

# Protein interactions and filler effects in heat-set gels based on egg

A. Kalkani, A. Paraskevopoulou, V. Kiosseoglou\*

*Laboratory of Food Chemistry and Technology, School of Chemistry, Aristotle University of Thessaloniki, 54 124 Thessaloniki, Greece*

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## Abstract

The effect of incorporation of emulsified oil with yolk on the strength of gel network systems, produced by heating of egg yolk or egg white dispersions, was investigated. The yolk gels exhibited a decrease in their mechanical strength as a result of oil incorporation. The white-based gels, on the other hand, showed a higher strength, following incorporation of oil droplets, the result depending on the droplet size but more on oil levels. The interaction between the globular proteins of egg white and the lipoprotein-covered oil droplet surfaces was established by applying SDS-PAGE electrophoresis. The results are discussed in terms of the differences in structure between the yolk and the white proteins that may affect the way they denature and interact following heating.

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**Keywords:** Egg white; Egg yolk; Emulsion gel; Filler effects; Protein interactions

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## 1. Introduction

Egg constitutes a highly complex food system both in terms of composition and physicochemical structure. The main constituents of egg are lipids and proteins of exceptionally high biological value which also exhibit remarkable functional properties. As a result of this, whole egg or its fractions, the yolk and the white, are extensively used as functional ingredients in foods such as salad dressings, cakes, omelets, sauces, pie filling, confectionery, meat products, etc. where they play an important role in product preparation as well as in improving its physicochemical stability (Kiosseoglou, 1989, 2003a,b; Mine, 2002; Powrie & Nakai, 1984). One key functional property of egg proteins that determines the rheological and textural characteristics of foods such as heat-set creams, omelets, cakes, etc., is their ability to coagulate and form a gel network exhibiting a solid-like behavior (Kiosseoglou, 2003a,b; Paraskevopoulou, Alevisopoulos, Kasapis, & Kiosseoglou, 2000). Both the white and the yolk tend to form upon heating gel structures. The egg white fraction, however, produces stronger and more resilient gel networks compared to yolk, when heated at relatively high tempera-

tures, i.e. above 85 °C where ovalbumen denaturation takes place (Woodward, 1990). The yolk, on the other hand, is well known for its remarkable emulsifying ability, attributed to the presence of lipoproteins. More specifically, the apolipoprotein molecules of the lipovitellenins, the main constituents at the yolk micelles, exhibit a high surface-penetrating power, due to their structure flexibility, that enables the molecules to rapidly adsorb and rearrange at the oil–water interface of the emulsion droplets (Aluko, Keeratiurai, & Mine, 1998; Kiosseoglou & Sherman, 1983; Martinet, Beaumal, Dalgalarondo, & Anton, 2002). On the other hand, globular and less flexible proteins, such as those of egg white or the yolk plasma livetins, adsorb less effectively at the droplet surfaces in the presence of yolk lipoproteins and are competitively excluded from the droplet surfaces (Shenton, 1979). In food emulsion systems containing whole egg, therefore, it is expected that the absorbed membrane around the droplets will be built up mainly by yolk lipoproteins while the continuous phase of the emulsion is expected to be dominated by the non-adsorbing egg white and the yolk plasma globular proteins. When such a system is heat-treated an emulsion gel structure results made up of oil droplets embedded in a gel structure. Depending on parameters such as droplet size and droplet surface network interactions, the droplets may act as fillers and modify the mechanical and textural

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\*Corresponding author. Tel.: +30 231 0 997834; fax: +30 231 0 997779.  
E-mail address: [kiosse@chem.auth.gr](mailto:kiosse@chem.auth.gr) (V. Kiosseoglou).

properties at the resulting gel. Examples of products where such a situation may be encountered are foods such as creams, cake batters and pie-fillings. Furthermore, a better understanding of the role of yolk-stabilized emulsion gels may aid in the development of novel gel structures where the emulsifying potential of yolk lipoproteins with the gel-forming ability of egg albumen are combined.

