

Growth and Development: Early Development and Fetal Programming

635 Evaluation of the NCAPG I442M locus, a major gene for bovine prenatal growth, for effects on postnatal development compared to a disruptive mutation in the myostatin encoding gene GDF8. C. Kuehn*, P. Widmann, R. Pfuhl, and R. Weikard, *Leibniz Institute for Farm Animal Biology (FBN), Dummerstorf, Germany.*

Recently, we identified the I442M mutation in the bovine non-structural maintenance of chromosomes condensin I complex, subunit G (NCAPG) gene affecting bovine prenatal growth in an F2 resource population. Due to consistent application of embryo transfer during generation of the resource population, the direct genetic background of divergent postnatal growth dissected from putatively persistent maternal allelic effects could be addressed. In addition to the NCAPG I442M locus, also the disrupting mutation Q204X of the growth differentiation factor 8 (GDF8) segregated in our resource population. Thus, the effects of both loci could be evaluated on an identical genetic background. For our study, male calves were fed a milk replacer/hay/concentrate diet until d 121 followed by a semi ad libitum feed ration of concentrates and chaffed hay. Body weight was recorded monthly until 18 mo of age. All P0, F1 and F2 individuals were genotyped for NCAPG I442M and GDF8 Q204X. Association analyses were performed with a single locus and a 2 locus model fitting a fixed effect of the year of birth, an additive genetic SNP effect and an infinitesimal polygenic animal effect. The NCAPG I442M allele that had been associated with increased birth weight, showed a significant effect ($P = 0.0001$) increasing body weight at 18 mo of age. The locus explained 9.3% of the variance in the model. The effect of the loss-of-function allele GDF8 Q204X on body weight was also significant ($P = 0.006$). The difference between alleles amounted to 25.4 kg (std. err. 6.34 kg) for NCAPG I442M and 30.0 kg (std. err. 12.5 kg) for GDF8 Q204X, respectively. For the 2-locus model, effects of essentially the same magnitude were obtained. In conclusion, both, the NCAPG I442M locus and the GDF8 Q204X locus, exhibit significant, independent effects on postnatal growth.

Key Words: postnatal growth, cattle, NCAPG

636 Maternal nutrition differentially influenced gene expression responsible for fetal bovine adipocyte development. T. D. Jennings*, K. R. Underwood, A. E. Wertz-Lutz, and A. D. Weaver, *South Dakota State University, Brookings.*

Maternal nutrition during mid-late gestation influences adipose development in the fetus of various species, however bovine research is limited. The objective of this experiment was to determine the effects of maternal nutrition on the expression of genes in bovine fetal tissues. Genes of interest were selected because each has been demonstrated previously to influence body composition. Twenty-two Angus cross-bred heifers (BW = 527.73 ± 8.3 kg) were assigned randomly to 3 dietary treatments. Maternal dietary treatments were formulated and intake was controlled to provide 150% (HIGH; n = 7), 100% (INT; n = 7), and 80% (LOW; n = 8) of maintenance energy requirements for growing pregnant heifers. Heifers received dietary treatment from d 85 to d 180 of gestation, at which time fetuses were removed via cesarean section and muscle, subcutaneous fat, and liver samples were collected. At trial initiation, dam BW was similar among treatment groups. Final BW was lowest for the LOW dams ($P < 0.05$), however final BW for INT and HIGH were similar. Ribfat thickness increased in the HIGH treatment group compared with LOW and INT dams ($P < 0.05$). Thus, dam growth was influenced by diet during the treatment period, however dietary treatment did not influence fetal weight, crown rump length, or liver weight.

Preadipocyte factor-1 showed increased expression in fetal LM ($P < 0.05$) of HIGH fetuses as compared with INT and LOW. Peroxisome proliferator-activated receptor gamma and C/EBP α did not differ as a result of dietary treatment. However, LOW fetuses showed increased C/EBP β expression as compared with INT ($P < 0.05$). Collectively these results suggest that fetal growth characteristics are not affected by maternal nutritional manipulation during mid-gestation in beef cows. However, differences in expression of fetal genes regulating adipose tissue growth and development could lead to differences in composition of growth and warrants further investigation.

Key Words: adipose tissue, beef cattle, fetal programming

637 Lipid accumulation and fibrosis in skeletal muscle of offspring born to obese dams. X. Yan*, Y. Huang¹, M. J. Zhu¹, N. M. Long¹, A. B. Uthlaut¹, R. J. McCormick¹, S. P. Ford¹, P. W. Nathanielsz², and M. Du¹, ¹Department of Animal Science, University of Wyoming, Laramie, ²University of Texas Health Science Center, San Antonio.

