**T212 Comparison of milk fatty acids composition from buffalo, camel, cow, goat, and yak.** J. H. Yang, D. P. Bu*, J. Q. Wang, L. Ma, J. X. Zhang, and J. T. Chen, *Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China.*

Milk originated from different mammals has its own characteristic. The objective of this study was to investigate variation of FAs profile in milk from different dairy mammals. Milk samples collected from buffalo (n = 22), camel (n = 19), cow (n = 108), goat (n = 23) and yak (n = 22) were analyzed in this study. The FA methyl esters were determined via Agilent 6890 GC (Agilent Technologies, Hewlett Packard Co., Avondale, PA) fitted with a flame-ionization detector and expressed as grams per hundred grams. Data was processed through multi-comparison and principle component analysis (PCA), completed by SAS 9.0 (SAS Institute Inc., Cary, NC) and the Unscrambler 9.8 (Camo Software AS, Oslo, Norway), respectively. Results showed that composition (%) of camel milk contained the lowest C4-C12 (all below 0.90) and the highest C16:1 (3.51), C18 (21.30), C18:1 (35.58), c9c12C18:2 (2.66), c9t11C18:2 (1.85), C18:3 (1.81), C20:1 (0.22), C20:2 (1.52) and C20:3 (0.53). Goat milk had the highest C8 (2.92), C10 (10.24)and C12 (5.30). C4 (6.43) in buffalo milk and C14:1 (0.94) in cow milk were higher than in others, while yak milk highlighted FAs above 14 carbons without unsaturated bond like C15 (1.94), C17 (1.56), C20 (0.48) and C22 (0.35). Moreover, there was no difference in saturated FA (71.67–73.47%) and monounsaturated FA (MUFA, 24.10–25.39%) among bovine milk samples, whereas camel had maximum MUFA and polyunsaturated FAs. Short and median chain FA (sum of C4-C16:1) in buffalo (63.96), goat (62.67), cow (60.51), and yak (59.03) milk were higher than in camel milk (32.95). Scores and loading plots of PCA showed that high t11C18:1, c9C18:1, C18:0 in camel milk and high C8, C10, C12 in goat milk separated their samples from others, while buffalo, cow and yak milk clustered together for similar FA composition. It was suggested that camel milk highlight characteristic on long chain and unsaturated FA, and goat milk had special effects on short chain FA, while bovine milks exhibited much similarity.

**Key Words:** milk, fatty acid, different mammals

**T213 Characterizing the relationship between peroxidase activity and enzymatic bleaching in fluid whey.** R. E. Campbell*[1], P. D. Gerard[2], and M. A. Drake[1], *[1]North Carolina State University, Raleigh, [2]Clemson University, Clemson, SC.*

The carotenoid norbixin (annatto) is added to Cheddar cheese milk to impart desired color to cheese. Norbixin is also present in whey and is bleached using benzoyl or hydrogen peroxide. Due to increased regulations and off-flavors produced by these bleaching agents, alternative bleaching agents have been explored. Lactoperoxidase (LP), naturally in milk, or an external peroxidase (EP) may be used to bleach fluid whey or retentate with the addition of low concentrations (<50 ppm) of hydrogen peroxide. A working knowledge of enzyme activity and how activity relates to bleaching efficacy is required. The objective of this study was to characterize the relationship between enzyme activity and bleaching efficacy in fluid whey and whey protein retentate. A range of pH (5.5–6.5) and temperature (4–60°C) was evaluated. Colored Cheddar cheese whey was manufactured in triplicate from pasteurized whole milk and fat-separated pasteurized whey and whey protein retentate (10% solids, 80% protein) were manufactured. Subsequently, the effects of selected temperatures and pH in both fluid whey and whey protein retentate were evaluated for their effects on LP and EP activity and bleaching efficacy using a response surface model – central composite design (RSM-CCD) matrix. Peroxidase activity was determined using an established colorimetric method and bleaching efficacy was measured by quantifying norbixin destruction via high performance liquid chromatography. Across the pH and temperature ranges evaluated (5.5–6.5, 4–60°C), peroxidase activity associated with LP and bleaching efficacy increased with increasing temperature. The effects of pH were not as pronounced as temperature effects ($P < 0.05$). Increased peroxidase activity with the addition of EP was not evident while increased bleaching was observed, suggesting that peroxidase activity and bleaching are not necessarily linear. These results suggest that peroxidase activity is not always an accurate prediction of bleaching efficacy and that addition of exogenous EP may be beneficial to manufacturers to improve bleaching consistency across processing variables.

