270P Comparing the use of bird-level versus room illumination on broiler production, fear, and stress. B. Rocha* and G. S. Archer, Texas A&M University, College Station, TX.

Lighting can greatly alter broiler production and behavior. Currently, more energy-efficient means of illuminating broiler barns are being sought. Using LED light bulbs that are easily retrofit into existing electrical sockets are starting to become an alternative to incandescent bulbs. However, they still do not maximize the energy savings capacity of this technology as the bulbs must light the barn from the ceiling down. This project investigated using LED lighting attached to the feeders and waterers to light only the bird level. The treatments (n = 120 birds per treatment) were incandescent lighting (Incan), LED lighting (LEDhi) and LED lighting at bird level (LEDlo). All lighting was 23L:1D at 20 lx for 14 d and then was changed to 20L:4D at 5 lx for the remaining 31 d. Fearfulness was determined using several fear tests and stress susceptibility was assessed using a composite asymmetry score determined by middle toe length and metatarsal length and width. The Incan (2.73 ± 0.08 kg) birds weighed less (P < 0.05) after 45 d than both the LEDhi (2.95 ± 0.06 kg) and LEDlo birds (3.02 ± 0.05 kg). The LEDlo (1.61 ± 0.02) birds had better (P < 0.05) FCR than the Incan (1.71 ± 0.03) and LEDhi birds (1.67 ± 0.01). The LEDlo birds were less fearful (20.6 ± 9.15 s; 13.3 ± 2.1 s; P < 0.05) than the other birds in the emergence (Incan, 160.7 ± 6.7 s; LEDhi, 148.9 ± 7.4 s) and isolation tests (Incan, 27.7 ± 3.7 s; LEDhi, 23.2 ± 3.7 s). Both LED groups were less fearful (P = 0.05) during tonic immobility (LEDlo, 230.5 ± 27.6 s; LEDhi, 225.3 ± 25.9 s) and had lower composite asymmetry (LEDlo, 13.32 ± 0.15 mm; LEDhi, 1.68 ± 0.15 mm), when compared with the Incan birds (312.3 ± 26.6 s; 2.82 ± 0.19 mm). All data were analyzed using PROC GLM and significant differences were considered at P < 0.05; the LSD post hoc procedure was used for mean separation. The results indicate that illuminating the birds at their level not only increases growth and feed conversion but results in birds that are less fearful and less stress susceptible. This method of illuminating birds might save energy and improve production and bird welfare.

Key Words: lighting, LED, broiler, fear, stress

271P The effect of dietary supplementation of Agrado on performance, egg quality, and behaviors in Bovans Brown hens. J. Y. Hu*, R. L. Dennis, and H. W. Cheng, Purdue University, West Lafayette, IN, 1Livestock Behavior Research Unit, USDA-ARS, West Lafayette, IN, 2University of Illinois, Urbana, IL.

Agrado is an antioxidant (ethoxyquin, propylene glycol, and propyl gallate) with functions in reducing lipid oxidation and preventing harmful free radical formation that negatively affects health and productivity in animals. The objective of this study was to determine if dietary supplementation of Agrado improves hen performance, egg quality, and behaviors following transportation and regrouping for housing. Forty-eight Bovans Brown hens at 28 wk of age were regrouped and transferred from floor pens to conventional cages (2 birds/cage) at 12 replicates per treatment. The cages were randomly assigned to either control (fed a commercial diet) or 1 of 3 treated groups receiving Agrado (low, 200 ppm; medium, 400 ppm; and high 600 ppm, respectively) for 2 wk. Egg production was recorded daily; feed intake, egg quality, and body weight gain were recorded weekly. Behaviors (eating, drinking, and cage pecking) were measured twice per week. Data were analyzed using an ANOVA in the MIXED model procedure of SAS. Egg production was increased in all treated hens compare with controls (P < 0.001) but without dose effects (P > 0.1). The incidence of broken eggs was greatest in the control group compared with treated hens (P = 0.0004). BW was reduce in all hens over the time period, however, compared with controls, medium treated birds tended to lose less (P = 0.06), together with a tendency to have a higher feed intake (P = 0.1). The medium group hens also exhibited less stereotypic behaviors such as cage pecking (P = 0.001) than controls. Our data suggest that dietary supplementation of Agrado, especially at 400 ppm, has positive effects on hen egg production and well-being.

Key Words: Agrado, antioxidant, hen, egg performance, behavior

272P The effect of thermally cooled perches installed in cages on White Leghorn hen performance. H. W. Cheng*, M. M. Makagon, R. S. Gates, J. Y. Hu, S. A. Enneking, and P. Y. Hester, 1Livestock Behavior Research Unit, USDA-ARS, West Lafayette, IN, 2Purdue University, West Lafayette, IN, 3University of Illinois, Urbana, IL.

