

Dairy Foods: Dairy Foods/Cheese

M57 Relationship between base and process cheese characteristics.

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The objective of this research was to study the relationship between properties of base and process cheese food. Full fat Cheddar cheese was made with exopolysaccharides (EPS)-producing or nonproducing cultures. Process cheese food was made from 2 day or 1 month old EPS-positive and negative base Cheddar cheese. The EPS-positive base Cheddar cheese contained 2% higher moisture and 1.5% lower fat than the EPS-negative one. Process cheese food contained about 44% moisture, 23% fat, 2% salt, and 21% protein. There were no differences ($P > 0.05$) in composition of process cheese made from EPS-positive and negative base Cheddar cheese. Texture profile analysis, meltability, and viscoelastic properties were evaluated in both base and process cheeses. The 2 day and 1 month old EPS-positive base Cheddar cheeses were softer, and less gummy and chewy ($P < 0.05$) than the corresponding EPS-negative cheeses. No differences ($P > 0.05$) were observed in the texture profile analysis of process cheese food made from 2 day old EPS-negative and positive Cheddar cheeses. However, process cheese food made from 1 month old EPS-negative Cheddar was harder, and more gummy and chewy ($P < 0.05$) than that made from the 1 month old EPS-positive base Cheddar cheese. This is due to less extensive proteolysis in the former base cheese. Although lower viscoelastic moduli were seen in the 2 day old EPS-positive than in the EPS-negative base Cheddar cheese, no differences ($P > 0.05$) were found in process cheese food made from these base cheeses. Interestingly, whereas the viscoelastic moduli in the base cheese were not affected by age (2 day or 1 month), they were higher ($P < 0.05$) in process cheese food made from base cheese aged for 1 month. Also, aging of base cheese for 1 month improved softening and melting properties. However, such improvements were not seen in the corresponding process cheese food. Modifications in the base cheese texture do not necessarily reflect on process cheese characteristics. Proteolysis of base cheese is a major factor affecting process cheese physical properties.

Key Words: process cheese food, base cheese, texture

M58 Fate of aflatoxin M₁ during manufacture and brining of feta cheese.

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Aflatoxins are toxic carcinogenic compounds that can be present in milk if there is growth of *Aspergillus* on feedstuffs consumed by livestock. Because of health risks from consuming large amounts of aflatoxins, maximum amounts allowable in milk are 0.50 µg/L in the United States and 0.05 µg/L in the European Union. Our objective was to determine the fate of aflatoxin M₁ (AFM₁) in milk after pasteurization followed by manufacture of feta cheese and its storage in brine, and whether this would reduce the level of dietary exposure of consumers to AFM₁. Feta cheese was made from 4 kg of milk spiked with 1 and 2 µg AFM₁ per kg and AFM₁ contents were studied using an enzyme immunoassay technique. Milk pasteurization at 63°C for 30 min caused minimal (<10%) destruction of AFM₁. The remaining AFM₁ in the pasteurized milk was partitioned between curd and whey during cheesemaking. The yield of curd was 0.22 kg per kg of milk, and AFM₁ levels in the curd were 2.95 and 6.00 µg/kg for cheese made from the 1- and 2-µg/kg AFM₁ milks, respectively. The AFM₁ levels in the corresponding wheys were 0.37 and 0.69 µg/kg. Thus, the curd contained only 64% of the

original AFM₁ added to the raw milk. Cheeses containing 2.95 µg/kg AFM₁ were placed in 8, 10 and 12% brine solutions for 60 d and stored at 6 and 18°C. There was a 22 to 27% reduction of AFM₁ during the first 10 d of storage, with slightly more loss as salt concentration increased and when the cheese was stored at 18°C. Further storage caused only slight decrease in AFM₁ and after 60 d of brining the cheeses contained 2.2 and 2.1 µg/kg AFM₁ for cheese brined at 6 and 18°C, respectively, for a loss of 25 and 29% of AFM₁ from the curd into the brine. Through the conversion of milk into feta cheese (brined for at least 50 d) there was a loss of about 50% of the AFM₁ originally present in the raw milk. Thus, for consumers in regions with high aflatoxin levels in milk, health risk could be reduced by converting the milk into a brined cheese. A 30-g serving of such cheese would contain only 0.07 µg aflatoxin compared to the 0.25 µg aflatoxin from a 250-g serving of such milk.

Key Words: aflatoxin, milk, cheese

M59 The ELISA test to determinate the κ-casein B contents in bulk milk samples: Practical use.

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κ-Casein is one of the most important part of milk casein involved in the cheese making. The κ-casein family consists of one major carbohydrate-free component and several "minor" glycosylated ones, with the same amino acid sequence but with different nature and number of the carbohydrate groups. The most frequent variants in dairy milk are A and B, which differ for two amino acids, one in position 136 and one in 148. Substantial differences in cheese making properties were found due to better coagulation properties; more cheese was obtained from milk samples with B variant compared with the same quantity of milk with A variant. The "test kappa" is a commercial ELISA test useful to quantify κ-casein B content in bulk milk samples. Using this test is possible to follow the yearly farm's variation of milk κ-casein B content. Four different ratio of milk dilution were tested: 1:1000, 1:1500, 1:2000 and 1:3000. The best results were obtained with the lowest dilution, due to the better matching of samples intensity color with the calibration curve.

Key Words: κ-casein, milk quality, cheese

M60 Aroma profile characterization of traditional Algerian Bouhezza cheese.

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Bouhezza cheese is an Algerian traditional cheese from goat, sheep or cow milk. It can be also produced from mixed milks in different proportions. The manufacturing of Bouhezza cheese requires the preparation of a natural container "chekoua" and a spontaneous fermented milk, rather skimmed and little acid "lben". The chekoua is a bag prepared from goat or sheep skin, treated with salt and juniper berry. It has also an important role as separator of serum and solid phase (ultrafilter). The manufacturing process is carried out by addition of "lben" and raw milk, to correct the acidity and salty. The cheese making is completed after 70 days, on the average, when pepper spice is added. The aim of this preliminary study was to characterize the volatile organic compounds (VOCs) of

Bouhezza cheese produced in different farms. Five Bouhezza samples (F1-F5) were analyzed by SMart Nose (SN) system which allowed the direct analysis by MS of VOCs. The ion fragments obtained by SN were then processed by a multivariate statistical PCA). Dynamic headspace purge and trap apparatus (Tekmar 8900) in combination with a gas chromatograph olfactometry/mass selective detector to analyze the odour active compounds. PCA applied to SN results showed a good separation (PC1 77,93%; PC2 13,05%) among the Bouhezza cheeses indicating the uniqueness of the homemade products. Olfactometry results confirmed high variability for the aroma profile. Cheese F1 had the richest profile (23 VOCs), followed by F4 (19 VOCs), F2 (18 VOCs), F3 (15 VOCs) and F5 (13 VOCs). All cheese samples were mainly characterized by aldehyde and ester compounds. Among aldehydes there were octanal, nonanal, (E,E)-2,4-decadienal, (E)-2-nonenal that gave, respectively, orange, apple, hay, and green notes. Free fatty esters detected were: ethyl octanoate, ethyl hexanoate, ethyl butanoate and propyl-1-methylacetate that gave, respectively, wine, floral, and fruity notes. The high variability of Bouhezza cheese was clearly detected by SN and GCO analyses confirming that the farm traditional cheese-making process, the milk and the different spices used during manufacturing contribute to the uniqueness of this product.

Key Words: Bouhezza cheese, SMart Nose, GCO

M61 Molecular characterization of Algerian cheese Bouhezza by PCR-TTGE. C. Pediliggieri¹, S. Carpino^{*1}, and G. Licitra^{1,2}, ¹CoRFiLaC, Regione Siciliana, Ragusa, Italy, ²D.A.C.P.A. University of Catania, Italy.

