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Effects of added whey protein aggregates on textural and microstructural properties of acidified milk model systems

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ABSTRACT

Non-fat milk model systems containing 5% total protein were investigated with addition of micro- or nanoparticulated whey protein at two levels of casein (2.5% and 3.5%, w/w). The systems were subjected to homogenisation (20 MPa), heat treatment (90 °C for 5 min) and chemical (glucono-delta-lactone) acidification to pH 4.6 and characterised in terms of denaturation degree of whey protein, particle size, textural properties, rheology and microstructure. The model systems with nanoparticulated whey protein exhibited significant larger particle size after heating and provided acid gels with higher firmness and viscosity, faster gelation and lower syneresis and a denser microstructure. In contrast, microparticulated whey protein appeared to only weakly interact with other proteins present and resulted in a protein network with low connectivity in the resulting gels. Increasing the casein/whey protein ratio did not decrease the gel strength in the acidified milk model systems with added whey protein aggregates.

1. Introduction

The use of milk protein ingredients in fermented dairy products for texture enhancement has become a common practice due to the good nutritional and functional properties (Hau & Bovetto, 2001; Liu et al., 2016a; de Wit, 1998). Depending on the required characteristics, milk protein ingredients such as skimmed milk powder (SMP), whey protein concentrate (WPC) or sodium caseinate are commonly added in yoghurts. Adding these ingredients, however, may lead to a powdery taste, excessive firmness, higher whey expulsion and grainy texture (Guzmán-González, Morais, & Amigo, 2000; Sandoval-Castilla, Lobato-Calleros, Aguirre-Mandujano, & Vernon-Carter, 2004).

Microparticulated whey protein (MWP) are colloidal particles manufactured in diameters ranging from 1 to 10 μm, which are based on acid precipitation and thermal denaturation of whey protein in WPC and further aggregation by shearing (Liu et al., 2016a; Renard, Lavenant, Sanchez, Hemar, & Horne, 2002). Nanoparticulated whey protein (NWP) is also produced by adjusting pH and subsequent heat treatment, but exhibit particle sizes smaller than 1 μm (Bovetto, Schmitt, Beaulieu, Carlier, & Unterhaslberger, 2006; Nicolai & Durand, 2013; Nicolai, Britten, & Schmitt, 2011). The early interactions that occur during heating of the milk are known to be of importance for the following acidification in yoghurt manufacture (Guyomarc'h, Law, & Dalgleish, 2003; Lucey, Teo, Munro, & Singh, 1997; O'Kennedy & Kelly, 2000). Heating induces unfolding of whey proteins, aggregation and thereby formation of complexes with κ-casein, α_{S2}-casein and other whey proteins via covalent and non-covalent interactions (Famelart, Tomazewski, Piot, & Pezennec, 2004; Fox, Uniacke-Lowe, McSweeney, & O'Mahony, 2015; Guyomarc'h, Jemin, Le Tilly, Madec, & Famelart, 2009; Nguyen, Wong, Guyomarc'h, Havea, & Anema, 2014). Tamime, Kalab, Muir, and Barrantes (1995) reported that MWP could become an integral part of yoghurt microstructure as a fat substitute, but gave a softer texture. Lobato-Calleros, Martínez-Torrijos, Sandoval-Castilla, Pérez-Orozco, and Vernon-Carter (2004) and Sandoval-Castilla et al. (2004) found that the yoghurt with MWP showed lower firmness and rheological properties, but blends of WPC and MWP could be more advantageous since they ensured textural characteristics close to full-fat yoghurt. Heat-induced nanoparticles of whey protein have been reported to enhance gel strength and increase whey retention (Andoyo,

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Guyomarc'h, Cauty, & Famelart, 2014; Morand, Dekkari, Guyomarc'h, & Famelart, 2012). In previous studies, MWP and NWP has also been shown to provide desired mouthfeel (creaminess) as well as increased viscosity and denser microstructure when incorporated into low-fat yoghurt (Janhøj, Petersen, Frøst, & Ipsen, 2006; Liu et al., 2016a; Torres, Janhøj, Mikkelsen, & Ipsen, 2011; Torres, Amigo Rubio, & Ipsen, 2012). Liu et al. (2016a) furthermore showed that increasing the ratio of whey protein isolate (WPI) or micellar casein isolate (MCI) could be important for the formation of firm acidified gels with added commercial whey protein aggregates; however, the influence of commercially added whey protein aggregates on overall texture and functionality of acidified milk gels needs to be further investigated.

In the present study, our objective was to investigate how two types of whey protein particles (MWP and NWP) interact with other milk components and how they have effect on the textural properties and final microstructure of acidified milk model systems. To do that, milk model systems were prepared from MCI, WPI and whey protein aggregates dispersed at different ratios in dissolved whey permeate powder mimicking the serum phase of milk. The consequences of changing the whey protein aggregates/WPI ratio and the casein/whey protein ratio were investigated with respect to the textural, rheological and microstructural properties of the acidified milk model systems.