Enhancing adipogenesis in muscle increases intramuscular adipocytes, while increasing fibrogenesis would affect meat tenderness. The objective was to evaluate the effects of maternal obesity on the intramuscular fat and collagen content of offspring muscle. Multiparous ewes (Rambouillet/Columbia cross) were fed a control diet (100% energy requirement, Con, n = 8) or an obesogenic diet (150% energy requirement, OB, n = 9) from 2-mo before pregnancy to parturition. Then, offspring lambs were fed commercial feeds to 22 mo old. The Longissimus dorsi (LD) muscle (2g) was biopsied at the left 12th rib for histochemical examination, mRNA and protein expression analyses and collagen content assessment. Mean ± standard errors of means are reported. No difference was observed in maternal body weight (68.3 ± 2.9 kg vs. 71.6 ± 3.2 in Con and OB) or body condition score (4.9 ± 0.4 in Con and 5.0 ± 0.3 in OB) before dietary treatments. Following 2 mo treatment (before mating), both maternal body weight (73.1 ± 4.0 vs. 108.8 ± 3.1 in Con and OB, $P < 0.05$) and maternal body condition score (4.9 ± 0.3 vs. 8.6 ± 0.2, $P < 0.05$) were higher on OB compared with Con diet. More intramuscular adipocytes were observed in OB offspring muscle compared with Con muscle; the mRNA expression of peroxisome proliferator-activated receptor (PPAR) γ , an adipocyte marker, was 33.6 ± 15.6% higher ($P < 0.05$) in OB, and the protein content was 51.1 ± 5.1% greater ($P < 0.05$), consistent with the 32.1 ± 9.8% higher intramuscular triglyceride content in OB compared with Con muscle ($P < 0.05$). The mRNA and protein contents of fatty acid transport protein 1 (FATP1) were increased in OB group by 61.8 ± 24.8% and 40.8 ± 9.3% ($P < 0.05$) respectively. We also detected 39.4 ± 8.8% higher mRNA expression for fatty acid translocase (FAT)/CD36 ($P < 0.05$). In addition, 50.6 ± 15.3% higher collagen content was detected in OB compared with Con offspring muscle ($P < 0.05$). In conclusion, maternal obesity increases intramuscular fat and collagen contents in offspring muscle, which results in changes of skeletal muscle composition and might affect the quality of resulting meat.

Key Words: maternal obesity, collagen, muscle

638 Enhanced transforming growth factor β (TGF- β) signaling and fibrogenesis in ovine fetal skeletal muscle of obese dams at late gestation. Y. Huang*, X. Yan¹, M. J. Zhu¹, R. J. McCormick¹, S. P. Ford¹, P. W. Nathanielsz², and M. Du¹, ¹Department of Animal Science, University of Wyoming, Laramie, ²University of Texas Health Science Center, San Antonio.

Maternal obesity is increasing at an alarming rate. The objective was to evaluate the effect of maternal obesity on fibrogenesis in fetal skeletal muscle at late gestation when ovine fetal muscle matures. Non-pregnant ewes were assigned to a control diet (Con, fed 100% of NRC nutrient recommendations, n = 6) or obesogenic diet (OB) fed 150% of NRC recommendations, n = 6) from 60 d before conception, and fetal semitendinosus (St) muscle was sampled at 135 d of gestation (term 148 d). The total concentration and the area of collagen in the cross-sections of muscle increased by $27.0 \pm 6.0\%$ ($P < 0.05$) and $105.1 \pm 5.9\%$ ($P = 0.05$) in OB compared with Con group. The expression of precursor TGF- β was $177.3 \pm 47.6\%$ higher ($P < 0.05$) in OB fetal muscle. The concentration of phospho-p38 was $74.7 \pm 23.6\%$ higher ($P < 0.05$) in OB group. An increase of $327.9 \pm 168.0\%$ ($P < 0.05$) and $188.9 \pm 82.1\%$ ($P < 0.05$) respectively for the mRNA expression of Smad7 and fibronectin was observed in OB compared with Con samples. In addition, enzymes involved in collagen synthesis, including lysyl oxidase, lysyl hydroxylase 2b and prolyl 4-hydroxylase $\alpha 1$ were increased by $350.2\% \pm 90.0\%$ ($P < 0.05$), $236.5 \pm 25.2\%$ ($P < 0.05$) and $82.0 \pm 36.2\%$ ($P = 0.05$) respectively in OB muscle. In conclusion, maternal obesity enhanced fibrogenesis in fetal muscle at late gestation which was associated with upregulation of TGF- β /p38 signaling pathway. Because muscle fibrosis is a hallmark of aging, enhanced fibrogenesis at such an early stage of development is expected to negatively affect the properties of offspring muscle.