Bouhezza cheese is an Algerian traditional cheese from goat, sheep or cow milk. It can be also produced from mixed milks in different proportions. The manufacturing of Bouhezza cheese requires at first the preparation of a natural container “*chekoua*” and a spontaneous fermented milk rather skimmed and little acid “*lben*”. The *chekoua* is a kind of bag prepared from goat or sheep skin, treated mainly with salt and juniper berry. It has also an important role as separator of serum and solid phase (ultrafilter). The end of the manufacturing is characterized by the addition of whole raw milk, what allows to correct the acidity and salty of the cheese. The manufacturing process is carried out by addition of “*lben*” and raw milk and it is completed after 70 days, on the average, when pepper spice is added. The bacterial ecosystem of Bouhezza cheese was explored by PCR-temporal temperature gel electrophoresis (PCR-TTGE). PCR-TTGE is a rapid molecular method based on direct analysis of DNA in the environment and on the separation of DNA molecules that differ by singles based allowing to analyse diversity within bacterial communities. Molecular fingerprinting was carried out on cheese samples collected at different time during two experimental cheese-making: cheese at 7 days, at 15d, 21d, 41d, 56d, and at 70 d with and without pepper. By the use of universal primers, PCR-TTGE revealed the predominance in all samples of *L. lactis*, which presence was confirmed by specific PCR tests. Furthermore the presence of *Lb. plantarum*, *Lb. haelveticus* and *Lb. delbruekii* was detected. More bands (correspondent to *S. equorum*, *Lb. delbruekii* , and a band maybe identifiable with *S. thermophilus*), were detected in PCR-TTGE profiles of cheeses after 41 days probably due to addition of whole raw milk. The presence of a few high-GC-content species, like *coryneform bacteria*, were observed too.

Key Words: Bouhezza cheese, PCR-TTGE, lactic bacteria

M62 Characterization of bacterial ecosystem in Pecorino Siciliano cheese produced in different areas of Sicily. C. Pediliggieri¹, S. Carpino^{*1}, and G. Licitra^{1,2}, ¹CoRFiLaC, Regione Siciliana, Ragusa, Italy, ²D.A.C.P.A. University of Catania, Italy.

Pecorino Siciliano is a PDO ewe’s milk cheese produced in Sicily. The aim of this study was to evaluate the influence of different production areas on the dynamics of the microbial communities of artisanal cheeses by Temporal Temperature Gel Electrophoresis (TTGE). PCR-TTGE is a rapid molecular method, allowing to analyse diversity within bacterial communities, based on direct analysis of DNA molecules that differ by singles based. Among 21 farms allocated in seven different areas of Sicily classed as: Iblean (I), Etnea (E), South Center (SC), North Center (NC), Western (W), Western Center (WC), and Peloritana (P) seven experimental cheeses were selected on microbiological composition of cheese samples. Cheese ripened at two, four and eight months were sampled from each farm. Comparison of TTGE profiles showed differences among the different areas, allowing a further classification amenable to three main areas of Sicily (East, Center and West) as confirmed by cluster analysis of milk samples. The TTGE profiles of cheese samples obtained from East area farms resulted richer of microorganisms than the others areas, specially for the high GC bacteria (Propionibacteria, Artrobacteria and Corinebacteria). This result was further confirmed comparing the TTGE profiles of the rind of cheese samples. The main species detected in cheese samples from all areas were: *S. equorum*, *S. thermophilus*, *S. galloyticus* subsp. *macedonicus*, and *Lc. lactis*, subsp. *cremoris*, however these microorganisms were more consistent in East area. Furthermore, an high relationship between microorganisms detected in milk samples and those ones detected in cheese samples, was observed. These results indicated that the quality of starter milk, probably affected by geographic area, might be also directly related to variability development in bacterial ecosystem of cheese.

Key Words: Pecorino Siciliano cheese, PCR-TTGE, bacterial ecosystem

M63 Persistency of conjugated linoleic acid and vaccenic acid on Tybo cow cheese. G. A. Gagliostro^{*1}, M. Martínez², V. I. Cejas³, M. A. Rodríguez³, and M. Balán⁴, ¹Instituto Nacional de Tecnología Agropecuaria, Balcarce, Buenos Aires, Argentina, ²Instituto Nacional de Tecnología Agropecuaria, Salta, Argentina, ³Instituto Nacional de Tecnología Industrial, PTM Miguelete, Buenos Aires, Argentina, ⁴PRODEO S.R.L., Chivilcoy, Buenos Aires, Argentina.

The study was designed to test concentration of 9-cis 11-trans C18:2 (CLA) and trans-11 C18:1 (VA) on Tybo cheese elaborated from milk with high CLA and VA contents. Eight Holstein cows (570 kg liveweight, 109 days in milk) producing 23.4 kg milk consumed (DM) 7.3 kg /cow of concentrate, 1.9 kg of a TMR composed by corn silage (70%), soybean oil (22%), fish oil (5.4%) and urea (2.6%) and 8 kg of pasture. After 34 days of adaptation milk was collected and transformed into Tybo cheese reproducing industrial conditions. Fatty acid (FA) composition of milk and cheese was analyzed by GLC and differences in FA content between milk and cheese were stated using the T-test for paired observations. Milk fat, protein and lactose contents were 24.2, 34.5 and 46.6 g/kg. Intake of 418 g of soybean oil and 103 g of fish oil contained in the TMR reduced the atherogenicity index (AI = [(C12 + 4C14 + C16)/Total unsaturated FA]) of milk from a pre-supplementation basal value of 2.06 to 1.16 Basal concentration (g/100g) of the atherogenic FA C12:0 (4.04) C14:0 (12.52) and C16:0 (29,16) were decreased by feeding oils (C12:0 = -1,84, 45%, C14:0 = -3,64, 29% and C16:0 = -3,29, 11%). CLA increased in milk from a basal value of 1.42 to 2.86

g/100g and VA from 2.56 to 3.55 g/100g FA after oil supplementation. Elaborated cheese contained 21.6% fat and 26.8% protein (w/w) with an atherogenicity index of 1.29. Differences between milk and cheese FA content were detected only for C4:0, C14:0 and VA. Concentrations of C4:0 (+0.42 g/100g, 36%), C14:0 (+0.85 g/100g, 10%) and VA (+0.93 g/100 g, 26%) increased in cheese. Transfer of 9-cis 11-trans CLA to Tybo cheese was 95%. Assuming that the cheese fat contain 95% FA, intake of 143 g/d of Tybo cheese rich in CLA may allow to obtain the suggested anticancer dose (800 mg) of CLA achieving also the proposed dose (250 mg CLA) that confers protection against cardiovascular disease for the consumer.

Key Words: cheese, conjugated linoleic acid, vaccenic acid

M64 Influence of microfiltration and adjunct culture on quality of Egyptian soft white cheese. S. Awad*, N. Ahmed, and M. El Soda, *Alexandria University, Alexandria, Egypt.*

Classical techniques used to improve milk shelf life and safety are based on heat treatments, like pasteurization and sterilization. Those techniques modify some physico-chemical properties of milk, for example its coagulation by rennet. Microfiltration constitutes an alternative to heat treatment to reduce the presence of bacteria and improve the microbiological safety of dairy products without modifying the physico-chemical properties of milk. Pasteurization and/or microfiltration of milk are recommended before cheese making to improve the hygienic quality of cheese. These will have a negative impact on the natural flora present in raw milk. Eight adjunct cultures of lactic acid bacteria, isolated from Egyptian dairy products, were evaluated in experimental Egyptian soft white cheese for flavour development. The effect of microfiltration (MF) and pasteurization on proteolysis, lipolysis and flavor development in Egyptian soft white cheese during 4 months of ripening were also studied. The chemical composition of cheeses seems to be affected by microfiltration rather than the culture types. The moisture content was higher and the pH was lower in pasteurized milk cheeses compared to microfiltered milk cheeses at day one of manufacture. Chemical composition of experimental cheeses was within the legal limit for Egyptian soft white cheese in Egypt. All adjunct cultures used in this study had no effect on chemical composition of Egyptian soft white cheese. Proteolysis and lipolysis during cheese ripening were lower in microfiltered milk cheeses comparing to pasteurized milk cheeses. Very significant variations in free amino acids, free fatty acids and sensory evaluation have been found among the adjunct cultures used in Egyptian soft white cheese. Cheese made using adjunct culture of *Lactobacillus delbrueckii* subsp. *Lactis*, *Lactobacillus paracasei* subsp. *paracasei*, and *Lactobacillus plantarum* received the highest scores of flavor and texture.

