Physiology and Endocrinology: Feed Intake, Metabolism and Maternal Nutrition

1121 Expression of neuropeptide Y and its receptors as affected by nutrition and leptin infusion in Zebu heifers. J. Diniz-Magalhães, M. V. Carvalho, A. B. S. Machado, R. A. Ribeiro, and L. F. P. Silva*, *Universidade de São Paulo*, *Pirassununga, SP, Brazil*.

Leptin has been proposed to act via NPY in the hypothalamus to modulate the effect of nutrition on sexual maturation. The objective was to evaluate the effect of oLeptin infusion and of high or low energy intake on the expression of neuropeptide Y (NPY), and 2 NPY receptors, NPY-Y1 and NPY-Y2. Thirty 6 prepubertal Nelore heifers, 18 to 20 mo-old, 275.8 ± 17.2kg BW and BCS of 5 ± 0.5 (1 to 9 scale) were randomly assigned to each of 3 treatments (n = 12): H (high energy diet), L (low energy diet), and LL (low energy diet + oLeptin). Diets were formulated to promote weight gain of 0.4 kg/day (groups L and LL) or 1.2 kg/day (H group). Heifers were fed ad libitum once a day, they were weighed and had their BCS evaluated twice weekly. After 21 d of adjustment, heifers in LL group received subcutaneous injections of oLeptin at 4.8µg/kg BW twice a day, for 56 d. Groups H and L received similar injections of 2mL saline solution. Age at puberty was considered to be the age on first detection of a corpus luteum by twice weekly transrectal ultrasonography, confirmed by plasma concentrations of progesterone of >1ng/ml. Twenty 4 heifers were slaughtered at the time of puberty for harvesting of the hypothalamus. Total RNA was extracted with Trizol, treated with DNaseI and reverse transcribed to cDNA. Expression of transcripts of NPY, NPY-Y1 and NPY-Y4 was quantified by real-time PCR. Changes in gene expression were calculated by relative quantification with the ΔΔCt method, using the gene ribosomal protein L-19 as the reference gene. There was no effect of leptin administration on expression of NPY ($P = 0.70$), NPY-Y1 ($P = 0.72$) and NPY-Y4 ($P = 0.96$) at the time of puberty. However, high energy intake reduced expression of NPY-Y1 ($P = 0.02$), without affecting expression of the other 2 genes. Downregulation of NPY-Y1 could indicate lower sensibility of the hypothalamus to NPY action. NPY is a well known inhibitor of puberty therefore this result indicates that a reduction in NPY-Y1 expression could permit attainment of puberty at a younger age in Zebu heifers.

Key Words: *Bos indicus*, hypothalamus, puberty

1122 Blocking µ-opioid receptors alters short-term feed intake and oro-sensorial preferences of weaned calves. C. Montoro*[1], I. Ipharraguerre[2], and A. Bach[1,3], [1]*Ruminant Production, IRTA*, *Caldes de Montbui, Barcelona, Spain*, [2]*LUCTA S.A.*, *Barcelona, Spain*, [3]*ICREA*, *Barcelona, Spain*.

In rats and humans, opioids play a role in controlling short-term regulation of feed intake through oro-sensorial reward mechanisms. Thirty-four Holstein calves (BW = 86.5 ± 1.73kg, age = 77 ± 0.6 d) were housed in individual pens and submitted to 4 treatments to determine whether naloxone, a µ-opioid receptor antagonist, affects feed intake and oro-sensorial preferences in weaned calves. The study was conducted in 2 periods with 17 animals per period. Treatments followed a 2 × 2 factorial arrangement combining a fasted (FA) or fed (FE) state with an i.v. injection of saline (0.9% NaCl) solution (S) or naloxone (1 mg/kg of BW; N). Each factor combination involved 8 animals, except FA-S and FE-N which had 9 calves. All calves were offered a double choice of the same feed unflavored or flavored with a sweetener (Luctarom SFS-R, Lucta) *ad libitum*. Feed consumption was recorded every 60 min during 6 h (from 0800 to 1400) following the morning feed offer. Data were log-transformed and analyzed using a mixed-effects model with repeated measures. Calves in the FE-N group tended ($P = 0.10$) to consume less concentrate (3.97 ± 0.398 log g/h) than FE-S (4.86 ± 0.422 log g/h), FA-N (5.30 ± 0.422 log g/h), and FA-S (5.07 ± 0.398 log g/h). Furthermore, S calves showed a preference for the sweetened feed ($P < 0.05$) whereas N calves did not show any preference (Table 1). These data suggest that µ-opioid receptors are involved in short-term feed intake regulation by mediating oro-sensorial feed preferences and may interact with energy-mediated mechanisms in regulating the amount of feed consumed.

