

## Graduate Student Poster Competition: National ADSA Dairy Foods Poster

**M173** Angiostatin-like peptides in milk: Potential development for dairy products capable of cancer prevention. E. Stefanutti\* and R. Jiménez-Flores, *California Polytechnic State University, San Luis Obispo*.

Angiogenesis, the development of new blood vessels from pre-existing vascularization, represents one of cancer's hallmarks. Blood supply is essential for a tumor to progress toward a malignant condition since this allows the delivery of nutrients and oxygen necessary to support cancerous cells' growth as well as entrance into the circulation causing metastasis to arise. Study on angiogenesis inhibitors has shown promising results on animal models. Blocking cancerous cells' blood access resulted in a reduction on primary tumor size and number of metastatic colonies. One of the peptides that showed interesting results on this study was angiostatin, the internal fragment of the fibrinolytic enzyme plasminogen in blood. It is well known that bovine plasminogen gets transferred into milk during lactation. The objective of this research was to investigate enzymes capable of releasing the antiangiogenic peptide from bovine plasminogen and compare the anticancer activity of the fragment with that of human angiostatin. We tested enzymes such as *Lactobacillus* (*L. casei*, *L. reuteri*, *L. acidophilus*) and *Bacillus* (*B. subtilis*, *B. cereus*, *B. coagulans*) originated proteases, *Bacillus polymyxa* metalloprotease dispase as well as elastase. Techniques such as SDS-PAGE, column chromatography, MALDI-TOF, and Western blot were utilized. After hydrolysis, results from electrophoresis showed bands in the molecular range of 37 kDa, which were confirmed through sequence analysis to belong to the kringle 1–4 region of plasminogen. Test for anticancer activity on melanoma and colon malignant cells, demonstrated a reduced proliferation of cancerous cells for samples treated with angiostatin and plasminogen; interestingly highest inhibition was obtained for treatments from bovine sources. We believe that these results represent a starting point for future development of novel dairy products capable of cancer prevention.

**Key Words:** plasminogen, angiostatin, proteases

**M174** The effect of different inulin types on the formation of rennet-induced gels. A. Foo\*, A. R. Hill, and M. Corredig, *University of Guelph, Guelph, Ontario, Canada*.

The objective of this research was to determine the effect of the prebiotic fiber inulin on rennet induced gelation of skim milk, and to determine the potential for utilization of inulin in rennet cheese applications. Four chicory inulin types (CLR, ST, XL, and HPX) with different degree of polymerization (DP) were studied. Inulin was added to untreated milk at concentration up to 3% (w/v) and the rennet-induced gelation was followed using dynamic oscillatory rheology and confocal microscopy. Experiments were performed in triplicates and differences were determined statistically using 2-way ANOVA and Tukey ( $P < 0.05$ ). Gelation time, gel stiffness and strain at break changed depending on type and concentration of inulin. XL, and HPX (high DP molecules) significantly increased gelation time at 2 and 3% compared with the control sample. All inulin-fortified gels were stiffer and were less brittle than control milk gels. Rennet-induced gels containing 0.02% FITC were observed after 3h incubation at 30°C with confocal laser scanning microscope, and results showed that type and concentration of inulin affected the gel structure formation. Low DP inulins (CLR and ST) produced similar structures as control, while high DP inulins (XL and HPX) produced more branched structures. The degree of branching seemed to increase with concentration. Syneresis was tested after gel formation at 3h. CLR

and ST fortified gels had almost identical syneresis rates as control, while XL and HPX fortified gels showed slower syneresis rates, especially at 3% inulin concentration. A modified HPLC method using an evaporative light scattering detector was used to analyze the amount of inulin present in the whey. Majority of CLR and ST ( $\sim 90 \pm 2-4\%$ ) were loss into the whey, while XL and HPX retained  $\sim 50 \pm 2-4\%$  in gels. The results indicated that high DP inulins were more retained in the gel than low DP inulins, but they also affected the gelling properties of caseins and network formation.

**Key Words:** inulin, rennet curd, rheological properties

**M175** Impact of temperature and fat content on bleaching of liquid whey. M. A. D. Listiyani\*<sup>1</sup>, R. E. Campbell<sup>1</sup>, R. E. Miracle<sup>1</sup>, D. M. Barbano<sup>2</sup>, and M. A. Drake<sup>1</sup>, <sup>1</sup>North Carolina State University, Raleigh, <sup>2</sup>Cornell University, Ithaca, NY.

