are postulated to be intermediates in myofibrillar turnover. It is unclear whether ERMs are indeed a subset of myofilaments on the surface of myofibrils or whether they are simply removed by shearing during trituration and more filaments can be removed by repeated trituration. We have found that ERMs can be prepared from either rat or bovine muscle; that the yield is less than previously reported (0.5-0.8 % of myofibrillar protein); that once removed, repeated trituration does not yield more ERMs; and that ERMs can not be obtained from thoroughly washed myofibrils where presumably the ERMs have already been removed. Hence, ERMs seem to be a real subset of filaments. Mild treatment with calpain increases the yield of ERMs by 2 to 2.5-fold, so the calpains can release ERMs as proposed over 30 years ago. Supported by NRI, NIH, MDA.

Key Words: Calpain, Proteasome, Myofibrillar Protein

633 The mTOR-signaling pathway in regulating metabolism and growth. X. Yang*, C. Yang, A. Farberman, C. F. M. de Lange, J. France, and M. Z. Fan, *University of Guelph*, *Guelph, Ontario, Canada*.

The mammalian target of rapamycin (mTOR) plays key roles in cell growth and the cell cycle and acts as a central regulator of protein synthesis and ribosome biogenesis at transcriptional and translational levels. mTOR senses and integrates signals from mitogens and nutrients. Ribosomal protein S6 protein kinase S6K1 and eukaryotic initiation factor 4E binding protein 4E-BP1 are currently the two best-known downstream effectors of mTOR signaling. Interactions of mTOR with raptor or rictor result in two types of mTOR complexes with the former being the primary controller of cell growth and the latter mediating effects that are insensitive to rapamycin such as cytoskeletal organization. Upstream elements of mTOR signaling include Ras-homolog enriched in brain (Rheb), and tuberous sclerosis complex 1 and 2 (TSC1/2) with TSC2 as the linker between PI3K/Akt or Ras/Raf/MEK/ERK pathways and the mTOR pathway. AMP activated kinase (AMPK), an important cellular energy sensor, can work with mTOR signalling to maintain cellular energy homeostasis. Nutrients and hormonal factors can differentially mediate metabolism and cellular growth via the mTOR pathway with effectors specific to organ or tissue types involved.

Key Words: Growth, Metabolism, Mammalian Target of Rapamycin (mTOR)

Physiology & Endocrinology - Livestock and Poultry: Endocrinology

634 Relationship between leptin and carcass quality and yield grade in a population of Certified Angus Beef–type cattle. D. L. McNamara*[1], T. B. Schmidt[3], E. L. Walker[4], M. M. Rolf[1], A. N. Brauch[1], W. Pittroff[2], and D. H. Keisler[1], [1]*University of Missouri*, *Columbia*, [2]*University of California*, *Davis*, [3]*Mississippi State University*, *Starkville*, [4]*Missouri State University*, *Springfield*.

Leptin is a protein hormone secreted by adipocytes. Serum concentrations of leptin increase with adiposity in various species, including beef cattle. In this investigation, we utilized a relatively uniform population of cattle — i.e. those destined for a Certified Angus Beef–type market, to determine the relationship between serum concentrations of leptin and phenotypic variables associated with carcass quality. This work differs from our prior investigations in which we utilized a non–uniform and random population of cattle from a commercial slaughter facility. Our hypothesis was that serum concentrations of leptin would be higher in heifers than steers and that serum concentrations of leptin would be an accurate, positive indicator of carcass quality and yield grades in a Certified Angus Beef–type population of cattle. In the current investigation blood samples were collected at slaughter and analyzed for serum concentrations of leptin from 2,815 black slaughter steers and heifers. The PROC GLM method of SAS was used with leptin as the dependent variable and all carcass merit variables analyzed as independent variables within the model. Any independent variables that were not significant within the model were removed from the final analysis. We observed that leptin levels were significantly greater in heifers than steers (25.12 vs 20.94 ng/ml, respectively; $P < 0.0001$). Independent of gender however, leptin concentration at the time of slaughter was a significant predicative indicator of the carcass quality grades: select, low choice, upper two-thirds choice, high choice, and prime (20.60, 22.16, 23.38, and 25.99 ng/ml, respectively; $P < 0.006$). Likewise, USDA Yield grades were resolvable by leptin levels at Yield grades 2, 3, and 4 (20.95, 23.32, and 25.74, respectively; $P < 0.001$), but incapable of resolving Yield grade 4 vs. 5 cattle — a threshold point (yield grade >3) at which carcasses typically begin to be discounted for excessive fat. We suggest that these data provide evidence that serum concentrations of leptin are indicative of the greater fat mass across gender and carcass merit.

