

Dairy Foods: Physico-Chemical Properties

631 Development of whey protein concentrate incorporated dietetic kulfi. H. G. Ramachandra Rao*¹ and A. Giri², ¹*Dairy Science College, Bangalore, Karnataka, India*, ²*National Dairy Research Institute, Karnal, Haryana, India*.

Kulfi, a traditional Indian frozen dairy product has a similar composition to that of ice cream. Kulfi is made by concentrating whole milk (5% fat and 8.5% SNF) by 2 fold and then added with sugar, sodium alginate, glycerol monostearate and cardamom. The mix is then pasteurized, placed in molds and hardened to the consistency of ice cream. The objective of our study was to produce kulfi with reduced sugar and enriched with whey protein concentrate without compromising the sensory quality. Stevia, non-caloric natural sweetener, which is about 300 times sweeter than sucrose and heat stable up to 200°C was used as a sugar replacer to an extent of 50, 60 and 70 by 0.05, 0.06 and 0.07% respectively. At higher levels of sugar replacement, there was significant decrease of specific gravity, melting rate, and significant increase of freezing point, hardness and fat, protein, ash and moisture percentage. 50% sugar replaced kulfi was adjudged by a panel of 5 experts (using ice cream score card) on par with the control in sensory characteristics. Above 50% sugar replacement there was lack of brownish appearance, increased bitterness and presence of icy texture. Further, the study was conducted with incorporation of different levels of whey protein concentrate (WPC) at 0, 2, 3 and 4% respectively on the basis of concentrated milk and mixing thoroughly and hardening at -20°C for 8 h. When the levels of WPC addition increased there was significant decrease of freezing point (due to higher levels of soluble constituents - whey protein, lactose and mineral from WPC), melting rate (for improved water binding ability and stability for whey proteins), hardness (for water binding property which imparts reduced iciness) and moisture percentage but significant increase of specific gravity (for increase water binding ability of WPC), protein percentage and total calorie content. Among control and treated samples up to 3% addition of WPC overall acceptability score increased due to increase body and texture for excellent functional properties of whey protein but above 3% addition of WPC there was detectable whey flavor and excess softness. Statistically significance was tested by employing ANOVA and comparison between means was made by critical difference (CD) value. It was concluded that kulfi made with 50% replacement of sugar using stevia and 3% WPC incorporation was acceptable.

Key Words: kulfi, stevia, whey protein concentrate

632 Application of ultrasound spectroscopy to monitor lactose crystallization. J. K. Amamcharla*¹, L. E. Metzger¹, and R. Tweedie², ¹*Midwest Dairy Foods Research Center, Dairy Science Department, South Dakota State University, Brookings*, ²*Industrial Tomography Systems plc, Manchester, UK*.

Lactose accounts for about 75 and 85% of the solids in whey and deproteinized whey, respectively. Production of lactose is usually carried out by a process called crystallization. Several factors including rate of cooling and mixing speed influence the crystal characteristics. Therefore, it is important to monitor the crystallization process. The objective of the present work was to use ultrasound spectroscopy as a tool to monitor the lactose crystallization process. Unlike conventional optical spectroscopy techniques, ultrasound spectroscopy can be used to characterize highly concentrated or optically opaque samples. An in situ ultrasound probe operating in pulse-echo mode connected to an ultrasound spectrometer (U2s, Industrial Tomography Systems, Manchester, UK) was used to acquire attenuation and velocity of ultrasound over the frequency range

5 and 20 MHz. In the first preliminary experiment, it was found that the attenuation of ultrasound in under-saturated lactose solutions was not influenced by temperature and lactose concentration. In the second preliminary experiment, it was found that the ultrasound attenuation increased along with an increase in the amount of crude lactose crystals. Lactose crystallization studies were conducted in a batch crystallizer (1000 mL). Supersaturated lactose solutions (70 g/100 g water) were prepared and crystallized using an isothermal batch process at 25°C. At regular intervals, 1mL of the crystal suspension was removed from the crystallizer and centrifuged to separate the crystals from the aqueous phase. The dissolved lactose in the aqueous phase was measured using a refractometer to determine crystallization kinetics. The separated crystals were washed with cold water (<5°C) and an image was acquired under 10× magnification using a camera attached to a microscope. As soon as the crystals appeared (20 µm), the ultrasound attenuation increased at frequency 12 MHz. Subsequently, the attenuation at other frequencies also increased as the concentration of lactose in soluble phase decreased. Overall ultrasound spectroscopy shows potential for monitoring the lactose crystallization process.

