

**132 Novel effects of nutrition on reproduction in lactating dairy cows.** M. C. Wiltbank\*, R. Sartori, S. Sangsritavong, H. Lopez, J. M. Haughian, P. M. Fricke, and A. Guen, *Department of Dairy Science, University of Wisconsin-Madison.*

A number of reproductive parameters are altered in lactating dairy cows including: twinning rate, expression of estrus, conception rate, pregnancy loss, and incidence of anovulation. We hypothesized that the high feed consumption associated with lactation has fundamentally altered reproductive physiology due to increased metabolism of steroids. We evaluated liver blood flow and steroid metabolism under different physiological conditions using continuous infusion of bromosulfothalein (90% metabolized by one pass through liver) and/or estradiol and progesterone. In lactating or non-lactating cows, acute feeding produced an increase in liver blood flow of 20-30% that was maximal by 2 h after feeding. There was a corresponding decrease of 30% in circulating estradiol and progesterone concentrations following acute feeding of cows continuously infused with steroids. The baseline liver blood flow was much greater in lactating (1600 l/h) as compared to non-lactating (800 l/h) cows. Similarly, estradiol and progesterone metabolism was much greater in lactating as compared to non-lactating cows. Analysis of normal estrous cycles showed striking differences in lactating cows vs. heifers that are consistent with high steroid metabolism. Peak serum estradiol near estrus was lower in single-ovulating lactating cows (7.15 pg/ml) than heifers (9.46) in spite of larger preovulatory follicle diameter in cows (17.45 mm) than heifers (14.82). Similarly, circulating progesterone concentrations were lower for cows than heifers from d 5 to 14 of the estrous cycle (on day 14: 4.13 vs. 6.14 ng/ml); whereas, luteal volume was greater for cows than heifers (on day 14: 8272932 vs. 5415276 mm<sup>3</sup>). There were also differences in double ovulation rate (cows-25.0%; heifers-1.8%) and embryo quality on day 5 after normal ovulation (n = 27-28 per group each flushed 3 different times). The percentage of degenerated embryos (Grade 5) was greater (p<0.05) in lactating cows (61.9%) vs. heifers (6.3%) during summer or comparing lactating (41.7%) vs. non-lactating cows (17.6%) during winter. Thus, changes in reproductive physiology may be a consequence of the high feed consumption, high liver blood flow and resulting increase in steroid metabolism associated with high milk production.

**Key Words:** Reproduction, Liver blood flow, Progesterone

**133 The influence of nutrient intake on ovarian form and function in meat-type chickens.** F. E. Robinson\*<sup>1</sup>, R. A. Renema<sup>1</sup>, and M. J. Zuidhof<sup>2</sup>, <sup>1</sup>University of Alberta, <sup>2</sup>Alberta Agriculture, Food and Rural Development.

The chicken is an ideal model in which to examine ovarian form and function, including follicular steroidogenesis, recruitment, yolk deposition, atresia and ovulation rate. Meat-type chickens that have been intensively selected for appetite exhibit very fast rates of growth. These stocks are reproductively unfit when allowed to feed ad libitum. Our research has focussed on the degree of feed restriction during rearing and lay, as well as the rate of change in feed intake during sexual maturation (SM) on the above mentioned ovarian parameters in breeding stock. During SM, dietary energy allocation to pullets is increased from approx. 285 kcal/day (20 wk) to 460 kcal/day (30 wk). Birds that are allocated this increase quickly, and early during this period have at least one extra large yellow follicle (LYF). This excess follicular development is associated with a reduction of about 10 eggs, as under estrogen domination following photostimulation (PS), hepatic vitellogenesis partitions excess dietary energy to follicular development. Considering the increased gonadotrophin levels seen in over-fed pullets, it is hypothesized that increased estrogen output was stimulated. Increased estrogen levels stimulate hepatic fatty acid synthesis and the formation of phospholipids and proteins to package lipid for yolk deposition. Recently, the consequences of over-feeding BB females for one of seven 14-d periods during the period from 18 to 32 wk of age were investigated. It was observed that the most critical period for exposing pullets to a bonus feeding allocation (30 g more feed/bird/d) was between 14 and 28 after PS. It is proposed that this period represents the time of maximal sensitivity of hepatic lipogenesis to circulating estrogen. Plasma levels of FSH and LH increase within 48 h of PS in all birds and levels are higher in heavy weight birds. These heavy birds reach sexual maturity more quickly and have a greater number of LYF than do small birds.

