506 Control of cow hypocalcemia: Field application of ion-selective field effect transistor technology. E. M. Rodríguez*[1], A. Bach[1,2], N. Abramova[3], A. Bratov[3], A. Ipatov[3], and A. Arís[1], *[1]Department of Ruminant Production, IRTA, Caldes de Montbui, Spain, [2]ICREA, Barcelona, Spain, [3]BioMEMS Group, IMB-CNM, CSIC, Bellaterra, Spain.*

The early detection of subclinical levels of blood calcium (Ca) in the cow can avoid later health and production problems. Therefore, a field Ca sensor could be a powerful tool. The aim of this study was to develop a reliable prototype to quantify blood Ca in the cow, to define its specifications and to determine the influence of blood Ca daily circadian fluctuations in the periparturient cow. An analytical system based on ion-selective field effect transistor (ISFET) with Ca selective photocurable membranes was developed to offer a semiautomatic Ca analysis of serum (obtained by thrombin coagulation or centrifugation), plasma or direct blood. The sensor Ca measurements were validated by comparing the results with those obtained using the ICP-OES reference method. Cleaning solutions based on proteolytic enzymes in a neutral solvent, detergent or HCl were tested. Twelve Holstein multiparous cows were sampled at 24–48 h postcalving to analyze daily blood Ca changes at 0700, 1400, and 2000 h. Data were analyzed with a mixed-effect model with repeated measures. The precision of the analysis by ISFET was greater than reported for double charged ions with a σ within 3–7%. A linear response in the $0.1–10^{-5}$ *M* Ca concentration range and a detection limit of 3×10^{-6} *M* were obtained. Each analysis takes <20 min and only 100 μL of sample is needed. The serum samples obtained after thrombin coagulation or centrifugation and the plasma samples did not differ from the total Ca concentration determined using the reference method. However, direct blood measures differed from the reference measures and shortened the life of the ISFET to a single use. The best way to recuperate the sensors' response was the cleaning of the system with the proteolytic solution, although the detergent yielded positive outcomes as well. Blood Ca levels were not affected by daily circadian fluctuations. In conclusion, the results demonstrate that ISFET technology can be applied efficiently to analyze serum or plasma Ca in the cow as a new fast, reliable and inexpensive method. Also, blood samples from the periparturient cow can be obtained any time of the day.

Key Words: cow, hypocalcemia, ion-selective field effect transistor (ISFET)

507 Calcium urinary excretion in dairy cows with different levels of glucose tolerance. E. Schwegler*[1], F. da Rosa[1], A. Silva[1], E. Oliveira[1], P. Montagner[1], M. Weschenfelder[1], A. Krause[1], C. Brauner[1], E. Schmitt[2], V. Rabassa[1], A. Schneider[1], E. Xavier[1], F. Del Pino[1], and M. Correa[1], *[1]Federal University of Pelotas, Pelotas, RS, Brazil, [2]Brazilian Agricultural Research Corporation, EMBRAPA-CPAFRO, Porto Velho, RO, Brazil.*

During the peripartum dairy cows experience a transitory period of insulin resistance. In humans, insulin resistance is associated with increased urinary excretion of calcium (Ca). Therefore, the aim of this study was to assess the Ca urinary excretion during the peripartum in dairy cows with different levels of glucose tolerance during the prepartum period. Nineteen pluriparous Holstein cows were enrolled in this study. Glucose tolerance tests (GTT) were conducted at 20d prepartum. The GTT was based on an infusion of 500 mg/kg body weight of glucose and posterior determination of serum glucose concentrations at several time points up to 180 min and the calculation of the glucose area under the curve (AUC). The cows were categorized according to the rate of glucose metabolism into sensitive group (GS: higher glucose metabolism, 8,194 ± 388.6 mg/dl), intermediate group (GI, 12,079 ± 528.2 mg/dL) and resistant group (GR: lower glucose metabolism, 15,507 ± 292.4 mg/dL). Blood and urine samples were collected on d −23, −14, −7, −3, 0, 3, 6, 9, 16 and 23 from calving. Concentrations of creatinine (Creat) and Ca were analyzed in serum (S) and urine (U). The Ca excretion in the urine was estimated by calculating the fractional excretion (EF) using the formula: (UCa/SCa) × (SCreat/UCreat) × 100. Statistical analysis was performed using the SAS. According with the AUC categorization, 6 cows were in the GS, 7 cows were in the GI and 6 cows in the GR. The GR had higher fractional excretion of calcium in the prepartum (96.3 ± 10.4%) than the GS (59.2 ± 8.8%) ($P < 0.01$). Calcium excretion in GI (59.5 ± 7.6%) was similar to GS ($P > 0.05$), but lower than GR ($P < 0.05$). In the postpartum period the GR (27.7 ± 3.7%) had an increased Ca excretion than GS (16.1 ± 3.3%) ($P = 0.02$), although GI similar between GR and GS (21.9 ± 3.0%) ($P > 0.05$). In summary, the present study indicates that prepartum cows less tolerant to glucose excrete more urinary calcium in the pre and postpartum periods. More studies to understand the potential effects of these results on the etiology of hypocalcemia are necessary.

