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**GRADUATE STUDENT COMPETITION:  
ADSA DAIRY FOODS DIVISION  
POSTER COMPETITION**

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**1129 (M112) The effect of native phospholipids on the flavor and flavor stability of bleached cheddar whey.** C. Park\*, and M. Drake, *Southeast Dairy Foods Research Center, NCSU, Raleigh, NC.*

Off-flavors due to processing of whey protein ingredients negatively influence consumer acceptance of ingredient applications. Previous research has demonstrated that bleaching of Cheddar whey increases lipid oxidation and off-flavors in liquid whey and resulting whey protein ingredients. Due to the high content of unsaturated fatty acids, native phospholipids have a high potential for lipid oxidation. The phospholipid to neutral lipid content ratio in fat free milk is reported to be significantly higher than in whole milk. The objective of this study was to determine the impact that native phospholipids in Cheddar whey have on the flavor and flavor stability of liquid whey. Liquid Cheddar whey was produced from whole milk, reduced fat milk, and fat free milk. The wheys were then fat separated to the same total fat content, pasteurized, bleached (250 ppm hydrogen peroxide) and stored at 4°C for up to 48 h. The wheys were sampled at 0 h, 24 h, and 48 h. The entire experiment was replicated 3 times. Flavor was analyzed by sensory and instrumental analyses. Phospholipids were quantified by UHPLC with an evaporative light scattering detector using hydrophilic interaction chromatography. Fatty acid profiles of the neutral and polar lipids were analyzed by FAME using GC-FID. All wheys increased in the lipid oxidation compounds pentanal, hexanal, heptanal, and nonanal and cardboard flavor from 0 h to 48 h ( $P < 0.05$ ). Liquid whey produced from fat free milk had higher concentrations of hexanal, pentanal, heptanal, and DMTS as well as increased cardboard flavor intensity after 24 or 48 h compared to other wheys ( $P < 0.05$ ). Phospholipids were significantly higher in the whey made from fat free milk compared to wheys made from whole or reduced fat milk ( $P < 0.05$ ). The polar lipid fraction contained higher concentrations of the unsaturated fatty acids 18:1, 18:2, 18:3, and 20:4 and lower concentrations of saturated fatty acids compared to the neutral lipid fraction. These results identify native phospholipids as a major source of off-flavors in liquid whey due to their unsaturated fatty acid profile and susceptibility to lipid oxidation.

**Key Words:** whey, phospholipids, flavor

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**1130 (M113) The effect of norbixin destruction or removal on flavor and functionality of 80% whey protein concentrate.** Y. Qiu\*, T. Smith, A. Foegeding, and M. Drake, *Southeast Dairy Foods Research Center, NCSU, Raleigh, NC.*

The residual annatto colorant (norbixin) in fluid cheddar cheese whey is bleached to provide a neutral-colored final product. Currently, chemical and enzymatic applications are utilized for bleaching liquid whey. These approaches bleach by free radical formation and can cause off-flavors due to lipid oxidation and protein degradation. The objective of this study was to compare three bleaching/norbixin removal methods: hydrogen peroxide (HP), lactoperoxidase (LP), and microfiltration (MF), and their effects on the flavor and functionality of 80% whey protein concentrate (WPC80). Cheddar cheese whey was manufactured from colored, pasteurized milk. The fluid whey was pasteurized and fat separated. Whey was subjected to 1 of 4 different treatments: Control (no bleaching; 50°C, 1h), HP (250 mg hydrogen peroxide/kg; 50°C, 1h), LP (20 mg hydrogen peroxide/kg; 50°C, 1h), or MF (microfiltration; 50°C, 1h). The treated whey was then ultrafiltered, diafiltered, and spray-dried to 80% whey protein concentrate (WPC80). The entire experiment was replicated three times. Proximate analysis, color, functionality, descriptive sensory and instrumental volatile analysis were conducted on WPC80. Norbixin was decreased by 50, 46 and 95% for MF, HP, and LP bleached WPC80 treatments, respectively ( $P < 0.05$ ). The HP and LP WPC80 had higher cardboard flavor and distinct cabbage flavor compared with the unbleached and MF WPC80. Volatile compound results were consistent with sensory results. The HP and LP WPC80 were higher in lipid oxidation compounds (especially heptanal, hexanal, pentanal, 1-hexen-3-one, 2-pentylfuran, octanal) compared to unbleached and MF WPC80. Protein solubility of WPC80 at various pH values was not different ( $P > 0.05$ ). All WPC80 had  $> 85\%$  solubility at the pH range evaluated. Gelation under all conditions showed similar trends in small strain viscoelastic properties ( $P > 0.05$ ). Based on bleaching efficacy, flavor and functionality results, MF may be a viable alternative to chemical or enzymatic bleaching of fluid whey.

