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Egg yolk hydrolysed granules: Characteristics, rheological properties and applications

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ABSTRACT

Given its low-cholesterol content feature, granules from egg yolk can be used as a substitute of the whole egg yolk. However, the functional properties of the granular fraction should be improved. In this sense, hydrolysis of proteins frequently produces improvements in some of its nutritional and technological properties. For that reason, in this work egg yolk granules were treated with a proteolytic enzyme, trypsin (E.C. 3.4.21.4) with the purpose of making a comparative characterization of the products.

Results showed that the enzymatic reaction produced a degree of hydrolysis of 12%, being the size of the different peptides obtained and quantified by chromatographic and electrophoretic techniques. Mayonnaises made with these hydrolysed granules resulted more stable to temperature changes between 4 and 20 °C than the one made with non-hydrolysed ones. In the rheological tests carried out, the mayonnaise elaborated with hydrolysed granules has the most similar rheological behaviour to that of a commercial one used as reference. In general, the results obtained suggest that the recipe elaborated with hydrolysed granules had better rheological characteristics than those prepared using non-hydrolysed granules, maintaining the low-cholesterol feature.

Keywords: Egg yolk; Granules; Hydrolysis; Emulsification; Mayonnaise; Rheological properties

1. Introduction

People's concern about the rise of cardiovascular diseases and the obesity has led the consumers to select the ingredients that form part of the food products, trying to avoid these health issues. In response to this question, the researchers have focused their attention on two particular nutritional facts, namely the lipids and cholesterol content of foods. In this sense, an ingredient with high levels of cholesterol and lipids, commonly used in the food industry, is the egg yolk.

Egg yolk is broadly recognized to contain many substances with biological functions beyond basic nutritional ones, and for that reason, its substitution is complicated and usually implies a quality loss in the final product.

However, egg yolk can be easily separated by centrifugation into two fractions: the plasma and the granular one. Plasma is mainly composed of 85% low density lipoproteins (LDL) and 15% livetins. This fraction contains about 73% of lipids and 3/4 of the whole egg yolk cholesterol. On the other hand, egg yolk granules are mainly composed of 70% high-density lipoproteins (HDLs), 16% phosvitin and 12% low density lipoproteins. This fraction is high in protein content and it has approximately 1/4 of the total cholesterol found in egg yolk (Laca et al., 2010a).

In a study about egg yolk fractionation, it has been shown that egg yolk granules keep, even after lyophilization treatment, good emulsifying, gelling and other properties, with the additional advantage of its lower cholesterol content (Laca et al., 2010a). However, the emulsifying properties of this granular fraction are lower than those found in the whole egg yolk or in the plasma fraction (Le Denmat et al., 2000).

Concerning proteins, their functional properties are those physicochemical properties that govern their performance and behaviour in food systems during their preparation, processing, storage and consumption. These properties can be enhanced through enzymatic modification of food proteins by controlled proteolysis over a wide pH range, and other processing conditions. Choosing the right proteolytic enzyme,

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environmental conditions and degree of hydrolysis (DH) is crucial for enhancing the functional properties of proteins. Owing to the complex nature of proteins it is very difficult to reach high DH values (Panyam and Kilara, 1996).

The emulsifying capacity of the egg yolk has been broadly investigated: the effect of a high-pressure treatment (Anton et al., 2001a), the stability and rheology when adding another agent (Kontogiorgos et al., 2004) or the egg yolk protein gels and emulsions (Kiosseoglou, 2003) are some examples. However, there is little information on the effect of controlled enzymatic hydrolysis of the egg yolk and its fractions on their foaming and emulsifying properties. When used as emulsifiers, the hydrolysis of the surfactant proteins, either before or after the formation of the emulsions, can affect the stability of the emulsion system by making the emulsion inherently unstable or by altering its sensitivity to external influences (e.g., calcium ions, reduced pH, or high temperature). Nevertheless, in some cases hydrolysis may even promote stability, as it has been observed in the increase in calcium stability of caseinate emulsions treated with a serine protease, trypsin. This enzyme cleaves peptide bonds at carboxyl terminals of arginine and lysine, except when linked to a proline residue (Olsen et al., 2004). Hence, if hydrolysed proteins are used as emulsifiers, there is a risk that the stabilizing effect of the protein will be lost affecting their potential applications, but, otherwise, it must be set also the possibility that the disruption of the protein structure may permit more efficient adsorption of some peptides (Singh and Dalgleish, 1998). Besides, whenever enzymatic hydrolysis of protein causes breakdown of protein molecules, the protein solubility increases, and it is well known that solubility is essential for most proteins to provide good functionalities such as foaming and emulsification.