The use of whey protein as an ingredient in foods and beverages is increasing, and thus demands for colorless and mild tasting whey protein are also rising. Bleaching is commonly applied to fluid colored cheese whey to decrease color, and different temperatures and bleach concentrations are applied. The objectives of this study were to compare the effects of hot and cold bleaching and the point of bleaching (before and after fat separation) on bleaching efficacy and volatile components of liquid Cheddar whey. Liquid colored Cheddar whey was produced in triplicate and pasteurized. Part of the whey was collected (no separation, NSE) and the rest was subjected to fat separation (FSE). NSE and FSE whey were then subdivided and bleaching treatments (benzoyl peroxide (BPO) 50 or 100 mg/kg and hydrogen peroxide (HP) 250 or 500 mg/kg) at 68C for 30 min or 4C for 16 h were applied. Control NSE and FSE with no added bleach were also subjected to each time temperature combination. Volatile compounds from wheys were evaluated by gas chromatography mass spectrometry (GCMS) and norbixin (annatto) was extracted and quantified to compare bleaching efficacy. Proximate analysis, including total solids, protein, and fat content were also conducted. Liquid whey subjected to hot bleaching at both concentrations of HP or 100 mg/kg BPO had significantly higher lipid oxidation products (aldehydes) compared with unbleached wheys, 50 mg/kg BPO hot-bleached whey, or cold-bleached wheys. Fat separation had no impact on the relative abundance of volatile lipid oxidation products ( $P > 0.05$ ). Wheys bleached with BPO had lower norbixin recovery compared with wheys bleached with HP ( $P < 0.05$ ). HP bleaching efficacy was decreased at 4C compared with 68C ( $P < 0.05$ ), BPO bleaching efficacy was not impacted by temperature ( $P > 0.05$ ). These results suggest that fat separation has no impact on bleaching efficacy or lipid oxidation and that hot bleaching may result in increased lipid oxidation in fluid whey.

**Key Words:** whey, bleach, flavor

**M176** Bleaching liquid Cheddar whey using ultraviolet radiation. E. J. Kang\* and M. A. Drake, *North Carolina State University, Raleigh*.

Two of the desirable attributes of whey protein (WP) are a neutral color and bland flavor. The residual annatto colorant from Cheddar cheese production, therefore, has to be removed by bleaching. Currently, in the United States, hydrogen peroxide and benzoyl peroxide are the only bleaching agents approved to bleach colored whey. Recent studies have demonstrated that peroxide bleaching can negatively impact WP flavor.

Alternative bleaching methods may be valuable to the dairy industry. The objective of this study was to investigate the efficacy of UV radiation on bleaching of liquid Cheddar whey. Fluid colored Cheddar whey was manufactured on duplicate occasions. Following pasteurization and fat separation, whey was subjected to UV radiation for 30, 60, 90 or 120 min or cooled with no bleaching (control). The UV reactor consisted of a stainless steel feed tank (4 L), peristaltic pump, and a UV reactor. A 385 mm length low pressure mercury lamp was enclosed in a 24 mm diameter glass tube inside the stainless steel UV reactor. This UV reactor had a 151 mL working volume. Liquid whey circulated through the UV reactor, back into the feed tank and then re-circulated while the feed tank was held at 60°C. Wheys were then subjected to norbixin extraction and quantitation to measure bleaching efficacy. Control Cheddar whey with no UV treatment had 4.12 ppm of norbixin. Whey exposed to UV radiation for 30, 60, 90 or 120 min had 0.65ppm, 0.39ppm, 0.52ppm, and 0.37ppm norbixin, respectively. Norbixin decreased ( $P < 0.05$ ) compared with control whey when exposed to UV light for 30 min; but increasing time up to 2 h did not increase the bleaching effect ( $P > 0.05$ ). Overall norbixin concentration in fluid whey was reduced 84–91% when treated with UV radiation. UV radiation may be an alternative to peroxide bleaching of fluid whey.

**Key Words:** UV radiation, liquid whey, bleach

**M177** Development and analysis of a dairy-based nutrient dense gel food rich in milk bioactives. M. Cleveland\* and R. Jiménez-Flores, California Polytechnic State University, San Luis Obispo.

Milk products are a rich source of nutrients and bioactive compounds beneficial to human health. Some of these include immunoglobulins, phospholipids, casein and whey proteins and peptides, and components of the milk fat globule membrane (MFGM). These products although originally in milk, can be found in higher concentrations in other products, such as colostrum, buttermilk, and whey protein powders. Addition of ingredients such as probiotics add additional value and are apt to be easily incorporated into a dairy system due to their natural residence in milk. The application of dairy ingredients in product development has great potential to create a strong nutrition delivery system with benefits beyond those of fluid milk. Exploitation of dairy ingredients in this way is economical and promotes use of products that still have untapped potential in the marketplace. Development of dairy-based ready-to-eat foods is an excellent way to provide a rich source of the aforementioned milk bioactives. These products would serve as a compact, convenient source of energy and specialized nutrition. The health benefits include immune system development and function, stimulation of gastrointestinal function, and probiotic activity. We have created and biochemically analyzed a dairy-based high nutrient density gel food. It has a pleasing sensory profile, provides significant whey protein, and due to its ingredient profile is an excellent source of milk bioactives. The protein quality and quantity were designed to promote satiety and healthy muscle mass. We have analyzed the gel for immunoglobulin concentration (ELISA), phospholipid content (HPLC method), probiotic survival and binding preference to phospholipids, and basic macronutrient content (FTIR). The results of these assessments show a significantly higher level of milk bioactivity in the gel than in an equivalent gram quantity of fluid milk. From our studies, we conclude that we have created a highly valuable, convenient dairy-based nutrition delivery system with potentially great health benefits. Future work includes design of large-scale manufacture and studies of bioactivity in cell culture.