Key Words: Egyptian soft white cheese, microfiltration, proteolysis

M65 Development of a flavor lexicon for processed and imitation cheeses. S. L. Drake*, M. D. Yates, and M. A. Drake, *North Carolina State University, Raleigh.*

Processed cheeses and cheese foods are made using natural cheese, but imitation products made with caseinate and nondairy proteins are also available. Functionality and meltability are primary characteristics of these products, but flavor is also important and can be a defining characteristic. Studies have not focused on flavor of these products. This study was conducted to identify and define the sensory proper-

ties of processed and imitation cheeses. An experienced sensory panel (n=9) evaluated an array of processed and imitation cheeses (n=48 products). Following language development and refinement, selected processed and imitation products (n=20) were evaluated in duplicate by each panelist. Data was evaluated by univariate and multivariate analyses. Twenty aromatics and five basic tastes were documented in the products. However, a language of fourteen attributes could be used to differentiate most products. Processed cheeses were characterized by sweet aromatic, cooked/milky, diacetyl, whey, and milkfat flavors. Processed cheeses labeled as sharp were characterized by the previous terms plus sulfur, fruity, free fatty acid, and brothy flavors. Processed cheese foods were characterized by similar flavors but lower or absent in cooked/milky flavor. Imitation cheeses with casein/caseinates were characterized by sweet aromatic, minty and hay flavors while imitation cheeses with other protein sources had cardboard, fatty, oxidized, and fishy flavors. This study demonstrated an array of flavor profiles among processed cheese and between processed and imitation cheeses. This flavor lexicon can help the industry to better define and differentiate processed and imitation cheeses and to understand consumer flavor preferences for these products.

Key Words: processed cheese, flavor, cheese food

M66 Effect of cream cheese made from freeze-dried milk powder on physicochemical properties. S. H. Kim¹, S. Y. Lee¹, J. Ahn², and H. S. Kwak*¹, ¹*Sejong University, Seoul, Korea*, ²*Jungwon University, Chungbuk, Korea.*

The cream cheese was manufactured from freeze-dried milk powder (FDMP) that was made by newly developed continuous multi-stage process. Physicochemical properties examined were composition, specific gravity, color, short-chain fatty acids and texture during 4 week storage at 4°. The compositions of the FDMP cream cheese were 32.7% fat, 8.3% protein and 57.1% water which were comparable to that of the cheese from control and spray-dried powder milk, respectively. Specific gravity of the FDMP cream cheese was 1.072 and it was not significantly different (P<0.05) from others. In color observation, L-, a-, and b-value were slightly different in each samples, however, they were not significant. Total short-chain fatty acid of all samples increased significantly during storage up to 4 weeks, but the FDMP cream cheese showed the lowest concentration among samples. This result indicated that the FDMP cream cheese could be extended storage time longer than others. In texture study, hardness scores decreased significantly in all samples during storage. The FDMP cream cheese showed that the scores were lower than control but higher than the cheese from spray-dried milk powder. Cohesiveness scores of the FDMP cream cheese were significantly increased during storage period and were higher than that of control and similar to the cheese from spray-dried milk powder. In conclusion, the quality of the FDMP cream cheese is similar to that of control and better than that of the cheese from spray-dried milk powder.

Key Words: cream cheese, freeze-dried milk powder, spray-dried milk powder

M67 Optimization of recovery of key Cheddar cheese flavor compounds from full fat and reduced fat Cheddar cheeses. D. M. Watson*, R. E. Miracle, and M. A. Drake, *North Carolina State University, Raleigh.*

Fat reduction in the American diet concerns both consumers and food manufacturers. Studies indicate that cheese consumers are interested in low fat cheese if a similar flavor profile to full fat cheese is assured. Recent flavor chemistry studies have identified twenty key flavor compounds in full and low fat Cheddar cheeses. Precise and high throughput extraction and quantitation of these compounds are crucial to identify methods to improve flavor of low fat Cheddar cheese. The objective of this study was to optimize a headspace solid phase microextraction (SPME) method for the recovery and quantitation of these key aroma compounds from full and reduced fat Cheddar cheese. Commercial full fat and 75% reduced fat Cheddar cheeses were purchased and used for the study. Sample size, salt content, extraction time, and extraction temperature were evaluated for their effect on recovery of the twenty previously identified compounds in a response surface model – central composite design (RSM-CCD) design matrix. The entire design was repeated using three SPME fiber types (Polydimethylsiloxane, PDMS; Carboxen-PDMS, CAR/PDMS; and Divinylbenzene-CAR/PDMS, DVB/CAR/PDMS) to determine which fiber was most efficient for key volatile compound recovery. Compounds were identified by gas chromatography mass spectrometry. The 3-phase DVB/CAR/PDMS fiber provided the most complete recovery of volatile compounds from the full and reduced-fat Cheddar cheeses. Sample conditions of 51 C for 40 min resulted in optimal recovery of low molecular weight compounds, especially acetic and butyric acids and 3-methylbutanal. Increasing sample weight under the same conditions resulted in the highest recovery of higher molecular weight compounds such as hexanoic acid, two lactones and phenylethanal. In contrast, a lower temperature time profile (44C for 20 min) allowed the highest recovery of both low molecular weight and low boiling point compounds. These results indicate that different sample extraction conditions can be used to achieve rapid and precise quantitation of key cheese flavor volatiles.

Key Words: cheese flavor, volatile compounds, volatile recovery

M68 The influence of sodium chloride on flavor of natural Cheddar cheese. M. A. Drake^{*1}, R. E. Miracle¹, and D. J. McMahon², ¹North Carolina State University, Raleigh, ²Utah State University, Logan.

Sodium reduction concurrent with fat reduction in the American diet are currently key issues for food processors. Dietary guidelines currently suggest a maximum intake of 2300 mg of sodium per day, while the average consumer intake is 9 g per day. Natural Cheddar cheese is not a low sodium food (~175 mg per serving) and salt plays a crucial role in Cheddar cheese ripening and flavor. The objective of this study was to evaluate the impact of sodium chloride on sensory properties of low fat Cheddar cheese. Duplicate batches of low fat Cheddar cheese (6.0% fat, 51% moisture) were manufactured with 1% sodium chloride and aged at 3C for 4 months. Following aging, cheeses were comminuted to particles of size 1 to 4 mm, divided into five 2.7-kg portions, and sufficient salt added to produce cheese containing 1.0, 1.4, 1.8, 2.2, and 2.6% salt. Cheeses were then re-formed, vacuum-sealed and stored at 3C for 1 mo prior to sensory testing. This procedure allowed the impact of sodium chloride on sensory properties to be evaluated without altering cheese biochemistry. A trained panel documented the sensory properties of cheeses. Consumer acceptance testing (n=75 consumers) was subsequently conducted to document consumer perception of cheeses with different salt contents. Salty taste, and to a lesser extent umami taste, increased with increasing sodium chloride concentration by descriptive sensory analysis (p<0.05). Sodium chloride addition and salty taste did not cross-modulate other trained panel sensory attributes. Consumers documented differences in salty taste intensity between all

salt concentrations except 2.2 and 2.6% (p<0.05). Salty taste liking scores suggested that salt concentrations of > 1.8% resulted in cheeses with acceptable salty taste intensity. A salt reduction of as little as 25% from typical salt content (1.8 decreased to 1.4%) in low fat Cheddar cheese was noticed by consumers and negatively impacted acceptance (p<0.05). Salty taste intensity is a consumer expectation in Cheddar cheeses, and exploration of sodium chloride alternatives is warranted if sodium reduction in this product is desired.

Key Words: salt reduction, cheese flavor, low sodium

M69 Automatic detection of microstructural features using a statistical image processing method. G. Impoco¹, L. Tuminello¹, N. Fucà¹, M. Caccamo^{*1}, and G. Licitra^{1,2}, ¹CoRFiLaC, Ragusa, Italy, ²D.A.C.P.A., University of Catania, Catania, Italy.

Detecting features in digital micrographs of cheese samples is an important pre-processing step for automated quantitative analysis. A statistical Image Processing algorithm has been developed for automated detection, requiring no user intervention. Scanning Electron Microscope (SEM) and Confocal Laser Scanning Microscope (CLSM) were used to acquire images of Ragusano pasta-filata cheese. SEM images were taken at 15kV, working distance 18mm, 1000×. Confocal images were taken at 10×, numerical aperture 0.30. The main microstructural features of the images were gathered into specific classes: fat globules, generic pores, protein matrix, and pore clusters, for SEM images; fat, protein, empty areas, and gas holes, for CLSM images. Images were manually labelled using an ad-hoc tool. The labelled pictures and the original micrographs were used to train a statistical algorithm, on the basis of pixel intensity and gradient neighborhoods, computed as multi-scale N,S,W,E circular sections around each pixel. The result of this learning stage was 51 probability density functions (PDFs), describing the probability of pixels to take certain numerical values for each feature class. Different PDFs were generated for SEM and CLSM images, and collected into two different libraries. After training, new images can be classified using the learned statistics. The algorithm associates to each pixel its probability to belong to a certain feature class. Notice that, although an intensive manual effort is necessary for training, this work is carried out only at the very beginning on a new type of cheese. Once the method has been trained, the operative stage requires no user intervention at all. Satisfactory classification results have been obtained for SEM images. Results for CLSM images were not accurate and can be probably improved using geometric features. Nevertheless, this statistical analysis suggested that in the observed images gas holes lie close to fat with high probability.