**Key Words:** whey protein, enzymatic bleaching, lactoperoxidase

**T214 Factors that influence the required membrane area of a multi-stage microfiltration process to separate serum protein and lactose from micellar casein in skim milk.** E. E. Hurt* and D. M. Barbano, *Northeast Dairy Foods Research Center, Department of Food Science, Cornell University, Ithaca, NY.*

Our objective was to determine how to minimize the microfiltration (MF) membrane area required to produce a micellar casein concentrate (MCC) from skim milk (SM). Several factors were studied. The 1st factor was feeding a MF system with ultrafiltered (UF) SM versus SM. Other factors (when UF SM was the MF feed) were the concentration of the UF SM, the number of MF stages and MF flux. To determine the effect of these factors on MF membrane area, a theoretical model based on a mass balance was developed. The model was based on previously measured performance of a 0.1µm ceramic uniform transmembrane pressure MF system producing a final MCC of 9% protein with ≥ 95% of the serum protein (SP) removed and <0.2% lactose from SM. The model was used to minimize the total mass of MF permeate removed by changing the concentration factor (CF) of the UF SM feeding the MF (if UF SM was the feed) and the CF and diafiltration factor for each MF stage. The MF permeate removed was converted to membrane area by specifying flux (54 kg/m$^2$ per h), processing time (18h) and mass of SM before UF (150,000 kg). It was assumed that 76% of the lactose had been removed from SM by UF. A 95% removal of lactose (equal to the SP removal) from this UF SM by MF would produce a MCC with <0.2% lactose. If the feed was UF SM, the MF membrane area required for a 5-stage process was 182 m$^2$ compared with 315 m$^2$ when SM was the feed. For 5-stages, increasing the UF SM CF from 1.0 to 1.7 decreased MF membrane area from 202 m$^2$ to 182 m$^2$. The UF SM CF that minimized MF membrane area for 2, 3, 4 and 5-stages was 0.8, 1.2, 1.5 and 1.7 respectively. Increasing the number of stages also decreased the required MF membrane area. When UF SM was used, increasing the number of stages from 2 to 5 decreased MF membrane from 297 m$^2$ to 182 m$^2$. Finally, for 5-stages, increasing the flux from 54 to 60 kg/m$^2$ per h decreased the MF membrane area from 182 m$^2$ to 164 m$^2$. Using UF SM, the UF SM CF, the number of MF stages and flux were all important factors for minimizing the membrane area of a MF system.

**Key Words:** microfiltration, micellar casein concentrate, optimization

**T215 The impact of bleaching on functionality of whey protein isolate.** T. J. Smith*, E. A. Foegeding, and M. A. Drake, *North Carolina State University, Raleigh.*

Whey protein is a highly functional food ingredient used in a wide variety of applications. A large portion of whey produced in the United States is derived from Cheddar cheese manufacture and contains annatto, and therefore must be bleached. We have demonstrated bleaching effects on whey protein flavor but bleaching effects on protein functionality have not been fully elucidated. The objective of this study was to compare functional properties of bleached and unbleached whey protein isolate (WPI). Cheddar whey was manufactured followed by addition of no bleach (control), hydrogen peroxide (HP, 250 mg/kg) or benzoyl peroxide (BP, 50 mg/kg) at 50°C for 1 h. WPI were then produced and spray dried. WPI were manufactured in triplicate. Functional properties were evaluated by measurement of foam stability, protein solubility, native PAGE, and the effect of sodium chloride (NaCl) concentration on gelation. In addition to functional properties, descriptive analysis and gas chromatography-mass spectrometry were performed to characterize flavor differences among treatments. Overrun and yield stress were not different between bleached and control WPI ($P < 0.05$). Foam stability of WPI was increased in both bleached treatments relative to the control ($P < 0.05$). Soluble protein loss at pH 4.6 was decreased by bleaching with either HP or BP ($P < 0.05$). The effect of NaCl concentration on gelation was also distinct with bleaching agent ($P < 0.05$). Lastly, native PAGE results suggested that both HP and BP contributed to protein degradation, which was likely a factor in the functional differences observed between unbleached and bleached WPI. These results suggest that chemical bleaching does have a significant effect on many of the functional characteristics of whey proteins.

**Key Words:** functionality, WPI, flavor

**T216 Optimizing methods for improved raw milk analysis by NIR spectroscopy.** T. J. Reuter*[1], X. Xiong[2], G. Rolland[2], and T. C. Schoenfuss[1], *[1]University of Minnesota, St. Paul, [2]BHI Labortechnik AG, Flawil, St. Gallen, Switzerland.*

Raw milk is hard to analyze by NIR because of excessive light scattering effects caused by large size variations in the fat component. How the sample is prepared and presented for NIR analysis could improve predictions of component quantities. In this study, sample presentation (static and dynamic) and fat particle size reduction by various homogenization processes were investigated for 160 diverse samples to predict 5 component quantities in raw milk (fat, moisture, ash, protein, casein) by FT-NIR. Spectra were acquired with a polarization inferometer in transflection in the wavelength range 1000 to 2500 nm, and preprocessed using standard normal variation scatter correction and Savitzky-Golay smoothing. The calibration models were created using PLS method. Excellent results were obtained for the standard errors of prediction following no homogenization (0.02, 0.02, 0.01, 0.01, and 0.01) for fat, moisture, ash, protein, and casein, respectively. Results from homogenization with sonicator and tube dispersion methods were not significantly improved from non-homogenized samples, but were still better compared with 2-stage valve homogenization for moisture and casein components (0.12 and 0.07, respectively) possibly because of whey protein denaturation that occurred during 2-stage processing. Relatively high sample temperatures during analysis could have led to more positive results for all component calibrations compared with reference values. For sample presentation, both the static and dynamic flow cell methods were shown to be very accurate based on the lowest standard errors of prediction (0.02, 0.02, 0.01, 0.01, and 0.01) for fat, moisture, ash, protein, and casein, respectively. The Petri dish presentation method was similarly accurate but may have been limited by the poor structural design of the transflectance cover, which allowed for sample dehydration and loss of reflected light.

