

Growth and Development I

419 Growth, intake, and health of Holstein heifer calves fed an enhanced diet pre-weaning with or without exogenous estradiol immediately post-weaning. Adam J. Geiger^{*1}, Robert E. James¹, Catherine L. Parsons¹, Anthony V. Capuco², and Robert M. Akers¹, ¹Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA, ²United States Department of Agriculture-Agricultural Research Service, Beltsville, MD.

Enhanced feeding (HP) of heifers post-weaning but before puberty can reduce mammary gland (MG) development and milk yield. Better pre-weaning (PW) nutrition is likely beneficial. Our objectives were to determine effects of PW HP and exogenous estradiol (E₂) on growth, intake, and health. Thirty-six Holstein heifer calves were reared on: 1) a control milk replacer (MR) fed at 454 g powder/day (CON; 20% crude protein [CP], 20% fat), or 2) an accelerated MR fed at 1135 g powder/day (ACC; 28% CP, 25% fat). MR feeding was reduced 50% at wk 8. Starter was offered after wk 4 but balanced between treatments. BW and frame measures were taken weekly with intakes and health monitored daily. At weaning a subset of calves were killed (n = 6/diet). Remaining calves received E₂ implants and were killed at wk 10. The 4 treatments were: 1) CON, 2) CON + E₂ (EST), 3) ACC, and 4) ACC + E₂ (ACCEST). Data were analyzed using PROC GLIMMIX in SAS and treatment differences were separated with the PDFIFF function. Significance was declared when $P < 0.05$ and tendencies when $0.10 > P > 0.05$. ACC calves were heavier at weaning (75 vs 51 kg, $P < 0.01$). E₂ did not influence BW. ACC calves had greater average daily gain (ADG) between wk 1 to 7 ($P < 0.01$). Post-weaning ADG was similar between CON, ACC, and ACCEST calves, but greater in EST calves ($P < 0.05$) than ACC calves. At first slaughter, ACC calves had greater carcass, MG, thymus, liver, and spleen weights ($P < 0.01$). EST calves had heavier MG than CON calves ($P < 0.01$) and ACCEST calves heavier reproductive tracts and uteri compared with ACC calves ($P < 0.01$). ACC calves consumed more MR DM, CP, fat, and ash PW ($P < 0.01$). CON calves consumed more starter DM, CP, fat, NDF, ADF, and ash PW ($P < 0.01$). These differences were not observed post-weaning. Fecal scores (FS) were greater for ACC calves (1.7 vs 1.5; $P < 0.01$) PW. FS were lower for EST calves post-weaning. Results show that PW HP increases weight and frame measures pre and post-weaning. PW HP increased ADG PW, but not post-weaning. E₂ may elicit beneficial responses in some calves. Analysis of collected samples will allow us to determine cellular and molecular processes responsible for tissue differences.

Key Words: mammary gland, estradiol, milk replacer

420 Plane of nutrition affects Holstein bull calf growth, bone mineral density and organ size. Meghan E. MacGhee^{*}, Sarah R. McCoski, Camilla H. K. Hughes, Sally E. Johnson, and Alan D. Ealy, Virginia Polytechnic Institute and State University, Blacksburg, VA.

Early postnatal milk replacer diet affects ADG and feed efficiency and can have long-term consequences on calf growth, health and productivity. This study examined how plane of nutrition affects calf body weight, height, bone mineralization, and organ size. On postnatal d 1, Holstein bull calves (n = 4–5/diet/time point) were assigned randomly to a low plane of nutrition (LPN; 20% CP, 20% fat; 441 g DM/d) or high plane of nutrition (HPN; 27% CP, 10% fat; 882 g DM/d during wk 1 and 1131 g DM/d thereafter). Starter grain (25% CP, 4% fat) was offered at 1% BW beginning at wk 3. At 2, 4, or 8 wk of age, calves were euthanized and bone mineralization and body composition were