Key Words: Leptin, Adipose, Carcass

635 Variation in maintenance energy requirements of gestating beef cows and relationships with calf performance and plasma IGF-I. M. J. Prado-Cooper*, N. M. Long, R. P. Wettemann, G. W. Horn, L. J. Spicer, and C. R. Krehbiel, *Oklahoma Agricultural Experiment Station*.

Variation in maintenance energy requirements (MR) was determined in spring-calving Angus × Hereford cows during gestation in each of two years (yr 1, n = 27; yr 2, n = 32). A second objective was to determine if MR were related to plasma concentrations of IGF-I and postnatal calf growth. Nonlactating cows (4 to 7 yr of age) with a BCS of 5.0 ± 0.2, and BW of 582 ± 37 kg, in the second to third trimester of gestation, were individually fed a complete diet in amounts to meet predicted MR (Model 1, NRC 2000). After 2 wk, daily feed intake was adjusted each 7 d until constant BW was achieved. Regression analysis was used to determine constant BW. Final BCS averaged 5.0 ± 0.2 (yr 1) and 4.6 ± 0.4 (yr 2). Daily MR averaged 0.0892 (yr 1) and 0.0930 (yr 2) (Mcal/$BW_{kg}^{.75}$). Cows were classified based on MR as low (> 0.5 SD less than mean, L), moderate (± 0.5 SD of mean, M) or high (> 0.5 SD more than mean, H). The greatest differences in MR for all cows were 29% (yr 1) and 24% (yr 2). ADG and IGF-I were

analyzed using the GLM and MIXED procedures (SAS), respectively. In yr 1, 205 d BW of calves (200 kg) was not influenced by MR. Plasma concentrations of IGF-I (yr 2), after consuming NRC predicted MR, were greater ($P < 0.005$) for M cows (66.5 ± 4.8 ng/mL) compared with L cows (38.9 ± 5.7 ng/mL), and H cows (52.8 ± 4.8 ng/mL) were not different from M or L cows. After 3 wk of constant BW, concentrations of IGF-I were not different ($P = 0.14$) among L, M and H cows (40.2, 52.1 and 51.2 ± 4.4 ng/mL, respectively). Concentrations of IGF-I in plasma were not correlated with dietary intake (Mcal/d) or BW. With ad libitum intake (yr 2), ADG of the cows was influenced by MR; L cows had less ($P < 0.01$) ADG compared with M and H cows (1.0, 1.4, 1.6, ± 0.3 kg/d, respectively). Variation in MR of cows during gestation was not associated with performance of calves. Identification of cows that require less energy to maintain weight may increase efficiency of production.

Key Words: Maintenance, Beef Cattle, Calves

636 Negative energy balance increases prandial ghrelin and growth hormone concentrations in lactating dairy cows. B. J. Bradford* and M. S. Allen, *Michigan State University*, *East Lansing*.

The reported effects of feeding on growth hormone (GH) secretion in ruminants have been inconsistent, and may be influenced by energy status of animals. High-producing dairy cows in early lactation and late lactation were used to assess the effects of energy balance on temporal variation of plasma metabolites and hormones. Cows were fed a TMR once daily, and feed was withdrawn for 90 min prior to feeding. Beginning at the time of feed withdrawal, plasma samples were collected via jugular catheters hourly for 24 h. Concentrations of non-esterified fatty acids and GH were measured for all samples, while insulin, glucose, and acylated (active) ghrelin were quantified for 4 sample times around feeding. Blood plasma data was analyzed using a mixed model with repeated measures over time. As expected, calculated energy balance was significantly lower in early lactation than late lactation cows (-10.4 vs. 1.7 Mcal retained /d, $P < 0.001$). Following the primary meal of the day, a GH surge was observed in early lactation but not in late lactation cows (time × stage of lactation interaction: $P < 0.01$). This difference was not explained by temporal patterns in non-esterified fatty acid, insulin, or glucose concentrations. However, a preprandial ghrelin surge was observed in early lactation only (76.8 vs. 40.1 pg/mL, $P < 0.05$), suggesting that ghrelin was responsible for the prandial GH surge in this group. Results of a stepwise regression statistical analysis showed that both preprandial ghrelin concentration and energy balance were significant predictors of prandial GH increase over baseline. Adaptations to negative energy balance in lactating dairy cattle likely include enhanced ghrelin secretion and greater GH response to ghrelin.