Key Words: cheese microstructure, image processing, feature detection

M70 Lactic acid bacteria enhance levels of conjugated linoleic acid in Cheddar cheese. A. J. Pandit, S. K. Anand^{*}, A. N. Hassan, and K. F. Kalscheur, South Dakota State University, Brookings.

The present study was undertaken to produce Cheddar cheese with enhanced levels of conjugated linoleic acid (CLA) using two strains of lactic acid bacteria previously identified as high CLA producers (*Lactococcus lactis* 4b isolated from Cheddar cheese and produced 1.12 g CLA/100g fatty acids in milk, and *Lactobacillus helveticus* ATCC 15807, which produced 0.54 g CLA/100g fatty acid in milk). These cultures were evaluated for their suitability for cheese making by the

cheese slurry method. The four treatments employed were: 1) cheese made with a CLA negative culture (DVS), 2) cheese made with *Lc. lactis* 4b, 3) cheese made with a combination of DVS and *Lb. helveticus* ATCC 15807, and 4) cheese made with a combination of *Lc. lactis* 4b and *Lb. helveticus* ATCC15807. The control cheeses made with DVS had a CLA level of 0.62g/100g FA (day 1). The CLA levels in cheese manufactured using *Lc. lactis* 4b increased from 0.86g/100g FA on day 1 to 0.97g/100g FA after 6 months of ripening. The addition of *Lb. helveticus* (treatments 3 and 4) although showed an initial increase in CLA levels to 0.84 and 0.83 g/100g FA on day 1, did not show any significant enhancement throughout the ripening period. There was no difference ($P>0.05$) in the chemical composition of cheeses among treatments. Due to higher acid production, the pH values of cheeses made with *Lc. lactis* 4b were lower than those made with DVS. Proteolysis (water soluble nitrogen and trichloroacetic acid soluble nitrogen) was higher in the CLA-positive cheeses than in the control cheese. The CLA-positive cheeses had lower ($P>0.05$) levels of free oil and less meltability than control cheeses. Textural properties of CLA-positive cheeses differed from those in the control cheese during the first 3 months but not after 6 months of ripening. Bacterial counts on M17 and MRS agar were higher ($P>0.05$) in CLA-positive cheeses than in the control cheese throughout the ripening period. The CLA-positive cheeses had high sensory scores. Our study demonstrates the possibility of increasing levels of CLA in Cheddar cheese using lactic cultures without the need of substrate addition to milk.

Key Words: lactic acid bacteria, CLA, cheese

M71 Effect of aging on the rheology of full fat and low fat Cheddar-like caprine cheese. D. L. Van Hekken^{*1}, Y. W. Park², and M. H. Tunick¹, ¹*Dairy Processing & Products Research Unit, Agricultural Research Service, Wyndmoor, PA*, ²*Agricultural Research Station, Fort Valley University, Fort Valley, GA*.

The rheological properties of aging full fat (FF) and low fat (LF) caprine milk cheeses were characterized to determine the changes in the cheese matrix during storage. Three batches each of high moisture, Cheddar-like caprine FF and LF cheeses were manufactured from whole or skim milk from the University's goat herd and were aged at 4°C for up to 6 months. Rheological properties (texture profile, torsion, and small amplitude oscillatory shear analyses) and protein distribution (SDS-PAGE) were measured after 1, 3, and 6 months of storage. The LF cheeses contained 55.1% moisture, 35.7% protein, and 1.3% fat while the FF cheeses contained 47.7% moisture, 22.5% protein, and 25.6% fat. Structurally, the LF cheeses were significantly harder, chewier, and more cohesive than the FF cheeses throughout storage; overall averages for LF versus FF cheeses were 160 N versus 17.4 N for hardness, 326 mJ versus 27.2 mJ for chewiness, and 0.24 versus 0.18 for cohesiveness, respectively. Higher values for the point of fracture and viscoelastic properties were noted as well for the LF cheeses when compared to FF cheeses; overall means were 92.8 kPa versus 17.8 kPa for shear stress, 108 kPa versus 23.6 kPa for shear rigidity, 96.6 kPa versus 34.7 kPa for elastic modulus, and 34.4 kPa versus 12.8 kPa for viscous modulus, respectively. Although overall changes between months 1 and 6 were not always significant, the LF cheeses showed more variation in their rheological properties than the FF cheeses. The LF cheeses demonstrated that fat is critical in creating a soft flexible cheese matrix and the results can be used to identify fat levels that will meet consumer demands for lower fat cheeses with acceptable texture.

Key Words: caprine milk, cheese, rheology

M72 Effect of renneting pH on calcium balance in cheese making process. N. Remillard* and M. Britten, *Food Research and Development Centre, Agriculture and Agri-food Canada, St-Hyacinthe, QC, Canada*.

Texture and melting properties of Italian cheese (Mozzarella) are influenced by calcium content. Decreasing whey drainage pH or curd washing are common practices to control calcium concentration in cheeses. In the present work, renneting pH was studied as a potential factor controlling calcium level in cheese. The effect of renneting pH (between 6.65 and 6.2) on: 1) the calcium transfer to the soluble phase during milk acidification, 2) the characteristics of rennet gels and 3) the cheese mass balance were determined. Cheese milk was standardized to a protein to fat ratio of 1.28 and a casein concentration of 2.65%. Decreasing the pH of milk was carried out by inoculation with thermophilic starter at 33.7°C. During the acidification and coagulation process soluble calcium concentration was monitored and calcium distribution between colloidal and soluble phases was established. The kinetics of gel formation as well as rennet gels permeability were determined as a function of renneting pH. Model cheeses were produced at a laboratory scale. Moisture, protein, fat and calcium distribution were determined from mass balance and composition of milk, cheese and whey. During acidification, the increase of soluble calcium concentration (d[Ca]/dpH) was 2-fold higher before renneting (liquid state) than after (gel state). As a consequence, decreasing renneting pH by 0.1 units reduced micellar calcium content by about 0.5mg per g casein in cheese curd at drainage. Lower renneting pH was associated with rapid gel formation and rennet concentrations had to be reduced to maintain constant gel formation kinetics. The permeability coefficient of rennet gels increased with decreasing renneting pH, suggesting faster dehydration of the gel matrix. Micellar calcium concentration in model cheese was significantly reduced by lowering renneting pH. Furthermore, cheese moisture and fat retention coefficients were both increased when the renneting pH was reduced.

Key Words: renneting pH, calcium, cheese

M73 Denaturation of proteins measured in liquid whey. M. Allen* and P. Tong, *California Polytechnic State University, San Luis Obispo*.

Protein structure affects the functionality of whey protein ingredients in food systems. The degree of denaturation of whey proteins has become important for characterizing how whey protein ingredients will perform in a food system. Several methods have been developed to quantify protein denaturation of purified whey proteins, whey protein concentrates and isolates, milk powders, fluid milk, liquid whey and other more complex dairy systems. The purpose of this experiment was to compare three different methods to measure denaturation of whey proteins in liquid whey obtained by various methods of separation and with varying degrees of heat treatment. A split-split plot experimental design was used. Raw bovine milk was skimmed and liquid whey was separated from the skim milk. Three separation methods: centrifugation, membrane filtration and enzyme coagulation, made up the first split plot. Each sub-plot of liquid whey was then divided into three split-split plots to receive heat treatment. Heat treatments were no heat, 76°C for fifteen seconds and 85°C for three minutes. Each of the resulting nine sub-sub plots was analyzed by polyacrylamide gel electrophoresis, bicinchoninic acid-soluble Protein assay and fluorescence spectroscopy to determine the amount of denatured protein in the liquid whey. The total protein in the liquid whey samples varied based on the separation method and ranged from 0.6%-1.0% protein by weight. To account for the differ-

ences in protein content, the amount of protein denatured was expressed as a percentage of the original total protein in each sample. Of the total protein, the amount of denatured protein ranged from 10%-40%. Differences in denaturation due to separation method and heat treatment were detected by all three methods of quantification. However, data indicates that each of the three methods of quantification yield different amounts of denaturation, which indicates there are also differences are due to the method of quantification. Subsequent work will attempt to gain a more complete understanding of how different methods of measuring whey protein denaturation can be used to characterize whey protein ingredients functionality.