Increased plane of nutrition can double the initial increases in plasma levels of FSH and LH and further accelerate SM. Increased follicular development is not advantageous, as evidenced by significantly lower egg output in aggressively fed flocks.

**Key Words:** Chicken, Ovarian morphology, Follicular recruitment

**134 Relationships between Bovine follicular steroids and components of the extracellular matrix.** C.M. Field\*<sup>1</sup>, A.R. Williams<sup>1</sup>, A.B. Moore<sup>1</sup>, J.N. Oyarzo<sup>2</sup>, M.E. Bellin<sup>2</sup>, and R.L. Ax<sup>2</sup>, <sup>1</sup>Mississippi State University, Starkville, MS, <sup>2</sup>University of Arizona, Tucson, AZ.

Objectives were to confirm presence of TIMP-2 (T2) on surface membranes of bovine follicular cells, determine the relationship of estradiol-17 $\beta$  (E), progesterone (P), and glycosaminoglycans (GAGs) to T2 in follicular development, and quantify binding of T2 in follicular content. Ovaries were obtained within 30 minutes of slaughter or by ovariectomy. Follicles (n=441) were aspirated from ovaries and follicular fluid and follicular cells separated by centrifugation, and frozen. E and P concentrations in individual follicles were determined by RIA from follicular fluid. Follicles were classified into 4 groups by high/low E and P and large/small size (n=40). Atresia was defined by low E concentration. Follicular fluid GAGs were determined spectrophotometrically using an Alcian blue dye procedure. Granulosa cell content per follicle was quantified using a Pierce BCA protein assay and spectrophotometry. The quantity of T2 present was measured with a fluorometer after incubation with fluorescent-labeled human T2 antibody at 210  $\mu$ g/ml per sample. Data were analyzed using SAS GLM procedures to determine relationships of E and P to GAGs and T2 expression and correlation procedures to establish relationship of T2 to GAGs. An inverse relationship between T2 and GAGs to E and P was revealed (P<.05). Regardless of size, high E and P follicles had low GAGs and T2 binding, while low E and P follicles displayed elevated GAGs and T2 binding. The amount of T2/ $\mu$ g protein indicated a trend toward additional T2 binding on follicular cells from large atretic follicles, compared to healthy large follicles and small atretic follicles (P<.10). A strong relationship (r=0.85) between T2/ $\mu$ g protein and GAGs existed in large, high E follicles. Results confirmed follicle health was related to T2 and GAG expression, with atretic follicles, on the basis of steroid concentrations, displaying higher GAGs and T expression.

**Key Words:** Bovine, Follicle, TIMP-2

**135 Relationship between preovulatory follicle growth and postovulatory luteal function in the cow.** GE Mann\*<sup>1</sup>, ECL Bleach<sup>2</sup>, GR Starbuck<sup>1</sup>, and MD Fray<sup>3</sup>, <sup>1</sup>University of Nottingham, Sutton Bonington, Loughborough, UK, <sup>2</sup>University of Reading, Whiteknights, Reading, UK, <sup>3</sup>Institute for Animal Health, Compton, Newbury, UK.

