

ABSTRACTS
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1 Early maturation of broiler breeder males. C.A. Pietsch*¹, J.B. Hess¹, R.J. Lien¹, and W.D. Berry¹, ¹*Department of Poultry Science, Auburn University.*

Sex separate rearing allows for manipulation of the development of breeder males. Advantages to early maturation include reduced cost of feed and ability to grow replacement males in less time. However, birds reared to early maturation may require a higher level of protein in the diet to compensate for reduced cumulative protein intake. For this experiment, 3 pens, each containing 100 cockerel chicks, were started on a 20% protein diet. After 3 weeks, chicks were divided into 3 treatments with 4 replicates containing 25 chicks. A linear body weight goal was developed for the fast growth birds targeting the goal body weight suggested in the management guide for 22 weeks of age (3.32 kg) by 16 weeks. The fast growth birds were fed either a high protein grower diet (HP) with 20% protein or a normal grower diet (NP) with 15% protein; light stimulation occurred at 16 weeks. The control group (CD) was fed the normal grower diet (15%) and reared according to guidelines, with light stimulation starting at 22 weeks. Weekly average body weight was entered into an energy needs formula to determine amount of feed required for the week. Plasma samples, growth measurements, and organ weights were collected throughout the experiment. No treatment differences were found. Weight uniformity was higher for the NP and HP groups than for the CD groups. In the second part of the trial, 3 roosters per pen were marked and semen collected for analysis weekly, starting at 20 weeks of age for the HP and NP groups and at 26 weeks of age CD groups. Also in this part of the experiment two replicates from the same growth treatment were combined using 4 roosters per pen; these birds were then placed with 80 hens, resulting in 2 replicates per treatment with 8 roosters per breeder pen. Weekly, 90 eggs per pen were set for 3 days and then opened to determine fertility. No treatment differences existed for fertility and sperm count. It was concluded that it is feasi-

ble to rear roosters using a fast growth program and introducing light stimulation at an earlier age; a higher level of protein is not required.

Key Words: Breeder males, Maturation, Growth, Fertility

2 The impact of sperm mobility and sperm morphology on older flock fertility. E.R. Bowling*¹, D.P. Froman², and J.L. Wilson¹, ¹*The University of Georgia, Athens GA*, ²*Oregon State University, Corvallis OR.*

In the commercial broiler breeder setting, flock fertility is one of the most important factors, and it is common for fertility to decrease as the birds age. A population of males was screened using the sperm mobility assay in order to determine low and high sperm mobility phenotypes. Roosters in the low (n=10) and high (n=10) sperm mobility phenotypes were randomly selected for use in artificial insemination. For each semen sample (n=20), 15 White Leghorn hens were inseminated with 75×10^6 sperm in a 50 μ L volume. Eggs were collected 2-14 days post insemination and incubated once per week. Insemination occurred when males were 50, 54, 58, 62, and 65 weeks of age. Breakout analysis was performed on all nonviable eggs at day 12 of incubation. Fertility in both the low and high sperm mobility phenotypes declined as the flock aged, and high sperm mobility males produced more fertilized eggs than low sperm mobility males at all ages (p<0.05). A transmission electron microscope (TEM) was used to evaluate differences in the sperm cells of three males from each phenotype. Fresh ejaculates (motile sperm) were collected from individual males, and on the following day sperm cells (non-motile) were extracted from the left deferent duct of the same roosters. Mitochondrial ultrastructure was examined in staggered serial sections (n= an average of 400 sperm per male) using the TEM. Any midpiece containing disorganized or swollen mitochondria were categorized as aberrant. Percentages of total aberrant mitochondria were expressed as mean SEM. Percentages for high and low sperm mobility phenotypes were 4.63 0.236 and 19.07 1.321, respectively. These data support the contention that differences in sperm mobility are related to mitochondrial status, and sperm

mobility is an important factor in predicting the fertilizing ability of older broiler breeder males.

Key Words: Broiler breeder, sperm mobility, fertility, transmission electron microscope, mitochondria

3 Semen dilution prior to analysis influences the ability of the sperm quality index to predict fertility whether inseminating with a constant number of sperm or a constant volume of semen. H. M. Parker* and C. D. McDaniel, *Mississippi State University, Mississippi State, MS.*

Previous research has shown that the sperm quality index (SQI) is positively correlated with semen characteristics as well as fertility when semen is diluted 10-fold prior to analysis. Extensive research has not been conducted to establish if semen dilution rate affects the ability of the SQI to predict fertility when inseminating a constant number of sperm or a constant volume of semen. As a result, two experiments were undertaken. The objective of the first study was to identify potential semen dilution rates for the SQI by obtaining the range in live sperm concentration to which the SQI is most sensitive. The purpose of Experiment 2 was to determine which semen dilution rate, 10-, 25-, 50-, 75-fold, or a constant 100×10^6 sperm/mL, yields an SQI that is the most predictive of fertility. Once a week for three weeks, 20 hens were individually inseminated with either 20 μ L of 4-fold diluted semen or a constant 25×10^6 sperm from each of 29 broiler breeder males. To determine which semen dilution rate yielded an SQI that was most predictive of fertility, Pearson's correlation coefficients were obtained between the SQI at each dilution rate and fertility. Sperm quality index values declined logarithmically as live sperm concentration decreased. The SQI was most sensitive to live sperm concentrations from 11 to 232×10^6 sperm/mL. The SQI for semen diluted 10-fold was the best predictor of fertility when inseminating with a constant volume of 4-fold diluted semen ($r=0.71$). When hens were inseminated with a constant 25×10^6 sperm/hen, the SQI for semen diluted 10- and 25-fold as well as to a constant 100×10^6 sperm/mL was equally effective at predicting fertility ($r=0.59, 0.52, \text{ and } 0.61$, respectively). Apparently, the SQI is very predictive of fertility when semen samples are diluted 10-fold prior to analysis regardless of insemination method used.

Key Words: Sperm Quality Index, Fertility, Broiler Breeders, Semen, Artificial Insemination

4 Utilization of cottonseed meal during the rearing period of broiler breeder pullets does not affect future reproductive performance. M. M. Lordelo*, A. J. Davis, J. L. Wilson, and N. M. Dale, *University of Georgia, Athens, GA USA.*

Due to its lower nutrient density, cottonseed meal (CSM) may be a potential replacement for soybean meal (SBM) in broiler breeder pullet grower diets. By promoting a slower growth rate, it may be possible to improve pullet body weight uniformity. To investigate this hypothesis, 1500 day-old Hubbard breeder pullets were divided into 6 groups. From 2 to 18 weeks of age, 3 of the groups received a standard corn/SBM pullet grower diet, the remaining 3 groups receiving a diet containing 20% CSM. To lower gossypol intake, the CSM employed was manufactured with no added soapstock. At 18 weeks all birds returned to a standard corn/SBM diet. 300 Hubbard cockerels were reared on the standard diet. Individual bird weight was monitored every 2 weeks until 20 weeks of age, when hens previously reared with CSM or with the standard diet were separated into 8 breeder pens per treatment. Each pen contained 60 hens and 10 roosters. Egg production, fertility and hatchability were monitored until 32 weeks of age. Livers were collected for gossypol analysis every 4 weeks during the rearing period and once a week during the breeding period.

To achieve the breeder guideline ideal weight, more feed was provided to the birds consuming the CSM based diet. Consequently, the coefficient of variation of bird weight was significantly lower at 18 weeks of age for the birds reared with the CSM based diet compared to those fed the standard diet. This difference in uniformity was maintained until the onset of egg production. Hens from both treatments reached 25 and 50 percent egg production at the same time. Although not significantly different, by 32 weeks of age, birds reared on the CSM diet had produced on average about 1 more egg per bird than those reared on the standard diet. Egg weight, hatchability and fertility were not significantly different between the 2 treatments throughout the breeding period. Liver gossypol analysis showed that during the rearing period, gossypol levels reached 416 micrograms/g DM. By the onset of egg production average gossypol levels

had decreased to 77 micrograms/g DM. The results suggest that broiler breeder pullets can be successfully reared with a diet containing CSM as the major protein source so as to improve flock uniformity without a detrimental impact on future reproductive performance.

Key Words: Cottonseed meal, Gossypol, Broiler breeders

5 Effect of dietary available phosphorus and phytase on broiler breeder performance. C. V. Williams*, B. A. Lenfestey, and J. Brake, *North Carolina State University, Raleigh, NC USA.*

Two experiments were conducted to determine the effects of low dietary available phosphorus (AP) and phytase (Alltech) on performance of broiler breeders. In Exp. 1, Arbor Acres FSY broiler breeder pullets were grown on starter and grower diets containing 0.90% calcium and 0.45% AP to 8 wk of age followed by a second grower diet with 0.80% calcium and 0.35% AP to housing. Levels of AP were decreased at 8 wk of age to prevent excessive accumulation of phosphorus reserves after primary development of the skeletal system was complete. Arbor Acres yield males received a diet with the initial level of calcium and AP throughout rearing. From 21 wk males and females were mixed and fed a third grower diet to 5% lay. Photostimulation was at 21 wk of age in a slat-litter house. The third grower diet and the layer diet (corn-soy-wheat bran) were formulated to created eight treatments that represented a normal level of AP ($\sim 0.40\%$), the lowest level possible created by removing all added dicalcium phosphorus ($\sim 0.10\%$), and two intermediate levels ($\sim 0.20\%$ and $\sim 0.30\%$) with each diet fed with and without phytase to two pens of broiler breeders each. Exp. 2 was conducted as for Exp. 1 except that Ross 308 pullets and Ross males were fed the highest and lowest AP diets only with and without phytase (corn-soy only) to comprise four treatments with four replicate pens each. The wheat bran used in Exp. 1 was found to contribute phytase. There were no differences in fertility in either experiment but hatchability of fertile eggs was decreased by phytase in Exp. 1 due to increased late deads. There were no significant effects on egg production or mortality in Exp. 1 due to phosphorus level or phytase but while hen-day production was also not affected in Exp. 2, hen-housed production was decreased in the lower phosphorus without phytase treatment due to high female heat stress related mortality. These data show that broiler breeders may be fed diets without added phosphorus if phytase is included.