As producers convert from conventional to enriched cages, the use of thermally cooled perches as a tool to minimize the deleterious effects of heat stress on hens requires evaluation. Our objective was to determine if use of water cooled perches during the summer of 2013 affected hen performance, feather condition, and foot health. White Leghorn pullets (n = 162), 16 wk of age, were assigned to 1 of 3 banks of 6 cages each. A bank consisted of 3 deck levels with 2 cages per deck. Each bank was assigned to 1 of 3 treatments from 16 to 32 wk of age: (1) conventional cages with circular perches that were cooled with circulating water when temperature within the perch exceeded 25°C during June through September, (2) conventional cages with the same perches but with ambient air and no coolant, and (3) conventional cages without perches. Hens were subjected to 4 h of acute stress of 32.0 to 34.6°C at 27.6 wk of age. Numbers of eggs, including cracked and dirty eggs, were recorded daily. If use of water cooled perches during the summer of 2013 affected hen performance, feather condition, and foot health. The medium group hens also exhibited less stereotypic behaviors such as cage pecking (P < 0.001) than controls. Our data suggest that dietary supplementation of Agrado, especially at 400 ppm, has positive effects on hen egg production and well-being.

Key Words: cooled perches, egg production, shell traits, hyperkeratosis, foot health

Behavior and Well-Being
273P  The musculoskeletal health of caged White Leghorn hens with access to thermally cooled perches.  P. Y. Hester*1, M. M. Makagon1, R. S. Gates2, J. Y. Hu1, S. A. Enneking3, and H. W. Cheng3, 1Purdue University, West Lafayette, IN, 2University of Illinois, Urbana, IL, 3Livestock Behavior Research Unit, USDA-ARS, West Lafayette, IN.

Enriched cages that include perches are being installed into commercial egg laying facilities to meet some behavioral needs of laying hens. Because excessive heat can compromise animal welfare causing stress and inducing mortality, the use of thermally cooled perches to minimize the deleterious effects of heat stress in caged laying hens needs assessment. The objective of the current study was to determine if use of water-cooled perches during the summer of 2013 affected bone mineralization and muscle deposition of 32-wk-old White Leghorn hens. Pullets, 16 wk of age, were assigned to 1 of 3 banks of 6 cages each. A bank consisted of 3 deck levels with 2 cages per deck. Each bank was assigned to 1 of 3 treatments from 16 to 32 wk of age: (1) conventional cages with round metal perches that circulated cooled water when perch temperature exceeded 25°C, (2) conventional cages with identical perches except there was no coolant, and (3) conventional cages with no perches. A 4-h acute heating episode where temperatures were increased to a range of 32.0 to 34.6°C was instigated when hens were 27.6 wk of age. Hens (n = 12/treatment) were euthanized and BW determined at 32 wk of age. Perch temperature exceeded 25°C, (2) conventional cages with identical perches except there was no coolant, and (3) conventional cages with no perches. A 4-h acute heating episode where temperatures were increased to a range of 32.0 to 34.6°C was instigated when hens were 27.6 wk of age. Hens (n = 12/treatment) were euthanized and BW determined at 32 wk of age. Perch temperature exceeded 25°C, (2) conventional cages with identical perches except there was no coolant, and (3) conventional cages with no perches. A 4-h acute heating episode where temperatures were increased to a range of 32.0 to 34.6°C was instigated when hens were 27.6 wk of age. Hens (n = 12/treatment) were euthanized and BW determined at 32 wk of age.

Mineralization of the left femur, tibia, keel, humerus, ulna, and radius was determined using dual energy x-ray absorptiometry. The muscles and tendons of the breast, thigh, and drum were excised from bone and weighed. Data were subjected to an ANOVA or an analysis of covariance with BW as the covariate where appropriate using the MIXED model procedure of SAS. Bone mineralization and muscle deposition were not affected by treatment. As an example, the bone mineral densities of the keel were 0.091 ± 0.002, 0.095 ± 0.004, and 0.088 ± 0.003 g/cm² (P = 0.58) for hens of the cooled perch, non-cooled perch, and control (no perch) treatments, respectively. In conclusion, cooled perches had no effect on musculoskeletal health of 32-wk-old White Leghorn hens perhaps because the summer of 2013 was characterized as mild and the 4 h of acute heat stress occurred 5 wk before assessing musculoskeletal health.

Key Words: cooled perches, bone mineralization, muscle deposition, White Leghorn, heat stress

274P  Pre-stun shocks in electrical water-bath stunners in broiler: Indicative of animal welfare and effects on carcasses.  C. M. Giacomazzi1, A. M. Silveira1, G. P. Bergmann1, R. D. Sainz2, and L. Kindlein*1,2, 1Federal University of Rio Grande do Sul, Porto Alegre, Brazil, 2University of California, Davis, CA.

