

The role of Tween in inhibiting heat-induced destabilization of yolk-based emulsions

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Abstract

The process of heat-induced destabilization of yolk-based emulsions and the role of Tween addition in inhibiting droplet aggregation/coalescence in the thermally treated emulsions were investigated. The aim of the study was to understand the mechanism behind yolk emulsion destabilization during the application of processes such as pasteurization/sterilization and/or cooking. Data on emulsion particle size distribution were combined with results on yolk protein adsorption to clarify the role of the unadsorbed yolk protein fraction in the destabilization of the thermally treated emulsion. Surface tension measurements were also conducted to investigate yolk protein–Tween interactions at the air/water interface and their effect on emulsion stability. The presence in the emulsion continuous phase of unadsorbed yolk protein is crucial for the thermal destabilization of the system. Tween addition inhibits droplet flocculation/coalescence phenomena by shielding the reactive groups of protein molecules adsorbed at the droplet surfaces and those of unadsorbed proteins in the emulsion continuous phase which become available for interaction following heating and protein denaturation.

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1. Introduction

Egg yolk is a key ingredient of a number of food products. Its importance stems from the fact that it is a material rich in proteins of high biological value and lipids made up of essential ω -3 fatty acids. On the other hand, the functional properties of yolk e.g. emulsifying and thickening ability, attributed mainly to its lipoprotein constituents, make the yolk an indispensable ingredient in the preparation of certain semi-liquid products e.g. dressings and creams, where the dispersion of relatively high amounts of oil/fat in the form of emulsified droplets is required. (Kiosseoglou, 2003).

Protein-based emulsions, including those of yolk, are often subjected to thermal treatment for pasteurization/sterilization or cooking purposes. Heat processing of emulsions at temperatures above the denaturation point of the stabilizing protein may result in the physicochemical

destabilization (droplet aggregation and/or coalescence) of emulsion systems (Demetriades, Coupland, & McClements, 1997; McSweeney, Mulvihill, & O'Callaghan, 2004). According to Kim, Decker, and McClements (2002), droplet aggregation in heat-treated β -lactoglobulin emulsions at temperatures above the protein denaturation point, is the result of extensive unfolding of the adsorbed protein molecules that leads to exposure of reactive groups previously located in the molecule. Under suitable conditions, e.g. reduced repulsive electrostatic interaction forces, droplet aggregation may take place involving the protein film adsorbed on neighbouring droplet surfaces, which interact through hydrophobic bonds and disulfide “bridges”. Euston, Finnigan, and Hirst (2000), suggested that droplet-droplet interaction in β -lactoglobulin emulsions depends on the presence in the continuous phase of non-adsorbed protein molecules which unfold and aggregate following heat treatment. The protein aggregates then act as “glue” between adsorbed protein layers at the surface of neighbouring droplets, leading to droplets aggregation.

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Heat-induced destabilization of protein emulsions can be prevented by suitably manipulating the emulsifier composition. Thus, small additions of casein to β -lactoglobulin emulsions eliminated any heat-induced droplet aggregation phenomena (Parkinson & Dickinson, 2004) while the same was observed for soy protein emulsions, when the protein was conjugated to dextran by dry-heating (Diftis & Kiosseoglou, 2005). The improvement in stability of these systems against heat-induced aggregation was attributed to enhancement of repulsive steric forces resulting from the adsorption of casein or dextran molecules which prevented the droplets from approaching and interacting through their adsorbed protein layers. Addition of hydrophilic emulsifiers such as Tween may also aid in the improvement of stability upon heating of protein-based emulsions. This emulsifier displaces the protein molecules from the droplet surfaces and at the same time prevents non-polar protein groups from interacting through hydrophobic bonding (McClements, 2004). Parkinson and Dickinson (2004) reported that Tween addition at low levels to β -lactoglobulin-stabilized emulsions resulted in the intensification of heat-induced droplet aggregation effects. This was attributed to the displacement of some non-polar protein segments from the droplet surfaces and the enhancement of hydrophobic interaction forces between the emulsion droplets.

