Physiology and Endocrinology: Effects of nutrition and metabolism on ruminant reproduction

M209 Consistency of metabolic responses to nutrient deficiencies in early and mid-lactation of dairy cows. Josef J. Gross* and Rupert M. Bruckmaier, *Veterinary Physiology, Vetsuisse Faculty University of Bern, Bern, Switzerland.*

The adaptive response to a given metabolic load during energy deficiencies varies considerably among animals. The aim of this study was to investigate if individual cows respond in a repeated manner to a negative energy balance (NEB) in early and mid-lactation. Twenty-five multiparous Holstein dairy cows (3.0 ± 1.1 parities; mean ± SD) experienced a NEB during the first weeks of lactation, and after getting into a positive EB, cows were exposed to a 3-wk feed-restriction period providing 50% of estimated requirements starting at around 100 DIM. Dry matter intake and milk yield were recorded daily. Blood samples for analysis of plasma concentrations of glucose, NEFA, BHBA, cholesterol, and IGF-1 were obtained once weekly. Retrospectively, cows were ranked according to plasma NEFA concentration in early lactation. The cows with the 33% highest and 33% lowest NEFA concentrations (8 animals each) were selected and classified either a high response (HR) or a low response (LR) group, respectively. Data were analyzed using MIXED models in SAS including wk, group (HR or LR), parity, and the wk × group interaction as fixed effects and the individual cow as repeated subject. Differences between HR and LR over time were detected by the Bonferroni *t*-test. Significant effects were assumed at a level of $P < 0.05$. Before parturition, no differences were detected between LR and HR. After parturition, milk yield and ECM were higher for HR (milk yield: +7.4 kg/d, ECM: +4.9 kg/d from wk 2 to 14 pp) compared with LR ($P < 0.05$). Although plasma concentrations of glucose and cholesterol showed group differences in early lactation, but not during feed-restriction, concentrations of NEFA, BHBA, and IGF-1 showed a consistent similarly directed, but different response to a NEB at the 2 stages of lactation despite a similar EB. HR had higher NEFA (+0.8 mmol/L in wk 2 pp, +0.2 mmol/L in wk 2 of feed-restriction), BHBA (+1.3 mmol/L in wk 3 pp, +0.3 mmol/L in wk 1 of feed-restriction) and lower IGF-1 concentrations (−48.7 ng/mL in wk 2 pp, −54.9 ng/mL in wk 2 of feed-restriction) compared with LR at the 2 stages of lactation ($P < 0.05$). Cows responding to high milk production and negative energy balance with higher plasma NEFA and BHBA did again respond with higher NEFA and BHBA to a negative energy balance in later lactation as compared with cows with lower NEFA and BHBA concentrations with similar production levels.

Key Words: adaptation, metabolic plasticity, negative energy balance

M210 Relationship of dietary and serum phosphorus during the transition period to fertility measures. Ellen R. Jordan[1], Kevin J. Lager[1,2], J. Armando Garcia Buitrago*[3], Don R. Topliff[4], and Pablo J. Pinedo[5], *[1]1. Texas A&M AgriLife Extension, Dallas, TX, [2]2.West Texas A&M University, Canyon, TX, [3]3. New Mexico State University, Clovis, NM, [4]4. Angelo State University, San Angelo, TX, [5]Texas A&M AgriLife Research, Amarillo, TX.*

The objective was to evaluate the relationship between dietary and serum phosphorus (P) during the transition period and its relationship to fertility measures. Blood samples were collected into vacuum tubes at the morning feeding from cows (n = 4129) in 8 Holstein (H) herds in the summer (S) and winter (W), as well as 8 Jersey (J) herds in the S. Samples were placed on ice immediately. After processing, samples were stored at −20°C until analysis. On the day of sampling, total mixed ration samples were collected for subsequent analysis. Associations were tested by logistic regression, correlation analysis, and ANOVA. During S, P in the ration (% DM) averaged 0.35 ± 0.05, prepartum H; 0.45 ± 0.12, prepartum J; 0.42 ± 0.03, postpartum H; and 0.47 ± 0.07, postpartum J; and during W averaged 0.35 ± 0.05, prepartum H and 0.42 ± 0.03, postpartum H. Serum P values (mg/dl) for wk −3, −2, −1, 1, 2, and 3 relative to calving were 5.23 ± 0.09, 5.26 ± 0.19, 5.34 ± 0.10, 4.79 ± 0.12, 4.97 ± 0.12, and 5.40 ± 0.10 for S-H; 6.38 ± 0.09, 6.30 ± 0.07, 6.21 ± 0.07, 5.88 ± 0.09, 5.78 ± 0.08, 5.65 ± 0.10 for S-J; and 5.23 ± 0.12, 5.29 ± 0.20, 5.71 ± 0.1, 5.08 ± 0.13, 5.44 ± 0.11, and 5.53 ± 0.10 for W-H. The ration and serum P correlation coefficients ($P < 0.0001$) ranged from 0.14 over all weeks to a high of 0.34 for wk 3. During S, serum P concentrations were significantly associated with breed for wk −3, −2, −1, 1 and 2 ($P < 0.0001$), while lactation was significant for wk −1, wk 2, and wk 3 ($P = 0.0157$) and tended to be significant during wk −2 ($P < 0.0575$). In univariable analysis, P serum concentration appeared to be associated with days to first service (DFS) and days open (DO); however when season and dairy were added in multivariate analysis, P serum concentration was no longer significant ($P > 0.10$). When estimating the odds of pregnancy in either the first 90 (P90) or 150 (P150) d postpartum, serum P concentration was not significant in the multivariate analysis; when season and lactation were included. Serum P concentrations in the peripartum period were correlated with P intake; however, the main factors influencing the reproductive factors of DFS, DO, P90, and P150 were breed, season and lactation rather than serum P concentration.

Key Words: phosphorus, transition cow

M211 Reproductive performance of lactating dairy cows with an extended duration of the postpartum voluntary waiting period and injectable trace mineral supplementation. Matias L. Stangaferro*[1], Robert Wijma[1], Magdalena Masello[1], Rodrigo C. Bicalho[2], Mark J. Thomas[3], and Julio O. Giordano[1], *[1]Department of Animal Science, Cornell University, Ithaca, NY, [2]Department of Population Medicine and Diagnostic Sciences, Cornell University, Ithaca, NY, [3]Dairy Health and Management Services, Lowville, NY.*