Key Words: collagen, TGF, muscle

639 Up-regulation of nutrient transporters in the placenta of nutrient restricted pregnant ewes. Y. Ma*¹, M. J. Zhu¹, P. W. Nathanielsz², and S. P. Ford¹, ¹Center for the Study of Fetal Programming, Univ. of Wyoming, Laramie, ²Center for Pregnancy and Newborn Research, Univ. of Texas Health Sciences Center, San Antonio.

In sheep, maternal:fetal exchange occurs in placentomes, comprised of uterine caruncular and placental cotyledonary (COT) tissues. Glucose transporters (GLUT) and fatty acids transporters (FATP) in COT deliver glucose and long chain fatty acids (LCFA) to the fetal compartment. We have reported that fetuses from ewes fed to 50% NRC recommendations from 28 to 78 d of gestation (dGA; nutrient restricted, NR) weighed ~30% less than fetuses gestated by control ewes (C, 100% of NRC) at 78dGA. In contrast, NR fetuses exhibited a marked increase of LCFA storage in their lung, liver and muscle. When NR ewes were re-alimented to a C diet from 78dGA, their fetuses exhibited weights similar to C fetuses by 135dGA. COT tissue collected on 78 and 135 dGA was used to investigate GLUT and FATP systems via Realtime PCR and Western blot. Ewes assigned to C (n = 5) and NR (n = 6) groups were necropsied on 78dGA, while 6 C and 7 NR-realistmented ewes were fed the C diet from 78 to 135dGA before necropsy. At 78dGA, COT GLUT1 mRNA and protein levels were greater ($P < 0.05$) in NR than C ewes. Similarly, COT FATP4 mRNA and protein levels were greater ($P < 0.05$ and $P = 0.06$, respectively), and CD36 mRNA and Protein levels tended to be greater ($P = 0.06$) in NR versus C ewes. At 135dGA, COT FATP4 mRNA and protein levels tended to remain elevated ($P = 0.06$ and $P = 0.09$, respectively) in NR-realistmented versus C ewes. CD36 and GLUT3 protein levels also tended to remain elevated ($P = 0.08$ and $P < 0.05$, respectively) in NR-realistmented versus C ewes on 135dGA. The increased COT GLUT and FATP expression in NR versus C ewes at 78dGA is consistent with increased placental efforts to increase maternal nutrient transfer to the fetus. The continued elevation in COT GLUT3, FATP4 and CD36 expression after realimentation of NR ewes, would facilitate delivery of the increased blood levels of maternal nutrient

to the fetus, accelerating its growth, and possibly causing metabolic problems in postnatal life.

Key Words: maternal nutrient restriction, placental nutrient transport, sheep

640 Effect of grouping calves post-weaning according to pre-grouping feed intake on animal performance. C. M. Matuk*¹, M. Chahine¹, A. Bach^{2,3}, B. Ozer¹, M. E. de Haro Martí³, J. B. Glaze Jr.¹, T. Fife¹, and M. Nelson¹, ¹University of Idaho, Twin Falls, ²IRTA, Caldes de Montbui, Spain, ³ICREA, Barcelona, Spain, ⁴University of Idaho, Gooding.