Key Words: Phytase, Phosphorous, Broiler breeders, Egg production, Mortality

6 Effect of delayed emergence from eggs of different age broiler breeder flocks on chick quality and subsequent live performance for further-processing. N. S. Joseph* and E. T. Moran, Jr., *Auburn University, Auburn, AL.*

An experiment was conducted to compare the quality and subsequent performance of chicks that emerge early in the hatcher versus those delayed until "pull". A total of 960 hatching eggs were obtained from Ross \times Ross 308 breeder flocks at 32 and 41 weeks of age. Prior to incubation, amounts of yolk, albumen, and shell weight were measured on 60 eggs and confirmed the expectation that those from 41 week old hens had more yolk and less albumen in all respects than those from 32 week old hens. Remaining eggs were incubated at 36.9°C (54%RH) to extend duration of emergence such that mean incubation length of early-hatch chicks was 497-hr (20.7 days), 12 hr less than late-hatch chicks (509 hr or 21.2 days). Total chick yield was similar between 32 and 41 week old hens (82 and 83% of eggs set, respectively) and greater for early (88%) than late (68%) emergence. Young breeders produced chicks having reduced body and yolk sac weights while chicks from the older flock had an advantage in both respects. Higher yolk sac weight was also associated with late emergence. Body weights of chicks after access to feed and water for 3 and 8 days continued to be at a disadvantage with birds from young hens, particularly with early emergence. Decreases in yolk sac at one day of age as well as liver at 8 days indicated that a diminished nutrient reserve was the basis for reduced early live performance. Males and females responded to treatments similarly. By 6 weeks of age, greater total feed conversion and carcass abdominal fat existed with broilers from 41 compared to those from 32 week old breeders while yield of skinless boneless breast meats were unaltered. Difference in chick emergence had

no major effects on final live performance, carcass quality, and breast meat yield.

Key Words: broiler, incubation length, live performance, carcass yield, hen age

7 Affect of storage time on Growth of *Salmonella Typhimurium* in egg components. Z Howard*¹, K Medvedev¹, R Moore¹, S Birkhold¹, and S Ricke¹, *Department of poultry science, Texas A&M University.*

Salmonella Typhimurium is responsible for about 23% of all nontyphoidal cases of Salmonellosis worldwide. An increasing trend in disease incidence makes it a necessity to try to understand the mechanisms of *Salmonella*'s invasion of the egg. Approximately 150 eggs were collected over a 24 hour period from a flock of single comb white leghorn hens at the Texas A&M Poultry Science farm. Eggs were held at refrigeration temperatures until time points of one-week intervals from zero to eight weeks and then removed for analysis. Each week, ten eggs were cracked, separated into yolk and albumen components, and inoculated with 10⁸ CFU/mL of novobiocin and naladixic acid (NO/NA) resistant *Salmonella Typhimurium* onto the vitelline membrane of the egg. Yolks were then covered with albumen. Eggs were incubated for twenty-four hours at 25C and again separated into albumen, yolk, and vitelline membrane samples. Samples were diluted ten fold and enumerated on brilliant green agar supplemented with NO/NA. Non-inoculated samples were also tested to assure eggs were not initially contaminated with *Salmonella*. Membrane sample counts where observed to increase significantly by eight weeks of storage after inoculation. Membrane counts increased from about 10⁵ to 10⁷ CFU/mL. Albumen samples also increased by eight weeks, but showed peaks of growth at two and five weeks of storage. After inoculation and incubation, albumen counts at zero weeks of storage were observed at 2.3X10⁵. By eight weeks, counts in albumen had reached 1.1X10⁹. Yolk samples showed no clear increase over time suggesting that the membrane was still mostly impermeable to *Salmonella Typhimurium* at eight weeks.

Key Words: *Salmonella Typhimurium*, Vitelline membrane, egg

8 Blood characteristics of commercial egg laying hens between 20 and 58 wk of age when inoculated with S6-strain of *Mycoplasma gallisepticum* at 10, 22, or 45 wk of age. E. Y. Basenko*¹, E. D. Peebles¹, S. L. Branton², P. G. Gerard¹, M. R. Burnham¹, and S. K. Whitmarsh¹, ¹*Mississippi State University, Mississippi State, MS*, ²*USDA ARS, SCPRL, Mississippi State, MS.*

The effects of S6-strain *Mycoplasma gallisepticum* (S6MG) inoculation and its timing on blood characteristics of layer chickens between 22 and 58 wk of age were investigated. A total of 160 Hy-Line W36 strain hens housed in 16 isolation units with 10 birds per unit. There were 4 replicate units per treatment. Control birds received sham inoculations at 10 wk of age. Treated birds received S6MG inoculations at 10, 22, or 45 wk of age. Birds were bled for determination of hematocrit, plasma total protein, and serum cholesterol (CHOL), triglycerides, and calcium (CA) at Weeks 20, 24, 43, 47 and 58. Main effects due to hen age were found for hematocrit, plasma total protein, and serum triglycerides. There were no significant differences between control and treated birds for any of the blood parameters investigated. However, across Weeks 47 and 58, CA was significantly higher in birds inoculated with S6MG at 10 wk of age when compared to those inoculated at 22 wk of age. Furthermore, at Week 47, CHOL was significantly higher in birds inoculated at 22 wk of age in comparison to those inoculated at 45 wk. These data suggest that timing of S6MG inoculation between 10 and 45 wk of age differentially influences the levels of CA and CHOL in commercial laying hens during post-peak lay.

Key Words: *Mycoplasma gallisepticum*, Blood characteristics, Commercial layers, Inoculation, Serum calcium

9 Relationships between Breeds, Coccidiosis Control Programs, and Lighting Programs. J. L. Bray*, T. E. Cherry, P. W. Weatherford, E. R. Oviedo, S. Thompson, S. Antle, J. David, and C. Roberts, *Stephen F. Austin State University, Nacogdoches, Tx, USA.*

RELATIONSHIPS BETWEEN BREEDS, COCCIDIOSIS CONTROL PROGRAMS, AND LIGHTING PROGRAMS J.L. Bray* and T.E.

Cherry, Department of Agriculture, Stephen F. Austin State University, Nacogdoches, TX 75962

This study evaluated the effects on performance and the interactions between two breeds of commercial broiler chickens, a coccidiosis control program compared to a coccidiosis vaccine program, and two lighting programs (0.01fc; 0.05fc). This trial was conducted as a completely random block factorial design, within a tunnel-ventilated, 96-floor pen research facility (63 birds/pen). Data collection for this trial consisted of average body weight, average feed conversion, adjusted feed conversion, mortality, and yield performance. The treatment assignments for this study were as follows: Trt 1 # Breed A, Salinomycin, Lighting Program 1. Trt 2 # Breed B, Salinomycin, Lighting Program 1. Trt 3 # Breed A, Coccidiosis Vaccine, Lighting Program 1. Trt 4 # Breed B, Coccidiosis Vaccine, Lighting Program 1. Trt 5 # Breed A, Salinomycin, Lighting Program 2. Trt 6 # Breed B, Salinomycin, Lighting Program 2. Trt 7 # Breed A, Coccidiosis Vaccine, Lighting Program 2. Trt 8 # Breed B, Coccidiosis Vaccine, Lighting Program 2. All 8 treatments were fed Bacitracin at 50 mg/ton, in the starter, grower, and withdrawal feeds. At 45 days of age, Breed A statistically had the best body weight and adjusted feed conversion. At the end of the study, 3 birds per pen were randomly selected and yield performance was evaluated. Statistically, Breed B, while on a coccidiostat program, and grown under lighting program 2 (Trt 6), had the best Breast Yield %. The results show that Breed B significantly has the best Breast Yield %.

Key Words: Breeds, Coccidiosis Control Program, Coccidiostat Program, Lighting Program, Broiler Chickens

10 Litter Characteristics and Nitrogen Mass Balance in Recycled Rice Hull Broiler Litter. C. D. Coufal*, C. Chavez, P. L. Niemeyer, and J. B. Carey, *Poultry Science Dept., Texas A&M University.*

The amount of nutrients in broiler litter is an important concern to broiler producers when making waste management decisions. As litter is recycled from flock to flock, the accumulation of nutrients in the litter needs to be taken into consideration. Emerging concerns over the fate of nitrogen in broiler production warrant investigation of the nitrogen mass balance in modern commercial broiler production. An experiment was conducted to characterize recycled broiler rice hull litter including the associated nitrogen mass balance. Broilers were reared in four concrete-floored experimental pens under simulated commercial conditions. Seven flocks were reared in succession on recycled rice hull litter, with the first flock starting on litter that had been previously used for one flock. Beginning rice hull litter depth was approximately 8-10 cm. Commercial broiler feed was provided ad libitum, as was water via nipple drinkers. Caked litter was separated from loose litter in each pen after removal of the flock at 40-41 days of age. The wet weight, moisture content and nitrogen content of all birds, feed, litter and caked litter were measured. Day-old chicks and feed accounted for all nitrogen entering the pens. At the end of each trial, residual nitrogen not accounted for in marketed broilers, mortality, litter or caked litter was assumed to have dissipated as dust or ammonia. Over the seven flocks, the percent of nitrogen (dry matter basis) in litter at the end of the flock increased through flock 3 (2.5 to 3.2 %) then remained relatively constant (3.25%). Litter nitrogen expressed as g Nitrogen / kg live weight tended to decrease as successive flocks were housed on the litter (9.3 g/kg, flock 1 to 1.7 g/kg, flock 6). Nitrogen lost as dust and ammonia tended to increase as successive flocks were housed on the litter (10.8 g/kg, flock 1 to 20.8 g/kg, flock 6). This data demonstrates the accumulation of nitrogen in recycled broiler litter is dependant on the number of flocks housed on the litter. Additionally, the partitioning of nitrogen through the broiler production process has been quantified over several successive flocks on recycled litter.