Key Words: ultrasound, lactose crystallization, in situ

633 Heat induced aggregation of whey proteins as influenced by shear, pH, and protein concentration. M. Dissanayake, L. Ramchandran, and T. Vasiljevic,* *Advanced Food Systems Faculty Research Unit, School of Biomedical and Health Sciences and Institute for Sustainability and Innovation, Victoria University, Werribee Campus, VIC, Australia*.

Microparticulation of whey proteins (WP) is understood to improve their functional properties that could enhance their utilization. However, shear forces used in microparticulation of WP can modulate their functional properties by altering their structure. The main objective of this study was to monitor the effect of simultaneous heating (90°C/20 min) and shearing (100, 500 or 1000/s) on WP aggregation at different protein concentrations (5, 10, 17.5 or ~25% w/w) and pH (3, 5 or 7). WP retentates (Warrnambool Cheese and Butter Factory, VIC, Australia) were simultaneously subjected to shear and heat using rheometer geometry. The sheared and heat treated WP were then analyzed for the conformational and structural changes by measuring turbidity, gel color, PAGE and surface hydrophobicity. Results from these tests indicated that application of shear reduced the formation of molecular associations including covalent bonds particularly at pH 7 while the reverse was true at pH 5. Surface hydrophobicity of WP increased significantly ($P < 0.05$) when sheared at pH 5 suggesting greater denaturation than the control and at pH 3 and 7, which further increased significantly ($P < 0.05$) with increase in shear rate at protein concentrations up to 10%. Turbidity tests indicated that shear forces significantly increased ($P < 0.05$) aggregate growth at pH 5, while at pH 3, protein concentration significantly increased ($P < 0.05$) the size of aggregates. Thus, the rate of denaturation and size of resulting aggregates was dependent on pH, protein concentration, and intensity of the shear forces applied. However, shear rate alone did not affect ($P > 0.05$) the color (lightness, L^*) of the shear heat-induced WP gels but the effect was significant ($P < 0.05$) along with changes in pH and protein concentration. It was concluded that simultaneous heating and shearing of WP could modify the surface properties of the WP that could change the direction and extent of heat-induced WP denaturation and aggregation.

Key Words: whey proteins, aggregation, heat/shear

634 Effect of pH and protein concentration on denaturation kinetics of whey proteins. M. Dissanayake, L. Ramchandran, and T. Vasiljevic,* *Advanced Food Systems Faculty Research Unit, School of Biomedical and Health Sciences and Institute for Sustainability and Innovation, Victoria University, Werribee Campus, VIC, Australia.*

Microparticulation of whey proteins (WP) has been implied to help overcome the loss of protein functionality accruing from traditional heat processing. This study examined the reaction kinetics of WP aggregation as a function of pH (4–6) and protein concentration (10, 17.5 and 25%, wt/wt) to help devise a more feasible pathway to produce microparticulated WP powder having small particle size. WP retentates (Warrnambool Cheese and Butter Factory, VIC, Australia) were heat treated at 140°C for 5–20 s in an oil bath. The thermal behavior of the WP was assessed using differential scanning calorimeter while the kinetics of denaturation was examined using capillary electrophoresis. The extent of protein aggregation was monitored by measuring solubility and turbidity. The denaturation and aggregation of β -Lg appeared to follow the first order reaction kinetics under steady state conditions. The reaction rate constant (k) obtained suggested distinctly higher rate of denaturation and aggregation when WP dispersions having 10% protein concentration were heated at pH 5. Also, the decline in k values was more noticeable at pH 4 than at pH 6 indicating a slower rate of denaturation at pH 4. At higher concentrations of protein, the rate of aggregation was not prominent at pH 5. Heat induced changes at low protein concentration reduced the solubility of WP regardless of pH, the reduction being significant ($P < 0.05$) during the first 5 s of heating at pH 5 and 6. The rate of decrease in solubility was significantly ($P < 0.05$) greater at pH 6 than at pH 4 and 5. The turbidity of WP dispersions at pH 4 were significantly higher ($P < 0.05$) than those at pH 5 and 6, suggesting that WP were most stable against heat induced denaturation at high protein concentration and low pH. Results from differential scanning calorimetric studies indicated that rate of WP denaturation varied with protein concentration and pH, being highest at 10% concentration and pH 5. It was concluded that microparticulation of WP at low pH and high protein concentration could produce heat stable WP ingredients.

