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Aggregation and gelation characteristics of high protein dairy ingredient powders

Key external stakeholders:

Dairy Ingredient manufacturers, food formulators.

Practical implications for stakeholders:

The provision of new information on the aggregation behaviour of reconstituted dairy protein ingredients and how this is affected by degree of whey protein denaturation during ingredient manufacture, extent of protein mineralization, and composition of the solvent used for reconstitution. The information is of particular relevance to the manufacturers of skim milk powders, micellar caseins, and milk protein concentrates.

Main results:

- The type of dairy protein powder used to fortify the protein content of skim milk significantly affected rennet gelation, heat stability at pH 6.2 – 7.2, and ethanol stability at pH 6.4.
- Reducing the calcium phosphate content of phospho-casein (PC) through the addition of sodium citrate/citric acid to skim milk prior of microfiltration and diafiltration significantly impacted on functionality of the reconstituted PC.
- The temperature and pH during milk heat treatment had a notable impact on the functionality of reconstituted skim milk powder, including rennet gelation, heat stability at pH 6.2 – 7.2, and ethanol stability at pH 6.4
- The heat treatment applied to skim milk during the manufacture of milk protein concentrate (MPC) affected rennet gelation, heat coagulation time as a function of pH at 6.2–7.2, and acid-gel formation to an extent dependent on the composition of the solvent used for reconstitution (water or milk permeate).

Opportunity / Benefit:

The project provides new insights into:

- The manipulation of dairy protein ingredient functionality by altering the conditions during ingredient manufacture and reconstitution.
- The development of potentially-new ingredients (κ-casein, β-lactoglobulin rich powders; κ-casein depleted micelle) by the partitioning of milk protein into soluble and sedimentable phases.

Collaborating Institutions:

Teagasc, University College Cork

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1. Project background:

Milk protein ingredients, in the form of dairy powders, are used extensively in manufacture and stabilization of formulated foods (e.g., protein bars, ice cream, imitation cheeses, fresh-cheese products, dairy spreads, coffee whitener, confectionery, bakery, cooked meat products, mayonnaise, and yoghurt) and beverages (e.g., UHT beverages, infant milk formula, high protein beverages for sports performance /health, cream liqueurs). They are also frequently used in formulating recombined milk, protein-fortified milk or reassembled 'milk' that is converted into products such as cheese and yoghurt. In these foods, protein provides a range of techno-functionalities including water binding, viscosity, surface activity, rennet-gelation, acid-gelation and heat stability. These functionalities are ultimately determined by the degree of protein aggregation. A low tendency of the proteins to aggregate (e.g., during heating, acidification, blending with ethanol) is conducive to stability and liquidity, while higher aggregation favours the formation of more viscous or stiffer structures such as gels (e.g., on quiescent acidification or rennet treatment) or flocs (e.g., on heating while agitating)

The current project focused on the aggregation behavior of milk protein ingredients, and how this is affected by the type of dairy ingredient, manufacturing conditions, and the composition of the solvent used for reconstitution. Protein aggregation was evaluated primarily by the tendency of dispersions of the milk protein ingredients to undergo heat-or ethanol-induced flocculation, and acid- or rennet-induced gelation.

2. Questions addressed by the project:

- How does ingredient type affect the aggregation behavior of milk proteins, and hence the processing characteristics/functionality of milk to which the ingredients are added to fortify protein content?
- How do selected manufacturing parameters and composition of solvent influence the functionality of reconstituted skim milk powder, phosphocasein and milk protein concentrates?
- Are differences in the aggregation behavior of reconstituted milk protein ingredients related to changes in physicochemical parameters (e.g., protein hydration, particle size and charge) and/or the partitioning of milk proteins and minerals between the serum and non-sedimentable phases?

3. The experimental studies:

The protein content of skim milk was fortified from 3.3 to 4.1% using skim milk powder (**SMP**), sodium caseinate (**NaCas**), calcium caseinate (**CaCas**), native phosphocasein (**NPC**) or calcium-reduced phosphocasein (**CaRPC**). The effects of these ingredients on the composition, physicochemical properties, and aggregation behavior (e.g., heat coagulation and ethanol stability in the pH range 6.2-7.2, rennet gelation) of the fortified milk were investigated.

The effects of altering manufacturing steps during the production of skim milk powder on the aggregation behavior, composition and physicochemical properties of the reconstituted skim milk powder was evaluated. The manufacturing steps investigated included milk heat treatment (72 °C for 15 s or 120°C for 120 s), pH during heat treatment (6.2, 7.2 or 7.5), evaporation and drying.

Milk protein concentrate powders (MPC, ~81% protein) were made from skimmed milk which was heat-treated at 72°C for 15 s (LHMPC) or 85°C for 30 s (MHMPC). The resultant MPC powders were reconstituted to 4.1% protein in water or milk permeate. The aggregation behavior and physicochemical properties of MPC dispersions (4.1% protein) were studied.

4. Main results:

Effects of adding ingredients on the aggregation behavior of protein-fortified milk

- The use of NPC instead of SMP to fortify milk protein from 3.3 to 4.1% enhanced the rennet gelation properties but reduced heat coagulation time in the pH region 6.7 – 7.2. In contrast, fortification with NaCas, CaCas or CaRPC had an opposite effect, resulting in poorer rennet gelation, enhanced heat stability in the pH region 6.7 - 7.2, and ethanol stability at pH 6.4.