Key Words: Fhrelin, Growth Hormone, Energy Balance

637 Effect of ghrelin and obestatin infusion on milk production, body condition score, and energy balance in dairy cows. J. R. Roche*[1,2], A. J. Sheahan[1], L. M. Chagas[1], D. Blache[3], D. P. Berry[4], and J. K. Kay[1], [1]*Dexcel*, *New Zealand*, [2]*University of Tasmania*, *Australia*, [3]*University of Western Australia*, *Australia*, [4]*Teagasc Moorepark*, *Ireland*.

Ghrelin is an endogenous ligand of the growth hormone secretagogue receptor and a potential orexigenic agent in monogastrics and ruminants. Obestatin has been reported to have the opposite effect, reducing appetite. Fifty one multiparous cows were randomly allocated to one of three groups (n=17 cows/treatment); a control (C) and cows continuously infused with either 0.74 μmol/d of ghrelin (G) or obestatin (O) subcutaneously. Infusions began 20 days in milk and treatments continued for 8 wk. Milk yield was recorded daily and milk composition weekly. Blood was sampled every two wk. Generalized linear models were used to determine the treatment effect on average daily and cumulative milk production and daily composition, plasma ghrelin, growth hormone, IGF-1, leptin, NEFA and glucose. Mixed models, with cow included as a repeated effect, were used to determine treatment effects on weekly milk production, body weight and body condition score (scale 1-10). Parity, breed, wk of the yr at calving, treatment, wk post-calving and the 2 wk pre-experimental average of each measure (covariate) were included as fixed effects. Despite a numerical tendency for G cows to produce more milk (1,779kg) than either C (1,681kg) or O (1,714kg) cows during the eight wk period, differences were not significant ($P=0.39$). Similarly, there was no significant effect of treatment on milk fat, protein or lactose per cent or yield. Similarly, treatment did not affect DMI. Cows infused with G lost significantly ($P<0.05$) more BCS (0.68 BCS units) over the 8 wk study period than the O infused (0.40 BCS units) or C (0.25 BCS units), which did not differ significantly from each other. Treatment had no significant effect on BW change. Although, treatment did not significantly affect plasma ghrelin, IGF-1, or growth hormone concentration, plasma NEFA concentration was elevated ($P<0.05$) and there was a tendency ($P<0.10$) for plasma leptin to be reduced in G cows. Results indicate an effect of ghrelin infusion on lipolysis.

Key Words: Ghrelin, Obestatin, Dairy

638 Expression of ghrelin and the growth hormone secretagogue receptor 1a (GHS-R1a) in the reproductive tissues of Holstein heifers. M. L. Rhoads*, J. B. Wheelock, L. L. Hernandez, R. P. Rhoads, and R. J. Collier, *University of Arizona*, *Tucson*.

The hormone ghrelin and the active form of its receptor (growth hormone secretagogue receptor 1a; GHS-R1a) are expressed in the reproductive tissues of several species and may be involved in the metabolic regulation of reproductive function. However, little is known about the expression of ghrelin and GHS-R1a in the reproductive tissues of dairy cattle. The objective of this study was to characterize the expression of ghrelin and GHS-R1a in the reproductive tissues of dairy cattle. Reproductive tissues (follicle, CL, ampulla, isthmus, uterine horn, endometrium and uterine body) were collected from three Holstein heifers immediately following slaughter. Samples of the GI tract (reticulum, rumen, omasum, abomasum, duodenum, jejunum, ileum and colon) were simultaneously collected for comparative purposes. Ghrelin and GHS-R1a mRNA abundance were evaluated by real-time RT-PCR. Within the reproductive tract, ghrelin and GHS-R1a gene expression was detectable in all tissues with the greatest expression in the ampulla (0.99 ± 0.09 and 1.94 ± 0.14 AU, respectively; $P<0.001$). Ghrelin expression did not differ based on proximity to the corpus luteum. However, GHS-R1a expression was lower in the ipsilateral ampulla than in the contralateral ampulla (1.14 ± 0.20 vs. 2.75 ± 0.20 AU; $P<0.02$). GHS-R1a mRNA was detected in bovine oocytes and IVF-produced d 8 embryos whereas ghrelin expression was detectable, but low. Among the GI tissues, ghrelin