**Key Words:** near-infrared spectroscopy, milk analysis, sample preparation

**T217 The mechanism of resistance to plasmin activity through protein succinylation: A model study using β-casein.** H. Bhatt*[2,1], A. Cucheval[1], C. Coker[1], H. Patel[3], A. Carr[2], and R. Bennett[2], *[1]Fonterra Research & Development Centre, Palmerston North, Manawatu, New Zealand, [2]Institute of Food, Nutrition and Human Health, Massey University, Palmerston North, Manawatu, New Zealand, [3]Dairy Science Department, South Dakota State University, Brookings.*

Plasmin-induced proteolysis is a major concern in the dairy industry, leading to defects in the texture and taste e.g., gelation and bitterness, of various milk systems. It is therefore important to control the activity of plasmin in milk systems. Several techniques have been investigated; the present work explores inhibition of the plasmin-catalyzed hydrolysis of β-casein, which is the primary target of plasmin action. As plasmin hydrolyses proteins on the carboxyl site of lysine-X and arginine-X bonds with a preference for the lysine-X bond, lysine residues on the β-casein backbone were targeted by succinylation. The target lysine sites were identified by liquid chromatography-tandem mass spectrometry to be Lys-28, 29, 99, 105 and 107. To get greater insight into the effect of different levels of succinylation on the hydrolysis of β-casein by plasmin, the reaction was monitored by quantifying the formation of the hydrolyzed product using sodium dodecyl sulfate PAGE and reverse-phase high performance liquid chromatography. This allowed the primary stage of the hydrolysis of β-casein, i.e., the formation of γ-caseins and proteose peptones, and the secondary stage, i.e., further hydrolysis of the γ-caseins and proteose peptones, to be distinguished. The results clearly indicated that succinylation affected both stages of hydrolysis negatively. Succinylated β-casein and proteose peptones became resistant to hydrolysis by plasmin. The following mechanism is proposed. The formation of succinyl-lysine renders the casein unrecognizable to the substrate-binding pocket of plasmin. Thus, the modified substrate cannot be positioned into the pocket and is not hydrolyzed by the catalytic triad of plasmin. These results indicate that succinylation may be useful for controlling the plasmin-induced hydrolysis of milk proteins and that the proposed mechanism may be useful as a base hypothesis for developing plasmin-resistant proteins by other means of modification.

**Key Words:** plasmin, succinylation, β-casein

**T218 Effect of processing and storage temperatures on the physical stability of sodium-caseinate-stabilized emulsions.** Y. C. Liang*[1,2], H. Patel[3], L. Matia-Merino[2], A. Q. Ye[4], G. Gillies[1], and M. Golding[2,4], *[1]Fonterra Research and Development Centre, Palmerston North, New Zealand, [2]Institute of Food, Nutrition and Human Health, Massey University, Palmerston North, New Zealand, [3]Dairy Science Department, South Dakota State University, Brookings, [4]Riddet Institute, Massey University, Palmerston North, New Zealand.*

Oil-in-water emulsions are an important basis of many food and nutraceutical products. Liquid beverages are processed at high temperature (e.g., retort or UHT processing) and maybe stored at different temperatures (e.g., at 40°C). There is little information on the effect of high heat treatment and high storage temperatures on the caseinate and droplet sizes, depletion potential, and creaming stability of caseinate-stabilized emulsions. This study investigated the influence of processing and

storage conditions on the structure and stability of caseinate-stabilized emulsions. Sodium caseinate was reconstituted to a 3% (wt/wt) solution. Corn oil (60% w/w) was mixed with the protein solution. The mixture was homogenized to yield a stock emulsion, which was then mixed with stock caseinate (20% wt/wt) to produce 30% oil-in-water emulsions containing 2 to 10% caseinate. The pH was adjusted to 6.8 ± 0.04 for all model emulsions. Each model emulsion was prepared at least in duplicates. The experimental data was analyzed by Student's *t*-tests, significant differences among the means were determined at a 95.0% confidence level. Heat-treated (120°C, 10 min) caseinate emulsions phase separated much more rapidly than unheated emulsions ($P < 0.05$). The droplet size was found to be unchanged after heating. The change in phase separation behavior can be attributed to the change in caseinate size because of the heat-induced polymerization of casein molecules. A small change in caseinate size resulted in a moderate change in the depletion interaction potential. In addition, unheated caseinate emulsions stored at 40°C phase separated much more rapidly than unheated emulsions stored at 20°C ($P < 0.05$). This change appeared to be influenced by the continuous phase viscosity. The formation of the droplet network and its rearrangement as a function of the caseinate concentration at 40°C were well reflected by small deformation rheology and Turbiscan curves. We show the interrelations between the formation of the droplet network and the processing conditions.

