manure moisture and NH_3 losses compared to conventional systems. Although many strategies are available to address NH_3 and other air emissions, ultimately regulations and costs will be determining factors guiding implementation.

Key Words: Ammonia, Air Emissions, Management

195 Emissions, regulations and impact in the EU and The Netherlands. H. Ellen*, *Applied Research of Animal Sciences Group of Wageningen, Lelystad, The Netherlands.*

Air pollution has been one of Europe's main political concerns since the late 1970's. The aim of EU-policy is to develop and implement appropriate instruments to improve air quality. The Sixth Environment Action Programme, "Environment 2010: Our future, Our Choice" regards the period from 2001 until 2010. One of the important issues of the EAP is the reduction of greenhouse gasses. This also supports the aims of the Kyoto-Protocol, because EU-countries have promised to decline their emissions with 8% in total in 2010 in comparison with 1990. In the EAP each country of the EU has an Emission Ceiling for gasses as SO_2, NO_x and NH_3. According to an EU-directive these emission ceilings have to be reached in 2010. In addition to the EU-rules, each country have to implement its own national programme for a progressive reduction of the national emissions. For poultry, the reduction of NH_3-emission is the most important issue in the EAP. Already there was EU-directive in 1996 concerning the integrated pollution prevention and control (IPPC). According to this directive no new poultry farm larger than 40,000 bird places may be operated without a permit. From 2007 all farms need a permission and have to use a so called BAT. These are techniques that aim to reduce emission of ammonia at acceptable costs. Large farms have to make a Environmental Impact Assessment when they apply for a new permit. For already many years in Holland poultry farms require a permit in which also the amount of ammonia and odour emission is registered. Lists of ammonia and odour emission factors for all the housing systems in the different poultry categories has been established. To fulfil the EU-directive 2001/81/EC, the Dutch government has set maximum values for the ammonia emission from farms. Together with the environment regulations, poultry farmers also have to deal with EU-directives on animal welfare. Most of the housing systems that give good welfare, have higher emissions of ammonia. These systems also consume more energy, which doesn't harmonise with the need to reduce the emission of CO_2.

Key Words: Ammonia Emission, Environment, Poultry

Food Safety in Animal Production

196 A USDA multi-agency project: Collaboration in animal health, food safety & epidemiology (CAHFSE). R. R. Kraeling*[1], E. J. Bush[2], D. A Dargatz[2], N. E. Wineland[2], S. Ladely[1], and P. J. Fedorka-Cray[1], [1]*ARS, USDA, Athens,* [2]*APHIS-VS, USDA, Fort Collins, CO.*

The emergence of antimicrobial resistant zoonotic bacteria continues to be a global concern. In response to growing surveillance needs, USDA-ARS, APHIS and FSIA collectively developed CAHFSE. CAHFSE will enhance our overall understanding of pathogens that pose a food-safety risk by tracking these organisms from farm to plant. Risk analysis, antimicrobial use information, resistance and animal health will also be assessed. The first commodity of CAHFSE is pork. Currently, blood and fecal samples are being collected quarterly on sentinel farms in four states. Herd health and management data are also being collected from these farms. To date, fecal samples from 48 site visits have been collected and cultured for Salmonella, Campylobacter and E. coli. Salmonella was recovered from 8.1% (146/1811) of the samples. Sixteen serotypes were identified, of which the predominant serotypes included; S. derby (31.5%), S. typhimurium var. copenhagen (26%), S. heidelberg (8.9%) and S. give (7.5%). Across all serotypes, resistance was most commonly observed for tetracycline (90.4%), streptomycin (63.5%), sulfamethoxazole (43.5%) and ampicillin (43.5%). Among generic E. coli, resistance to tetracycline (92.2%), sulfamethoxazole (27.2%), streptomycin (27.1%) and ampicillin (25.4%) was most common. All Campylobacter isolated to date were identified as C. coli. Resistance to tetracycline (73.6%), azithromycin (60.3%) and erythromycin (60.1%) were observed most often. Determination of risk factors related to prevalence and antimicrobial resistance of these organisms will lead to practical methods of mitigating food borne illness.

