1338 Ca and P endogenous losses and true absorption of alfalfa and fescue diets when fed to dairy cows. M. F. Weiss*, F. A. Martz, R. L. Belyea, and A. T. Belo, *University of Missouri, Columbia MO*.

Eight holstein cows of high milk potential were paired by production and parity, then assigned to a fescue(F) or an alfalfa(A) diet in order to study dietary Ca and P use. Dry cow diets were 50% forage, plus corn silage with 0.91% Ca and 0.42% P for A and 0.50% Ca and 0.27% P for F. Diets fed during lactation were 24% forage, plus corn silage and grain mix resulting in 0.55 and 0.57% Ca, while 0.37and 0.45% P for A and F respectively. Endogenous fecal loss(EFL) and diet true absorption(TA) were measured using isotope dilution of 45-Ca and 32-P single injection tracers iv. Plasma and fecal specific activity(SA) of both tracers were measured over 7d during the dry(D) period, early(EL) and post-peak(PP) lactation. Data were analyzed as a split-plot in time. The ratio of integrals of the fecal SA curve to the plasma SA curve is theoretically justified as the first step for precursor-product single injection tracer studies (to find fractional EFL). Alternatively simple fecal SA/plasma SA equilibrium ratios with various time delays are often used for convenience. Calculated EFL and TA varied with the method used, and values from different methods changed rank dependent upon stage of lactation and mineral. For Ca EFL, the differences among methods were greatest for EL period effect, with the integral method value 9.4g/d intermediate to the equilibrium ratio methods with 24 and 48h delays, 11.3 and 6.5 g/d respectively(P<.01). Values for TA reflect this same pattern for EL, with the integral solution value 40% being likewise intermediate. The Ca EFL for main effect of D was 6.7g/d(diff. from EL, P<.05) comparing integral values. Main effects for diet were not significant. For P EFL, the differences among methods were also greatest for EL period effect, with the integral method(Pi based) value 22.4g/d, being greater than the equilibrium ratio methods with 24 and 48h delays, 15.5 and 12.9g/d respectively(P<.01). Corresponding TA values were 70, 62, and 59% respectively. The P EFL for main effect of D was 12.2g/d(diff. from EL, P<.05) comparing integral values. Main effects for diet were not significant(P<.05) for P EFL.

Key Words: Dairy, True absorption, Phosphorus

1339 Continuous vs 8-paddock rotational stocking of rye-ryegrass pastures at three stocking rates. F.M. Rouquette, *Texas Agricultural Experiment Station*.

During two successive years, Simmental crossbreed steers and heifers, and Angus X Braham (F-1) steers (total n=272) grazed Maton rye and TAM-90 annual ryegrass pastures under grazing methods of either continuous (CNT) or rotational (RTN) stocking, and each at three (LO, ME, HI) stocking rates (SR). All pastures were stocked without interruption each year from early-December to mid-May for 159 d in Year 1 and 156 d in Year 2. A fixed SR for both CNT and RTN were the same within a year for LO, ME, and HI, respectively, and for Year 1 was 4.2, 5.7, and 7.2 hd/ha, and for Year 2 was 3.7, 5.2, and 6.7 hd/ha. At initiation of grazing in December, stocker cattle averaged 275 kg and were weighed at 28-d intervals. Pastures were sampled for nutritive value and DM at about 14-d intervals from both CNT and the 8-paddock RTN pastures. Residence time in each paddock of RTN stocked pastures averaged 2 days with about a 14-day rest period. The overall statistical analyses showed differences for ADG between years (P=.0001), grazing method (P=.03), stocking rates (P=.0001), and sex (P.0001), but no difference for breeds (P=.25). In Year 1, ADG was similar for grazing methods, CNT vs RTN; different for all three SR with 1.25 kg/d (LO), 1.06 kg/d (ME), and 0.69 kg/d (HI); and different for steers (1.06 kg/da) vs heifers (0.86 kg/da). In Year 2, ADG was highly significant for grazing method, stocking rate, breed, and sex. A significant interaction showed ADG from RTN to be greater than from CNT and was attributable principally to HI (P=.0008) SR (0.39 vs 0.20 kg/d) rather than to ME (P=.09) (0.74 vs 0.65 kg/d) or LO (P=.55) (1.06 vs 1.02 kg/d). The rest period in RTN paddocks enhanced forage DM production in all SR; however, only at HI SR was ADG increased. When forage DM limits intake and SR can not be reduced, then a RTN grazing method will likely increase gains per animal and per ha via increased forage production. Selection of CNT vs RTN for cool-season annual grass pastures was not as important as selection of SR for expected animal performance. //

Key Words: Grazing, Stocking Rate, Ryegrass

1340 Use of dosed and endogenous herbage alkanes as markers for estimating intake of alfalfa and alfalfa:sainfoin pastures by grazing steers. Y. Wang*[1], T.A. McAllister[1], L.R. Barbieri[1], B.P. Berg[2], and D.M. Veira[3], [1]*Agriculture and Agri-Food Canada, Lethbridge, AB,* [2]*Alberta Agriculture, Food and Rural Development, Lethbridge, AB,* [3]*Agriculture and Agri-Food Canada, Kamloops, BC*.