**Key Words:** whey bleaching, functionality, flavor

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**1131 (M114) Storage and temperature effects on the solubility, Maillard browning, and sensory characteristics of milk protein concentrates.** T. Smith\*, R. Campbell, and M. Drake, *Southeast Dairy Foods Research Center, NCSU, Raleigh, NC.*

Milk protein concentrates (MPC) are a relatively young, increasingly important category of dairy ingredients. MPC production in the US increased by 38% between 2008 and 2012. MPC are a highly functional protein, however, solubility and a mild flavor across storage are required for success. The ob-

jective of this study was to determine the effect of storage time and temperature on the solubility, sensory characteristics, and Maillard browning of low (45%) and high (80%) protein MPC. MPC45 and 80 were manufactured in triplicate and stored at low (4°C), medium (25°C) and high (40°C) temperatures for 0, 1, 3, 6, and 12 mo. Solubility was evaluated by measurement of turbidity and protein at pH 7 before and after centrifugation. Maillard browning was determined by measuring furosine levels by high performance liquid chromatography (HPLC). Descriptive analysis and gas chromatography-mass spectrometry (GC-MS) were also performed to evaluate sensory and volatile compound characteristics. Solubility of MPC45 was higher than MPC80 ( $P < 0.05$ ), and a significant decrease in solubility in both MPC 45 and 80 occurred over time and as temperature increased ( $P < 0.05$ ). Furosine increased with storage time and temperature, and this change was higher for MPC80 than for MPC45 ( $P < 0.05$ ). MPC45 were characterized by sweet aromatic/milky and cardboard flavors while MPC 80 were characterized by lower sweet aromatic/milky and higher cardboard flavors, as well as distinct tortilla/grapey flavor. Cardboard, tortilla/grapey and animal flavors increased with storage time and temperature ( $P < 0.05$ ). Key volatile flavor compounds in MPC were o,p,m 2-amino-acetophenone (tortilla), 2 2-methyl-butanal (green/fruity), 1-octen-3-one (earthy/mushroom), and methional (potato brothy), and concentrations of these compounds increased with storage time and temperature. An understanding of storage and environmental effects on MPC lays the foundation for optimizing quality.

**Key Words:** milk protein, flavor, solubility

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**1132 (M115) The salt, pH and thermotolerance of a novel nonstarter lactic acid bacterium that might be associated with slit defect in ripened cheddar cheese.** F. Ortakci<sup>1</sup>, J. R. Broadbent<sup>1</sup>, C. J. Oberg<sup>1,2</sup>, and D. J. McMahon<sup>3</sup>, <sup>1</sup>*Dep. of Nutrition, Dietetics, and Food Sciences, Western Dairy Center, Utah State University, Logan*, <sup>2</sup>*Dep. of Microbiology, Weber State University, Ogden, UT*, <sup>3</sup>*Western Dairy Center, Utah State University, Logan*.

An obligate heterofermentative lactic acid bacteria, *Lactobacillus wasatchii* WDC04 (WDC04), isolated from an aged Cheddar cheese, was studied for salt, pH and thermotolerance. We investigated the pH and salt tolerance of WDC04 in MRS+1.5% Ribose (MRS-R) under conditions that mimic Cheddar cheese ripening. In addition, the thermotolerance of WDC04 was tested in 2% milk to estimate its pasteurization tolerance. WDC04 was inoculated in MRS-R at two different pH levels (5.2 or 6.5), each containing 0, 1, 2, 3, 4, or 5% salt-in-moisture (S/M) levels (w/w). Growth was monitored by OD<sub>600</sub> measurements every 8 h under anaerobic conditions at 23°C for 60 h. After 48 h, an OD<sub>600</sub> of 2.0 was reached in all media except for 5% S/M at pH 5.2 which had an OD<sub>600</sub> of