The effect of grouping calves post-weaning according to pre-grouping feed intake on animal performance was evaluated using 752 replacement Holstein calves raised on a large operation in southern Idaho. In 4 different periods, individual feed intake was recorded 4 times a week during the last 3 wk that calves were individually hutched (56 d of age). Calves were classified as 'high eaters' (highest feeding level quartile) and 'low eaters' (lowest feeding level quartile). When leaving the individual hutches in each period, calves formed 6 groups: 20 animals randomly chosen without considering their level of feed intake (Control), 20 calves within the highest quartile of feed intake during the 3 wk prior leaving the hutches (HH), 20 within the lowest quartile (LL), 5 calves from the lowest and 15 from highest feeding level (LHH), 15 calves from the lowest and 5 from highest feeding level (LLH), and 10 calves from the highest and 10 from lowest feeding level (HL). Thus, out of 752 initially-tracked heifers, 480 heifers were chosen to form the 20 groups (6 groups per period) that were studied. Pen feed intake was recorded during the first 4 wk after grouping. After grouping, calves received a TMR composed of 95% starter and 5% alfalfa. Final weight was recorded at the end of the 12 wk of study. Pen was the experimental unit. Data were analyzed using a mixed-effects model with repeated measures accounting for the random effect of period and pen and the fixed effects of treatment, intake level class, time of measurements, and their 2-way interaction. Average DMI after grouping was greatest ($P < 0.05$) in HH (2.24 kg/d) and HHL (2.15 kg/d) followed by HL (2.07 kg/d), Control (2.06 kg/d), LLH (1.92 kg/d) and LL (1.77 kg/d). Similarly, ADG was greatest ($P < 0.05$) in HH (694 g/d) and HHL (658 g/d) than in HL (584 g/d), LLH (571 g/d), Control (546 g/d), and LL (531 g/d). The coefficient of variation of final BW (at 84 d of age) was lowest ($P < 0.05$) for HH (9.3%) and LL (11.7%), followed by Control (12.9%), LLH (15.8%), HHL (13.5%), and HL (17%). Grouping calves according to pre-weaning intake improves overall animal performance and diminishes variation.

Key Words: calves, heifers, intake

641 Evaluation of serum protein-based arrival formula and serum protein (Gammulin) on growth, morbidity, and mortality of stressed dairy calves. A. Pineda*¹, J. K. Drackley¹, and J. M. Campbell², ¹University of Illinois, Urbana, ²APC, Inc., Ankeny, IA.

Appropriate nutrition is a crucial factor to decrease morbidity and mortality of pre-weaning dairy calves. Several nutritional additives are available that may help to achieve this goal; however, their effectiveness is uncertain. The objective of this study was to evaluate a serum protein-based arrival formula (AF) and use of a commercial serum protein supplement (G; Gammulin, APC Inc.) in milk replacer for stressed (transport, cold) male calves on performance, morbidity, and mortality. Ninety-three male Holstein calves were stratified by arrival BW and plasma protein, and then randomly assigned to 1 of 4 treatment groups: Treatment A = AF, milk replacer without G (n = 22); Treatment B =

control electrolyte, milk replacer without G (n = 25); Treatment C = AF, milk replacer with G (n = 22); and Treatment D = control electrolyte, milk replacer with G (n = 24). At arrival, calves were fed either AF or a control electrolyte solution. At the next feeding, all calves received either a commercial calf milk replacer (20% CP, and 20% fat; no G supplementation) or the same milk replacer supplemented with G (50 g/d during the first 14 d only). Feed offered and refused was recorded daily. Calf health was assessed by daily assignment of scour and respiratory scores. Body weight, withers height, body length, heart girth, hip height, and hip width were measured weekly. Calves remained in the experiment until d 56. Data were analyzed using the MIXED procedure of SAS (v. 9.2). Results indicated that, during the first 2 wk of dietary treatment, calves fed AF had significantly greater heart girth ($P = 0.05$) and body length ($P = 0.03$), while G supplementation resulted in greater BW ($P = 0.05$). In addition, a significant ($P = 0.03$) interaction of G \times week was observed for ADG. Mortality was greater ($P = 0.007$) for calves that did not receive G. Addition of a serum protein product improved early growth and decreased mortality in transported male calves.

Key Words: calves, serum protein, Gammulin

642 The effect of maternal exercise on gestating gilts on neonatal piglet organ weight. E. K. Harris*, K. A. Vonnahme, J. D. Kirsch, J. D. Magolski, T. L. Neville, and E. P. Berg, *North Dakota State University, Fargo.*

