



# Effect of egg white protein-pectin electrostatic interactions in a high sugar content system on foaming and foam rheological properties



Mitie S. Sadahira <sup>a,\*</sup>, Maria I. Rodrigues <sup>b</sup>, Mahmood Akhtar <sup>c</sup>, Brent S. Murray <sup>c</sup>, Flavia M. Netto <sup>d,\*\*</sup>

<sup>a</sup> Instituto de Tecnologia de Alimentos/ITAL, Av. Brasil, 2880, CEP, 13070-178 Campinas, Brazil

<sup>b</sup> Protimiza Consultoria e treinamento em planejamento de experimentos e otimização de processos, Campinas, Brazil

<sup>c</sup> School of Food Science and Nutrition, University of Leeds, Leeds LS2 9JT, UK

<sup>d</sup> Faculdade de Engenharia de Alimentos, Universidade Estadual de Campinas/ UNICAMP, Rua Monteiro Lobato n° 80 - CEP, 13.083-862, Campinas, Brazil

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## ABSTRACT

The aim of this study was to evaluate the effect of electrostatic interaction between egg white protein (EW) and pectin in a high sugar content system (80 wt% total solid) on the foaming properties (density, overrun and stability) and foam rheological properties. A central composite rotatable design was carried out to study the effects of biopolymer concentration (1.40–5.60%, w/w) and EW:pectin ratio (7:1–63:1) on the apparent viscosity before whipping, foaming capacity (density and overrun) and foam rheological properties (storage modulus  $G'$ , loss modulus  $G''$  and phase angle  $\delta$ ) of sugar/EW/pectin mixtures at pH 3.0. The apparent viscosity increased as biopolymer concentration increased while EW:pectin ratio had no significant effect ( $p > 0.10$ ) on this response. At 7:1 EW:pectin ratio, the mixture presented low foaming capacity, resulting in foam with less solid character and low stability, possibly due to the pectin excess in the system. At 49:1 EW:pectin ratio, the mixture showed higher foaming capacity and foam elasticity. The formation of soluble complexes between EW and pectin possibly increased the continuous phase viscosity and enhanced the foam stability by slowing liquid drainage.

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## 1. Introduction

Food foam is formed by air, liquid and surface-active agent such as proteins (Kinsella, 1981). The formation of air bubbles modifies the texture and the rheological properties of aerated food (Campbell & Mougeot, 1999).

In the confectionery industry, aeration is used to obtain products such as nougat, marshmallow, chews and pulled sugar. The density of these products varies between 0.2 and 1.0 g/mL. In aerated confectionery, foams are produced by aeration of a mixture of sugar syrups and proteins. Egg white protein (EW) is the most widely used surface active agent for production of marshmallows and nougats (Jackson, 1995). Polysaccharides are also used due to

their thickening and gelling properties; their addition to foam can improve stability because they control the rheology and network structure of the continuous phase (Dickinson, 2003, 2008).

Pectin is a carboxylated anionic polysaccharide of high molecular weight. Its functional properties depend on the degree of esterification (DE). High-methoxyl pectins ( $\geq 50\%$  DE) require high sugar concentration and low pH to form gels, whereas low-methoxyl pectins form gels in the presence of calcium (Dickinson, 2003; Akhtar, Dickinson, Mazoyer, & Langendorff, 2002).

Proteins and polysaccharides contribute to food structural and textural properties due to their aggregation and gelation properties (Benichou, Aserin, Lutz, & Garti, 2007). Mixtures of polysaccharide and protein solutions can exhibit one of the three behaviours: miscibility, complex coacervation and thermodynamic incompatibility. Miscibility usually occurs at low biopolymer concentrations. Coacervation takes place due to attractive interactions between protein and polysaccharide leading to the formation of soluble and/or insoluble complexes. Thermodynamic incompatibility results in a separation into two distinct phases, due to the limited thermodynamic compatibility between proteins and polysaccharides in aqueous solution (Dickinson, 2003; Doublier, Garnier, Renard, & Sanchez, 2000; Rodríguez Patino & Pilosof, 2011).