expression was greatest in the duodenum (12.71 ± 0.65 AU; P<0.001) and was also high in the abomasum (3.27 ± 0.65 AU; P<0.13) while, numerically, GHS-R1a mRNA concentrations were greatest in the ileum (4.45 ± 0.37 AU) and jejunum (3.88 ± 0.37 AU). Both ghrelin and GHS-R1a are expressed throughout the reproductive tract, albeit at lower concentrations than in some GI tissues. The observed patterns of expression suggest that the metabolic regulation of reproduction may be mediated through ghrelin signaling during the early stages of embryo development.

Key Words: Ghrelin, GHS-R1a, Dairy Cattle

639 Seasonal effects on twenty-four hour patterns of melatonin in blood and milk of dairy cows. N. Castro*[1,2], M. T. Kollmann[3], V. Lollivier[4], S. Richter[1], A. Baumert[1], O. Wellnitz[1], and R. M. Bruckmaier[3], [1]*University of Bern*, *Bern, Switzerland*, [2]*Las Palmas de Gran canaria University*, *Las Palmas, Spain*, [3]*Technical University Munich*, *Germany*, [4]*INRA*, *France*.

Pineal secretion of melatonin (MEL) is usually low during light exposure and high during the night. In this study MEL concentration in blood and milk was measured every h during 24 h (7.00 AM-6.00 AM) in June and December in 12 high yielding dairy cows which were housed under natural light conditions in Switzerland, i.e.at 16 and 9 h daylight respectively. Blood samples were taken and the whole available milk was collected by machine milking after an i.v. injection of 1 i.u. of oxytocin every h. No artificial light was used throughout the experiment except for small head lamps during sampling in the dark nights. MEL in blood and milk was determinated by ELISA. For all statistical analyses a general linear model procedure with repeated measures and post hoc Tukey analysis was used. Both MEL blood and milk concentration showed a diurnal pattern with high levels during scotoperiod and low levels during photoperiod. In June melatonin blood levels (pg/ml) were 2.3 ± 0.2 during the day (08.00 to 22.00 h), started to increase at 23.00 h (16.0 ± 3.8) and decreased to baseline at 07.00 h (5.3 ± 2.2). Peak MEL was observed at 01.00 h (25.3 ± 6.2). In December MEL blood levels increased already at 17.00 h (16.3 ± 3.3) and the decrease started at 07.00 h (12.2 ± 2.2) Peak MEL was 40.4 ± 5.2 at 22.00 h. Areas under the curve (AUC) were calculated for samples from 23.00 h to 06.00 h. AUC/h in blood and in milk was higher ($p < 0.05$) in winter than in summer (22.4± 2.4 and 16.2 ± 2.3, 7.2 ± 1.0 and 2.2 ± 0.5, resp.). In all individuals MEL was higher in blood than in milk in both seasons. AUC of milk melatonin concentration was 20 ± 7 % in summer and 35 ± 7 % in winter of the MEL in blood. In conclusion, milk MEL is elevated, in parallel with blood MEL, albeit at a lower level, most likely being transferred by the way of diffusion with water.

Key Words: Melatonin, Daylight, Milk

640 Effect of restricted feeding and monopropylene glycol postpartum on metabolic hormones and postpartum anoestrus in grazing dairy heifers. L. M. Chagas*[1], P. J. S. Gore[1], K. A. Macdonald[1], and D. Blache[2], [1]*Dexcel Limited*, *Hamilton, New Zealand*, [2]*The University of Western Australia*, *Crawley, Australia*.