In dairy cows the timing and strength of the postovulatory progesterone rise is a key determinant of early embryo development and survival. The aim of this study was to determine whether postovulatory luteal function is related to preovulatory follicle development. Luteal regression was synchronized in 25 non-lactating Holstein Friesian cows which subsequently underwent daily trans rectal ultrasonography to determine the growth pattern and size of the ovulatory follicle. Following ovulation, daily plasma samples were collected for 6 days to determine progesterone concentrations. The mean (sem) diameter of the ovulatory follicle rose from 10.9 $\pm$ 0.8 mm at luteolysis to 15.9 $\pm$ 0.5 mm at ovulation 4.0 $\pm$ 0.2 days later. There was a significant relationship between initial follicle diameter and time to ovulation with smaller follicles taking longer to ovulate (r<sup>2</sup> = 0.63; p<0.001). The mean plasma concentration of progesterone (from days 4-6 post ovulation) increased significantly with increasing initial (r<sup>2</sup> = 0.34; p<0.01) and final (r<sup>2</sup> = 0.24; p<0.05) diameter of the ovulatory follicle and decreased with increasing time from luteolysis to ovulation (r<sup>2</sup> = 0.22; p<0.05). The mean interval from ovulation to the onset of the postovulatory progesterone rise (>1ng/ml) decreased with increasing initial (r<sup>2</sup> = 0.34; p<0.01) and final follicle diameter (r<sup>2</sup> = 0.22; p<0.05) and increased with increasing days to ovulation (r<sup>2</sup> = 0.34; p<0.01). The results demonstrate that increased progesterone secretion by the developing corpus luteum is related to the ovulation of a larger follicle that took less time to ovulate. Whether it is

follicle size or time to ovulation that is the more important determinant of subsequent luteal function remains to be determined.

**Key Words:** Cow, Follicle, Corpus Luteum

**136 Effects of Acute Nutritional Restriction of Beef Heifers on LH in serum and Anovulation.** C. A. Lents\*, F. J. White, L. N. Floyd, N. H. Ciccioli, I. Rubio, and R. P. Wettemann, *Department of Animal Science, Oklahoma Agricultural Experiment Station.*

Angus x Hereford heifers (BCS =  $5.5 \pm 0.1$ ;  $387 \pm 7$  kg BW) exhibiting normal estrous cycles at 15 mo of age were used to determine the effects of acute nutritional restriction on LH in serum and the incidence of anovulation. Heifers were maintained in individual pens and adapted to a 1.2 x maintenance (M) diet for 10 d. On d 0, heifers were randomly assigned to 0.4 M (n = 16) or 1.2 M (n = 7). Heifers were treated with PGF $_{2\alpha}$  on d -10, 0, and 10. Daily blood samples were obtained by tail venipuncture and progesterone in plasma was quantified. Jugular catheters were inserted on d 8. Blood samples were collected every 10 min for 8 h on d 9, 10 and 11, and LH in serum was quantified. Beginning on d 12, blood samples were collected from 0.4 M (n = 9) and 1.2 M (n = 4) heifers every 4 h for 48 h to quantify estradiol in plasma and the ovulatory surge of LH. Heifers with plasma progesterone less than 0.5 ng/mL on d 14 to 21 were classified as anovulatory. On d 12, 0.4 M heifers weighed less ( $P < 0.01$ ) than 1.2 M heifers ( $353 \pm 9$  kg vs  $391 \pm 7$  kg, respectively), but BCS was similar for 0.4 M and 1.2 M heifers ( $5.4 \pm 0.1$ ). On d 14, 44% (4 of 9) of 0.4 M heifers were anovulatory and all 1.2 M heifers ovulated. Concentrations of LH in serum and frequency of LH pulses on d 9, 10, or 11 were not influenced by treatment. Anovulatory heifers lacked an ovulatory surge of LH and concentrations of estradiol were similar and minimal on days 2, 3, and 4 after PGF $_{2\alpha}$  ( $1.1 \pm 0.1$  pg/mL). Heifers on 1.2 M had a preovulatory increase in plasma estradiol ( $2.7 \pm 0.4$  pg/mL) preceding the ovulatory surge of LH. In conclusion, acute nutritional restriction for 14 d induced anovulation in 44% of heifers, did not influence serum concentrations of LH or frequency of LH pulses during follicular growth, but inhibited the proestrous increase in estradiol and the ovulatory surge of LH.