Table 1. Feed consumption rate (log of g/h) as affected by treatments

Treatment	Unflavored concentrate	Sweetened concentrate	P-value
FA-S	4.78±0.417	5.36±0.42	0.02
FA-N	5.31±0.442	5.29±0.44	0.93
FE-S	4.36±0.442	5.36±0.44	<0.001
FE-N	4.14±0.417	3.79±0.42	0.15

Key Words: taste, intake, regulation

1123 Evidence that nesfatin-1 is a satiety factor in the pig and that the hypothalamus controls its expression in adipose tissue. C. A. Lents*[1], C. R. Barb[2], G. J. Hausman[2], L. Lee-Rutherford[2], C. J. Rogers[1], N. L. Heidorn[1], R. S. Cisse[1], and M. J. Azain[1], [1]*University of Georgia*, *Athens,* [2]*USDA-ARS Richard B. Russell Agriculture Research Center*, *Athens, GA*.

Two experiments (Exp) were conducted to test if nesfatin-1 is part of the adipose tissue-hypothalamic loop regulating appetite and energy balance of the pig. In Exp 1, prepuberal gilts were adapted to a twice-daily feeding schedule (0800 and 1600 h) and received intracerebroventricular (i.c.v.) injection of 100 µg of either recombinant human leptin or nesfatin-1 (nearly equal mass to leptin) in 0.9% saline. Control animals received 0.9% saline alone (n = 4/group). Four hours after i.c.v. injection, feeders were placed in all pens (1600 h) for determination of cumulative intake at 4, 20, 44, and 68 h after feed presentation. Food consumption of nesfatin-1 treated pigs was suppressed ($P < 0.01$) during the first 20 h compared with saline controls (0.54 ± 0.3 and 3.15 ± 0.3 kg, respectively), but was not different from leptin-treated pigs (1.04 ± 0.3 kg). Although food intake of nesfatin-1 and leptin-treated pigs was increasing ($P < 0.05$) after 20 h, it was still less ($P < 0.001$) at 68 h than that observed for saline-treated pigs (4.04, 4.50 and 8.76 ± 0.31 kg for nesfatin-1, leptin and saline, respectively). Subcutaneous (SC) adipose in the pig is innervated by hypothalamic neurons that are sensitive to secreted adipokines. Nesfatin-1 is proposed to work through the melanocortin 3/4 receptor (MC3/4R) pathway, which when activated alters gene expression in fat. In Exp II, gilts received i.c.v. injection of 10 µg of the MC3/4R agonist NDP-MSH or 0.9% saline alone (n = 9/group). Pigs were sacrificed 24 h later and SC adipose tissue was collected for isolation of RNA. Abundance of mRNA for nesfatin was quantified with real-time RT-PCR. Relative differences in expression were calculated by the REST procedure with 18S rRNA as the reference control. mRNA for nesfatin in SC adipose tissue of NDP-MSH treated pigs was reduced 1.7 fold ($P = 0.05$) compared with saline-treated pigs. We conclude that nesfatin-1 is a satiety factor in the pig and that activation of the MC3/4R pathway suppresses expression of nesfatin-1, which may be mediated by sympathetic neuronal outflow to adipose tissue.