**Key Words:** caseinate, physical stability, depletion interaction potential

**T219 Effect of reconstitution temperatures on the solubility of different protein fractions present in milk protein concentrates (MPC 80).** H. Patel*[1,2], [1]*Dairy Science Department, South Dakota State University, Brookings,* [2]*Animal Sciences and Industry, Kansas State University, Manhattan.*

The solubility of high protein ingredients such as milk protein concentrates (MPC) influences important functional properties such as emulsification, foaming and gelling. The rate of solubility of MPC and different protein fractions present in MPC may vary depending on the reconstitution conditions. However, there is lack of reliable information on the solubility behavior of individual protein fractions during reconstitution process. Better understanding of such factors would help the manufacturers to identify optimum conditions for reconstitution. Considering this, the objective of present study was to study the order in which individual protein fractions are solubilize during reconstitution process. Five representative samples of MPC80 with similar protein (81–82%), lactose contents were obtained. Solutions (5% w/w) of these MPC were prepared by reconstituting MPC at 20, 40, 50 and 60°C. During reconstitution, the aliquots from each of this were withdrawn at different time interval (2, 10, 20, 30, 45 and 60 min). Percent solubility of these samples was analyzed using standard methods. The rate of solubility of different protein fractions was determined using PAGE under different reconstitution condition. The data were analyzed using SAS. Percent solubility of all MPC samples increased significantly ($P < 0.05$) with increase in the temperature and time of reconstitution up to 50°C and up to 30 min. The results of PAGE showed that the whey proteins were solubilized within initial 2 min of reconstitution process. The order of solubility of different proteins was found to be β-lactoglobulin = α-lactalbumin > $\alpha_{s1}$-casein > β-casein > $\alpha_{s2}$-casein > κ-casein. It required almost double time to reconstitute MPC at 20°C compared with 50°C. It was found that the disulfide interaction of casein and whey proteins were responsible for the poor solubility of some MPC samples. The manufacturers can use such insights for identifying processing conditions and, for achieving desired functionality of MPC in the final products.

**Key Words:** milk protein concentrate (MPC), solubility, polyacrylamide gel electrophoresis (PAGE)

**T220 Comparison of the in vitro digestion of raw pasture milk and commercial HTST and UHT pasteurized milk.** D. X. Ren[1,2], D. L. Van Hekken*[1], M. H. Tunick[1], and P. M. Tomasula[1], [1]*USDA, ARS, ERRC, Dairy and Functional Foods Research Unit, Wyndmoor, PA,* [2]*Zhejiang University, Institute of Dairy Science, College of Animal Science, Hangzhou, China.*

Consumption of raw milk from pasture-fed cows, typically purchased at local farms, is steadily increasing in the US because many consumers believe that high-temperature short-time (HTST) or ultrahigh temperature (UHT) pasteurization affects the digestibility of milk proteins and thus the bioavailability of their nutrients. The objective of this study was to compare the evolution of curd or clot size distribution with time during in vitro digestion of protein under simulated fasting gastro-intestinal conditions for commercial whole and skim milk treated by HTST or UHT pasteurization and raw whole and skim milk from pasture-fed cows. Milk digestion procedures followed the 2012 US Pharmacopeia with simulated gastric fluid (SGF) using pepsin and simulated intestinal fluid (SIF) using pancreatin. The in vitro protein digestibility of whole milk samples was greater than that of skim milk samples ($P < 0.05$), with milk fat acting as a barrier to the protein aggregation observed in skim milk samples. In a second series of experiments, the degradation kinetics of clots that formed upon initiation of SGF digestion were followed for 3 h using a light-scattering particle-size analyzer. The average clot sizes of the proteins upon initiation of digestion decreased in the order raw pasture > HTST > UHT milk with the clot sizes for skim milk (130.5, 128.3 and 52.9 μm, respectively) being larger than those for whole milk (98.5, 86.3 and 32.7 μm; $P < 0.05$). After 3 h of digestion, a single particle-size peak was observed for all samples. With the exception of the UHT milk samples, the clot sizes in skim milk samples averaged 38.5 μm and were larger than those in whole milk which averaged 29.3 μm ($P < 0.05$). Clot sizes of skim and whole milk UHT samples averaged 8.0 μm ($P < 0.05$), indicating that UHT milk was the most digestible since the smaller clot sizes offered increased surface area for enzyme contact. The results showed an inverse relationship between clot size and extent of in vitro digestibility. Moreover, the results indicated that raw pasture milk is as digestible as commercial HTST milk and not as digestible as commercial UHT milk.