Key Words: Swine, Food Safety, Antimicrobial Susceptibility

197 Monitoring the safety of edible poultry tissues: Antibiotic residue concentrations can vary between different muscle tissues. I. Reyes-Herrera*[1], M. J. Schneider[2], K. Cole, P. J. Blore, and D. J. Donoghue, [1]*Department of Poultry Science, University of Arkansas, Fayetteville,* [2]*USDA/ARS/ERRC, Wyndmoor, PA.*

The use of veterinary antibiotics is an important tool for the treatment of disease in poultry. However, misuse of these antibiotics can create antimicrobial residues in edible animal tissues exceeding the FDA established safety tolerances. To ensure the safety of the U.S. food supply, the Federal Government monitors foods, including poultry, for illegal residues. For the veterinary antibiotic enrofloxacin (Baytril ®), the FDA, in the Code of Federal Regulations, has listed muscle as the tissue to be monitored in poultry, irrespective of the type of muscle tissue. This study was conducted to determine if either breast or thigh meat is a better indicator of the highest enrofloxacin concentrations. One hundred and sixty five, 5-wk old chickens were dosed with the FDA approved dose of enrofloxacin in water. The 4 treatment groups were: 25 ppm for 3 days, 25 ppm for 7 days, 50 ppm for 3 days and 50 ppm for 7 days. Five chickens from each treatment group were randomly selected and samples of breast and thigh muscle tissue were collected prior to dosing (controls, n=5), during the dosing (n=5/group/day) and for a 3-day withdrawal period (n=5/group/day). Each sample was prepared and assayed using an agar diffusion microbiological method (Schneider and Donoghue, Poultry Science, 2004). The results demonstrated that greater overall concentrations of enrofloxacin were present in breast versus thigh muscle tissues during the dosing period (443 $\pm$ 22 ppb vs. 386 $\pm$ 24 ppb. $P < 0.05$). These data indicate that, at least for enrofloxacin, not all muscle tissues incorporate antibiotics at the same concentrations. Therefore, the Federal Government should consider monitoring specific muscle tissues to ensure established safety tolerances are not being exceeded.

Key Words: Enrofloxacin Residues, Muscle Tissues, Chicken

198 Survival of *Salmonella* species following sodium hypochlorite treatment during commercial broiler processing. R. M. Lipscomb*, L. T. Walker, W. L. Hurlock, L. L. Williams, and M. Verghese, *Alabama A&M University, Normal.*

Technological advances and mass production in poultry processing present new challenges for providing a microbiologically safe product. Research has shown that Salmonella can survive acidic conditions. The objective of this study was to determine the effect of Acidified Sodium Hypochlorite (ASH) on survival of *Salmonella* species on commercially produced broiler carcasses. Broiler carcasses were sampled from four processing steps (rehang, reprocessing, prechill, and postchill) to determine the effect of ASH against Salmonella. Ten carcasses were sampled at each processing step. Carcasses were dipped (20 secs) in an ASH solution dip tank at prechill and rinsed (1 min) with 400ml Butterfield's Phosphate Dilution water (BPD). A 1:10 dilution was plated onto Brilliant Green and Bismuth Sulfite agars, incubated for 24 hours at 37C, and Salmonella counts were determined. Salmonella O Poly A and H Poly A antigens were determined by serotyping. *Salmonella* species were confirmed by PCR analysis. The experiment was replicated three times over three periods. The results showed that average Salmonella counts were 1.2888 log cfu/ml (rehang), 0.9403 log cfu/ml (reprocessing), 0.9779 log cfu/ml (prechill), and 0.2039 log cfu/ml (postchill). Means for 48 and 72 hours were not significantly different at p#88050.05. Of a total of 53 suspect Salmonella isolates (including all reps), 37 were positive for the Salmonella O Poly A antigen, and 50 samples were positive for the Salmonella H Poly A antigen. PCR analysis confirmed 32 samples to be *Salmonella* species.

Although ASH was effective in reducing microbial contamination, *Salmonella* species survived ASH treatment. Several factors can influence the effectiveness of ASH such as pH, temperature, and ASH concentration. Regulation of pH, temperature, and ASH concentration is the key to ASH achieving its full potential as a disinfectant used in the poultry industry.