*Abbreviations:* EW, egg white protein; ANOVA, analysis of variance; CCRD, central composite rotatable design;  $G'$ , storage modulus;  $G''$ , loss modulus;  $\delta$ , phase angle;  $R^2$ , percentage of variance explained.

\* Corresponding author.

\*\* Corresponding author.

*E-mail addresses:* [mitie@ital.sp.gov.br](mailto:mitie@ital.sp.gov.br) (M.S. Sadahira), [protimiza@protimiza.com.br](mailto:protimiza@protimiza.com.br) (M.I. Rodrigues), [M.Akhtar@food.leeds.ac.uk](mailto:M.Akhtar@food.leeds.ac.uk) (M. Akhtar), [B.S.Murray@leeds.ac.uk](mailto:B.S.Murray@leeds.ac.uk) (B.S. Murray), [fmnetto@unicamp.br](mailto:fmnetto@unicamp.br) (F.M. Netto).

Electrostatic interaction between a positively charged protein, where  $\text{pH} < \text{pI}$  ( $\text{pI}$  = protein isoelectric pH) and a negatively charged polysaccharide ( $\text{pH} \gg \text{pKa}$  of the polysaccharide acidic functional groups) can result in soluble and/or insoluble complex formation (Benichou et al., 2007; Dickinson, 2008). The physicochemical parameters that influence the electrical charge of protein and polysaccharide and the electrostatic complex formation are: pH, ionic strength, temperature, protein:polysaccharide ratio and total biopolymer concentration (Schmitt & Turgeon, 2011). Studies have shown that the electrostatic interaction between pectin ( $\text{pKa} \sim 2.9\text{--}3.5$ ) and EW ( $\text{pI} \sim 4.5\text{--}4.9$ ) are effective in increasing foam stability in aqueous solution (Ibanoglu & Ercelebi, 2007; Surh, Decker, & McClements, 2006; Ralet, Dronnet, Buchholt, & Thibault, 2001; Sadahira, Lopes, Rodrigues, & Netto, 2014).

Foam stability is affected by creaming, drainage, disproportionation and coalescence (Damodaran, 2005; Walstra, 2003). Stability is an important property for aerated products because the foam structure must be maintained during additional processing and shelf-life (Foedgeding, Luck, & Davis, 2006). Density difference between dispersed phase (air) and the continuous phase (aqueous phase) leads to creaming. The liquid drainage from the lamella film is followed by the approach of bubble surface leading to coalescence. Even the drainage and coalescence are reduced, diffusion of gas from small to large bubbles with different internal pressures can occur. This process causes shrinkage of small bubbles and expansion of large bubbles and it is called disproportionation (Murray & Ettelaie, 2004; Murray, 2007).

It has been shown that neutral complexes of  $\beta$ -lactoglobulin-pectin,  $\beta$ -lactoglobulin-Acacia gum and ovalbumin-pectin build dense viscoelastic interfacial networks at the air–water interface leading to low gas permeability and increasing foam stability. Total biopolymer concentration and protein–polysaccharide ratio are physicochemical parameters that influence electrical charge within the range of pH where both biopolymers have opposite charges and can form electrostatic complexes ( $\text{pKa}_{\text{polysaccharide}} < \text{pH} < \text{pI}_{\text{protein}}$ ) (Ganzevles, Zinviadou, van Vliet, Cohen, & de Jongh, 2006; Kudryashova, Visser, van Hoek, de Jongh, 2007; Liz et al., 2006).

Aerated confectioneries are produced using boiled sugar syrup and proteins; therefore, the aim of this study was to evaluate the effect of electrostatic interaction between EW and pectin in a high sugar content system on the foaming properties (density, overrun and stability) and foams rheological properties.