2. Materials and methods

2.1. Raw materials

Microparticulated whey protein (81.0% protein, 7.0% fat, 1.5% lactose, 2.3% ash), nanoparticulated whey protein (52.0% protein, 5.0% fat, 31.0% lactose, 5.4% ash), whey protein isolate (89.5% protein, 0.1% fat, 3.8% ash), micellar casein isolate (83.3% protein, 1.1% fat, 2.8% lactose, 7.7% ash) and ultrafiltrated (UF) whey permeate powder (2.6% protein, 87.1% lactose, 8.4% ash) were obtained from Arla Foods Ingredients (Nr. Vium, Denmark).

2.2. Pilot scale processing of milk model systems

Six model systems and four references (5 L for each) (see Table 1 for details) were prepared by mixing UF whey permeate from 5% (w/w) to 8% (w/w) (to maintain a similar ionic environment as in milk), whey protein particles, whey protein isolate and micellar casein in different combinations. All systems were made up to a final protein concentration of 5% (w/w) and lactose concentration of 6–7% (w/w). To ensure proper solubilisation all systems containing MWP were subjected to an initial two step homogenisation (15 MPa/5 MPa) using an APV R-5-14-38 homogeniser (APV, Gatwick, UK). The model systems were stored overnight at 4 °C to allow hydration of the powders prior to processing. All experiments were performed in triplicate.

Following overnight cold storage, the model milk systems were preheated to 63 °C in a water bath and thereafter subjected to one-pass homogenisation at 20 MPa though an APV R-5-14-38 homogeniser. The homogenised samples were then pasteurised at 90 °C for 5 min and cooled down to about 10 °C using a UHT/HTST Lab Direct & Indirect Processing system (MicroThermics Inc., Raleigh, NC, USA). Samples were subsequently heated up to the acidification temperature (45 °C) in a water bath and 1.5% (w/v) of GDL (glucono-δ-lactone; Sigma-Aldrich Co., St Louis, MO, USA) was added. When the pH reached 4.6, the gels were manually stirred with a stainless steel perforated disk and were subsequently smoothened and cooled to 22 °C by use of a post-treatment unit (Scandinox A/S, Hoven, Denmark) by passing the acidified gel through a pipe system (ø12 mm) at a backpressure of 0.1 MPa (Zhang, Folkenberg, Amigo, & Ipsen, 2016). Finally the samples were packaged in 250 mL plastic cups and kept at 5 °C for 7 d before analyses were performed. Samples were collected after homogenisation, heating and gelation and subsequently analysed.

2.3. Determination of thermal denaturation degree by reverse phase-high performance liquid chromatography analysis

The degree of denaturation of native β-lactoglobulin (β-lg) and α-lactalbumin (α-la) was determined by reverse phase-high performance liquid chromatography (RP-HPLC) according to the method mainly described by Tolkach and Kulozik (2005) with some modifications. The HPLC analysis was carried out on a Waters Alliance e2695 Separations Module (Waters, MA, USA) equipped with a Waters 2487 Dual λ Absorbance Detector. Relative quantification of peaks was performed by automated integration of peak areas, and when required, by manual integration, using the Empower 3 Chromatography Data software. Peak detection followed at 214 nm, and a reverse phase analytical C4 column (Jupiter 5 μm C4 300 Å 250 × 4.6 mm, Phenomenex) was used. A linear gradient of eluent A (650 μL triflouroacetic acid in 1 L MilliQ water) in eluent B (560 μL triflouroacetic acid in 1 L acetonitrile) was applied. The gradient was set to start with 15% of eluent B and subsequently increased from 15 to 45% in 30 min, held constant at 45% for 2 min and increased from 45% to 90% B in 0.5 min, and then remain constant at 90% for 5 min before returning to the initial conditions.

2.4. Particle size distribution and fractionation

Particle size distribution was measured for the milk model systems collected during each processing step (initial,

Table 1
Experimental design and composition of the milk model systems.[a]

Model system code	Casein	Total whey protein	Microparticulated whey protein	Nanoparticulated whey protein	WPI
M1	2.5	2.5	1.5	–	1.0
N1	2.5	2.5	–	1.5	1.0
M2	2.5	2.5	2.5	–	–
N2	2.5	2.5	–	2.5	–
M3	3.5	1.5	0.9	–	0.6
N3	3.5	1.5	–	0.9	0.6
RW	2.5	2.5	–	–	2.5
RN	–	5.0	–	5.0	–
RM	–	5.0	5.0	–	–
RC	5.0	–	–	–	–

[a] WPI, whey protein isolate; compositions given as % (w/w).