Key Words: nesfatin-1, feed intake, swine

1124 Endocannabinoid and PPARα signaling gene network expression in liver of peripartal cows fed two levels of dietary energy prepartum. M. J. Khan*, D. E. Graugnard, and J. J. Loor, *University of Illinois*, *Urbana.*

Fatty acid ethanolamides (FAE) have anorexic and anti-inflammatory properties and are endogenous ligands for cannabinoid receptors subtype 1 (CNR1) or 2 (CNR2). Some FAE mediate peripheral metabolic effects in non-ruminants through the activation of PPARα. Because of the potential for endogenous FAE to mediate inflammation and satiety at the level of the liver, we sought to establish the longitudinal mRNA expression of *CNR1*, *CNR2*, *PPARA*, *FAAH*, *ASAH*, several PPARα targets (*CPT1A*, *ANGPTL4*, *FGF21*), and *RXRA* in cows (n = 14/diet) assigned to a control (high-straw, S; NEL = 1.34 Mcal/kg) or moderate-energy (ME; NEL = 1.62 Mcal/kg) diet during the dry period and until parturition. A percutaneous liver biopsy was collected at −14, 7, 14, and 30 d relative to parturition for transcript profiling via quantitative PCR. Normalized/log-transformed data were analyzed using ANOVA. Estimated prepartal energy balance (EBAL) in cows fed ME was greater ($P < 0.05$) and averaged 159% of requirements compared with 102% in cows fed S. However, EBAL during the first week postpartum tended ($P = 0.10$) to be lower in cows fed ME (83% vs. 89% of requirements) which had greater ($P = 0.06$) serum NEFA (560 vs. 351 mEq/L). *CNR2* expression remained high (diet × time $P < 0.05$) with ME by d 7 after which it decreased to levels similar to cows fed S. Following a decrease from −14 to 7 d, *PPARA* and RXRA increased ($P < 0.05$) gradually through 14 d regardless of diet. However, cows fed ME had overall greater ($P < 0.05$) *RXRA*. The PPARα targets *ANGPTL4* (3-fold) and FGF21 (20-fold) increased dramatically between −14 and 7 d in cows fed ME (diet × time $P < 0.05$). Thereafter, expression of both genes declined and reached lowest levels at 30 d regardless of prepartal diet. Whereas *CPT1A* decreased between −14 and 7 d and then reached peak expression by 30 d with S, expression increased gradually between −14 and 14 d with ME (diet × time $P < 0.05$). Preliminary results revealed a potential role of negative EBAL and high NEFA induced by prepartal energy overfeeding on endocannabinoid signaling and PPARα activation.

Key Words: metabolism, inflammation, transition cow

1125 Endoplasmic reticulum (ER) stress gene network expression in liver of peripartal cows fed two levels of dietary energy prepartum. M. J. Khan*, D. E. Graugnard, and J. J. Loor, *University of Illinois*, *Urbana*.

X-box binding protein 1 (XBP1) is a key regulator of the mammalian unfolded protein response or ER stress response. We examined expression of genes associated with the ER stress response in cows assigned (14/diet) to a control (high-straw, S; NEL = 1.34 Mcal/kg) or moderate-energy (ME; NEL = 1.62 Mcal/kg) diet prepartum and until parturition. A percutaneous liver biopsy was collected at −14, 7, 14, and 30 d relative to parturition for transcript profiling via quantitative PCR. Normalized/log-transformed data were analyzed by ANOVA. Estimated prepartal energy balance (EBAL) in cows fed ME was greater ($P < 0.05$) and averaged 159% of requirements compared with 102% in cows fed S. However, EBAL during the first week postpartum tended ($P = 0.10$) to be lower in cows fed Me (83% vs. 89% of requirements) which had greater ($P = 0.06$) serum NEFA (560 vs. 351 mEq/L). *XBP1* was 185% greater at −14 d in cows fed S and increased 24% by d 7 postpartum (diet × day $P < 0.05$) after which it decreased close to prepartal levels by 14 d but still was 93% greater in cows fed S vs. ME. There was a diet × time effect ($P < 0.05$) for the expression of *EIF2AK3*, a kinase induced by ER stress leading to a halt of translation, due to a decrease between −14 and 7 d in cows fed S which also had greater (25%) *EIF2AK3* at −14 d. *EIF2AK3* returned to prepartal levels by 14 through 30 d. The stress-induced transcription factor DDIT3 decreased ($P < 0.05$) between −14 and 7 d regardless of prepartal diet after which it remained below prepartal values. Expression of stearoyl-CoA desaturase (*SCD*), whose downregulation in rodent liver leads to robust ER stress, was lower at −14 d with S but increased by 4-fold at 7 d leading to a diet × time effect ($P < 0.05$). There was no change in *SCD* between −14 and 7 d in cows fed ME, but expression increased gradually to peak values at 30 d. Despite differences in EBAL and NEFA postpartum, preliminary results indicated moderate effects of prepartal energy level on ER stress at calving potentially driven the by marked upregulation of *SCD* postpartum.

