

559 Effects of EcoCare® Feed on mineral excretion of pigs during the finishing phase. T. Walraven*, S. Carter¹, M. Lachmann¹, J. Bundy¹, J. Jarrett¹, and B. De Rodas², ¹Oklahoma State University, Stillwater, ²Land O'Lakes Purina Feed, Gray Summit, MO.

We have previously reported that EcoCare® Feed reduced N and P excretion of finishing pigs without affecting growth performance or carcass traits. To determine mineral excretion, eighty crossbred (D x (L x Y)) pigs (30 kg BW) were blocked by BW and sex, and randomly allotted to 1 of 2 dietary treatments. Pigs were housed in an environmentally-controlled building with 4 identical rooms (20 pigs/room, 2 rooms/trt). Each room contained a shallow pit, pull plug system. A fortified corn-soybean meal-based diet served as the control. The test diet (EcoCare®, EC) was similar to the control diet except that CP and available P were reduced (~2.6% units for CP; ~0.11% unit for avail. P) with additions of Lys, Thr, Met, EC Pak (containing phytase) and EC premix. To maintain similar Ca:P ratio, Ca content of the EC diet was reduced. The EC premix provided approximately 20% less Fe, Zn, Cu, and Mn than the mineral premix used in the control diet. All trace minerals were included in an inorganic form with the exception of Se. Pigs and feeders were weighed at each phase change, and pit volume was measured. Feed and pit samples were collected for mineral analysis. On d 0 and 122, 6 and 24 pigs (6/room), respectively, were ground to estimate initial and final body composition. There was no difference ($P > 0.10$) in initial or final ash or mineral concentration; thus, accretion was similar ($P > 0.10$) for pigs fed the control or EC diet. The intake of minerals was reduced ($P < 0.05$) for pigs fed EC. The excretion (g/d) of Ca (7.44 vs. 4.51), P (8.2 vs. 5.7), K (18.4 vs. 14.6), Mg (2.91 vs. 2.55), S (2.12 vs. 1.79), Fe (0.433 vs. 0.343), Zn (0.247 vs. 0.205), Cu (0.026 vs. 0.020), and Mn (0.080 vs. 0.064) was reduced ($P < 0.05$) for pigs fed EC compared to those fed the control. In general, the excretion of minerals as a % of intake was reduced ($P < 0.05$) for pigs fed EC. These results suggest that EcoCare® Feed reduced mineral excretion without affecting mineral accretion or growth performance.

Key Words: pigs, mineral, excretion

560 Effects of combining multiple dietary manipulations on growth performance and nutrient excretion of finishing pigs. T. Walraven*, S. Carter, J. Jarrett, M. Bible, and H. J. Kim, Oklahoma State University, Stillwater.

Eighty-eight crossbred (D x (L x Y)) pigs (32 to 114 kg BW) were used to evaluate the effects of reducing dietary CP, Ca, and P with the additions of phytase and organic trace minerals on nutrient excretion during a 94-d finishing period. Pigs were stratified by sex, weight, ancestry and randomly allotted to 1 of 2 dietary treatments. Pigs were housed in an environmentally-controlled building with 4 identical rooms. Each room contained a shallow pit, pull plug system (22 pigs/room, 2 rooms/trt). Dietary treatments were fed in 4 dietary phases and consisted of a fortified corn-soybean meal diet and a Reduced Excretion (REx) diet. The REx diet had a 3% units decrease in CP with Lys, Thr, Met, and Trp added as needed and a reduction in available phosphorus of 0.10% with phytase inclusion. Also, in the REx diet, monocalcium P replaced dicalcium P and CaCl replaced 50% of the CaCO₃ in the control diet. Furthermore, organic sources of Fe, Zn, and Cu replaced inorganic sources of these minerals in the control diet. All diets within phase were formulated on a SID lysine basis (0.92, 0.79, 0.65, and 0.56% for Phases 1 to 4). Feed and slurry samples were collected weekly along with pig weights, feed intake, pit volume, and pH of slurry. Diet did not affect ($P < 0.10$) ADG (822 vs. 839 g), ADFI (2.27 vs. 2.28 kg), or G:F (0.365 vs. 0.367). Daily DM intakes were similar ($P > 0.10$), but N (57.8 vs. 47.4 g/d) and P (11.1 vs. 8.6 g/d) intakes were reduced ($P < 0.05$) for pigs fed REx. Also, slurry pH for pigs fed REx was reduced (7.4 vs. 6.8; $P < 0.05$). Slurry DM concentration was similar ($P > 0.05$), but slurry N (15.1 vs. 11.8%) and P (2.84 vs. 1.98%) tended to be reduced ($P < 0.10$) with REx. The daily excretion of DM, N (33.9 vs. 24.4 g) and P (6.8 vs. 4.5 g) for pigs fed REx was reduced ($P < 0.05$) compared to those fed the control. These results suggest that combining dietary manipulations is an effective method of reducing N and P excretion. *This project was supported by the National Pork Board.*

Key Words: pigs, diet, excretion

Physiology and Endocrinology: Impact of Gonadal Steroids on Brain Development and Function

561 Feedback and fitness: Consequences of non-classical estrogen receptor α signaling in the brain. J. E. Levine*, Northwestern University, Evanston, IL.