Key Words: nitrogen, balance, broilers, litter

11 Impact of Supplementary Methionine Source on Volatile Compound Concentrations in Broiler Excreta. C. Chavez*¹, C. D. Coufal¹, P. L. Reynolds¹, J. B. Carey¹, R. E. Lacey¹, R. C. Beier², and J. A. Zahn³, ¹*Texas A&M University, College Station, TX*, ²*USDA/ARS Southern Plains Research Center, College Station, TX*, ³*National Swine Research Center, USDA/ARS, Ames, IA.*

The focus of this research was to determine the effect of supplemental dietary methionine sources on volatile compounds in broiler excreta. Three trials were conducted utilizing straight run broilers in battery cages. All excreta were collected in litter pans lined with aluminum foil. Each

trial consisted of 5 treatment groups with 3 replications of 13 randomly distributed broiler chicks per pen. Treatment groups consisted of no supplemental methionine (control group), sodium methioninate aqueous solution, dry methionine hydroxy analogue, liquid methionine hydroxy analogue, dl-methionine. The methionine activity of each methionine source was 45.9, 52, 88, and 98% respectively. In trials 1 and 2, starter and grower diets were formulated to contain 0.8% total methionine activity, (except control group, 0.35% total methionine activity). In trial 3, starter and grower diets were formulated to contain 0.5% and 0.38% total methionine activity, respectively. Trial 3 control levels of methionine were as in earlier trials. There were no significant differences in body weight, feed consumption or feed conversion among the treatments in any trial. Odor volatiles were evaluated in all trials by an electronic nose (EN) (Cyranose 320, Cyranose Sci., Inc., Pasadena, CA), gas chromatography, mass spectrometry (GC/MS) in trial 2 and olfactory sensory panel (OSP) in trial 3. EN evaluation indicated significant differences in odor compounds in broiler excreta among the treatments. GC/MS revealed significant differences in odor compound concentrations among the treatments, with control levels consistently lower than other treatments. OSP evaluation indicated that the detection threshold of control samples was significantly higher than that of other treatment groups. These findings demonstrate that supplemental dietary methionine sources significantly influence odor volatile concentrations in broiler excreta and that even small levels of supplemental methionine have a significant impact on concentration of odor compounds and odor intensity.

Key Words: Methionine, Broiler, Odor, Excreta

12 Analysis of Different Sampling Strategies Over-time for the Determination of *Salmonella* Status in Broiler Houses. M. Rybolt*¹, T. Doler¹, R. Wills¹, J. Byrd², and R. Bailey¹, ¹Mississippi State University College of Veterinary Medicine, Starkville, MS, ²USDA ARS, SPARC, Food and Feed Safety Research Unit, College Station, TX.

The first step in the analysis of food safety hazards within the food animal production environment is a thorough assessment of factors that may influence the risks. Identification and utilization of the most appropriate sampling methods for performing biological risk assessment is of the utmost importance and can be challenging. The sensitivity, specificity, and repeatability of sampling techniques for enteric pathogens of human food safety concern are not well documented in broiler production. Previously, our laboratory compared four different methods of *Salmonella* isolation and found that incorporation of a secondary enrichment step significantly increased the isolation rate when compared to the other three protocols studied. Therefore, this superior method was used to evaluate different sampling strategies for classification of the *Salmonella* status of broiler

houses over a nine week period. Variation in the recovery of *Salmonella* was observed between weeks, houses, and sample types. These findings underscore the importance of proper method selection to be used for sample collection as well as the most appropriate bacterial isolation protocol utilized when attempting to characterize the *Salmonella* risk associated with poultry production.

Key Words: Salmonella, Risk Analysis, Food Safety, Methodology

13 Utilization of thermophilic microbes for digestion of poultry carcasses. P.L. Niemeier*¹, J.B. Carey¹, C.D. Coufal¹, and B.M. Hargis², ¹Poultry Science Department, Texas A&M University, ²Department of Poultry Science, University of Arkansas.

Disposal of poultry carcasses has become a growing environmental and economic problem. Burial, incineration, and composting are common means of disposing of poultry carcasses, but have been under scrutiny as ground water and air quality concerns grow. Thermophilic digestion of animal carcass waste can provide a rapid means of decomposition and disposal without the potential for a strong odor or flies, and could eliminate the disease causing agents due to the increased temperature. Thermophiles were isolated from poultry mortality compost using Raw Chicken Agar (RCA), producing colonies of thermophiles which would thrive on chicken carcasses as a sole nutrient source. Biological degradation processes of the thermophiles convert organic matter into carbon dioxide and other volatile compounds resulting in a loss of dry matter. Reduction in dry matter content of the reaction vessels was used to evaluate the extent of degradation. Trials 1-2 used small vessels containing ground chick carcasses, phosphate buffered solution (PBS) and bacterium from RCA plates. In trials 3-5, whole chick carcasses were used in small vessels. Trials 6-9 used large vessels consisting of viable broth from previous trials and several chicks to evaluate whole carcass degradation. The culture was continuously under selection for capability to degrade chick carcasses throughout the trials. At the end of each trial, the residue was strained through dense cheesecloth prior to dry matter determination of the un-degraded portion. Some unknown quantity of the dry matter would have remained in the solution and the cheesecloth, thus the reduction in dry matter can only be taken to indicate the relative degree of the degradative process, not a quantitative measure of microbial degradative process. Dry matter reduction was 33-42% for ground chick trials and 17-90% for whole chick trials. The pH of the vessels was 7.96-8.31 throughout the trials. It is apparent that a thermophilic species exists that will biodegrade chick carcasses. This procedure was found to be effective in the decomposition of broiler chicken carcasses without reduced potential for nuisances.

Key Words: thermophiles, composting, broiler carcasses

Monday, January 20, 2003 Nutrition Room:B313

14 Econometric feeding of commercial layers: 1. Optimal methionine + cystine to lysine ratio in corn-soy diets for maximum profits. D. A. Roland*, A. Bateman, M. M. Bryant, S. S. Sohail, and R. A. Voitle, Auburn University, Auburn, AL.

The purpose of this paper is to explain why the commonly used and recommended M+C/L ratio of 0.83 to 0.85 is incorrect. Significant implications of this research on producer profits and nutrient requirement of hens are discussed. In Exp.1, three strains (Hy-LineW-36, and two strains of Bovans) and three M+C/L ratios (0.83, 0.79 and 0.75) in a low protein (14.6%) diet were used. Hens (n=1440; housed at 68F to allow for high feed consumption) were fed diets from 46 to 54 wks of age. In Exp.2, two protein levels (19.8 and 18.7%) and six M+C/L ratios in 0.04 increments starting at 0.58 or 0.60 for the 19.8 and 18.7% protein diets respectively were used. The W-36 hens (n=1920; housed at 78F) were fed diets from 21-36 wks of age. Criteria used in both experiments were egg production (EP), egg weight (EW), and feed consumption (FC). Results of Exp.1 indicated a significant strain effect on EP, EW and FC. Hy-Line W-36 hens ate less feed (99 vs. 108 g/hen/d) and produced fewer (79 vs. 88%) but larger (61.9 vs. 60.9g) eggs than the Bovans. There were no interactions or M+C/L ratio effects. The optimal M+C/L ratio for each strain was 0.75 or less. In Exp. 2, M+C/L ratio had no effect on EP. All hens peaked at 93 to 95%, and averaged 91 to 92% (wk 21-36). Average FC was 83 g/hen/d and was not influenced by M+C/L ratio. EW was

significantly influenced by M+C/L ratio at both protein levels. The optimal ratio for hens consuming 19.8 and 18.7% protein was 0.70 and 0.76 respectively. The results indicate that producers using a M+C/L ratio of 0.85 in corn-soy diets are feeding an excess of approximately 1.7 lbs methionine/ton feed. Under conditions of this study, the lysine and M+C requirement for optimal profits (W-36 hens, phase 1) was 925 and 647 mg/hen/d in hens consuming 19.8 and 18.7% protein diet respectively. Based on economic analysis, it was concluded that only the M+C/L ratio can be fixed. No fixed requirement for protein, lysine or methionine can be stated because the factors (environmental temperature, energy intake, egg and feed prices) which determine the requirement vary continually.

Key Words: Econometric, lysine, methionine, protein, strains

15 Econometric feeding of commercial layers: 2. Relative bioefficacy of methionine hydroxy analog compared to DL-methionine. A. Bateman*, Z. Liu, M. M. Bryant, and D. A. Roland, Sr., Auburn University, Auburn, Alabama.

Three studies were conducted to compare the relative bioefficacy of liquid DL-methionine hydroxy analogue-free acid (MHA-FA) and DL-methionine (DLM). Biological efficacy was determined for egg production (EP), feed consumption (FC), egg weight (EW) and egg mass (EM)

using exponential nonlinear regression analysis, as well as linear (slope-ratio) regression analysis. In Trial 1, five graded levels of DLM (0.023, 0.045, 0.068, 0.090 and 0.113%) and MHA-FA (0.026, 0.051, 0.077, 0.102 and 0.128%) were added on an equimolar basis to a basal diet containing 14.97% protein and 0.27% Met. This trial used 1760 first cycle, Phase II Hy-Line W-36 hens. There was no response above the basal diet in any of the criteria measured, so no regression analysis was performed. In Trial 2, four graded levels of DLM (0.012, 0.024, 0.036, and 0.048%) and MHA-FA (0.014, 0.027, 0.041, and 0.054%) were added on an equimolar basis to the basal diet used in Trial 1. This trial used 1440 first cycle, Phase I Hy-Line W-36 hens. Exponential nonlinear regression analysis revealed that the bioefficacy of liquid MHA-FA vs. pure DLM on an equimolar or weight for weight basis respectively was 81.7 or 72.7% (EP), 97.5 or 86.0% (FC), 111.2 or 98.4% (EW) and 89.9 or 77.4% (EM), while linear (slope-ratio) analysis revealed that the bioefficacy was 86.1 or 75.8% (EP), 101.4 or 89.2% (FC), 113.9 or 100.2% (EW) and 92.6 or 81.5% (EM). In Trial 3, five graded levels of DLM (0.012, 0.024, 0.036, 0.048 and 0.060%) and MHA-FA (0.014, 0.027, 0.041, 0.054 and 0.068%) were added on an equimolar basis to the basal diet used in Trial 1. This trial used 1760 second cycle, Phase I Hy-Line W-36 hens. Exponential nonlinear regression analysis revealed that the bioefficacy of liquid MHA-FA vs. pure DLM on an equimolar or weight for weight basis respectively was 94.0 or 89.7% (EP), 102.1 or 90.2% (FC), 120.7 or 107.8% (EW) and 101.9 or 93.8% (EM), while linear (slope-ratio) analysis revealed that the bioefficacy was 89.5 or 78.8% (EP), 91.0 or 80.1% (FC), 111.4 or 98.0% (EW) and 96.5 or 85.0% (EM).

Key Words: DL-methionine, MHA-FA, egg production, egg mass, laying hen

16 Econometric Feeding of Commercial Layers: 3. Bioefficacy of Methionine Hydroxy Analogue Compared to DL-Methionine on Layers with Linear and Nonlinear Models. Z. Liu, A. Bateman, S. S. Sohail, M. M. Bryant, and D. A. Roland, Sr., Auburn University, Auburn, Alabama.