determined with Dual Energy x-ray Absorptiometry (DEXA). Visceral organs were removed and weighed, and empty carcass weight (ECW) was recorded. Data were analyzed by least squares ANOVA. Final BW and ADG were greater ($P < 0.05$) in calves fed HPN at each time point. Hip heights and bone mineral densities were greater ($P < 0.05$) for HPN calves than LPN calves at 8 wk but not at 2 or 4 wk. ECW-adjusted organ weights also were affected by diet. Thymus weight was greater ($P < 0.05$) in HPN calves than LPN calves at 4 and 8 wk (0.37% vs. 0.13% and 0.64% vs. 0.22%, respectively; SEM = 0.03). Spleen weight was greater ($P < 0.05$) in HPN calves than LPN calves at 8 wk (0.93% vs. 0.46%; SEM = 0.05). Also, kidney and liver weights were greater ($P < 0.05$) in HPN calves at 4 wk, and lung weights were reduced ($P < 0.05$) in HPN calves at 2 wk. Weights of the heart and pancreas did not differ based on plane of nutrition within each time point. Calf ECW was greater ($P < 0.01$) in the HPN group at each time point. In summary, deviations in growth rates, bone mineralization, and organ weights were evident between calves fed HPN and LPN diets. These outcomes are consistent with the contention that HPN feeding accelerates growth and improves health in young calves. The beneficial effects of HPN on bone mineral density and immune-centric organs provide new possibilities for understanding the effects of early nutrition on calf immune responses and productive lifespan of the cow.

Key Words: milk replacer, organ growth, bone density

421 Daily growth rate in Holstein Friesian heifers is affected by fasting insulin levels as newborn calves. M. Van Eetvelde^{*}, M. M. Kamal, H. Bogaert, and G. Opsomer, Department of Reproduction, Obstetrics and Herd Health, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium.

Body growth and reproduction traits were followed in 51 female Holstein Friesian calves at one herd in Flanders (Belgium). After an overnight fast, 3-d-old calves were weighed and blood samples were taken to determine fasting glucose (GLU_F) and insulin (INS_F) levels. All calves were kept indoors during the first year of life, and were fed according to their requirements for maintenance and growth. Every 3 mo until their first calving, body weight was recorded, as well as insemination dates and date of calving. Calves weighed 39.1 ± 4.26 kg at birth and were classified in 3 categories dependent on their daily growth rate during the first 6 mo of life: slow (<750 g/day; n = 6); moderate (750 to 950 g/day; n = 40) and fast growing (>950 g/day; n = 5). Analysis of variance was used to compare Glu_F and body weight between categories and the differences in Ins_F were assessed by the Kruskal-Wallis test. Glu_F was 6.0 ± 0.63 mmol/L without significant differences between the 3 groups ($P = 0.16$). Ins_F was 7.2 ± 4.76 mU/L in moderately growing calves. Slow growing calves had higher INS_F of 12.9 ± 9.10 mU/L in comparison to the fast growing ones (4.3 ± 2.39 mU/L, $P < 0.05$). Calves were inseminated at 14.9 mo. Heifers that were growing > 950 g/day weighed 472 ± 45 kg at first insemination, which was significantly more than the moderately and slow growing calves (respectively 414 ± 42 and 378 ± 29 kg, $P < 0.05$). The effect of early growth was still visible at first calving, as the fastest growing heifers weighed 734 ± 144 kg, which was significantly more than the heifers with a moderate growth rate (615 ± 72 kg, $P < 0.05$). In human medicine, low INS_F is seen in newborns that are small for gestational age, which is indicative for an increased insulin sensitivity to glucose and resembles conditions of prolonged fasting. Although in our research, no difference in birth weight was observed, the association of low insulin levels with subsequently high daily

growth rates resembles the human situation of catch up growth. Based on these preliminary results, we suggest that low fasting insulin levels in newborns could be the result of a suboptimal environment during gestation and could be a forecast of catch-up growth during early life.

Key Words: catch-up growth, insulin, Holstein Friesian heifers

422 Maternal obesity (MO) during ovine pregnancy leads to increased collagen content and cross-linking in the myocardium of adult F₁ but not F₂ offspring. Adel Bashir Ghnenis*, John F. Odhiambo, Richard J. McCormick, and Stephen P. Ford, *Department of Animal Science, University of Wyoming, Laramie, WY.*