**Key Words:** in vitro digestion, raw milk, pasteurized milk

**T221 Computer simulation to predict energy use, greenhouse gas emissions and costs for production of extended shelf-life milk using microfiltration.** P. M. Tomasula, W. C. F. Yee, A. J. McAloon, and L. M. Bonnaillie*, *USDA, ARS, ERRC, Dairy and Functional Foods Research Unit, Wyndmoor, PA.*

Extended shelf-life (ESL) milk has a shelf life between that of high-temperature short-time (HTST) and ultrahigh temperature (UHT) pasteurized milk. ESL milk is usually pasteurized at temperatures exceeding 125°C which may give the milk a cooked taste. ESL milk produced using crossflow microfiltration (MF) before HTST pasteurization at 72°C produces milk with the fresh taste of HTST milk and depending on raw milk quality, packaging and cold-chain handling, may have a shelf-life of up to 30 d. Little information is available on the additional energy use, greenhouse gas (GHG) emissions and operating costs of installing MF in an existing HTST processing plant. The objective of this study was to develop a model for milk MF and incorporate it into a computer simulation model of the fluid milk process that was recently

developed for processors to benchmark their current energy use, GHG emissions, and capital and operating costs. MF was modeled as 2 MF skids in series, each containing the housings for 1.4 µm membranes. In the simulator, skim milk leaves the separator at 55°C as the feed to the first skid. The retentate from the first skid fed the second MF skid. Permeates from both skids were blended with cream and pasteurized to produce 3.25% whole milk. Retentate (3% of the total feed stream) was added to the remaining cream stream and heated to 130°C before storage. Simulations conducted for medium–sized milk plants processing 27,300 L/h of milk showed GHG emissions, electricity and natural gas use were 88.6 $gCO_2eq$/kg milk, 0.38 MJ/L and 0.12 MJ/L using HTST pasteurization alone and 93.6 $gCO_2eq$/kg milk, 0.38 MJ/L and 0.14 MJ/L for the HTST/MF plant, showing the additional electricity and natural gas used by the MF process and for heating of the cream/retentate stream. The difference in operating costs between HTST and HTST/MF was estimated as 0.10 cents/L in agreement with literature estimates. This study demonstrates that computer simulation allows rapid assessment of process changes and evaluation of new technologies in an existing processing line without costly pilot testing.

**Key Words:** greenhouse gas, microfiltration, energy

**T222 Effect of preheating temperature and time on the properties of evaporated milk.** B. Chen*, A. Grandison, and M. Lewis, *University of Reading, Reading, Berkshire, UK.*

The objective of this study was to investigate the effect of different preheating temperature/time combinations on the properties of evaporated milk. Pasteurized whole milk was preheated at 85°C, 90°C and 95°C for 10, 20 and 30 min at each temperature. After evaporation to 28% total solids, different concentrations of disodium hydrogen phosphate and trisodium citrate were added (8 to 24 m*M*) and the density and heat stability of evaporated milk were measured after in-container sterilization. Heat stability was assessed by measuring the amount of sediment in the evaporated milk. Concurrently, pH, $Ca^{2+}$and viscosity were monitored at every processing stage. Overall, the pH and $Ca^{2+}$of raw milk was around 6.72 and 1.54 m*M*. Preheating caused only a small pH reduction and a fall in $Ca^2$ of about 0.30 m*M*, whereas sterilization caused a dramatic reduction in pH but an increase of $Ca^{2+}$. Also, viscosity of evaporated milk increased after preheating and also after evaporation but the most dramatic increase was brought about by sterilization. Higher preheating temperatures and longer preheating times resulted in evaporated milk with significantly lower pH and $Ca^{2+}$. The viscosity of evaporated milk without pre-heating was significantly higher than those which had been preheated. Addition of stabilizing salts significantly reduced the viscosity of evaporated milk. However, higher additions of up to 24 m*M* stabilizing salts sometimes resulted in a viscosity which was higher and above the range found for commercial samples. Adding increasing amounts of these stabilizing salts to evaporated milk increased pH, decreased $Ca^{2+}$ and slightly increased the dry sediment. In addition, dialysis was used for measuring pH and $Ca^{2+}$at preheating temperatures. The pH of dialysates decreased as pre-heating temperature increased but $Ca^{2+}$ did not change significantly. It was concluded that 85°C or 90°C for 10 min pre-heating were the best conditions for the evaporated milk manufacture to obtain a satisfactory viscosity.

**Key Words:** evaporated milk, preheating temperature and time, viscosity

**T223 Predicting color change of skim milk during high pressure thermal processing.** A. F. Devi[1,2], R. Buckow*[2], Y. Hemar[3], and S. Kasapis[1], *[1]School of Applied Sciences, RMIT University, Melbourne, VIC, Australia, [2]CSIRO Animal, Food and Health Sciences, Werribee, VIC, Australia, [3]School of Chemical Sciences, The University of Auckland, Auckland, New Zealand.*

The demand of extended shelf-life with fresh-like quality and retained nutrients has driven studies on high pressure treatment of milk. Inactivation of bacterial spores for milk sterilization purpose requires the incorporation of heat in the high pressure process. Currently, little is known about the influence of high pressure thermal (HPT) processing on physicochemical properties of milk. Therefore, the objective of this study was to develop a predictive model describing kinetics of color change (from white to caramel brown) of skim milk during HPT processing. Reconstituted skim milk (10% wt/wt) was treated under isothermal/isobaric conditions at 100 to 140°C and 0.1 to 600 MPa for up to 60 min. Its color was measured within 4 h after treatment using a chromameter and reported in CIE-Lab system. The total color change was calculated referring to the initial color of skim milk and represented as total color difference ($\Delta E_{ab}$*). $\Delta E_{ab}$* increased with time (t) and was described with an empirical equation: $\Delta E_{ab}^* = 38\ t/(k+t)$. Coefficient k varied with temperature and pressure, following a third order polynomial equation and reached the minimum value at 400 MPa. Consequently, $\Delta E_{ab}$* was largely enhanced during HPT at 400 MPa. Meanwhile, $\Delta E_{ab}$* increased progressively with temperature regardless of the applied pressure. Compared with atmospheric pressure, 60 min HPT treatment at 110°C and 200, 400, and 600 MPa increased $\Delta E_{ab}$* by approximately 50, 200, and 100%, respectively. Selected images of processed skim milk, mathematical models, and pressure-temperature diagrams of color conversion as a function of pressure, temperature, and treatment time will be presented. The resultant knowledge can possibly help to identify suitable HPT process conditions for improved sensory quality of sterilized milk.