**Key Words:** Heifers, Nutrition, LH

**137 Estradiol benzoate (EB) inhibits secretion of LH and induces atresia of dominant follicles within 36 hours in cyclic heifers.** C.R. Burke<sup>\*1,2</sup>, S. Morgan<sup>2</sup>, M.L. Mussard<sup>1</sup>, D.E. Grum<sup>1</sup>, and M.L. Day<sup>1</sup>, <sup>1</sup>The Ohio State University, Columbus OH, <sup>2</sup>Dexcel Ltd, Hamilton, New Zealand.

The aim of this study was to characterize LH secretion and timing of functional atresia in dominant follicles (DF) of cyclic heifers treated with EB. At  $5.6 \pm .1$  d after estrus, heifers received 1 mg EB/500 kg BW (designated h 0; T; n=15) or remained untreated (C; n=15). Ovarian structures were monitored by ultrasonography at least daily from estrus to ovariectomy or new follicle wave emergence. Blood samples were collected every 20 min for 12 h beginning at h -12, 0, 24 and 48 from seven heifers in each treatment to characterize LH secretion. Four heifers from each of these groups were ovariectomized 12 h after new wave emergence. The remaining eight heifers per treatment were ovariectomized at either h 12 or 36. Concentrations of estradiol-17 $\beta$  (E $_2$ ) and progesterone (P $_4$ ) were determined in the fluid of DF. Maximum diameter of DF was less in the T ( $12.0 \pm .8$  mm) than the C ( $14.0 \pm .4$  mm) treatment (treatment x time;  $P < .01$ ) but the interval to new wave emergence ( $4.6 \pm .2$  d) was similar among treatments. Mean E $_2$  and E $_2$ :P $_4$  ratio in DF of T heifers at 36 h ( $48.6 \pm 4.25$  ng/ml and  $2.37 \pm .6$ ) was less ( $P < .01$ ) than in C heifers at 36 h ( $258.13 \pm 26.7$  ng/ml and  $7.3 \pm .7$ ), and comparable ( $P > .1$ ) to that in C heifers after a new follicle wave had emerged ( $27.8 \pm 17.6$  ng/ml and  $1.3 \pm .1$ ). Mean concentrations of LH and amplitude of LH pulses were lower in the T than C treatment at h 24 and 48 (treatment x time,  $P < .05$ ) while LH pulse frequency was similar between treatments. Maximum P $_4$  in plasma after treatment was lower ( $P < .01$ ) in T ( $3.2 \pm .2$  ng/ml) than C treatment ( $4.87 \pm .5$  ng/ml). The results indicate that EB-induced atresia of dominant follicles is achieved by 36 h and is accompanied by a reduction in LH pulse amplitude and mean concentration. It is also suggested that the interval from EB treatment to emergence of a new follicle wave is not entirely dependent on the functional status of the present DF.

**Key Words:** Estrous synchronization, Follicular atresia, Estradiol

**138 Effect of Heat Stress in Follicular Development of Dairy Cows in Intensive Production in North-Central Mexico.** R.R. Lozano-Dominguez<sup>1</sup>, C.F. Arechiga<sup>2</sup>, and E. Gonzalez-Padilla<sup>\*1</sup>, <sup>1</sup>Universidad Nacional Autonoma de Mexico, Mexico, <sup>2</sup>Universidad Autonoma de Zacatecas, Zacatecas, Mexico..