Objectives were to evaluate (1) the effect of extending the duration of the voluntary waiting period (VWP) from 60 to 88 DIM; and (2) the effect of injectable trace mineral (Zn, Mn, Se, Cu) supplementation (TMS) on reproductive performance of dairy cows. Holstein cows [n = 1,105; 441 primiparous (PP) and 664 multiparous (MP)] were blocked by parity and total milk production in their previous lactation (MP only) and assigned to a 2x2 treatment arrangment to receive: TMS or NoTMS and first timed-AI (TAI) at 60 ± 3 (SVWP) or 88 ± 3 DIM (LVWP) after synchronization of ovulation with the Double-Ovsynch protocol (GnRH-7d-PGF-3d-GnRH-7d- GnRH-7d-PGF-56h-GnRH-16h-TAI) resulting in the following groups: TMS-SVWP (n = 315), NoTMS-SVWP (n = 325), TMS-LVWP (n = 228) and NoTMS-LVWP (n = 237). Cows in TMS received 3 SQ injections of 5 mL of TMS (Multimin 90®) as follows: 2 injections prepartum at 229 ± 3 and 259 ± 3 d of gestation for MP and 243 ± 3 and 263 ± 3 d of gestation for PP. A third injection was given at 20 ± 3 d before first TAI. The effect of VWP duration, TMS, parity, and their interaction on pregnancy per AI (P/AI) at 39 d after AI, pregnancy loss (PL), and cows pregnant by 90 DIM (PG90) were evalu-

ated by logistical regression. At 39 d after AI, P/AI was greater for cows in the LVWP (47.5%) than SVWP (39.1%) group. At 39 d after AI, TMS did not affect ($P = 0.96$) P/AI (42.7% and 42.5% for TMS and NoTMS). Primiparous cows had greater ($P < 0.01$) P/AI than multiparous cows (49.0% and 38.4%). Pregnancy losses were similar ($P = 0.68$) for cows in LVWP (5.8%) and SVWP (4.7%) and for cows in TMS (5.7%) and NoTMS (4.9%). Also, PL were similar ($P = 0.13$) for PP (3.1%) and MP (7.1%) cows. No differences ($P = 0.59$) in PG90 were observed for cows in LWVP (46.2%) and SVWP (48.0%) and for cows in the TMS (48.3%) and NoTMS (46.0%) group ($P = 0.49$). Conversely PG90 was greater ($P < 0.01$) for PP (52.5%) than MP (43.7%) cows. We conclude that extending the duration of the VWP from 60 to 88 DIM improved P/AI of lactating dairy cows. Conversely, TMS supplementation during the prepartum period and 20 d before TAI did not improve fertility of lactating dairy cows.

Key Words: voluntary waiting period, trace mineral, dairy cow

M212 Resumption of ovarian cycle postpartum in dairy cows is affected by metabolic load in herbage-based feeding systems. Josef J. Gross*[1], Chiho Kawashima[2], Frigga Dohme-Meier[3], Akio Miyamoto[2], and Rupert M. Bruckmaier[1], *[1]Veterinary Physiology, Vetsuisse Faculty, Bern, Switzerland, [2]Obihiro University of Agriculture & Veterinary Medicine, Obihiro, Japan, [3]Agroscope Institute for Livestock Sciences ILS, Posieux, Switzerland.*

Due to topographic and climatic conditions, milk production in Switzerland is mainly based on herbage feeding with only little input of concentrates. This study investigated the effects of a solely herbage-based diet on production, metabolic, and endocrine parameters of dairy cows, and determined the factors affecting the resumption of ovarian cycle postpartum (pp). Twenty-three multiparous Holstein dairy cows (were divided into 2 groups according to their previous lactation yield (4679–10808 kg): a control (C, n = 13) and a treatment group (nC, n = 10) from wk 3 antepartum until wk 8 pp. While C received fresh cut herbage plus additional concentrate according to their estimated energy and nutrient requirements, no concentrate was fed to nC throughout the experiment. Milk yield and DMI were recorded daily. Blood samples were taken weekly and analyzed for IGF-1, glucose, NEFA, and BHBA. Milk progesterone (P4) concentrations were measured every 3 d in morning milk samples (skim milk) by RIA, and considered as resumption of ovarian cycle if the P4 level reached >1 ng/mL. Data were analyzed using mixed models. Plasma NEFA and BHBA concentrations pp were higher in nC (0.82, 1.18 mmol/L) compared with C (0.55, 0.63 mmol/L, $P < 0.05$). Days to resumption of ovarian cycle was similar between 2 groups (C = 29.8 d, nC = 33.3 d), thus the cows were divided into further 2 sub-groups with earlier/later than the above mean value, and categorized as earlier ovulation (E-OV; C = 22.6 d, n = 7; nC = 21.4 d, n = 6) and delayed ovulation (D-OV; C = 38.3 d, n = 6; nC = 45.2 d, n = 4). In C group, only BCS pp revealed lower in D-OV (2.81) compared with E-OV (3.21, $P < 0.05$). In nC group, however, D-OV showed lower BW (571 vs. 697 kg in E-OV) and glucose (3.5 vs. 3.6 mmol/L in E-OV + nC), higher BHBA (1.39 vs. 1.04 mmol/L in E-OV + nC) and NEFA pp (0.89 vs. 0.76 mmol/L in E-OV + nC, $P < 0.05$), suggesting an energy deficiency/stress on lipid metabolism. Other metabolic and welfare-related parameters were similar between E-OV vs. D-OV. In conclusion, in herbage-based feeding system without supplementary concentrate, dairy cows experience a higher metabolic load, which is very likely to affect resumption of ovarian cycle.

Key Words: metabolic load, ovulation, herbage feeding

M213 Blood β-hydroxybutyrate (BHBA) concentrations during the first two weeks after calving affect pregnancy establishment in postpartum dairy cows. Monica O. Caldeira*[1], Matthew C. Lucy[1], Ricardo O. Rodrigues[1], and Scott E. Poock[2], *[1]Division of Animal Sciences, University of Missouri, Columbia, MO, [2]College of Veterinary Medicine, University of Missouri, Columbia, MO.*

Negative energy balance (NEB) occurs in early postpartum dairy cows as a normal process when the energy demand at the onset of lactation exceeds feed energy intake. A poor response to NEB leads to greater blood concentrations of nonesterified fatty acids (NEFA) and BHBA, and also lesser blood glucose concentration. The objective was to determine whether blood BHBA and glucose concentration early postpartum affected pregnancy establishment after 3 inseminations in dairy cows. Blood samples were collected from lactating Holstein dairy cows (n = 204) twice weekly for the first 4 wk postpartum (n = 8 samples per cow). Blood BHBA and glucose were measured cowside by using a hand-held meter (Precision Xtra; Abbott Diabettes Care, Alameda, CA). Body weight (BW) and body condition score (BCS) were measured at calving, 2 wk, and 4 wk. Data were analyzed using the GLIMMIX, GLM, and MIXED procedures of SAS. The mean (SD) d postpartum of sample collection was 2.1 (1.3), 5.6 (1.5), 9.2 (1.4), 12.7 (1.6), 16.3 (1.6), 19.7 (1.6), 23.4 (1.6), and 26.8 (1.7) for samples 1 to 8. There were 171 cows with pregnancy data and 127/171 (74%) were pregnant after 3 inseminations. The blood BHBA concentration for sample 1 to 3 affected whether the cow became pregnant postpartum (lesser BHBA favorable toward pregnancy). The BHBA for non-pregnant and pregnant (respectively) was 0.91 ± 0.07 and 0.73 ± 0.04 (sample 1; $P < 0.029$), 0.92 ± 0.07 and 0.76 ± 0.04 (sample 2; $P < 0.043$) and 0.76 ± 0.04 and 0.68 ± 0.02 (sample 3; $P < 0.08$). The BHBA concentration for samples 4 to 8 did not affect pregnancy. Greater blood glucose concentration for sample 2 tended to be associated with pregnancy (51.3 ± 2.2 and 55.3 ± 1.3 for nonpregnant and pregnant; $P < 0.11$). Glucose in sample 1 and 3 to 8 were not associated with pregnancy later postpartum. BW loss and BCS loss postpartum did not affect pregnancy. In conclusion, BHBA concentrations within the first 10 d postpartum affected pregnancy outcome later postpartum.