Externally dosed (C_{32}) and naturally occurring (C_{31}) alkanes were used to estimate intake of three alfalfa:sainfoin pastures (0 to 36% sainfoin) by 12 ruminally cannulated steers ($n = 4$) to determine the effects of sainfoin in the pasture on intake. The study comprised three 12-d periods during which the steers grazed continuously for 6 h/d (0830 to 1430 h). Samples of pasture herbage and pure swards of component species were collected every 48 h for alkane analysis by gas chromatography. The majority (95%) of the alkanes in herbage were odd-numbered from C_{25} to C_{33}, predominantly C_{29} and C_{31}, and these findings were consistent between pastures and between grazing periods. Alkane composition profiles of pure alfalfa and sainfoin were also similar. Each day, the steers were dosed intraruminally with 10 g of ground, extracted alfalfa hay coated with 1 g exogenous alkane (dotriacontane, $C_{32}H_{66}$) immediately prior to entering the pastures. Fecal samples were collected by rectal grab on d 8 to d 12, immediately prior to and following grazing. Intake was estimated as excretion of marker/indigestibility of forage. Average intake of organic matter (OMI) by steers, as estimated by the combination of C_{31} and C_{32} methods, was 4 to 5 kg per 6-h grazing bout. Within periods, OMI did not differ ($P > 0.05$) among pastures; further, OMI estimates were numerically similar in each of the three periods. In all pastures and all periods, estimates of OMI derived from afternoon fecal samples were numerically higher than those derived from samples collected in the morning; this was closely associated with the lower fecal concentration of external marker alkane (C_{32}) in the afternoon as compared to morning samples. It is concluded that sainfoin incorporated into alfalfa pastures at up to 36% (DM basis) did not affect feed intake of grazing beef cattle.

Key Words: n-Alkanes, Grazing, Feed Intake

1341 Withdrawn. , .

Growth and Development
Dairy Calf and Heifer Growth

1342 Development of a repeatable procedure for rumen tissue sampling. K. E. Lesmeister*, A. J. Heinrichs, and P. R. Tozer, *The Pennsylvania State University, University Park, Pennsylvania*.

To aid the understanding of rumen development and papillae growth in young calves and to increase repeatability in rumen tissue sampling techniques, a procedure for rumen sampling and measuring was developed. An extensive statistical analysis of the procedure's results was conducted to determine its efficacy. With the reticulo-rumen lying upon its left side, the esophageal groove facing away, an incision was made around the circumference of the organ in line with the esophageal groove. A 6 cm section of the caudal portion of the caudal ventral blind sac was maintained intact. The rumen pillars were incised in line with the initial incision, and the muscles forming the rumen pillars separated. The reticulo-rumen was then opened and laid flat, creating a right and left side separated by the portion of the rumen maintained intact. The rumen pillars separate the rumen into distinct sampling areas: the caudal portion of the caudal ventral blind sac (CaV), the caudal dorsal blind sac (CaD), the cranial dorsal sac (CrD), the cranial ventral sac (CrV), and the ventral portion of the CaV. Right and left sides of the rumen were sampled. A 1-cm^2 section was removed from the four corners and center of each area and measured for papillae length (n=20) and width (n=20), rumen wall thickness (n=5), and number of papillae per cm^2 (n=5). Rumens utilized in the development of this procedure were obtained from 12 calves that were assigned to 1 of 3 treatments. Means

and standard errors were obtained for each area and a power analysis conducted to determine necessary sample size with a power of .80. The power analysis suggests areas CrV, CrD, and CaD for papillae length determination and area CrV to determine differences for papillae width. Samples should be taken from the CaD and CrD to find differences for rumen wall thickness, and from the CrD, CaD, and CaV to find differences for papillae per cm^2. However, additional differences between treatments may result from different analysis of variance procedures.

Key Words: Procedures, Rumen Sampling, Power Analysis

1343 Absorption of IgG from maternal colostrum or fractions of bovine or porcine plasma proteins. J. D. Quigley* and T. A. Wolfe, *APC, Inc.*

Absorption of an adequate mass of IgG from maternal colostrum is required to minimize the risk of morbidity and mortality in neonatal calves. Colostrum supplements (CS) or replacers (CR) have been developed to allow producers to manage maternal colostrum (MC) programs. However, formulation of CS and CR have not been optimized and the potential use of porcine serum in CS has not been determined. Our objective was to compare IgG concentrations in calves fed MC, CR or CS containing bovine (BS) or porcine (PS) serum as the IgG source. Both CS were formulated to contain approximately 50 g of IgG/feeding and CR was formulated to contain approximately 100 g of IgG/feeding. Holstein and Jersey calves (n = 44) were collected immediately after birth and fed 1.9 L of MC or reconstituted CR, BS, or PS at 1.1 and 8.1 h of age. Jugular blood was collected at 0 and 24 h for measurement of hematocrit, total protein and IgG. Data were analyzed as a completely randomized design with treatment, sex of calf and breed in the model. Mass of IgG consumed by calves was 168, 100, 81, and 253 g for calves fed MC, BS, PS and CR, respectively. Plasma IgG was not measurable in samples taken at 0 h of age. Mean plasma IgG concentrations at 24 h of age were 13.8, 10.6, 6.7, and 13.9 g/L, respectively. Least squares means of total protein at 24 h were 5.73, 5.42, 5.08, and 5.45 g/dl, respectively. Least squares means of hematocrit at 24 h of age were 34.4, 29.7, 30.6, and 34.2%, respectively. Proportion of calves with FPT (<10 g of IgG/L of plasma at 24 h) for calves fed MC and CR were 40 and 14%, respectively. More calves had FPT when fed PS compared to BS. Apparent efficiency of IgG absorption did not differ among treatments, suggesting that differences in plasma IgG were due to differences in IgG intake. Relationship between total protein and plasma IgG concentrations at 24 h of age varied among treatments, suggesting that use of the refractometer to predict IgG concentrations in neonatal calves may be inappropriate when CS or CR are fed. Production of PS containing large amounts of IgG was difficult and resulted in CS with reduced IgG concentration.