Key Words: transition cow, metabolism, transcriptomics

1126 Effects of a hyperinsulemic euglycemic clamp administered during heat stress or pair feeding on plasma ghrelin concentrations of lactating dairy cattle. S. E. Cossel*, M. E. Field, M. V. Skrzypek, S. R. Sanders, L. H. Baumgard, R. P. Rhoads, and M. L. Rhoads, *University of Arizona*, *Tucson*.

The objective of this study was to measure the effects of a hyperinsulinemic euglycemic clamp (HEC) on plasma ghrelin (GHR) concentrations during heat stress (HS) or pair feeding (PF). Lactating Holstein dairy cows (n = 11; 136 ± 8 DIM; 560 ± 32 kg BW) were housed in environmentally controlled chambers and assigned to 1 of 2 treatment groups. During period 1 (9 d), both groups were maintained in thermal neutral (TN) conditions (18°C; 20% RH) and fed ad libitum. During period 2 (9 d), cows were exposed to: 1) HS (n = 6; cyclical temperatures; 31.1–38.9°C; 20% RH) with ad libitum feed or 2) TN conditions while being PF to match the intake of the HS cows (n = 5). On d 8 of each period, 7 control (CON) blood samples were collected at 30-min intervals for each group. On d 9 of each period, a HEC was conducted and 7 blood samples were collected at 30-min intervals beginning 240 min after the administration of an insulin bolus. Rectal temperatures (RT) and respiration rates (RR) increased during the HS period compared with the TN period (39.7 ± 0.1 vs. 38.3 ± 0.1°C; $P < 0.01$ and 90 ± 2 vs. 38 ± 2; $P < 0.01$, respectively), while DMI decreased (15.1 ± 0.4 vs. 19.75 ± 0.4 kg; $P < 0.01$). The RT and RR of PF animals did not differ from TN conditions, while DMI decreased as designed. Plasma GHR concentrations were not affected by HS or PF alone (CON-HS or CON-PF vs. CON-TN concentrations). Likewise, mean plasma GHR concentrations in CON samples were similar to those from the HEC during the TN and HS treatments. In contrast to the HS group, PF cows had lower mean plasma GHR concentrations during the HEC compared with CON (187.2 ± 53.9 vs. 336.6 ± 53.9 pg/mL, respectively; $P < 0.01$). During the HEC of both the HS and PF treatments, plasma GHR concentrations increased from the beginning to the end of the sampling period ($P < 0.03$ and $P < 0.01$, respectively). Thus, regardless of similar feed intake, profiles of plasma GHR concentrations differed between PF and HS cows.

Key Words: ghrelin, dairy, glucose

1127 Effects of heat stress on insulin action in lactating Holstein cows. M. V. Skrzypek*[1], R. P. Rhoads[1], S. R. Sanders[1], K. Flann[1], L. Cole[1], J. W. Perfield[2], M. R. Waldron[2], and L. H. Baumgard[3], [1]*University of Arizona*, *Tucson,* [2]*University of Missouri*, *Columbia,* [3]*Iowa State University*, *Ames*.

