ELSEVIER

Food Hydrocolloids 19 (2005) 239–248

FOOD HYDROCOLLOIDS

www.elsevier.com/locate/foodhyd

Interactions of polysaccharides with β-lactoglobulin adsorbed films at the air–water interface

Rosa Baeza[a], Cecilio Carrera Sanchez[b], Ana M.R. Pilosof [a,1], Juan M. Rodríguez Patino[b,*]

[a]*Departamento de Industrias, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Ciudad Universitaria (1428), Buenos Aires, Argentina*
[b]*Departamento de Ingeniería Química, Facultad de Química, Universidad de Sevilla, c/. Prof. García González, 1. 41012 Sevilla Spain*

Received 8 April 2004; revised 11 May 2004; accepted 8 June 2004

Abstract

In the present work we have studied the adsorption (dynamic surface pressure) of β-lactoglobulin (βLG), polysaccharides and their mixtures at the air–water interface at 20 °C and at pH 7. The measurements were performed on a automatic drop tensiometer. As polysaccharides with interfacial activity we have used propylene glycol alginates (PGA). To evaluate the effect of the degree of PGA esterification and viscosity, different commercial samples were studied—Kelcoloid O (KO), Kelcoloid LVF (KLVF) and Manucol ester (MAN). Xanthan gum (XG) and λ-carrageenan (λ-C) were studied as non-surface active polysaccharides. The results reveal a significant effect of surface-active and non-surface-active polysaccharides on dynamic characteristics of β-lactoglobulin adsorbed films. To explain the observed effects on the rates of diffusion, penetration, and rearrangement of these biopolymers at the air–water interface, three phenomena were taken into account: (i) the competitive adsorption; (ii) the complexation, and (iii) the existence of a limited thermodynamic compatibility between protein and polysaccharide at the air–water interface and in the bulk aqueous phase. Surface-active polysaccharides (MAN, KO) are less effective than non-adsorbing polysaccharides (XG) for increasing the surface pressure of protein films, because a competitive behaviour with protein. Highly hydrophilic polysaccharides that do not adsorb by their own at the interface (XG, λ-C) or surface-active polysaccharides with low hydrophobicity (KLVF) show a cooperative behaviour with protein that promotes a significant increase of surface pressure of adsorbed films.

Keywords: Protein; Polysaccharide; Gums; Hydrocolloid; Air–water interface; Adsorbed film; Surface tension

1. Introduction

β-lactoglobulin (β-LG), the most abundant protein in whey, is a globular protein of molecular mass 18.3 kDa, stabilised by two internal disulfide cross-linking, that exhibits good foaming properties (Tornberg, 1979). The structural and dynamic properties of β-lactoglobulin at the air–water interface, have been extensively studied in the past years (Horne & Rodríguez Patino, 2003; Rodríguez Niño, Carrera, Cejudo, & Rodríguez Patino, 2001; Rodríguez Patino, Carrera, Rodríguez Niño, &Cejudo, 2001) so that this protein is a good model to study the interactions of non-surface active and surface active polysaccharides with proteins at the air–water interface.

Some of the following questions are expected to be addressed in the present work: (1) Does the hydrophilic character of polysaccharides as xanthan or λ-carrageenan mean that they do not adsorb at air–water interfaces in protein + polysaccharide mixtures? (2) What are the mechanisms by which polysaccharide can influence the interfacial properties of proteins? (3) Is a surface-active polysaccharide more effective than a non-adsorbing polysaccharide for decreasing the surface tension in a protein + polysaccharide mixture?

In order to answer the questions above it seems appropriate to study the protein–polysaccharide mixtures at a pH above the isoelectric point of protein, where complex formation as a result of net electrostatic interactions does not occur. Above the isoelectric point of

* Corresponding author. Tel.: +34-954-556446; fax: +34-954-557134.
E-mail addresses: apilosof@di.fcen.uba.ar (A.M.R. Pilosof), jmrodri@us.es (J.M. Rodríguez Patino).
[1] Fax.: 54-11-45763366.

0268-005X/$ - see front matter © 2004 Elsevier Ltd. All rights reserved.
doi:10.1016/j.foodhyd.2004.06.002

the protein thermodynamic incompatibility between the protein and polysaccharide generally occurs because of the repulsive electrostatic interactions and different affinities towards the solvent (Tolstoguzov, 1997). Therefore, protein and polysaccharide may co-exist in a single phase (miscibility) in domains in which they mutually exclude one another or, above a critical concentration, segregate into different phases. Excluded volume effects can have the following manifestations: enhancement of the association of macromolecules, reduction in the critical concentration for gelation and increase in the rate of gelation (Baeza & Pilosof, 2001; Capron, Nicolai, & Durand, 1999), enhancement of protein adsorption at fluid interfaces (Tsapkina, Semenova, Pavlovskaya, & Tolstoguzov, 1992), etc.