Key Words: Calves, Immunoglobulin, Colostrum

1344 Colostrum intake in the newborn calves. R. Skrzypek*[1], D. Hofmanski[2], and S. Osieglowski[3], [1]*Agricultural University, Poznan, Poland,* [2]*Kombinat 2000, Smigiel, Poland,* [2]*National Research Institute of Animal Production, Balice, Poland.*

The study was carried out on 61 single-born Holstein x Black-and-White calves of both sexes (33 heifers, 28 bulls). Within the first 5 days after birth, the calves were fed twice a day with fresh mother's colostrum, using a bucket with nipple. The first feeding was practiced within the first hour of birth. For the first 3 days of life, the colostrum was offered ad libitum, and for the next 2 days the amount of colostrum fed was limited to 4L per feeding. Daily colostrum intake increased from 4.86L on day 1 to 7.41L on day 5 after birth. This trait was associated significantly with the share of Holstein genes in calves (range from 50.0 to 96.9%) and with their birth weight (39.5±5.9 kg). The share of Holsteins genes was associated with colostrum intake in the first day of life only (R^2=0.16; P<0.01), whereas birth weight was correlated with colostrum intake from day 2 (r=0.50; P<0.01) to day 5 of life (r=0.32; P<0.01). In the first day of life, colostrum intake ranged from 4.2 to 6.1L in calves carrying 96.9% and 50.0% Holstein genes, respectively. This corresponds with earlier results of Skrzypek (2000), who found nearly 3-fold lower colostral immunity in first- and second generation sucking Holstein x Black-and-White crosses, as compared to the purebred native calves. Within the days 3 through 5 after birth, colostrum intake was correlated significantly with daily body weight gains (r from 0.25; P<0.05 to 0.43; P<0.01, respectively), while at the younger age the correlation was insignificant (r from -0.02 to 0.22).

Key Words: Calves, Colostrum intake

1345 Feeding liquid whey to newborn Holstein dairy calves. R. Valizadeh*, M. Jamchi, and A. Naserian, *Ferdowsi University, Agriculture college, Animal Sci. Dep.,Mashhad, Khorasan, Iran.*

A study was conducted to evaluate the effects of sweat liquid whey on feed intake and daily gain of newborn Holstein dairy calves using a completely randomized design with factorial arrangement of 2×4. Sixteen male calves with mean body weight of 43±4 Kg and 16 female calves with mean body weight of 40±4 Kg fed individually from birth to weaning time (65 days post-partum) by the following regimes: A; (control), fresh milk (10% of body weight)+ starter (ad libitum) + tap water, B; fresh milk (8% of body weight) + liquid whey (6% of body weight) + starter (ad lib.) + tap water, C; fresh milk (6% of body weight) + starter (ad lib.) + liquid whey (12% of body weight) + starter (ad lib.) + tap water, D; fresh milk (10% of body weight) + starter (ad lib.) + liquid whey (ad lib.). ME and CP contents of the starter component were 2.86 MCal/Kg and 15.5% respectively. Average daily gain of male calves was higher than females (499 vs. 405 g/d). Mean daily intakes of male male calves for treatments A, B, C and D was 2.05, 2.43, 2.19 and 2.12 Kg. These figures for female calves were 1.91, 2.10, 2.30 and 2.21 Kg respectively. Although, a large amount of liquid whey was consumed by the calves in treatments C (3.8 Kg/d), D (3.4 Kg/d) and B (2.01 kg/d) no significant difference was observed between the daily gain of experimental calves. This finding has an important economical and practical application. In Iranian dairy industry liquid whey is produced in large amount and is mostly discarded in the enviroment, which can be a big source of pollution. In this situation this cheap unconventional feed resource can be fed to suckling or young calves without any adverse effects. The minimum and maximum figures for rumen pH in this study were 6.07 and 6.70. These are in the normal range for a healthy and functional stomach. The blood glucose and BUN contents for the calves were into the normal ranges. It was concluded that utilization of liquid whey in feeding of suckling and growing calves could be an economical and practical recommendation for Iranian dairy farms.

Key Words: Liquid whey, Dairy calves, Gain

1346 Analysis of body composition of Jersey bull calves fed varying levels of fat and protein with dual energy X-ray absorptiometry. S. S. Bascom*, C. S. Huffard, S. M. Nickols-Richardson, E. P. Hovingh, R. E. James, and M. L. McGilliard, *Virginia Polytechnic Institute and State University.*

Week-old Jersey bull calves (n=20) were fed one of four diets for four weeks to determine differences in body composition. In addition, two calves were slaughtered at 7 days of age to establish baseline body composition. Calves assigned to diet MM (n=5) were fed 21% fat:21% protein milk replacer (MR) at 15% of body weight. Calves on diets HH, HL, and JM were fed 180g of CP per day. Calves assigned to diet HH (n=5) received a 27% CP:33% fat MR. Calves assigned to HL (n=5) received a 29% CP:16% fat MR. Calves assigned to diet JM (n=3) received whole milk (4.7% fat:3.2% true protein). Calves were fed three times daily; MRs were reconstituted to 12.5% dm. Calves were sacrificed and liver, organs (including empty GI tract), and right half of the carcass were used to estimate body composition. Tissue fat content was determined by dual energy X-ray absorptiometry (DXA; Hologic 4500A, Bedford, MA) using the Whole Body Analysis (version 8.25a) software. Differences in fat% in liver and organs were not significant. Carcass half fat% were 20.85±1.37, 12.79±0.81, 13.93±0.81, 12.80±0.86, 15.83±1.15, respectively for baseline, and diets MM, HH, HL, and JM. When compared to the baseline body composition all calves fed MR had lower carcass fat%. The carcass fat% of JM calves was not different from baseline. Calves fed MR lost carcass fat from 1 week of age to 5 weeks of age but calves fed JM maintained baseline body composition.

