Journal of Food Engineering 195 (2017) 40–51

Contents lists available at ScienceDirect

Journal of Food Engineering

journal homepage: www.elsevier.com/locate/jfoodeng

Effect of dry heat treatment of egg white powder on its functional, nutritional and allergenic properties

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ARTICLE INFO

Article history:
Received 23 June 2016
Received in revised form
19 September 2016
Accepted 22 September 2016
Available online 25 September 2016

Keywords:
Egg white powder
Dry heating
Interfacial property
In vitro digestion
Antigenicity

ABSTRACT

Egg white is a key ingredient in many food products as it combines high nutritional quality with excellent functional properties. However, it is also one of the leading causes of food allergy in childhood. Dehydrated egg white is a common form of industrial egg white. To increase the functional properties, egg white powders are heated which may change protein antigenicity and susceptibility to digestion. The present work highlighted the effect of a wide range of dry heating rates (from 1 to 10 days between 60 and 90 °C) simultaneously on the interfacial properties, antigenicity and susceptibility to *in vitro* digestion of egg white proteins. Thanks to a powerful statistical methodology, i.e. multiple factor analysis (MFA), that enables to consider all the data in a common space, intermediate dry heating treatments (2–5 days at 70 °C or 1–2 days at 80 or 90 °C) were found to be good compromise to improve egg white functionality without enhancing too much protein resistance to digestion or protein antigenicity.

1. Introduction

Egg white is a key ingredient in many food products as it combines high nutritional quality (Seuss-Baum et al., 2011) with excellent functional properties (Lechevalier et al., 2011). However, egg white is also one of the leading causes of IgE mediated food allergy in childhood (Moneret-Vautrin, 2008).

Egg white is commercialised as liquid, frozen or spray dried, the last one being a common and convenient form. After spray-drying, egg white powders are stored in hot rooms under controlled temperature (over 60 °C) and humidity conditions during a few days to ensure their microbiological safety. This industrial practice, called dry heating, is also very effective to improve the gelling and foaming properties of egg white when performed at higher temperatures (over 70 °C) (Kato et al., 1989, 1990a,b). Many studies have thus been carried out to investigate the effect of dry heating on foaming and gelling properties of either egg white, or purified egg white proteins (Baron et al., 2003; Hammershoj et al., 2006b; Desfougères et al., 2008; Matsudomi et al., 1991; Mine, 1996, 1997). Fewer studies were dedicated to egg white emulsifying properties (Kato et al., 1989). Most of these studies were performed at a given dry heating temperature, often 80 °C, for 5–10 days. Talansier et al. (2009) explored the consequences of different time-temperature tables (from 1 to 7 days and 60–80 °C) on the interfacial properties of egg white and its protein structure. These authors highlighted that mild treatments (between 70 °C for 3 days, and 80 °C for1 day) offered a good compromise to optimize both foaming capacity and foam stability.

Because of the well-known relationship between protein functionality and protein structure, many studies were also performed to understand the effect of dry heating treatment on the structure of egg white proteins (Hammershoj et al., 2006a; Matsudomi et al., 2001; Nicorescu et al., 2011; Van der Plancken et al., 2007; Watanabe et al., 1999; Xu et al., 1998). Protein aggregation and an increase in protein surface hydrophobicity were the main changes

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http://dx.doi.org/10.1016/j.jfoodeng.2016.09.022
0260-8774/

noticed by the authors. Recently, Desfougères et al. (2011a) highlighted chemical modifications of lysozyme induced by dry heating (formation of one to five cyclic imide rings) leading to the increase in the protein net charge and hydrophobicity but without any changes in protein secondary or tertiary structures. These modifications were correlated with the strong increase of the lysozyme interfacial properties after dry heating. These improved properties were due to a faster and more efficient adsorption of the protein at the air-water interface, and to the thick and strong viscoelastic interfacial film the dry heated lysozyme created (Desfougères et al., 2011b). Undoubtedly, dry heating modifies protein properties, and one can thus wonder if such changes may alter other protein properties such as digestibility and/or antigenicity.

The high content of essential amino acids in egg white proteins and the high bioavailability of these proteins are of great benefit to human nutrition. However, the effects of industrial processing such as dry heating on the nutritional quality of egg white proteins are poorly documented. Some studies considered the effect of dry heating on the *in vitro* digestibility of proteins as it is a prerequisite to nutritional quality. Schmidt et al. (2007) did not find any significant effect of dry heating duration at 70 °C on the *in vitro* digestibility of egg white proteins. Watkins and Veum (2010), however, showed a negative effect of dry heating (30 min at 121 °C) on the growth performance of neonatal pigs. Some studies also investigated the heat treatment effect on the digestibility of purified proteins in solution. Jimenez-Saiz et al. (2011) thus highlighted an increase of the *in vitro* digestibility of ovalbumin treated up to 15 min at 95 °C, whereas the digestibility of ovomucoid was not affected by this treatment.