Key Words: glucose tolerance, peripartum, calcium urinary excretion

508 The association of postpartum calcium concentration with body weight change and milk production in dairy herds with automatic milking systems during the first 30 days in milk. L. S. Caixeta*[1], P. A. Ospina[1], S. K. Johnson[1], M. Capel[2], and D. V. Nydam[1], *[1]Cornell University, Ithaca, NY, [2]Perry Veterinary Clinic, Perry, NY.*

The objectives were to characterize calcium concentration variability and evaluate the association between subclinical hypocalcemia (HPC) and changes in body weight (BW) and milk production (MP) within the first 30 d in milk (DIM) in herds with automatic milking systems. In a prospective cohort study of 3 herds, 105 dry cows were enrolled and followed until 30 DIM. Serum samples were analyzed for calcium at 1, 2 and 3 DIM. Based on previous reports, HPC was defined as having at least one reading between 6 and 8 mg/dL and treated as a dichotomous variable in the analysis. Daily measurements of BW and MP were used to estimate BW change over time and total MP in the first 30 DIM. Occurrence of any disorders: displaced abomasum, ketosis, milk fever, retained placenta or metritis, was also included in the analysis as a dichotomous covariate. The Mixed procedure in SAS was used to evaluate the association between HPC, disease, herd, and any biologically relevant interactions with BW and MP. HPC was found in 17% of primiparous and 65% of multiparous animals. Both disease and HPC status were significant predictor variables ($P < 0.01$), thus the result of the interaction between them is reported in Table 1. Subclinical hypocalcemia was not a significant predictor of milk production in primiparous animals, but multiparous cows with HPC produced 45.5 kg ($P < 0.01$) less milk than those without HPC during the first 30 DIM. Mature animals with HPC exhibited more rapid weight loss and produced less milk than their normocalcemic counterparts.

Table 1. Body weight change (kg) per day over the first 30 DIM and the interaction between disease (Dz) and subclinical hypocalcemia (HPC) by parity group; ll interactions had $P < 0.01$.

	Parity 1	Parity 2	Parity ≥3
No Dz × No HPC	−0.42	−1.44	−1.25
No Dz × HPC	−1.35	−1.22	−1.75
Dz × No HPC	−1.67	−2.40	−0.75
Dz × HPC	−1.10	−0.66	−2.13

Key Words: hypocalcemia, body weight, milk

509 Effects of protein supplementation of fall calving cows during breeding and lactation on growth and concentrations of IGF-I in plasma of beef calves. K. J. McLean*, B. H. Boehmer, L. J. Spicer, and R. P. Wettemann, *Oklahoma Agricultural Experiment Station, Stillwater.*