Key Words: Calves, Milk Replacer, DXA

1347 Growth hormone influences growth performance, but does not affect gluconeogenesis from lactate or propionate in 60-d old veal calves. H.M. Hammon*[1] and S.S. Donkin[2], [1] *University of Berne, Berne, Switzerland,* [2] *Purdue University, West Lafayette, IN.*

The somatotropic axis becomes the main endocrine growth regulatory system after birth and growth hormone (GH) enhances glucose supply for bone and muscle growth. The objective was to investigate the influence of GH on growth performance, plasma glucose concentrations, and hepatic gluconeogenesis in veal calves. Thirteen male calves were randomly assigned to one of two treatment groups starting at d 3 of age. Calves received colostrum on first d of life followed by milk replacer at a rate of 2% of body weight (BW) on a dry matter basis. Calves (GrGH, n = 6) were treated with 500 mg GH as Posilac (Monsanto, St. Louis, MO) on d 3, 17, 31, 45, and 59 of age and control calves (GrC, n = 7) received 0.9% saline. BW was measured weekly and blood samples were obtained on d 3, 7, 14, 28, 42, and 60 of age. On d 60 liver biopsy samples were obtained, and liver slices were prepared to determine incorporation of [U-^{14}C]lactate and [2-^{14}C]propionate into glucose and CO_2 (nmol of substrate converted to product/mg tissue/h). BW increased with age ($P < 0.001$) and ADG tended to be higher ($P = 0.06$) for GrGH (0.87 ± 0.01 kg/d) compared with GrC (0.80 ± 0.03 kg/d) calves. Plasma glucose concentrations decreased in both groups to d 28. Plasma glucose concentrations were higher ($P < 0.05$) on d 7, and tended to be higher ($P = 0.07$) during the whole experimental period for GrGH compared with GrC calves. Conversion of 2.5 mM [2-^{14}C]propionate and 2 mM [U-^{14}C]lactate into glucose was 1.9 ± 0.2 and 0.42 ± 0.05, respectively. Metabolism of 2.5 mM [2-^{14}C]propionate and 2 mM [U-^{14}C]lactate into CO_2 was 3.7 ± 0.3 and 0.31 ± 0.04, respectively. Gluconeogenesis and CO_2 production did not differ between groups. The data indicates effects of GH to accelerate growth performance in veal calves and to alter plasma glucose concentrations, but no effect of GH on hepatic gluconeogenesis.

Key Words: Veal Calves, Growth Hormone, Gluconeogenesis

1348 Effect of feed intake and genetic potential for milk yield on somatotropin (ST) response to growth hormone releasing factor (GRF) in Holstein heifers. W. J. Weber*, S. H. Wu, H. Chester-Jones, L. B. Hansen, and B. A. Crooker, *Department of Animal Science, University of Minnesota.*

Heifers (123 ± 5 d of age) from control (CL, n=6) and select (SL, n=7) lines were used to determine effects of selection on ST response to GRF. Milk yield of CL and SL cows in 1999 were 6,200 and 11,100 kg/305 d. Successive GRF challenges (C1, C2) were conducted when heifers were fed 150% (FF), 50% (HF) and 150% (RF) of maintenance during a 37 d study. Each heifer received 4 μg/100 kg BW of human GRF (1-29) analog (Hoffman-LaRoche, Ro23-7863) at 0 and 120 min. Blood samples were obtained at -30, -20, -10, -5, 0, 2.5, 5, 7.5, 10, 15, 20, 30, 45, 60, and 90 min relative to GRF challenge. Plasma ST was determined by RIA. Mean pre-challenge ST concentrations (PCST) were determined during the 30 min interval prior to C1 and C2 and transformed to natural logarithms. Area under the ST response curve was quantified (0 to 60 min post-dosing, AUC) by trapezoidal summation after subtracting baseline ST. Effects of line, intake, challenge and their interactions were assessed using GLM of SAS. Means differed when $P < 0.05$. PCST was greater for C2 than C1 suggesting potential carryover effects, thus only C1 data were used to evaluate PCST. There was a line by intake interaction with a reduced PCST during HF for CL ($\log_e$: 1.49^a, 0.85^b, 1.44^a ± 0.11 ng/ml) but not for SL ($\log_e$: 1.58^a, 1.40^a, 1.51^a ± 0.11 ng/ml). Plasma ST at time 0 was similar for C1 and C2 ($\log_e$: 1.40, 1.49 ± 0.05 ng/ml and used for AUC baselines. AUC was greater for C1 than C2 (1896, 1404 ± 138 ng·min·ml^{-1}), less at HF than FF or RF (1895^a, 889^b, 2164^a ± 168 ng·min·ml^{-1}) and did not differ between CL and SL (1501, 1798 ± 140 ng·min·ml^{-1}). Feed deprivation caused a reduction in PCST for CL but not SL heifers and reduced AUC in both CL and SL. Selection for milk yield had no affect on AUC. Results suggest mechanisms in the growing heifer that regulate adaptation of the somatotropic axis to feed deprivation may be affected by genetic potential for subsequent milk yield.

