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Influence of pH and NaCl on rheological properties of rennet-induced casein gels made from UF concentrated skim milk

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Abstract

Milk concentrates are used in the production of cast cheese. The effects of pH (5.19–6.21) and NaCl concentration (0, 1.75% and 3.50%, w/w) on the rheological and microstructural properties of rennet-induced casein gels made from ultra filtered skim milk (19.8%, w/w casein) were investigated. Low pH and high NaCl concentration reduced the development rate of the gel elasticity after coagulation of the ultra filtrated skim milk. Strain at fracture and stress at fracture from uniaxial compression of casein gels 48 h after coagulation showed maximum and minimum values at pH ~5.8 and 5.29, respectively. Young's modulus from uniaxial compression of the same gels was almost constant between pH 5.52 and 6.21 but much lower at pH 5.28. Addition of NaCl resulted in increased Young's modulus in the interval pH 6.21–5.52. As pH decreased, the level of colloidal calcium phosphate decreased concomitantly, giving less cross-links in the casein network and partly explaining the changes in the rheological properties. Increased ionic strength by adjusting pH and addition of NaCl also influenced rheological results. The microstructure examined with confocal laser-scanning microscopy was unaffected by the changes in pH and concentrations of NaCl in the range studied as revealed by image analysis and calculations of two- and three-dimensional data from micrographs.

Keywords: Colloidal calcium phosphate; Image analysis; Rennet-induced casein gel; Ultrafiltration

1. Introduction

The casein micelles in milk contain four main types of caseins (α_{S1}-, α_{S2}-, β- and κ-casein) of which some bind colloidal calcium phosphate (CCP) (Walstra, Geurts, Noomen, Jellema, & van Boekel, 1999). CCP enables cross-linkage between caseins and contributes to the structure of the micelle. As the casein micelles constitute the basic building blocks of rennet-induced casein gels, CCP is also important to the structure of the gel network and its rheological properties (Lucey, 2002). Decreasing CCP while keeping the pH constant in rennet-induced casein gels made from dialysed milk resulted in a lower elastic modulus (Zoon, van Vliet, & Walstra, 1988).

In cheese manufacture, the amount of CCP is changed through addition of $CaCl_2$ and NaCl and by decreasing pH. At decreased pH more CCP is solubilized, especially below pH 5.8 (Le Graët & Gaucheron, 1999). This is considered as one of the main causes for the weakening of rennet-induced skim milk gels when pH is decreased (Lucey, Johnson, & Horne, 2003) but changed ionic strength and the pK_a of individual amino acids are also important (van Vliet, Roefs, Zoon, & Walstra, 1989). Addition of NaCl, generally, increases hydration of casein, decreases pH, increases calcium ion activity in the milk serum and increases the screening of charges on caseins (Karlsson, Ipsen, Schrader, & Ardö, 2005; Zoon, van Vliet, & Walstra, 1989). Depending on pH, NaCl has been reported to have different effects on the rheological properties of casein gels (Zoon et al., 1989) and cheese (Guinee, 2004). However, except for the results of Zoon et al. (1989), who studied the coagulation properties of milk after addition of NaCl, the effect of NaCl in casein gel systems on rheological properties has not been thoroughly evaluated.

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The relationships between microstructure of the casein network and the rheological properties of casein gels have been considered important by some authors (Bremer, Bijsterbosch, Schrijvers, van Vliet, & Walstra, 1990; Wium, Pedersen, & Qvist, 2003). Wium et al. (2003) described relationships between increased coarseness of the casein network and increased fracture and elastic properties of a model cheese from UF concentrates at pH 4.80 when the rennet concentration and the coagulation temperature varied. When observing the microstructure of casein gels from unconcentrated skim milk, lower pH and higher temperature have been reported to have the effect of increasing the rate of rearrangements of the protein network during gel aging (Mellema, 2000), but at the stage of casein micelle aggregation, only small differences have been observed in the fractal dimensionality of aggregates between pH 5.60 and 6.58 (Vétier, Banon, Ramet, & Hardy, 2000).