Fall calving cows grazing dormant native grass pasture were used to evaluate effects of protein supplementation during breeding and the first trimester of gestation on postnatal growth and concentrations of insulin and IGF-I in plasma of calves. Cows calved in Sept-Oct, grazed in one pasture, and were individually fed control (C, 1.82 kg/d of 38% CP supplement, n = 22) or low (L, 0.2 kg/d of 8% CP supplement, n = 21) from Nov 17 to March 20. Cows were exposed to bulls for 60 d commencing Dec 1. During lactation the subsequent year, half of the cows on C and L prenatal treatments was assigned to C and the other half was assigned to L. Birth weight, weaning weight, insulin, IGF-I, and plasma proteins were analyzed as a 2 × 2 factorial using the MIXED procedure of SAS. Birth weight and BW at weaning (205 d) were not influenced ($P = 0.76$) by prenatal treatment. Weaning weight (205 d) was greater ($P = 0.02$) for calves on postnatal C (200 ± 5 kg) compared with L (184 ± 5 kg). Neither prenatal nor postnatal treatment of dams influenced preweaning concentrations of insulin in plasma of calves ($P > 0.14$). Plasma concentrations of proteins and IGF-I were not influenced by prenatal or postnatal treatment in Dec ($P > 0.50$ and $P > 0.22$, respectively). Postnatal C calves had greater ($P = 0.03$) concentrations of IGF-I (21.5 ± 2.1 and 14.8 ± 2.1 ng/mL, respectively) and less plasma proteins (6.6 ± 0.1 and 6.8 ± 0.1 g/100 mL, respectively, $P = 0.05$) compared with L calves in Jan. Steer calves tended to have greater ($P = 0.06$) concentrations of IGF-I compared with heifers at weaning (29.9 ± 2.6 and 22.1 ± 2.9 ng/mL, respectively). Calves with L dams both prenatally and postnatally, had less plasma proteins at weaning ($P = 0.02$) compared with the other treatments. Postnatal performance of calves was not influenced when dams were supplemented with protein during breeding and early gestation that resulted in less BW loss and greater plasma IGF-I concentrations (J. Anim. Sci. 90 (E-Suppl. 3): 325, 2012), however, protein supplementation of dams during lactation increased growth of calves.

Key Words: nutrition, IGF-I, calf

510 Lipogenic-associated gene activity of adipose tissue from beef heifers and relation to production and reproductive traits. L. A. Rempel*, R. A. Cushman, T. G. McDaneld, J. R. Miles, L. A. Kuehn, and A. K. Lindholm-Perry, *USDA, ARS, U.S. Meat Animal Research Center, Clay Center, NE.*

Transition from adolescence to puberty is marked by changes in metabolic activity. Fatty acid synthase (FASN) catalyzes the de novo synthesis of fatty acids and increased expression has been linked to excess energy intake and obesity. The enzyme, DNA-protein kinase (DNA-PK) plays a role in DNA damage repair, but can also influence transcription of lipogenic genes, including FASN. The objectives of our study were to (1) evaluate the expression of these lipogenic-affiliated genes, FASN and DNA-PK, at one year of age and (2) evaluate their association with production and reproductive traits in crossbred heifers. Subcutaneous adipose tissue was collected at approximately one year of age from 130 crossbred beef heifers. Total RNA was extracted from adipose tissue and reverse transcribed to generate cDNA for real-time PCR. Raw expression data was log-transformed and normalized against 2 housekeeping genes. Relative expression of FASN and DNA-PK were not correlated ($P = 0.6148$) with each other. The relationship between gene expression and growth and reproductive traits was modeled using a regression analysis with covariates heterosis and breed included. Relative expression of adipose FASN in beef heifers at one year of age had a positive association ($P \leq 0.0019$) with ADG (0.08 ± 0.024 kg) and weight at puberty (85.3 ± 25.86 kg). Similarly, relative DNA-PK expression was also positively associated ($P \leq 0.0042$) with ADG (0.12 ± 0.041 kg) and weight at puberty (146.7 ± 40.75 kg). Moreover relative DNA-PK expression at one year of age in beef heifers was positively associated ($P = 0.0004$) with age at puberty (75.8 ± 20.94 d) while a negative association ($P = 0.0610$) was observed with weight at first breeding (−37.7 ± 19.94 kg). Relative expression of FASN and DNA-PK in adipose from heifers at 1 yr of age was related to growth parameters; however, age at puberty and weight at first breeding were affiliated with DNA-PK expression only. DNA-PK may be contributing to metabolic gene activation, which influences growth and puberty.

Key Words: heifer, adipose, pubertal

511 Endocrine profile and hepatic gene expression in Holstein cows with different nutritional managements during early lactation. A. L. Astessiano*[1], P. Chilibroste[2], M. Fajardo[2], J. Laporta[1], J. Gil[3], D. Mattiauda[2], A. Meikle[4], and M. Carriquiry[1], *[1]School of Agronomy, UDELAR, Montevideo, Uruguay, [2]School of Agronomy, UDELAR, Paysandú (EEMAC), Uruguay, [3]School of Veterinary Medicine, UDELAR, Paysandú (EEMAC), Uruguay, [4]School of Veterinary Medicine, UDELAR, Montevideo, Uruguay.*