Pastoral dairying in New Zealand requires a 12 months calving interval to balance cow nutrient demands with pasture growth, however, in 30% of cows postpartum anoestrous interval (PPAI) exceeds 80 days resulting in lost productivity. Supplementary feeding can overcome low pasture availability and a previous study showed that 250 ml of monopropylene glycol (MPG) given twice a day to Holstein-Friesian heifers calving in low body condition score (BCS; 2.8) reduced PPAI. In that study 82% of heifers given MPG were cycling by 12 weeks postpartum compared to 28% of controls. A further study was undertaken with MPG supplements and feed restriction using heifers calving in good BCS (3.1). Treatments comprised of *ad libitum* pasture (Adlib; n=18) and two groups given restricted pasture (RES; n=18) and restricted pasture with MPG (250 ml twice daily; RESM; n=13) given from calving for 150 days. *Ad libitum* feeding resulted in greater BW, milk, protein and fat yield than other groups (P<0.05). Plasma concentration of insulin, IGF-I, growth hormone (GH), leptin, glucose and non-esterified fatty acids (NEFA) were measured weekly. Treatment effects on these constituents were minor, with low concentrations of insulin at week 3 (P<0.05) and greater NEFA concentrations in weeks 2-5 than in the RES heifers than other groups (P<0.05). Restricted feeding lowered leptin concentration relative to Adlib group in weeks 3, 5-7 and 12, and at week 6 when compared to the RESM group (P<0.05). During the metabolic challenge with MPG insulin secretion was stimulated. In the present study there was no difference in PPAI among the groups. We concluded that the most important impact on PPAI in heifers is BCS at calving. Restricted feeding and MPG supplementation of heifers calving in good BCS, affects milk production rather than PPAI. Supplementation with MPG can reduce PPAI of heifers with low BCS (2.8), but when BCS is 3.1 supplements are of little benefit to pasture fed heifers.

Key Words: Postpartum Anestrous, Monopropylene Glycol, Dairy Heifer

641 Hypothalamic genes expression in early- and late-maturing *Bos indicus* heifers. A. Vaiciunas*[1], L. L. Coutinho[2], and L. F. P. Silva[1], [1]*University of São Paulo*, *Pirassununga, SP, Brazil*, [2]*University of São Paulo*, *Piracicaba, SP, Brazil*.

The molecular mechanism by which leptin signaling in the hypothalamus might permit the initiation of puberty has not been elucidated. One possible mechanism for leptin molecular action on the reproductive axis is affecting NPY signaling. It was our objective to test whether early-maturing *Bos indicus* heifers have altered expression of hypothalamic genes related to leptin signaling. Among a population of 500 heifers between 20 and 25 months of age, 100 heifers were selected base on breed attributes (Nelore), month of birth, and body weight (280 to 300 kg). These 100 heifers were scored as prepubertal or pubertal according to the presence or not of a palpable corpus luteum (CL). Ten heifers without a CL and ten heifers with a palpable CL received a prostaglandin injection, and according to visual observation of heat and rectal palpation, 6 prepubertal and 6 pubertal heifers were selected for the experiment. These 12 heifers were slaughtered and samples of hypothalamus were collected and frozen in liquid nitrogen. Expression of Ob-Rb, SOCS-3, NPY, NPY-Y1 and NPY-Y4 was quantified by real-time PCR using the ribosomal protein RP-L19 as a reference gene. Hypothalamic expression of Ob-Rb, SOCS-3 and NPY was not different between groups of heifers. It was thought that late-maturing heifers could be resistant to leptin due to an increased expression of SOC-3, or a decreased expression of Ob-Rb at the hypothalamus, but our results did not corroborate with this hypothesis. There was a tendency for NPY-Y1 and NPY-Y4 expression to be reduced in

heifers that reached puberty earlier (P=0,10). Expression of NPY-Y1 was 8.3-folds lower and NPY-Y4 expression was 14.3-folds lower in early-maturing than in late-maturing heifers. When analyzed together, there was an 11-fold reduction in NPY receptors expression in early-maturing heifers, and this effect was statistically significant (P=0.03). These results suggest that, because of the lower expression of NPY receptors, the hypothalamus of early-maturing heifers could be less sensitive to NPY inhibition, and therefore reach puberty with lower levels of circulating leptin.