Multiparous cows (n = 12; parity = 2; 136 ± 8 DIM, 560 ± 32 kg BW) housed in climate chambers were fed a TMR consisting primarily of alfalfa hay and steam-flaked corn and subjected to 2 experimental periods

(P): 1) thermoneutral (TN) conditions (18°C, 20% humidity) with ad libitum intake for 9d and 2) heat-stress (HS) conditions (cyclical temperature 31.1–38.9°C, 20% humidity: min THI = 73, max THI = 80.5) and ad libitum intake (n = 6) or pair-fed (PF in TN conditions, n = 6) for 9d. Rectal temperature (Tr) and respiration rate (RR) were measured thrice daily at 0430, 1200 and 1630h. To evaluate insulin sensitivity, an 8-h hyperinsulinemic-euglycemic clamp (HEC) was performed on d9 of P1 and P2. The HEC was a primed, continuous infusion of bovine insulin (3.8 μg/kg BW/h followed by 2 μg/kg BW/h) and euglycemia maintained by varying intrajugular exogenous glucose infusion rates. During P2, HS cows had ($P < 0.01$) a 1.48°C increase in Tr and a 2.4-fold increase in RR compared with PF cows. HS reduced ($P < 0.01$) DMI by 8 kg/d and by design PF cows had similar intake reductions. Milk yield was decreased similarly (30%) in HS and PF cows and both groups entered into a similar (−4.5 Mcal/d) calculated negative energy balance during P2. Compared with P1 ($P < 0.05$), basal glucose levels increased (5%) in PF cows but decreased (5%) in HS cows. Basal NEFA levels increased (34%, $P < 0.01$) in PF cows compared with P1, but did not change from P1 to P2 in HS cows. The HEC increased ($P < 0.01$) plasma insulin levels similarly for both treatments during both periods (0.6 vs. 5.9 ng/ml). The overall rate of glucose infusion (ROI) to maintain euglycemia during the HEC did not differ between treatments, but was markedly reduced (29%; $P < 0.01$) during P2. Compared with P1, the ratio of ROI to basal glucose concentration decreased (21%; $P < 0.01$) in PF cows, but did not differ in HS cows during P2. Because a comparable ROI was needed to maintain a smaller blood glucose pool in HS cows, the HEC data imply HS cows have greater glucose disposal as a result of enhanced insulin action.

Key Words: heat stress, insulin

1128 The effect of insulin glargine on the metabolism of lactating Holstein cows. L. A. Winkelman*, D. M. Barbano, M. E. Van Amburgh, and T. R. Overton, *Cornell University*, *Ithaca, NY*.

Two studies were conducted to determine the effects of an insulin analog, insulin glargine (SRI) (Lantus , Sanofi-Aventis), on metabolism of lactating Holstein cows. In study one, 16 multiparous cows (213 ± 10 DIM) were divided into 2 groups and randomly assigned to one of 4 treatments (control, 0.1 IU SRI/kg BW (L), 0.2 IU/kg BW (M), and 0.4 IU/kg BW (H)). Subcutaneous (SQ) injections of SRI or water were given at 0900 h. Cows were fed hourly and milked at 1500, 2300, and 0700 h. Blood samples were taken hourly via jugular catheter for 24 h after treatment injections. Administration of increasing doses of SRI resulted in a linear ($P = 0.004$) decrease in plasma glucose (66.0, 62.3, 61.0, 54.1 mg/dl glucose for control, L, M, and H doses, respectively). Endogenous insulin secretion decreased linearly ($P = 0.028$) with SRI dose (1.04, 0.88, 0.79, 0.64 ng/ml for control, L, M, and H doses, respectively). In study 2, 3 multiparous cows (101 ± 22 DIM) fitted with indwelling intercostal arterial and mammary vein catheters were used to determine effects of SRI in a 2-period crossover design. Periods lasted 4 d with a 2 d washout in between periods. In period one, 2 cows received 0.15 IU/kg BW of SRI, 2x/d at 12-h intervals, while the remaining cow was a control. Treatments were reversed in period 2. On d 4 of each period, simultaneous blood samples were taken from the arterial and venous catheters hourly for 12 h. Dry matter intake, milk yield, and all major milk components, except lactose, were not different ($P > 0.10$). Lactose content ($P = 0.094$) and yield ($P = 0.091$) were higher for the control treatment. Casein content and yield did not differ between treatments ($P > 0.10$), but whey content ($P = 0.012$) and yield ($P = 0.015$) were higher for SRI. Arterial and venous glucose were lower ($P < 0.10$) for SRI (arterial: 63.1, 56.9 mg/dl for control vs. SRI, respectively; venous: 45.4, and 40.7 mg/dl for control vs. SRI, respectively). Based on these studies, insulin glargine is able to lower plasma glucose across different doses and its effect on milk protein composition warrants further research.