Key Words: Salmonella, Broilers, Sodium Hypochlorite

199 Herd-level factors associated with cow and calf *Salmonella* shedding in a multi-state study of 129 dairy farms. C. P. Fossler*[1], S. J. Wells[1], J. B. Kaneene[2], P. L. Ruegg[3], L. D. Warnick[4], J. B. Bender[1], L. E. Eberly[1], S. M. Godden[1], and L. W. Halbert[2], [1]*University of Minnesota, St. Paul,* [2]*Michigan State University, East Lansing,* [3]*University of Wisconsin, Madison,* [4]*Cornell University, Ithaca, NY.*

This study evaluated associations between herd characteristics and the isolation of *Salmonella* from dairy cows and calves. 129 conventional and organic farms in Minnesota, Wisconsin, Michigan, and New York were enrolled without regard to previous *Salmonella* history. Herds were sampled up to 5 times at 2-month intervals over one year. *Salmonella* was detected in fecal samples from 4.9% of 20,089 cows and 3.8% of 4,673 preweaned calves. Risk factors were evaluated using multivariable logistic regression with the generalized estimating equations approach, and within-herd prevalence by visit in cows and calves was the outcome used for cow and calf models, respectively. Factors associated with an increased odds for *Salmonella* shedding in the final cow model ($p<0.05$) included lack of use of tiestall or stanchion facilities for lactating cows, not storing all purchased concentrate or protein feeds in an enclosed building, not using monensin in weaned calf or bred heifer diets, cow access to surface water, disposal of manure in liquid form (slurry or irrigation, as opposed to broadcast/solid spreader) on owned or rented land, and eating or grazing roughage by cows from fields having surface application of manure during the growing season. Factors associated with an increased odds for *Salmonella* shedding in the final calf model ($p<0.05$) included lack of routine feeding of medicated milk replacer, and use of calving pen as a hospital area for sick cows>once a month. Herd size and farm type (organic vs. conventional) were not associated with *Salmonella* shedding in the final cow or calf models. A number of herd-level risk factors were identified in this study that could potentially be implemented in *Salmonella* control programs on dairy farms.

Key Words: *Salmonella*, Dairy Cattle, Risk Factors

200 Investigation of cattle feedlot management practices to reduce *Escherichia coli* O157. J. R. Ransom*, K. E. Belk, J. A. Scanga, J. N. Sofos, and G. C. Smith, *Colorado State University, Fort Collins.*

Studies have shown that prevalence of *Escherichia coli* O157 on/in cattle entering the packing plant range from 10 to >70%. The objective of this study was to determine the effectiveness of single and multiple preharvest pathogen intervention strategies on the prevalence of *E. coli* O157 on/in cattle before transport to harvest. Cattle from 24 pens [approximately 200 head of cattle (419 kg) per pen] were randomly allocated (3 pens/treatment) to one of 8 treatment groups: Control (CT; No treatment), Bovamine (Bov; a *Lactobacillus acidophilus* product), NEOMIX (Neo; neomycin sulfate), an *E. coli* O157:H7 bacterin vaccine (Vac) and all combinations of the single treatments. Treatments of Bov and Vac began 60 d preharvest, and Neo was administered for 3 d before harvest with a 24 h withdrawal period. Twenty-five cattle per pen were randomly sampled using rectal fecal collection and by sponge-swabbing 500 cm<2> of the dorsal-thorax region. Cattle were sampled within a 10 d timeperiod and samples were collected no more than 48 h before harvest. Results showed that CT cattle had the highest pathogen prevalence levels (45.8 and 40.3%, for fecal and hide samples, respectively), and cattle receiving treatments had a lower prevalence of *E. coli* O157 than did CT. Trends suggested that Neo was the most effective single intervention, reducing *E. coli* O157 prevalence levels to 0.0 and 8.5%, in feces and on hides, respectively. Bovamine and Vac used singly, were equally effective, as pathogen prevalence was reduced to 22.7 and 20.0%, respectively, on hide samples and to 13.3 and 14.7%, respectively, for fecal samples. When Bov, Vac and Neo were used in combination, pathogen prevalence for fecal and hide samples were reduced to 2.7 and 6.7%, respectively. In general, combinations of interventions resulted in lower pathogen prevalence than when a single intervention was used. This study demonstrated that preharvest mitigation strategies used singly or in combination can be effective in reducing the prevalence of *E. coli* O157 in market-ready feedlot cattle.