**Key Words:** milk bioactives, nutrition, dairy foods

**M178** Identification of bioactive peptides derived from fermentation of organic milk. S. R. Pritchard\*, M. Phillips, and K. Kailasapathy, University of Western Sydney, East Richmond, New South Wales, Australia.

The aim was to identify peptides isolated from bacterial fermentations of organic milk that may have antimicrobial, antihypertensive and antioxidant properties. Organic milk was fermented in duplicate with *Lactobacillus acidophilus* LAFTI L10, *Lactobacillus casei* 2603, *Lactobacillus rhamnosus* 2625 (2, 6 and 9 d) and *Lactobacillus helveticus* (10% v/v) (2, 4 and 6 h). The pH was adjusted to 4.6 and soluble and insoluble peptide fractions were extracted by centrifugation followed by filtration. The extracts were screened for the presence of antimicrobial, antihypertensive and antioxidant activity. All analysis was carried out in triplicate. Antioxidant activity was measured by the inhibition of the free radical 1, 1-diphenyl-2-picrylhydrazyl (DPPH). The soluble fractions had the greatest inhibition of DPPH compared with the insoluble fractions. The peptides extract that inhibited DPPH the greatest were derived from organic milk fermented by *L. rhamnosus* for 6 d ( $42.83\% \pm 1.26$  standard deviation (SD)), followed by the peptide extracts derived from milk fermented by *L. helveticus* for 2 h ( $34\% \pm 3.45$ ) and *L. helveticus* for 4 h ( $32\% \pm 0.45$ ). Antimicrobial peptides were determined by inhibition of 3 bacteria namely *Escherichia coli* ATCC 8739, *Bacillus cereus* ATCC 11778 and *Staphylococcus aureus* ATCC 6538. Overall, *E. coli* was inhibited the greatest by soluble peptide fractions including the peptide extracts derived from organic milk fermented by *L. acidophilus* for 9 d ( $108.38\% \pm 8.85$  SD), with *L. rhamnosus* for 6 d ( $101.13\% \pm 1.95$ ) and with *L. casei* for 9 d ( $90.04\% \pm 3.52$ ). Antihypertensive activity was determined by the inhibition of the angiotensin-converting enzyme (ACE) using RP-HPLC to detect the amount of hippuric acid produced. Overall, the soluble fractions had the lowest IC50 values. The peptide extract derived from organic milk fermented for 9 d by *L. casei* had the lowest IC50 value ( $0.124\text{mg/ml} \pm 0.008$  SD). This research has shown that peptide extracts derived from fermented organic milk have antimicrobial, antihypertensive and antioxidant properties.

**Key Words:** bioactive peptide, milk, fermentation

**M179** Increasing stringiness of low fat mozzarella cheese using polysaccharides. E. N. Oberg\*<sup>1</sup>, K. M. Larsen<sup>1</sup>, D. A. Irish<sup>1</sup>, M. M. Motawee<sup>2,1</sup>, and D. J. McMahon<sup>1</sup>, <sup>1</sup>Western Dairy Center, Utah State University, Logan, <sup>2</sup>National Organization for Drug Control and Research, Cairo, Egypt.

Removing fat from mozzarella cheese decreases fiber formation because protein strands fuse together during stretching and extruding. This study examined the ability of polysaccharides to act as fat mimetics and aid in fiber formation in manufacture of string cheese. Low fat mozzarella cheese curd made from 273 kg of 0.7%-fat milk was salted at a rate of 10 g/kg then divided into 2.7-kg batches that were hand-stretched in 5% (wt/wt) hot brine (80°C) and formed into a homogeneous mass. The hot cheese was hand mixed with a hot (80°C) polysaccharide slurry and then placed into a small piston-driven extruder and cheese forced through a 16-mm die to form the string cheese and cut manually into about 15-cm lengths. Cheeses were analyzed for fat, salt, pH, and moisture. After 1 wk of storage at 4°C, extent of stringiness was measured by pulling apart the cheese longitudinally, visually observing and photographing size and appearance of individual strings of cheese, and measuring their length. In one trial, 3 starches (waxy corn, waxy rice, and instant tapioca starches) were tested using 8% (wt/vol) slurries at a ratio 7% (wt/wt) of stretched curd. Waxy rice and waxy corn starches did not mix well with hot cheese and in a second trial, xanthan gum and guar gum slurries were added at a ratio of 2% (wt/wt), and polydextrose and instant

tapioca starch added at 7%. In the control low fat cheese there was no apparent fiber formation and the cheese was homogeneous in texture. All cheeses made with added polysaccharides had substantial fiber formation and were more similar to regular commercial string cheese. Low fat cheese with added xanthan gum had the most pronounced fiber formation with the longest strings (i.e., the full length of the cheese stick) and best string separation. Adding polydextrose produced cheese with the least string formation and was only slightly better than the control. Overall, extent of stringiness was greatest immediately after extrusion and tended to decrease during storage with string diameter increasing and string length decreasing.