Key Words: liquid whey, denatured, protein

M74 Use of fluorescence spectroscopy for monitoring changes and predicting browning reactions during whole milk powder storage. P. Salunke*, J. Amamcharla, and L. E. Metzger, *Midwest Dairy Foods Research Center, South Dakota State University, Brookings.*

Maillard reaction products and browning intermediates are products of a series of chemical reactions between lactose and proteins and have an impact on the quality of dairy based ingredients including whole milk powder (WMP). Maillard browning in dairy products can be determined by reacting furfural compounds with 2-thiobarbituric acid (TBA) to yield a yellow color which can be quantitatively measured by UV-VIS spectrometry. Using this approach the level of free hydroxymethylfurfural (FH), browning intermediates (BI) and total hydroxymethylfurfural (TH) can be determined. However, measurement of FH, BI and TH in WMP using this approach is tedious and time consuming. The objective of this study was to determine if front face fluorescence spectroscopy (FFFS) could be used to characterize the Maillard reaction products of WMP caused by manufacture and storage. Two WMP samples were collected from 4 different manufactures (total of eight samples) and stored for 12 months. At 0, 2, 10 and 12 months of storage the FH, BI and TH content were determined on each sample using the standard TBA based protocols. FFFS was also performed on each sample (emission scan at 400-600nm with excitation wavelength of 360nm) at each time point. For FFFS analysis three scans were performed on three sub-samples of each WMP sample (9 scans total). The nine scans were then averaged to obtain a single spectrum. The spectra were then normalized and used as input for the prediction of the FH, BI and TH. Root mean square error of cross validation (RMSECV) and relative standard error of prediction (RSEP) were used to evaluate the performance of the model. RMSECV of the samples were 1.53, 1.28 and 1.58 HMF μ moles/L, whereas RSEP was 10.48, 3.44 and 3.10% for FH, BI and TH, respectively. There was a good agreement between measured and predicted values of BI ($R^2=0.92$) and TH ($R^2=0.87$). However, the partial least squares (PLS) prediction model for FH had a high RMSECV and RSEP. The results indicate that FFFS has potential to characterize WMP and predict HMF values.

Key Words: whole milk powder, front face fluorescence spectroscopy, browning reaction prediction

M75 Profiling changes in amino acids and organic acids in Cheddar cheese during ripening using infrared spectroscopy. A. Subramanian*, J. Harper, and L. Rodriguez-Saona, *The Ohio State University, Columbus.*

Composition and flavor quality of Cheddar cheese, which influence the consumer acceptance, price and food processing application,

develop during the ripening process. Organic acids and amino acids play important roles in flavor development. Therefore determination of their levels and the changes during ripening is important to understanding their influence on cheese quality. Fourier-transform infrared (FTIR) spectroscopy is an attractive technique for rapid and sensitive analysis of food composition. The goal of this research was to combine FTIR spectroscopy with a simple extraction technique to rapidly profile changes in organic acids and amino acids during cheese ripening. Twelve different cheese samples were ripened for a period of 73 days. Samples were collected on days 7, 15, 30, 45 and 73 during ripening, powdered and extracted with water, chloroform and ethanol. The extracts were analyzed by gas chromatography for amino acids, liquid chromatography for organic acids and FTIR for infrared spectra ($4000-700\text{cm}^{-1}$). The collected spectra were correlated with the amino acid and organic acid concentrations and analyzed by partial least squares regression (PLSR) analysis. Analysis of variance (ANOVA) of the chromatographic data was used to identify the compounds that exhibited significant changes during ripening. The FTIR spectra correlated well with amino acid and organic acid concentrations. The PLSR models showed excellent fit with coefficient of correlation >0.95 . Amino acids and organic acids could be simultaneously determined in less than 20 min. The estimated standard errors for predicting unknown samples were $<4\%$. Some of the prominent infrared marker bands that were responsible for the correlation were 1411 (amino acids), 1710-1450 (fatty acids), and 1200-1050 cm^{-1} (organic acids). Lactic acid, glutamic acid, leucine, asparagine, phenylalanine and valine are some of the compounds that exhibited significant changes during ripening. This method could potentially speed up amino acid and organic acid determination due to the possibility of simultaneous analysis and thereby save time and money.

Key Words: Cheddar cheese, rapid analysis, infrared spectroscopy

M76 Production of nisin-containing whey protein concentrate. H. Abd El-aal¹, R. Dave¹, A. Khattab², and A. Hassan*¹, ¹*South Dakota State University, Brookings,* ²*Alexandria University, Alexandria, Egypt.*

The objective of this study was to optimize conditions for the production of nisin by two strains of *Lactococcus lactis* subsp. *lactis* in ultrafiltered (UF) whey, and study its stability in freeze dried whey protein concentrate. Pasteurized reconstituted (6% w/v) dried sweet whey concentrated 5 times by UF was fermented at 30°C for 24 hours by each of two strains of *Lactococcus lactis* subsp. *lactis* (ATCC 7962 and B10). The pH of the media was maintained at 6.5 using 5 M solution of ammonium hydroxide during the first 8 hours of fermentation followed by a free drop to a value of 4.7 to 4.9. Fermented media were freeze dried, packaged in polyethylene bags, and stored at 4 or 30°C for 3 months. Freeze drying was used as initial trials showed that the single stage drying decreased nisin activity by almost 50%. Freeze drying did not affect nisin activity. Nisin was determined by the agar well diffusion method in the fermented media before and after drying. No differences ($P > 0.05$) in nisin production were seen between the two strains or in regular and UF whey. Boiling of cells prior to nisin assay increased ($P < 0.05$) nisin activity. Whereas nisin activity in UF whey fermented with BS10 and ATCC 7962 was 183 and 167 UI/ml in the absence of yeast extract, it reached 1617 and 1667 UI/ml in the presence of 1% yeast extract, respectively. Nisin activity in the 1 month old whey protein concentrate powder stored at 4°C was about 40 to 45% of that before storage. This value further reduced to 30 to 35% during storage at 30°C. Boiling of reconstituted powder at pH 2 for 5 minutes released nisin from the producing cells and increased ($P < 0.05$) activity. Boiled whey maintained 75% of the

original nisin activity after storage at 4°C for 1 month. By the end of the second month of storage, powder maintained only 15 to 20% of the initial activity. No differences ($P > 0.05$) in nisin activity were observed between 2 and 3 months of storage. Whey protein concentrate powder containing nisin was successfully produced. Future research will study stability of nisin in whey protein concentrate produced using a multistage spray dryer and stored in modified atmosphere.

Key Words: nisin, whey protein concentrate, storage

M77 Bovine milk based infant formula promote the growth and acid production of bifidobacteria. K. Mohamedali* and S. A. Ibrahim, *North Carolina A & T State University, Greensboro.*

Several infant formulas are currently available in the market that are fortified with different nutrients to nourish infant development. Because recent research has demonstrated that bifidobacteria promote good health, it is important to determine the effects that infant formula may have on the growth of bifidobacteria. Therefore the objective of this research was to determine whether different commercial infant formulas are associated with differences in the growth and acid production of bifidobacteria in skim milk. Fifteen different strains of bifidobacteria were used in this study. Skim milk samples (100ml) were fortified with one of the 15 infant formulas and sterilized at 65 °C for 30 min. Samples were then cooled to 40°C and inoculated with individual overnight strains of bifidobacteria to achieve a final inoculum level of 3.0 log CFU/ml. Samples were then incubated at 37°C for 24 hrs. Bacterial growth was monitored at different time intervals by measuring pH values and total titratable acidity. At the end of the incubation period, samples were withdrawn and appropriate dilutions were surface plated on MRS agar. Results showed that the different rates growth of bifidobacteria were observed due to different formulas. Bovine milk protein hydrolysate based formulas enhanced the growth of bifidobacteria (as measured by the acid development and total bacterial population) significantly ($P < 0.05$) compared with other formula base types. These results suggest that bovine milk protein hydrolysate based promote the growth of bifidobacteria and should be considered when selecting formula as an alternative for mother's milk.

