

quality regardless of light and intensity.

Key Words: light-emitting diode, milk, packaging

0707 Efficient removal of spores from skim milk using microfiltration: Spore size and surface property considerations.

E. R. Griep*, Y. Cheng, and C. I. Moraru, *Cornell University, Ithaca, NY.*

Presence of spores in milk can cause numerous quality and shelf-life issues for dairy products. Microfiltration (MF) using a 1.4- μm pore size can effectively remove vegetative bacterial cells from milk and is used in commercial applications. However, this pore size may not be equally effective in spore removal. The objective of this study was to determine the effectiveness of MF using 1.4- and 1.2- μm pore sizes for removing spores of *Bacillus licheniformis* (BL) and *Geobacillus* spp. (GEO) from skim milk. Cell size of both spores and vegetative cells was evaluated by scanning electron microscopy, surface charge by zeta potential analysis, and surface hydrophobicity by contact angle measurements, in triplicate. Commercially pasteurized skim milk was inoculated in a sterilized feed tank with a spore suspension, at about 10^6 spores/mL, and then treated by MF (in triplicate) using ceramic Isoflux membranes at 6°C, cross-flow velocity of 4.1 m/s, and transmembrane pressure between 69 and 74 kPa. Total aerobic plate count and spore count of the permeate were conducted. An unpaired *t* test was used to determine significant differences between samples at a $P < 0.05$ significance level. Vegetative cell length ranged between 2.40 and 3.82 μm and the width ranged between 0.39 and 0.64 μm . Spores were shorter and wider, averaging 1.39 to 1.58 μm in length and 0.63 to 0.88 μm in width, therefore having a higher probability to pass through a 1.4- μm membrane. Indeed, for BL (1.39- μm length \times 0.63- μm width) an average spore reduction of only 2.17 log was achieved by 1.4- μm pore size. For the 1.2- μm membrane, a 4.57 log reduction was achieved. For GEO spores, their larger spore size (1.58- μm length by 0.81- μm width) allowed a practically complete removal using both pore sizes (spore counts in permeate below the detection limit). The surface properties of BL and GEO indicated that they may interact differently with the membrane. Both spore species and the ceramic membrane had negative surface charge at the milk pH, indicating slight electrostatic repulsion between them. *Geobacillus* spp. spores were hydrophilic, whereas BL spores were slightly hydrophobic; the ceramic membrane surface changes from hydrophilic (in unfouled state) to hydrophobic after adsorption of caseins during MF. Consequently, BL spores may experience slight attractive force to the membrane through hydrophobic interactions, which will facilitate their passage through the membrane. A good understanding of all factors that affect the removal of spores using MF can lead to the production of milk with lower spore count, higher quality, and increased shelf life.

Key Words: microfiltration, skim milk, spore removal

ADSA DAIRY FOODS GRADUATE STUDENT POSTER COMPETITION

0708 Unit operations before and during spray drying influence the flavor of milk protein concentrate and whole milk powder.

C. Park* and M. Drake, *Southeast Dairy Foods Research Center, North Carolina State University, Raleigh.*

Flavor is a limiting factor in the application and shelf life of dried dairy ingredients. Many off-flavors are caused during ingredient manufacture, which carry through into ingredient applications and decrease consumer acceptance. The objective of this research was to investigate the effect of spray drying parameters on the flavor of milk protein concentrate (MPC) and whole milk powder (WMP). Liquid MPC70 was produced from pasteurized skim milk by ultrafiltration/diafiltration to 19% solids (wt/wt) and evaporated to 32% solids (wt/wt). Spray drying was performed with varying inlet temperature (160, 210, or 260°C) and feed solids concentration (12, 22, or 32%). Whole milk powder was produced from standardized pasteurized whole milk that was evaporated to 50% solids (wt/wt), homogenized in two stages with varying pressures (0/0, 55.1/13.8, 110/27.6, or 165/41.4 bar), and spray dried. Whole milk powder was evaluated at 0, 3, and 6 mo storage at 22°C. Sensory properties were evaluated by descriptive sensory analysis and volatile compounds were evaluated by headspace extraction (SPME) with gas chromatography mass spectrometry. Fat globule size in condensed whole milk and particle size of powders were measured by laser diffraction. Surface free fat of WMP was measured by solvent extraction. Furosine in MPC70 was analyzed by UPLC-MS. Spray drying of MPC70 at 160°C increased cardboard flavor and volatile lipid oxidation products and decreased sweet aromatic flavor and furosine concentration compared with 210 or 260°C ($P < 0.05$). Solids concentration during drying had no effect on furosine concentration ($P > 0.05$). Decreasing feed solids concentration decreased sweet aromatic flavor and increased cardboard flavor and volatile lipid oxidation products ($P < 0.05$). Increased homogenization pressure decreased cardboard flavor, volatile lipid oxidation compound concentrations, fat globule size in condensed milk, and surface free fat in WMP ($P < 0.05$). Surface free fat in powders increased cardboard flavor and lipid oxidation. These results indicate that off-flavors are decreased with increasing feed solids concentration and inlet temperature in MPC70 and with increased homogenization pressures in WMP. To decrease off-flavor intensities in WMP and MPC70, manufacturers should evaluate these parameters during ingredient manufacture.