**Key Words:** mozzarella, string cheese, low fat

**M180** Enrichment of low fat Cheddar cheese with dietary fiber. R. Wadhvani\*, D. J. McMahon, and D. A. Irish, *Utah State University, Logan*.

Dietary fiber intake of 20 to 35 g/d is recommended for lowering coronary heart disease, cancer, and other health benefits such as satiety and improved digestion. However, the average American daily intake is only 12 to 18 g/d. Fiber intake can be increased by enriching more foods and this study examines the feasibility of enriching low fat Cheddar cheese (6% fat) with dietary fibers and their effect on cheese organoleptic acceptability. Low fat Cheddar was made and stored for 15 d, then comminuted to 1.5 mm particle size. Then, inulin, pectin, polydextrose, or resistant starch were mixed into 1.82 kg batches of comminuted cheese at 50 g/kg with or without addition of 50 g/kg of water. The comminuted batches were then repressed individually in cheese molds, vacuum packaged and stored at 4°C. All cheese samples were analyzed at 90 d for composition and fiber was calculated by difference. Texture profile analysis was performed at 90 and 180 d of storage. Sensory flavor analysis was performed at 180 d with a full fat Cheddar cheese comparison. Chewiness was evaluated by counting the number of bites before swallowing cheese. No liquid expulsion from repressed cheese mixed with fiber was observed which resulted in 100% retention of fiber in cheese except for polydextrose which experienced liquid expulsion of < 0.5%. When added with water, cheese mixed with inulin or pectin resulted in better knitting and uniform mixing of cheese particulates which was confirmed by increased cohesiveness from 0.48 to 0.65 for inulin with water and 0.50 for pectin with water. Hardness for inulin (46.41N) and pectin (55.87N) cheeses were significantly lower than non-repressed control cheeses (80.94N) and chewiness was also significantly reduced from 44.18N to 12.45N. Polydextrose and resistant starch cheese were poor in appeal and lacked smooth texture. Chewdown method showed that full fat cheese and comminuted cheeses required 12 bites before swallow whereas non-comminuted cheese control was reported 24 bites by trained panel which is clearly double the number. This study indicated a feasibility to enrich comminuted low fat cheese with fiber contributing to better texture and no impact on overall flavor.

**Key Words:** fiber, Cheddar, texture

**M181** Development of a rapid method for determination of lactose in process cheese using blood glucose meter. A. C. Biswas\*, J. Amamcharla, and L. E. Metzger, *Midwest Dairy Foods Research Center, Dairy Science Department, South Dakota State University, Brookings*.

The lactose content can influence the functional and textural attributes of process cheese (PC). Hence, the development of a rapid and accurate method for determination of lactose in PC will be beneficial to the industry. Previously, a blood glucose meter method (BGMM) was developed for the determination of lactose in model solutions and raw

milk. The current objective was to modify the BGMM for the determination of lactose in PC. The previous method was modified by utilizing a phosphate buffer to dilute the sample and used a new generation of glucose meter for analysis. For sample preparation, 1 g of PC was added to 20 g of 0.01 M phosphate buffer (pH 7.4) and mixed vigorously at 60°C. Five milliliters of the mixture was transferred to a test tube, and 0.01 mL of  $\beta$ -galactosidase was added. The solution was incubated at 40°C for 10 min to hydrolyze the lactose into glucose and galactose, and then analyzed for glucose in duplicate using the ReliOn Confirm glucose meter and its compatible test strips. To evaluate the variation between different test strip lots, 4 lots of test strips were utilized. For all 4 lots of test strips an individual calibration curve was developed using PC with known lactose concentrations between 3.2 and 8.2%. A universal calibration curve was also computed by pooling the data from all 4 lots of test strips. A sample of the original dilution of PC and buffer was also analyzed for lactose using an HPLC based method. The new rapid method was validated using 10 PC. The slopes and intercepts of individual calibration curves ranged from 1.23 to 1.37 and from -105 to -60, respectively. The slope and intercept of the universal curve were 1.32 and 88, respectively. The mean absolute bias was found to be between 0.11 - 0.32% for individual calibration and 0.15 - 0.24% for the universal calibration curve. The observed moderately high bias could be caused by variability in the test strip lots and intrinsic components of PC. Overall the novel rapid method is promising. However, modifications in sample preparation and calibration need to be developed to improve the method.