Key Words: Bovine, Neuropeptide-Y, Puberty

642 Evaluating reproductive and immune consequences of endocrine disrupting chemicals in an avian bioassay. M. A. Ottinger*[1], E. T. Lavoie[1], and M. J. Quinn[2], [1]*University of Maryland, College Park*, [2]*U.S. Army Center for Health Promotion and Preventive Medicine, Aberdeen, MD*.

The avian embryo provides a self contained system for studying the consequences of exposure to endocrine disrupting chemicals (EDCs). Therefore, the avian embryo is an appealing ′bioassay′ for detecting EDC effects in birds. Japanese quail were used to evaluate endocrine disruption by several EDCs, including estradiol, trenbolone, vinclozolin, methoxychlor, DDE, atrazine, and PCB 126. A number of endocrine, neuroendocrine and immune endpoints were evaluated for endocrine disruption in the Japanese quail embryo, juvenile/maturing, and adult, including serum or fecal estradiol and androgen concentrations, histology and morphology of the bursa and gonad, and hypothalamic aromatase activity, monoamine content and GnRH-I concentration. Maturation and male mating behavior were evaluated post-hatch to compare discernable EDC effects in hatchlings compared to long-term effects impacting reproductive function in adults. Results showed that all chemicals impaired male sexual behavior, regardless of androgenic or estrogenic mode of action. The GnRH-I system in hatchlings was sensitive to many of the EDCs, with up to a 50% decrease in content at some EDC doses. Similarly, hypothalamic dopamine levels were decreased by some EDCs in hatchlings; fewer effects were observed in adults. Gonadal steroids and thyroid gland hormones were decreased with EDC exposures. Immune response show long-term effects of EDC exposure, relative to bursal morphology and antibody responses. Our data provide supporting evidence that even low level exposure to selected EDCs exerts effects on the development of the reproductive axis, with additional impacts on other physiological systems, including the immune system. The embryo appears to be more vulnerable than the adult to sublethal and low level exposure; possibly due to compensation of physiological systems as the animal matures. Multiple endpoints appear to be necessary for detection of endocrine disruption due to differing modes of action and toxicity of EDCs. Supported by EPA R826134010 (Star Grant), NSF 9817024, and EPA R-2877801(MAO).

Key Words: Endocrine Disrupting Chemicals, Reproductive and Immune Systems, Japanese Quail

643 Differential expression of adiponectin, adiponectin receptor 1 (AdipoR1) and leptin mRNA in different adipose depots in sheep. A. Lemor*[1], M. Mielenz[1], M. Altmann[2], E. von Borell[2], and H. Sauerwein[1], [1]*University of Bonn, Germany*, [2]*Martin-Luther-University, Halle-Wittenberg, Germany*.

The adipokines adiponectin and leptin are important in lipid metabolism and glucose homeostasis. Adiponectin is inversely related to leptin secretion and is associated with insulin resistance. Data on the expression of adiponectin and its receptor AdipoR1 are mainly limited to monogastric species, whereas for ruminants corresponding data are scarce. We aimed to characterize mRNA expression for adiponectin, AdipoR1 and leptin mRNA in different sheep fat depots. Subcutaneous (sc) and visceral (vc) fat was dissected from carcasses of 10 male crossbred sheep (40 kg b.w.). Sc fat was sampled at 3 different sites: close to the sternum (S), withers (W) and the base of the tail (T); vc fat was sampled from perirenal (P) and mesenteric (M) depots. The mRNAs of adiponectin, AdipoR1 and leptin were quantified by real-time RT-PCR. For adiponectin, no differences were found between depots (p=0.36); whereas AdipoR1 mRNA concentrations were higher ($p<0.05$) in P compared in W, T and M. Leptin mRNA concentrations were higher in S than in both vc depots ($p<0.05$). Sc leptin mRNA values were correlated with the total sc fat fraction of the carcass ($p<0.05$, r=0.41) but not with vc fat. In contrast, vc adiponectin tended to be correlated with the vc carcass fat (p=0.1, r=0.40) but not with sc. A trend for a correlation was found between AdipoR1 with adiponectin (p=0.08, r=0.36 (sc), p=0.09; r=0.41 (vc)) in both adipose depots. In conclusion, our findings indicate a constant expression of adiponectin mRNA in different sheep fat depots, which is in contrast to reports from monogastric species. AdipoR1 as well as leptin mRNA seems to be differentially expressed in different adipose depots of sheep.