Key Words: whey proteins, kinetics, denaturation

635 Comparison of heat stability of bovine milk subjected to UHT and in-container sterilisation. B. Chen,* F. Ren, A. Grandison, and M. Lewis, *University of Reading, Reading, UK.*

The objective of this study was to compare how the heat stability of bovine milk was affected by the UHT and in-container sterilization. In addition, stabilizing salts are added to bovine milk to decrease ionic calcium concentration in milk and the roles of different stabilizing salts (di-sodium hydrogen phosphate and tri-sodium citrate) in both heat treatments were evaluated. Heat stability was assessed by measuring the amount of sediment in the bovine milk. Sediment formation provides a robust way of measuring heat stability during in-container sterilization, as standard deviations were small for replicated determinations of the same sample. Without stabilizing salts, bovine milk produced more sediment when subjected to UHT processing compared with in-container sterilization. Addition of up to 12.8 mM stabilizing salts resulted in a significant ($P < 0.05$) increase in sediment for in-container sterilization. This arises due to the considerable increase in measured casein micelle size during in-container sterilization. The existence of this lower heat stability region has not been specifically pointed out previously, since it is not intuitive that reducing ionic calcium will decrease heat stability. In contrast, adding 6.4 mM DSHP (di-sodium hydrogen phosphate) initially reduced sediment formation in UHT treated milk but this increased on addition of 12.8 mM DSHP. However, Adding up to 12.8 mM TSC

(tri-sodium citrate) resulted in a continuous increase in sediment in UHT processing. Adding these stabilizing salts to bovine milk increased pH, decreased ionic calcium and increased casein micelle size. Adding up to 2 mM calcium chloride increased sediment formation significantly ($P < 0.05$) more after UHT treatment than after in-container sterilization. These results for bovine milk are in agreement with trends found for heat stability of caprine milk, which have been published previously. It was concluded that there is no single mechanism or set of reactions that cause milk to produce sediment during heating and that the kinetics are different for UHT and in-container sterilization processes. Poor heat stability could be induced by both increasing and decreasing ionic calcium.

Key Words: heat stability, UHT, in-container sterilization

636 Investigating the influence of phospholipids on the viability of *Streptococcus thermophilus* and *Bifidobacterium lactis*. B. Chinnasamy* and S. Clark, *Food Science and Human Nutrition, Iowa State University, Ames.*