In the present work we have studied the adsorption dynamics of single β-lactoglobulin and polysaccharides in comparison to (β-LG)+polysaccharides mixtures at the air–water interface, at 20 °C and at pH 7. As polysaccharides with interfacial activity we have used propylene glycol alginates (PGA). To evaluate the effect of the degree of PGA esterification and viscosity, different commercial samples were studied. Xanthan gum (XG) and λ-carrageenan (λ-C) were studied as non-surface active poly-saccharide.

Xanthan (XG) is an anionic polysaccharide that produces high viscosities at low concentrations (Imeson, 1992). Xanthan, being highly hydrophilic and without any significant hydrophobic bonds, is not adsorbed at the air–water interface (Yilmazer, Carrillo, & Kokini, 1991). Nevertheless, xanthan promoted soy protein subunits aggregation at the air–water interface in foams based on native soy proteins (Carp, Bartholomai, & Pilosof, 1999). Those specific effects further influenced the foam stability of the mixed systems (Carp, Bartholomoi, Relkin, & Pilosof, 2001). One possible interpretation of these findings is that the gum adsorbs onto the protein, forming a combined structure of a primary protein layer predominantly in contact with the air phase. Following this last hypothesis, the surface tension might be dominated by the primary protein layer, covered by and adsorbed polysaccharide layer in strong electrostatic contact, dominating the surface rheology (Galazka & Dickinson, 1995). Sulphated polysaccharides like carrageenans can interact with charged groups in a protein more strongly than carboxylated hydrocolloids like xanthan gum at pH above the protein isoelectric point (Dickinson, 2003).

One distinct group of surface-active polysaccharides are the propylene glycol esters of alginic acid (PGA), a high molecular weight linear polysaccharides composed of 1,4 linked-D-mannuronic acid and L-guluronic acid (Dziezak, 1991). They are produced with a range of viscosities and degrees of esterification. The increase in the degree of esterification reduces the overall hydrophilic character of the molecules and imparts surface-active properties. The resulting ability of PGA to reduce the surface tension of water as well as increase the viscosity of the water phase make them suitable as stabilisers and foaming agents. PGA is an example of a polysaccharide used in food foams, such as beer, to aid 'head' retention of the foam (Sarker & Wilde, 1999). Due to its surface-active character, competitive adsorption of PGA could occur in mixtures of this polysaccharide and proteins. Numerous studies have been published on competitive adsorption of milk proteins and low-molecular weight surface-active agents such as monoglycerides (Dickinson & Tanai, 1992; Rodriguez Patino, Rodríguez Niño, & Carrera Sánchez, 2003) surfactants (Mackie, Gunning, Wilde, & Morris, 1999, 2000), lecithins (Courthaudon, Dickinson, & Christie, 1991). However, no reports were found on competitive adsorption of proteins and polysaccharides. In addition, formation of protein–PGA complexes at the interface could also occur (Ahmed & Dickinson, 1991). The foaming properties were strengthened at pH value above 7, where both biopolymers carried negative charges, revealing the possibility of formation of electrostatic complexes between positively charged patches in the proteins and carboxyl groups in PGA. A more quantitative indication of non-covalent complex formation between PGA and caseinate, whey protein isolates and β-LG at the oil-water interface was provided by film surface viscosity (Dickinson & Euston, 1991). The principal mechanism is thought to be non-covalent cross-linking of the protein molecules at the interface.

2. Materials and methods

2.1. Materials

β-lactoglobulin (β-LG) was supplied by Danisco Ingredients (Denmark). The powder composition was: protein 92%, β-lactoglobulin > 95%, α-lactalbumin < 5%. The polysaccharides λ-carrageenan (λ-C) and xanthan gum (XG) were provided by BIOTEC (Argentina) and propylene glycol alginates (PGA) were from ISP Alginates. The PGA used were Kelcoloid O (KO), Kelcoloid LVF (KLVF), and Manucol ester (MAN). The degree of esterification and viscosity of PGAs are shown in Table 1.

2.2. Method

The existence of β-lactoglobulin–polysaccharides interactions at the air–water interface was determined by monitoring the dynamics of surface pressure (π) of single components and mixed systems. For time-dependent surface

Table 1
Degree of esterification and viscosity of propylene glycol alginates

PGA	Degree of esterification	Viscosity[a] (cps)
Manucol ester (MAN)	High	High (11.8)
Kelcoloid LVF (KLVF)	Medium	High (13.9)
Kelcoloid O (KO)	High	Low (4.7)

[a] Viscosity (60 s^{-1}) of 0.5% wt/wt solution.