Key Words: Adiponectin, Adiponectin Receptor 1, Leptin

644 Prolactin levels and ovulation rate in crossbreed ewes with induced oestrus during the anoestrous season and the effect of bromocryptine and naloxone. V. O. Fuentes-Hernandez*[1], R. Orozco[1], J. J. Uribe[1], V. M. Sanches[2], and P. I. Fuentes[3], [1]*Universidad de Guadalajara*, [2]*FMVZ Universidad Michoacana de San Nicolas Hidalgo*, [3]*Hospital Pemex Sur de Alta Especialidad Mexico DF*.

With the objective of studying the effect on naloxone and bromocryptine on the ovulation rate and the plasmatic levels of Prolactin in the crossbreed ewe during the anoestrous season. 40 crossbreed ewes were allocated at random in to four groups of 10. All ewes received an intravaginal sponge with 40 mg Medroxiprogesterone acetate for a period of 14 days and on sponge withdrawal 250 I.U. of Ecg was administered by intramuscular injection. During the period of sponge treatment, ewes were medicated as follows: Group 1 (n = 10) was treated with 0.5 mg bromocryptine im at 12 hour intervals. Group 2 (n = 10) was treated as group 1 and an implant of 15 mg of naloxone was applied subcutaneously. Group 3 received a subcutaneous implant of 15 mg NaloxoneGroup and group 4 (n = 10) was sham treated. In group 1 it was observed that bromocryptine decreased significantly ($p<$ .001 Prolactin plasma levels, In group 2 bromocryptine + Naloxone treated ewes plasma Prolactin levels were decreased significantly ($p<.001$) in a similar pattern as observed in group 1. In group 3 prolactine levels were decreased significantly ($p<.05$). Control group 4 prolactine levels remained unchanged. After sponge withdrawasl and eCG injection oestrus was present in all groups. 6 to 8 days after estrus CL were observed by laparoscopy under ketalar xylazne anaesthesia, ovulation rate in bromocriptine and control groups was not significantly different (2.1 ± .4 and 2.2 ± .5 respectively). In bromocryptine

naloxone and naloxone treated ewes ovulation rate was significantly increased as compared with controls (2.9 ± .5 and 3.1 ± .4 Respectively p< .01). It was concluded that endogenous opioids are important modulators of ovulation.

Key Words: Ewe, Estrus, Naloxone

645 Luteinizing hormone-releasing hormone immunization alters pituitary hormone synthesis and storage in bulls and steers. K. J. Wells*[1], T. W. Geary[2], D. M. de Avila[1], J. de Avila[1], V. A. Conforti[1], H. Ulker[1], D. J. McLean[1], A. J. Roberts[2], and J. J. Reeves[1], [1]*Washington State University*, *Pullman*, [2]*USDA ARS Fort Keogh*, *Miles City, MT*.

Objectives of this study were (1) to determine if trenbolone acetate (TBA) co-administered with LHRH immunization would suppress reproductive function in beef bulls and (2) to examine the effects of LHRH immunization and TBA treatment on pituitary function. To address these objectives 44 Angus x Hereford bull calves (mean BW = 225 ± 2 kg; mean age = 187 ± 6 d) were randomized into eight treatments in a 2 × 2 × 2 factorial experiment, with castration, LHRH immunization, and TBA administration as treatment factors. Calves immunized against LHRH received a primary injection of ovalbumin-LHRH-7 fusion protein on d 0, followed by two booster injections on d 42 and 196. Calves treated with TBA were implanted on d 224. Mean LHRH antibody binding activity in serum increased after each booster for immunized calves, but was negligible in non-immunized animals throughout the experiment. Concentrations of testosterone in serum were lower ($P < 0.0001$) by d 84 and scrotal circumference smaller ($P < 0.05$) by d 168 in LHRH immunized bulls compared to non-immunized bulls. Treatment with TBA tended ($P = 0.07$) to decrease concentrations of testosterone in serum from bulls. Testes + epididymides weights at slaughter (d 272) were lighter ($P < 0.0001$) for immunized compared to non-immunized bulls. Both LHRH immunization and castration resulted in decreased anterior pituitary stores of LH and FSH ($P < 0.001$). Immunization against LHRH suppressed expression of the LH β, and common α-subunit genes ($P < 0.0001$), while castration increased expression of the same two genes ($P = 0.02$). Synthesis and storage of LH and FSH, as measured by pituitary LH and FSH content and expression of the LH β-subunit and common α-subunit genes, was suppressed by LHRH immunization.