**Key Words:** lactose, glucose meter, process cheese

**M182** Monitoring changes in the non-casein nitrogen fraction of raw milk during storage using casein/fat standardizer (CFS). P. Salunke\*, J. Amamcharla, and L. E. Metzger, *Midwest Dairy Foods Research Center, South Dakota State University, Brookings*.

Prolonged storage at refrigerated temperatures causes changes in the nitrogen distribution of raw and pasteurized milk. An increase in the non-casein nitrogen (NCN) fraction during storage is the result of psychrotrophic microorganisms and proteolytic enzymes. This phenomenon has direct implications on both cheese yield and quality. The Casein-Fat-Standardizer (CFS; Tetra Pak CPS, MN and Metron Instruments, OH) is being used as an on-line tool for standardizing the casein and fat of cheese milk. CFS uses a patented Brewster analysis method of laser light scattering to measure the casein and fat in the cheese milk. The objective of this study was to determine if the CFS can be used to follow the increase in the NCN fraction of cheese milk during refrigerated storage. Four raw milk samples (1 L) were collected immediately after milking. Each of the 4 samples was divided into 6 sub-samples and stored at 4°C. On each experimental day (for 6 consecutive days), one of the 6 sub-samples was withdrawn and tempered to 40°C. The sample was analyzed for fat, total protein (TP) and NCN in triplicate using the respective standard methods. The sample was also analyzed using the CFS to measure the casein and fat. It was found that there was no significant ( $P > 0.05$ ) effect of storage time on the TP and fat of raw milk as analyzed by the standard methods. However, the NCN fraction of milks significantly ( $P < 0.05$ ) increased during the storage period leading to a significant ( $P > 0.05$ ) decrease in casein content after 2 d of storage. Similarly, the casein content analyzed by the CFS also showed significant ( $P > 0.05$ ) differences after 3 d of refrigerated storage. The fat content measured in CFS also showed a significant ( $P > 0.05$ ) decrease from d 0 to d 1, but showed no significant ( $P < 0.05$ ) difference between the remaining storage period. Paired *t*-test comparison between the standard methods and CFS measured fat and casein showed

no significant differences for casein ( $P = 0.76$ ) and fat ( $P = 0.72$ ). These results indicate that the CFS instrument is capable of monitoring changes in the NCN content of milk during refrigerated storage.

**Key Words:** NCN, raw milk storage, CFS

**M183** Impact of xylitol on the functional properties of low fat process cheese. A Kommineni\*, J Amamcharla, and L. E. Metzger, *Midwest Dairy Foods Research Center, South Dakota State University*.

Process cheese is a dairy food made by blending natural cheese, salt, emulsifying salts and other dairy and non dairy ingredients, and heating with continuous agitation to produce a homogeneous product. Due to increased health concerns the demand for low fat products has increased. Fat is a critical component of process cheese and plays an important role in its functional characteristics. Reducing or lowering the fat content of process cheese results in poor functional properties. The objective of the current study was to evaluate the effect of xylitol on the functional properties of low fat PC. Three different low fat PC formulations were made with 0%, 2% and 4% xylitol. All PC formulations contained 5% fat and 55% moisture and each treatment was manufactured in triplicate. Rheological characteristics including elastic modulus ( $G'$ ), Viscous modulus ( $G''$ ) and Temperature at  $\tan\delta = 1$  were determined using Dynamic Stress rheometry (DSR). The DSR was carried out at 1.5Hz frequency and 400 Pa stress levels using a temperature sweep from 200C to 900C. The hardness of the samples was also determined with texture profile analysis (TPA). Compositional analysis indicated that all treatments had a similar fat, protein, and moisture content. The elastic ( $G'$ ) and viscous ( $G''$ ) modulus results obtained with DSR decreased with increasing xylitol addition. The meltability index temperature was not significantly ( $P > 0.05$ ) different between all treatments. TPA analysis demonstrated that xylitol addition significantly ( $P < 0.05$ ) decreased the hardness of low fat process cheese. Based on the results obtained with DSR and TPA this study has demonstrated that xylitol addition improves the functional properties of low fat PC.

**Key Words:** process cheese, low fat, xylitol

**M184** Application of salt whey in process cheese food made from young Cheddar cheese containing exopolysaccharides. O. Janevski\*, A. N. Hassan, and L. Metzger, *South Dakota State University, Brookings*.