Key Words: β-hydroxybutyrate, glucose, reproduction

M214 Effects of prepartum diets supplemented with oilseeds on reproductive performance in dairy cows. R. Salehi*[1], M. G. Colazo[2], M. Oba[1], and D. J. Ambrose[1,2], *[1]University of Alberta, Edmonton, Alberta, Canada, [2]Alberta Agriculture and Rural Development, Edmonton, Alberta, Canada.*

We investigated the effects of dietary fat supplementation (oilseed vs. no-oilseed) and type of oilseed (sunflower vs. canola) during late gestation on postpartum (pp) reproductive performance. Pregnant Holsteins were, blocked by BCS and parity, assigned to 1 of 3 diets containing 8% DM rolled sunflower (SUN; n = 45) or canola seed (CAN; n = 43), or no oilseed (CON; n = 43), for the last 35 d of gestation and then a common lactation diet pp. Ovaries were scanned 2x/wk until 35 DIM in 95 cows to record first appearance of 10 (DF) and 16 mm (PreOVF) follicles, and ovulation. Blood samples were collected strategically to evaluate NEFA, BHBA and glucose, and endometrial cytology performed at 25 ± 1 d pp (n = 12/diet). Data were analyzed using the Mixed procedure of SAS. Multiparous CON cows consumed more DM (19.5 ± 0.2) than those fed oilseed (18.0 ± 0.2 kg) during pre- and pp, until wk+3 pp. Cows fed SUN consumed more than those fed CAN (23.5 ± 0.2 vs 22.5 ± 0.3 kg) pp from wk+2 to wk+5. Cows fed oilseed had higher NEFA (mEq/mL) than CON at wk-3 (108.0 ± 10.3 vs. 70.8 ± 10.3), wk+1 (388.7 ± 56.2

vs. 271.0 ± 56.2), and wk+4 (242.0 ± 36.6 vs. 163.5 ± 36.6), but pre and pp energy balance (EB) did not differ (8.1 ± 1.6 Mcal). Prepartum diets did not affect BHBA (12.1 ± 1.6 mg/dL) and glucose (54.5 ± 2.9 mg/dL) pre or pp. Prepartum oilseed and type of oilseed supplemented neither altered the intervals (d) from calving to DF (9.4 ± 0.5), PreOVF (14.7 ± 0.8), or ovulation (21.4 ± 1.5), nor conception rate to first AI (27.2%) and cumulative pregnancy rate (67.3%) up to 5 AI. The interval to DF was correlated with % of polymorphonuclear cells (PMN; R: 0.38), DMI (R: −0.28) and glucose (R: −0.32) on wk+2. Interval to PreOVF was determined by % of PMN (R: 0.38) and DMI on wk −4 (R:-0.27), −3 (R:-0.27), −2 (R:-0.33),-1 (R:-0.38), +1 (R:-0.38), +2 (R:-0.45) and +3 (R:-0.39). None of the metabolites, glucose, EB, DMI or PMN affected interval to ovulation, although the latter was associated with interval to PreOVF (R: 0.41). In summary, feeding oilseeds prepartum, particularly canola, reduced DMI and increased NEFA without affecting reproductive performance.

Key Words: endometrial cytology, oilseed, ovarian function

M215 Distribution of fatty acids in reproductive tissues of cows fed flax-, canola-, or sunflower seed-based rations. Patricia A. Dutra[1,2], Mohanathas Gobikrushanth*[2], Reza Salehi[2], Marcos G. Colazo[3], and Divakar J. Ambrose[2,3], *[1]Departamento de Zootecnia, Universidade Federal da Bahia, Salvador, Bahia, Brazil, [2]Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada, [3]Livestock Research Branch, Alberta Agriculture and Rural Development, Edmonton, Alberta, Canada.*

Essential fatty acids (FA) like linoleic (C18:2) and α-linolenic (C18:3) acids play key roles in reproductive function. The objective of this study was to determine the distribution of fatty acids in the reproductive tissues of cows fed 3 different lipid diets. Nonlactating Holstein cows were randomly assigned to receive one of the 3 diets supplemented with rolled oilseeds: canola (CAN = 5), sunflower (SUN = 5), or flax (FLX = 5) at 8%DM. After cows had been on the experimental diets for at least 60 d, they were superovulated, inseminated and slaughtered 14 d after insemination. While embryos were used in another study, tissue samples of the uterus, oviduct and corpus luteum (CL) were collected and kept frozen until analyzed for FA. Fats from the reproductive tissues were extracted and methylated using indirect methylation method, and FA profiles in each tissue type were determined by gas chromatography. Primarily, the effects of diet, type of tissue, and their interactions on FA of our interest [oleic (C18:1), C18:2, C18:3, eicosapentaenoic (C20:5) and docosahexaenoic (C22:6)] were evaluated using Mixed procedure of SAS. As the interactions between diet and type of tissue were not significant, FA concentration was finally modeled against effects of diet and type of tissue. Diet did not affect FA concentrations. Except for oleic acid ($P = 0.07$), all of the other aforementioned FA differed ($P < 0.05$) among tissue types. The concentrations (mg/50mg tissue) of C18:2, C18:3 and C20:5, respectively, were higher in CL (0.282, 0.020 and 0.019) than in uterus (0.075, 0.004 and 0.008) and oviduct (0.037, 0.003 and 0.001). However, the concentration of C22:6 was higher in uterus than oviduct or CL (0.008 vs. 0.005 or 0.004). In addition, the amount of total FA, saturated FA, polyunsaturated FA, n-3 and n-6 FA were significantly higher in CL than uterus or oviduct. Results indicate that CL accumulates most of the FA of our interest than the uterus or oviduct, whereas C22:6 was more concentrated in the uterus. The biological reasons for this differential accumulation of FA among reproductive tissues need further investigation.

Key Words: fatty acid, reproductive tissue, diet

M216 Fatty acid profile in follicular fluid and serum of dairy cows fed diets supplemented with rolled canola, sunflower or flax seed. Patricia A. Dutra[1,2], Mohanathas Gobikrushanth*[2], Reza Salehi[2], Marcos G. Colazo[3], and Divakar J. Ambrose[3,2], *[1]Departamento de Zootecnia, Universidade Federal da Bahia, Salvador, Bahia, Brazil, [2]Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada, [3]Livestock Research Branch, Alberta Agriculture and Rural Development, Edmonton, Alberta, Canada.*

Polyunsaturated fatty acids such as n-3 and n-6 are known to influence reproductive performance of dairy cows. The objective of this study was to determine fatty acid (FA) profile in serum and follicular fluid of cows fed 3 different lipid diets as well as to determine the association between serum and follicular fluid in terms of FA concentrations. Nine nonlactating Holstein cows were randomly assigned to receive 1 of 3 diets supplemented with rolled oil seeds: canola (CAN = 3), sunflower (SUN = 3) or flax (FLX = 3) at 8%DM. After cows received their respective diets for at least 60 d, they were superovulated, inseminated and slaughtered 14 d after insemination. The experimental diets continued until 12 h before slaughter. Embryos and reproductive tissues collected were used in another study. Blood samples were collected on the day of insemination, and follicular fluid was collected from large ovarian follicles within 4 h of slaughter. Serum and follicular fluid were kept frozen until analyzed for FA concentrations. Fats from the follicular fluid and serum were extracted, methylated and FA profiles were determined by gas chromatography. Data were analyzed using the MIXED and CORR procedures of SAS. In follicular fluid, cows fed SUN had higher concentrations (mg/mL) of linoleic acid (0.60 vs. 0.30, 0.32) and total n-6 FAs (0.62 vs. 0.31, 0.33) and higher n-6/n-3 ratio (9.25 vs. 2.09, 3.84) than those fed either FLX or CAN. Similarly, cows fed FLX had higher concentrations of α-linolenic acid (C18:3; 0.14 vs. 0.08, 0.06), total n-3 FAs (0.16 vs. 0.09, 0.07) and higher n-3/n-6 ratio (0.49 vs. 0.10, 0.11) in follicular fluid than those fed either CAN or SUN. None of the FA other than γ-linolenic acid, in FLX cows, was altered by diet in serum. No significant correlations were observed between follicular fluid and serum FA profiles. Although diets supplemented with oilseeds selectively altered FA concentrations in follicular fluid, none of the major FA of relevance to reproductive function was altered in serum.