1.75. At pH 6.5, WDC04 growth rates were similar at S/M levels of 0, 1, 2, and 3% after 24 hr. Growth rates for 0, 1, and 2% S/M levels at pH 5.2 after 24 h were also similar, while growth rates at S/M levels of 4, and 5% at either pH were slower. Two different thermotolerance tests were performed. Milks containing  $\sim 7 \times 10^6$  CFU/ml WDC04, was heated at 72°C for 15 s followed by a cooling by placing in a water bath at 31°C (set temperature for cheesemaking) for 2 h. In the second heat treatment, WDC04 inoculated milks were heated at 63°C for 30 min with samples collected at 0, 15 and 30 min intervals followed by incubation at 31°C for 2 h. Samples were plated on MRS-R agar in triplicate. There was a 4-log reduction of WDC04 after the 72°C for 15 s heat treatment with  $9.2 \times 10^1$  CFU/ml after cooling to 31°C. However, there were no detectable colonies ( $< 10^2$  CFU/ml) when heated for 30 min at 63°C. This suggests that WDC04 is maybe sufficiently thermotolerant for some cells to survive HTST pasteurization but perhaps not LTLT pasteurization. It also appears that WDC04 is able to grow in the same environment that occurs during Cheddar cheese ripening, high S/M (2.5 to 4.5%) and low pH. The ability to survive pasteurization and grow under cheese ripening conditions allow WDC04 to be considered a NSLAB, placing it in a position to be involved with late gas production and slit defect in ripened Cheddar cheese.

**Key Words:** nonstarter lactic acid bacteria, salt, pH, and thermotolerance

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**1133 (M116) Role of protein interactions on microstructure and rheological properties of Greek-style yogurt.** G. H. Meletharayil<sup>1</sup>, H. A. Patel<sup>2</sup>, and S. G. Sutariya<sup>1</sup>, <sup>1</sup>*South Dakota State University, Brookings*, <sup>2</sup>*Dairy Science Dep., South Dakota State University, Brookings*.

Disposal of acid whey is a major concern for the manufacturers of Greek-style yogurt (GSY) because of its potential environmental impact. An alternate way of preparing GSY is to eliminate de-whey step by using functional milk proteins and manipulating process-induced protein interactions. The objective of this study was to investigate the influence of micellar and non-micellar casein to globular proteins ratios on the properties of GSY prepared using this alternate process. GSY (7.5% w/w proteins, 15% w/w total solids) were prepared with either milk protein concentrates (MPC) as a source of micellar casein or carbon-dioxide treated milk protein concentrate (T-MPC) as a source of non-micellar caseins. Whey protein concentrates (WPC) and de-proteinized whey was used to adjust the globular protein and total solids level respectively. The casein to whey ratio was adjusted to 4:1, 2.3:1 and 1.5:1. All samples were pre-adjusted to pH 6.5 before heating at 90°C/10 min. Acid gels were prepared using Glucono- $\delta$ -lactone to obtain final pH 4.4 after 4 h of incubation at 30°C. The soluble (serum) phases obtained by centrifugation of heated and unheated milk samples at 25,000g/1h were characterized using sodium dodecyl

sulfate polyacrylamide gel electrophoresis (SDS-PAGE). Microstructure and rheological characterization of acid gels were performed using confocal laser scanning microscopy (CLSM) in the fluorescence mode and small amplitude oscillatory rheology (1% strain, 0.1 Hz frequency), respectively. ANOVA was used to test the results and statistical significance at  $P < 0.05$  was determined, using the statistical software SAS. SDS-PAGE results indicated significantly higher proportion ( $P < 0.05$ ) of soluble disulfide-linked aggregates in serum phase of heated milks prepared from T-MPC. Gels prepared using T-MPC as a source of non-micellar casein had significantly higher ( $P < 0.05$ ) elastic modulus ( $G'$ ) (e.g.,  $1.12 \times 10^3$  Pa) compared to gels prepared using MPC (e.g.,  $6.59 \times 10^2$  Pa). Acid gels containing T-MPC in different ratios with globular proteins had a significantly higher ( $P < 0.05$ )  $G'$  than acid gels prepared with MPC and WPC. CLSM images revealed that T-MPC gels had smaller, well-connected aggregates with uniform, homogenous pore sizes, which strongly supported the results of rheological characterization. It can be concluded that T-MPC as a source of non-micellar casein is an ideal ingredient to alter the ratio of non-micellar to globular protein ratio and thereby increase the gel strength of Greek-style yogurts. This invention is patent pending and can be used to produce protein structures having different gels strengths suitable for commercial applications.