**Key Words:** high pressure processing, skim milk, color

**T224 Feed substrates influence biofilm formation on reverse osmosis (RO) membranes and their cleaning efficacy.** S. Marka* and S. Anand, *Dairy Science Department, Midwest Dairy Foods Research Center, South Dakota State University, Brookings.*

In recent times, there is a greater use of RO membranes for concentration of various feed materials such as whey and ultrafiltration (UF) permeate in the dairy industry. This study compares the influence of skim milk UF permeate and cheese whey on membrane biofilm formation. A resistant *Bacillus* sp., previously isolated from a membrane biofilm, was used to develop 48-h-old static biofilms on the RO membrane pieces using UF permeate and whey as feed substrates. Biofilms were analyzed for viable counts by swab technique and for microstructure using scanning electron microscopy (SEM). The membrane cleaning process included 6 sequential steps; alkali, surfactant, acid, enzyme, a second surfactant, and sanitizer application. The data were statistically analyzed. The SEM images showed a significant difference in the microstructure of biofilms for the 2 feed substrates. In the case of UF permeate biofilms, the bacilli were found to be adherent in small groups of random arrangement that were covered with a thin film of exopolysaccharide material having protrusions. On the other hand, the bacilli in whey biofilms were found to be in bigger groups with cells arranged in long chains covered with smoother and thinner films of exopolysaccharide material without any protrusions. Differences were also noticed in the resistance pattern of these 2 types of biofilms. The mean pre-treatment counts of the 48-h UF permeate biofilms were log 5.39, which were much higher than the whey biofilms pre-treatment counts of log 3.44. After 6 steps cleaning cycle, there were 2.6 log survivors of the *Bacillus* isolate on UF permeate biofilms, whereas only 1.82 log survivors were found in whey biofilms. In conclusion, the UF permeate substrate biofilms resulted in a higher

resistance to cleaning process as compared with the biofilms developed by whey substrate. The biofilm microstructure variations for the 2 feed substrates might have caused differences in their resistance to the CIP protocol. Further studies are in progress.

**Key Words:** whey and UF permeate, biofilm, reverse osmosis membrane

**T225 Characterization of some changes in composition and physicochemical properties of casein micelles from cream to buttermilk.** M. Looney*[1], Y. Pouliot[2], M. Britten[3], and R. Jiménez-Flores[1], *[1]Dairy Products Technology Center, California Polytechnic State University, San Luis Obispo, [2]STELA Dairy Research Center, Institute of Nutrition and Functional Foods (INAF), Laval University, Quebec City, QC, Canada, [3]Food Research and Development Center (FRDC), Agriculture and Agri-Food Canada, St-Hyacinthe, QC, Canada.*

It is well-documented that the poor coagulation properties of buttermilk are attributed to the changes that occur to the casein micelles during the butter-making process. It was hypothesized that churning is a critical step for the changes that occur in composition of the casein micelles. Raw cream was processed using a rotary churn at 18°C for approximately 30 min, and buttermilk was collected for analysis. Raw milk was skimmed at 10°C by centrifuging at 3,000 × *g* for 20 min. Cream, skim milk and buttermilk were centrifuged at 60,000 × *g* for 40 min twice using imidazole buffer at pH 6.8 to isolate the micellar content in the pellet. Variation in physical properties of the casein micelles was determined using a Malvern Zetasizer. Protein profiles of UP cream, skim milk, and buttermilk were analyzed using 1- and 2-dimensional gel electrophoresis technique. Experiments were performed using 3 different batches of UP cream, skim milk and buttermilk. Statistical analyses showed that processing the buttermilk significantly increased the surface charge ($P < 0.05$) of the micelle but had no significant effect on their size ($P > 0.05$). Our results also indicate that churning of cream promoted interactions between casein micelles and MFGM proteins as shown by the more complex 2D-gel electrophoresis pattern obtained for casein micelles sedimented from buttermilk.