Three experiments were conducted to determine the bioefficacy of methionine hydroxy analogue free acid (MHA) compared to DL-methionine (DLM) on commercial layers. Experiment 1 was designed for linear and nonlinear regression models to determine the bioefficacy of MHA compared to DLM. In this experiment, five graded supplemental levels (0.012, 0.024, 0.036, 0.048 and 0.060%) of methionine from DLM and MHA were added to a basal diet (containing 0.27% methionine) on equimolar basis. Regression analysis showed that bioefficacy on molar or weight basis respectively were 88.03% or 77.47% (egg production), 87.67% or 77.15% (egg mass) and 84.93% or 74.74% (egg weight) with an exponential model, and were 89.72% or 78.95% (egg production), 89.59% or 78.88% (egg mass) and 86.75% or 76.34% (egg weight) with a slope-ratio model. Experiment 2, a 3 x 2 x 2 factorial experiment, was designed to determine the sensitivity of bioefficacy comparison between DLM and MHA on layers. There were three protein levels (15.06%, 16.18% and 17.44%), two methionine levels (0.02% and 0.04%) and two methionine sources (DLM and MHA). The analysis of egg production and egg mass data showed that it was difficult to detect the difference between DLM and MHA if it exists, since no difference due to levels, which was supposed to be larger than the difference due to sources, was obtained. The analysis of egg weight data showed that a significant difference due to methionine levels was obtained, and the numerical difference due to methionine sources suggested that the bioefficacy was below 100% on molar basis or 88% on weight basis. Experiment 3 was designed based on assuming 65% bioefficacy of MHA compared to DLM. In this experiment, five graded supplemental levels of methionine (0.012, 0.024, 0.036, 0.048 and 0.060%) were added to a basal diet, but MHA was calculated as 65%. The results showed that the bioefficacy of MHA was significantly higher than 65% ($P \leq 0.05$) with egg production and egg mass as the criteria, and was not significant higher than 65% ($P \leq 0.05$) with egg weight as the criterion.

Key Words: DL-methionine, Methionine hydroxy analogue, bioefficacy, layer

17 Econometric Feeding of Commercial Layers: 4. Comparison of Performance and Nutrient Requirements of Two Strains after Molting. M. M. Bryant*, D. A. Roland, Sr., A. Bateman, and P. Curtis, Auburn University, Auburn, Alabama.

A study was conducted to compare the performance and nutrient requirements of two strains of commercial leghorns (Bovans White and DeKalb

White) after molting. Three hundred and sixty hens from each strain were force molted. After molting, hens were divided into groups and fed one of three diets yielding a 2 x 3 factorial design. Diets were formulated to reflect commercial diets with low density (16.2% protein, 0.67% methionine + cystine, 0.83% lysine), medium density (17.3% protein, 0.75% M+C, 0.92% lysine) or high density (18.7% protein, 0.73% M+C, 1.02% lysine). Due to cold weather and high feed consumption, diets were reformulated after six weeks to reduce diet density by 10% for each diet. These diets were fed for an additional six weeks. Dekalb Whites had higher egg weights, higher egg specific gravity, lower feed conversion and a lower percent egg solids than Bovans White. After molt peak egg production was 89% for DeKalb Whites and 92% for Bovans Whites. There was a significant diet effect in which hens fed the high density diets had higher egg production, higher egg weights and lower feed conversion than hens fed the low density diets. Data were subjected to econometric analysis. Results indicate that the requirements of Bovans White for maximum profit under trial conditions were 950 mg/h/d of lysine (18.4 mg/g egg), 713 mg/h/d of total sulfur amino acids (13.8 mg/g egg), 18.5 g/h/d of protein (0.358 g/g egg) and 323 kcal/h/d of metabolizable energy (6.26 kcal/g egg). Requirements for Dekalb whites were 836 mg/h/d of lysine (15.7 mg/g egg), 627 mg/h/d of total sulfur amino acids (11.8 mg/g egg), 16.7 g/h/d of protein (0.315 g/g egg) and 315 kcal/h/d of metabolizable energy (5.93 kcal/g egg). These requirements could change with different egg prices, feed costs and environmental temperature (energy intake).

Key Words: strain, protein, laying hen

18 Arginine and Coccidiosis Responses in Broiler Chicks. S. A. Thornton*¹, S. J. Barber¹, W. S. Virden¹, J. P. Thaxton¹, B. J. Kerr², and M. T. Kidd¹, ¹Mississippi State University, Mississippi State, MS, ²United States Department of Agriculture, Ames, IA.

Arginine (Arg) is an essential amino acid in broilers that has numerous physiological and immunological functions, in addition to being required for growth. This experiment consisted of a 3 x 2 factorial design of dietary Arg (1.00, 1.25, and 1.50% of diet) and coccidiosis (with and without a field isolate of *Eimeria tenella* and *acervulina*) in Ross x Ross 308 male broilers from Days 4 to 18 (6 treatments with 8 replications). All chicks received a common pre-starter diet to Day 4. Birds were reared in starter batteries (9 birds/pen) that were thermostatically controlled and had raised wire floors. Arginine treatments were randomized across two batteries in separate rooms to prevent cross-contamination of oocysts. At Day 11, birds were inoculated with 60,000 sporulated oocysts per os. Gross lesion scores were greater ($P < 0.05$) in birds inoculated with oocysts 3 d postinoculation. At 7 d postinoculation, gross lesions and relative liver and spleen weights were greater ($P < 0.05$) in inoculated birds than controls, but inoculated birds had depressed ($P < 0.05$) relative thymus weight. In addition, birds inoculated had poorer ($P < 0.05$) BW gain, feed conversion, and livability. Birds fed 1.50% dietary Arg had lower ($P < 0.05$) corrected feed conversion than birds fed the diet containing 1.00% Arg. Birds fed diets supplemented with Arg to 1.25 or 1.50% had depressed ($P < 0.05$) relative liver weights at 7 d postinoculation in comparison to birds fed the diet containing 1.00% Arg. An interaction occurred for percentage N retained that was measured 4 d postinoculation. The interaction indicated a depression in percentage N retained in inoculated birds fed 1.50% Arg in comparison to inoculated birds fed 1.25% Arg or uninoculated birds fed 1.00 or 1.50% Arg. Results indicate that birds fed diets containing low dietary Arg levels in comparison to birds fed diets containing Arg levels to optimize feed conversion (1.25 to 1.50% of diet) may have differing physiological and nutrient utilization responses during a coccidiosis infection.

Key Words: Arginine, Coccidiosis, Broiler

19 Tryptophan Need of Broiler Males from 42 to 56 Days of Age. A. Corzo*¹, E. T. Moran, Jr.¹, D. Hoehler², and B. Zinner³, ¹Poultry Science Department - Auburn University, ²Degussa Corporation, ³Statistics Department - Auburn University.

Tryptophan need of broiler males from 42 and 56 days of age has not been established. Ross x Ross 308 males (32 floor pens; 35 chicks/pen) were provided nutritionally adequate common feeds from 0 to 42 days of age. Subsequently, a corn, soybean meal, corn gluten meal, and gelatin combination of feedstuffs (18% CP, 3.25 Kcal/g ME, 0.85% lysine, 1.00% arginine, and 0.74% isoleucine) provided 0.12 tryptophan to which 0.04%

increments of L-tryptophan were supplemented at the expense of an isonitrogenous amount of L-glutamic acid to 0.24%. Birds receiving 0.12% tryptophan exhibited aberrant behavior based on the spillage of considerable amounts of feed from the trough while greatly contaminating adjacent waters with floor litter. Tryptophan supplementation of the feed to attain and exceed 0.16% eliminated this behavior. Body weight gain, feed conversion, and chilled carcass weights improved as tryptophan increased with maximum overall performance being attained at 0.17%. Abdominal fat removed from the chilled carcass after processing increased to a maximum corresponding to 0.16% tryptophan while the weights and proportions of fillets, tenders and total breast meat deboned from the chilled carcass maximized at 0.16 and 0.15%, respectively. Percentage nitrogen retention measured using the same experimental feeds and sample birds at 48-49 days of age linearly increased with dietary tryptophan to 0.24%. Plasma uric acid, albumin, total protein, and aspartate-transferase measured immediate to nitrogen retention were not altered; however, glucose linearly increased with dietary tryptophan. Overall results suggests that the broiler male needs about 0.17% dietary tryptophan between 42 and 56 days of age which agrees with the NRC (1994) recommendation of 0.16% estimated from modeling.

Key Words: Amino acid requirement, Broiler, Carcass quality, Live performance, Tryptophan

20 Impact of choline, betaine and sulfur amino acid level on growth performance and hepatic homocysteine flux in broilers during the starter and grower periods. P. B. Pillai*¹, L. N. Loupe¹, K. W. Beers², and J. L. Emmert¹, ¹University of Arkansas, Fayetteville, AR, ²Safe Foods Inc., Rogers, AR.

An experiment was conducted to assess the impact of supplemental choline or betaine on growth performance and hepatic homocysteine (HCY) metabolism of broilers from 8-21 d. Five pens of 5 male chicks were fed a CYS- and MET-deficient diet (0.227 and 0.253% digestible MET and CYS, respectively) or a CYS- and MET-adequate diet (0.427 and 0.453% digestible MET and CYS, respectively) to which were added 0.25% choline or 0.28% betaine. Addition of choline or betaine to the deficient diet increased weight gain and feed intake ($P < 0.05$), whereas feed efficiency was improved only by betaine ($P < 0.05$). Growth performance was not affected ($P > 0.05$) by addition of choline or betaine to the adequate diet. Hepatic tissue analysis (using a stable isotope of betaine) indicated that deficiency of sulfur amino acids (SAA) in the diet stimulated the remethylation of HCY. Choline or betaine addition to the deficient and adequate diets further stimulated HCY remethylation. A second experiment was conducted to assess the efficacy of liquid MET (HMB) in supporting growth of broilers fed SAA-deficient diets from 21 to 42 d. Five pens of 20 male chicks were fed a basal diet deficient in MET and CYS (0.214% digestible MET, 0.236% digestible CYS), and an additional four pens of 20 male chicks were fed a basal diet deficient in MET alone (0.214% digestible MET, 0.436% digestible CYS). Graded levels of DL-MET or MHA (0, 0.04, or 0.08%) were added to each of the basal diets. Weight gain increased linearly ($P < 0.05$) with increasing levels of DL-MET or HMB. Using slope-ratio methodology in which weight gain was regressed versus supplemental MET source intake, HMB was found to be 93.4% ($P > 0.05$) as efficacious as DL-MET when added to a diet deficient in MET and CYS, and 81.6% ($P < 0.05$) as efficacious as DL-MET when added to a diet deficient in MET alone. Liver samples were taken for future assessment of HCY flux.