Key Words: Selection, GRF, GH

1349 Effect of feed intake, bST administration, and genetic potential for milk yield on hepatic IGF-I and growth hormone receptor mRNA in growing Holstein heifers. S. H. Wu*, W. J. Weber, H. Chester-Jones, L. B. Hansen, and B. A. Crooker, *Department of Animal Science, University of Minnesota.*

Heifers from an unselected (since 1964) stable milk yield line (Control, CL) and a contemporary line (Select, SL) were used to assess effects of selection for milk yield, bST, and energy and nutrient supply on components of the somatotropic axis. During the 37 d study, heifers (CL: n = 6, 120 ± 5 d old, 116 ± 10 kg BW; SL: n = 7, 124 ± 7 d old, 157 ± 12 kg BW) were fed 150% (d 1 to 18), 100% (d 19), 75% (d 20 to 21), 50% (d 22 to 26), 75% (d 27), 100% (d 28) and 150% (d 29 to 37) of their maintenance energy needs. Heifers received 30 μg bST/kg BW (IM) prior to the a.m. feeding on d 4 to 10. Blood samples were obtained prior to feeding and analyzed for IGF-I. Liver biopsies were collected on d 1, 8, 23, 26, 30, 33, and 36 and analyzed for IGF-I and growth hormone receptor (GHR) mRNA by RPA (reported as pixel densities relative to 18S RNA). Results were analyzed as a repeated measures analyses. Means differed when $P < 0.05$. Plasma IGF-I was greater in SL than CL heifers (170, 145 ± 4 ng/ml). Effects of bST, feed restriction, and refeeding were similar (223, 202; 65, 40; and 186, 175 ± 7 ng/ml) for each line. Relative to d 1, hepatic IGF-I mRNA decreased during feed restriction and increased with refeeding. There was a strong trend ($P = 0.07$ for line x day) for a more rapid recovery of IGF-I mRNA in SL heifers. Magnitude of the decrease in IGF-I mRNA (25% of d 1) was similar in CL and SL on d 23 but decreased in CL and increased in SL (13, 54% of d 1) on d 26. On d 30, IGF-I mRNA increased in CL and SL (80, 160% of d 1) and continued to increase through d 36 (160, 200% of d 1). Although not significant ($P = 0.14$), a similar line x day interaction occurred for liver specific GHR-1A mRNA. GHR-1A but not IGF-I mRNA was decreased by bST. Results suggest mechanisms in the growing heifer that regulate adaptation of the somatotropic axis to feed deprivation may be affected by genetic potential for subsequent milk yield.

Key Words: Selection, Liver, Somatotropic axis

1350 Comparison between measurement of parenchymal development of heifer mammary glands by computed tomography and traditional dissection techniques. J. M. Smith*[1], S. S. Block[1], N. L. Dykes[2], D. E. Bauman[1], and M. E. Van Amburgh[1], [1] *Cornell University, Ithaca, NY,* [2] *College of Veterinary Medicine, Cornell University, Ithaca, NY.*

In conjunction with a study of the effects of feeding diets containing specific fatty acids prior to puberty on mammary development in Holstein heifers, mammary glands were removed and imaged by computed tomography prior to traditional dissection. Pubertal heifers had been fed a ration containing calcium salts of saturated and unsaturated fatty acids at 2.6% DM for an average of 221 d and were, on average, 361 d of age at slaughter. Half-glands from three prepubertal (pre-treatment) and three pubertal heifers were scanned in transverse 2 mm sections, 8 mm apart, by computed tomography (Picker, PQS, Cleveland, OH). Digital images were analyzed using an image analysis program (QuantIm, Zedec Technologies Inc., Durham, NC). Several levels of contrast were selected as criteria for calculating parenchymal area using the program. Areas were converted to volumes by assuming constant area over the distance between scans. Correlations between the mass of dissected parenchyma and the calculated parenchymal volumes were 0.94 and 0.91 when contrast criteria 80 and 90 were used, respectively. Table 1. Age, body weight, days on treatment, parenchymal mass, and parenchymal volume[a] of mammary gland halves from six heifers.

Time	ID	Age, d	BW, kg	D on Trt	Mass, g	Vol 80	Vol 90
PreTrt	B2	134	131.5	0	72	42	39
	B3	132	131.1	0	70	29	27
	B7	153	107.0	0	33	6	5
Post	C3	356	340.6	203	637	452	285
	C6	396	342.0	287	630	636	433
	C10	332	298.5	174	554	717	516

[a]Volume in cubic cm calculated with contrast criteria set at value indicated.

Key Words: Dairy heifer, Mammary development, Computed tomography

1351 Effect of feeding a calcium salt of conjugated linoleic acid (CLA) prior to puberty on body composition and mammary development in Holstein heifers. J. M. Smith*, S. S. Block, D. E. Bauman, and M. E. Van Amburgh, *Cornell University, Ithaca, NY.*

Our objective was to determine whether feeding mixed isomers of CLA to Holstein heifers between weaning and puberty would affect body composition and mammary development measured at puberty. Twenty-eight purchased heifers between 3 and 5 mo of age were enrolled. Eight were slaughtered at 134 ± 7 d of age (119.2 ± 4.1 kg BW) for baseline data. The remaining 20 were assigned to either the control diet (CTRL) containing a commercial fat supplement at 2.6% DM or to the diet containing calcium salts of CLA (CLA) at 2.7% DM (total isomers at 1% DM). The diets were formulated to provide 1.01 kg/d ME- and 1.07 kg/d MP-allowable gain (CNCPS v. 4). Heifers were slaughtered during their third estrus cycle. One heifer on CTRL was lost from the study. Days on treatment were 251 and 259 d, respectively, for CTRL and CLA. Age at puberty, BW, body condition, body and mammary composition were analyzed by GLM (SAS System for Windows, v. 8). Significance required $P < 0.05$. Puberty was reached at 347 and 334 d, respectively, for CTRL and CLA ($P > 0.05$). Body weights at puberty (319.4 ± 13.7 kg) and slaughter (357.2 ± 13.1 kg) and body condition (3.16 ± 0.07) were not different. Two body components from CLA were lost. Body and mammary composition were not different between treatments. Fatty acids were extracted and methylated from samples of subcutaneous and mammary fat from all heifers. Component fatty acid profiles were analyzed by gas chromatography (Hewlett Packard, Wilmington, DE). Total CLA isomers were detectable only in samples from CLA. Table 1. Body and mammary composition of heifers slaughtered after puberty.