The objective of this study was to utilize salt whey in making process cheese food (PCF) from young (3-week-old) Cheddar cheese. To maximize the level of salt whey in process cheese, low salt (0.6%) Cheddar cheese was utilized. Since salt reduction causes undesirable physico-chemical changes during extended cheese ripening, young Cheddar cheese was used in making process cheese. An exopolysaccharides (EPS) producing culture (JFR) was used to reduce rigidity and improve meltability of young Cheddar cheese. A non-EPS-producing culture (DVS) was applied in making the control cheese. To obtain similar composition in the EPS-positive and negative Cheddar cheeses, the making protocol was modified in the later cheese to increase its moisture level. The composition of Cheddar cheese was determined and used to formulate the corresponding PCF (JFR-PCF and DVS-PCF). Three-week-old Cheddar cheese containing 41.4% moisture, 31% fat, and 21.2% protein, was shredded, and stored frozen until used for PCF manufacture. No differences in pH and level of water-soluble nitrogen were seen between the EPS-positive and negative Cheddar cheeses. The utilization of low salt Cheddar cheese allowed up to 13% of salt whey containing 9.1% salt to be used in process cheese making. The preblend was mixed in the Rapid Visco Analyzer (RVA) at 1000 rpm, heated at 95°C for 3

min, and process cheese was transferred into copper cylinders, sealed and kept at 4°C. Process cheese contained 43.28% moisture, 23.7% fat, 18.9% protein, and 2.1% salt. No difference in composition was seen between the JFR-PCF and DVS-PCF ( $P > 0.05$ ). The texture profile analysis showed that JFR-PCF was softer, and less gummy and chewy ( $P \leq 0.05$ ) than DVS-PCF. The end apparent viscosity and meltability in JFR-PCF were higher ( $P \leq 0.05$ ) than those in DVS-PCF, whereas the emulsification time was shorter ( $P \leq 0.05$ ) in the former cheese. In conclusion, process cheese, containing up to 13% salt whey, with improved textural and melting properties could be made from young EPS-positive Cheddar cheese.

**Key Words:** process cheese, salt whey, exopolysaccharides producing cultures

**M185** Prediction of water activity of natural cheese using a model cheese system. J. Grummer\* and T. C. Schoenfuss, *University of Minnesota, St. Paul*.

The ability to predict the water activity of natural cheese in modified formulas is critical in developing products such as reduced or low sodium cheese. In published attempts to develop reduced sodium cheese, replacement salts have been added at levels too low to create the equivalent microbial and enzymatic stability of full sodium controls. The objective of this study was to develop a model system that could be used to measure  $A_w$  directly to verify samples formulated using predictive equations. Flour, butter and water were blended at levels that represented typical solids, fat, and water levels in Cheddar cheese. Predictive equations were used to calculate the levels of the various salt replacers needed to achieve the same water activity as samples with 600 mg sodium/100g. Sodium chloride and salt replacers (potassium chloride, modified potassium chloride (Nu-tek Products, Minnetonka, MN), magnesium chloride hexahydrate, and calcium chloride dehydrate) were added to the model blends to produce cheese with less than 300 mg sodium/100g of cheese. The measured water activities did not match the predicted values in some cases. This could have been due to differences in purity and moisture levels between the salts or the solubility of the salts at the percent moisture tested. The discrepancy demonstrated the benefit of using this model system for direct testing of water activity in a model system before producing batches of cheese.

**Key Words:** water activity, prediction, cheese

**M186** Optimization of a CO<sub>2</sub> injection method for increasing the permeate flux in cold microfiltration of skim milk. T. J. Tan\*, A. Sauer, and C. I. Moraru, *Cornell University, Ithaca, NY*.

Cold microfiltration (MF) was proven effective for microbial removal from skim milk. One of the challenges associated with MF is the low permeate flux, caused by fouling. To address this, a CO<sub>2</sub> injection technique capable to counteract membrane fouling was developed and optimized. The experimental MF unit consisted of a feed tank connected to a variable-speed centrifugal pump, a tubular heat exchanger and a tubular ceramic membrane of Tami design, placed inside a stainless steel housing. The membrane had a nominal pore size of 1.4  $\mu\text{m}$  and total membrane area of 0.35 m<sup>2</sup>. Three portable CO<sub>2</sub> injection ports were attached to the membrane housing. The combinations tested included: no CO<sub>2</sub> injection (control), 1 port, and 3 ports. Four combinations of CO<sub>2</sub> injection frequencies and durations were tested: no injection (control), 60s frequency with 2s duration (60s/2s), 60s frequency with 1s duration (60s/1s), and 120s frequency with 1s duration (120s/1s). The MF experiments were performed at a temperature of 6°C, a cross-flow velocity of 7 m/s and a transmembrane pressure of 83 kPa. A data acquisition

port was used to collect the temperature, pressure and permeate weight data; the latter was then used to calculate the permeate flux. In all runs, permeate flux showed an initial rapid decrease followed by a long and gradual decline. A series of 45min experiments were first performed. At similar water flux values of the membrane ( $1500 \pm 50 \text{ L/m}^2\text{h}$ ), the combination of 3 ports with 120s/1s produced the highest flux ( $46.09 \text{ L/m}^2\text{h}$  at 45min) followed by 3 ports with 60s/1s ( $41.32 \text{ L/m}^2\text{h}$ ) and 3 ports with 60s/2s ( $39.41 \text{ L/m}^2\text{h}$ ). Three hour experiments were then conducted: without injection (control), and with optimal  $\text{CO}_2$  injection (3 ports, 120s/1s). After 3 h, the flux for the control was  $26.36 \pm 3.38 \text{ L/m}^2\text{h}$ , while for the optimized  $\text{CO}_2$  injection experiment, a flux of  $33.65 \pm 3.68 \text{ L/m}^2\text{h}$  was obtained. In addition, a smaller drop in flux was observed for the  $\text{CO}_2$  injection experiment (23% after 3h) as compared with the control (31.5% after 3h). The developed method is effective and can be used to increase the flux in microfiltration applications.