Key Words: Broiler, Choline, Betaine, Sulfur amino acids, Homocysteine

21 Digestible lysine requirements of broilers under conditions of continuous or reduced levels of crude protein in test diets. C.A. Fritts*, M.W. Greenwood, and P.W. Waldroup, University of Arkansas, Fayetteville AR.

Controversy exists on total or digestible lysine needs of broilers, due in part to various times in which the test diets were fed. In this study diets were formulated for starter (0-14 d) and grower (14-42 d) periods to provide diets adequate in all amino acids but Lys. Corn, soybean meal, corn gluten meal and peanut meal of known composition were used with soybean meal and peanut meal at a 2:1 ratio. Digestible Lys (DigLys) was estimated from a composite of reported data. Benchmark starter and grower diets that exceeded current industry standards were used. One group of chicks was fed test diets based on starter feed from 0 to 42d with DigLys ranging from 0.75 to 1.2% in 0.05% increments. A second group was fed the benchmark starter to 14 d then placed on test diets

based on grower feed from 14 to 42 d. DigLys levels ranged from 0.60 to 1.1% in increments of 0.05%. Four replicate pens of 12 male chicks were fed each level of DigLys. Birds and feed were weighed weekly. Total and supplemental Lys agreed with calculated values. DigLys needs were estimated weekly by nonlinear regression. When based on continuous feeding of starter formulas DigLys needs were estimated to be 0.99, 0.98, 0.96, 0.98, 0.96, and 0.92 for BW, and nonestimable, 1.16, 1.08, 1.04, 1.05, and 1.0% for FCR for weeks 1 to 6, respectively. However, when chicks were fed starter diets for 14 d then placed on grower test diets the estimates were 0.79, 0.81, 0.81, and 0.82% for BW and 0.81, 0.85, 0.89, and 0.85% for FCR for weeks three through six, respectively. These results indicate that estimates of Lys should consider time fed and previous nutritional status.

Key Words: Digestible lysine, Broilers, Amino acid requirements

22 Efficacy of Microbial Phytase in Broiler Diets. R. N. Dilger*¹, E. M. Onyango¹, J. S. Sands², and O. Adeola¹, ¹Purdue University, West Lafayette, IN, ²Danisco Animal Nutrition, Marlborough, UK.

Two studies were conducted to evaluate the efficacy of a microbial phytase. Study 1 used 192 eight-day-old male broiler chicks in a 14-d trial to assess growth performance and nutrient utilization. Birds were blocked by BW and assigned to 4 dietary treatments (222 g kg⁻¹ CP) according to a RCBD with 6 chicks per cage and 8 replicate cages per diet. Dietary treatments included a positive control (4.5 g kg⁻¹ available P), basal (0.8 g kg⁻¹ available P), and basal plus either 500 or 1000 microbial phytase units/kg. A linear phytase effect was observed for weight gain, feed intake, feed efficiency and toe ash ($P < 0.01$) as well as tibia ash (linear and quadratic, $P < 0.05$). Phytase also increased apparent ileal P digestibility (linear, $P < 0.01$) with an additional linear effect shown for retention of DM and P ($P < 0.05$). Study 2 utilized 576 day-old male broilers in a 42-d growth performance trial arranged as a RCBD with 12 chicks per pen and 8 replicate floor pens per diet. Six dietary treatments were formulated for both starter (223 g kg⁻¹ CP) and grower (202 g kg⁻¹ CP) phases and consisted of a positive control (starter and grower, 4.5 and 3.3 g kg⁻¹ available P, respectively), basal (starter and grower, 1.9 and 1.4g kg⁻¹ available P, respectively), basal plus 500, 750, or 1000 microbial phytase units/kg, and basal plus 500 phytase units/kg from Natuphos. Pen body weights and feed consumption were recorded weekly and tibia and toe were collected from 2 birds per pen at the middle and end of the study. A linear phytase effect ($P < 0.05$) was observed for weight gain (d 0-21 and overall), feed intake, feed efficiency (d 0-21), as well as tibia and toe ash content (d 0-21 and d 0-42) relative to the basal diet. In conclusion, this microbial phytase gave growth and nutrient utilization responses similar to Natuphos phytase. Furthermore, growth performance responses of chicks were similar between phosphorus-adequate and phytase-supplemented diets.

Key Words: Microbial phytase, Growth, Nutrient metabolism, Chicken

23 Low phytate barley and phytase reduces phosphorus excretion in broilers fed diets for 42 days. L. B. Linares*¹, J. N. Broomhead¹, E. A. Guaiume¹, D. R. Ledoux¹, T. L. Veum¹, and V. Raboy², ¹University of Missouri Columbia, MO USA, ²USDA-ARS Aberdeen, ID USA.

An experiment was conducted to evaluate the efficacy of low phytic acid barley (LPB) and phytase in increasing P utilization by broilers. Low phytate mutant (M955) and wild-type (HC) barleys made up 54.5% of the diets which were fed to 420 d-old male chicks from hatch to day 42. There were 7 dietary treatments with 15 replicate pens of 12 chicks allotted randomly to each treatment. Treatments included: (1) Positive control corn-soybean meal diet containing 0.45% nonphytate P (npP), 1.0% Ca; (2 and 3) HC and M955 diets containing 0.45% npP, 1.0% Ca; (4 and 5) HC and M955 diets containing 0.35% npP, 0.9% Ca; and (6 and 7) HC and M955 diets containing 0.35% npP, 0.9% Ca but supplemented with 600 U/kg of phytase. Barley diets were supplemented with Ronozyme[®] B (β -glucanase, xylanase, and α -amylase). These diets were fed from day 1 to 21. From day 22 to 42, npP and Ca in all diets were decreased by 0.10%. Chicks fed diets 2 and 3 consumed similar amounts of feed as chicks fed diet 1. However, compared to chicks fed diet 1, chicks fed diet 2 gained less weight ($P < .05$), whereas chicks fed diet 3 were less efficient in converting feed to body weight gain ($P < .05$). Compared with chicks fed diet 1, chicks fed diets 4 and 5 consumed less feed, gained less weight, and were less efficient ($P < .05$). Chicks fed diets 6 and 7, performed

as well as chicks fed diet 1. Tibia ash of chicks fed dietary treatments averaged 41.9, 41.5, 41.5, 39.4, 39.0, 40.8, and 41.7% for diets 1, 2, 3, 4, 5, 6, and 7, respectively. Litter P concentrations were 1.38, 1.27, 0.87, 0.87, 0.54, 0.96, and 0.53% for diets 1, 2, 3, 4, 5, 6, and 7, respectively. Results indicate that feeding LPB will significantly reduce P excretion (31%) in diets containing NRC levels of npP. Feeding LPB, reducing the npP level by 0.1%, and using supplemental phytase reduced P excretion by an additional 27% while maintaining chick performance equivalent to that of birds fed NRC levels of npP.

Key Words: Phytic acid, Barley, Phosphorus excretion, Chicks, Phytase

24 The use of dietary phytase, 25-hydroxycholecalciferol, and 1-alpha-hydroxycholecalciferol in broiler chick nutrition. R.B. Shirley* and H. M. Edwards, Jr., *The University of Georgia, Athens, GA / USA.*

Two experiments were conducted to test the hypothesis that broiler performance is further improved when either 25-hydroxycholecalciferol [25-D₃ (68.75 ug/kg diet)] or 1-alpha-hydroxycholecalciferol [1-alpha-D₃ (5 ug/kg diet)] is added to a phytase-supplemented diet. Day-of-hatch, Ross x Cobb broiler chicks were used in each of the two 16-d experiments. The log-dose levels of phytase that were added to the corn-soybean meal basal diets of each experiment were: 0, 187.5, 350, 750, 1,500, 3,000, 6,000, or 12,000 U/kg diet. The basal diets of both experiments were formulated to contain 3,123 kcal of ME /kg diet, 22.4% CP, 0.95% Ca, and 0.45% total P. Referring to the main effect means of Experiments 1 and 2, supplemental phytase improved body weight gain (BWG) from 293 to 435 g/chick, feed intake (FI) from 386 to 534 g/chick, plasma P from 2.2 to 7.1 mg/dL, tibia ash from 29 to 40%, and decreased the incidence of P rickets from 81 to 1% in Experiment 1 ($P \leq 0.0001$). In Experiment 2, phytase increased BWG from 359 to 481 g/chick, FI from 449 to 510 g/chick, plasma P from 2.9 to 5.2 mg/dL, tibia ash from 30 to 40%, and decreased the incidence of P rickets from 71 to 3.9% ($P \leq 0.0001$). While there were no statistical differences in BWG, FI, or plasma P when 25-D₃ was supplemented to the phytase-treated diets of Experiment 1 ($P = 0.7208$, 0.3023, and 0.1077, respectively), there were phytase x 25-D₃ interactions for tibia ash and P rickets ($P \leq 0.0001$ and $P = 0.0140$, respectively). In Experiment 2, the opposite occurred, as there were phytase x 1-alpha-D₃ interactions for BWG, FI, and P rickets ($P \leq 0.0001$, $P \leq 0.0001$ and $P = 0.0397$, respectively), and no phytase x 1-alpha-D₃ interactions for plasma P or tibia ash ($P = 0.7190$ and 0.1200, respectively). Using a total P-deficient broiler diet, these data indicate that phytase supplementation above that of current broiler industry standards does improve broiler performance; and, that even though supplemental 25-D₃ or 1-alpha-D₃ reduces the incidence of P rickets in rapidly growing broiler chicks consuming phytase-supplemented diets, each affects BWG, FI, and tibia ash differently.

Key Words: Broiler chicken, Phytase, 25-hydroxycholecalciferol, 1-alpha-hydroxycholecalciferol

25 β -mannanase Removes Growth Inhibition Associated with Dietary Guar Meal. J. T. Lee*, C. A. Bailey, and A. L. Cartwright, ¹Texas A&M University System.