In the same way, the changes in protein structure induced by dry heating may hide, destroy or disclose protein allergenic epitopes. Moreover, the potential changes in protein digestibility can also change the ability to sensitize and elicit the immune response (Jimenez-Saiz et al., 2011). The effect of heat treatment on the allergenic properties of egg proteins has been focused on cooking since some children with egg allergy tolerate cooked eggs (Hirose et al., 2004; Jimenez-Saiz et al., 2011; Julia et al., 2007; Lemon-Mulé et al., 2008; Liu et al., 2013; Mine and Zhang, 2002; Shin et al., 2013; Tong et al., 2012; Urisu et al., 1997). In most cases, physicochemical changes caused by cooking enhanced either a decrease or no significant effect on protein allergenicity, depending on the susceptibility of the proteins to unfold and lose their conformational epitopes. But up to now, no study was performed to test the effect of dry heating on egg white antigenicity or allergenicity. Only Escudero et al. (2013) tested dehydrated egg white in oral food challenges as it is microbiologically safer than raw egg white. As they tested commercial dehydrated egg white, one can think it was dry heated egg white, even if the process conditions were not mentioned by the authors. They showed that dehydrated egg white was not less allergenic than raw egg white.

Up to now, the few studies carried out on dry heating of egg white powder focused on the effect on either functional properties or protein digestibility or again protein immunoreactivity or allergenicity. The present work proposes a multicriteria approach by testing a wide range of industrial dry heating conditions simultaneously on protein interfacial properties, antigenicity and digestibility. The aim is to determine the treatment conditions that optimize protein interfacial properties neither decreasing their digestibility nor increasing their antigenicity. To do so, a powerful statistical tool, i.e. multiple factor analysis (MFA) was applied to the data set. As explained by Lechevalier et al. (2015), MFA considers several sets of variables simultaneously as active elements, in order to take all of them into account in the definition of the distance between the conditions tested. Moreover, as a multivariate method, MFA reduces the complexity of the dataset and is less affected by data distribution or variance heterogeneity than univariate methods. The originality of this work rests on the use of this method that allows considering several treatment conditions on many measurements of different nature (protein interfacial properties, digestibility, immunoreactivity, …).

2. Material and methods

2.1. Experimental design

Raw egg white powder (93.7% of dry matter) was supplied by Igreca (Seiches sur le Loir, France). The powder was aliquoted into 17 samples of 100 g. Each sample was subjected to one dry heating treatment according to the following experimental design: complete factorial design 4^2 with 4 temperatures (60, 70, 80 and 90 °C) and 4 durations (1, 2, 5 and 10 days); the control sample was maintained at 20 °C.

2.2. Protein solubility

Egg white solutions were prepared by dissolution of 1 mg of egg white powders in 1 ml of distilled water, and equilibration for 1 h at room temperature under continuous stirring with a magnetic stir bar. Protein content was measured in these solutions, and in the corresponding supernatants obtained after centrifugation for 20 min at 10,000*g*, and 10 °C. Protein content was determined according to Markwell et al. (1978) after calibration with bovine serum albumin as a standard. Protein solubility was calculated as follows:

$$solubility(\%) = \frac{Cs}{Ci} \times 100 \quad (1)$$

where Cs and Ci are the protein concentrations (in $g.l^{-1}$) in supernatant and initially, respectively.

2.3. Interfacial properties

2.3.1. Langmuir isotherms at air-water interface

Measurements of the surface pressure (Π) - surface area (*A*) isotherms have been performed by compression – expansion cycles using the Wilhelmy plate method as described by Rannou et al. (2015). Egg white powders were solubilised in 50 mM phosphate buffer pH 7, NaCl 300 mM, and stirred for 2 h at 20 °C, before centrifugation for 20 min at 10,000*g* and 10 °C. The final concentration of supernatant was adjusted to 1 mg ml^{-1} after measurement of protein content according to Markwell et al. (1978). One hundred microliters of 1 mg ml^{-1} protein solutions were dropped with a micrometric syringe all along the surface of the phosphate buffer. In order to allow protein spreading, adsorption and rearrangements, samples were dropped 30 min before compression. The compression speed was maintained constant at 40 cm^2 min^{-1}, i.e. low enough to prevent secondary effects due to the barrier displacement. Three replicated isotherms were performed for each sample. The surface corresponding to an interfacial pressure of 10 mN m^{-1} (S_{10}) was selected as the indicator of the sample behaviour at the air-water interface.

2.3.2. Emulsifying properties

2.3.2.1. Preparation of oil-water emulsions. Emulsions were prepared as described by Rannou et al. (2015). The aqueous phase was the supernatant adjusted at 15 mg ml^{-1} obtained after centrifugation for 20 min at 10,000*g* and 10 °C of egg white powders dissolved at 20 mg ml^{-1} in 50 mM phosphate buffer pH7, NaCl 300 mM. For each sample, emulsions were done in triplicate.