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- A further study involving the use of NaCas at varying levels to fortify milk protein from 3.3 to 4.1% showed that: (i) the adverse effect of NaCas on rennet gelation became progressive with level added in the range 0.1 to 0.3%, and that rennet gelation failed to occur on addition at levels ≥ 0.4%; and (ii) the positive effect of NaCas on heat coagulation time and ethanol stability increased progressively with level added in the range 0.1 to 0.8%.
- The different ingredients exerted their effects on the evaluated aggregation characteristics by altering the degree of dissociation of casein and calcium from the casein micelle to the serum.

Effects of manufacturing steps on the aggregation behavior of reconstituted low-heat and high-heat skim milk powders

- Increasing the severity of heat treatment from 72 °C x 15 s to 120 °C x 120 s, prior to evaporation and drying had significant effects on the aggregation behavior of skim milk and skim milk concentrates prepared by reconstitution of the resultant skim milk powder. It resulted in higher heat coagulation time (HCT) at pH 6.3-6.6 and ethanol stability (ES) at pH 6.2 - 6.6, and a marked deterioration in the rennet-induced coagulability of the resultant skim milk. It also led to a significant increase in the heat coagulation time of skim milk concentrates with 20-25% total solids in the pH region 6.3 - 6.6. The effects of increasing heat treatment were associated with higher levels of whey protein denaturation and casein dissociation, an increase in casein micelle size, and a reduction the concentration of ionic calcium.
- Increasing the pH of skim milk from ~ 6.6 to 7.2 or 7.5 prior to high heat treatment (95 °C x 2 min), followed by restoration to pH 6.6 after heating, enhanced heat stability at pH 6.6-6.7, but did affect the rennet coagulability or ethanol stability of the milk prepared by reconstitution of the skim milk powder. The positive effect on heat stability coincided with increases in casein dissociation and the concentration of serum-soluble κ-casein/whey protein aggregates, and reductions in casein micelle size and concentration of serum calcium.
- The evaporation and drying stages of manufacture of skim milk powder had little, or no, effect of skim milk prepared on reconstitution of skim milk.

Effects of milk heat treatment during the manufacture of milk protein concentrate, and the composition of the solvent used for reconstitution, on the aggregation behavior of milk protein dispersions

- Increasing milk heat treatment during the manufacture of milk protein concentrate powder (MPC) from 72 °C x 15 s to 85 °C x 30 s significantly impaired the rennet gelation properties of MPC dispersions (4.0% protein), and reduced the viscosity of stirred skimmed milk yoghurt prepared from the MPC dispersion. However, it had little, or no, effect on the heat stability or ethanol stability of the dispersions. The effects were associated with a higher level of whey protein denaturation and lower concentrations of serum protein and serum calcium.
- The use of milk permeate instead of water (as solvent for the preparation of MPC dispersions) enhanced ethanol stability at pH 6.6–7.0, impaired rennet gelation, changed the heat coagulation time/pH profile from type B to type A, and reduced the viscosity of acid-induced gels. These effects were associated with differences in ionic strength of the solvent which affected the degree of κ-casein dissociation, micelle size and charge, and concentration of ionic calcium.

5. Opportunity/Benefit:

The data provide new knowledge and insights on the factors controlling protein aggregation and functional properties of reconstituted milk protein ingredients in applications involving heat treatment, acidification, rennet treatment, or blending with ethanol.

6. Dissemination:

Main publications:

Lin Y., Kelly, A.L., O'Mahony J.A. and Guinee T.P. (2016) 'Fortification of milk protein content with different dairy protein powders alters its compositional, rennet gelation, heat stability and ethanol stability characteristics' *International Dairy Journal* 61:220-227.

Lin Y., Kelly, A.L., O'Mahony J.A. and Guinee T.P. (2017a). Seasonal variation in the composition and processing characteristics of herd milk with varying proportions of milk from spring-calving and autumn-calving cows. *Journal of Dairy Research*, *84,* 444-452.

Lin Y., Kelly, A.L., O'Mahony J.A. and Guinee T.P. (2017b) 'Addition of sodium caseinate to skim milk increases non-sedimentable casein and causes significant changes in rennet-induced gelation, heat stability, and ethanol stability' *Journal of Dairy Science* 100: 908-918.

Lin Y., Kelly, A.L., O'Mahony J.A. and Guinee T.P. (2018a) 'Altering the physicochemical and processing characteristics of high heat-treated skim milk by increasing the pH prior to heating and restoring after heating' *Food Chemistry* 245: 1079–1086.

Lin Y., Kelly, A.L., O'Mahony J.A. and Guinee T.P. (2018b). 'Effect of heat treatment, evaporation and spray drying during skim milk powder manufacture on the compositional and processing characteristics of reconstituted skim milk and concentrate' *International Dairy Journal* 78: 53-64.

Lin Y., Kelly, A.L., O'Mahony J.A. and Guinee T.P. (2018c).Effects of milk heat treatment and solvent composition on physicochemical and selected functional characteristics of milk protein concentrate. *Journal of Dairy Science* (In Press)

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