To assess the effect of heat stress in lactating dairy cows in intensive farming conditions, on ovarian follicular development, a study was carried out with 56 Holstein cows located in different farms in the state of Aguascalientes, Mexico. Daily ultrasound observations of both ovaries were performed between two estrous cycles. Average production of the herds was around 9500 kg per cow per year and studied cows were cycling regularly and had  $71.2 \pm 17.9$  d in milk. Measurements were taken at four different times of the year according to temperature-humidity index 60 days prior or during the studied cycle. Therefore there were stressful (S) or neutral (N) conditions before or during evaluation: SN (n = 24), NN (n = 11), NS (n = 11) and SS (n = 10). The variables studied were: number of follicular waves per cycle (FW: 1, 2, and 3); maximum diameter of the dominant follicle (DF) in each wave (DDF, mm); growth rate of each DF (GRDF: 1 - first dominant follicle and 2 - second dominant follicle, mm/d); and growth length of each DF (GLDF, d). Data were analyzed by least square analysis of variance. Chi-square was utilized for analysis of proportions. In NN the DDF of FW1 was larger ( $20.9 \pm 0.6$ ,  $P < 0.05$ ) than in the other groups (SN,  $19.1 \pm 0.6$ ; NS,  $18.0 \pm 0.8$ ; SS,  $18.6 \pm 0.9$ ). In FW2, the DDF of NS ( $16.9 \pm 0.7$ ) and SS ( $16.7 \pm 0.8$ ) were smaller ( $P < 0.05$ ) than those of the SN ( $18.9 \pm 0.6$ ) and NN ( $19.2 \pm 0.8$ ). There were no differences in GRDF1 between groups ( $P < 0.05$ ); however GRDF2 in SS ( $2.2 \pm 0.3$ ) was larger than in the other groups (SN,  $1.4 \pm 0.2$ ; NN,  $1.5 \pm 0.3$ ; NS,  $1.3 \pm 0.3$ ) and had GLDF shorter ( $4.7$  d,  $P < 0.05$ ) than SN ( $8.6$  d), NN ( $9.1$  d) or NS ( $7.1$  d). Estrous cycles in SS group had more FW (2.6) ( $P < 0.05$ ) than the rest. In SS + NS, in 36% of the cycles the ovulated follicle was of FW3 while this occurred in 9.1% of the cycles of NN+SN ( $P < 0.05$ ). Thermal-humidity stress induces shorter periods of follicular growth and dominance and increases estrous cycles of three follicular waves, which may be related with lowered fertility in the warmer seasons of the year.

**Key Words:** follicular development, dairy cow, Mexico

**139 Expression of insulin-like growth factor-binding protein-2, -3, -4, and -5 messenger RNA in fresh versus cultured bovine granulosa and theca cells.** J.L. Voge\*, D.T. Allen, J.R. Malayer, and L.J. Spicer, *Oklahoma State University.*

The objective of this study was to determine the presence of messenger RNA (mRNA) for IGF-binding protein (IGFBP)-2, -3, -4, and -5 in fresh versus cultured bovine granulosa and theca cells. Granulosa cells (GC) from small follicles (1-5 mm) and theca cells (TC) from large follicles ( $> 7.9$  mm) were collected from cattle and RNA isolated either before or after in vitro culture. For in vitro derived RNA, GC and TC were cultured for 2 d in medium with 10% fetal calf serum and an additional 2 d in serum-free medium. RNA from fresh and cultured cells was isolated using the Trizol extraction method. The presence of mRNA for IGFBP-2, -3, -4, and -5 was assessed using reverse transcriptase-PCR. IGFBP-2 mRNA was detected in 2 of 2 fresh and 4 of 5 cultured GC samples, and in 2 of 2 fresh and 4 of 6 cultured TC samples; the proportion of TC (85.7%) and GC (75%) samples that had detectable IGFBP-2 mRNA did not differ ( $P > 0.50$ , chi-square). IGFBP-3 mRNA was not detectable in either fresh or cultured GC, whereas IGFBP-3 mRNA was detectable in all fresh and cultured TC samples ( $P < 0.005$ ). IGFBP-4 mRNA was expressed in 1 of 2 fresh GC samples and 3 of 5 cultured GC samples. IGFBP-4 mRNA expression was found in all fresh TC samples and in 4 of 6 cultured TC samples; the proportion of TC samples (87.5%) that had detectable IGFBP-4 mRNA tended to be greater ( $P < 0.10$ ) than that of GC samples (42.9%). IGFBP-5 mRNA was detected in 1 of 2 of the fresh and 3 of 5 of the cultured GC. All of fresh TC and 4 of 6 cultured TC samples had IGFBP-5 mRNA; the proportion of TC (75%) and GC (57%) samples that had detectable IGFBP-5 mRNA did not differ ( $P > 0.25$ ). These results indicate that IGFBP-3 may be produced by TC and not GC, whereas IGFBP-2, -4, and -5 are produced by both TC and GC. Furthermore, in vitro culture had no effect on whether or not IGFBP-2, -3, -4, and -5 mRNA was expressed in small GC or large TC.