In this work, since the granular egg yolk fraction is low in cholesterol content, and with the aim to test the possibility of enhancing the functional properties of the granular proteins, they were hydrolysed to a value close to 12% degree using trypsin (E.C. 3.4.21.4). The procedure was developed at the optimal temperature and pH for the activity of trypsin, 37 °C and pH over 7.0 (Chelulei Cheison et al., 2011). Then, the applicability of the product of hydrolysis was tested, in particular elaborating mayonnaises with non-hydrolysed and hydrolysed granules. Rheological properties were measured in order to evaluate the mayonnaises characteristics compared to those of a first quality mayonnaise acquired from a local market, which was used as a reference.

2. Materials and methods

2.1. Fractionation of egg yolk

The fractionation method was developed modifying the procedure by Laca et al. (2010a). The general scheme for egg yolk fractionation is shown in Fig. 1.

The granules fraction was frozen at −80 °C overnight and then lyophilized at −70 °C and 0.1 mBa in a Telstar Cryodos Lyophilizator for 24 h, in order to increase its shelf life.

2.2. Enzymatic hydrolysis

The hydrolysis reaction was carried out in a 5 l bioreactor with a pH-STAT automatic titration (pH-Burette 24 2S, Crison) connected to an iso-thermal shaker.

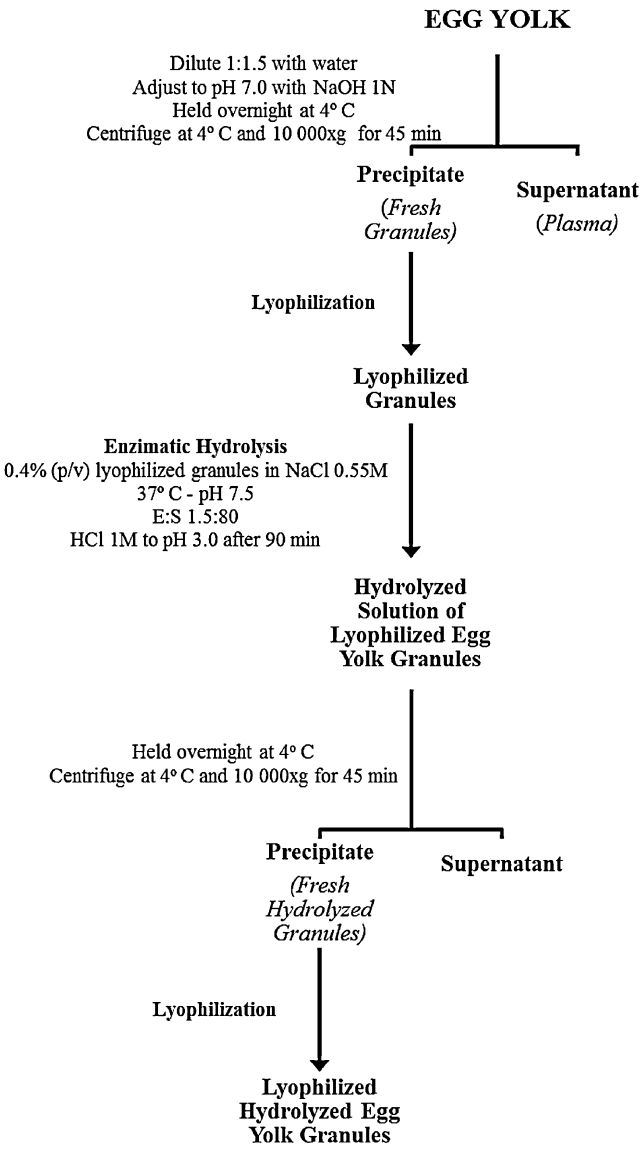

Fig. 1 – General scheme procedure to obtain lyophilized hydrolysed egg yolk granules.

Lyophilized egg yolk granules were dissolved at 0.4% (p/v) in 0.55 M sodium chloride solution for their total disruption (Antón et al., 2000) and stirred to fully disperse the protein. The protein content of this solution was calculated by Bradford method. Then, it was heated at 37 °C and pH adjusted to 7.5 by using 0.1 M sodium hydroxide solution. Trypsin from porcine pancreas E.C. 3.4.21.4 (T7409, Sigma–Aldrich) 1900 u/mg solid was added to the bioreactor (1.5:80 enzyme–substrate relation). Inactivation of the enzyme was done by acidifying with 1 M hydrochloric acid to pH 3.0 after 90 min of reaction.

The hydrolysed solution was kept at 4 °C overnight and then centrifuged (KUBOTA 6500 Centrifuge) at 10 000 × *g* and 4 °C for 45 min to separate into supernatant and the hydrolysed granule fraction (precipitate). The supernatant was separated from the hydrolysed granules by decantation.

The hydrolysed granules fraction was lyophilized by the same method as for the non-hydrolysed granules. The protein contents of both lyophilized hydrolysed and non-hydrolysed granules were calculated according to the Dumas combustion method using a CNHS/O Vario EL analyzer (Elementar).