Key Words: fatty acid, follicular fluid, serum

M217 Effects of rumen-protected methionine and choline supplementation on gene expression of follicular cells of the first postpartum dominant follicle. Diego A. Velasco Acosta*[1,2], Ines M. Rivelli[2], Cassandra Skenandore[2], Daniel Luchini[3], Marcio Corrêa[1], and Felipe Cardoso[2], *[1]Universidade Federal de Pelotas, Pelotas, Rio Grande do Sul, Brazil, [2]University of Illinois, Urbana, IL, [3]Adisseo NA, Alpharetta, GA.*

This study aimed to determine the effects of rumen-protected methionine and choline supplementation during the transition period on mRNA expression of follicular cells of the 1st postpartum dominant follicle in Holstein cows. Multiparous cows were assigned in a randomized complete block design into 4 treatments from 21 d before calving to 30 DIM. Treatments were: CON (n = 10, fed the close-up and fresh cow diets with a Lys:Met = 3.5:1), MET (n = 9, fed the basal diet + methionine, Smartamine M to a Lys:Met = 2.9:1), CHO (n = 9, fed the basal diets + choline 60 g/d, Reashure), and MIX (n = 12, fed the basal diets plus Smartamine M to a Lys:Met = 2.9:1 and 60 g/d Reashure). Follicular development was monitored via ultrasound every 2 d starting at 7 DIM until the first dominant follicle reached a diameter of 16 mm. Follicular fluid from each cow was aspirated and cells were retrieved immediately

by centrifugation and stored at −80°C until RNA extraction. Statistical analysis was performed using the MIXED procedure of SAS. Gene expression of *LHCGR, STAR, 3β-HSD, P450scc, P450c17, CYP19A1, IRS1, IGF, MAT1A, SAHH, TLR4, TNF, IL1-β, IL8* and *IL6* was measured by real-time PCR. Treatments did not affect mRNA expression of *LHCGR*, *STAR*, *P450scc*, *CYP19A*, *SAHH*, *MAT1A* and *IL6* ($P > 0.05$) however, *3β-HSD* expression was higher ($P < 0.05$) for MET (1.46 ± 0.3) and MIX (1.25 ± 0.3) than CON (0.17 ± 0.04) and CHO (0.26 ± 0.1). For *TNF, TLR4* and *IL1-B* mRNA expression was higher ($P < 0.05$) for CON (11.70 ± 4.6, 21.29 ± 10.4, 6.28 ± 1.4) than CHO (2.77 ± 0.9, 2.16 ± 0.9, 2.29 ± 0.7) and MIX (2.23 ± 0.7, 1.46 ± 0.6, 2.92 ± 0.8). There was higher ($P < 0.05$) *IL1*-β expression and a tendency ($P = 0.07$) for higher TNF expression in CON (6.27 ± 1.4, 11.70 ± 4.6) than MET (3.28 ± 0.6, 3.06 ± 0.8). There was no difference ($P = 0.43$) between CON and MET for *TLR4*. Expression of IL8 mRNA was lower ($P < 0.05$) for CHO (0.98 ± 0.3) than CON (4.90 ± 0.7), MET (6.10 ± 1.7) and MIX (5.05 ± 1.8). In conclusion, supplementing Smartamine M and Reashure during the transition period changed mRNA expression in follicular cells of the 1st postpartum dominant follicle in Holstein cows.

Key Words: methionine, choline, gene expression

M218 Effect of yeast culture plus enzymatically hydrolyzed yeast supplementation starting prepartum on acute phase protein profiles and reproductive performance in dairy cows. Vanessa Oliveira Freitas[1], Claudia Faccio Demarco[1], Tatiele Mumbach[1], Eduardo Gularte Xavier[2], Raquel Fraga Silva Raimondo[3], Fernanda Medeiros Gonçalves[1], Francisco Augusto Burkert Del Pino[1], Viviane Rohrig Rabassa[1], Sangita Jalukar[4], Marcio Nunes Corrêa[1], and Cassio Cassal Brauner*[1], *[1]Universidade Federal de Pelotas, NUPEEC, Pelotas, RS, Brazil, [2]Granjas 4 Irmãos S/A, Rio Grande, RS, Brazil, [3]Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil, [4]Arm & Hammer Animal Nutrition, Princeton, NJ.*

The aim of this study was to evaluate the effect of yeast culture plus enzymatically hydrolyzed yeast supplementation starting prepartum on acute phase protein profile and reproductive performance in dairy cows. Twenty 9 multiparous Holstein cows were blocked by milk production on the previous lactation and randomly assigned into 2 groups which consisted in one (n = 15) receiving 28 g/d top-dressed yeast culture plus enzymatically hydrolyzed yeast (YC-EHY; Celmanax, Arm & Hammer Animal Nutrition, Princeton, NJ), while the control group (n = 14) did not receive the supplement. The experimental period lasted from d −35 relative to calving to 150 d postpartum. Plasma samples collected on −21, −14, −7, 0, 3, 7, 14, 21, 28, 35, and 42 d relative to calving were analyzed for acute phase proteins (haptoglobin, paraoxonase and albumin). To assess estrous cyclicity resumption, blood samples were collected weekly from 14 to 35 d after calving to evaluate the concentration of progesterone. Reproductive performance was also monitored until 150 d after calving. Data were analyzed using mixed models with repeated measures over time. Interval from calving to conception was analyzed using Kaplan-Meier survival curves. The YC-EHY group had lower ($P = 0.04$) interval from calving to conception than control group respectively, 95.35 ± 10.78 vs.130.73 ± 10.35 d. The interval from calving to the first ovulation were similar ($P = 0.18$) between groups, 23,33 ± 2,8 d vs. 29,16 ± 2,8 d, respectively for YC-EHY and control groups. The YC-EHY tended ($P = 0.07$) to have lower activity of paraoxonase during the postpartum period (112.54 ± 3.49 U/L vs. 106.39 ± 3.60 U/L) than the control group. No differences ($P > 0.05$) were observed in albumin and haptoglobin concentrations during the transition period. In conclusion, dairy cows supplemented since prepartum with yeast culture plus enzymatically hydrolyzed yeast, had lower interval from calving to conception, without affect calving to first ovulation interval and tend to have lower activity of paraoxonase during the postpartum period.