There is accumulating evidence that MO increases offspring risk of obesity and cardiovascular disease in later life. Utilizing a well-established (J. Anim. Sci. 2010. 88:3546) ovine model of diet-induced MO from conception through gestation, we reported that gestational d 135 fetal heart left ventricle weight and collagen content were markedly increased by MO. Further, in a Langendorff system, the d 135 fetal heart from MO fetuses could not sustain high work levels (FASEB J. 2010. 24:2066). Here we examined the effect of MO on increasing collagen content and crosslinking in the adult F₁ and F₂ offspring myocardium. Ewes were assigned randomly as controls (CON) and fed at 100% of NRC recommendations or to an MO group fed 150% of NRC from 60 d before conception to term. Left ventricular free wall myocardial samples were collected from adult (22 mo old) male F₁ and F₂ offspring of CON and MO ewes at necropsy following a 12-week ad lib feeding challenge. Myocardial tissue was either snap frozen in liquid nitrogen for collagen analyses or fixed and paraffin embedded for histological evaluation. Collagen concentration was determined by colorimetry as hydroxyproline equivalents. Collagen crosslinking was also determined by measuring the amount of hydroxylslypyridinoline by immunoassay (Microvue, PYD EIA Kit, Quidel, San Diego, CA). Data were analyzed by mixed procedures of SAS. Myocardial collagen concentration was greater ($P < 0.05$) in MOF1 compared with CON F1 offspring (1.73 ± 0.10 vs. 1.42 ± 0.07 $\mu\text{g}/\text{mg}$, $n = 6$). The differences in OBF1 and CONF1 collagen concentration were confirmed by visualizing trichrome stained sections of myocardial tissue. Myocardial collagen crosslinking was 11% greater ($P < 0.05$) in OBF1 vs. CONF1 offspring. In contrast, myocardial collagen content and crosslinking did not differ between MOF2 and CONF2 offspring. These data provide evidence for programming of increased myocardial fibrosis in adult MOF1 offspring, which potentially results from MO-induced changes in fetal life and might impair cardiac muscle function in postnatal life.

Key Words: maternal obesity, myocardium, collagen

423 Growth and lactation during gestation decrease placental efficiency in cattle. M. Van Eetvelde*, M. M. Kamal, H. Bogaert, and G. Opsomer, *Department of Reproduction, Obstetrics and Herd Health, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium.*

In adult pregnancy, the gravid uterus receives a high priority status for nutrient partitioning. A shift in this hierarchy in favor of maternal tissues, as seen in adolescent pregnancy, compromises placental growth and fetal development. This situation occurs during gestation in growing heifers and lactating cows, as a high priority status of respectively maternal tissue growth and lactation is then observed. To assess the effect of dam lactation and growth on placental and fetal development, placentas of 49 multiparous (MP) Holstein Friesian (HF) cows and 43 growing heifers [27 HF and 16 Belgian Blue (BB)] were compared with afterbirths of

27 non-growing, nonlactating MPBB cows. Placentas were weighed and cotyledons were removed, after which their surface was calculated by the formula $\text{area}(\text{ellips}) = \pi ab$; with $a = \text{half of major diameter}$ and $b = \text{half of minor diameter}$. Placental efficiency (PE) was assessed by the calf weight:cotyledonary surface ratio. Analysis of variance was used to compare placental weight and cotyledonary surface, differences in cotyledon number and PE were assessed by the Kruskal-Wallis test. Placentas of MPBB dams weighed 5.9 ± 1.79 kg with a cotyledonary surface of 0.53 ± 0.110 m². Placentas of MPHf dams were 0.9 kg lighter ($P < 0.01$) but tended to have a larger total cotyledonary surface (0.58 ± 0.106 m², $P = 0.05$). Placentas of heifers were smaller (4.7 ± 1.34 kg and 0.48 ± 0.085 m², $P < 0.05$), without significant breed effect. The number of cotyledons in MPBB placentas (91 ± 27.2) was lower than in placentas of heifers and MPHf (respectively 114 ± 29.0 and 115 ± 28.2 , $P < 0.05$). In addition, MPHf dams had a greater proportion of small (<60 cm²) cotyledons; as mentioned in nutrient-restricted ewes. MPBB cows had the highest PE of 107 ± 25.1 kg/m², compared with 74 ± 13.5 kg/m² in MPHf dams ($P < 0.01$). BB and HF heifers had a similar PE of 96 ± 23.0 and 84 ± 18.0 kg/m² respectively. These preliminary findings suggest maternal growth and lactation during gestation to have a similar nutrient restrictive effect on placental development, which may result in a lower PE. This could be, at least partly, responsible for the differences in size of the offspring. Further research is necessary to assess the influence on organ development and future health of the calves.

Key Words: growth, lactation, placental development

424 Effects of GH and IGF-I on proliferation and apoptosis of bovine mammary epithelial cells. Hongrong Wang*, Yun Ji, Xueyan Pang, Qing Tian, Mengzhi Wang, and Lihuai Yu, *College of Animal Science and Technology, Yangzhou University, Yangzhou, Jiangsu Province, China.*