**Key Words:** microfiltration, fouling,  $\text{CO}_2$  injection

**M187 Polysaccharide addition to lowfat Cheddar cheese to improve texture.** R. Kumar\* and T. C. Schoenfuss, *University of Minnesota, St. Paul.*

Fat plays a role in cheese texture by acting as a plasticizer and inhibiting cross-linking between protein chains. Reductions in fat to produce lowfat cheese (less than 3 g fat/50 g cheese) results in texture defects. The objective of this study was to improve the texture of lowfat cheese by incorporating a polysaccharide gel as a filler to disrupt protein cross-links. Polysaccharides examined were alginate, xanthan, pectin, carageenan (Danisco USA Inc., New Century, KS) and Novagel RCN 15 (FMC Biopolymer, Philadelphia, PA). The level of polysaccharide needed to produce a soft gel was mixed with whey protein concentrate (Avonlac 180, Glanbia Nutritionals, Monroe, WI) on a high shear mixer. The gel (calculated at 10% of the pressed cheese if all was retained) was homogenized (Niro panda, GEA, Hudson, WI) at 160 bar with all the cream and a portion of the skim milk. It was then blended with the remaining skim. Control lowfat cheese was made with homogenized cream without polysaccharides. The cheesemaking procedure used a stirred curd method with pre-acidification to pH 6.2, and a final curd pH of 5.7 at salting. Curds were pressed in 0.7 kg Wilson-style hoops overnight. Cheeses were evaluated at 2 mo of age by sensory and texture profile analysis. Untrained panelists were instructed to place cheese samples (treatments and controls presented blindly) spatially on a  $61 \times 61$  cm sheet of paper based on differences they perceived in texture and flavor. The distance of each sample from a full-fat sample was measured in cm. Novagel and pectin treatments were placed significantly closer ( $P < 0.05$ ) to the full fat cheese than low fat control. Samples measured on a TA.XT-Plus Texture Analyzer (Texture Technologies, Scarsdale, NY) showed pectin and carageenan samples were significantly lower in percent energy recovered ( $P < 0.05$ ) than lowfat control. Moistures of the lowfat samples were not significantly different, (52 to 54%). It was concluded that pectin, carageenan and Novagel had a positive effect on cheese texture using this method.

**Key Words:** lowfat, Cheddar, polysaccharides

**M188 Effect of concentration and temperature on the rheological properties of 95% serum protein (SP) reduced micellar casein concentrates (MCC).** A. Sauer\*, C. Beliciu, and C. I. Moraru, *Cornell University, Ithaca, NY.*

The use of casein preparations obtained by membrane separation is receiving increasing interest from the dairy industry, as well as other industries. Currently, there is a lack of knowledge regarding the flow

behavior of these protein concentrates under various processing conditions such as temperature and shear. This work focused on evaluating the rheological properties of micellar casein and on understanding how they are affected by concentration, temperature and shear. MCC with 95% SP reduced were obtained from skim milk by microfiltration followed by spray drying. MCC preparations of concentrations ranging from 5% to 12.5% were obtained by dispersing the MCC powders in deionized water, under vigorous stirring. Large amplitude and small amplitude rheological analyses were performed to evaluate the viscosity and flow behavior, as well as the network structure of these protein preparations. Steady shear experiments at temperature ranging from 0C to 80C were performed using an ARES strain controlled rheometer (TA Instruments). The viscosity vs. shear rate curves were used to evaluate the effect of shear on viscosity, and apparent viscosity at a shear rate of 100s<sup>-1</sup> was used to make direct comparison between various concentrations and temperature conditions. All protein preparations displayed a shear thinning behavior, which was more pronounced as casein concentration increased. The apparent viscosity of MCC increased exponentially with casein concentration and decreased with temperature. The dependency of apparent viscosity on temperature followed an Arrhenius relationship. The activation energy for viscous flow ( $E_a$ ) in the Arrhenius relationship increased with concentration.  $E_a$  values of 21280 kJ/mol, 26730 kJ/mol and 40167 kJ/mol were obtained for MCC concentrations of 5%, 7.5% and 10%, respectively. The relationships developed will allow the prediction of rheological properties under desired temperature and concentration conditions, which will provide the dairy and food industry with critical rheological data necessary for developing applications of micellar casein preparations.