**Key Words:** Greek-style yogurt, carbon dioxide, rheology

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**1134 (M117) Assessment of consumer perceptions and preferences regarding fluid milk at the beginning and end of printed code date.** M. E. Paterson\*, *Iowa State University, Ames.*

The objective of this study is to understand consumers' expectations and actual sensory perceptions about fluid milk at the beginning and end of code. Eleven sessions were carried out ( $n = 103$ ). Sessions began with explanation of consent form and the experiment process, then panelists filled out a survey about demographics and milk purchasing and consumption behaviors. Consumers were blindly served two pairs of milk samples (2% within 2 to 3 days of production (fresh) and 2% with 2 to 3 days to end of code (end); skim (fresh and end) and asked to indicate preference and the level of acceptability for each sample using a seven-point scale. All samples tasted by consumers were simultaneously evaluated by a panel of eight judges who were trained to evaluate milk quality attributes on a 15-cm line scale. All milk was from the same source, processed on the same timeline for each session; milk was stored in the warehouse until transport to the sessions. Eighty-five participants (82%) indicated they check for the farthest out code date more than half the time they shop. However, upon tasting, consumers did not have a preference for 2% fresh milk over 2% end, or for skim fresh over skim end ( $P > 0.05$ ). These findings were in agreement with their acceptability scores, which were 4.7 for skim fresh, 4.6 for skim end, 5.1 for 2% fresh and 5.1 for

2% end ( $P > 0.05$ ). Trained panelists did not detect a difference in lacks freshness flavor in skim fresh (1.9 cm) or skim end (1.3 cm). Trained panelists also did not detect a difference in cooked, feed, flat, foreign or oxidized flavors for 2% or skim milk samples. Trained panelists detected a significant difference in lacks freshness flavor of 2% fresh (2.3 cm) and 2% end (0.3 cm) ( $P < 0.05$ ). When the one off-flavored batch of 2% fresh milk was removed from analysis, trained panelists could not distinguish a difference in lacks freshness between 2% fresh (1.0 cm) and 2% end (0.4 cm) ( $P > 0.05$ ). After tasting and receiving an educational message about the meaning of code dates, 83% of consumers stated the information would impact their future purchases. These results confirm that although many consumers go out of their way to buy the freshest milk, they cannot distinguish fresh milk from milk at the end of code. Additional research must be conducted to confirm impact of educational messages about code date on purchasing behavior.

**Key Words:** milk, sensory, code-date

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**1135 (M118) Performance of cross-linked and calcium-reduced milk protein concentrate ingredients in model high-protein nutrition bars.** J. C. Banach<sup>1</sup>, S. Clark<sup>1</sup>, L. Metzger<sup>2</sup>, and B. P. Lamsal<sup>1</sup>, <sup>1</sup>*Iowa State University, Ames*, <sup>2</sup>*Midwest Dairy Foods Research Center, South Dakota State University, Brookings.*

Milk protein concentrate (MPC) and micellar casein concentrate (MCC) have the potential to be the major protein source in high-protein nutrition bars. However, MPCs are known to produce bars that harden too quickly and also result in a crumbly texture. Less is known about the performance of MCC in bars. The objective of this study was to evaluate the performance of transglutaminase (TGase) cross-linked MPC and MCC, and calcium-reduced MPC in high-protein bars. MPC and MCC retentate were treated with TGase at 0.3 units (low; MPC-L and MCC-L) and 3.0 units (high; MPC-H and MCC-H) per g protein. Controls (MPC-C and MCC-C) were not treated with the enzyme. Separately, CO<sub>2</sub> was injected during MPC ultrafiltration to produce calcium-reduced MPC. Retentates were spray-dried to produce ingredients with 72 to 78% protein. Bars were prepared by hand pressing dough containing 30% protein, 8.9% HFCS, 20.7% glycerol, 11% maltitol syrup, and 17.6% palm oil into molds (ID = 21 mm, H = 13 mm) and water activity sample cups. Model bars were stored at 32°C for up to 42 days, during which texture, color, water activity, moisture content, and pH were evaluated. Bar hardness and fracturability were determined with compression, after which a sieve analysis was used to evaluate crumbliness. Data, separated via Tukey's adjusted p-value ( $P < 0.05$ ), are the average of two bar preparations. On day 0, no statistical difference was detected for fracturability and hardness between bars ( $P < 0.05$ ) and crumbliness reduced when cross-linked ingredients were compared with their respective controls ( $P < 0.05$ ). Throughout storage, bars formulated with a commercially