Growth hormone (GH) may exert its function through direct activation of the the growth hormone receptor (GHR) in mammary gland or through stimulating insulin-like growth factor-I (IGF-I). We hypothesized that the reduction of milk yield caused in part by apoptosis of mammary epithelial cells depends on the ratio of GH and IGF-I which could be influenced by nutrition, however, evidence to support this suggestion is lacking. Therefore, the purpose of this study was to investigate the effects of GH and IGF-I alone and in combination on proliferation and apoptosis of bovine mammary epithelial cells cultured in vitro. Mammary epithelial cells isolated from bovine mammary tissue were identified by cytomorphology, immunocytochemistry and specific gene expression. Subcultured and purified cells were treated with GH (100 ng/mL) and IGF-I (100 ng/mL) alone or a combination of GH and IGF-I (each 100 ng/mL) in growth medium without fetal calf serum for 24 h. The mRNA abundance was quantified by RT-qPCR, cell proliferation was determined with a CCK-8 kit, and apoptosis was identified by an FITC-Annexin V/PI kit. Statistical analysis was carried out by using ANOVA procedure of SAS 9.1 software with Duncan's multiple-range test. Cell proliferation was not enhanced by adding GH alone ($P > 0.05$) while it was promoted by IGF-I at about 6 h ($P < 0.05$). The combination of GH and IGF-I increased cell proliferation from 4 to 24 h ($P < 0.05$), which had no significant difference with IGF-I group ($P > 0.05$); The supplementation of GH or IGF-I individually had no significant effect on the early apoptosis rates of cells ($P > 0.05$), however, the late ($P < 0.01$) and total apoptosis rates ($P < 0.05$) were reduced. The late and total cell apoptosis rates were decreased by adding GH and IGF-I together ($P < 0.01$). In addition, adding GH or IGF-I alone tended to decreased the mRNA abundance of IGFBP-5 ($P < 0.1$) compared with the control, and the inhibitory effect was strongest when GH was combined with IGF-I

($P < 0.05$). This study demonstrated that GH and IGF-1 can regulate the number and activity of bovine mammary epithelial cell through promoting cell proliferation and inhibiting cell apoptosis to stimulate lactation.

Key Words: mammary epithelial cell, growth hormone, insulin-like growth factor-I

425 Effects of recombinant bovine somatotropin (bST) administration at breeding on the cow, conceptus, and subsequent offspring performance of beef cattle. Vitor R. G. Mercadante*, Francine M. Ciriaco, Darren D. Henry, Pedro L. P. Fontes, Danilo D. Demeterco, Pedro H. S. Pereira, Nicolas DiLorenzo, and G. Cliff Lamb, *North Florida Research and Education Center, University of Florida, Marianna, FL.*

To determine the effects of administration of a low dose of slow-release bST (Posilac, Elanco, Greenville, IN) on hormone concentration and conceptus development, a total of 190 suckled beef cows were exposed to the 7-d CO-Synch+CIDR fixed-time AI (TAI) protocol. Cows were blocked by days postpartum, BCS, breed and randomly assigned to receive one of the following treatments: (1) 2 injections of 325 mg bST, one at TAI and a second injection 14 d after TAI (D-bST, $n = 40$); (2) 1 injection of 325 mg bST at TAI and a placebo (saline) injection 14 d after TAI (TAI-bST, $n = 48$); (3) a placebo injection at TAI and one injection

of 325 mg bST 14 d later (14D-bST $n = 49$); and (4) 2 injections of placebo, one at TAI and a second injection 14 d after TAI (Ctrl, $n = 53$). Pregnancy was determined via transrectal ultrasonography 35 d after TAI and conceptus development was assessed by measuring crown to rump length (CRL) on d 35 and crown to nose length (CNL) on d 65 after TAI. Blood samples were collected on d 0, 7, 14, 21, 35 and 65 relative to TAI to determine concentrations of and IGF-1. Plasma concentrations of pregnancy-specific protein B (PSPB) were also assessed on d 35 and 65 after TAI. Individual calf birth weight and sex were determined at birth. Procedure GLIMMIX of SAS was used to analyze all data with the appropriate models. There were no differences ($P = 0.767$) among treatments on pregnancy to TAI ($48.7 \pm 0.5\%$). Administration of bST at TAI increased ($P < 0.01$) plasma concentration of IGF-1 on d 7, 14 and 21. However, CRL and CNL (12.11 ± 0.4 mm and 17.09 ± 0.2 mm, respectively) did not differ ($P = 0.231$) among treatments. Concentration of PSPB did not differ ($P = 0.18$) among treatments and between days ($P = 0.30$; 2.69 ± 0.11 ng/mL), and gestation length (282 ± 9 d) did not differ ($P = 0.49$) among treatments. In addition, calf birth weight was similar ($P = 0.52$) among treatments. We conclude that administration of 325 mg bST during the time of TAI to suckled beef cows enhanced concentrations of IGF-1, but failed to improve pregnancy rates, fetal size, PSPB concentrations, and had no effect on calf birth weight.

Key Words: bovine somatotropin, fetal programming, IGF-1