Guar meal is composed of germ and hull fractions. Hull fraction is a high protein ingredient with potential for poultry rations. However, the hull fraction depresses growth and feed efficiency due to a galactomannan polysaccharide that increases intestinal viscosity. Correlations between growth depression and increased intestinal viscosity are positive. Broiler starter diets with 2.5% hull fraction reduce growth and increase intestinal

viscosity. Hemicell[®], a β -mannanase degrades galactomannan polymers and reduces viscosity in broilers fed guar germ fraction. A 3 x 3 factorial experiment with three hull (0, 2.5, and 5.0%) and b-mannanase concentrations (0, 1, and 4 times the manufacturer's recommended dose, 1.09×10^5 units/kg) was used to determine the effect of guar hull fraction on growth parameters and ileal viscosity. The lowest level of guar hull significantly reduced body weight and increased feed conversion ratio, ileal viscosity, and relative ileal weight to 20 days of age. β -mannanase inclusion reduced ileal viscosity, increased body weight and reduced feed conversion ratio to control levels. Recommended b-mannanase level increased body weight and reduced feed conversion to control values in the 2.5% hull fraction starter diet while the highest concentration had no further impact. Supplementation of 5.0% hull fraction diet with β -mannanase significantly increased body weight and decreased feed conversion although not to control values, but the highest concentration of enzyme significantly improved body weight and feed conversion to control levels. β -mannanase supplementation did not affect measured parameters for the control diet. β -mannanase removed inhibitory effects associated with guar hull feeding.

Key Words: guar meal, β -mannanase, intestinal viscosity

26 Effects of adjusting the amino acid profile of a broiler diet when using a soybean meal derived from a high protein cultivar. B. A. Lenfestey*¹, P. W. Plumstead¹, J. W. Burton², R. F. Wilson³, and J. Brake¹, ¹North Carolina State University, Raleigh, NC USA, ²USDA-ARS, Raleigh, NC USA, ³USDA-ARS, Beltsville, MD USA.

Higher protein "designer" soybeans are now being developed. A consequence of an increase in soybean protein can be a change in the ratio of 11S (glycinin) and 7S (beta-conglycinin) storage proteins. As a result of this different ratio the amino acid profiles can differ from that of conventional soybean meal (SBM). In order to maximize bird performance when using designer SBM it may be necessary to use amino acid supplementation or blending with conventional SBM. Analysis of the meal produced from one cultivar, Prolina, indicated a higher than normal level of protein but a change in the amino acid profile of Prolina was evident when compared to a conventional SBM. Previous research has shown that diets containing a blend of Prolina and conventional SBM (BLEND) resulted in the highest BW when compared to broilers fed diets with SBM or Prolina alone. The present experiment was designed to adjust the amino acid profile of Prolina to resemble that of SBM and to further evaluate the BLEND performance phenomenon. Nine hundred male Cobb 500 broilers were randomly distributed among 36 pens. Starter, grower, and finisher diets were formulated using a conventional SBM amino acid profile. SBM, a BLEND, and Prolina soybean meal alone were substituted as the soybean meal source. Each diet was then fed with or without supplemental synthetic L-threonine (THR) (to reach 105% of NRC) in a 3 x 2 factorial. Addition of THR to Prolina treatments was found to improve BW at 42 d (2845g) as compared to the non-supplemented diet (2828g). No benefit was seen with THR supplementation in the SBM or BLEND diets. Birds fed the BLEND diet without THR had the highest 42 d BW (2851g) when compared to SBM (2788g) with Prolina being intermediate (2829g). Prolina had the best 42 d FCR among the treatments (1.68 g:g). These results indicate that THR is one of the limiting components of the Prolina amino acid profile. These data also support previous research showing that blending SBM and Prolina produces a beneficial amino acid balance that maximizes broiler BW. Prolina can be beneficial in broiler diets with proper amino acid profile adjustments.

Key Words: Soybean meal, Broiler, Threonine, Prolina, Designer soybeans

**Monday, January 20, 2003
Pathology
Room:B315**

27 Intestinal mucosal mast cell and morphological response to *Eimeria acervulina* isolates in broiler chickens. B. C. Morris*¹, H.D. Danforth², D.J. Caldwell³, and A.P. McElroy¹, ¹Virginia Tech, ²USDA/ARS/LPSI/PBEL, Beltsville, MD, ³Texas A&M University.

Immunovariability between coccidial species in commercial poultry rearing facilities has emerged as a potential problem for the prevention of coccidiosis. The host-pathogen interaction and intestinal immune responses

must be further investigated to understand immunity to and pathophysiology of *Eimeria* parasites in chickens. Five experiments were conducted to compare two isolates of *Eimeria acervulina* (EA), EA1 and EA2. In three experiments, commercial broiler chicks were divided into control (non-challenged) and EA1 or EA2 challenged (14 days of age) groups. Duodenal tissue samples were taken 6 days post-challenge (PC) to evaluate morphometric alterations and mucosal mast cell responses. The ratio of villus height to crypt depth was reduced in EA1 challenged birds when

compared to controls in all experiments, with EA2 challenged birds significantly different from controls in Experiment 3 only. Mast cells were significantly decreased in EA1 challenged birds in Experiment 1 only, with no differences in Experiments 2 or 3. Subjective observations of intestines from EA2 challenged birds were suggestive of an intestinal secretory response. In Experiment 4, tissues were taken for analysis from day 2 through day 6 PC. Villus atrophy and crypt hyperplasia were heightened on day 5 PC in both challenged groups. Mast cell counts were significantly altered on days 3 and 4 PC in EA1 challenged birds. In Experiment 5, EA2 oocysts were cleaned with 5.25% sodium hypochlorite to evaluate the possibility of a contaminant contributing to the pathogenesis of intestinal alterations. Villus heights and crypt depths were significantly altered in challenged birds, resulting in lower villus to crypt ratios, however, there were no differences in mast cell number. These data are indicative of differential host response and immunovariability between different isolates of the same *Eimeria* species and are suggestive of a role of mast cells in coccidiosis immunity.

Key Words: Eimeria, immunovariability, broilers, immunity, mast cell

28 Bacteriophage Attachment and Penetration to Host Organisms may not Influence Host Specificity. K. L. Guenther^{*1}, S. E. Higgins¹, B. M. Hargis¹, W. E. Huff², and L. A. Newberry¹, ¹University of Arkansas, ²PP&PSRU, ARS, USDA.

The ability of bacteriophages (phage) to effectively treat enteric bacterial infections has previously been demonstrated. Host-specificity is a desirable trait for bacteriophages as it offers an advantage for the preservation of beneficial flora. However, extreme host specificity implies the necessity for custom selection of phage for field application. Studies in our laboratory have confirmed the presence of phage:host specificity that is related to the phage:host cell ratio. It was noted that by increasing phage titer, lysis occurred in a broader range of host bacteria. In some bacterial species, a single 10x dilution made the difference between no lysis or complete lysis with no increase in plaque formation. Lack of plaque formation in alternate hosts suggests that lysis may be from without and not a result of actual phage replication. The ability of bacteriophage isolate (PHL-4) to attach and replicate in an alternative host, *Salmonella kentucky* NVSL 1271-94 (*SK*) was tested. PHL-4 was added to either the original host, *Salmonella enteritidis* PT 13A (*SE*) or *SK* in two experiments, and either provided adequate time to replicate, or removed from the host cell culture before replication could occur. In trial one PHL-4 was allowed to replicate for three passes at 1.5 hr in both hosts. PHL-4 only increased in titer with *SE* as a host, suggesting that PHL-4 was not capable of replicating in *SK*. In trial two, PHL-4 was in contact with each host for 10 min. Results indicated less phage were present in the *SK* vs. the *SE* suspension, suggesting that attachment of PHL-4 to *SK* was higher. Collectively these observations imply that failure of PHL-4 to replicate in the alternate host is not a factor of attachment or penetration. Apparent broad host specificity resulting from increasing the phage:host cell ratios may reflect mechanical cell death.

Key Words: Bacteriophage, Phage, Host Specificity, *Salmonella*

29 Identification of Alternative Host Bacteria for Amplification of Wild-Type Bacteriophages Isolated Using *Salmonella enteritidis*. L. R. Bielke^{*1}, S. E. Higgins¹, K. L. Guenther¹, G. Nava¹, and B. M. Hargis¹, ¹University of Arkansas.

Bacteriophages are viruses that exclusively infect bacteria. Generally, it is believed that these viruses will amplify in only a narrow host range of closely-related bacteria, a limiting factor for the use of bacteriophage as an alternative to antibiotic therapy. Presently, we have selected bacteriophages with the ability to infect more than one genus of bacteria. Bacteriophages were initially selected from a waste water treatment plant for ability to form plaques in *Salmonella enteritidis* PT13A (*SE*). For determination of host specificity, 44 combined isolates of bacteriophages were evaluated for ability to propagate in 35 individual non-pathogenic enteric bacterial isolates. Titer (pfu) of the pooled 44 bacteriophage was determined by incubation of serial dilutions of the bacteriophage mixture in tryptic soy agar (TSA) soft overlay with *SE*. This *SE*-selected bacteriophage mixture was filtered using a 0.2 μ m filter and combined with each individual bacterial isolate at a ratio of one part bacteriophage, three parts turbid bacterial broth culture, and five parts fresh tryptic soy broth. This mixture was incubated for 2h, sterile filtered, and recombined using the above ratio with fresh bacterial culture and media for a total of four serial passes. The pfu's of bacteriophages were then determined

using TSA soft overlay with *SE*. The entire procedure was repeated to confirm amplification with each of the same enteric isolates. Four different bacterial isolates, one *Klebsiella* and three different *Escherichia coli* isolates, successfully amplified some bacteriophage(s) from the *SE*-selected bacteriophage pool. Resulting plaques were then reisolated and serially passed in their respective enteric bacterial isolate. Amplification in each species was confirmed by the formation of increased pfu's in a TSA soft overlay with the enteric bacteria. These experiments suggest that bacteriophage host range is not limited to a specific genus. In the present studies, identified alternative hosts were limited to closely-related genera. Ongoing studies are evaluating the potential for more phylogenetically distant non-pathogenic isolates to support replication of *Salmonella*, which may allow for improved safety for bacteriophage application to commercial poultry.

Key Words: bacteriophage, poultry, *Salmonella enteritidis*, host range, enteric bacteria

30 Evaluation of a simple in vitro-selected probiotic consisting of nine non-pathogenic bacteria to prevent *Salmonella* infection in turkey poults. S.E. Higgins^{*1}, L.R. Bielke¹, G.M. Nava¹, K.L. Guenther¹, G.I. Tellez¹, L.A. Newberry¹, and B.M. Hargis¹, ¹University of Arkansas.