Multiparous cows (n = 25) were used in a randomized block design to study the effects of nutrition during the first 60 d postpartum on endocrine profiles and hepatic gene expression. Cows were assigned to 3 treatments (TREAT): TMR = total mixed rations (30 kg DM/d offered; 45% forage, 55% concentrate); G1 = 50% pasture in one (am) grazing session (6 h; pasture allowance = 15 kg DM/d) + 50% TMR (15 kg DM/d offered); and G2 = 50% pasture in 2 (am/pm) grazing sessions (9 h; pasture allowance = 15 kg DM/d) + 50% TMR (15 kg DM/d offered). Plasma and liver biopsies were collected at −40, −20, +10 and +55 DPP. Gene expression was quantified by real time PCR. Means from a repeated measures analysis differed when $P < 0.05$. Insulin decreased around calving in G1 and G2 cows and in early lactation tended to be greater ($P = 0.06$) for TMR than G1 cows (19.4, 15.1 and 17.3 ± 1.3 uIU/mL for TMR, G1 and G2). Adiponectin increased from pre to postpartum (77%) and during postpartum was greater in G2 than G1 cows (52.5, 87.1 and 132.7 ± 18.2 ng/mL for TMR, G1 and G2). Leptin decreased from −40 to +10 d and increased thereafter recovering values of prepartum, and during the postpartum were greater in G2 than G1 cows (4.1, 3.1 and 4.5 ± 0.4 ng/mL for TMR, G1 and G2). Hepatic INSR mRNA increased 1.5-fold at +55 d for all TREAT. The LEPR-b mRNA was greater in G2 than G1 cows during early lactation (1.57, 1.10 and 1.91 ± 0.3 for TMR, G1 and G2) as its expression increased from pre to postpartum in TMR and G2 but not in G1 cows. Although ADIPOR1 mRNA did not vary due to TREAT or days of lactation, ADIPOR2 mRNA increased 2.5-fold from pre to postpartum in all TREAT. Expression of ANGPTL4 mRNA tended ($P = 0.07$) to be affected by TREAT being greater in TMR than G2 cows during the postpartum (2.36, 1.97 and 1.05 ± 0.4 for TMR, G1 and G2) as it increased from pre to postpartum in TMR and G2 but remained stable in G1 cows. Cows fed TMR showed better metabolic status (greater BCS, insulin and, ANGPLT4 mRNA) while G2 showed a differential partitioning of energy (body reserves vs. activity) during early lactation associated with the feeding strategy postpartum

Key Words: nutrition, dairy cattle

512 Effects of maintenance energy requirements of gestating beef cows on plasma concentrations thyroxine, triiodothyronine, and rectal temperature. B. H. Boehmer*, K. J. McLean, and R. P. Wettemann, *Oklahoma Agricultural Experiment Station, Stillwater.*

Spring-calving, Angus cows (n = 29) were used to evaluate the effects of maintenance energy requirements on plasma concentrations of thyroxine (T_4), triiodothyronine (T_3), and rectal temperature (RT). Nonlactating cows (5 to 10 yr of age, 144 ± 2 d of gestation) with a BW of 552 ± 8 kg and BCS of 4.3 ± 0.1 were individually fed a maintenance diet (Ne_m, Model 1, NRC 1996) for 31 d. Body weights were obtained twice weekly and daily feed intake was adjusted every 2 wk until constant BW was achieved (maintenance, MR). Mean MR of cows was 83.4 kcal/kg $BW^{0.75}$/d. Cows (n = 20) were infused with TRH (0.33 μg/kg BW) after consuming maintenance diets for 21 d. Blood plasma was collected at 0, 30, 60, 90, 120, 150, 180, and 240 min after TRH challenge and T_3 and T_4 were quantified in plasma by RIA. Maintenance energy requirements were used to classify cows as low (L; >0.5 SD less than mean, 79.4 kcal/kg $BW^{0.75}$/d), moderate (M; ± 0.5 SD of mean, 83.4 kcal/kg $BW^{0.75}$/d) or high (H; > 0.5 SD more than mean, 86.8 kcal/kg BW $^{0.75}$/d). Thyroxine, T_3 and RT were analyzed with PROC MIXED (SAS Inst. Inc.) and BW was analyzed with PROC GLM (SAS Inst. Inc.). Plasma concentrations of T_3 were greater in H and M cows (P = 0.04; 0.63 and 0.66 ng/mL, respectively) compared with L cows (0.58 ng/mL). Plasma T_3 increased after TRH to a maximum at 180 min in H (0.74 ng/mL) and M cows (0.78 ng/mL) and at 120 min in L cows (0.63 ng/mL). There was a MR × time effect ($P < 0.05$) for T_4 in plasma. Maintenance requirement of cows did not influence T_3:T_4 in plasma (P = 0.59) or RT (P = 0.46). Thyroid hormones influence metabolism and may be useful in determining maintenance requirements of beef cows. Production efficiency of beef cows may be improved by identifying cows that require less energy input while retaining performance.