Key Words: Cattle, Preharvest, *Escherichia coli* O157

201 Colicin E1, N and A treatment inhibits growth of *Escherichia coli* O157:H7 strains in vitro. T. R. Callaway*[1], C. H. Stahl[2], T. S. Edrington[1], K. J. Genovese[1], L. M. Lincoln[2], R. C. Anderson[1], R. B. Harvey[1], and D. J. Nisbet[1], [1]*USDA/ARS, Food and Feed Safety Research Unit, College Station, TX,* [2]*Department of Animal Science, Iowa State University, Ames.*

Escherichia coli O157:H7 is a highly virulent food-borne pathogen that causes severe human illnesses and inhabits the intestinal tract of ruminants. Although abattoir do an excellent job of controlling the cross-contamination of pathogens, too many human illnesses linked to animal-derived food products still occur. Therefore, researchers have investigated methods to reduce the overall burden of *E. coli* O157:H7 entering the abattoir within the live animal. Antibiotic treatment has been proposed to reduce *E. coli* O157:H7 populations in the live animal; however, with the growing worldwide concern over the prophylactic use of antibiotics in food animals, alternative strategies to the use of traditional antibiotics must be explored. Colicins are antimicrobial proteins produced by some *E. coli* strains that are inhibitory or bactericidal against other *E. coli*. In the present study, the efficacy of three pore-forming colicins E1, N and A were quantified in vitro against *E. coli* O157:H7 strains 86-24 and 933. All three colicins reduced the growth of *E. coli* O157:H7 strains, but the efficacy of each colicin varied between strains. Colicin E1 was most effective against strain 86-24 and 933. Colicin N, while less effective than E1, was very effective against strain 86-24, however, it was unable to produce more than a 50% reduction in growth against strain 933. Colicin A did not show more than 50% of the efficacy of either of the other colicins, even when added to incubations at 10-fold or greater protein concentrations. Colicin E1 was more effective against both strains of *E. coli* O157:H7 than colicins A and N, and reduced populations of *E. coli* O157:H7 more than 1000-fold at concentrations less than 5 μg/ml. Colicin N reduced populations of strain 86-24, but not 933; and colicin A did not affect the populations of either *E. coli* O157:H7 strain tested. These potent antimicrobial proteins may potentially provide an effective and environmentally sound pre-harvest strategy to reduce *E. coli* O157:H7 populations in ruminant animals.

Key Words: E. coli O157:H7, Intervention Strategy, Colicin

202 Effects of lactic acid treatments on microbiological, chemical, and sensory properties of stored fresh beef trimmings. S. E. Rose*, K. E. Belk, J. N. Sofos, J. A. Scanga, K. L. Hossner, and G. C. Smith, *Colorado State University, Fort Collins.*

The objective of this study was to evaluate the effectiveness of warm (55°C) lactic acid (LA) at various concentrations (1.25, 2.00, or 2.50%) on microbiological, chemical, and sensory properties of fresh beef trimmings stored at 4°C for 0, 2, 5, or 8 d on Styrofoam™ trays wrapped with air-permeable cellophane film. Two replicates of four beef short plate (BSP) pieces (5 cm x 2.5 cm x 1 cm; 40 cm^2) per treatment were immersed in treatment solutions or water control at 55°C for 30 s. Aerobic plate counts (APC) were determined using Petrifilm™ Aerobic Count Plate (ACP), and Total Coliform Count (TCC) and *Escherichia coli* (ECC) populations were determined using Petrifilm™ *E. coli*/Coliform Plate (ECCP). Lactic acid concentrations of the BSP pieces were determined by conducting enzymatic colorimetric assays. A sensory panel was used to determine differences in odor of BSP pieces after treatment and during storage. A MiniScan XE was used at each storage time to evaluate color (L*, a*, and b* values) of muscle and adipose tissue. Bacterial populations associated with BSP pieces treated with 1.25% LA were not different ($P > 0.05$) from controls; however, 2.00 or 2.50% LA reduced ($P < 0.05$) APC, but did not ($P > 0.05$) reduce TCC, when compared to controls for all storage days. All ECC were below the detection limit in all treatments and the control at all storage days. Lactic acid concentrations associated with BSP pieces treated with 2.00 or 2.50% LA at all storage days were higher ($P <$

0.05) compared to control BSP pieces. There were no differences ($P > 0.05$) in odor between treatments and the control BSP pieces for all storage days. Lactic acid treated BSP pieces were more likely to be lighter, greener, and more yellow throughout the storage period when compared to control BSP pieces. Lower microbial counts and indistinguishable odor differences were achieved with 2.00% LA treated BSP pieces. Using 2.00% LA in beef trimmings may provide a microbiologically cleaner product to the consumer without major adverse effects on quality.