Key Words: milk proteins, whole milk powder, unit operations

0709 The effect of bleaching agents on the degradation of vitamins and carotenoids in WPC80.

M. A. Stout*¹, C. Park², and M. Drake², ¹North Carolina State University, Raleigh, ²Southeast Dairy Foods Research Center, North Carolina State University, Raleigh.

Previous research has demonstrated that chemical or enzymatic bleaching impact flavor and functionality of whey proteins. The role of bleaching on vitamin and carotenoid degradation is unknown. The objective of this study was to determine the effects of bleaching with hydrogen peroxide (HP), benzoyl peroxide (BP), or native lactoperoxidase (LP) on vitamin and carotenoid degradation. The role of an alternative colorant on whey protein vitamin profile was also evaluated. Colored cheddar whey (15 mL/454 L milk) was manufactured, pasteurized, and fat separated and then assigned to 250 ppm HP, 25 ppm BP, or 20 ppm HP (LP system) at 50°C for 1 h. An unbleached control (Con) as well as whey from cheese milk with an alternative colorant (AltC) were also included. The Con and AltC were also heated to 50°C for 1 h. Wheys were concentrated to 80% protein by ultrafiltration and spray dried. The experiment was replicated in triplicate. Vitamin, norbixin, and carotenoid contents were determined by HPLC and volatiles by gas chromatography mass spectrometry. A trained panel documented sensory properties of the rehydrated WPC80. Volatile compound and sensory results were consistent with previous studies on bleached and unbleached whey proteins. WPC80 that were chemically or enzymatically bleached had decreased retinol, β -carotene, ascorbic acid, α -carotene, thiamine, lutein, and α -tocopherol ($P < 0.05$) compared with Con WPC80 or AltC WPC80. Benzoyl peroxide WPC80 contained less of these compounds than HP or LP WPC80 ($P < 0.05$). Riboflavin, pantothenic acid, pyridoxine, and nicotinic acid concentrations were not impacted by bleaching ($P > 0.05$). Alternative colorant WPC80 contained 6% more β -carotene than Con WPC80 ($P < 0.05$). Bleaching to remove norbixin decreases fat soluble vitamin and carotenoid concentrations in the final spray dried whey protein.

Key Words: bleaching, vitamin degradation, alternative colorant

0710 Characterization of flavor and functional properties of liquid and dried WPC 80, WPI, MPC 85, and micellar casein concentrates.

B. Carter*¹, H. Patel¹, D. M. Barbano², and M. Drake³, ¹North Carolina State University, Raleigh, ²Cornell University, Department of Food Science, Northeast Dairy Foods Research Center, Ithaca, NY, ³Southeast Dairy Foods Research Center, North Carolina State University, Raleigh.

Traditionally most protein ingredients are sold as a powder due to its ease to transport and longer shelf life. Many