To determine the effects of maternal exercise of gestating gilts on fetal piglet development and growth, Yorkshire gilts (n = 8), bred to a common boar, were placed in individual gestation stalls at d 30 of gestation. Treatments were assigned and initiated at d 40 of gestation. Exercise gilts (EX) were housed in individual gestation stalls but were individually exercised 3 times per week for 30 min until d 105 of gestation. Control gilts (CON) remained in gestation stalls for the duration of gestation. All farrowings were attended. Within 12 h of completion of farrowing, the lightest (LWT) and heaviest (HWT) male and female from each litter, excluding piglets <800 g, were selected for necropsy. Adrenal glands, brain, digesta, heart, kidneys, intestines, liver, lung, pancreas, spleen, stomach, gonads, semimembranosus (SM) and semitendinosus (ST) were dissected and weighed. Organ weights (g) and organ weight/live BW (g/g) were analyzed by PROC MIXED. Live weight of necropsied piglets was greater in offspring from EX gilts ($P = 0.03$; 1282.06 and 1698.70 \pm 125.40 g). Piglets from EX gilts had larger adrenal glands (g) ($P < 0.01$), kidneys (g) ($P < 0.01$), liver (g) ($P < 0.01$), and stomach (g) ($P < 0.01$) than CON gilts. Piglets from EX gilts had a tendency ($P < 0.10$) to have more digesta (g), heavier hearts (g), intestines (g), spleen (g), and SM (g/g). The brain and pancreas were the only organs not affected by treatment. Treatment by weight interaction occurred in liver (g/g) ($P = 0.05$), ovaries (g) ($P = 0.02$), and uterus (g/g) ($P < 0.01$). Light weight EX offspring had larger liver (g/g) ($P < 0.001$) compared with LWT CON offspring but were not different compared with HWT CON and EX treatments. LWT CON livers were also smaller than HWT EX ($P = 0.01$) but not different than HWT CON livers. Ovaries (g) were lightest in gilts from LWT CON gilts compared with all other treatment groups ($P < 0.05$). Light weight CON offspring had heavier uteri (g/g) compared with all other treatment groups ($P < 0.05$). Maternal activity during mid to late gestation influenced the developmental composition of the neonate.

Key Words: neonatal offspring, organ weights, pigs

643 Changes in gene expression during pituitary morphogenesis and organogenesis in embryonic chicks. M. Proszkowiec-Weglarz*,

S. E. Higgins, and T. E. Porter, *University of Maryland, Department of Animal and Avian Sciences, College Park.*

The anterior pituitary gland (AP) plays an important role in the regulation of many physiological processes in vertebrates. Formation of Rathke's pouch (RP), the precursor of the AP, involves invagination of the oral ectoderm (OE) and a multi-step process regulated by cell interactions, signaling pathways and transcription factors. In contrast to mammals, the molecular mechanisms underlying development of the AP in birds are poorly understood. Thus, the aim of this study was to evaluate tissue-specific gene expression patterns in the developing chicken AP. OE/RP and the adjacent neuroectoderm (NE) were collected by laser capture microdissection from 60 paraffin-embedded chicken embryos at 12-h intervals from embryonic day (e) 2.5 to e7 (n = 3 replicates). After RNA isolation and amplification, quantitative real-time PCR was performed to determine RP- and NE-specific gene expression. RP was formed by invagination of OE around e2.5, and by e6-e6.5 RP lost its connection with the oral cavity and proliferated to form the AP. Among genes involved in early pituitary organogenesis, *Pitx1*, *Pitx2* and *Hesx1* showed high expression at e2.5 in RP that decreased during development ($P < 0.05$). Expression of pituitary cell lineage specification and differentiation genes (*Pit1* and *Tbx19*) increased gradually, reaching the highest level by e7 and e6.5 in RP, respectively ($P < 0.05$). *Alpha-GSU*, encoding a common glycoprotein subunit of gonadotropins and thyrotropin, showed increasing mRNA expression from e4 ($P < 0.05$). *BMP4*, *BMP2*, *Wnt5a*, *Isl1* and *Noggin* were expressed in both RP and NE, while *Nkx2.1* showed NE-specific expression during AP formation. Taken together, we present a gene expression profile of the developing AP in the chicken. Our results will be helpful in better understanding the functional development of this gland, which is critical for controlling animal growth, reproduction, metabolism, and stress responses.