Effective probiotics or competitive exclusion cultures have been shown to accelerate development of normal microflora in chicks and poults, providing increased resistance to infection by some enteric bacterial pathogens. Our objective was to create a simple, safe, defined, air tolerant, and effective culture. We have previously selected bacteria isolated from the intestinal tract of normal chickens based on their ability to inhibit *Salmonella enteritidis* (*SE*) growth in vitro and demonstrated that a culture consisting of 24 organisms was efficacious in vivo. In the present experiments, seven members of the family *Enterobacteriaceae* and 2 lactic acid bacteria isolates, each capable of in vitro inhibition of *SE*, were selected and combined to form the putative probiotic. In experiment 1, day-of-hatch poults were orally gavaged with tryptic soy broth (control) or four different doses of the culture. Five treatment groups (20 birds per group) were treated by oral gavage and inclusion of the culture in the drinking water. The control was gavaged with tryptic soy broth, and received no probiotic in the water. The group receiving the highest dose received 1.68×10^8 cfu by oral gavage and 1.1×10^5 cfu/ml in the drinking water for four days. The other two treatment groups received serial 100-fold dilutions of the highest dose. All poults were challenged 48 h after placement with 2.5×10^4 cfu *SE*. Cecal tonsils were sampled 48 h post-challenge and cultured for *SE* recovery. *SE* was recovered with a lower ($p < .05$) incidence from the group that received 1.68×10^4 cfu by gavage and 11.2 cfu/mL in the drinking water (25%) as compared to controls (95% positive). In experiment 2, four treatment groups (4 groups total, 30 birds per group) were treated with the culture and challenged 48 h later as in experiment 1. The highest dose was 4.16×10^5 cfu by gavage and 600 cfu/ per mL of culture in the drinking water daily, and the other two groups received 10-fold serial dilutions of the highest dose. Treatment with 4.16×10^3 cfu by gavage and 6 cfu/mL of water caused reduced ($p < .05$) recovery of *SE* (7%) as compared to controls (50%). These data suggest that relatively simple defined cultures consisting of air-tolerant bacteria can inhibit *Salmonella* colonization in poults.

Key Words: competitive exclusion, *Salmonella enteritidis*, poultry

31 Effect of spent media collected from a PREEMPTTM continuous flow culture on *Salmonella* Typhimurium virulence expression. M.M. Kunding¹, R.W. Moore^{*1}, I.B. Zabala Diaz¹, J.A. Byrd², D.J. Nisbet², and S.C. Ricke¹, ¹Dept. of Poultry Science, Texas A&M University, College Station, TX, ²USDA-ARS, SPARC, Food and Feed Safety Research Unit, College Station, TX.

Salmonella is an important human enteric pathogen, responsible for causing severe gastroenteritis in infants and adults. PREEMPTTM is an FDA-approved competitive exclusion culture developed for chicks to control *Salmonella* colonization in the gut. Bacterial virulence is regulated in response to a combination of environmental stimuli and involves a coordinated genetic response. Therefore it is important to understand the relevant features of the host (and its indigenous associated microflora) that affect *Salmonella* development of virulence *in vivo*. Our objective in this study is to determine virulence expression of *Salmonella* Typhimurium (ST) when exposed to PREEMPTTM spent media. A frozen 100 μ l aliquot of ST was inoculated into 10 mL Tryptic Soy Broth (TSB)

and incubated at 37°C for 24 h. This procedure was repeated 2 times, transferring 100 µl aliquots of the culture each time. The final culture was collected and centrifuged at 4000xg for 10 min and then filter sterilized through a 0.2µm membrane. PREEMPTTM spent media was from an anaerobic continuous flow fermentation culture apparatus containing a commercial preparation of PREEMPTTM and prepared similar to the ST spent media. The filtered spent media was then stored at 4°C until needed. To assess virulence gene response of ST, a microtiter Miller assay was performed. There was a significance difference in expression of *hilA* between ST spent media when compared to BHI (Average MU values 1221.8197.8 and 32.110.0 respectively $p>0.05$). However, there was no significance difference in expression of *hilA* between PREEMPTTM and BHI Media (Average MU values 477.3 102.8 and 32.110.0 respectively). Exposure of ST cells to ST spent media resulted in 2.5 fold increase in values of β -galactosidase than when exposed to PREEMPTTM spent media (Average MU values 1221.8197.8 and 477.3 102.8 respectively, $p>0.05$). In conclusion, presence of PREEMPTTM cultures does not appear to stimulate *hilA* expression, and that probiotic bacteria may decrease the expression of virulence in ST.

Key Words: PREEMPTTM, Probiotic, *hilA*, *Salmonella*, Virulence

32 Organophosphate toxicosis in chickens: A case report. R. R. Hubbard*, F. D. Clark, and J. England, University of Arkansas, Center of Excellence for Poultry Science.

Two cases of organophosphate toxicity were diagnosed at the University of Arkansas Poultry Science Department poultry research farm in the spring of 2002. In both cases the birds were treated with the organophosphate RaVap[®] for Northern Fowl Mites (*Ornithonyssus sylvarium*) infestations. In case one, 57 birds died and 7 were severely affected by organophosphate toxicosis three days after treatment. In case two, four male Australorps died after treatment and another seven developed clinical symptoms. In both cases, surviving birds were treated with atropine sulfate. A total of 61 birds died and 13 were treated successfully with atropine sulphate. It was noted that the most severe mortality was seen in the purebred Australorp and Australorp-Smyth line crosses.

33 Effect of various combinations of alfalfa and standard layer diet on susceptibility of laying hens to *Salmonella enteritidis* during forced molt. R. W. Moore*¹, L. F. Kubena², K. L. Medvedev¹, C. L. Woodward¹, M. R. Burnham², D. J. Nisbet², and S. C. Ricke¹, ¹Texas A&M University, College Station, TX, ²USDA-ARS, SPARC, Food and Feed Safety Research Unit, College Station, TX.

Feed deprivation is commonly used by the poultry industry to induce molting and stimulate multiple egg-laying cycles. However, feed deprivation has been observed to increase susceptibility to *Salmonella* infections. Previously, we have reported that alfalfa meal induces molting similar to feed deprivation, while reducing colonization of *Salmonella*. The present study was designed to investigate the efficacy of combined alfalfa and layer diets on *Salmonella* colonization. Leghorn hens over 50 wk of age were divided into groups of 12 hens and placed in individual laying cages. One wk prior to dietary changes, hens were put on an 8-h light and 16 h-dark photoperiod that continued for the 9-day experiment. Hens were challenged orally with 10⁴ cfu of *Salmonella enteritidis* (SE) on the fourth day of treatment, and cultured for SE at the termination of the study. Treatments were a 100% alfalfa meal diet (100A), a 90% alfalfa meal/10% standard commercial layer diet (90A), a 70% alfalfa meal/30% standard commercial layer diet (70A), non-fed hens (molted, M), and 100% standard commercial layer diet (non-molted, NM). Incidence and enumeration of SE in crops of hens fed 90A and NM were non-detectable and significantly ($P>0.05$) reduced from that of M (1.0 log cfu, 42% positive). SE counts from ceca were reduced ($P>0.05$) in 100A (0.4 log cfu), 70A (0.4 log cfu), and NM (no SE detected) as compared to M (2.8 log cfu); however, only NM (0%) had a reduced ($P>0.05$) incidence of cecal SE as compared to M (M=58.3%). Organ incidence of SE was reduced ($P>0.05$) in ovaries (100A=0%, 70A=0%), liver (100A=0%, 90A=16.7%, 70A=16.7%, and NM=0%), and spleen (70A=0% and NM=0%) as compared to M (41.7%, 58.3%, 33.3%, respectively). Ovary weights as a percent of body weight were similarly reduced ($P>0.05$) in 100A, 90A, 70A, and M from NM (0.46 g, 0.45 g, 0.44 g, 0.46 g, and 2.57 g respectively). Body weight losses were significantly ($P>0.05$) greater in M (25.8%) and 100A (23.2%) than in NM (1.9%). Treatments 90A (20.9%) and 70A (17.8%) had body weight losses that were significantly less than M and more than NM. These data suggest that alfalfa can potentially be

combined with layer ration to limit SE infection and still induce a molt comparable to feed withdrawal.

Key Words: *Salmonella enteritidis*, Molting, Laying Hen, Alfalfa

34 Reproductive organ characteristics of commercial layer hens inoculated with F-strain *Mycoplasma gallisepticum* and housed in a caged layer facility. M. R. Burnham*¹, E. D. Peebles², and S. L. Branton³, ¹USDA-ARS, SPARC, Food and Feed Safety Research Unit, College Station, Texas 77845, ²Department of Poultry Science, Mississippi State University, Mississippi State, Mississippi 39762, ³USDA-ARS, SCPRL, Mississippi State, Mississippi 39762.

Age of inoculation with F-strain *Mycoplasma gallisepticum* (FMG) and its subsequent effects on reproductive and digestive organ characteristics in commercial egg laying hens were investigated. At 12 wk of age, sixty layer hens were randomly assigned to individual cages in each of two enclosed ends of a temperature regulated caged layer facility and were either FMG (treated)- or sham (control)- inoculated. At 22 wk of age an additional sixty layer hens were randomly assigned to individual cages in each enclosed end of the same facility and were likewise FMG- or sham-inoculated. Each inoculation period group on each side of the house (treatment) were replicated 6 times, with ten hens in each replication. Birds were euthanized by cervical dislocation and their organs were harvested at trial termination (60 wk of age). Variables investigated included liver weight and composition, ovary weight and follicular hierarchy, segmental weights, lengths, and histologies of the intestine and oviduct, and percentage incidence of Fatty Liver Hemorrhagic Syndrome. Relative liver weight, moisture, and lipid contents were significantly decreased in birds inoculated with FMG at 22 wk of age when compared to 22 wk sham-inoculated (FMG-free) birds. Relative (% of BW) oviduct, infundibulum, isthmus, and uterus weights were significantly increased in birds inoculated with FMG at 22 wk of age when compared to 22 wk sham-inoculated (FMG-free) birds. No reproductive organ characteristics were significantly affected in birds inoculated with FMG at 12 wk of age in comparison to 12 wk sham inoculated controls in the caged layer facility. These data suggest that timing of an FMG-inoculation affects the liver and reproductive organs of commercial layers with subsequent effects on their egg production. More specifically, there were associated effects between the organ characteristics and performance parameters of hens inoculated with FMG at 22 wk of age and when housed in a caged layer facility.