The objective of the present study was to test a relationship between pre-stunning shock in electrical water-bath stunners and the occurrence of hemorrhagic lesions in broilers carcasses. Thus, the hypothesis was that meat quality could be an indicator of animal welfare. A review of the literature has suggested that some birds receive a painful electrical shock before their stunning, detrimentally affecting their welfare. This would be a strong failure in the welfare of these animals. Thus, a total of 40,006 broilers from 29 different origins, raised in a traditional intensive system, were slaughtered in a plant in Rio Grande do Sul, Brazil. The presence of an electrical shock to the birds immediately before stunning in the immersion tank was checked by visual assessment of the birds’ behavior. The voltage, current, and frequency of the stunning tub were standardized to 70 V, 1.4 A, and 1.2 kHz, respectively. During postmortem inspection, the carcasses were evaluated for the presence or absence of hemorrhagic lesions and location of these lesions (wings, breast, and legs). Data were analyzed using the Chi-squared test (5%) using SPSS software, and the correlations between “occurrence of pre-shock” and “occurrence of carcass lesions,” were performed considering the location of lesions and body weight. Within all 29 lots evaluated, 82.75% of the birds displayed pre-shock behavior. The percentage of bruises in the chest was significantly higher (2.95 ± 2.7) for birds that suffered pre-shock compared those that did not (0.6 ± 0.89; P < 0.05). The hemorrhagic lesions occurrence on the wings and legs did not differ with the absence (10.4 ± 2.40% and 14.2 ± 6.37%) or presence (9.04 ± 3.31% and 9.5 ± 4.75%) of pre-shock. Birds with lower body weight (2.71 ± 0.21 kg) had a higher incidence of pre-shock compared with animals with higher weight (3.17 ± 0.11 kg; P < 0.05). The very high percentage of pre-shock occurrences demonstrated an animal welfare problem. This failure in the slaughter process resulted in an increased incidence of hemorrhagic lesions in the carcass detected only in the chest.

Key Words: animal welfare, hemorrhagic lesion, pre-stun shock, meat quality

275P  Effects of dietary tryptophan on serotonin and behavior in laying hens.  R. L. Dennis* and H. W. Cheng, Livestock Behavior Research Unit, USDA-ARS, West Lafayette, IN.

Serotonin (5-HT), an indolamine neurotransmitter synthesized from tryptophan (TRP), has been shown to regulate aggressive and impulsive behaviors including feather pecking and stereotypic behaviors. The present study investigated the effects of feeding excess TRP on behavior and neurophysiology. White Leghorn hens (n = 144) were assigned to 2-bird cages and fed either a standard layer diet (C; 0.16% TRP) or diets supplemented with TRP at a final concentration of 0.4, 0.8 and 1.6%. Diets were fed from 18 to 26 wk of age. Behaviors were recorded and analyzed for frequency of feather pecking, aggressive behaviors and stereotypic displays, by continual sampling for two 1-h periods at 25 wk of age (n = 18 cages/trt). Blood and brain samples (raphe nucleus and hypothalamus) were taken from one bird per cage at 26 wk of age (n = 12/trt). Blood was analyzed for 5-HT concentrations and the left brain regions were analyzed for 5-HT and dopamine (DA) concentrations, and their metabolites, 5-hydroxyindoleacetic acid (5-HIAA) and homovanillic acid (HVA) by HPLC. The right brain regions were analyzed by qRT-PCR for differential expression of 5-HT1A and 5-HT1B receptor genes. Data were analyzed using an ANOVA. Hens fed 1.6% dietary TRP displayed fewer aggressive behaviors and body shakes (P < 0.004 and 0.05, respectively). Cage pecking was decreased in 0.4 and 0.8% fed hens compared with both C and 1.6% hens (P < 0.02 and 0.04, respectively). Compared with C, whole blood 5-HT concentrations were increased in all treated groups (P < 0.01). Hens fed 1.6% had higher 5-HT concentrations in the hypothalamus (P < 0.03) but not the raphe nucleus (P = 0.06), compared with C. DA and HVA were also greatest in the raphe nucleus of 1.6% fed hens compared with C (P < 0.001 and 0.03, respectively). There was no treatment effect on the expression of 5-HT1A or 5-HT1B receptor genes (P = 0.66 and 0.72, respectively). These data show that dietary TRP caused modification of the neural systems is concentration and region dependent. Our results suggest a role for dietary TRP to improve hen well-being by reducing the expression of cage pecking and aggressive behaviors.