The potential health benefits and improved extraction methods of dairy phospholipids from by-products such as buttermilk and whey have opened new avenues for dairy phospholipids (PL) application in foods; one such application is fortification of yogurt with PL. However, before fortification, it is critical to evaluate if PL inhibit common lactic acid bacteria (LAB) used in yogurt. The objective of this research is to determine the influence of PL on viability of *Streptococcus thermophilus* (ST), *Bifidobacterium lactis* (BL), *Lactobacillus delbrueckii* ssp. *bulgaricus* (LB) and *Lactobacillus acidophilus* (LA) in yogurt. In preparation for work in yogurt, ST and BL were grown in M17 and MRS broth respectively, fortified with one of 5 PL: phosphatidylcholine (PC), phosphatidylethanolamine (PE), phosphatidylinositol (PI), phosphatidylserine (PS), and sphingomyelin (SM). Growth was monitored at a) optimum growth temperatures and b) 5°C above the optimum growth temperatures (42°C \pm 1 and 47°C \pm 1 for ST and 37°C \pm 1 and 42°C \pm 1 for BL, respectively). The temperature spike of 5°C from optimum growth temperatures was selected to “stress” bacteria in a way that may occur during yogurt processing. The PL were fortified at the rate of 0.01% wt/vol. The LAB were enumerated at 0, 3, 6 and 9 h time intervals using the pour plate technique. At optimum temperature, after 9 h, ST and BL counts tended to be higher when media were fortified with PL than when not. At elevated temperatures, most PL showed a similar trend, except ST, which was slightly inhibited by PS and PI, and BL by PE. No statistically significant differences ($P > 0.05$) were recorded in the viability of ST and BL in the presence or absence of PL or across temperatures. The results suggest that phospholipids do not have profound beneficial or inhibitory influences on ST and BL. However, viability studies in yogurt and on the remaining lactic acid bacteria must be conducted to determine fortification of PL in yogurt is a promising venture.

Key Words: phospholipids, *Streptococcus thermophilus*, *Bifidobacterium lactis*

637 Elucidating the role of α_2 -casein in the superior functionality of acid gels prepared from high-pressure-treated milks compared with heat-treated milks. H. Patel^{1*}, P. Salunke³, L. Creamer², and H. Singh², ¹*Fonterra Research Centre, Palmerston North, New Zealand*, ²*Riddet Institute, Massey University, Palmerston North, New Zealand*, ³*South Dakota State University, Brookings, SD.*

The objective of this study was to compare the functional properties of heat- and high-pressure-treated milks and elucidate the mechanisms

for differences in their functionality. Samples of low heat skim milk powder reconstituted to 10% total solids (wt/wt) were heat treated at 90°C for 10 min or pressure treated at 600 MPa for 3 min. Acid gels were prepared from these heat- and pressure-treated milk samples and rheological measurements (G' , G'' , $\tan\delta$) were carried out. Samples were also centrifuged into a soluble (serum) phase and an insoluble casein micelle (pellet) phase and analyzed using PAGE techniques to study the interactions of proteins as affected by heat and pressure treatments of the samples. Both the heat- and pressure-treated samples generated a range of disulfide-linked aggregates, which were much larger in the heat-treated samples than the pressure-treated samples. The acid gel strengths were significantly greater in pressure-treated samples (1000 Pa) compared with heat-treated samples (200 Pa). The PAGE techniques suggested that the casein-whey protein (WP) complexes in the soluble phase consisted mainly of disulfide-linked complexes of κ -casein with β -lactoglobulin and other WPs, with little evidence of the presence of α_{s2} -casein, in the heat-treated samples, but contained significantly higher proportions of both α_{s2} - and κ -casein in the pressure-treated samples. The higher acid gel strengths of the pressure-treated samples, compared with the heat-treated samples, can be explained by the presence of greater amounts of soluble casein-WP complexes and the involvement of α_{s2} -casein (in addition to κ -casein) in the disulfide-linked interactions with WPs. Generally, α_{s2} -Casein is buried inside the casein micelles and is not easily accessible for casein-WP interactions whereas κ -casein protrudes outside the micelles and is readily available. High-pressure treatment affects the colloidal calcium phosphate complex; it dissociates/solubilizes the casein micelles, therefore exposing α_{s2} -casein and making it readily available, whereas only the κ -casein is accessible in heat-treated samples. These results clarify that α_{s2} -casein has a special role to play in improving the functional properties of high-pressure-treated milks.

Key Words: high pressure, heat, casein

638 Coagulation properties of the casein micelle by combination of ultrafiltration and dilfiltration measured using rheology and diffusing wave spectroscopy. J. G. Luo*^{1,2}, E. Kristo¹, and M. Corredig¹, ¹Department of Food Science, University of Guelph, Guelph, ON, Canada, ²Gay Lea Foods Co-operative Ltd., Guelph, ON, Canada.