Ovarian estrogens exert critically important actions in hypothalamic neurons to regulate ovulatory cyclicality, reproductive behaviors, and energy homeostasis. Estrogen receptor alpha (ER α) appears to mediate most of these effects, as disruption of ER signaling leads to infertility and metabolic syndrome. ER signaling mechanisms may include "classical genotropic" effects mediated by direct binding of receptor dimers to DNA, "non-classical genotropic" effects involving tethering of ERs to other transcription factors, and "non-classical non-genotropic" actions mediated by cytoplasmic ERs coupled to membrane-initiated signal transduction pathways. Our studies make use of novel ER mutant mouse models to ascertain the cellular mechanisms by which ER mediates E2 effects on these physiological and behavioral processes. We have utilized a novel mutant ER knock-in mouse model, which confers non-classical genotropic and non-genotropic signaling in the absence of classical signaling, to determine that non-classical ER signaling can convey E2 effects integral to homeostatic feedback control of reproductive hormone secretions, as well as E2 actions governing paracopulatory behavior and body weight regulation.

Key Words: estrogen, body weight, reproduction

562 Nongenomic actions of estrogens directly on the ovine pituitary facilitates LH secretion. T. Nett*, A. Arevalo-Arreguin¹, and T. Davis², ¹Colorado State University, Fort Collins, ²University of Idaho, Moscow.

In addition to the well known genomic actions of steroid hormones, there are nongenomic effects of these hormones that are only now being recognized. Our laboratory has investigated the nongenomic effects of estradiol (E) on secretion of LH in sheep. For these studies, we have employed E conjugated to either BSA (E-BSA) or to a novel 15 amino acid peptide (E-PEP), neither of which appear to induce classic genomic effects attributed to estradiol. Both compounds (as well as E) inhibited ($P < 0.01$) GnRH-induced secretion of LH from cultured ovine pituitary cells in a dose dependent manner. Each of the estrogens essentially abolished the LH secretion induced by 2 nM GnRH when present in the culture media at 20 nM or greater. Based on these data, ovariectomized (OVX) ewes were administered E or E-BSA and concentrations of LH were monitored for the next 24 hr. Both E and E-BSA resulted in a rapid decrease (< 20 min, $P < 0.01$) in serum concentrations of LH. Interestingly, only E decreased secretion of FSH. Likewise, E, but not E-BSA stimulated an ovulatory-like surge of LH beginning approximately 12 hr after treatment. Although there was no ovulatory-like surge of LH in ewes treated with E-BSA, there was a slight increase in LH at the

time an ovulatory-like LH surge would have been expected. These data indicate that acute inhibition of GnRH-induced LH secretion is mediated by a nongenomic action of estradiol directly on gonadotropes. *Supported by NRI grant No. 2005-35203-15376 from the USDA-CSREES.*

Key Words: estrogen receptors, nongenomic, luteinizing hormone

563 Actions of androgens in regulating sexual differentiation of the sheep brain and consequent effects on sexual behavior. C. E. Roselli*^{1,2} and F. Stormshak², ¹*Oregon Health and Science University, Portland*, ²*Oregon State University, Corvallis*.

During a critical period in early life, the male-typical mammalian brain develops via direct actions of testicular testosterone, while the female-typical brain develops in the absence of significant exposure to testosterone. Many sex-specific behaviors and neuroendocrine functions that will later be expressed in adults are programmed during this time. In our laboratory the domestic ram is used to study the early programming of the neural mechanisms underlying same sex partner preference. As many as 8% of domestic rams are sexually attracted to other rams (male-oriented) in contrast to the majority of rams that are

attracted to estrous ewes (female-oriented). Male-oriented rams thus express a female-typical sexual attraction that may have arisen from atypically low exposure to testosterone during early fetal life. To test this hypothesis we compared the structure of the preoptic area/anterior hypothalamus among female-oriented rams, male-oriented rams and ewes. We found that sexual partner preferences correlate with the volume of the ovine sexual dimorphic nucleus (oSDN) and the number of aromatase-expressing neurons it contains. We also studied the ontogeny of the oSDN and found that it develops prenatally under the influence of testosterone arguing that the circuitry for sexual attraction is assembled prior to birth. The high level of aromatase mRNA in neurons of the oSDN raised the question of whether local aromatization of testosterone to estradiol is required to masculinize brain structure and sexual attraction. Prenatal treatment with an aromatase inhibitor did not alter the expression of male-typical behaviors, including sexual preference for females or neuroendocrine responses; nor did it reduce the size of oSDN in genetic males. These results argue that male-typical sexual differentiation of the sheep brain does not depend on locally synthesized estradiol, but rather is programmed through androgen receptor mechanisms. *Supported by NIH grant R01 RR014270.*