A prerequisite for emulsified oil/fat particles to perform as active fillers in a protein gel network is the interaction between the absorbed protein membrane and the proteins of the network (Aguilera & Kessler, 1988; Aguilera, 1992; Dickinson & Hong, 1995; Matsumura, Kang, Sakamoto, Motoki, & Mori, 1993; Shenton, 1979). As Anton, Le Denmat, Beaumal, and Pilet (2001) pointed out, the incorporation of oil droplets of a mean size of about 1  $\mu\text{m}$ , appears to have a weakening effect on a yolk-based gel structure. According to these workers the negative effect of oil droplets on the mechanical properties of the gel was the result of the domination of the yolk gelation mechanism by the micellar low-density lipoproteins of the yolk and the absence of any interaction of the gel network proteins with the yolk lipoprotein-covered droplet surfaces. Similar results were reported by Koidis, Paraskevopoulou, and Kiosseoglou (2002) for a gel network based on yolk protein concentrate and incorporating oil droplets of a mean diameter of around 2  $\mu\text{m}$ . Since, however, the oil droplet size is one parameter that may have an effect on the filler properties of the droplets incorporated in the protein gel (Dickinson & Hong, 1995), an attempt was made in the present investigation to reduce the droplet diameter to submicron dimensions by applying high pressure homogenization in order to check whether the negative effect of droplets with respect to mechanical properties in the previous investigations might have been the lack of gel network–droplet surface interactions and not the relatively large diameter of the droplets. The main target of this work, however, was to investigate the role of oil droplets stabilised with yolk in gel network systems based on egg white proteins, in an attempt to advance our knowledge about the formation and the mechanical properties of heat-set products that contain both whole egg and oil/fat.

## 2. Materials and methods

### 2.1. Materials

Dehydrated egg white, grade II (CAS 9006-59-1) and freeze-dried BSA (CAS 9048-46-8) were purchased by Sigma (Steinheim, Germany). Egg yolk in the liquid form was obtained by first breaking fresh hen's eggs and, following complete removal of the adhering white by rolling the intact yolks on tissue paper, the vitelline membrane was punctured and the liquid yolks of a number of eggs were collected, pooled and stored at 2 °C in the presence of 0.02%  $\text{NaN}_3$ . Refined corn oil was bought from the local market. All the reagents used were of analytical grade.

### 2.2. Emulsion preparation

Oil-in-water emulsions were prepared by first dispersing the liquid yolk in phosphate buffer (1/15 M  $\text{Na}_2\text{HPO}_4$ , 1/15 M  $\text{KH}_2\text{PO}_4$ ) at pH 7. The oil was then added under continuous stirring with the aid of a mechanical stirrer and the resulting crude emulsion was finally homogenized by employing either in APV (Type: 0401005) pressure homogenizer (Albertslund, Denmark) or in Ultra-Turrax T25 mechanical homogenizer (IKA Labortechnik, Staufen, Germany) to obtain a number of dispersions varying in droplet size.

### 2.3. Mean droplet size determination

To determine the emulsion droplet size, 1 g of each sample was dispersed in 500 mL of distilled water in the measuring vessel of a light scattering instrument (Malvern Mastersizer, Worcestershire, UK) under continuous stirring at 3000 rpm for 2 min. Droplet size distributions and average droplet diameter,  $d_{32}$ , were calculated by using the following parameters: refractive index of corn oil and water: 1.4673 and 1.33, respectively; absorption: 0.002. The particle size distributions were monomodal as determined by light scattering and the droplets were not flocculated when observed under the light microscope.

### 2.4. Gel preparation

To obtain the gel sample based on yolk, the liquid yolk was first dispersed in phosphate buffer of pH 7 and the resulting dispersion (500 mg/mL in liquid yolk) was stirred for 30 min with the aid of a magnetic stirrer. The mixture was then placed in cylindrical cells of 1 cm diameter constructed from aluminum foil and reinforced with plastic tape. The ends were tightly sealed and the cells were immersed in a water bath at 90 °C for 30 min. The cells were then removed from the bath, stored overnight at room temperature, the aluminum foil was removed and the gel was cut to obtain cylindrical samples 1 cm in height. Emulsion gels based on yolk were prepared as described above from a liquid yolk-buffer mixture incorporating 15% corn oil in the form of emulsified with yolk oil droplets of a mean size ( $d_{32}$ ) of 0.15  $\mu\text{m}$ , obtained by homogenization of the crude emulsion with the aid of the APV at 800 bar.

To prepare gel samples based on egg white or BSA, the appropriate quantity of the dehydrated protein was dispersed in the phosphate buffer and mixed thoroughly with the aid of a magnetic stirrer for 1 h, followed by heating in cylindrical cells at 90 °C for 30 min. To obtain egg white gels incorporating emulsified oil droplets a series of o/w emulsions containing 15% corn oil and 2% liquid yolk were first prepared and homogenized either with the APV or with the Ultra-Turrax homogenizer at different pressure or speed values, respectively. Dehydrated egg white was then incorporated in the emulsion and

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