The objective of this work is to understand how casein micelles (CM) are affected during milk protein concentrates (MPC) manufacturing and what effect this may have on its coagulation properties. The changes in structure function of the CM occurring in MPC are a topic of great interest in dairy technology. There is limited research of coagulation properties of the casein micelle during MPC manufacturing. This study investigates the changes in the renneting functionality of fresh retentates and reconstituted retentates by combination of ultrafiltration and dilfiltration. Reconstitution was carried out in milk or milk serum (permeate). The rennet gelation behavior of reconstituted retentate was also compared with a control reconstituted freeze-dried skim milk powder (6% protein level). The renneting behavior of the CM was followed using rheology and diffusing wave spectroscopy (DWS). The release of caseinomacropptide was also quantified. Soluble calcium and total calcium were measured to understand the effect of this ion on the mechanism of the gelation. Concentrated fresh retentate and reconstituted MPC (17.5% protein) formed significantly stiffer gels

than regular samples (6% protein) due to increased number of bonds in the network. There were no significant difference in casein micelle size between fresh retentate and reconstituted retentates. The reconstituted retentates at 6% protein were dialyzed against skim milk to ensure the ionic equilibrium of the samples were similar to skim milk. The release of the caseinomacropptide during renneting was also not significantly different among the samples tested at the same protein concentration. No significant differences were noted in the rennet coagulation time as tested by both DWS and rheology. After dialysis against milk, the gelation time of the samples was shorter and the gel modulus higher than for the nondialyzed samples. Based on our result, we conclude that ionic equilibrium is important in the reconstitution of MPC for rennet-induced gelation of milk.

Key Words: ultrafiltration, dilfiltration, rennet

639 Composition and physical properties of dairy products in the UK. B. Chen,* A. Grandison, and M. Lewis, *University of Reading, Reading, UK.*

The objective of this study was to investigate the effect of raw milk composition on some selected properties of milk products. Raw bulk cow milk was collected and its composition and physical properties were measured every 2 weeks. This milk was then converted to a range of products using standardized methodology and selected properties of these products were measured. Products include evaporated milk, soft cheese, skim milk powder, whipping cream, UHT milk and in-container sterilized milk. Results presented are those from the first 6 batches of raw milk, collected over the period August to December 2011. This project is ongoing and will be replicated with a minimum of 25 batches of raw milk. The range of values for fat and protein were 4.04 to 4.77% and 2.89 to 3.29% respectively. Ranges for lactose (4.26 to 4.69%), total solids (12.45 to 13.32%), pH (6.74 to 6.78), buffering capacity (pH change from 0.83 to 0.88) and ethanol stability (87 to 94%) were narrower. Ca^{2+} concentration ranged from 1.75 to 2.55 mM. Sediment in raw milk was very low, ranging from 0.10 to 0.12% (dry weight basis). Viscosity ranged from 1.56 to 2.31 cp and density from 1.026 to 1.031 g/cm³. Casein micelle size ranged from 160 to 200 nm and freezing point depression from -0.525 to -0.514. The longest foaming times were 75 and 61 s for raw and skim milk, respectively, and the shortest were 24 s and 19 s. Foam stability was more variable, ranging from 76 s and 72 s to 213 s and 435 s for raw and skim milk, respectively. The viscosity range for evaporated milk was wide and was also affected by the level of added stabilizer. The range of for sediment for UHT and in-container sterilized milk was from 0.16 to 0.29% and from 0.18 to 0.31% respectively. Sediment formation was always accompanied by an increase in casein micelle size. For the whipping cream, the overrun and hardness were constant, ranging from 151 to 153% and 0.13 to 0.15 N, but the range of stability was more variable (14.63 to 18.75 mL). The heat stability of skim milk powder showed considerable variability. The ranges of the moisture content and hardness in soft cheese were 55.5 to 59.1% and 1.27 to 2.26 N respectively. It was concluded that variations in raw milk composition influence the properties of manufactured milk products.

Key Words: raw milk quality, physico-chemical properties, best use for milk