Component	CTRL	CLA
Total body fat, % dry EBW	57.4 ± 1.2	55.2 ± 0.9
Total body CP, % dry EBW	40.4 ± 1.9	41.9 ± 1.3
Total body ash, % dry EBW	7.9 ± 1.0	8.2 ± 0.3
Mammary half-gland wt, g	1354 ± 206.0	1229 ± 122.9
Parenchyma (half-gland), g	711 ± 82.1	609 ± 55.6
Parenchymal fat, % DM	89.0 ± 0.8	89.2 ± 0.7
Parenchymal CP, % DM	8.8 ± 0.9	9.1 ± 0.8
Parenchymal ash, % DM	0.5 ± 0.07	0.5 ± 0.06

1352 Leptin reduces proliferation of a bovine mammary epithelial cell line (MAC-T). L.F.P. Silva*, M.J. VandeHaar, M.S. Weber Nielsen, and B.E. Etchebarne, *Michigan State University, East Lansing MI.*

High-energy diets promoting body growth rates above 1 kg/d before puberty impairs mammary development. These rapid-growth regimens enable earlier calving but also lead to reduced milk production and increased volume of adipocytes in mammary parenchyma. We hypothesized that leptin, a protein produced by adipocytes, mediates the inhibitory effect of energy on mammary development. Our objective was to determine the effect of leptin on mammary epithelial cell proliferation. MAC-T cells, a bovine mammary epithelial cell line, were seeded onto 24-well plates coated with collagen-I, at a density of $5x10^3$ cells per well. Initial medium was composed of Dulbecco's Modified Eagle Media/F12 (DMEM/F12) supplemented with 10% fetal bovine serum (FBS). Cells were allowed to attach for 24h. Cells were then washed with Dulbecco's phosphate buffered saline and cultured in serum-free medium (SFM) for 48 h, after which the treatments were added. SFM consisted of DMEM/F12 containing bovine serum albumin (5μg/ml), insulin (1ng/ml), glutathione (1μg/ml), sodium selenite (1ng/ml), and transferrin (5μg/ml). The treatments consisted of increasing doses (0 to 160 ng/ml) of either ovine (oLeptin) or bovine leptin (bLeptin) together with 5 ng/ml IGF-I; or increasing doses of oLeptin (0 to 1,000 ng/ml) together with 1% FBS. Cells were grown in treatment media for three days with [^{3}H]-thymidine present (0.5 μCi/well) during the last day of incubation to measure rate of DNA synthesis. Excess of [^{3}H]-thymidine was removed, cells were lysed, and radioactivity measured in a liquid scintillation counter. Addition of recombinant ovine leptin (oLeptin) to media containing 5 ng/ml of IGF-I decreased cell proliferation linearly. The minimum dose of oLeptin that decreased ($P<0.05$) cell proliferation was 64 ng/ml (13% decrease). The highest dose of oLeptin (160 ng/ml) decreased IGF-I stimulated cell proliferation by 20% ($P<0.001$). Similar pattern was obtained using bLeptin. When MAC-T cells were stimulated to grow with 1% FBS, addition of oLeptin at 100 or 1,000 ng/ml decreased ($P<0.001$) cell proliferation by 27%. We conclude that leptin reduces IGF-I- or FBS-stimulated proliferation of MAC-T cells. We suggest that leptin may mediate the inhibitory effect of high-energy diets on mammary development.

Key Words: Prepubertal, Heifer, Mammary gland

1353 Effect of dietary energy and protein density on body condition and ovarian follicular dynamics in peripubertal dairy heifers. P.K. Chelikani*[1], J.D. Ambrose[2], and J.J. Kennelly[1], [1]*University of Alberta,* [2]*Alberta Agriculture, Food & Rural Development, Edmonton,Canada.*

Nutritional regulation of ovarian follicular dynamics in dairy heifers has received little attention. Our objectives were to determine the effects of diets varying in energy and protein levels on body condition and ovarian follicles during the pre and peripubertal period. Thirty Holstein heifers were randomly allotted (n=10/diet) at 100 kg body weight (BW) to either a Low (L: 2.25 ME Mcal/kg DM, 14% CP, ADG 0.5kg/d), Medium (M: 2.50 ME Mcal/kg DM, 16% CP, ADG 0.8kg/d) or High (H: 2.75 ME Mcal/kg DM, 18% CP, 1.1kg/d) diet. Ultrasound imaging of ovaries was performed on alternate days (d) for 22 d at 8 months (m) and for 45 d at 10 m of age (n=5/treatment). Serial blood samples for LH analysis were collected for 8 h at 8 and 10 m. At 1st ovulation, L heifers were 5.9 and 4.4 m older (P<0.01) than H or M heifers, respectively (L: 456 d, M: 324 d, H: 278, SE 16). BW at 1st ovulation did not differ (P>0.05) between treatments (H: 281 kg, M: 282 kg, L: 312 kg, SEM 11 kg). BCS and back-fat thickness (BF) at puberty were lower (P<0.05) in L compared to H or M (BCS L: 2.82, M: 3.20, H: 3.36, SEM 0.09; BF (mm) L: 2.62; M: 2.84; H: 3.36) heifers. Mean concentrations, pulse frequency and amplitude of LH at 8 m did not differ (P>0.05) between treatments. At 10 m, LH pulse frequency was lower (P<0.05), and amplitude greater (P=0.06) in L than H heifers (Pulses/8h: L: 1.17, M: 1.67 H: 2.6 SEM 0.40; Amplitude (ng/ml) L: 1.70, M: 1.14 H: 0.84, SEM 0.32). At 8m, the mean number of follicles #d10mm was lower (P=0.08) in L compared to H or M heifers (L: 0.27, M: 0.43, H: 0.47, SEM 0.07). Maximum diameter (mm) of dominant follicle (DF) at 8m was lower (P<0.01) in L than H or M heifers (L: 11.17, M: 13.00, H: 13.50, SEM 0.52). Size (mm) of the anovulatory DF at 10m was lower (P<0.05) in L than H or M (L: 11.25, M: 12.83, H: 13.40, SEM 0.48). Growth rate (mm/d) of the anovulatory DF was greater (P<0.01) in M than H or L (L: 0.64, M: 1.19, H: 0.67, SEM 0.12) heifers at 10 m. Size of the ovulatory DF was greater (P=0.05) for H than M (M: 11.80, H: 14.90, SEM 0.93) heifers. These findings suggest that diets differing in energy and protein density have a significant influence on body condition, onset of puberty and follicular dynamics during the pubertal transition in dairy heifers.