Key Words: growth factor, bifidobacteria

M78 Induction of α and β galactosidases from *Lactobacillus reuteri* by different metal ions. A. Y. Alazzeh*¹, S. A. Ibrahim¹, D. Song¹, A. Shahbazi¹, and A. A. AbuGhazaleh², ¹*North Carolina A & T State University, Greensboro,* ²*Southern Illinois University, Carbondale.*

Probiotics are food grade bacteria that could be used safely and directly as food supplements. Probiotics have many health benefits to the host such as better digestibility of sugars mainly lactose and raffinose, antimicrobial activity and decrease blood cholesterol of the host. Health benefits of probiotics are strain specific and *Lactobacillus reuteri* has been shown to have good potential for the production of digestive enzymes (α and β -galactosidases). Bacterial media could be an important factor in over producing these enzymes. The induction of α and β -galactosidases in probiotic bacteria is one of the interesting areas in food science. The objective of our study was test the induction of α and β -galactosidases by metal ions. 10 mM of Na⁺, K⁺, Fe²⁺, Cu²⁺, Mg²⁺ and 1 mM of Mn²⁺ were added separately to the growth media to test the induction of α and β -galactosidases in six strains of *L. reuteri* (CF2-7F, DSM20016, MF14-C, MM2-3, MM7 and SD2112). Results

showed that the activity of α and β galactosidases in *L. reuteri* in response to added metal ions is strain specific. The addition of Cu²⁺ lead to a complete inhibition of both enzymes, while the addition of Fe²⁺ lead to partial inhibition of both enzymes. *L. reuteri* (strain CF2-7F) showed the highest α and β -galactosidase activity when grown on a media with added Mn²⁺ ions (22.66 and 19.33 Gal U/ml, respectively). One mM Mn²⁺ ions could be added to the growth media of CF2-7F to induce the activity of both enzymes.

M79 Immobilization of *Lactobacillus acidophilus* in apple pieces (*Pyrus malus* L.) and mamey sapote (*Pouteria sapota*) for whey fermentation. M. E. Yañez-Villar¹, E. Paz-Gamoba*¹, A. Perez-Silva¹, H. S. García², and M. Montero-Lagunes³, ¹*Instituto Tecnológico de Tuxtepec, Tuxtepec, Oax, Mexico,* ²*Instituto Tecnológico de Veracruz, Veracruz, Ver, Mexico,* ³*INIFAP Campo Experimental, Veracruz, Ver, Mexico.*

Lactobacillus acidophilus is a homofermentative probiotic bacterium which has been employed to treat digestive disorders. Immobilized cell preparations have been used to provide stability and better process conditions. In this work we evaluated cell viability of *L. acidophilus* immobilized on apple and mamey sapote chunks. Enriched whey was used as propagation medium with cell concentrations of 1, 5 and 10 gL⁻¹. *L. acidophilus* cells were immobilized at 37 °C for 12 h without stirring on fruit pulp cubes of 0.5 or 1 cm. The preparations fermented whey supplemented with 2% glucose and 1% yeast extract, and a control was included which contained free cells, under the same reaction conditions. Cell viability, pH values and acidity as % lactic acid were monitored in stored fermented whey samples that were kept at 4°C for 90 days. Fermented wheys developed acidity with decreased pH values and cell populations of 10⁸ CFU/mL were reached with a 10 gL⁻¹ inoculum in 8 h. Cells immobilized on mamey sapote cubes produced less acid but were more stable than free cell preparations and their populations reached >10⁸UFC/mL with the same inoculum after 90 days of storage. Immobilized cell preparations showed good stability and re-usability, showed potential as carrier of probiotic bacteria as *L. acidophilus* and can be used to develop probiotic drinks containing fruit pulp and whey with a fermentation time of 6-8 h.

Key Words: probiotic, immobilization, *Lactobacillus acidophilus*

M80 A simple on-farm technique for early detection of foreign substances in milk. M. H. Hathurusinghe*¹, A. Alazzeh¹, A. Shahbazi¹, S. A. Ibrahim¹, and A. A. AbuGhazaleh², ¹*North Carolina A & T State University, Greensboro,* ²*Southern Illinois University, Carbondale.*

High accessibility to dairy farms and poor level of biosecurity creates the possibility of intentional contamination of milk with biological or chemical agents. If such an event occurred contaminated milk from a single farm could affect large fraction of dairy output. Current available contamination detection methods require sophisticated techniques and cumbersome procedures. To improve detection capabilities, simple, robust, and rapid and effective techniques are needed. The main objective of this project was to investigate the use a simple and rapid test using Lactic Acid Bacteria (LAB) as indicator organisms to detect contaminants in milk. Preliminary studies have shown that LAB are sensitive to a number of toxins including arsenic, cadmium and cyanide. In this study, the ability of LAB to detect rat poison and its major component chemicals (Brodifacoum) was tested. Five strains of *Lactobacillus reuteri* were added to test tubes separately containing 10ml of MRS broth

and different concentrations of either a commercial brand of rat poison or a single chemical component. The test tubes were then incubated at 37 C. Growth of the bacteria was monitored by measuring the optical density. pH values of each sample was measured at the end of the incubation period. Four out of the five strains showed sensitivity for rat poison. Further studies will be carried out with the other LAB to select the strains that show fastest and greatest sensitivity and an organic dye will then be selected, which gives rapid color change for the pH change. This system will be improved to a highly sensitive, environmentally safe, quick and accurate test kit which could be used as a universal marker for early detection of terrorist attacks to the food supplies.

Key Words: biosecurity

M81 Fatty acid composition in ewe's milk fat produced in lowland, hill and highland areas of Sardinia. M. G. Manca, F. Puggioni, R. Boe, R. Rubattu, G. Battacone*, and A. Nudda, *Dipartimento di Scienze Zootecniche, University of Sassari, Italy.*

Aim of this work was to determine the fatty acid (FA) profile of sheep milk from farms located at different altitudes and characterized by different feeding practices. The FA profile, including the content of conjugated linoleic acid (CLA c9, t11), vaccenic acid (VA; C18:1 t11) and n3 FA, was determined by gas-chromatography. Milk bulk samples were collected every two weeks from April to July 2008 from 15 sheep farms of Sardinia: five farms in the lowland, five in the hill and five in the highland. Data were analyzed with a linear model using sampling period, altitude and sampling period × altitude as fixed factors. During the trial, sheep feeding was widely based on pasture, with some differences among the 3 areas. The FA composition was significantly affected by sampling period. The highest contents of CLA, VA and n3 FA (2.1, 4.6 and 2.0 g/100g of FAME, respectively) were observed in April when pasture availability was the highest, and the lowest ones were in July (1.1, 1.3 and 1.0 g/100g of FAME, respectively) when grass was more mature. Altitude influenced significantly all FA analyzed (Table 1). Interaction was significant for almost all FA, except for VA and CLA. The contents of VA, CLA, and C18:3 n3 were higher in lowland farms compared to hill and highland. A possible explanation for this pattern could be the differences in sheep farming system among the areas, especially the greater availability of fresh grass in the lowland, due to a larger presence of cultivated grass pasture.

Table 1. Fatty acid profile in milk produced in sheep farms localized at different altitude

Fatty Acid (g/100g of FAME)	Altitude		
	Lowland	Hill	Highland
C4-C16	52.1 ^b	51.6 ^{ab}	52.5 ^a
C18	10.9 ^b	12.53 ^a	12.21 ^a
C18:1 t11	2.7 ^a	2.5 ^{ab}	2.3 ^b
C18:1 c9	22.0 ^b	22.7 ^{ab}	23.1 ^a
C18:2 c9,c12	3.4 ^a	2.9 ^b	2.6 ^c
C18:3 n-3	1.5 ^a	1.2 ^b	0.8 ^c
CLAc9, t11	1.5 ^a	1.3 ^b	1.2 ^b
PUFA n-3	1.9 ^a	1.6 ^b	1.1 ^c
n6/n3	2.0 ^b	2.2 ^b	3.0 ^a

^{a,b,c}Means within a row with different superscripts differ.