**Key Words:** Insulin-like Growth Factors, Binding Proteins, Ovarian Follicles

**140 Insulin plays a key role in re-coupling the IGF-somatotropin axis in the early postpartum dairy cow.** S.T. Butler\* and W.R. Butler, *Cornell University, Ithaca, NY.*

Negative energy balance associated with the onset of lactation results in hypoinsulinemia, uncoupling of the IGF-somatotropin axis, attenuation of gonadotropin release and delayed first ovulation. Our objectives were to examine the effects of elevated insulin during the immediate postpartum period on circulating IGF-I concentrations, ovarian follicular growth, estradiol secretion and LH pulse profiles. Holstein cows ( $n=14$ ) were subjected to either a hyperinsulinemic-euglycemic clamp (INS) or saline infusion (CTL) for 96 hours starting on day 10 post-calving. Blood samples were taken on days 8-9 to establish baseline glucose values. Insulin was infused continuously ( $1 \mu\text{g/kg BW/hr}$ ) via a jugular catheter. Blood samples were collected hourly, and euglycemia was maintained by infusion of exogenous glucose. During infusion, insulin concentrations were increased 8-fold in INS cows over those in CTL cows ( $2.4 \pm 0.1$  vs.  $0.3 \pm 0.1$  ng/ml;  $P<0.001$ ), while blood glucose concentrations were not different between the treatments ( $45.4 \pm 2.2$  vs.  $41.7 \pm 2.3$  mg/dl;  $P=0.27$ ). Plasma IGF-I increased continuously during the insulin infusion, and reached the highest values at the end of the clamp, being almost 4-fold higher in INS compared with CTL cows ( $122 \pm 11$  vs.  $33 \pm 2$  ng/ml;  $P<0.001$ ). Ultrasound measurements of ovarian follicular growth revealed that 2 cows ovulated, both of which were CTL cows. Among non-ovulatory cows, the dominant follicle reached a greater maximum diameter in the INS cows compared with CTL cows ( $13.4 \pm 0.5$  vs.  $11.6 \pm 0.6$  mm;  $P=0.04$ ). Excluding ovulatory cows, no difference in plasma estradiol was observed between groups during the infusion period ( $1.3 \pm 0.2$  vs.  $1.1 \pm 0.3$  pg/ml;  $P=0.69$ ). Blood samples collected every 10 minutes for 8 hours prior to and at the end of the infusion periods showed no differences between groups in LH pulse frequency, pulse amplitude or area under the curve. In conclusion, insulin appears to be a key metabolic signal in coupling the IGF-somatotropin axis, orchestrating the observed marked elevation in circulating IGF-I.

