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Structure and rheological properties of acid-induced egg white protein gels

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Abstract

This study compares the rheological properties of acid-induced gels prepared of industrial spray-dried egg white proteins (EWP) with the acid-induced gels prepared of ovalbumin (OA) and whey protein isolate (WPI). Also we aimed to form transparent gels of EWP by means of the cold-gelation process. We showed that it was not possible to prepare cold-set gels because ovotransferrin (OT), present in EWP, was found to interfere with fibril formation. Therefore, we developed a new purification method in which first OT was selectively denatured by a heating step, subsequently precipitated by acidification and removed by centrifugation. Finally, the supernatant was desalted by ultra filtration. This resulted in a preheated EWP preparation, which mainly contains OA (>80%). By removing OT using this new preheat procedure transparent gels were obtained after acid-induced gelation. Fracture properties of various EWP preparations were determined and compared with those of acid-induced gels of OA and WPI. Gels formed from different EWP preparations were weak (fracture stress 1–15 kPa, fracture strain 0.3–0.7), and the networks consisted of thin strands with hardly any additional disulphide bonds formed during the gelation step. In conclusion, the microstructure of the aggregates formed in the first step of the cold-gelation process and the amount of additional disulphide bonds formed during the second step appeared to be the determining factors contributing to the hardness and deformability of acid-induced gels of egg white proteins.

Keywords: Ovalbumin; Hen egg white proteins; Purification; Aggregation; Acid-induced gelation; Turbidity; Ovotransferrin; Whey protein isolate

1. Introduction

Consumers continuously demand more tasty, natural, healthy, convenient and new (e.g. transparent) food products. Fracture properties are important because the perception of texture is in part an evaluation of fracture properties when a gel-type food is consumed (Li, Errington, & Foegeding, 1999). Texture is a quality parameter and often caused by protein structures. Therefore, improving protein structures can provide answers to the constant requests for innovation. Typical food proteins of interest include proteins derived from milk, soy, fish and egg, which are used in a number of foods including beverages, confectionary, desserts, dairy products and meat and fish products.

In traditional food applications, protein solutions are heated at relatively high temperatures (>60 °C) to induce denaturation. Heating above a critical gelation concentration or percolation threshold, C^*, results in the formation of gelled systems, whereas heating below this concentration thickens solutions but does not cause gelation. The application of heat-induced gelation has limitations or is not always desired (Barbut & Foegeding, 1993). For instance, heat gelation is relatively simple and fast, but not 100% efficient in terms of protein usage. Cold gelation is easier to control, more efficient and an advantage might be that heat-labile or volatile compounds can be added (in the gelation step) without any losses or off-flavor occurring. However, it is more complex than the heat-induced gelation process.

The formation of acid-induced cold-set gels involves two stages. In the first stage, the preparation of a heat-denatured protein solution of soluble aggregates occurs at neutral pH

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and low ionic strength. In the second step, at ambient temperature, gelation is induced by changing the pH towards the iso-electric point of the protein. This reduces the electrostatic repulsion between the aggregates and therefore promotes the formation of a percolation network (Alting et al., 2004; Bryant & McClements, 1998). Dependent on the type of aggregates produced in the first stage, different gel network structures and rheological properties can be obtained. Bryant & McClements. (1998) published a clear and complete overview on cold-set gels derived from heat-denatured whey proteins. We previously reported the formation of transparent cold-set gels of ovalbumin (OA), whereas whey protein isolate (WPI) forms turbid gels.

Similar to WPI, egg white powder consist of a mixture of proteins, of which OA is the most predominant (~54%), followed by ovotransferrin (OT, ~12%) and ovomucoid (OM, ~11%) (Table 1). The composition and properties of the egg white have been extensively described in literature (Hammershøj, Larsen, Ipsen, & Qvist, 2001; Handa, Takahashi, Kuroda, & Froning, 1998; Li-Chan & Nakai, 1989; Mine, 1995; Nakamura, Umemura, & Takemoto, 1979; Powrie & Nakai, 1986). Also, interactions between the various purified proteins present in egg white during heating have been published (Cunningham & Lineweaver, 1967; Matsuda, Watanabe, & Sato, 1981, 1982; Matsudomi, Oka, & Sonoda, 2002; Matsudomi, Takasaki, and Kobayashi, 1991; Matsudomi, Yamamura, & Kobayashi, 1986; Yamashita, Ishibashi, Hong, & Hirose, 1998). In those papers, aggregate properties are discussed by means of turbidity measurements and SDS-PAGE analysis. However, the influence of OT on the fibril formation of OA has not been described so far and is of major importance to produce transparent gels by means of acid-induced gelation.

Large strain rheological measurements were performed to characterize the fracture properties of acid-induced egg white protein gels and compared with known behaviour of WPI gels. In general, fracture stress reflects hardness and strain reflects deformability or brittleness of gels. Previous publications on WPI gels revealed that gels that fracture at low strain values have networks composed of relatively thin strands and small homogeneous pores, whereas gels which fracture at high strain values are composed of thicker strands and relatively large homogeneous pores (Roff & Foegeding, 1996). For WPI gels it was demonstrated that fracture stress and strain can be influenced by the presence of disulphide bonds (Alting, Hamer, de Kruif, & Visschers, 2000; Errington & Foegeding, 1998). However, it is not clear what the effect of microstructure and disulphide bonding is on the turbidity and rheological properties of acid-induced gels of EWP.

Since purified OA is not food grade available, industrial egg white powder was used for further studies. These preparations were not able to form cold-set gels, since they already formed turbid gels at low protein concentration, because the ionic strength in egg white powders is relatively high. Recently, we described the procedure to prepare transparent gels from industrial egg white powder (Visschers, van de Velde, & Weijers, 2004). This opens new possibilities for applications in food products. This paper describes the molecular background of the formation of transparent acid-induced gels of industrial egg white powder, and compares the rheological properties of acid-induced gels prepared of industrial spray-dried egg white proteins (EWP) with the acid-induced gels prepared of ovalbumin (OA) and whey protein isolate (WPI). The main objective of this study was to investigate the molecular background of the formation of transparent cold-set gels

Table 1
Overview of egg white protein composition and molecular properties

Protein	% (w/w)	p*I*	M_w (kDa)	T_d (°C)	Cysteines	–SH	S–S
Ovalbumin[a–e]	54	4.5–4.9	45	75–84	6	4	1
Ovotransferrin[a–d,f] (conalbumin)	12–13	6.0–6.1	77.7	61–65 (76.5, Al^{3+})	30	–	15
Ovomucoid[a,b,f,g]	11	4.1	28	77	18	–	9
Ovomucin[a,b,f,g]	1.5–3.5	4.5–5.0	110, 5500–8300, 220–270,000		(2)	–	
Lysozyme[a–c,f,g]	3.4–3.5	10.7	14.3–14.6	69–77	6		4
G2 ovoglobulin[a,b,f]	1.0	4.9–5.5	47–49				
G3 ovoglobulin[a,b,f]	1.0	4.8, 5.8	49–50				
Ovoflavoprotein[b,f,g]	0.8	4.0	32–35, 80		5		2
Ovostatin[b,f,g]	0.5	4.5–4.7	760–900				
Cystatin[b,f]	0.05	5.1	12				
Avidin[a,b,f,g]	0.05	10.0	55–68.3		2		1

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[f] http://www.food-allergens.de/symposium-vol1(1)/data/egg-white/egg-composition.htm.
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