Key Words: beef cattle, maintenance, thyroid

513 Comparisons of the transcriptome profiles of adipose tissue from beef and dairy cattle. J. Thomson*[1,2], P. Stothard[2], and J. P. McNamara[3], *[1]Montana State University, Bozeman, [2]University of Alberta, Edmonton, AB, Canada, [3]Washington State University, Pullman.*

RNA-seq, an application of next-gen nucleic acid sequencing technologies has enabled us to look at the transcriptomes of tissues at a given point of time with extremely high resolution. This technique has been used with great success to evaluate the effect of perturbations or treatments on gene expression. In this study we evaluated the functional and metabolic differences in transcriptome profile of 2 breeds of cattle with a history of divergent selective pressure and resulting differences in metabolism and gene expression: Holstein dairy cows and crossbred beef cows of Hereford and Angus genetics. RNA was isolated from 8 Holstein animals and 10 beef animals. The RNA was pooled within breed, enriched using poly-T oligo attached beads, and then cDNA libraries were synthesized and sequenced. Reads were aligned to UMD 3.1 and normalized read counts were used to calculate expression values for coding genes, alternative splice variants, and non-coding RNA transcripts. Functional analysis was conducted using Genesifter software from Geospiza and DAVID bioinformatics software version 6.7. Over 10 million reads were mapped to the reference sequence and expression values were generated for over 15,000 genes and over 30,000 potential splice variants in each breed. Differentially expressed genes were determined using Genesifter software pairwise *t*-test and Bonferroni correction for multiple comparisons with a significance threshold of adjusted $P < 0.05$. As expected the adipose tissue in both beef and dairy highly expresses genes associated with lipid transport and metabolism such as fatty acid binding protein 4 (FABP4) and perilipin (PLIN) both not statistically different. Definite differences were observed with dairy transcripts being enriched for oxidative phosphorylation (113 transcripts, $P < 0.005$) and cell cycle pathways (109 transcripts, $P < 0.005$) while beef adipose transcripts were enriched for insulin signaling (105 transcripts, $P = 0.01$) and adipocytokine signaling (58 transcripts, $P = 0.003$). This may provide insight into the different roles of adipose tissue in breeds of cattle selected for different purposes.

Key Words: RNA-seq, genomics, physiology

514 Alterations in body mass and inflammometabolic indices in Holstein cows fed different levels of energy and receiving 2,4-thiazolidinedione. A. Hosseini*[1], E. Trevisi[2], F. T. da Rosa[1], G. Bertoni[2], J. K. Drackley[1], and J. J. Loor[1], *[1]University of Illinois, Urbana, [2]Università Cattolica del Sacro Cuore, Piacenza, Italy.*

Overfeeding energy during the dry period increases the incidence of metabolic disease postpartum. Dry matter intake (DMI), body condition score (BCS) and blood inflammometabolic markers can serve as reliable indicator of health, inflammation and liver function. We evaluated DMI, BCS, metabolism, inflammation, and liver function in response to level of dietary energy and 2,4-thiazolidinedione (TZD) administration. Fourteen dry non-pregnant Holstein cows were assigned to treatments in a randomized block design. All cows were fed a control diet (CON; NEL = 1.32 Mcal/kg) to meet 100% of NRC requirements for 3 wk, after which half of the cows were assigned to a moderate-energy diet (OVE; NEL = 1.54 Mcal/kg) and half of the cows continued on CON for 6 wk. The OVE diet was fed ad libitum and resulted in cows consuming ~180% of NRC. CON cows were fed to consume only to 100% of NRC. All cows received 4 mg TZD/kg of BW daily starting 2 wk after the initiation of treatments and for 2 additional wk. The last 2 wk of the study served as the washout period. BW and BCS were recorded twice a week, while DMI was recorded daily during the entire study. Blood was harvested frequently during wk −1 to 6 for measurement of metabolites and hormones. Data were analyzed using PROC MIXED of SAS. In OVE compared with CON, the BW, DMI and DMI as a percentage of BW increased over time ($P < 0.001$), while BCS remained unchanged. The concentration of glucose, hydroxybutyrate (BHBA), cholesterol and aspartate aminotransferase-oxaloacetic transaminase (AST-GOT) increased ($P \leq 0.04$) in OVE, but paraoxonase (PON) decreased ($P \leq 0.03$) over time compared with CON. Overall concentration of NEFA and haptoglobin was lower ($P < 0.05$) in OVE than CON. An improvement of energy balance status in OVE cows was observed without a negative effect of TZD on DMI. The changes in AST-GOT and PON might reflect the effects of OVE on hepatic function. Excess dietary energy did not enhance inflammation and oxidative stress. In contrast with previous studies, TZD did not improve insulin sensitivity beyond what was observed with OVE alone.