Key Words: Lactic Acid, Beef Trimming, Residue

203 Influence of aflatoxin B1 on milk production and health in dairy sheep. G. Battacone[1], M. Palomba[2], M. Pascale[3], A. Mazzette[1], and G. Pulina*[1], [1]*Dipartimento di Scienze Zootecniche, University of Sassari, Italy via Enrico de Nicola 9, 07100 Sassari, Italy,* [2]*Dipartimento Farmaco Chimico Tossicologico, University of Sassari, Italy via Muroni, 07100 Sassari, Italy,* [3]*CNR Istituto di Scienze delle Produzioni Alimentari, Bari, Italy via le Einaudi 51, 70125 Bari, Italy.*

The transfer of aflatoxin B1 in the diet of lactating sheep into the milk were investigated, and also its effects on milk yield and animal health. Twenty lactating Sarda sheep were divided into four groups of five. Three groups were used for the experiment and the fourth was a control group. The experimental design was a 33 Latin square with one additional group. The experimental groups were given 32, 64 or 128 μg per day of pure aflatoxin B1 (AFB1) in two daily doses, given immediately before milking at 7:00 a.m. and 7:00 p.m.. The treatment continued for 7 days, followed by a 5 day clearance period. The control group was fed an aflatoxin-free diet. Individual milk production was recorded and milk sampled at each milking. Blood samples were collected after 7 days. Aflatoxin M1 (AFM1) levels in milk were determined using an immunoaffinity column - HPLC method. AFM1 appeared in the milk of all treated groups 12 h after the beginning of administration. No AFM1 was found in the milk of the control group. AFM1 concentrations above the EC tolerance level (0.05 μg/kg) were detected even in the milk of the group that received only 32 μg/d of AFB1. The mean AFM1 concentration in milk reached a steady state after 2-3 days. At this point AFM1 concentration did not differ at morning and evening milking. The AFM1 concentration was linearly related to the dose. No AFM1 was detected 3-4 days after the end of treatment with AFB1. This suggests that the Latin square is an appropriate experimental design for mycotoxicological studies. The milk production traits of the AFB1 groups were no different from those of the control group. These doses of AFB1 had no effect on hematological and biochemical blood parameters. The results indicate that the level of AFB1 used did not adversely affect animal health and milk production traits, while considerable amounts of AFM1 were excreted in the milk. (Partly supported by MIUR-MiPAF SISPROLAT project)

Key Words: Aflatoxin, Sheep, Milk

ADSA-ASAS Northeast Graduate Student Competition

204 Fractional removal of amino acids by the small intestines and whole gastrointestinal tract of sheep remains constant across levels of protein supply. S. W. El-Kadi*, N. E. Sunny, M. Oba, S. L. Owens, and B. J. Bequette, *University of Maryland, College Park.*

We hypothesized that the net removal of amino acids (AA) by the mesenteric (MDV; small intestine) and portal (PDV; whole gut) drained viscera of sheep would remain fixed in amount, even with increasing supply of protein to the gastrointestinal tract (GIT). Wethers (n=4, 33 $\pm$ 2.0 kg) were fitted with catheters for duodenal infusion of casein and for measurements of PDV and MDV appearance of AA. Animals were fed a forage-based diet low in protein (9.5 % CP) to 1.4 $\times$ maintenance and given 5-d duodenal infusions of casein (0, 35, 70 and 105 g/d) in a 4 $\times$ 4 Latin square design. On day 5 of each period, a blood flow marker was infused and blood continuously withdrawn over 1-h intervals during a 4-h period. Plasma concentrations of AA were determined by isotope dilution with mass spectrometry. Net absorption of AA across the PDV and MDV were calculated as the product of veno-arterial difference and blood flow. Regression analyses were performed to establish equations describing net appearance of AA in relation to AA infusion rate as casein. Regression curves were found to best fit a first-order model (R^2 = 0.60-0.95; at least $P < 0.05$) for all AA except Val ($R^2 = 0.50$; $P = 0.06$). The large removals of the branched chain AA (43-51%; $P < 0.05$) by the GIT probably relates to their catabolism for energy production. For Lys, however, it is unclear why the GIT net metabolizes (33%; $P < 0.05$) this potentially limiting AA to a much greater extent than for His, Phe and Met, whose fractional removals by the GIT were lower (7-19%). Our data indicate that AA removal by the GIT is not fixed in amounts, but rather that the amount removed increased with greater protein supply. The fates and factors affecting AA metabolism by the GIT have yet to be elucidated. However, if AA are metabolized by the GIT to provide energy then perhaps there may be opportunity to reduce catabolism by providing energetically equivalent non-AA substrates.