**Key Words:** buttermilk, casein micelle, churning

**T226 Investigation of the mechanism of membrane fouling in cold microfiltration of skim milk: A proteomics study.** T. J. Tan* and C. I. Moraru, *Department of Food Science, Cornell University, Ithaca, NY.*

The main challenge in milk microfiltration (MF) is membrane fouling, which leads to a significant decline in permeate flux over time. This work aims to elucidate the mechanism of membrane fouling in cold MF of skim milk, by identifying and quantifying the milk proteins and minerals involved in the external and internal fouling of the membrane. Skim milk was subjected to MF using a 1.4 µm ceramic membrane, at a temperature of 6 ± 1°C, cross-flow velocity of 6 m/s and transmembrane pressure of 159 kPa, for 90 min. First, RO water rinses (20°C, 5 min) were conducted to collect the loosely attached external foulants (S1) and then loosely attached internal foulants (S2) from the membrane. After that, pressurized hot water extractions (70°C, 10 min) were performed to collect the strongly attached external foulants (S3) and the strongly attached internal foulants (S4). The collected foulants were analyzed for protein content using the Bradford method. The foulants were further analyzed by liquid chromatography coupled with tandem mass spectrometry for protein identification and relative quantitation of individual proteins. The mineral composition of the foulants was analyzed using Inductively Coupled Plasma Spectrophotometry and potentiometric titration. All experiments and analyses were carried out in triplicate. Significant differences among samples were determined by Tukey's HSD test at $P \leq 0.05$. The protein concentrations for S1, S2, S3, and S4 were 62.2 ± 10.1 µg/mL, 10.7 ± 1.0 µg/mL, 6.6 ± 0.7 µg/mL, and 1.0 ± 0.1 µg/mL, respectively. All major milk proteins were identified in the 4 foulants. α-Lactalbumin and BSA were found in higher levels in S4 than in the other foulants. This suggests that these proteins strongly attached to the internal pores of the membrane, which likely results in pore constriction and subsequent flux reduction. In general, the concentration for all mineral was very small (below 7.7 ppm) in all foulants, and likely they do not have a major contribution to membrane fouling. The knowledge generated in this study can be used to identify solutions to minimize membrane fouling and thus increase the efficiency of skim milk MF.

**T227 Evaluation of thermization and $CO_2$ addition as methods of raw milk preservation.** P. R. Rocha, V. P. Voltarelli, V. O. Gaino, C. M. V. B. de Rensis, and P. C. B. Vianna*, *Universidade Norte do Paraná (UNOPAR), Londrina/PR/Brazil.*

The aim of this study was to evaluate the use of thermization or dioxide carbon ($CO_2$) addition as methods of raw milk preservation before processing. Raw milk was divided into 3 treatments: (1) Control milk (without treatments), (2) thermized milk (65°C/20 s) and (3) $CO_2$ added raw milk (until pH 6.2). The samples were stored in plastic bottles hermetically sealed at 4 ± 1°C. Raw milk was evaluated upon receipt (d 0) for physicochemical composition, standard plate count and psychrotrophic bacteria count. Samples were randomly selected and evaluated after 3, 6, 10, 13 and 16 d for proteolysis and the same microbial counts evaluated at the reception. Decreased in casein as a percentage of true protein (CN/TP) was used as an index of proteolysis. Split-plot design was used with 3 replications and the results were evaluated by ANOVA and Tukey's test, at 5% significance level. The lag phase of psychrotrophic bacteria count was analyzed by Gompertz model. Raw milk presented typical whole milk composition and the standard plate count and psychrotrophic count were $3.1 \times 10^4$ cfu/mL and $3.1 \times 10^2$ cfu/mL, respectively. Standard plate count and psychrotrophic bacteria count increased during refrigerated storage for all the samples, however, this increase was significantly higher for control milk. Based on the initial standard plate count, the time required for control milk reaches the critical count regulated by Brazilian legislation ($6.0 \times 10^5$ cfu/mL) was 10 d. For thermized and $CO_2$ added milk, microbiological counts remained below critical limits during the 16 storage days. The $CO_2$ addition and thermization extended the lag phase of psychrotrophic bacteria in ~2.2 and ~10.3 times, respectively, when compared with raw milk control. Proteolysis significantly increased during refrigerated storage for all samples. However, in this study, the increased proteolysis could not be related to greater psychrotrophic count of raw milk control. The results showed that both $CO_2$ addition and thermization are effective and can be used to preserve the quality of raw milk before processing and to prevent quality problems in dairy products.

**Key Words:** raw milk, carbon dioxide, thermization

**T228 Utilizing clean label starches in yogurts processed with challenging temperature and homogenization pressures.** B. Roa*, A. Perez, E. Yildiz, I. Potrebko, T. Shah, and L. Carr, *Ingredion Incorporated, Bridgewater, NJ.*

Formulating yogurts with thermally inhibited, clean label starches has often been challenging because of the harsh processing conditions