Key Words: dairy cow, metabolic adaptation, 2,4-thiazolidinedione (TZD)

515 Adipose tissue insulin sensitivity in response to level of dietary energy and 2,4-thiazolidinedione in Holstein cows. A. Hosseini*, J. S. Osorio, F. T. da Rosa, J. K. Drackley, and J. J. Loor, *University of Illinois, Urbana.*

Mechanisms regulating insulin sensitivity in subcutaneous adipose tissue (SAT) of dairy cattle fed different levels of dietary energy during the prepartal period remain largely unknown. In monogastric SAT, 2,4-thia-

zolidinedione (TZD), a ligand of peroxisome proliferator-activated receptor-g (PPARG), has insulin-sensitizing effects. The specific aim of our study was to examine mechanisms whereby feeding a control or moderate-energy diet alter insulin sensitivity before, during, and after injection of TZD by evaluating insulin signaling-related genes in SAT. Fourteen dry non-pregnant Holstein cows were assigned to treatments in a randomized block design. All cows were fed a control diet (CON; NEL = 1.32 Mcal/kg) to meet 100% of NRC requirements for 3 wk, after which half of the cows were assigned to a moderate-energy diet (OVE; NEL = 1.54 Mcal/kg) and half of the cows continued on CON for 6 wk. The OVE diet was fed ad libitum and resulted in cows consuming ~180% of NRC. CON cows were fed to consume only to 100% of NRC. All cows received an intravenous injection of 4 mg TZD/kg of BW daily into the jugular vein starting 2 wk after the initiation of treatments and for 2 additional wk. The last 2 wk of the study served as the washout period. Biopsies of SAT were harvested at 2 (before TZD injection), 3, 4 (end of TZD), and 5 wk. Genes chosen for study included insulin signaling-related (INSR, IRS1, SLC2A4) and adipogenic/lipogenic enzymes/inducers (CEBPA, ADIPOQ, ADIPOR1, PPARG, SREBF1, FASN, SCD, DGAT2, INSIG1). Data were analyzed using PROC MIXED of SAS. In OVE cows, expression of PPARG and FASN was greater ($P < 0.05$) before and at the end of TZD injection. However, TZD injection decreased expression of IRS1 ($P = 0.04$), PPARG ($P = 0.05$), FASN ($P < 0.001$) and SREBF1 ($P = 0.02$) in OVE but not in CON cows. Results indicated that OVE did not decrease insulin sensitivity, and TZD actually decreased it.

Key Words: insulin sensitivity, 2,4-thiazolidinedione (TZD), dairy cattle

516 Overfeeding energy increases visceral fat deposition and alters metabolic indices in Holstein cows. A. Hosseini*[1], E. F. Garrett[3], E. Trevisi[2], F. T. da Rosa[1], G. Bertoni[2], J. K. Drackley[1], and J. J. Loor[1], *[1]University of Illinois, Urbana, [2]Università Cattolica del Sacro Cuore, Piacenza, Italy, [3]Department of Veterinary Clinical Medicine, University of Illinois, Urbana.*