**Key Words:** micellar casein, rheology, viscosity

**M189 Formation of bacterial biofilms on spiral wound reverse osmosis whey concentration membranes and its influence on retentate quality.** M. Avadhanula\*, A. C. Biswas, S. Anand, and A. Hassan, *Midwest Dairy Foods Research Center, Dairy Science Department, South Dakota State University, Brookings.*

Fouling is a major operational hurdle in the membrane processing of whey. In addition to the deposition of organic matter, bacterial biofilm formation on membranes may play a role in their reduced performance. The present investigation was conducted to study the formation of bacterial biofilms on spiral wound, reverse osmosis (RO) whey concentration membranes using the standards plate counting technique. Membrane cartridges from a commercial whey RO system were drawn at intervals of 2 mo up to a total duration of 12 mo to evaluate the effect of membrane aging on biofilm formation. Swab samples were taken from  $1 \times 1 \text{ cm}^2$  membrane pieces drawn randomly from each cartridge. Counts of different bacterial groups were monitored in the feed, retentate and on the surface of membranes using selective media. The results confirmed the presence of multispecies bacterial biofilms on the whey RO membranes. Considerable variations were noticed in the distribution pattern of bacterial constituents in biofilms as the membranes aged. The average colony forming units (cfu/cm<sup>2</sup>) for mesophiles, coliforms, lactic acid bacteria, staphylococci, and  $\beta$ -hemolytic colonies were 1.16, Nil, 0.78, 0.72, and 1.26 on the 2 mo old membrane, 4.11, 3.15, 4.05, 1.4, and 3.0 on the 6 mo old membrane, and 3.86, 2.38, 4.04, 1.91 and 2.86 on the 12 mo old membrane, respectively. The average log count (cfu/ml) in the feed whey was 5.44, which increased to 7.67 at the end of a 24 h cycle. In comparison to that, the average retentate count (cfu/ml) increased from log 5.30 to log 7.93. The higher increase in the retentate counts than that in the feed may be due to contamination from the membrane biofilms. This study provides a qualitative analysis of bacterial constituents of the

biofilm consortia obtained from whey concentration membranes from active industrial processes and its influence on retentate quality.

**Key Words:** whey retentate, reverse osmosis membrane, multispecies biofilm

**M190** Thermal aggregation of whey proteins in the presence of buttermilk. M. Saffon\*<sup>1</sup>, M. Britten<sup>2</sup>, and Y. Pouliot<sup>1</sup>, <sup>1</sup>*STELA Dairy Research Center, Institute of Nutraceuticals and Functional Food (INAF), Université Laval, Québec, QC, Canada,* <sup>2</sup>*Food Research and Development Center (FRDC), Agriculture and Agri-Food Canada, St-Hyacinthe, Québec, Canada.*

Reincorporating heat-denatured whey proteins in cheese milk is widely used in the cheese industry. The use of buttermilk is however limited because it increases cheese moisture. It was hypothesized that a co-denaturation process involving a mixture of whey and buttermilk proteins increases the use of buttermilk in cheesemaking. In preliminary experiments, mixed dispersions were prepared from powdered concentrates acidified to pH: 4.6; 5.4 and 6.2 using HCl 1N and heated at 2 temperatures (80°C and 90°C). From these trials, heating the mixed dispersions to 90°C and pH 4.6 showed the best results and this condition was chosen to test different ratios of whey to buttermilk protein. Cheese whey and buttermilk were concentrated by ultrafiltration up to

9.5% (w/v) protein content. Mixtures were heated to 90°C for 25min (including come-up time). After cooling, samples were homogenized 5 times at 9500 psi. Aggregated material was separated by centrifugation at 15000g for 20 min. Protein aggregation was calculated from protein content in the supernatant and water holding capacity (WHC) was determined on the pellet. Heat denaturation was also applied to protein mixtures containing thiol blocker (N-ethylmaleimide) to understand the impact of disulfide bonds on the formation of aggregates. All the heating experiments were repeated 3 times, statistical analysis of the data was performed using ANOVA and the results were considered significantly when  $P < 0.05$ . Increasing temperature significantly increased protein aggregation from 58% to 75% when heating temperature was raised from 80°C to 90°C. Decreasing heating pH significantly decrease WHC. Minimum WHC being observed at pH 4.6. Increasing the fraction of buttermilk protein in the mixture increased significantly protein aggregation and reduced WHC up to a ratio of 25:75 (78% and 1.80 g water/g protein). The use of NEM significantly increased protein aggregation by 6.2% and decreased WHC by 0.73 g water/g protein. Overall, our results show that increasing buttermilk fraction resulted in higher protein aggregation and lower WHC and suggest that disulfide bonds are formed in the early stages of aggregation.

**Key Words:** cheese whey, buttermilk, aggregation