Key Words: Rathke's pouch and neuroectoderm, pituitary development, chicken embryo

644 Effects of in ovo feeding of carbohydrates and arginine on the energy metabolism, protein status and perinatal growth in Pekin ducks. M. Tangara*, W. Chen, F. R. Huang, and P. Jian, *Laboratory of Animal Molecular Nutrition, Department of Animal Nutrition and Feed Science, Huazhong Agricultural University, Wuhan, Hubei, P. R. China.*

The objective of the present study was to determine the effects of in ovo feeding of exogenous nutrients on the glycogen reserves, protein status and early growth of Pekin ducks. To this end, based on randomized completely block design, 750 fertile eggs were divided into following 5 groups of 150 eggs: 1) Uninjected; 2) 0.35% sodium chloride (NaCl); 3) 2.5% sucrose + 3% maltose (CHO); 4) 0.22% arginine (Arg); and 5) 2.5% sucrose + 3% maltose + 0.22% arginine (CHO+Arg). At 23 d of incubation, 1.2 mL of each solution was injected into amniotic fluid of each group using a 22-gauge needle. Ten eggs/ducklings per treatment were sampled at 25 d of incubation, hatch, 3 and 7 d of age to determine liver and muscle glycogen, glucose-6-phosphatase (G6P) activity and different protein expression including S6K1 (S6 kinase1), phosphorylated S6K1 and phosphorylated adenosine monophosphate-activated protein kinase (AMPK) using iodine reduction test, colorimetric oxidase test and Western blot, respectively. Maximal hatchability was found in the group ($P < 0.05$) fed with CHO+Arg (94%) followed in order by Arg (90%), CHO (89%), uninjected control (85%) and NaCl (80%). All the ovo fed ducklings improved BW at hatch, 3, 7, 14, 21, 28 and 35 d of age related to uninjected ($P < 0.05$). Arg and CHO+Arg had significantly enhanced the liver glycogen by 188% and 249%, respectively at hatch ($P < 0.01$) compared with that of uninjected group.

CHO and CHO+Arg significantly increased muscle glycogen level ($P < 0.01$) by 22% and 42%, respectively at 25 d of incubation over the uninjected group. CHO and Arg had significantly decreased ($P < 0.01$) G6P by 41% and 30%, respectively at 25 d of incubation, whereas NaCl and CHO+Arg increased G6P by 30% and 20%, respectively at hatch in comparison with the uninjected group ($P < 0.01$). At 25 d of incubation, hatch, 3 and 7d posthatch, greater values of S6K1 and S6 phosphorylation were observed in duck embryos and neonates fed with Arg and CHO+Arg. The activation of AMPK was also detected in the group fed with Arg and CHO+Arg. The present results indicated that in ovo feeding CHO and Arg may improve glycogen storage and muscle protein deposition in ducks

Key Words: in ovo feeding, energy, protein, metabolism, growth, ducks

645 The effect of induced moisture loss on embryonic development of pekin ducks. C. Noonan* and M. S. Lilburn, *Ohio State University/OARDC, Wooster.*

Moisture loss in commercial Pekin duck eggs during incubation is often variable and accelerated moisture loss may contribute to excessive hatchling dehydration and adversely affect later developing systems such as the intestine. Two experiments were conducted to determine the effect of induced moisture loss on embryonic development. In both experiments, commercial duck eggs were individually weighed at set. At 12 d, all eggs were reweighed and randomly allocated to one of 4 treatments consisting of 0, 1, 2, or 3 holes (Experiment 1; $n = 40$ per treatment) or 0, 1 or 2 holes (Experiment 2; $n = 120$ per treatment). The holes (<1 mm) were drilled above the air sac. All eggs were reweighed at approximately 2-d intervals. All data was analyzed by ANOVA using the PROC Mixed program (SAS Inc.) and least squares means were separated using LSD. In Experiment 1, there were no differences in initial egg wt or moisture loss (4.67%; $P > 0.05$) at 12 d. On D 14 and all sample days thereafter, there were incremental increases in moisture loss with each additional hole ($P < 0.01$). At D 19, the range was 7.35% (0-hole) to 20.86% (3-hole) and the experiment was terminated. In Experiment 2, the range was from 5.32% (0-hole) to 7.34% (2-hole) on D 14 ($P < 0.01$) and 8.03% (0-hole) to 15.15% (2-hole) on D 19. On Day 20, a sample of embryos from each treatment ($n = 25$) was broken out for embryo weight determination. Wet embryo weight was heavier ($P < 0.01$) in the 1-hole treatment (36.7 g) compared with the 0-hole (33.9 g) and 2-hole (31.8 g) treatments. The same pattern was observed for dry embryo weight. There was a progressive decrease in hatch weight ($P < 0.05$) as moisture loss increased (0-hole, 55.3 g; 1-hole, 53.6 g; 2-hole, 52.3 g) but no effects on intestinal measures (villus height; crypt depth). In conclusion, major differences in moisture loss during incubation in Pekin duck embryos have only a small effect on body weight or physiological status at hatch.