Our objective was to examine the effect of overfeeding a moderate-energy diet on performance, visceral depot weights, body condition score (BCS), body weight (BW), and blood metabolites in dry non-pregnant cows. Fourteen Holstein cows (BCS = 3.31 ± 0.14) were assigned to treatments in a randomized block design. All cows were fed individually a control diet (CON; NEL = 1.32 Mcal/kg) to meet 100% of NRC requirements for 3 wk, after which half of the cows were assigned to a moderate-energy diet (OVE; NEL = 1.54 Mcal/kg) and half of the cows continued on CON for 6 wk. The OVE diet was fed ad libitum and resulted in cows consuming energy at ~180% of NRC. CON cows were fed to consume only to 100% of NRC. The BW and BCS were measured from wk −3 to 6, while the blood samples were collected before slaughter and several metabolites and hormones were measured. The DMI was recorded from −1 wk through slaughter on a daily basis. The wk before slaughter, OVE cows had greater concentration of BHBA (0.43 vs. 0.22; $P < 0.001$), cholesterol (3.77 vs. 2.65; $P = 0.008$) and AST-GOT (78.77 vs. 64.33; $P = 0.04$); whereas, the concentration of NEFA (0.07 vs. 0.17; $P = 0.002$) and bilirubin (0.89 vs. 1.5; $P = 0.005$) was lower in OVE cows. OVE cows had greater ($P < 0.001$) BW (757.5 vs. 692.5), DMI (kg/d; 17.20 vs. 8.02) and DMI as a percentage of BW (2.20 vs. 1.18); whereas, the BCS (3.4 vs. 3.6) and empty carcass weight (468.43 vs. 525.91) remained unchanged. In OVE cows, weight of the mesenteric (15.49 vs. 8.1; $P \leq 0.01$) and perirenal fat mass (11.17 vs. 3.39; $P \leq 0.04$), and liver (11.4 vs. 7.82; $P < 0.001$) was greater. Omental fat mass (15.16 vs. 23.41) did not differ. The similar BCS between the 2 diets and the fact that OVE cows had greater internal fat deposition suggests that BCS provided little information on visceral fat mass. Overfeeding energy did not impair insulin sensitivity but seemed to affect hepatic function.

Key Words: dairy cow, plane of energy, visceral fat

517 Level of dietary energy alters in vitro bovine adipose tissue insulin sensitivity and inflammatory response to TNF-α. A. Hosseini*[1], K. M. Moyes[2], F. T. da Rosa[1], J. K. Drackley[1], and J. J. Loor[1], *[1]University of Illinois, Urbana, [2]University of Maryland, College Park.*

Exogenous TNF-α increases liver triacylglycerol accumulation in late-lactation dairy cattle. In rats, an acute food deprivation resulted in greater periepididymal adipose TNF-α production. The objective of this study was to determine the acute in vitro effects of bovine insulin, recombinant bovine TNF-α, and their combination on subcutaneous adipose tissue (SAT) insulin sensitivity in dairy cattle fed different levels of dietary energy. Adipose tissue from the tail head was obtained at slaughter from 7 cows fed a control diet to meet 100% of NRC requirements and 7 cows fed a moderate-energy diet (OVE; ~180% of NRC requirements) for 6 wk. Tissue was transported on ice to the laboratory in an endotoxin free DMEM/Ham's F-12, L-Glutamine medium supplemented with Pen/Streptomycin as basal medium. After removing blood and connective tissue, 500 mg were dissected and cut to smaller pieces. Incubations were performed at 37°C and 5% CO_2 in duplicate for 2 h using 4 mL of basal media (control) or control with 1 μmol/L of bovine insulin (INS), 5 ng/mL of bovine recombinant TNF-α (TNF), or the same concentration of INS and TNF (IN-TNF). Data were analyzed using PROC MIXED of SAS. Genes chosen for study included insulin signaling-related (INSR, IRS1, SLC2A4), inflammatory and anti-inflammatory regulators (NFKB1, TNF, IL-1, IL-6, IL-10, SAA3 and HP) and adipogenic/lipogenic enzymes/inducers (CEBPA, ADIPOQ, ADIPOR1, PPARG, SREBF1, FASN, SCD, DGAT2, INSIG1). In CON cows, TNF and IN-TNF reduced ($P < 0.05$) the mRNA of IRS1. In OVE cows, TNF and IN-TNF led to greater ($P < 0.001$) mRNA of NFKB1 compared with control cultures; whereas, in CON cows the mRNA of TNF was lower ($P < 0.05$) in cultures with INS than control. Compared with control cultures, the mRNA of NFKB1 increased ($P < 0.001$) with TNF, and IN-TNF in CON cows. Our results indicate that SAT of OVE cows is more responsive to inflammatory cytokines. Moreover, exogenous insulin did not seem to improve its sensitivity in SAT during the inflammatory insult.

Key Words: subcutaneous adipose tissue, TNF-α, explant