produced native MPC80 were less crumbly and maintained a larger geometric mean diameter after compression. A high-level of TGase helped maintain cohesiveness in MPC-H and MCC-H compared with their controls through day 16 and day 28, respectively ( $P < 0.05$ ). Sample moisture content on the day of preparation was the same ( $P > 0.05$ ), whereas water activity was different ( $P < 0.05$ ). A significant increase ( $P < 0.05$ ) in water activity was seen ~24 h after preparation, but after the initial increase it remained stable within each batch. Moisture content and pH remained fairly constant during storage, while visual color change was apparent within the samples. TGase treated and calcium-reduced milk protein ingredients have altered performance in high-protein nutrition bars, which potentially could lead to improved commercial feasibility.

**Key Words:** milk protein concentrate, micellar casein concentrate, protein bar

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**1136 (M119) The effects of post-exercise consumption of a kefir beverage on performance and recovery during intensive endurance training.** K. V. O'Brien\*, Louisiana State University, Baton Rouge.

This study was designed to determine whether kefir accentuates the positive health benefits assessed by measures in fitness and/or body composition, as a measure of cardiovascular disease risk as well as the biomarker c-reactive protein (CRP). Thirty-eight adult males and females aged 18 to 24 yr were assigned to one of four groups: 1) endurance training + control beverage (ETC), 2) endurance training + kefir beverage (ETK), 3) active control + control beverage (ACC) or 4) active control + kefir beverage (ACK). The E groups (ETK and ETC) completed 15 weeks of structured endurance training. The AC groups (ACK and ACC) maintained their usual exercise routine. Additionally, each group was assigned to either a kefir (ETK and ACK) or a calorie/macronutrient matched placebo (ETC and ACC) beverage that was consumed twice per week. The kefir beverage and the control beverage were developed and manufactured in the Louisiana State University Creamery and were identical in ingredients used with the only difference being the fermentation of the milk used in the kefir beverage. Pre/post measures included: body mass and composition, waist hip ratio and 1.5 mile run. Serum CRP was measured using an ELISA (Alpco Diagnostics, Salem, NH). A MANOVA was used to identify significant interactions and significance was set at  $P < 0.05$ . RESULTS: There was a significant time x training group interaction ( $P = 0.0124$ ) with the E groups (ETK and ETC) experiencing an average of 4.11% improvement in 1.5 mile time. There were no significance interactions among groups with respect to all other outcome variable with the exception of serum CRP. Serum CRP increased over time ( $P = 0.103$ ). However, there was also a trend for a time x kefir effect ( $P = 0.0778$ ). The ETK and ACK groups experienced less (21.18%, 5.45%) of an increase when compared to the ETC and ACC (22.36%, 64.71%). The

endurance training was effective in improving 1.5 mile times and kefir supplementation may have been a factor in attenuating the increase in CRP that was observed over the course of the intervention period. This preliminary study suggests that kefir may be involved in improving the risk profile for cardiovascular disease as defined by CRP.

**Key Words:** kefir, endurance training, cardiovascular disease

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**1137 (M120) Manufacture of high protein yogurts with low-Ca MPC.** A. Kommineni<sup>\*1</sup>, C. Marella<sup>2</sup>, A. C. Biswas<sup>1</sup>, and L. Metzger<sup>3</sup>, <sup>1</sup>Dairy Science Dep., South Dakota State University, Brookings, <sup>2</sup>Dairy Science Dep., California Polytechnic State University, San Luis Obispo, <sup>3</sup>Midwest Dairy Foods Research Center, South Dakota State University, Brookings.