Key Words: Dairy heifers, Nutrition, Reproduction

1354 Analysis of the spatial, hormonal, and developmental regulation of the estrogen receptor α gene in cattle. M. J. Meyer*[1], R. P. Rhoads[1], A. L. Marr[2], W. R. Butler[1], Y. R. Boisclair[1], and M. E. Van Amburgh[1], [1]*Cornell University, Ithaca, NY,* [2]*Elanco Animal Health, Greenfield, IN.*

Estradiol elicits its biological effects by binding nuclear hormone receptors resulting in transcription of estrogen responsive genes. Two subtypes of this receptor, estrogen receptor α (ER α) and ER β, are known to exist. Despite the essential role of estradiol in driving a number of developmental processes, regulation of ER α has not been extensively studied in the bovine. A partial cDNA fragment corresponding to exon 1 of the bovine ER α was amplified by RT-PCR of total RNA extracted from bovine uterine tissue, and used to develop a ribonuclease protection assay (RPA). This assay was used to evaluate the spatial regulation of ER α gene expression in mature Holstein cows. ER α was expressed in the spleen, liver, adipose tissue, ovarian follicule, and uterus, but not in the heart, lung, or kidney. ER α was also expressed in parenchymal and extraparenchymal mammary tissues collected from both pre- and postpubertal Holstein heifers. In order to investigate transcriptional regulation of the ER α gene, nine nulliparous Holstein heifers were ovariectomized (OVX) at 2 years of age. OVX heifers were left untreated (n=3) or received estradiol delivered via an ear implant alone (E; n=3) or in concert with progesterone delivered via an intrauterine implant (E/P; n=3). After nine days of treatment, heifers were slaughtered and uterine tissue was collected for total RNA analysis. Uterine ER α gene expression was 2 fold greater in OVX than in either E and

E/P treated heifers ($p < 0.01$); E and E/P treated heifers did not differ. This RPA will be used to further evaluate the possible role of ER α receptor in regulating important processes such as mammary gland development.

Key Words: Bovine, Steroid, Gene Expression

1355 Effects of chronic in vitro growth hormone treatment on insulin receptor substrates and PI3 kinase in adipose tissue. F. Castro*[1], E. Delgado[2], and D. Lanna[2], [1]*University of California, Davis/ CA/ USA,* [2]*Esalq-USP/ SP/ Brazil.*

Growth hormone (GH) has profound effects on carbohydrate and lipid metabolism, including a reduction in adipose tissue sensitivity to insulin. The objective of the present study was to characterize some components that may be involved in the cellular mechanism of this insulin resistance. Concentrations of insulin receptor substrates (IRS) including IRS-1, IRS-3 and phosphatidylinositol 3-kinase (PI3K); phosphorylation of IRS-3; and association of IRS-1/PI3K and IRS-3/PI3K were evaluated in adipose tissue cultured in the absence or presence of GH. Male Wistar rats had their epididymal fat pads removed and placed in medium 199. In a first protocol adipose tissue explants were incubated in medium 199 for 48h with: a) 100 ng/ml of insulin plus 10 nM dexamethasone or b) 100 ng/ml of insulin plus 10 nM dexamethasone and 100 ng/ml of hGH (human GH). The second protocol evaluated the effects of chronic exposure to hGH on adipose tissue response to short-term insulin stimulus. Explants were incubated for 24h in medium 199 with: a) no additions or b) 100 ng/ml hGH. After 24h, half of the explants were stimulated for 20min with 1μg/ml of insulin and homogenized in extraction buffer. The concentration, phosphorylation state and association of the proteins were studied using western blots. In adipocytes exposed to hGH for 48 hours, the amounts of the IRS-1, IRS-3 and PI3K decreased by 336% ($P<.01$, n=6), 278% ($P<.05$, n=7) and 268% ($P<.05$, n=7), respectively. Consistent with effects observed after 48 hours, adipose tissue treated with hGH for 24h had concentrations of IRS-1, IRS-3 and PI3K decreased by 294% ($P<.01$, n=5), 153% ($P<.01$, n=8) and 295% ($P<.01$, n=6), respectively. Short-term insulin stimulation increased degree of phosphorylation and associations of IRSs ($P<.01$). Short-term insulin effects were altered by chronic in vitro incubation with hGH, including: amount of phosphorylated IRS-3 was reduced by 286% ($P<.05$, n=7), and the amounts of IRS-1 and IRS-3 associated with PI3K were reduced by 4420% ($P<.10$, n=6) and 284% ($P<.01$, n=6), respectively. Results of this study suggest that chronic GH treatment in vitro alters the early steps of insulin signal transduction in rat adipose tissue, including decreased IRS-3 concentration.