**Key Words:** Insulin, IGF-I, Ovary

**141 Postpartum nutrition influences concentrations of leptin, IGF-I, and pregnancy rate of primiparous beef cows.** N. H. Ciccioli<sup>1</sup>, R. P. Wettemann<sup>1</sup>, L. J. Spicer<sup>1</sup>, D. H. Keisler<sup>2</sup>, C. A. Lents<sup>1</sup>, and F. J. White<sup>1</sup>, <sup>1</sup>Oklahoma Agricultural Experiment Station, Stillwater, <sup>2</sup>University of Missouri-Columbia.

The influence of nutrition on plasma concentrations of leptin and IGF-I and reproductive function was determined at the first estrus in Hereford x Angus spring calving primiparous cows. At parturition, cows (BCS =  $4.3 \pm 0.1$ ; BW =  $385 \pm 17$  kg) were blocked by BCS and calving date and randomly assigned to gain 0.45 (M;  $n = 17$ ) or 0.90 (H;  $n=17$ ) kg/d for 11 wk. Then, all cows were fed the same (M) diet until the first estrus. A second replication was added to assess pregnancy rate (M;  $n = 13$ ; H;  $n=17$ ). Leptin and IGF-I were quantified weekly and progesterone thrice a week in blood plasma. Estrous behavior was detected with Heatwatch<sup>®</sup> and ovulation was determined using plasma progesterone. The dominant follicle was measured by ultrasonography at 4 to 14 h after onset of estrus. All cows were AI between 12 and 20 h after onset of estrus. During treatment, H cows gained ( $P < 0.01$ ) BW

and BCS and had greater ( $P < 0.01$ ) concentrations of leptin and IGF-I compared with M cows. From the end of treatment to first estrus, concentrations of IGF-I were greater ( $P < 0.01$ ) in H cows; however, concentrations of leptin decreased ( $P < 0.01$ ) in H cows after treatment termination and did not differ ( $P = .16$ ) from those of M cows through the first estrus. Cows that exhibited estrus and ovulated on or before 19 wk postpartum had greater ( $P < 0.06$ ) concentrations of leptin on wk 13 to 15 than anestrus cows. During treatment, leptin and IGF-I concentrations were positively correlated ( $P < 0.05$ ) with changes in BW and BCS. H cows had a larger ( $P < 0.05$ ) dominant follicle, more mounts ( $P < 0.06$ ), and a shorter ( $P = 0.06$ ) interval to estrus than M cows. Pregnancy rate at the first postpartum estrus was greater ( $P < 0.01$ ) for H ( $n=34$ ; 82.3 %) than for M ( $n=30$ ; 60 %) cows. We conclude that increased nutrient intake after calving results in increased concentrations of IGF-I in plasma, and increased follicle size and pregnancy rate at the first estrus.

**Key Words:** IGF-I, Leptin, Reproduction

**142 Concentrations of leptin and insulin like growth factor-I (IGF-I) during acute nutritionally induced anovulation and realimentation.** F.J. White<sup>\*1</sup>, C.A. Lents<sup>1</sup>, N.H. Ciccioli<sup>1</sup>, R.P. Wettemann<sup>1</sup>, L.J. Spicer<sup>1</sup>, and D.H. Keisler<sup>2</sup>, <sup>1</sup>Oklahoma Agricultural Experiment Station, Stillwater, <sup>2</sup>University of Missouri, Columbia.