Key Words: nutrition, reproduction, supplement

M219 Nutritional level of the recipient ewe, but not of the donor, determines the embryo fate when good quality embryos are transferred. Victoria de Brun*[1], Ana Meikle[1], Fernando Forcada[2], Inmaculada Palacín[2], Cecilia Sosa[3], and José Alfonso Abecia[2], *[1]Laboratorio de Técnicas Nucleares, Montevideo, Uruguay, [2]Departamento de Producción Animal y Ciencia de los Alimentos, Zaragoza, Spain, [3]Departamento de Anatomía Patológica, Medicina Legal y Forense y Toxicología, Zaragoza, Spain.*

We aimed to isolate effects of undernutrition on the embryo or the mother and whether or not the endocrine signals differ from ewes that presented late embryo mortality. Forty-five donors and 52 recipient Rasa Aragonesa ewes were fed 1.5 (control group; donor n = 20; recipient n = 25) or 0.5 (low group; donor n = 25; recipient n = 27) times the daily requirements for maintenance. These regimens were maintained up to day of embryo collection and transfer. Embryos were collected 7 d after the onset of estrus, and 2 good quality embryos per ewe were transferred into recipient ewes. Blood samples were collected at days −14, −1, 7 and 18. Pregnancy and late embryonic mortality were analyzed using the GENMOD procedure, and hormone and metabolite concentrations using a MIXED model in SAS. Nutritional treatment of donor did not affect fertility. Pregnancy evaluated by ultrasound on d 40 did not differ between control and undernourished ewes (72% vs. 55%, $P = 0.17$). Embryonic mortality between d 18 and 40 tended to be greater in undernourished than in control recipient ewes (35% vs. 14%, $P = 0.11$). Undernourished ewes had reduced live weight and body condition score ($P < 0.01$), and increased concentration of nonesterified fatty acid ($P < 0.05$). Pregnancy rates at d 18 and 40 were similar between groups, but recipient undernourished ewes presented a greater late embryonic mortality (d 18 to 40) than control ewes ($P = 0.11$). Pregnant ewes had greater ovulation rate than nonpregnant ewes ($P = 0.02$). In recipient ewes, pregnant undernourished presented greater P4 concentrations than pregnant control ($P < 0.05$) and nonpregnant underfed ewes ($P < 0.05$). Recipient undernourished ewes that suffered late embryo mortality (d 18 to 40) presented or tended to present lower insulin and progesterone concentrations than low pregnant ewes ($P = 0.05$ and $P = 0.07$, respectively). In conclusion, the failure in reproductive performances in underfed ewes transferred good quality embryos is caused principally by maternal factors, so a suitable maternal environment is crucial to ensure a normal embryo growth. In addition, the endocrine profiles such as progesterone during early luteal phase and insulin are associated with pregnancy outcome.

Key Words: sheep, undernutrition, embryo

M220 Evaluation of the hypothalamic kisspeptin system during the attainment of puberty in gilts. Eric S. Jolitz*[1], Waljit S. Dhillo[2], and Jeffrey A. Clapper[1], *[1]South Dakota State University, Brookings, SD, [2]Imperial College, London, UK.*

It has been demonstrated that circulating kisspeptin is increased during the attainment of puberty in humans. To determine if hypothalamic (MBH) expression of kisspeptin (Kiss1), anterior pituitary (AP) expression of GnRH receptor (GnRHr) and LH-β, and MBH and AP kisspeptin content increase at pubertal onset in the gilt, the following experiment was performed. Twenty-four crossbred gilts of similar age (150 d) and weight (102.7 ± 0.3 kg) were relocated and exposed to a mature

boar for estrus detection on d 1 and continuing for 6 d. Gilts that stood immobile within 24 h of slaughter (d 6) were considered to have attained puberty. Plasma samples were collected on d 1, 3, and 6. All gilts were slaughtered on d 6 when MBH, AP, and blood were collected. Relative expression of MBH Kiss1 and β-actin and AP GnRHr, LH-β, and β-actin was determined using real-time reverse transcriptase PCR. Fold changes in relative expression were determined using the Relative Expression software tool. Relative expression is based on the expression ratio of a target gene versus a reference gene. The expression ratio results of the transcripts were tested for significance by a pair-wise fixed reallocation randomized test with day compared as independent time effects. Hypothalamic and AP content of kisspeptin were determined by RIA and differences were determined using the MIXED procedure of SAS. Relative expression of Kiss1 was increased ($P = 0.005$) 2.2 fold in the gilts that had attained puberty. Relative expression of GnRHr did not differ ($P > 0.05$) in gilts that had attained puberty versus those that did not. Relative expression of LH-β tended to be decreased ($P = 0.09$) 0.80 fold in gilts that had attained puberty. AP concentrations of kisspeptin were not different ($P > 0.05$) between treatments. Kisspeptin content in the MBH was increased ($P = 0.03$) in gilts that attained puberty (0.72 ± 0.14 ng/μg protein) compared with gilts that did not (0.44 ± 0.05 ng/μg protein). These data provide preliminary evidence that hypothalamic expression of Kiss1 and content of kisspeptin are increased during the attainment of puberty in the pig, which may modulate the release of GnRH.

Key Words: kisspeptin, hypothalamus, pig

M221 Ovine maternal nutrient restriction from mid to late gestation induces steroid metabolizing enzyme activity in maternal and fetal reproductive and liver tissues. Megan P. T. Coleson*[1], Christa L. Gilfeather[1], Kimberly A. Vonnahme[2], and Caleb O. Lemley[1], *[1]Department of Animal and Dairy Sciences, Mississippi State University, Mississippi State, MS, [2]Department of Animal Sciences, North Dakota State University, Fargo, ND.*

The objective was to determine the effects of nutrient restriction on steroid metabolizing enzymes within maternal, placental, and fetal tissues. Singleton pregnant ewe lambs (n = 30) were allocated to receive either 100% [adequate (ADQ; n = 14)] or 60% [restricted (RES; n = 16)] of nutrient requirements from d 50 until d 130 of gestation. At slaughter both maternal and fetal livers and maternal (caruncle; CAR) and fetal (cotyledon; COT) placentas were collected for analysis. Activity of cytochrome P450 2C (CYP2C), cytochrome P450 1A (CYP1A), and cytochrome P450 3A (CYP3A), were determined using specific luminogenic substrates. Activities were expressed relative to mg of protein, total tissue weight, or BW. Data were analyzed using MIXED procedure of SAS and the model statement included nutritional plane. Activity of CYP2C was not detected in CAR or COT tissues. Activity of CYP1A and CYP3A were not different in CAR tissue between the 2 treatments. Activity of CYP1A relative to BW was increased ($P = 0.03$) in COT tissue of RES ewes compared with ADQ fed; however, CYP3A activity in COT was not different between treatments. Maternal liver activity of CYP2C was decreased ($P < 0.01$) in RES ewes compared with ADQ; however, activity of CYP1A (relative to mg of protein), and CYP3A (relative to mg of protein or maternal BW) were increased ($P \leq 0.05$) in RES ewes compared with ADQ. Fetal liver activity of CYP1A (relative to liver weight or fetal BW), CYP2C (relative to liver weight), and CYP3A (relative to mg of protein, liver weight, or fetal BW) were decreased ($P < 0.05$) in RES ewes compared with ADQ. In conclusion, maternal nutrient restriction increased activity of CYP1A and CYP3A in maternal liver, but decreased the activity of CYP1A and CYP3A in fetal liver. A similar downregulation of CYP2C activity was observed in both maternal and fetal liver. The differential regulation of hepatic cytochrome P450 in maternal and fetal tissues may influence peripheral steroid concentrations during late pregnancy and deserves future investigation.