Key Words: fatty acid, sheep milk, altitude

M82 Properties of nanopowdered chitosan-added and cholesterol-reduced yogurt during storage. M. H. Seo, Y. S. Lee, and H. S. Kwak*, *Sejong University, Seoul, Korea.*

The present study designed to develop the nanopowdered chitosan-added and cholesterol-reduced yogurt, and investigate the physico-chemical property and microbiological changes during storage. The percentages of powdered (PC, 150 µm) and nanopowdered chitosan (NPC, 500-600 nm) were 0.1, 0.3, 0.5 and 0.7%. The cholesterol removal was in the range of 92.7 to 93.5%. The pHs in PC and NPC-added yogurts were unchanged for 15 days, however, no chitosan added yogurt showed a pH decrease from 4.21 at 0 day to 4.05 at 10 days and was maintained thereafter. The counts of lactic acid bacteria decreased as percent of PC and NPC addition increased and storage time. At 15 day storage, the counts were 1.14×10^8 cfu/ml in no chitosan-added group, while in the range about 105 to 106 cfu/ml in both NPC and PC-added groups. In both PC and NPC-added yogurts, the viscosity decreased with storage period and it were significantly lower compared with that in no-chitosan-added group through the storage except 0.7% PC-added yogurt ($p < 0.05$). There was no trend found in color, however, a-value in the NPC-added yogurt was significantly different compared with other yogurts ($p < 0.05$). Especially, 0.7% PC or NPC-added yogurts showed a significant difference in most of sensory characteristics at all storage periods. The present study indicated that NPC or PC addition into yogurt resulted in pH maintenance during 15 day storage which was due to less counts of lactic acid, and less than 0.5% chitosan may be applicable to the nanopowdered chitosan-added and cholesterol-reduced yogurt development.

Key Words: functional yogurt, nanopowdered chitosan, storage

M83 Residual beta-cyclodextrin and entrapped nutrients in milk treated by crosslinked beta-cyclodextrin. H. J. Ha, J. E. Lee, and H. S. Kwak*, *Sejong University, Seoul, Korea.*

This study was carried out to investigate the residual beta-cyclodextrin(beta-CD) and entrapped nutrients, such as water-soluble vitamins(L-ascorbic acid, niacin, thiamine and riboflavin), free amino acids, short-chain free fatty acid and lactose when cholesterol was removed in milk (milk fat: 3.4%, cholesterol: 10.6mg/100ml) treated by various concentrations of crosslinked beta-CD (0.4, 0.6, 0.8, 1.0 and 1.2%). The beta-cyclodextrins provided average 92.7% removal of cholesterol when 1.0% of the beta-CD was added. The residual beta-CDs were 1.22 and 1.32 ppm when 0.4% of crosslinked and powdered beta-CD were added, respectively. And the residuals proportionally increased to 3.00 and 3.18 ppm in 1.0% both beta-CDs. In the loss of nutrients observation, the difference of loss was not found in any nutrients and the additions of crosslinked and powdered beta-CD in the milk. The content of lactose in control milk was 4.86% and the loss was ranged from 0.00 to 0.03%. The total concentration of free amino acid was 8.78 mol/ml and the loss was ranged from 0.26 to 0.71 mol/ml. The concentrations of L-ascorbic acid, niacin, thiamine and riboflavin were 3.24, 1.33, 0.51 and 1.31 ppm in the control milk and the losses of the vitamins were ranged 0.02~0.05, 0.01~0.06, 1.11~0.16 and 0.01~0.06 ppm, respectively. The concentrations of butyric, caproic, caprylic and capric acid in the control milk were 4.45, 2.26, 2.78 and 3.21 ppm and the losses ranged 0.00~0.03, 0.00~0.04, 0.00~0.05 and 0.00~0.04 ppm, respectively. In conclusion, this study indicated that the residual beta-CD was trace amounts and entrapped nutrients were negligible.

Key Words: residual beta-cyclodextrin, entrapped nutrients, cholesterol-removed milk

M84 Effect of reconstituted milk made from freeze-dried milk powder on physicochemical properties. S. H. Kim, S. I. Ahn, Y. H. Chang, and H. S. Kwak*, *Sejong University, Seoul, Korea.*

This study was carried out to investigate the reconstituted milk made from freeze-dried milk powder (FDMP) that was made by newly developed continuous multi-stage process. Physicochemical properties tested were composition, color, thiobarbituric acid value (TBA), short-chain free fatty acids, water-soluble vitamins and fat-soluble vitamins stored at 4°C for 18 days. The compositions of FDMP reconstituted milk were 3.6% fat, 3.2% protein, 4.8% lactose and 87.2% water which were similar to that of the control and spray-dried milk powder (SDMP) origin. In color, L-, a- and b-value were slightly different among samples, however, they were not significant ($P < 0.05$). TBA absorbance of FDMP reconstituted milk was not significantly different from that of SDMP milk, however, both products were higher than control milk. Total short-chain fatty acids of FDMP milk were similar to that of control during storage up to 18 days, however, SDMP milk showed significantly higher than FDMP milk in the fatty acids. The concentrations of water-soluble vitamins, such as L-ascorbic acid, niacin, thiamine and riboflavin showed decreasing trend during storage periods. The vitamins from FDMP were lower than that of control, but higher than that of SDMP milk. The fat-soluble vitamins, such as retinol and tocopherol showed similar decreasing trends to the water-soluble vitamins during storage. In conclusion, the results of this study indicated that the quality of reconstituted milk made from FDMP is much improved than that of SDMP milk.

Key Words: reconstituted milk, freeze-dried milk powder, vitamins

M85 Comparison of physico-chemical properties between freeze-dried and spray-dried milk powders during storage. S. H. Kim, J. H. Park, and H. S. Kwak*, *Sejong University, Seoul, Korea.*

The present study was carried out to compare the physico-chemical properties between freeze-dried milk powder (FDMP) and spray-dried milk powder (SDMP) stored at different temperatures (4, 20 and 37°C) during 80 days. There was no difference in moisture, protein, fat and ash between both milk powders ($p > 0.05$). Dispersibility and wetting time were higher in FDMP than in SDMP, which indicated the higher quality of FDMP in physical property. FDMP showed the slightly higher pH was found at 4 and 27°C throughout the storage period compared with SDMP, however, a profound decrease was observed in both FDMP and SDMP at 37°C after 50 day. Similarly, solubility was higher in FDMP than in SDMP at 4 and 27°C, however, the solubility in FDMP decreased dramatically from 30 day storage when stored at 37°C. As expected, FDMP showed the lower moisture content at 4 and 27°C, and an increase after 40 day storage was found at both FDMP and SDMP. During 80 day storage, non-protein nitrogen (NPN) content was greater in SDMP at every temperature and increased continuously with the storage. In FDMP, after 60 day storage, NPN content was over 0.20, however, SDMP showed over 0.20 at the beginning of storage. Therefore, the present study indicated that the FDMP maintained the higher quality in physico-chemical properties including pH, spreadability, and solubility than SDMP, especially at 4 and 27°C during 80 day storage.

Key Words: freeze-dried milk powder, spray-dried milk powder, physicochemical properties

M86 Phylogenetic analysis of dairy *Penicillium* rDNA. G. Petit* and S. Labrie, *Université Laval, Québec, Canada.*

Despite their industrial importance, little is known about the genetics of dairy *Penicillium* spp. At present, 13 *P. camemberti* (syn. *P. candidum* and *P. caseicolum*) genes and 13 *P. roqueforti* are available in public sequence databases. However, little information is available on the ribosomal DNA (rDNA) of either species since only the sequence of the ITS1-5,8S-ITS2 region has been deposited in databases. Based on this 600 bp sequence, it has been shown that *P. commune* and *P. camemberti*, which is considered a domesticated form of *P. commune*, are virtually identical at the phylogenetic level. The *P. commune* designation is still widely used in the scientific literature because of a number of differences at the phenotypic level, mainly growth rate and color variations. The objective of the present study was to determine the genetic variability over the entire rDNA operon sequence (4768 bp) of four terverticillate *Penicillium* spp. found in the dairy environment (*P. camemberti*, *P. commune*, *P. roqueforti*, and *P. chrysogenum*). Multiple sequence analyses revealed that there are clear phylogenetic similarities between the four species. In addition, the operons of *P. camemberti* and *P. commune* differed by a single nucleotide, confirming that there is no significant difference between these two species at the phylogenetic level. *P. chrysogenum* and *P. roqueforti* had the highest variability, with 26 polymorphisms. Overall, our results showed that rDNA is highly conserved among these four *Penicillium* species, with only 31 polymorphisms detected.

Key Words: dairy foods, phylogenetic analysis, *Penicillium* spp.

