of entanglements among polymeric molecules (21) but may be related to the interactions among emulsifier molecules located at the oil–water interface of adjacent droplets, favoring the formation

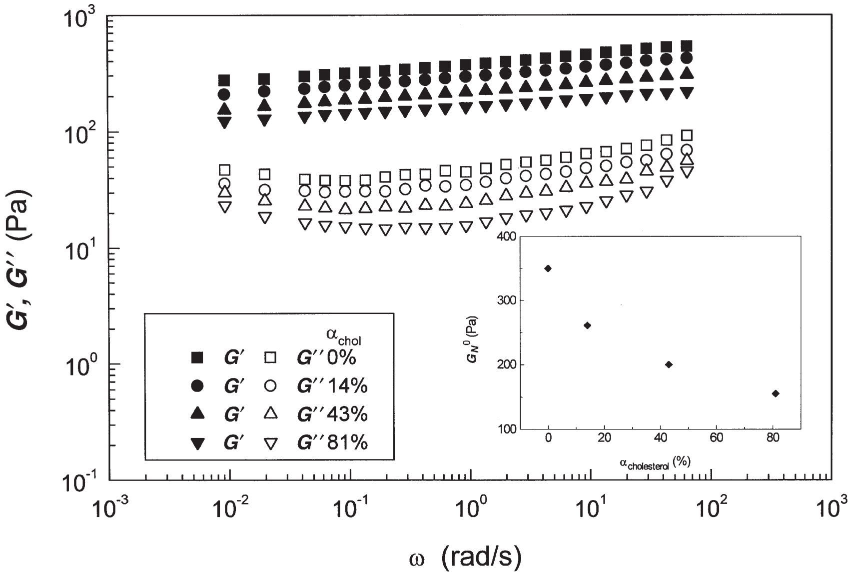

FIG. 3. Evolution of the storage, G', and loss moduli, G'', with frequency and values of the plateau modulus, G_N^0, for emulsions containing 70 wt% oil and 3 wt% protein, as a function of the level of cholesterol extraction.

of the above-mentioned structural network. G_N^0 may easily be easily estimated from the loss tangent ($\tan\delta = G''/G'$) as follows (20):

$$G_N^0 = [G']_{\tan\delta\rightarrow\text{minimum}} \qquad [4]$$

Although there are more accurate methods to calculate the plateau modulus, this method provided values that were not appreciably different despite its simplicity.

The linear viscoelastic functions, as well as the plateau modulus, of the emulsions studied significantly decreased with egg-yolk cholesterol concentration (Fig. 3) for systems having a constant concentration of surface-active components. Thus, for instance, a reduction of more than 50% in the values of G_N^0 was found (Fig. 3) for an emulsion stabilized by a highly cholesterol-reduced egg yolk (81 wt%). However, as can be observed in Tables 2 and 3, an opposite evolution generally was found in emulsions containing the same amount of cholesterol-reduced yolk. Once again, the generalized increase in viscoelastic parameters must be explained by an increase in the concentration of surface-active components as cholesterol and lipids are removed. In the same manner, a conformational alteration of lipoproteins induced by the cholesterol reduction seems to affect emulsions with higher oil content or lower yolk concentrations, predominantly using the maximum cholesterol-reduced egg yolk (81 wt%). In addition, a decrease in G_N^0 values may be observed for emulsions stabilized by a lower concentration of 14 wt% cholesterol-reduced yolk in comparison to those shown by fresh, spray-dried yolk-stabilized emulsions (Tables 2 and 3). All the results were quite similar to those found when the samples were submitted to steady shear tests.

Structural considerations. If the total amount of cholesterol-reduced egg yolk is fixed in the emulsion formulation, a higher level of cholesterol extraction implies a higher concentration of the main surface-active agents, lipoproteins and phospholipids, which favors a reduction in surface tension and, generally, lower mean droplet diameter (19). This process yields higher values of the viscous and linear viscoelastic functions of the emulsions. Mean droplet diameters similar to those found for unleached yolk-stabilized emulsions, however, were obtained using a highly cholesterol-reduced egg yolk (81 wt%), although this effect was not always associated with a reduction in the viscous and linear viscoelasticity functions (Table 3). Consequently, the rheological behavior of these emulsions cannot be explained only by taking into account the influence of droplet diameter. It is well known that bulk rheology of emulsions is highly dependent on interdroplet interactions, which are influenced by droplet diameter as well as composition and physical properties of the interfacial layer (22,23). Thus, a high level of cholesterol extraction induces both a slight increase in the moisture content and a conformational alteration of lipoproteins. These two phenomena are mainly relevant at high oil concentrations or low egg yolk contents, being related to a decrease in the interfacial viscoelasticity when the interfacial layer is not still saturated (22).

This effect may be confirmed if emulsions are prepared by adding the same amount of egg yolk surface-active agents (proteins and phospholipids). In this case, a decrease in the emulsion rheological parameters, accompanied by an increase in mean droplet diameter with cholesterol content, always was found. The free nonpolar lipid fraction extracted from egg yolk does not significantly affect the surface activity of egg yolk, which is mainly due to lipoproteins and phospholipids (2,5). As has been pointed out (14), however, lipoprotein complexes are structurally breakable. Thus, the total or partial extraction of cholesterol, together with the slightly higher water content, may affect the adsorption properties and consequently modify the interfacial layer, which implies different interdroplet interactions (8,9,19). This structural weakness is especially relevant for LDL. These LDL consist of 85% lipids, 70% of which are nonpolar lipids that are susceptible to extraction by CO_2. In addition, although the way in which cholesterol, lipoproteins, and phospholipids interact among themselves is still not clear, some authors (19) have indicated that the role of cholesterol molecules is to improve PE interactions at the interface. Consequently, a decrease in cholesterol content would reduce the interfacial properties of these phospholipids.