high-protein powder ingredients such as MPC 85 and micellar casein concentrate (MCC) have poor rehydration properties (e.g., solubility), which might be a limiting factor for using these ingredients in end applications. Previous research suggested that the spray drying may have some adverse effects on flavor and functional properties of dried ingredients. Moreover, spray drying is the costly unit operation in the manufacture of protein ingredients. Considering this, manufacturers of dried protein ingredients are considering an option to manufacture liquid retentate, which will not only save the cost of spray drying but may provide improved flavor and functional properties. The objective of this study was to determine what effect, if any, spray drying has on the flavor and functionality of high-protein ingredients. Liquid and dried protein ingredients (WPC 80, WPI, MPC 80, and MCC) were manufactured from the same lot of milk at the North Carolina State University pilot plant. These ingredients were characterized using native PAGE, particle size, and calcium activity, and functionality differences were evaluated by measurement of foam stability, protein solubility, and heat stability. Protein solubility was measured at pH 7 before and after centrifugation by micro-bicinchoninic acid assay (micro-BCA), and heat stability by heating at 90°C for 0, 10, 20, and 30 min followed by micro-BCA and turbidity loss. Flavor was evaluated by descriptive analysis, and volatile compound analysis was conducted by GC-MS to identify key flavor differences between the liquid and spray dried protein ingredients. No differences were detected in solubility and heat stability between liquids and powders ($P > 0.05$). WPC 80 (liquid or spray dried) did not produce a foam; powder WPI produced a more stable foam as opposed to the liquid, but with milk proteins, the liquids produced a more stable foam ($P < 0.05$). This result is likely due to the particle size difference between liquid and powder being much greater in milk proteins compared with whey proteins ($P < 0.05$). All powders had higher aroma intensity and cooked flavors compared with liquids ($P < 0.05$). Powder proteins also had low but distinct cardboard flavor concurrent with higher volatile aldehydes compared with liquids. An understanding of how spray drying effects both flavor and functionality will help producers better use the ingredients they have available to them.

Key Words: milk proteins, whey proteins, flavor, functionality

0711 Effect of milk protein concentrate (MPC 80) quality on susceptibility to fouling during thermal processing. G. Gandhi*¹ and J. K. Amamcharla², ¹Department of Animal Sciences and Industry/ Food Sciences Institute, Kansas State University, Manhattan, ²Food Science Institute, Animal Sciences and Industry, Kansas State University, Manhattan.

Milk protein concentrate (MPC) is incorporated into wide range of dairy beverages to improve the functional, nutritional,

and sensory properties. Various factors such as drying conditions, composition, storage, and dissolution conditions affect the overall functional characteristics of MPC. To be used for its intended purpose, it is essential to study the functional properties such as solubility, dissolution, and fouling characteristics of MPC. Fouling of the stainless steel (SS) surfaces during thermal processing of milk is a major problem in the dairy industry. It is important to understand the composition and structure of the fouling layer to minimize the fouling of the processing equipment. The objective of the present study was to understand the effect of MPC solubility on its susceptibility to initiate fouling on SS surfaces during thermal processing. Milk protein concentrate powder with 80% protein content was obtained from a commercial manufacturer, divided into two lots. To create powders with different solubility characteristics, the first lot was stored at 25°C and the second lot at 40°C for 2 wk. Immediately after the storage, the powder solubility characteristics were monitored using focus beam reflectance measurement technique and solubility index. As expected, the MPC stored at 40°C showed the slow dispersion rate of particles when compared with the powder stored at 25°C, indicating poor solubility characteristics. Fouling characteristics were studied using a custom-build benchtop heat exchanger (bPHE). The bPHE was designed to accommodate two SS coupons with 1" by 1" dimensions. The MPC was reconstituted to 10% (wt/wt) solution and pumped for 2 h through bPHE to an average outlet temperature of 71.9°C. Subsequently, the SS coupons were removed from the bPHE and fouled layer was characterized using weight of fouling, scanning electron microscopy, confocal laser scanning microscopy, energy dispersive X-ray spectroscopy, and SDS-PAGE. The average foulant accumulated over the SS coupons by the poor quality powder (stored 40°C) and the good quality (stored at 25°C) powder was 0.171 ± 0.031 and 0.093 ± 0.019 g, respectively. Storing the powders at 40°C significantly ($P < 0.05$) increased the amount of fouling on SS coupons. Microscopic investigations revealed the heterogeneity of the fouling layer with the discrete distribution of lipids and proteins with uniform calcium distribution. Therefore, the study will be helpful in designing effective strategies to reduce fouling during processing of high-protein dairy beverages.