M87 Effects of culture conditions on the growth and autoaggregation ability of bifidobacteria and *Lactobacillus reuteri*. O. A. Hassan*¹, S. A. Ibrahim¹, A. A. AbuGhazaleh², A. Shahbazi¹, and Y. Murad³, ¹North Carolina A & T State University, Greensboro, ²Southern Illinois University, Carbondale, ³National Research Council-Canada, Ottawa, Canada.

The use of bifidobacteria and lactobacillus as dietary adjuncts or as a source of probiotics is a subject of intense and growing interest. Several reports have indicated that probiotics have the ability to provide several health benefits. However, in order for these cultures to manifest beneficial effects, they need to achieve a viable mass through aggregation. A major factor affecting aggregation is the ability of these bacterial cells to adhere and grow within the host. This ability is a desirable property sought for use in commercial food preparations. The objective of this study was to evaluate the effects of different chemical factors on the growth and autoaggregating ability of bifidobacteria and *Lactobacillus reuteri*. Thirty-seven strains of bifidobacteria and eight strains of *L. reuteri* were cultivated in different culture media, (TPY, MRS) initial pH, and temperatures. Autoaggregation behavior was determined using the adhesion assay. Our results showed that the addition of calcium significantly induced autoaggregation for all of the strains in TPY media. We also observed that autoaggregation ability increased mainly at low initial pH levels (pH 5.5) and lower incubation temperatures (30-34°C). These findings indicate that autoaggregation ability can be significantly affected by several host environment factors including initial pH value and calcium. To achieve effective aggregations of probiotic bacteria, emphasis should be placed on optimizing growing conditions through introduction of calcium and pH lowering foods.

Key Words: autoaggregation, *Lactobacillus reuteri*

M88 80% whey (WPC) and serum protein (SPC) concentrate and 95% serum protein (SP) reduced micellar casein concentrate (MCC): Production and composition. J. Zulewska*², D. M. Barbano¹, M. Newbold¹, M. Drake³, E. A. Foegeding³, and C. Moraru¹, ¹Cornell University, Ithaca, NY, ²University of Warmia and Mazury, Olsztyn, Poland, ³North Carolina State University, Raleigh.

The research objective was to compare the production characteristics and composition of 80% WPC and 80% SPC. Raw whole milk (ca 1900 kg) was split into 2 portions: one was pasteurized (72C, 15s) and made into Cheddar cheese and whey and the other was cold separated into skim milk (SM) and cream. The SM was pasteurized (72C, 15 s), microfiltered (MF) to 3X (0.1 µm ceramic uniform transmembrane pressure) and diafiltered (DF) to 3X twice by adding reverse osmosis (RO) water back to the original SM weight to produce 95% SP reduced MCC. Separated whey (640 kg) and MF permeate (705 kg) from the first stage of MF of skim milk were ultrafiltered (UF) and DF using a spiral-wound 10 kDa polyethersulfone membrane to produce liquid 80% WPC and SPC. Whey and MF permeate were UF 5.2 and 5.5 X,

respectively. Protein content [measured by infrared spectrophotometer (IR)] was 47 and 41% of protein plus lactose in the retentate for whey and MF permeate after UF, respectively. Next, UF retentates from whey and MF permeate were diluted to their original weight with RO water and DF until the retentate protein content measured by IR was close to 90% of protein plus lactose. The DF retentates (ca 55 kg) were kept frozen (-40C) prior to spray drying (200C and 95C at inlet and outlet, respectively). All processing was replicated 3 times. Mean UF and DF flux was higher for WPC than SPC, but flux decreased more per hour for WPC than SPC over a 135 min run. Over a longer run, the WPC flux may be lower. Liquid SPC was clear while WPC was opaque; L-value was 18.5 vs. 52.5 for liquid SPC and WPC, respectively. Liquid SPC pH was higher than WPC (6.9 vs. 6.5, respectively). Spray dried (SD) 80% WPC contained more fat on a dry basis (db) than SPC, 0.5 vs. 8% and this may cause sensory differences between products. WPC contained more glycomacropeptide than SPC (about 4.9 vs. 0% of protein) and this may cause differences in functionality. The SD 95% SP reduced MCC contained 2.7% fat and 84.6% protein on db.

Key Words: whey protein, serum protein, micellar casein

Forages and Pastures: Forage Composition, Analysis and Utilization

M89 Utilizing near infrared (NIR) spectroscopy to predict carbohydrates (sugars) in forages. J. Horst*^{1,2} and G. Ayangbile^{1,2}, ¹Agri-King Inc., Fulton, IL, ²Analab, Fulton, IL.

The objective of this study was to determine if Near Infrared (NIR) can define the concentrations of carbohydrates in the cell contents of forages better than past methodologies. Total water soluble carbohydrates (WSC) and other acidic methods have been instituted in the past to quantify sugar contents. These methods are plagued with reproducibility problems, trying to determine a proper reference standard and numerous matrix interferences. NIR calibrations can be developed to assess carbohydrate concentration of mono and disaccharides in forages. The ability to rapidly predict Xylose, Fructose, Glucose and Sucrose in both fermented and non-fermented forages can provide useful nutritional information for optimizing animal performance. NIR also can provide accurate mannitol (sugar polyol) predictions on fermented silages, indicating undesirable heterofermentation. Calibrations were derived from reproducible wet chemistry techniques with extractions subjected to High Pressure Liquid Chromatography (HPLC) for analysis. The quantification of cell contents in feedstuffs including hay, haylage, corn silage, small grain silages and grain sources can potentially reduce incidents of laminitis in horses while improving feed efficiency in dairy, swine and poultry rations. NIR has achieved strong statistical correlations exceeding ≥ 90 1-VR (validation R-squared) on most analytes and forage matrices.

Key Words: carbohydrates, sugars, NIR Calibrations

M90 Investigation into the use of NIR predicted 12 and 30 hr IVNDFD as a measure of corn silage quality. R. T. Ward¹ and R. A. Patton*², ¹Cumberland Valley Analytical Service, Maugansville, MD, ²Nittany Dairy Nutrition, Inc., Mifflinburg, PA.

NDF digestibility in corn silage is important in determining the amount of nutrients provided in high producing dairy cattle with low rumen residence times. There is poor understanding of how to utilize available NIR predicted in vitro NDF digestibility (IVNDFD) evaluations to infer

differences in digestibility rates at rumen appropriate time points. This study characterized relationships between common nutrients in corn silage and IVNDFD at 12 hrs and 30 hrs measured by NIR to determine if the use of an earlier IVNDFD time point (12 hrs) allows for better characterization of rates of NDF digestibility. This study involved 14,576 samples of corn silage, including both normal and BMR types, for which both 12 and 30 hr NIR measured IVNDFD were reported for 3 crop years. Samples were analyzed for DM%, and CP%, soluble protein%, lignin%, ADF% and NDF% of DM. All nutrients were measured with NIR. The equations for 12 hr and 30 hr IVNDFD had R² of .883 and .853 for samples run by rumen incubation with digestibility of 36.7±1.57% and 60.9±2.41% respectively. Data were analyzed using proc mixed and proc reg of SAS. Fixed effects were crop year, geographic area (Northeast, Southeast, Midwest and Far West) and DM group. Dependent variables were IVNDFD at 12 and 30 hrs, the hourly rate of digestion for the first 12 hr and the last 18 hr. The Far West had lower 12 hr IVNDFD compared to other regions. Means for 12 hr and 30 hr IVNDFD were 38.7% and 58.3% respectively. Regression of 12 hr IVNDFD on 30 hr had R²=.51. Regression of rate of 12 hr IVNDFD with the final 18 hr yielded an R² of .18; thus speed of 12 hr digestibility was unrelated to speed of the last 18 hrs. Using all items analyzed, the maximum R² was .55 for 12 hr IVNDFD and .60 for 30 hr. We conclude 12 hr NIR predicted IVNDFD has the potential to be more appropriate than 30 hr NIR IVNDFD for characterizing NDF digestibility differences in corn silage. There appears to be a consistent geographical and DM% effect on NIR predicted 12 hr IVNDFD.

Key Words: NIR, IVNDFD, geographic effects

M91 Ethanol or amylase pretreatments affect estimates of in vitro NDF digestibility. A. L. Miller, J. P. Goeser, and D. K. Combs*, *University of Wisconsin, Madison.*

Our objective was to determine if pre-treating feed samples with ethanol or amylase affects estimates of NDF or in vitro NDF digestibility (iv NDFD). We also tested whether pre-rinsing Ankom F-57 fiber bags with