manufacturers utilize. This is the case when manufactures use high temperature and homogenization pressures, which degrade the integrity of many existing clean label starches. The loss of starch integrity affects the functionality and ultimately the texture of finished yogurt product. This presentation will focus primarily on the next generation clean label starch that extends the functionality of clean label starches in yogurts processed with demanding temperature/pressure parameters. The next generation clean label starch performance was compared with existing clean label starches and chemically modified counterparts, over a range of different parameters including upstream/downstream homogenization configurations. The downstream yogurts were processed over a pasteurization range of 185 to 195°F and 0 to 2610 psi total homogenization pressure. The upstream yogurts were processed over a preheat range of 140 to 170°F and 725 to 3625 psi total homogenization pressure. Based on instrumental characterization results from the Brookfield DV-II+ Viscometer and SMS TA-XT2 Texture Analyzer, the next-generation clean label starches were able to be processed with up to 1450 psi more total homogenization pressure at certain temperatures than with previously available clean label starches, while still providing the desired texture effect. Using a downstream homogenization configuration with 2610 psi total pressure, the next generation clean label sample had a viscosity of 5100 cps compared with the previously available clean label starch sample's viscosity of 1674 cps. The instrumental characterizations also demonstrated that the enhanced functionality of the next generation clean label starches yielded process tolerance comparable to highly inhibited chemically modified starches. Based on the results of this work, the next generation clean label starch was able to be processed with very demanding parameters, while still providing functionality in terms of viscosity and overall texture.

**Key Words:** yogurt, starch, processing

**T229 Proteomic evaluation of milk fat globule membrane proteins and bovine health status.** M. Vaiente*, L. Tomanek, M. Yeung, and R. Jimenez-Flores, *California Polytechnic State University, San Luis Obispo.*

Proteins associated with the milk fat globule membrane (MFGM) have been associated with many biological functions in the dairy cow. These proteins are also purported to confer nutritional and health benefits to human. However, there is little information about the effect of the health of the cow on the composition and expression of these membrane proteins. Since the overabundance of casein and whey in milk interfere with typical MFGM protein analysis, a contemporary proteomic approach offers an ideal solution. To this end, this study was aimed to observe and measure changes in the MFGM proteins collected from cow with different health status. MFGM fractions and somatic cell pellets were extracted from raw bovine milk collected from 3 healthy cows at the dairy farm on campus. These were separately pooled to form a MFGM sample and a somatic cell sample. Similarly, the samples were pooled from 3 cows exhibiting suboptimal health, indicated by elevated levels of somatic cell count. To generate the MFGM and somatic cell proteome maps, 2D gel electrophoresis was conducted and the gel images were analyzed using Delta2D program. Spot intensities were compared between samples derived from healthy and suboptimally healthy cow by using permutation ANOVA ($\alpha = 0.02$) available within Delta2D. The proteomes were also visualized using principal components analysis to explore differences between the proteomes and bovine health status. The proteome maps for somatic cells and MFGM proteins show differentially protein expression and distinct characteristics, including numerous putative novel proteins. Unique proteins were selected for MALDI-TOF mass spectrometry analysis and identification. Our results suggest that cow lactation physiology is markedly different under varying health status and lead to changes in the MFGM proteome. The outcome of this study further helped generate specific hypotheses to test the interaction between bovine health and individual protein species, and the role of these proteins in human health.

**Key Words:** milk fat globule membrane, proteomics, somatic cell

**T230 Production of sodium chloride nanoparticles by nanospray drying method.** M. Moncada*[1], K. Aryana[1,2], C. E. Astete[3], and C. Sabliov[3], *[1]School of Animal Sciences, Louisiana State University Agricultural Center, Baton Rouge, [2]Department of Food Science, Louisiana State University Agricultural Center, Baton Rouge, [3]Biological and Agricultural Engineering, Louisiana State University Agricultural Center, Baton Rouge.*

Reduction in sodium intake decreases blood pressure and could prevent hypertension; hypertension is a major risk factor in the development of cardiovascular diseases. One alternative to reduce sodium content without replacing sodium chloride (NaCl) in foods by other food additives is to reduce the particle size of salt without modifying its chemical composition to produce nanosalt. Reducing the particle size of sodium chloride crystal would increase dissolution rate in saliva leading to a more efficient transfer of the ions to the taste buds and hence a saltier perception of foods. The goal of this study was to develop nanosalt by using a nanospray drying method. The processing parameters were optimized to ensure formation of the smallest size nanosalt, as measured by scanning electron microscopy (SEM) and dynamic light scattering. A sodium chloride solution (3% wt/wt) prepared in deionized water was processed by nanospray drying (Nanospray dryer B-90, Büchi Corporation). The sodium chloride solution was sprayed through 2 different nozzle sizes (4 and 7 μm) at 20, 10, 5 and 3% (wt/wt). The gas flow (125 L/min), pressure (38 mbar), head temperature (95°C) and spray percentage (90%) were kept constant in all treatments. The resulting nanoparticles were analyzed by SEM to visually analyze shape and size. Of the 3 concentrations studied, the lower concentration (3% wt/wt) provided the smallest size nanosalt. Three replicates were conducted at this concentration, and particle size distribution from each nozzle size was analyzed using ANOVA of SAS. There was a significant ($P < 0.05$) difference in NaCl particle sizes using 4 and 7 μm nozzles. The NaCl solution subjected through the 4-μm nozzle had the smallest sodium chloride nanoparticle size (500–700 nm). The outcomes from these trials showed that smaller sodium chloride nanoparticles were produced when utilizing a 4-μm nozzle in the Nanospray dryer B-90 with a 3% (wt/wt) NaCl solution. Studies are on the way to test the nanosalt on the physicochemical, microbiological and sensory characteristics of surface-salted cheese crackers.

**Key Words:** nanosalt