Key Words: LHRH Immunization, Trenbolone Acetate, Pituitary

646 Glial cell line-derived neurotrophic factor enhances porcine oocyte developmental competence in vitro. K. Linher*[1], D. Wu[1,2], and J. Li[1], [1]*University of Guelph*, *Guelph, Ontario, Canada*, [2]*Sichuan Agricultural University*, *China*.

The success of early embryonic development depends on oocyte nuclear and cytoplasmic maturation. We have investigated whether glial cell line-derived neurotrophic factor (GDNF) affects the in vitro maturation (IVM) of porcine oocytes and their subsequent ability to sustain preimplantation embryo development. GDNF and both its co-receptors, GDNF family receptor α-1 (GFRα-1) and the rearranged during transformation (RET) receptor were expressed in oocytes and their surrounding cumulus cells derived from small and large follicles. When included in IVM medium, GDNF significantly enhanced cumulus cell expansion of both small ($P<0.05$) and large ($P<0.001$) cumulus-oocyte complexes. It also significantly increased the percentage of small follicle-derived oocytes maturing to the metaphase II (MII) stage ($P<0.01$), although the nuclear maturation of large oocytes was not significantly affected. Examination of cyclin B1 protein expression as a measure of cytoplasmic maturation revealed that in the presence of GDNF, cyclin B1 levels were significantly increased in large follicle-derived oocytes ($P<0.05$). Although not significant, cyclin B1 expression was also elevated in small follicle-derived oocytes to levels comparable to untreated large oocytes. After parthenogenetic activation, a significantly higher percentage of both small and large follicle-derived oocytes that were matured in the presence of GDNF developed to the blastocyst stage compared to untreated controls ($P<0.05$). Indeed, GDNF enhanced the blastocyst rate of small follicle-derived oocytes to levels comparable to those obtained for large oocytes matured without GDNF. Our study provides the first functional evidence that GDNF enhances oocyte maturation and preimplantation embryo developmental competence in a follicular stage-dependent manner. This finding may provide insights for improving the formulation of IVM culture systems, especially for small follicle-derived porcine oocytes.

Key Words: GDNF, Oocyte Maturation, Preimplantation Development

Ruminant Nutrition: Corn Milling Co-Products - Dairy

647 Maintaining milk components when feeding co-products of corn ethanol production. L. Armentano*, *University of Wisconsin*, *Madison*.

Use of DGS at 15 to 20% of ration DM may be economical, but requires caution to ensure optimal production of milk protein and fat. DGS can affect milk protein yield through altered diet carbohydrate and protein profile. Adequate dietary degradable protein should be supplied from other sources, and some attention to the lysine concentration in the remaining undegraded protein sources is warranted. However, recent research with modern DGS suggests that lysine content is less of a concern than previously thought. Starch content of the diet should be monitored for adequacy if DGS replaces grain. Laboratories may analyze DGS NDF with Na sulfite and NDFCP without sulfite. Using these values to calculate NFC as 100- ash - CP - NDF - Ether Extract + NDFCP, will overestimate NFC. Several characteristics of DGS may impact milk fat yield and composition. Excess oil may be present in diets with DGS. Different analytical methods (acid hydrolysis ether extract, ether extract variations) may not accurately measure fatty acids in DGS, and this content will vary. To the extent that DGS carbohydrate displaces starch, this will tend to have a positive effect on milk fat production, but care must be taken if DGS NDF replaces physically effective fiber from forages. Low forage fiber combined with high oil could trigger milk fat depression. If DGS is added to low oil diets, and diet oil is thereby increased, there may be less secretion of fatty acids shorter than 16 carbons, however, this may be compensated