Key Words: laying hen, tryptophan, serotonin, behavior, well-being
**276P** Effect of methionine on feather follicle development of chickens. J. H. Vilas-Da Silva*1, F. González-Cerón1, E. W. Howeth2, and S. E. Aggrey1, 1Department of Poultry Science, University of Georgia, Athens, GA, 2Veterinary School, University of Georgia, Athens, GA.

We studied the effects of methionine supplementation on feather follicle development in dorsal and ventral area of Cobb broiler chickens at 21 d. The diets were treatment 1 (basal-negative control, NC): 0 mg/kg betaine (BET), 1.57 mg/kg folic acid (FA), and 0.35% l-Met; treatment 2: NC + 0.19 mg/kg BET + 1 mg/kg FA; treatment 3 (positive control PC): supplemented with 0.19 mg/kg BET + 1 mg/kg FA + 0.19 mg/kg dl-Met. One hundred eighty male chicks were placed in completely randomized design heated batteries and fed 3 dietary treatments (starter), with 4 replicates and 15 chicks per replicate. Twenty-four chicks were killed and ventral and dorsal skin samples were taken (24 cm²) to evaluate skin and feather follicle histology at 21 d of age. The NC and NC+FA+BET groups had shallower follicle in ventral skin compared with PC group (186.7a, 238.3a and 552.8a⁸ mm) and smaller diameter in dorsal skin (122.3b, 131.8b and 743.8a⁸ mm). The PC diet increased the depth in dorsal (1565.0a vs 552.8b⁸ mm) and diameter (743.8a vs 226.5b⁸ mm) compared with dorsal area. The PC improves epidermis thickness compared with NC and NC+FA+BET (32.4a, 21.8b and 23.8b⁸ mm) and dermis compared with NC (243.4a and 119.9b⁸ mm). The PC improves hypodermis thickness in comparison with NC and NC+FA+BET in ventral (1,617.5b, 388.3b and 535.3a⁸ mm) and dorsal area (1,817.1a, 723.5b and 1,285.3b⁸ mm). Supplementation of methionine-deficient diets with either FA+BET can support a good early ventral and dorsal feather follicle development and skin thickness. However, methionine requirements need to be met for proper feather follicle and feathering development.

Key Words: methionine, feather follicle development, welfare, betaine, folic acid

**277P** Effect of dietary supplementation with creatine monohydrate during the finishing period on growth performance, carcass traits, meat quality, and muscle glycolytic potential of broilers suffering from long-term transport stress. L. Zhang1, J. L. Li1, T. Gao1, M. Lin1, X. F. Wang2, X. D. Zhu2, F. Gao*1, and G. H. Zhou1,

1College of Animal Science and Technology, Nanjing Agricultural University, Nanjing, China, 2College of Science, Nanjing Agricultural University, Nanjing, China.

This study evaluated the supplementation of creatine monohydrate (CMH) during the finishing period on growth performance and meat quality of broilers suffered from long-term transport stress. A total of 320 male Arbor Acres broiler chickens (28 d old) were randomly allotted to 1 of 3 experimental diets supplemented with 0 (160 birds), 600 mg/kg (80 birds), or 1,200 mg/kg (80 birds) CMH for 14 d. On the morning of 42 d, after an 8-h fast, the birds of CMH-free group were divided into 2 equal groups, and all birds of these 4 groups were transported according to the follow protocol: 45-min transport without CMH supplementation (as a lower stress control group), 3-h transport without CMH supplementation, 3-h transport with 600 mg/kg, or 1200 mg/kg CMH supplementation. Each treatment group was composed of 8 replicates with 10 birds. The results showed that supplementation of CMH did not affect the overall growth performance and carcass traits of stressed broilers (P > 0.05), but dietary supplementation with 1200 mg/kg CHM tended to decrease daily feed intake of birds (P = 0.083). A 3-h transport decreased plasma glucose concentration (P < 0.01), elevated plasma corticosterone level (P < 0.05), increased chicken death on arrival (P < 0.05) and live weight loss (P < 0.01), enhanced the breakdown of muscle glycogen as well as the accumulation of muscle lactate content (P < 0.05), and induced some detrimental changes to breast meat quality (lower ultimate pH, higher drip loss and L* value, P < 0.05). Nevertheless, supplementation of 1,200 mg/kg CMH significantly decreased chicken weight loss (P < 0.05), tended to reduce chicken stress response by decreasing plasma corticosterone concentration (61.81 ng/mL vs. 55.49 ng/mL, P = 0.092), and alleviated transport induced negative changes of breast meat quality by reducing muscle glycolysis rate (P < 0.05). These findings suggest that CMH could be an effective anti-stress additive in the management of broiler transportation.

Key Words: creatine monohydrate, broiler, transport stress