Key Words: Insulin, Growth Hormone, Insulin Receptor Substrate (IRS)

Milk Protein and Enzymes

1356 Characterization of carbohydrate structure of MUC1 and MUCX in porcine and bovine milk by exoglycosidase treatment and lectin blot test. C. Liu*, A.K. Erickson, and D.H. Francis, *South Dakota State University, Brookings, SD.*

Mucins are glycoproteins characterized by a high level of O-linked glycosylation of their core proteins. Two types of mucins, MUC1 and MUCX, are found to be present in porcine and bovine milk. Little information is available about their carbohydrate portion. This study employed exoglycosidase treatment together with lectin binding studies to determine the carbohydrate structure of MUC1 and MUCX in porcine and bovine milk. Treatment with neuraminidase reduced the mobility of both mucins on SDS gels in both species, indicating the existence of terminal sialic acid (NeuAc) residues; treatment with β-galactosidase had no effect on the mobility of native mucins but decreased the mobility of neuraminidase-treated mucins, suggesting terminal β-D-galactose occurs more commonly in the penultimate position. Twenty five lectins were used to further identify certain carbohydrate stucture associated with mucin core proteins. Regardless of species and mucin type, the presence of both N-linked and O-linked oligosaccharide chains was implicated by the binding of Concanavalin A and Jacalin. The presence of terminal N-acetyglucosamine (GlcNAc) was indicated by the binding of soybean agglutinin lectin (SBA) and *Vicia villosa* lectin (VVA), which both bind to MUC1 less strongly than to MUCX,implying a difference in terminal GalNAc abundance. Some carbohydrate structures were found to be species-specific and mucin type-specific. Bovine MUC1 contained exposed β-D-Gal-β(1,3)-D-GalNAc (T antigen) [peanut agglutinin (PNA) binding] while porcine MUC1 did not; terminal α(2,6)-linked NeuAc [elderberry bark lectin (EBL)] was found present in porcine MUC1 while it was lacking in bovine MUC1. Both porcine MUCX and Bovine MUCX contained exposed T antigen structure (PNA binding) and N-acetyglucosamine (GlcNAc) [*Solanum tuberosum* lectin (STL) binding]; however, bovine MUCX had α(2,6)-linked NeuAc [*Maackia amurensis* lectin II (MAAL II)] and terminal α(2,6)-linked NeuAc which were absent in porcine MUCX. The complexity and diversity of mucin glycosylation imply the functional importance of mucin carbohydrate. Further studies on the carbohydrate structure of milk mucins may contribute to an understanding of their possible functions between mother and young.

Key Words: Bovine milk mucins, Porcine milk mucins, Carbohydrate structure

1357 Structural studies of bovine β-casein by CD, FTIR and molecular modeling. P. X. Qi* and H. M. Farrell, Jr., *USDA-ARS-ERRC, Wyndmoor, PA, USA.*

The caseins of milk form micelles to carry the otherwise insoluble calcium and phosphate which are indispensable nutrients for humans. To better understand the molecular basis for the calcium-phosphate transport complex, we studied the major component of this complex: β-casein. The assumption that β-casein is #rheomorphic# remains controversial. In this work, we report our studies on the association reaction of β-casein to address the question of whether or not a conformational change in the monomer precedes aggregation, or occurs as a result of aggregation. Circular dichroism (CD) and Fourier transformation infrared (FTIR) spectroscopies were used to investigate the temperature-induced changes in the secondary structure of the β-casein under physiological relevant conditions (water, pH and low ionic strength). The degree of self-association under these conditions was assessed by analytical ultra centrifugation. CD and FTIR spectroscopies, as well as secondary structure predictions suggest the possible existence of polyproline II left-handed helices in β-casein. These short helices may play an important role in the self-association process. Furthermore, CD and FTIR results show that the β-casein may fold considerably prior to self-association, but may further respond to close packing in the polymer. The binding of β-casein to hydrophobic probe 1-anilino-8-naphthalenesulfonate (ANS) indicates it may be a molten globule-like protein. Molecular modeling techniques were used to not only generate a three-dimensional structure but also provide dynamic information for the self-association process as well as its function in calcium transporting.

Key Words: β-Casein, Structure, Polyproline II

1358 Conformational change in alpha-lacatalbumin produces an alternative biological function. K Stokes and B Alston-Mills*, *North Carolina State University, Raleigh, North Carolina, USA.*

Research within the past 10 years has suggested that the structure of alpha-lactalbumin is essential to its function, both as the modifier protein in the lactose synthase complex and as a modulator of mammary epithelial cell (MEC) proliferation. To directly test this hypothesis, we treated 3 mammary cell lines with 5 lots of bovine alpha-lactalbumin that differed in purity and tertiary conformation. Protein purity was determined by one- and two-dimensional PAGE and UV-spectroscopy and protein fold was determined by both flourescence and circular dichroism. Two lots were purchased from Sigma (98H7003 and 99H7029) and used without further purification. PAGE revealed several minor impurities in lot 98H7003, but the major species was alpha-lactalbumin. Two wild-type (wt) recombinant lots were expressed, folded and purified: lot wt01 was pure and correctly folded and lot wt02 was pure, but misfolded. A third recombinant protein, D87A, was pure, but unable to bind calcium and lacked a well-defined tertiary structure. Correctly folded native alpha-lactalbumin (99H7029) and wild-type (wt01) and mutant (D87A) recombinant proteins did induce inhibition of MEC