Key Words: cytochrome P450, nutrient restriction, steroid

M222 Effects of maternal nutrient restriction on bovine placentome and miRNA expression during mid-gestation. Regina K. Taylor*, Kayla S. Mangrum, Christopher T. LeMaster, Scott L. Pratt, and Nathan M. Long, *Clemson University, Clemson, South Carolina.*

Primiparous Angus-cross cows (n = 22) were synchronized and AIed with sexed semen from a single Angus sire. Animals were fed at 1.3× (Control [CON]) or 0.55× (Nutrient Restricted [NR]) of maintenance energy and protein requirements based on BW (NRC 1996). Animals were blocked by BCS and BW and assigned to 1 of 3 treatments: CON; (n = 8) d 30–190; NR/CON; (n = 7) NR d 30–110 then CON d 110–190; or CON/NR; (n = 7) CON d 30–110 then NR d 110–190. Cows were killed on d 190 of gestation, and the fetus and placenta collected. Isolation of RNA was performed from flash frozen cotyledon samples (3 subsamples/treatment) using the *mir*Vana microRNA Isolation kit and analyzed using a previously validated microarray. Placentome measures and microRNA expression was analyzed as an ANOVA analysis using appropriate procedures correcting for false discovery rate of microarray data. Total placentome weight and total caruncular weight tended to be decreased ($P = 0.056$, $P = 0.07$, respectively) in NR/CON animals vs. CON/NR and CON/CON. Total cotyledonary weight was increased ($P = 0.017$) in CON/NR animals vs. NR/CON and CON/CON. Ratio of cotyledon weight:caruncle weight was increased ($P = 0.02$) in NR/CON and CON/NR vs. CON/CON animals. Total placentome surface area tended to be increased ($P = 0.09$) in CON/NR animals vs. NR/CON and CON/NR. Cotyledons from CON had mdo-miR-195 upregulated ($P < 0.01$) and 8 downregulated ($P < 0.01$) miRNAs, including mmu-mir-5105-p3 and bta-miR-2484 vs. NR/CON and CON/NR cotyledons. Cotyledons from NR/CON had mmu-mir-5117-p3_1ss19TA and bta-miR-99b upregulated ($P < 0.01$) and mmu-mir-5105-p3_1ss24TC and bta-miR29a downregulated ($P < 0.01$) vs. CON/NR and CON cotyledons. Cotyledons from CON/NR had 11 upregulated ($P < 0.01$) miRNAs including bta-miR29a and bta-miR2484, and mdo-miR-195 and bta-miR-99b downregulated ($P < 0.01$) vs. CON and NR/CON cotyledons. The data show that maternal nutrient restriction during early or mid gestation causes asymmetrical fetal growth restriction and affects miRNA regulation differently depending on if the restriction is preceded or followed by a non-restriction period.

Key Words: fetal growth, undernutrition, fetal programming

M223 Maternal overnutrition/obesity (MO) in the ewe has multigenerational metabolic programming effects on adult granddaughters (F_2). Megan A. Walton*, John F. Odhiambo, Peter W. Nathanielsz, and Stephen P. Ford, *Department of Animal Science, University of Wyoming, Laramie, WY.*

We have reported on a model in which MO ewes are fed 150% of NRC recommendations from 60 d before conception to term or fed only to requirements (CON). Offspring (F1) of MO ewes exhibit hyperphagia, increased weight gain and adiposity, and insulin resistance in response to an ad libitum feeding challenge when compared with CONF1 (J. Anim. Sci. 2010. 88:3546). We recently reported that male grandsons (MOF2) also experience a significant increase in body mass and insulin resistance

in response to a similar bout of ad libitum feeding as 2-year-old adults, demonstrating a multigenerational effect of MO grandmothers on their grandsons. To evaluate potential offspring sex differences, we studied the multigenerational impact of MO on the response of 2-year-old adult female F2 to a 12-week ad libitum feeding challenge, MOF2 (n = 6) or CONF2 (n = 6). A dual x-ray absorptiometry (DEXA) scan was performed before and after the feeding challenge to determine changes in % body fat and % lean. Body weights and plasma samples were obtained bi-weekly. Glucose was evaluated via a colorimetric assay and insulin was analyzed via a validated RIA. Data (M ± SEM) were analyzed using the mixed procedure in SAS. No differences in body weight, % body fat or % lean were observed for CONF2 and MOF2 females either before or after the feeding trial. By the end of the feeding trial, body weight and % body fat were markedly increased ($P < 0.05$) in both groups. Plasma glucose was greater ($P < 0.05$) in MOF2 than CONF2 females, while plasma insulin tended to be greater ($P < 0.06$) in MOF2 than CONF2 females throughout the trial, resulting in a markedly elevated ($P < 0.01$) HOMA-IR in MOF2 vs. CONF2 females (7.8 ± 0.7 vs. 5.0 ± 0.7, respectively) demonstrating severe IR in MOF2. These data demonstrate that as previously reported for male OBF2, an ad libitum feeding challenge during adulthood induced marked insulin resistance in female OBF2. In contrast to the male OBF2, however, this elevated insulin resistance developed in the total absence of any increase in weight gain over that of female CONF2 in response to ad libitum feeding.

Key Words: programming

M224 Effect of prepartum dam supplementation, creep-feeding and post-weaning diet on age at puberty in Nellore heifers. Marcos V. C. Ferraz Junior*[1], Delci D. Nepomuceno[2,1], Marcos V. Biehl[2], Alexandre V. Pires[2,1], Marcelo H. Santos[1], Renan G. Silva[1], Vinicius N. Gouvea[1], Jose R. S. Gonçalves[3], Thiago S. Andrade[1], and Michael L. Day[4], *[1]University of São Paulo, Pirassununga, São Paulo, Brazil, [2]University of São Paulo, Piracicaba, São Paulo, Brazil, [3]Experimental Station Hildegard Georgina Von Pritzelwitz, Londrina, Paraná, Brazil, [4]The Ohio State University, Columbus, OH.*

The objective of this study was to evaluate the effect of various nutritional strategies on age at puberty in Nellore heifers. Nellore cows (n = 237 / BW 432 ± 50 kg / BCS 2.85 ± 0.4; 1 to 5 scale) were used in randomized design with a 2 × 2 x 2 factorial arrangement of treatments. Factors were as follows: factor 1 = prepartum supplementation with protein (PPS; n = 122; 0.5 kg soybean meal/cow daily) during the last trimester of gestation or no supplementation (NPPS; n = 115); factor 2 = provision of creep-feed to calves for 95 d before weaning (CF; n = 117) or no creep-feeding (NCF; n = 120; 22% CP and 72% TDN); and factor 3 = fed post-weaning from 7 to 14 mo of age in a feedlot (PWF; n = 117; 15% CP and 56% TDN) or no feedlot (NPWF; n = 120; *Brachiaria* pasture). After 14 mo of age, heifers were maintained in *Brachiaria* ssp. pastures. Heifers were weighed and reproductive status assessed by ovarian US (detection of presence of CL) monthly from 12 to 30 mo old. The proportion of heifers that had attained puberty by 14, 18, 26 and 30 mo of age, and BW at puberty were assessed using PROC MIXED and GLIMMIX. At weaning (220.1 ± 2.0), cow supplementation did not affect BW of heifer calves (168.2 ± 0.4 vs. 170.5 ± 0.5 kg for PPS and NPPS cows, respectively), but BW at weaning were greater ($P = 0.01$) in the CF (172.7 ± 1.4) than NCF (165.8 ± 1.4 kg) treatment. At the end of the post weaning period (Factor 3) BW were greater ($P < 0.01$) in the PWF (234.1 ± 2.2) than NPWF (187.2 ± 2.1) treatment and this difference was maintained through 18 mo of age. Proportions of heifers that attained puberty at 14 (7.1%), 18 (22.3%), 26 (70.0%) and 30 (93.2%) mo of age were not influenced by cow supplementation or creep feed. However, the post-weaning feedlot treatment (PWF) increased the proportion of heifers that reached puberty at 14 (13.7 vs. 0.9%; $P < 0.01$) and 18 (31.7 vs. 13.3%; $P < 0.01$) mo of age and tended ($P = 0.10$) to anticipate puberty at 26 mo of age (73.8 vs. 66.1%; PWS and NPWS, respectively). In conclusion, enhanced nutrition during the post weaning period increased the proportion of heifers that reached puberty between 14 and 26 mo of age and is an effective method to anticipate puberty.