Egg yolk owes its remarkable emulsifying ability to its lipoprotein constituents. These highly hydrophobic proteins are organized in the liquid yolk into supramolecular structures, e.g. the LDL micelles and the HDL granules (Kiosseoglou, 1989). Following emulsification, about 80% of the yolk proteins may become adsorbed to the droplet surfaces the extent of adsorption depending on parameters such as pH and/or NaCl concentration (Anton & Gandemer, 1999). Additionally, the yolk proteins suffer extensive molecular unfolding when heated at temperatures above 70 °C (Le Denmat, Anton, & Gandemer, 1999). The aim of the present study was to gain an insight into the mechanism behind the heat-induced destabilization of yolk-based emulsions and to establish whether droplet aggregation and/or coalescence effects can be controlled or totally prevented by the addition of Tween, a well known food emulsifier. Previous studies have shown that this surfactant may bring about the disorganization of the supramolecular yolk lipoprotein structures (Paraskevopoulou & Kiosseoglou, 1995) and, additionally, aid in preventing extensive gelation phenomena in heat-treated yolk solutions, probably by interrupting hydrophobic bonds between the yolk protein constituents (Kiosseoglou & Paraskevopoulou, 2005).

2. Materials and methods

2.1. Materials

Fresh hen eggs and refined corn oil were purchased from the local market. The eggs were broken manually and the

yolks were separated from the albumen. The vitelline membrane was then pierced and the liquid yolks of a number of eggs were collected and pooled. Sodium chloride and dithiothreitol (DTT) were obtained from Sigma Chemical Co. Tween 40 (polyoxyethylene-sorbitan-monopalmitate) was a product of Fluka.

2.2. Methods

Emulsion preparation: Egg yolk suspensions (6% w/v in liquid yolk) were initially prepared by dispersing the liquid yolk in deionized water with the aid of a mechanical stirrer. The pH of the yolk suspensions was then adjusted to the value of 7 using 0.1 N NaOH or HCl solutions and the oil was gradually added to the suspension under continuous agitation for 5 min with the mechanical stirrer, to obtain a crude emulsion 30%v/v in oil. This emulsion was then homogenized for 5 min at 900 bar employing a high-pressure value homogenizer (APV-2000). Portions from this initial stock emulsion were then drawn and diluted with deionized water to obtain emulsion samples 10% and 2% in oil and liquid yolk, respectively. The emulsion pH was checked and readjusted to the value of 7 when required.

Emulsions heating experiments and particle size measurement: Heat treatment of emulsions involved placing of a small quantity (1 ml) in glass tubes and holding the tubes isothermally at selected temperatures (up to 95 °C) in a heated water bath for 10 min followed by placing in an iced water bath. The samples required about 35–45 s to reach the bath temperature and therefore zero time was set after that time period. The particle size distribution of emulsions was determined using the laser light scattering technique with the aid of a Mastersizer 2000 (Malvern Instruments) following dilution of the samples with deionized water (about 1:1000) and gently stirring in the measuring vessel of the instrument. Particle size measurements are reported as weight-average mean diameters, $d_{4,3}$, that take into account both individual droplets and droplet aggregates, and are average values of measurements made at least twice on at least three freshly prepared emulsion samples. The following optical parameters were applied: corn oil refractive index: 1.4673; adsorption: 0.002; water refractive index: 1.3300. The emulsion samples were also treated with 1% Tween and 10 mM DTT for 2 h before the application of the laser scattering technique to determine the mean size of the emulsion droplets that constitute the particle aggregates. Heat treatment experiments were also conducted following addition of 1% Tween 40 or 1% Tween plus 10 mM DTT or after the removal of the unadsorbed egg yolk from the emulsion continuous phase by centrifugation at 10,000g for 30 min. The centrifugation step was repeated until no protein was detected in the continuous phase. The resulting cream was then dispersed in deionized water to obtain an emulsion with about 10% oil and heat-treated at 90 °C for 10 min.

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