Key Words: Amino Acid, Sheep, Gastrointestinal Tract

205 Regulation of urea recycling to the gastrointestinal tract and ammonia metabolism in ruminants. N. E. Sunny*, L. H. Hanus, S. W. El-Kadi, S. L. Owens, and B. J. Bequette, *University of Maryland, College Park.*

Urea recycling is the main N salvage mechanism allowing ruminants to maintain positive N balance on poor quality diets. Our aim was to determine the extent ruminants control urea recycling to the GIT, independent of rumen microbial metabolism. Thus, four wether lambs (28.1 kg BW) were fed to 1.5 x maintenance energy a pelleted diet containing (kg^{-1}, as-fed) 1.7 Mcal ME and only 68 g CP. Animals were assigned to four levels of urea-N infusion (0, 3.8, 7.5, 11.3 g/d) arranged in a 4 x 4 Latin square design. Urea was infused into a jugular vein for 10-d periods, and urea kinetics determined by continuous infusion of [$^{15}N^{15}N$]urea over the last 80 h. During [$^{15}N^{15}N$]urea infusion, total urine was collected by suction and feces by harness. Isotope enrichment of urinary urea ($^{15}N^{15}N$, $^{14}N^{15}N$ and $^{14}N^{14}N$) and fecal total ^{15}N was determined by mass spectrometry. Urea-N entry rate (UER) increased (5.1 to 21.8 g/d; $P < 0.0001$) with level of urea infusion, whereas the proportion of UER entering the GIT decreased progressively (80 to 61%; $P < 0.01$). The amount of urea-N partitioned to the GIT (recycling; 4.1 to 13.2 g/d; $P < 0.0001$) increased with each level, as did the amount excreted in urine (1.0 to 8.6 g/d; $P < 0.0001$). However, the proportion (44 to 69%; $P < 0.001$) and amount (1.8 to 9.2 g/d; $P < 0.0001$) of the GIT entry that returned to the liver for ureagenesis was greater with urea infusion. In consequence, the amount of urea-N used for anabolism (rumen microbial protein synthesis) reached a maximum (2.3 g/d) at the second level of urea infusion. Concurrently, fecal urea-N excretion reached a maximum (1.5 g/d) at the third level of infusion. The present study suggests that the ability of ruminants to partition urea-N to the GIT is less a limitation than the rumen environment. Thus, rather than up-regulating urea entry to the GIT, there appears to be more potential to improve N efficiency of ruminants by manipulating the rumen environment (eg. fermentable energy, pH) for optimal capture of recycled N.

Key Words: Urea Recycling, Ruminant, Metabolism

206 Urea supplementation increased rumen function in lactating dairy cows fed a corn silage based diet deficient in rumen-degradable feed protein (RDP). S. E. Ferguson*, R. S. Ordway, N. L. Whitehouse, P. J. Kononoff, and C. G. Schwab, *University of New Hampshire, Durham.*

Four lactating Holstein cows fitted with ruminal and duodenal cannulae were used in a 4 x 4 Latin square to determine the efficacy of adding urea to a corn silage based diet on ruminal fermentation and microbial protein synthesis. Dietary treatments were 0, 0.3, 0.6, and 0.9% urea in diet DM; urea was top dressed and manually incorporated into the diet. The basal diet contained (DM basis) 32% corn silage, 16% grass silage, 4% alfalfa hay, 19% corn, 6% barley, 4.5% soybean hulls, 3% citrus pulp, 3% beet pulp, 7% soybean meal, 1.3% ProvAAl^{TM}, 1.7% Megalac$^{®}$, and 2.7% vitamin/mineral mix. The basal diet was formulated to meet NRC (2001) requirements for energy and all nutrients except RDP. Cows were fed 3 times daily. Experimental periods were 14 d with a 9-d adaptation. Duodenal digesta (n=16), rumen samples (n=16), and milk samples were