Key Words: insulin, glucose

1129 The effects of maternal obesity and overnutrition on ovine fetal adipose tissue lipid composition. N. M. Long*[1,2], D. C. Rule[2], P. W. Nathanielsz[3], and S. P. Ford[1,2], [1]*Center for the Study of Fetal Programming*, *University of Wyoming, Laramie,* [2]*Department of Animal Science*, *University of Wyoming, Laramie,* [3]*Department of Obstetrics and Gynecology*, *University of Texas Health Sciences Center, San Antonio*.

The effects of maternal obesity and overnutrition on fetal adiposity and adipose tissue composition were evaluated. Multiparous ewes were allotted by BW and age and fed either 100% of NRC recommendations (Control; C) or 150% of NRC (Obese; OB) from d 60 before conception until necropsy on d 135 of gestation. Fetal body weights and fetal adipose depot weights were recorded. Fatty acid (FA) composition of perirenal, pericardial, and subcutaneous (SC) adipose tissue of 7 male twin fetuses per group was determined by GLC. Data were analyzed using the GLM procedure of SAS. Fetal BW tended to be reduced ($P = 0.08$), and eviscerated weight (EFW) was lower ($P = 0.05$) for fetuses from OB than C ewes (4.78 ± 0.23 vs. 5.40 ± 0.25 kg and 3.56 ± 0.18 vs. 4.12 ± 0.18 kg, respectively). Pericardial and perirenal adipose tissue weight as a % of EFW was greater ($P < 0.05$) in fetuses from OB than C ewes (0.195 ± 0.001 vs. 0.191 ± 0.001%; 0.81 ± 0.01 vs. 0.68 ± 0.01%, respectively). 12th rib fat thickness was greater ($P < 0.01$) in fetuses from OB than C ewes (0.9 ± 0.1 vs. 0.5 ± 0.1 mm). Total FA concentrations were greater ($P < 0.01$) in the perirenal adipose tissue of OB fetuses than C fetuses (913.4 ± 29.8 vs. 731.6 ± 29.8 mg/g tissue). Concentrations of 18:0, 18:1 c-9 and 18:1 c10/c11 in perirenal adipose tissue were greater ($P < 0.05$) in fetuses from OB than C ewes. Only 18:1 c-9 was greater ($P < 0.05$) in pericardial adipose tissue of fetuses from OB than C ewes. Total FA in SC adipose tissue tended to be greater ($P = 0.09$) in fetuses from OB than C ewes (114.5 ± 24.9 vs. 50.0 ± 24.9 mg/g tissue). Concentrations of 16:0, 18:1 c-9, and 20:4 n-6 were greater ($P < 0.03$) in SC adipose tissue from fetuses of OB than C ewes. Maternal obesity resulted in greater fetal adiposity and altered FA composition of adipose tissue in late gestation which could lead to permanent alteration in adipose tissue function.