Key Words: puberty, Nellore, supplemental management

M225 Effect of prepartum dam supplementation and creep-feeding on age at puberty in Nellore heifers. Elizangela M. Moreira[1], Alexandre V. Pires[2,1], Delci D. Nepomuceno[2], Marcos V. C. Ferraz Junior*[1], Jose A. Faleiro Neto[1], Marcos V. Biehl[2], Jose R. S. Goncalves[3], Renan G. Silva[1], Marcelo H. Santos[1], Vinicius N. Gouvea[1], and Michael L. Day[4], *[1]University of São Paulo, Pirassununga, São Paulo, Brazil, [2]University of São Paulo, Piracicaba, São Paulo, Brazil, [3]Experimental Station Hildegard Georgina Von Pritzelwitz, Londrina, Paraná, Brazil, [4]The Ohio State University, Columbus, OH.*

The objective of this study was to evaluate the effect of 2 nutritional strategies on age at puberty in Nellore heifers. Nellore cows were allocated to treatments by BW (438 ± 48 kg) and BCS (3.0 ± 0.5; 1 to 5 scale). The factorial experimental design (2 × 2) included 2 phases of supplementation: phase 1 – prepartum supplementation with protein (PPS; n = 73; 0.5 kg soybean meal/cow daily) during the last trimester of gestation or no supplementation (NPPS; n = 81); phase 2 – provision of creep-feed (creep diet was 22% CP, and 62% TDN) to calves for 118 d before weaning (CF; n = 78) or no creep-feeding (NCF; n = 75). After 7 mo of age (weaning), heifers were maintained in *Brachiaria* ssp. pastures. Heifers were weighed and reproductive status assessed by ovarian ultrasonography (detection of presence of CL) monthly from 14 to 30 mo old. The proportion of heifers that had attained puberty by 18, 26 and 30 mo of age, and BW were assessed using PROC MIXED and PROC GLIMMIX (differences with $P < 0.05$ considered significant). At weaning (6.4 mo), cow supplementation in last trimester of gestation did not affect BW of heifer calves (183 ± 1.9 vs. 184 ± 1.9 kg, $P = 0.98$ for PPS and NPPS cows, respectively). Creep feeding supplementation also did not influence BW at weaning (184 ± 1.9 vs. 183 ± 1.9 kg, $P = 0.78$ for CF and NCF, respectively). Prepartum supplementation did not change the proportion of heifers pubertal at 18 mo (4.1 vs. 6.2%, $P = 0.53$), 26 mo (18.5 vs. 16.4%, $P = 0.57$) and 30 mo (82.2 vs. 76.2%, $P = 0.36$ for PPS and NPPS, respectively) of age. Supplementation of suckled calves by Creep Feeding also did not change the proportion of heifers pubertal at 18 mo (3.8 vs. 6.7%, $P = 0.43$), 26 mo (15.8 vs. 19.1%, $P = 0.77$) and 30 mo (80.7 vs. 77.3%, $P = 0.59$ to CF and NCF, respectively) of age. In conclusion, neither the cow supplementation nor creep feeding used in the present study anticipated puberty in Nellore heifers.

Key Words: puberty, Nellore, supplemental management

M226 Effect of energy supplementation on plasmatic concentration of leptin in pre-pubertal beef heifers. Maria Carolina V. Miguel*[1], Siobhan W. Walsh[2], Heni F. Costa[1], Juliana S. Souza[1], Rafael S. Cipriano[1], David Giraldo-Arana[1], Marcos Antônio Maioli[1], Devani M. Pinheiro[1], Alexander C. Evans[2], and Guilherme P. Nogueira[1], *[1]São Paulo State University, Araçatuba, SP, Brazil, [2]University College Dublin, Dublin, Ireland.*

The objective was to evaluate nutritional influence on plasma leptin concentration in prepubertal Nellore heifers. Sixteen calves at birth

were assigned to 2 groups:1) calves received a concentrate of ground corn (2% of BW; Treated; n = 8); and, 2) calves received mineral salt without supplementation (Control; n = 8), calves from both groups had access to the cow's diet. After weaning (5 mo of age), both groups received sugar cane bagasse, citrus pulp and concentrate, but the Treated group also received a 2% of BW ground corn supplement until first ovulation or 22 mo of age. Heifers were weighed weekly to calculate monthly weight gain and blood samples were collected twice a week. Leptin was quantified with a RIA kit; the intra-assay CV were 7.64% for high (6.84 ng/mL) and 2.09% for low controls (0.58 ng/mL); the inter-assay CV were 21% for high and 12% for low controls, and assay sensitivity was 0,229 ng/mL. Data were analyzed by ANOVA with the Mixed procedure of SAS for repeated measures. The weight gain from weaning to puberty was 19.5 ± 0.03 kg/month for Treated and 13.2 ± 0.03 kg/month for Control heifers. The first ovulation occurred when the Treated heifers (n = 5) weighed 398.2 ± 10.6 kg (17.8 ± 1.1 mo) and the Control heifers (n = 1) 361kg (20.1 mo). After the 7th month of age there was a difference ($P < 0.0001$) in monthly weight between treatments. In both treatments, plasma leptin increased according to age ($P < 0.0001$). Plasma leptin concentration was greater ($P = 0.023$) in Treated compared with Control heifers at 13, 14, 16 and 17 mo of age, averaging, respectively, 5.67 ± 0.53 vs. 4.04 ± 0.53 ng/mL, 5.79 ± 0.53 vs. 4.34 ± 0.53 ng/mL, 7.12 ± 0.53 vs 5.21 ± 0.53 ng/mL and 6.74 ± 0.67 vs. 5.11 ± 0.53 ng/mL. The greatest weight gain in Treated heifers was observed close to first ovulation. The greatest plasma leptin concentration in Treated heifers was at the 16th month of age. Thus, supplementing heifers with corn hastened age at first ovulation, which is likely mediated by increased weight gain and leptin concentrations (r = 0.96; $P < 0.0001$), permissive factors that influence GnRH/LH secretion.