Key Words: fouling, milk protein concentrate, plate heat exchanger

0712 Oxygen barrier and light interference packaging properties for controlling light-induced oxidation in milk. H. Potts*, S. Duncan, M. L. Johnson, S. F. O'Keefe, J. E. Marcy, and K. Mallikarjunan, *Virginia Tech, Blacksburg.*

A recent shift to more energy efficient light-emitting diode (LED) lights in retail dairy cases has occurred, but the effects of LED light on fluid milk in retail conditions are not known. Our objective was to determine the efficacy of polyethylene

terephthalate (PET) packaging at preventing light-induced oxidation in 2% milk under LED and fluorescent retail light. Light interference effects were studied in combination with the oxygen barrier effects of PET. The extent of oxidation in 2% milk packaged in PET bottles (2 L; average wall thickness: 0.33 mm; treatments: clear with UV barrier and 2.1, 4.0, and 6.6% titanium dioxide [TiO_2]) under fluorescent and LED retail light up to 72 h was studied. Two control packages (clear PET = full light exposure and PET wrapped with foil and plastic = no light exposure) were used for comparison, creating a total of six packaging experimental treatments. Chemical measures of oxidation ($\alpha = 0.05$; ANOVA) included formation of secondary lipid oxidation products, riboflavin degradation, and headspace volatiles analysis by an electronic nose. Volatile analysis compared electronic nose smell-prints by canonical discrimination analysis. Sensory evaluation of milk (triangle test, 3 replications) compared milk from experimental packages to light-protected control milk for similarity ($\beta = 0.05$) and to light-exposed control milk for differences ($\alpha = 0.05$). Polyethylene terephthalate with 6.6% TiO_2 was an effective package for protecting fluid milk sensory quality for up to 8 h under LED light (936 ± 136 lux) but only for 4 h under fluorescent light ($1,447 \pm 1,072$ lux). Polyethylene terephthalate with 4% or less TiO_2 could not effectively protect milk flavor from light-induced changes through 4 h fluorescent or LED light exposure. Milk stored in PET packages retained 0.90 mg/L or higher riboflavin content over 72 h retail light exposure. Electronic nose technology differentiated ($P < 0.05$) volatile profiles among fresh milk with no light exposure and milk that remained under retail lights for 8 h or more, indirectly supporting the changes in sensory quality. The results conclude that LED light is less detrimental to milk quality than fluorescent light and higher levels of TiO_2 in PET packages were more effective at preventing light-induced oxidation in 2% milk.

Key Words: milk, oxidation, sensory

0713 Use of fluorescence-based Amaltheys analyzer for studying effect of pH and heat on whey protein interactions in reconstituted milk protein concentrate. K. Sajith Babu*, Z. Liu, and J. K. Amamcharla, *Food Science Institute, Animal Sciences and Industry, Kansas State University, Manhattan.*

Milk protein concentrates (MPC) are complete proteins that contain both casein and whey proteins in the same ratio as in milk. In comparison with skim milk powder or whole milk powder, MPC are higher in protein and lower in lactose and minerals. The objective of our study was to use fluorescence-based Amaltheys analyzer to study the effect of pH and heat on whey protein interactions in reconstituted milk protein concentrate. MPC85 from two different lots of same manufacturer was reconstituted into 5% solution and the pH values were adjusted between 5.5 and 7 and heated (70, 80, and 90°C) up to 20 min. The level of whey protein denaturation

and protein associations were examined for each milk sample. The level of native whey protein in the no-heat and heat-treated milk samples were determined using native PAGE. The whey protein nitrogen index (WPNI) and the FAST (fluorescence of advanced Maillard products and soluble tryptophan) index were measured by fluorescence-based Amaltheys analyzer (Spectralys Innovation, Romainville, France) and was compared with the native PAGE results. The results based on the WPNI, gives an indirect indication of the denaturation and aggregation of whey proteins. The level of denatured whey proteins was significantly dependent on the pH and thermal treatment, with high levels of interactions at pH 5.5 and low levels of changes at pH 7. Reconstituted MPC85 with no heat treatment (WPNI 1.84 ± 0.01 mg WPN·g⁻¹ powder at pH 7 and WPNI 1.69 ± 0.01 mg WPN·g⁻¹ powder at pH 5.5, respectively) retained most of the whey proteins in the native state. In contrast, the reconstituted MPC85 at 90°C for 20 min (WPNI 0.44 ± 0.01 mg WPN·g⁻¹ powder at pH 7 and WPNI 0.21 ± 0.01 mg WPN·g⁻¹ powder at pH 5.5) contained a comparatively small proportion of native whey proteins, although some α -lactalbumin was still present (shown in native PAGE). The degree of denaturation of β -lactoglobulin appeared to be crucial and could be related to the WPNI and FAST index. The amount of whey protein interactions increased with increase in temperature and decrease of pH. The results in this study indicate that the changes in whey protein induced by the heat treatment of reconstituted milk protein concentrate were affected by pH and thermal treatment time, and the Amaltheys analyzer was found to be a simple and rapid instrument for studying these interactions.