The purpose of this study was to evaluate and compare the effects of high protein yogurt produced with calcium reduced milk protein concentrate (low-Ca-MPC) and regular milk protein concentrate (MPC). The yogurt properties studied were viscosity, syneresis and quantity of acidifying agent required for the yogurt. Reduced Ca MPC was manufactured from ultra and dia-filtration process of skim milk that was injected with CO<sub>2</sub> prior to and during the ultra-filtration process. Three different high protein yogurt formulations containing 8% protein were prepared with regular MPC (T-1), low-Ca-MPC with the pH neutralized to 6.7 (T-2) and low-C-MPC at pH 5.9 (T-3). The MPC was the primary source of protein contributing approximately 97% in the all the formulations and the remaining 3% was coming from de-proteinized whey which was used for balancing the lactose content of each formula. All three treatments were manufactured in triplicate from three replicates of low-Ca MPC and regular MPC. Each yogurt formulation was heat treated to 93°C, held for 6 min and then cooled to 45°C. The formulations were then acidified with Glucono-delta-Lactone (GDL) at 45°C and then incubated for 2 hrs. The amount GDL required to reach a pH of 4.6 for 30g of yogurt was 0.7g, 0.6g and 0.55g respectively for T-1, T-2 and T-3. The T-3 yogurt formulation required 21% less GDL to reach the same pH due to the lower initial pH of the T-3 formulation. The T-2 yogurt formulation also used 14% less GDL than T-1 even though the initial pH of both the formulations was same. The undisturbed low-C yogurt formulations (T-2 & T-3) had approximately 40% higher viscosity than the control T-1 formulations. However after stirring all yogurt formulations had similar viscosities. The control yogurt formulation T-1 also had 10% higher syneresis than T-2 and T-3 yogurts. These results indicate that, the use of low-Ca MPC reduces both syneresis and amount of acid required to ferment yogurt.

**Key Words:** low-Ca MPC, calcium, yogurt, MPC

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**1138 (M121) Effect of titanium dioxide, annatto, and homogenisation on the translucency of reduced-fat cheddar cheese.**

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The appearance of cheese is greatly influenced by its composition. Low fat content is usually related to an increase in translucency. Many alternatives have been proposed to modify translucency of cheese, such as the addition of ingredients or through the modification of manufacturing protocols. Colorimetric methods have been used extensively to determine the degree of translucency. The Kubelka-Munk index (K/S) measures the relationship between the reflectance of a thin layer of sample above black and white backgrounds, which indicates the reflectance of a sample of infinite thickness. A method based on the measurement of L\* values and the application of K/S was proposed to investigate the effect of titanium dioxide, annatto and homogenisation on the translucency of reduced-fat Cheddar cheese during ripening. Three reduced-fat Cheddar cheeses were manufactured in parallel experiments. For titanium dioxide, levels of 0, 20 or 40 g of TiO<sub>2</sub>/100 kg were

added to cheesemilks. For annatto, levels of 0, 8.25 and 16.50 ml/100 kg were added to cheesemilks. Cheesemilks were also homogenised at 0, 10 or 20 MPa, using a two stage homogeniser (4:1 ratio) at 40°C. L\* values and K/S were obtained with a colorimeter at 20°C for all experimental cheeses during six months of ripening. A high correlation was observed between K/S and L\* values ( $r > 0.90$ ). Titanium dioxide, annatto and homogenisation significantly ( $P < 0.05$ ) modified K/S values compared to the control. Ripening significantly increased translucency (i.e., reduced K/S) for all treatments ( $P < 0.05$ ). An increase in K/S was observed when TiO<sub>2</sub> was added ( $P < 0.05$ ). A reduction of the K/S value was observed following addition of annatto ( $P < 0.05$ ). An increase of K/S was observed when milk was homogenised ( $P < 0.05$ ). However, no differences in K/S values were found between homogenisation pressures of 10 and 20 MPa. These results suggest that K/S is a useful tool to determine changes in the translucency of Cheddar cheese. Addition of titanium dioxide and homogenisation reduced translucency. On the other hand, the use of annatto increased translucency in experimental cheeses.

**Key Words:** translucency, L\* value, Kubelka-Munk