## 2. Material and methods

### 2.1. Material

Sucrose (Tate & Lyle, UK) was purchased from a local supermarket (Leeds). Glucose syrup (40 D.E., 83 wt% total solid) and invert sugar syrup (80 wt% total solid) were kindly donated by Brenntag UK & Ireland (Leeds, UK) and by British Sugar (Peterborough, UK), respectively. These sugars were used to prepare the multicomponent model systems of sugars. Dried egg white protein (EW) was supplied by Salto Alimentos LTDA (Salto, Brazil) and low methoxyl pectin (GENU Pectin type LM CG-22, degree of esterification 47.2%, molecular weight 90 kDa) by CPKelco (Grossenbrode, Germany) were used to prepare the biopolymer blends. EW represented, on a wet basis,  $79.9 \pm 1.2\%$  of protein  $10.20 \pm 0.02\%$  of moisture and  $5.64 \pm 0.22\%$  ash, determined according to methodologies described by AOAC (2010). SDS–PAGE analysis (Laemmli, 1970) of EW showed an electrophoretic profile with bands of 77.7, 44.5 and 14.3 kDa that correspond to conalbumin, ovalbumin and lysozyme, respectively. The other chemicals used were of analytical grade, the fluorescence dye Rhodamine B purchased from Aldrich (Dorset, UK) and Milli-Q water was used in all experiments.

### 2.2. Preparation of solutions and foams

The composition of the sugar mixture used as a model system to evaluate the foaming and rheological properties in aerated products was sucrose (42.5 wt% total sugar solid) + glucose syrup (42.5 wt% total sugar solid) + invert sugar (15 wt% total sugar solid). This sugar mixture resulted in foams with characteristics similar to aerated confectionery products such as marshmallow, with a density between 0.25 g/mL and 0.50 g/mL and a water activity range 0.778–0.665 (Jackson, 1995; Wills, 1998).

The sugar mixture (500 g) was heated in a beaker via a hot-plate stirrer to reach 80 wt% total solids then was cooled to beating temperature, 70 °C. The biopolymers, in appropriate amounts for each trial condition (Table 1), were hydrated together in 36 g of water with magnetic stirring for 1 h at room temperature. The pH was adjusted to 3.0 with 4 mol L<sup>-1</sup> citric acid.

The sugars mixture (at 70 °C) and EW/pectin blend were mixed in a Kitchen Aid 5KPM5 stand mixer (Havant, UK) with a flat beater for 1 min at speed setting 4. Then, the sugar/EW/pectin mixture was whipped using a whisk beater operating at speed setting 10 under atmospheric pressure for 6 min.

A Central Composite Rotatable Design CCRD (2<sup>2</sup> factorial design with 4 trials under the axial conditions and 3 repetitions at the central point) totaling 11 trials (Table 1) (Rodrigues & Iemma, 2015) was carried out to evaluate the effect of total biopolymer concentration (w/w%) and EW:pectin ratio (w/w) on apparent viscosity of sugar/EW/pectin mixture before whipping, foaming capacity (density and overrun) for fresh foam and rheological properties ( $G'$ ,  $G''$  and  $\delta$  at 1 Hz) for fresh foam and foam aged for 24 h. From the results, second-order models were obtained and evaluated statistically by analysis of variance (ANOVA) using the software Statistica 7.0 (Statsoft, USA).

To evaluate the effect of EW:pectin ratio on foaming properties (density, overrun and stability), trials were carried out under the best experimental conditions from CCRD to obtain good foamability (low density, high overrun) and solid character (high  $G'$  value, low  $\delta$  value) at different EW:pectin ratio under the model validation conditions (total biopolymer concentration, 80% total solid, 70 °C and pH 3.0). The results were analyzed for differences between means by Student t test ( $p < 0.05$ ). The model validation was performed under the same conditions.

### 2.3. Foaming properties

#### 2.3.1. Foaming capacity: density and overrun

Foaming capacity was studied by measuring density and overrun. Cylindrical containers ( $35.43 \pm 0.21$  mL) were carefully filled up with foam. The top of the container was leveled with a metal spatula to achieve a uniform and plane surface to obtain constant volume. The weight of foam was recorded and then the foam density and overrun were calculated as follows.

$$\text{Density} \left( \frac{\text{g}}{\text{mL}} \right) = \frac{m_f}{\text{volume of cylindrical container}} \quad (1)$$

whereas overrun is defined by (Lau & Dickinson, 2004):

$$\text{Overrun} (\%) = \frac{100(m_i - m_f)}{m_f} \quad (2)$$

where  $m_i$  is the mass of the initial solution (unwhipped sample) and  $m_f$  is the mass of the whipped sample with the same volume of  $m_i$ .

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