Key Words: milk protein concentrate, whey protein nitrogen index, protein denaturation and aggregation

0714 Use of ozonated water in removing *Bacillus cereus* biofilms from the dairy membranes.

R. Henderson*¹, G. Gandhi¹, N. Sevart¹, S. Gragg², R. Phebus¹, and J. K. Amamcharla³, ¹Department of Animal Sciences and Industry/Food Sciences Institute, Kansas State University, Manhattan, ²Department of Animal Sciences and Industry/Food Sciences Institute, Kansas State University, Olathe, ³Food Science Institute, Animal Sciences and Industry, Kansas State University, Manhattan.

Fouling of dairy membranes is a major problem and leads to biofilm development. This causes reduction in membrane performance and leads to premature replacement of membranes. Ozone is a potent bactericidal agent used in diverse applications. The objective of this study evaluates the efficiency of ozonated water on removal of biofilms from dairy processing membranes. To generate aqueous ozone, an ozone generator system supplied by CleanCore Technologies Inc. (Omaha, NE) was used. The system consists of six ozone generators connected to an injector that mixes ozone with water. It also

houses a dissolved ozone monitor and oxidation–reduction potential sensor. The half-life of ozone in ozonated water was evaluated using a factorial design with pH (2, 4, and 7) and temperature (5, 10, and 20°C) as independent factors with two replications. Reduction in ozone concentration in ozonated water was monitored at regular intervals until the final concentration dropped below 1.0 ppm. First order rate constants (k) were calculated using a first order decay model and ozone half-life was then calculated. Temperature and pH and their interaction significantly ($P < 0.05$) affected half-life. Maximum ozone half-life was achieved with pH 4 solution at 10°C, with an average half-life of 478 min; therefore, these conditions were used to evaluate biofilm removal from dairy membranes. Ultrafiltration of skim milk was performed in a bench-top plate and frame system. Flat sheet polyethersulfone (PES) membranes (Hannifin Corp., Oxnard, CA) were fouled during 5x ultrafiltration of pasteurized skim milk. Fouled PES membranes were submerged in Luria broth inoculated with *Bacillus cereus* (ATCC 10987) and incubated for 48 h at 37°C to promote biofilm formation. Biofouled membranes were installed in the plate and frame system and exposed to ozonated water for 6 min in recirculation mode. Subsequently, residual biofilm was removed by scraping a 6.45 cm² of the membrane, transferred into 10 mL of 0.1% peptone water, and vortexed. Serial dilutions were plated onto mannitol egg yolk polymyxin agar, and plates were incubated at 37°C for 24 h to quantify viable *B. cereus* populations, which were compared with corresponding population levels on nontreated control membranes. The *B. cereus* biofilms grown on fouled PES dairy membranes and treated with ozonated water under the described conditions were reduced by an average of 1.0 log cfu/cm². Data suggests ozonated water has potential as an effective and environmentally friendly means for removing biofilms from dairy membrane systems.

Key Words: ozonated water, ultrafiltration membranes, biofilms

0715 Development of a benchtop method to polymerize lactose to soluble fiber. A. F. Kuechel* and

T. C. Schoenfuss, *University of Minnesota, Department of Food Science and Nutrition, St. Paul.*

Twin-screw extrusion is used to polymerize lactose and glucose to an oligosaccharide, poly lactose. However, previous research in our lab demonstrated that the lactose in permeate and acid whey could not be polymerized using our standard method. There is a lack of understanding for what inhibits the polymerization reaction; the citric acid content and extruder feed rate have been researched, yet chemical properties such as moisture and mineral contents have not. The objective of this study was to develop a benchtop method for polymerization using a CEM Mars 6 microwave reaction system so that inhibition factors could be identified before scaling up to the extruder. A heating profile needed to be established that