Luteal activity and concentrations of leptin and IGF-I were evaluated during acute nutritional restriction and realimentation of beef heifers. Angus x Hereford heifers (14 mo old;  $n=19$ ) were housed in individual pens and fed a diet supplying 1.2 x maintenance (M) for 1 wk. Then heifers were randomly allotted on d 0 to either 0.4 or 1.2 M. Heifers were treated with PGF2 $\alpha$  on d -10, 0, and 10 to synchronize ovulation. After 30 d, 0.4 M heifers were gradually increased to 1.2 M during 10 d. Blood was collected 23 h after feeding on alternate days during restriction and 3 x per wk during 100 d realimentation. Heifers with progesterone  $<0.5$  ng/mL for 8 d were classified as anovulatory. Seventy percent (7 of 10) of 0.4 M heifers did not ovulate on d 14 while all 1.2 M heifers had normal luteal function. Plasma IGF-I in 0.4 M heifers decreased from  $49 \pm 3$  ng/mL on d 0 to  $33 \pm 4$  ng/mL on d 14; however, 1.2 M heifers had similar concentrations on d 0 and 14 ( $57 \pm 3$ ; day x diet effect,  $P < .01$ ). During restriction, heifers on 0.4 M tended ( $P = 0.1$ ) to have decreased concentrations of leptin (14 % less) in serum compared with 1.2 M. Five of the anovulatory heifers had luteal activity by 16 to 51 d of realimentation at 1.2 M (mean = 35 d); however, two heifers did not ovulate by 100 d. Realimentation with 1.2 M for 2 wk increased concentrations of IGF-I in 0.4 M heifers (day x diet effect,  $P < 0.05$ ) to concentrations similar to 1.2 M heifers. During realimentation, concentrations of leptin in blood samples taken after 23 h of fasting were similar ( $P > 0.1$ ) in 0.4 and 1.2 M heifers. Concentrations of IGF-I in plasma decreased with acute nutritionally induced anovulation and were similar to those in control heifers (1.2 M) by 2 wk of realimentation at 1.2 M. Systemic concentrations of IGF-I were more indicative of nutritional status than concentrations of leptin in these acute nutritionally restricted heifers.

**Key Words:** Beef heifer, IGF-I, Leptin

## ASAS/ADSA Ruminant Nutrition: Feed Additives

**143 Influence of length and ramification of the alcohol radical of esters of methionine and of 2-hydroxy-4 (methylthio) butanoic acid on methionine bioavailability.** J.C. Robert<sup>\*1</sup>, B.K. Sloan<sup>2</sup>, G. Etave<sup>1</sup>, and B. Bouza<sup>1</sup>, <sup>1</sup>Aventis Animal Nutrition, Antony, France, <sup>2</sup>Aventis Animal Nutrition, Alpharetta, USA.

In a series of five experiments, Methionine bioavailability from 8 DL-methionine and 3 HMB esters were tested according to the blood kinetics method based on determining the Area Under the Curve (AUC). The alcohol radical varied in number of carbons but all were arranged in a linear form. Smartamine<sup>TM</sup> M (a methionine coated with a pH sensitive polymer based coating) of which the methionine bioavailability is 80%, was used as a reference. Non lactating rumen cannulated Holstein cows, receiving 10 kg / animal / day of a ration comprising 75% hay and 25% concentrate delivered in equal quantities twice a day, were used. A single dose, 50 g of methionine equivalent, was supplied directly

into the rumen at 0800 on day two (D2) for the esters and at 1600 on day one (D1) for Smartamine<sup>TM</sup> M. Blood samples were obtained, on D2 at 0900,1000, 1100, 1300, 1500h and, thereafter, every three hours from 0900 to 1500h on D3 and D4. For Smartamine<sup>TM</sup> M, blood samples were collected every two hours on D2, starting at 0600 until 1000h and, thereafter, every three hours on D3 and D4 from 0600 until 1500h. Blood plasma methionine concentration (BPMC mg/100g) for base line determinations were measured on D1 at 0900,1100 and 1500h. Modelization of the AUC results for BPMC for the 11 esters resulted in the following relationship :  $Y = 40.6787 \exp(-0.2272 X)(R^2 = 0.89)(\text{SED} = 6.13)$  Y = bioavailability as a percentage of methionine equivalent ingested. X = alcohol carbon number. It shows that methionine bioavailability decreases with increasing number of carbons in the alcohol. Three esters with branched alcohol radicals were tested vs. their corresponding linear forms using the same methodology described above. In all cases, methionine bioavailabilities with the corresponding branched alcohol were higher.