Key Words: *Bos indicus*, heifer, feed supplementation

M227 Form of selenium in free-choice mineral mixes affects ovarian production of progesterone but not estradiol in cycling beef cows. Phillip J. Bridges*[1], Katheryn L. Cerny[1], Michelle Rhoads[2], Leslie H. Anderson[1], Walter R. Burris[1], and James C. Matthews[1], *[1]University of Kentucky, Lexington, KY, [2]Virginia Polytechnic Institute and State University, Blacksburg, VA.*

Selenium (Se) affects gonadal function, and the form of Se provided to cows affects tissue-specific gene expression. The objective of this study was therefore to determine whether the form of Se consumed by cows would affect follicular growth and the production of ovarian steroids. Pasture-fed Angus crossbred cows were randomly assigned to have ad libitum access to free-choice vitamin-mineral mixes containing Se (35 ppm) in either (TRT) inorganic (ISe, n = 41), organic (OSe, n = 42), or a 50/50 mix of ISe and OSe (MIX, n = 44) forms for 175 d. A subset of these cows (n = 9–11 per TRT) with a detectable corpus luteum were maintained on TRT and administered 25 mg $PGF_{2\alpha}$ to induce luteal regression, then assigned for further sampling on estrus (Day 0). Between Day 4 and 8, follicular diameter was determined by ultrasonography. On Day 6, cows were treated with 20 mg then 15 mg $PGF_{2\alpha}$, 8 to 12 h apart, to induce luteal regression and differentiation of the first-wave dominant follicle into a preovulatory follicle. On Day 8, 36 h after $PGF_{2\alpha}$, the contents of the preovulatory follicle were aspirated by ultrasound-guided follicular puncture. Progesterone and estradiol were determined in plasma collected on Day 6 and 8, and in follicular fluid collected on Day 8. TRT effects were assessed by ANOVA and Fisher's LSD. TRT affected ($P < 0.02$, OSe > MIX) total blood Se (OSe: 156 ± 5, ISe: 146 ± 4, MIX: 140 ± 6 µg/mL). TRT affected ($P < 0.04$, MIX > ISe) systemic progesterone on Day 6 (MIX: 5.1 ± 0.6, OSe: 4.6 ± 0.5, ISe: 3.4 ± 0.2 ng/mL) but not Day 8. TRT did not affect systemic estradiol on Day 6 or 8. TRT tended ($P = 0.07$, OSe > MIX, ISe) to affect follicular fluid progesterone (OSe: 58.9 ± 11.5, MIX: 44.4 ± 3.8, ISe: 30.5 ± 8.1 ng/mL) but not estradiol. TRT did not affect diameter of the dominant follicle on Days 4 to 6, and tended ($P = 0.08$, ISe > OSe, MIX) to affect the diameter of the preovulatory follicle on Day 8 (ISe: 12.6 ± 0.6, OSe: 11.4 ± 0.3, MIX: 11.3 ± 0.4 mm). Our results indicate that form of Se fed to cows affects progesterone but not estradiol synthesis, and tends to affect follicular diameter, in a manner independent of total blood Se concentrations.

Key Words: selenium, follicle, corpus luteum

M228 Relationship between plasma amino acid profile and ovarian function around the time of ovulation in beef cows. Taylor C. Geppert*[1], Allison M. Meyer[2], George A. Perry[3], and Patrick J. Gunn[1], *[1]Department of Animal Science, Iowa State University, Ames, IA, [2]Division of Animal Sciences, University of Missouri, Columbia, MO, [3]Department of Animal Sciences, South Dakota State University, Brookings, SD*

The objective of this experiment was to determine the relationship between plasma AA concentrations and reproductive parameters of beef cows (n = 26). Two studies evaluating the effects of excess MP on reproductive function were compiled for this analysis. Non-pregnant, nonlactating mature beef cows consuming ad libitum corn stalks were offered a once-daily supplement designed to meet NRC NE_m and exceed MP requirements by 25–50%. After a 20 d adaptation period, cows were synchronized for ovulation using the 5-d CO-Synch + CIDR protocol. Ten days after synchronization, 100 µL of GnRH was administered to reset ovarian follicular growth. Daily transrectal ultrasonography was performed to diagram ovarian activity, and blood samples were taken for hormone and AA analyses. Corpus luteum (CL) volume was determined via ultrasound, and supplementation ended 7 d after estrus. Blood samples collected between d 47 and d 49 underwent AA analysis. Data were analyzed using the CORR procedure of SAS. No significant correlations were observed between AA profile and ovulatory follicle size or antral follicle count ($P > 0.13$). However, a positive relationship between length of proestrus and total AA (r = 0.49, $P < 0.01$) and total essential AA (r = 0.48, $P < 0.01$) was observed. In addition, a positive relationship between length of proestrus and total glycogenic AA, branched-chain AA, urea cycle AA, arginine, threonine, valine and isoleucine ($r \geq 0.44$, $P \leq 0.04$) were observed. As a percent of essential AA, leucine and phenylalanine were negatively correlated ($r \leq -0.39$, $P < 0.05$) with length of proestrus. However, AA profile was not correlated with estradiol at estrus ($P > 0.05$). As a percent of total AA and essential AA, arginine was positively correlated with circulating progesterone 7 d post-estrus and ratio of progesterone to CL vol ($r \geq 0.41$, $P \leq 0.04$). Based on these data, total and essential AA concentrations may be related to several reproductive parameters around the time of ovulation; however, further research is needed to establish a causal relationship between individual AA and reproductive functions at ovulation.

Key Words: amino acid, correlation, ovulation

M229 Association between circulating blood or plasma urea nitrogen concentrations and reproductive efficiency in beef heifers and cows. Patrick J. Gunn*[1], Allie L. Lundberg[1], Robert A. Cushman[2], Harvey C. Freetly[2], Olivia L. Amundson[3], Julie A. Walker[3], and George A. Perry[3], *[1]Department of Animal Science, Iowa State University, Ames, IA, [2]USDA, ARS, US Meat Animal Research Center, Clay Center, NE, [3]Department of Animal Sciences, South Dakota State University, Brookings, SD.*

The objective was to examine the effect of circulating blood or plasma urea nitrogen concentrations (BPUNC) on reproductive efficiency in beef heifers and suckled beef cows. Data from nulliparous heifers (n = 284) as well as primiparous (n = 241) and multiparous (n = 806) beef cows were compiled across 15 experiments. A single blood sample collected from each female during estrous or ovulation synchronization was analyzed for BPUNC. Only females that were maintained on the same nutritional management scheme (pasture or coproduct-based drylot ration) for at least 1 wk before synchronization though the first 21 d of the breeding season were included in the analysis. To determine if BPUNC affected first service pregnancy rate, cattle were categorized as having BPUNC above or below each integer from 10 to 25 mg/dL. The GLIMMIX procedure of SAS was used for data analysis. The model for each BPUNC classification analysis also included the fixed effects of age classification and nutritional management scheme. Interactions among fixed effects were not significant and removed ($P \geq 0.10$). Days postpartum at synchronization was included in the model as a covariate when applicable and experiment was included as a random effect. Average first-service pregnancy rate across the data set was 55.6%. Irrespective of age and nutritional management scheme, a BPUNC that was associated with decreased pregnancy rates could not be established. In fact, there was a tendency for improved pregnancy rate as BPUNC increased ($P = 0.08$, r = 0.05) and cattle with BPUNC above 16 mg/dL tended to have greater pregnancy rates (57.0%) than those below 16 mg/dL (54.6%; $P = 0.07$). There were no differences ($P \geq 0.16$) in pregnancy rates between cattle that had BPUNC above or below any other integer from 10 to 25 mg/dL. Based on these data, when cattle are allowed to adapt to a nutritional management scheme before breeding and maintained on that diet through the first 21 d of the breeding season, BPUNC is not negatively associated with first-service pregnancy rates. USDA is an equal opportunity provider and employer.

Key Words: crude protein, cyclicity, pregnancy