would consistently polymerize a blend of citric acid (6%), glucose (20%), and lactose (74%). Seven-gram samples of the sugar acid blends were added to 8 Teflon MarsXpress vessels. The set temperature, ramp time, and hold time were varied to melt the powder blend and achieve polymerization, without reaching decomposition. All vessels were continuously monitored for temperature during the reaction via an infrared thermometer. The reacted samples were cooled and dissolved in water, passed through ion-exchange resins, and then separated and detected by HPLC-ELSD. An initial heating profile with a 5-min ramp time to a 180°C target temperature imitated extrusion conditions known to result in polymerization. Even though polymerization was observed with this heating profile, the reactants did not reach the target temperature and temperature variability between vessels occurred. These challenges led to modifications of the heating profile including an increase in the ramp time (15 min) and a reduction in the temperature (140°C). Uneven heating was still a challenge so the formula was modified by adding a small amount of water (<1% wt/wt) to increase dipole rotation due to the microwave energy. The inclusion of a polar solvent resulted in consistent, even heating. Product resulting from the lower-temperature, longer-time heating profile demonstrated successful polymerization. The elevated pressure in the microwave reaction system, when compared with the open extrusion system, allowed for polymerization at a lower temperature. This benchtop polymerization method allows for experimentation of numerous formulas and the identification of inhibitors. Understanding these factors for permeate or acid whey will allow for polymerization into a value added ingredient, soluble fiber.

Key Words: lactose, polymerization, microwave

0716 Effect of microencapsulated iron salts on cheddar cheese divalent cation balance and composition.

A. Arce* and Z. Ustunol, *Michigan State University, East Lansing.*

Milk is considered an important source of macro- and micro-nutrients but naturally low in iron content. Cheese and other dairy products had been fortified with iron with low success due to negative changes in composition and organoleptic attributes. There is limited information about using microencapsulation of iron compounds in dairy products. Minerals have the ability to displace one another in any system; consequently, it is expected that encapsulation will avoid divalent cation displacement within the cheese matrix. The objective of this study was to analyze divalent cation balance in fortified cheddar cheese with microencapsulated ferrous sulfate. Furthermore, proximate analysis was done to provide more information about any compositional changes after fortification. Cheddar cheese was manufactured using standard cheddar cheese procedures a total of three times. Cheddar cheese was fortified with either large microencapsulated ferrous sulfate (LMFS; 0.9536 g microencapsulated ferrous sulfate/

kg cheese and 700–1,000 µm diameter) or small microencapsulated ferrous sulfate (SMFS; 1.7801 g microencapsulated ferrous sulfate/kg cheese and 220 to 422 µm diameter). Iron treatment was incorporated to cheddar cheese processing in the salting step but omitted for the control. After 90 d of aging, calcium, iron, magnesium, and zinc content were analyzed using atomic absorption spectroscopy and percent recoveries were calculated. Moisture, ash, fat, and protein analysis were done using AOAC methods. All collected data was analyzed using one-way ANOVA and Tukey's HSD test ($P = 0.05$). Iron content for all treatments were significantly different ($P < 0.05$): approximately 0.030 mg Fe/g cheese for the control, 0.134 mg Fe/g cheese for LMFS, and 0.174 mg Fe/g cheese for SMFS. Results showed 81.3% iron recovery for LMFS and 90.0% iron recovery for SMFS. Proximate analysis and magnesium, zinc, and calcium content were not significantly different when comparing fortified cheeses with the control. Overall, microencapsulated ferrous sulfate caused no major changes in terms of cheddar cheese composition and successfully increased iron content. Microencapsulated ferrous sulfate with smaller diameter showed slightly better results for iron retention in cheddar cheese. The proposed fortified cheddar cheese can help increase total iron intake for children, pregnant women, vegetarians, and those whose diets are likely to be deficient in iron by providing at least 5 mg Fe (30% RDA) per serving.

Key Words: fortification, cheese, minerals

ADSA PRODUCTION DIVISION GRADUATE STUDENT ORAL COMPETITION: MS

0717 Rumen development in Holstein calves.

K. E. Mitchell*, *University of California, Davis, Davis.*

Feed intake in calves is very important for future production and health, but there are many issues that can influence starter intake such as weather, rumen development, and overall calf health. The objectives of this study were to observe the interaction of starter grain intake and rumen development. Data from 122 Holstein bull and heifer calves were collected from age 2 to 69 d, time of exit from hutch including fecal scores (1–3), DMI, medication, and milk intake. Daily starter grain samples were pooled by week and analyzed for nutrient content by Analab (Agriking, Fulton, IL). Blood samples were collected from a subset of 38 calves and analyzed for glucose (mg/dL) and β-hydroxybutyrate (BHBA; mmol/L) levels with Precision Extra (Abbott Diabetes Care, Inc., Alameda, CA) blood meters. At 1, 6, and 9 wk, blood samples were also analyzed using a VetScan Large Animal Profile rotor (Abaxis Inc., Union City, CA). The rotor tested for albumin (ALB), alkaline phosphatase (ALP), aspartate aminotransferase (AST),