Key Words: maternal obesity, fatty acid composition, adipose tissue

1130 Influence of metabolizable protein supplementation during late gestation on vasoreactivity of maternal and fetal placental arteries in sheep. L. A. Lekatz*[1], M. L. Van Emon[2], P. K. Shukla[3], S. T. O'Rourke[3], C. S. Schauer[2], K. M. Carlin[1], and K. A. Vonnahme[1], [1]*Center for Nutrition and Pregnancy, Department of Animal Sciences, North Dakota State University*, *Fargo,* [2]*Hettinger Research Extension Center, North Dakota State University*, *Hettinger,* [3]*Department of Pharmaceutical Sciences, North Dakota State University*, *Fargo*.

To examine the effects of metabolizable protein (MP) intake during late gestation on the vasoreactivity of placental arteries, 18 pregnant ewes received 100% (CON), 75% (LOW), or 125% (HIGH) of MP requirement from d 100 to 130 of gestation. On d 130, several caruncular (CAR) and cotyledonary (COT) arteries from placentomes of similar size and in close proximity to the umbilicus were selected for vasoreactivity studies. Arterial rings were suspended in organ chambers filled with 25 mL of physiological salt solution aerated with a mixture

of 95% O_2 and 5% CO_2 and kept at 38.6°C. Optimal tension was found by progressively stretching the rings until the contractile response to KCl (20 mM) was maximal. The presence or absence of endothelium was verified by testing the ability of bradykinin (BK; 10^{-7} M) to produce endothelium-dependent relaxation during contraction evoked by norepinephrine (10^{-6} M). Contractile dose response curves (DRC) were obtained by contracting with increasing concentrations of KCl and phenylephrine (PE). Rings were contracted with U46619 (10^{-8} M) and the DRC to either BK or sodium nitroprusside (SNP) was obtained in endothelium intact and endothelium removed rings, respectively. There was no effect on the KCl DRC in CAR arteries ($P \geq 0.36$). KCl exhibited a treatment × dose interaction in COT arteries where arteries from CON and LOW ewes were more ($P = 0.01$) sensitive to KCl compared with arteries from HIGH ewes. There was no effect of PE in CAR or COT ($P \geq 0.12$) arteries. There was no effect on either BK or SNP in CAR ($P \geq 0.56$) arteries. The COT arteries from CON and LOW ewes tended ($P = 0.09$) to be more sensitive to BK compared with arteries from HIGH ewes; however, there was no effect of SNP in these arteries ($P = 0.87$). This indicates that BK-induced vasodilation in COT arteries cannot solely be explained by a nitric oxide donor. Perhaps the vasodilator action of BK is through downstream affects of an endothelial derived hyperpolarizing factor and/or prostacyclin. Further analyses are needed to determine how protein supplementation impacts vascular function in the placenta.

1131 Maternal nutrient restriction (NR) upregulates phosphoenolpyruvate carboxykinase (PEPCK) expression in the livers of aged female offspring. L. Zhang*[1], Y. Ma[1], N. Tuersunjiang[1], L. A. George[1], S. P. Ford[1], and P. W. Nathanielsz[2], [1]*Center for the Study of Fetal Programming*, *Univ. of Wyoming, Laramie,* [2]*Center for Pregnancy and Newborn Research*, *Univ. of Texas Health Sciences Center, San Antonio*.