ACKNOWLEDGMENTS

This work is part of a research project (ALI96-0892) sponsored by the Comisión Interministerial de Ciencia y Technología, Spain. The authors gratefully acknowledge its financial support.

REFERENCES

1. Dickinson, E., and G. Stainsby, *Colloids in Food*, Applied Science Publishers, London, 1982, pp. 120–133.
2. Baldwin, R.E., Functional Properties of Eggs in Foods, in *Egg Science and Technology*, edited by W.J. Stadelman and O.J. Cotterill, Food Products Press, New York, 1990, pp. 341–383.
3. Bringe, N.A., and J. Cheng, Low-Fat, Low-Cholesterol Egg Yolk in Food Applications, *Food Technol. 49*:94–106 (1995).
4. Kiosseoglou, V., Egg Yolk, *Dev. Food Sci. 19*:63–91 (1989).
5. Paraskevopoulou, A., and V. Kiosseoglou, Cholesterol and Other Lipid Extraction from Egg Yolk Using Organic Solvents: Effects on Functional Properties of Yolk, *J. Food Sci. 59*:766–768 (1994).
6. Larsen, J.E., and G.W. Froning, Extraction and Processing of Various Components from Egg Yolk, *Poult. Sci. 60*:160–165 (1981).
7. Paraskevopoulou, A., V. Kiosseoglou, S. Alevisopoulos, and S. Kasapis, Influence of Reduced-Cholesterol Yolk on the Viscoelastic Behaviour of Concentrated O/W Emulsions, *Colloids Surf. B 12*:107–111 (1999).
8. Paraskevopoulou, A., V. Kiosseoglou, S. Alevisopoulos, and S. Kasapis, Small Deformation Properties of Model Salad Dressings Prepared with Reduced Cholesterol Egg Yolk, *J. Texture Stud. 28*:221–237 (1997).
9. Paraskevopoulou, A., V. Kiosseoglou, and S. Pegiadou, Interfacial Behavior of Egg Yolk with Reduced Cholesterol Content, *J. Agric. Food Chem. 45*:3717–3722 (1997).
10. Paraskevopoulou, A., K. Panayiotou, and V. Kiosseoglou, Functional Properties of Egg Yolk Concentrate with a Reduced Cholesterol Content Prepared by Treatment with Supercritical CO_2, *Food Hydrocoll. 11*:385–391 (1997).
11. Seideman, W.E., and O.J. Cotterill, Ion-Exchange Chromatog-

raphy of Egg Yolk. 2. Heating, Freezing and Spray-Drying, *Poult. Sci. 48*:894–900 (1969).

12. Franco, J.M., A. Raymundo, I. Sousa, and C. Gallegos, Influence of Processing Variables on the Rheological and Textural Properties of Lupin Protein-Stabilized Emulsions, *J. Agric. Food Chem. 46*:3109–3115 (1998).
13. Moros, J.E., J.M. Franco, and C. Gallegos, Rheology of Spray-Dried Egg Yolk-Stabilized Emulsions, *Int. J. Food Sci. Technol. 37*:297–307 (2002).
14. Guerrero, A., and H.R. Ball, Effect of Spray-Dried or Reduced-Cholesterol Yolk and Temperature on the Linear Viscoelastic Properties of Mayonnaise, *J. Texture Stud. 25*:363–381 (1994).
15. AOAC, *Official Methods of Analysis* of *AOAC International*, Association of Official Analytical Chemists, Arlington, 17th edition, 2002.
16. Beyer, J.D., F.X. Milani, M.J. Dutelle, and R.L. Bradley, Gas Chromatographic Determination of Cholesterol in Egg Products, *J. Assoc. Off. Anal. Chem. 72*:746–748 (1989).
17. Montgomery, D.C., *Diseño y análisis de experimentos*, Grupo Editorial Iberoamericana, México D.F., 1991, pp. 45–81.
18. Roselló, A., J. Cota, J.M. Moraga, and F. Carrillo, The Rate of Extraction of Cholesterol from Egg Yolk Using Liquid and Near-Critical CO_2, *Inf. Tecnol. 10*:93–96 (1999).
19. Mine, Y., and M. Bergougnoux, Effect of Cholesterol Reduction from Hen's Egg Yolk Low-Density Lipoproteins on Its Emulsifying Properties, in *Food Emulsions and Foams*, edited by E. Dickinson and J.M. Rodríguez, Elsevier Applied Science, London, 1998, pp. 129–139.
20. Franco, J.M., P. Partal, D. Ruiz-Marquez, B. Conde, and C. Gallegos, Influence of pH and Protein Thermal Treatment on the Rheology of Pea Protein-Stabilized Emulsions, *J. Am. Oil Chem. Soc. 77*:975–983 (2000).
21. Baumgaertel, M., M.E. De Rosa, J. Machado, M. Masse, and H.H. Winter, The Relaxation Time Spectrum of Nearly Monodisperse Polybutadiene Melts, *Rheol. Acta 31*:75–82 (1992).
22. Graham, D.E., and M.C. Phillips, Proteins at Liquid Interfaces, *J. Colloid Interface Sci. 76*:240–250 (1980).
23. Kiosseoglou, V., and P. Sherman, The Influence of Egg Yolk Lipoproteins on the Rheology and Stability of O/W Emulsions and Mayonnaise. 3. Viscoelastic Properties of Egg Yolk Films at the Groundnut Oil–Water Interface, *Colloid Polym. Sci. 261*: 520–526 (1983).

[Received August 15, 2001; accepted May 12, 2002]