Key Words: sexual partner preference, sexual differentiation, sexually dimorphic nucleus

Production, Management and the Environment: General

564 Heat stress does not alter immune status of Holstein calves but slick genotype confers reduced immune function. J. W. Bubolz*, S. Tao, B. C. do Amaral, M. J. Hayen, T. A. Olson, and G. E. Dahl, *University of Florida, Gainesville*.

Environmental factors such as photoperiod and heat stress influence health and hormone secretion in dairy cattle. Genetic background may also modulate immune status in cattle. The objective of the present study was to test the hypothesis that heat stress depressed immune function in cattle. A second objective was to examine the affect of the *Slick hair* gene on immune status. Calves defined as slick-haired possess a dominant gene of Senepol origin that when expressed produces a very short, sleek coat. Slick (n=4) and wild-type (n=4) calves were kept in controlled-temperature chambers for a period of 9 weeks. Calves were exposed to heat stress and neutral thermal conditions with a 1 week pre-treatment acclimation and 2 week recovery period between temperature treatments in a 2x2 cross-over design. Dry matter intake (DMI), water intake and infrared (IR) skin temperature were measured daily. Jugular blood samples were collected weekly and evaluated for lymphocyte proliferation, neutrophil phagocytosis and neutrophil oxidative burst activity. Relative to thermoneutral conditions, heat stress increased AM (35.0 vs. 30.6°C; P < 0.001) and PM skin temperatures (36.8 vs. 31.6°C; P < 0.001). Calves under heat stress increased daily water consumption (29.2 vs. 17.8 L; P < 0.04) and decreased DMI as percentage of body weight (2.29 vs. 3.83%; P < 0.001) compared with the thermoneutral period. Relative to thermoneutral treatment, no difference in any immune variable was observed during heat stress. However, neutrophils from wild type calves had greater phagocytic (P < 0.01) and oxidative burst (P < 0.07) activity compared with slick-haired calves. In addition, lymphocytes from wild type calves had greater proliferation relative to slick calves (P < 0.05). Results indicate that wild type calves had improved immune status compared to slick-haired calves regardless of environmental temperatures. Immune status of slick-haired calves was depressed relative to wild type, but heat stress did not influence immune status of Holstein calves in controlled-temperature chambers.

Key Words: slick-haired calves, heat stress, immune status

565 Clinical stopping rules in sequential field trials. D. B. Nielsen* and C. Enevoldsen, *Faculty of Life Sciences, Department of Large Animal Clinical Sciences, University of Copenhagen, Denmark*.

Trials performed by field veterinarians can be integrated in health programs in large dairy herds to enhance treatment efficiency. The objectives of this study were to identify the requirements to trial designs if they were to be accepted and performed non-subsidized by veterinarians in commercial herds and, by means of simulation, to illustrate a potentially useful design. Semi-structured research interviews of 12 veterinarians working within a similar type of health program were conducted. The veterinarians elaborated on their experience, present methods and future needs for evaluation of efficiency of metritis treatments (as a general disease model). Qualitative analysis was performed to identify coherent meanings across interviews. We performed a simulation of a sequential design (double triangular test with 'Christmas tree correction') with the PEST 4 program[®]. This design can potentially reduce the sample size by early stopping based on interim analysis of an important parameter and adjustable significance levels. The interviews showed that all the interviewed veterinarians preferred to evaluate efficiency by means of production parameters like milk production and fertility. However, the majority of the veterinarians emphasized that clinical cure (e.g., no need for follow-up treatment) imposed a constraint. They expressed unwillingness to perform trials without the possibility to apply early stopping in case of clinically significant adverse effects. The simulation (10,000 runs, alpha 0.05, power 0.8) showed that if an existing difference in probability of follow-up treatment was 0.3 in favor of the conventional treatment, a sample size of 54 cows (SD 19) would result in early stopping of the trial (fixed sample size of 72 cows). The simulation indicated 79% probability of showing inferiority of the experimental treatment and 21% probability of showing no difference between treatments. We conclude that implementation of scientifically acceptable production-oriented field trials could be accepted by field veterinarians as integrated parts of a health program if the designs include possibilities for early stopping based on valid statistical evaluations of clinical cure.

Key Words: field trial, stopping rules