Undernutrition often occurs in pregnant ruminants under range conditions. Maternal NR during pregnancy is linked to offspring insulin resistance and metabolic disease. In 2003, ewes carrying singleton fetuses were fed a control (C, 100% NRC recommendations) or NR (50% NRC) diet from 28 to 78d of gestation. After d78, all ewes were fed 100% NRC until lambing. C and NR lambs were managed together and assigned to this study in 2009, when female offspring of NR (n = 4) and C (n = 4) ewes were subjected to ad lib feeding for 12 wks. As previously reported (George et al., Ann. Mtg. Soc. Gynecol. Invest, 2010, abstract), feed efficiency, weight gain, glucose clearance and insulin resistance were greater ($P < 0.05$) in ewes from NR dams. After the feeding trial, ewes were necropsied, the liver quickly removed and tissue from the left lobe frozen in liquid nitrogen. Protein and mRNA expression of PEPCK and glucose-6-phosphatase (G6Pase) were quantified via Western blotting and Real-time PCR. Additional liver tissue was placed in a tissue cassette, fixed with paraformaldehyde and paraffin embedded for immunohistochemistry. Both mRNA and protein expression of PEPCK were elevated ($P < 0.05$) in NR vs C ewe offspring (4.6 ± 0.9 vs 1.9 ± 0.6 and 0.8 ± 0.1 vs 0.5 ± 0.1 arbitrary units, respectively). In contrast, no differences were observed between C and NR groups in G6Pase at the mRNA or protein levels (2.1 ± 0.7 vs 2.2 ± 0.3 and 0.6 ± 0.1 vs 0.6 ± 0.03 arbitrary units, respectively). PEPCK and G6Pase binding was localized to the cytoplasm of hepatic cells. Enhanced gluconeogenesis was associated with the greater glucose clearance index, weight gain and feed efficiency in these aged NR offspring during ad lib feeding. Further, lower insulin sensitivity in NR offspring may impair suppression of hepatic gluconeogenesis, fitting with the upregulation of PEPCK observed in this study.

Key Words: maternal undernutrition, aged offspring, liver glucoconeogenesis

1132 Maternal nutrient restriction (NR) from early to mid-gestation increases pancreatic β-cell number at mid-gestation but pancreatic weight and β-cell numbers are reduced by late-gestation. L Zhang*[1], L. A. George[1], S. P. Ford[1], and P. W. Nathanielsz[2], [1]*Center for the Study of Fetal Programming*, *Univ. of Wyoming, Laramie,* [2]*Center for Pregnancy and Newborn Research*, *Univ. of Texas Health Sciences Center, San Antonio*.

Both maternal obesity (MO) and NR are associated with increased rates of obesity and diabetes in offspring. We reported that MO in ewes results in increased fetal pancreatic β-cell numbers at mid-gestation, but reduced β-cell numbers by late gestation. Here we compare these data to a sheep model of early to midgestation maternal NR. Multiparous ewes were assigned to a control group (C, 100% NRC recommendations) or an NR group (50% NRC) from 28 to 78 d of gestation (dG). A subgroup of ewes was necropsied on 78 dG (C = 5, NR = 5), and the rest (C = 5, NR = 5) were fed 100% NRC from 78 to 135 dG and necropsied. A fetal blood sample was collected for insulin quantitation, and the fetal pancreas weighed and the splenic end paraffin embedded for determination of β-cell numbers per unit area. At 78 dG, NR fetal weight was lower ($P < 0.05$) than C fetuses (230.7 ± 10.3 vs. 312.0 ± 22.6 g). While no difference was observed in pancreatic weight on 78 dG, β-cell numbers per unit area islet tissue, were increased ($P < 0.05$) in NR versus C fetuses (134.9 ± 11.4 vs. 89.8 ± 9.9). No treatment difference in fetal weight was observed on d135 which averaged 4034.1 g, however, pancreatic weights of d135 NR fetuses were lower ($P < 0.05$) than those from C fetuses (2.8 ± 0.2 vs. 3.8 ± 0.3 g). The β-cell numbers were also markedly decreased ($P < 0.01$) in d135 fetuses from NR versus C ewes (113.3 ± 4.7 vs. 187.3 ± 19.3). Blood insulin levels from all d78 fetuses were below assay sensitivity, whereas on 135 dG concentrations were lower ($P = 0.07$) in fetuses from NR versus C ewes (1.8 ± 0.2 vs. 4.8 ± 1.4 mIU/L). These results suggest that maternal NR from early to midgestation, like MO, initial increased pancreatic growth and β-cell numbers, but lead to reductions in pancreatic growth and β-cell numbers by late gestation, possibly leading to pancreatic dysfunction in postnatal life.

Key Words: maternal nutrient restriction, fetal pancreatic development, sheep