The microstructure of rennet-induced casein gels from unconcentrated milk is not of direct industrial importance in traditional cheese making where the rennet-induced casein gels are cut some 20–40 min after rennet addition. In the production of cast cheese, however, the casein gel is not cut and the coagulation properties and microstructure during coagulation of UF concentrate are very different compared to those in unconcentrated milk (Karlsson, Ipsen, & Ardö, 2006). Cast cheese is produced from ultrafiltrated (UF) milk concentrates with the primary aim of achieving a more efficient production and increase of cheese yield through incorporation of whey proteins.

The main objective of the present study was to characterise the influence of pH and NaCl concentration on the rheological and microstructural properties of casein gels made from highly concentrated skim milk. Due to the high concentration of protein, the casein gels of the study exhibited no macrosyneresis and the gross composition of all studied gels (regardless of pH and NaCl concentration) was the same. Thus, it was possible to study the direct influence of pH and NaCl on the rheological and microstructural properties of the casein network in the rennet-induced gels made from concentrated skim milk. Texture of cheese is an important parameter for both the consumer and the further processing of cheese, e.g., shredding and slicing. Understanding the influence of pH and NaCl is of major importance in the production of cast cheese and for development of new types of cheese products made from UF concentrates.

2. Materials and methods

2.1. Production and chemical analysis of UF concentrate

The UF concentrate was produced as described by Karlsson et al. (2005). Ultrafiltration of the skim milk was stopped when the Brix value was 36.3°. Pasteurization, freezing and frozen storage of the UF concentrate, measurements of pH and chemical analysis of both UF concentrate and unconcentrated skim milk (which was used as feed in the UF process) were performed as previously described (Karlsson et al., 2006). All chemical analyses were performed at least in duplicate.

Prior to use in experiments, the UF concentrate was thawed in a water bath for 1 h at 30 °C and 0.02% (w/w) Thimerosal (Merck, Darmstadt, Germany) was added as preservative. After 24 h at 30 °C, glucono-δ-lactone (GDL; Acros Organics, Geel, Belgium) was added to obtain a desired approximate pH (6.1, 5.8, 5.5 or 5.2) and NaCl (Merck) was added to obtain a concentration of 0, 1.75 or 3.50% (w/w). Samples of the UF concentrate were then stored for additional 24 h at 30 °C before further use.

2.2. Amount of colloidal bound calcium and phosphate

The concentration of calcium and phosphate in the serum phase of the concentrate was found to be constant below pH 3.0, thus, all calcium and phosphate were solubilized. By adjusting the pH to 2.2–3.0 using 1 M HCl, the total amount of CCP could be determined. At pH 5.2–6.1 (i.e., adjusted by GDL), only a fraction of the CCP was dissolved in the serum phase. After adjustment of pH by HCl or GDL and separation of the serum phase as described by Karlsson et al. (2006) the amount of CCP in the UF concentrate of different pH could be determined. For analyses of calcium concentrations a Perkin Elmer atomic absorption spectrometer (Perkin Elmer, Boston, MA, USA) was used as described by Karlsson et al. (2006). Preparations of all solutions were performed in duplicate and every duplicate was measured twice.

Concentration of inorganic phosphate was determined spectrophotometrically using a modified method of Murphy and Riley (1962). The serum phase of the UF concentrate was diluted in a stepwise manner to obtain a phosphorus (P) concentration of approximately 10–40 ppm. To 0.5 mL of a diluted serum phase, 10 mL of a phosphate reagent solution and deionized H_2O to a final volume of 50 mL were added. The phosphate reagent solution contained 0.00518 M $(NH_4)_6Mo_7O_{24}$, 0.00024 M potassium antimonyl tartrate trihydrate ($C_6H_4K_2O_{12}Sb_2 \cdot 3H_2O$), 1.42 M H_2SO_4 and 0.0341 M ascorbic acid. The concentration of P in the solutions used for the measurement was 0.1–0.4 ppm. Calibration solutions containing 0, 0.1, 0.2, 0.3, 0.4 and 0.5 ppm P were prepared from a stock solution of 0.0323 M KH_2PO_4 (1000 ppm P). The calibration solutions contained the same amount of phosphate reagent solution as the sample solutions. The blue-coloured ammonium phosphorus molybdate complex developed after 30 min in the solutions before the light absorption was measured at 890 nm in a Gensys 10UV spectrophotometer (Spectronic Unicam, Rochester, NY, USA). Preparations of all solutions were performed in duplicate. Every duplicate was measured two times and the concentration in sample solutions were calculated from the calibration solutions.