Key Words: Pekin, embryo, incubation

646 Bone development of three breed crosses of broilers is affected by incubation profiles. E. O. Oviedo-Rondón*, M. J. Wineland, C. M. Ashwell, and P. R. Ferket, *North Carolina State University, Raleigh.*

Genetics, maternal nutrition, and incubation conditions affect bone development in broilers. One experiment was conducted to observe epigenetic effects of 2 incubation temperature profiles on bone development in 3 groups of broilers (A, B, C) differing in genetic background and maternal nutrition. Group A was the final cross of this strain; and B and C were the progeny of the male line either feed restricted or with no feed restriction. The first incubation profile followed standard (SS)

temperatures to maintain eggshell temperature (EST) at 37.5°C. The second profile (LH) was similar to what one observes when eggs are in a multi-stage incubation system, the first 7 d EST was maintained at 36.5°C EST, during the second 7 d at 37.5°C EST and the remaining days at 39°C EST. At hatch, a random sample of 8 chicks per treatment were collected, weighed, sacrificed, and residual yolk determined. Both legs were dissected and shank and femur weights, lengths, and thickness were obtained. Relative asymmetry and weight relative to BW without yolk (%) of each leg section were calculated, and bone density estimated as g/mm. A total of 288 chicks were randomly placed in 48 battery cages and raised until 21 d. Bones were collected again at 14 and 21 d and similar parameters evaluated. Data were analyzed as a 2x3 factorial design considering incubation profiles and genetic strain as main factors. No consistent interactions were observed. Chickens from C group were heavier than B chickens only at hatch. The B group had the shortest bones and the lowest femur density. The C group was heavier at 14 d, but no other effects of epigenetics were observed. The LH profile reduced yolk absorption, and weight, length, width and density of all bones at hatch. Similar effects of incubation were observed at 14 and 21 d of age. The LH profile caused shorter bones, with lower density, and higher asymmetry independently of the genetic group. These results indicated that incubation profiles may have more importance on bone development than the genetic background, and the epigenetic effects of parental feed restriction were only observed at hatch.

Key Words: bone development, incubation, epigenetics

647 Effect of in ovo selenium injection on chick embryo viability and tissue selenium levels. L. M. Macalintal*, A. H. Cantor, A. J. Pescatore, M. J. Ford, H. D. Gillespie, J. L. Pierce, K. A. Dawson, and R. F. Power, *Alltech-University of Kentucky Nutrition Research Alliance, Lexington.*

The effect of injecting graded levels of selenium (Se) as selenomethionine (SeMet) or sodium selenite (Na_2SeO_3) into the yolk of incubating eggs on embryo viability and liver Se levels was studied. Fertile eggs were obtained from white shell laying hens (Hy-Line W-36) that were fed a low Se corn-soybean meal diet. On Day 10 of incubation, eggs were candled to ensure embryo viability. The shell surface was disinfected with alcohol and a small hole was drilled over the air cell. The yolk of each of 30 eggs per treatment was then injected with 0.1 mL of a phosphate buffered saline solution providing 0, 2.5, 5, 10 or 20 μg Se as either SeMet or Na_2SeO_3 . In a control group of eggs holes were drilled in the shell, but no injection was administered. The holes were sealed with glue and eggs were returned to the incubator. On Day 20 of incubation, eggs were candled to determine viability. Viable embryos were then killed to obtain tissue samples. Liver samples were analyzed for Se using fluorometric analysis following digestion in nitric and perchloric acids. Embryo viability values for the non-injected eggs and eggs injected with buffer without Se were 100% and 94%, respectively. Viability values for eggs injected with 2.5, 5, 10 and 20 μg Se as SeMet were 97, 94, 90 and 83%, respectively, while the respective values for eggs treated with Na_2SeO_3 were 87, 94, 74 and 87%. Injecting graded doses of Se resulted in linear increases ($P < 0.001$) in liver Se. However, the regression coefficient for Na_2SeO_3 was greater than that for SeMet (0.059 vs. 0.014). The results indicate that in ovo injection of Se as SeMet or Na_2SeO_3 at levels up to 20 μg does not have a detrimental effect on embryo viability. The effects of the SeMet and Na_2SeO_3 on liver Se concentrations suggests that the compounds are metabolized differently by the chick embryo.

Key Words: in ovo injection, selenium, embryo viability