Key Words: Layer, Lipid, Liver, *Mycoplasma gallisepticum*, Reproductive tract

35 Changes in live performance of broiler chickens raised on built-up litter for eight production cycles following a coccidiosis challenge. E. Schildknecht, L. Rakebrand, L. Jensen, and J. Skinner*, Alpharma Inc., Fort Lee, NJ.

A study was conducted to evaluate changes in coccidia sensitivity during 8 sequential broiler chicken grow-out cycles on built-up litter. The following is a discussion of the performance of the chickens during each production cycle; however, changes in coccidia sensitivity will be presented in a subsequent paper. The coccidiosis control programs for the 8 cycles were as follows: Phase 1 alternate use of chemical anticoccidials (starter feeds) to salinomycin (grower feeds) programs (4 cycles), Phase 2 use of a coccidiosis vaccine for all birds (1 cycle) and Phase 3 alternate use of chemical anticoccidials (starter) to salinomycin (grower) programs (3 cycles). There were also pens of birds that received a coccidiosis vaccine for all 8 cycles. During Phase 3, a treatment receiving salinomycin in both the starter and grower feed was added. Chemical coccidiostats used included diclazuril (1 ppm), robenidine (33 ppm), nicarbazin (125 ppm), and zoalene (125 ppm). When included, salinomycin was at 55 ppm in feed during Phase 1 and 50 ppm in Phase 3. During Phase 1, Cycle 1 of the study all birds were challenged with recent (2001) field strains of *Eimeria maxima* and *E. acervulina*. Weight gains and feed conversion ratios of the broilers varied over time and in response to the different anticoccidial programs. The nicarbazin program was more suited to the cooler seasons. The anticoccidial program utilized on flocks prior to the use of a coccidiosis vaccine on a subsequent flock appeared to affect the performance of birds receiving the vaccine. Different anticoccidial programs following the use of a coccidiosis vaccine affected performance of broiler chickens differently. Within a coccidiosis control program, each chemical should be considered based on when it is to be used. Effects of previous anticoccidial programs on the current program and future

programs should be considered to maximize broiler weight gain and/or feed conversion.

Key Words: Chickens, Coccidiosis, Weight gain, Feed Conversion, Anticoccidial

36 Changes in anticoccidial sensitivity profiles of coccidia from broiler chickens raised on built-up litter for eight production cycles following a coccidiosis challenge. E. Schildknecht, L. Rakebrand, L. Jensen, and J. Skinner*, *Alpharma Inc., Fort Lee, NJ.*

A study was conducted to evaluate changes in sensitivity to anticoccidials of coccidia obtained from chickens during eight sequential grow-out cycles on built-up litter. The coccidiosis control programs for the eight cycles were as follows: Phase 1 alternate use of chemical anticoccidials (starter feeds) to salinomycin (SAL) in grower feeds (4 cycles), Phase 2 use of a coccidiosis vaccine (1 cycle) and Phase 3 subsequent use of alternate chemical anticoccidials (starter) to SAL in grower feeds (3 cycles). Additionally, there were pens of birds that were treated only with a coccidiosis vaccine for all eight grow-out cycles. In Phase 3, a treatment including SAL in the starter and grower feed was added. Chemical coccidiostats used included diclazuril (1 ppm), robenidine (33 ppm), nicarbazin (125 ppm), and zoalene (125 ppm). When included in the feed, SAL was at 55 and 50 ppm in Phase 1 and 3, respectively. During the first grow-out, all birds were challenged with recent (2001) field strains of *Eimeria maxima* and *E. acervulina*. An anticoccidial sensitivity test (AST) was conducted to determine their sensitivity to the drugs used in this study. AST were conducted similar to those reported by Schildknecht et al. (1980 Poultry Sci. 59:1145-1147) after the 3, 5 and 8 grow-outs using oocysts collected from litter at the selected grow-out periods. These data indicate that the coccidia retained their sensitivity to some drugs longer than others. The use of the coccidiosis vaccine did not appear to replace the field strains with a more drug sensitive population of coccidia. For some variables

there was a statistical improvement in anticoccidial efficiency; however, this improvement relative to the challenged controls was usually observed with a more pathogenic challenge. The use of a coccidiosis vaccine may introduce new coccidia into the production environment.

Key Words: Chickens, Coccidiosis, Sensitivity profiles, Anticoccidials, Oocysts

37 Anomaly increase among broiler chicks documented via computerized database. R.W. Keirs*¹, ¹Mississippi State University.

A computerized database constructed to collect viable information with directed consistency, broadly over extended time can be invaluable in depicting important changes. Hatching Efficiency Analysis System (HEAS) was so constructed and utilized and prior to 1998 the data base included at least 24 broiler complexes located in 9 U.S. broiler production states, 5,179 individual breeder flock records representing 55,581,571 eggs set of which over 500,000 plus hatching egg residue were categorized. Anomalies were @0.39%. In 1998 a specific HEAS study was completed in Mississippi (MS) that included all broiler complexes (13), all breeder flocks, and all hatcheries. HEAS data has been collated for 7/13 complexes. This includes 160 breeder flocks, and 17,596 residue. Anomalies were 0.64% of all eggs set. In April 1999 in a typical two-day HEAS study of a single hatchery that included all their breeder flocks, anomalies increased to 2% of all eggs set. A HEAS study was initiated among seven selected MS complexes to compare their current anomaly status to their previous 1998 HEAS database. This included 181 breeder flock records, 19,596 egg residue information with anomalies representing 1.77% of all eggs set. Anomalies had increased 176% over their collective 1998 HEAS data base value. A computerized data base has depicted a significant rise in broiler chick embryonic anomalies.

Key Words: Anomaly, Embryonic, Broiler, Chick, Database

Monday, January 20, 2003 Physiology Room:B316

38 Myofiber number in Pectoralis muscle of different chicken lines. G. N. Scheuermann*, S. F. Bilgili, S. Tuzun, and D. Mulvaney, *Auburn University, Auburn, AL - USA.*

Muscle growth is dependent upon myofiber numbers (MFN), established embryonically, and post-hatch enlargement in myofiber size. The objective of this study was to evaluate if MFN is related to muscularity of different lines of chickens. Day-old sexed chicks from three genotypes [two commercial broiler strain-crosses differing in breast yield (HY=high yield; LY=Low yield), and one leghorn] were reared separately, based on genotype and gender, to 35 d of age. A common commercial broiler feed was provided *ad libitum*, and lighting was used continuously. At Days 7, 14, 21, and 35, 24 chickens per genotype were sampled to assess body weight (BW) and Pectoralis muscle weight. Breast muscles sampled at Day 7 were sectioned transversally to the myofiber direction, traced for estimation of the total breast muscle area, frozen, sectioned, and stained for estimation of myofiber density (MFD=number of myofibers in a given cross-sectional area) and total apparent MFN. Statistical analysis for BW, breast muscle weight and yield was performed combining all the ages. A clear age effect ($P < 0.01$) was observed for all variables. As expected, Leghorns had lower ($P < 0.05$) BW and breast weight and yield than broilers, while HY and LY broilers differed ($P < 0.05$) only in breast yield. Analysis at Day 7 showed that Leghorns had about 45% higher MFD than the broiler strains ($P < 0.01$), indicating reduced cross-sectional area of the myofibers, but no MFD difference was observed between HY and LY. Besides the higher MFD of Leghorns, total apparent MFN of the broiler strains was more than twice that of Leghorns due to the broilers increased cross-sectional area of the breast muscles. HY broilers had 11% higher MFN than LY strain. MFN was correlated ($P < 0.01$) to BW ($r = .92$), breast weight ($r = .94$), and breast yield ($r = .87$), while MFD showed a negative correlation ($P < 0.01$) with the same variables ($r = -.84$ to $-.87$). However, exclusion of Leghorn genotype from the analysis reduced the correlation of MFN to 0.51 for BW ($P = .091$) and 0.68 for breast weight ($P = .016$), with no correlation being observed between MFN and breast yield. This study showed that broilers have a

higher MFN in the breast muscles than Leghorn-type chickens, and that high breast yield of broiler strains may be due to increased MFN.

Key Words: Chickens, Myofiber number, Pectoralis

39 Effect of alfalfa and feed deprivation molting techniques on various serum chemistry parameters in commercial laying hens. K Medvedev*¹, R Moore¹, C Woodward¹, D Landers², Z Howard¹, A Byrd³, J McReynolds³, L Kubena³, D Nisbet³, and S Ricke¹, ¹Department of poultry science, Texas A&M University, ²College of Veterinary Medicine, Texas A&M University, ³USDA-ARS SPARC, Food and Feed Safety Research Unit.

Alternatives to feed deprivation for the purpose of inducing molt in commercial laying hens have been developed over the past several years. Providing *ad libitum* access to alfalfa, a fibrous, plant based feedstuff, is one of many alternatives that show promising results in its ability to induce an effective molt. In order to further quantify the efficacy of such an alternative, it is important to ascertain what possible physiological implications could result from the use of the diet. In this study, two groups of hens were molted by either feed deprivation or alfalfa for a period of nine days. Body and ovarian weights were obtained. Blood was taken from a subset of hens daily until day 7 of the molt, including a day 0 control. Parameters measured included: ketone bodies, glucose, triglycerides, cholesterol, phosphorus, calcium, magnesium, uric acid, and total protein. Hens fed alfalfa had significantly lower levels of cholesterol, triglycerides, and ketone bodies than hens that were feed deprived ($p \leq 0.05$). Hens fed alfalfa also had significantly higher levels of uric acid and glucose than hens that were feed deprived ($p \leq 0.05$). No significant differences were noted between the two treatment groups for levels of total protein, magnesium, calcium, and phosphorus; however, the levels for magnesium, calcium, and phosphorus were significantly different than the day 0 control ($p \leq 0.05$). Hens fed alfalfa lost significantly less weight than hens that were feed deprived ($p \leq 0.05$). This, however, did not affect ovarian weights as the molting treatments did not differ significantly ($p \geq 0.05$). Therefore, while the serum chemistry parameters

indicate that alfalfa molted hens differ physiologically from feed deprived hens, the ovarian weights suggest that these differences do not alter the hen's capacity to undergo a complete molt.

Key Words: Alfalfa, Molt, Serum

40 Physiological stress in laying hens. J. A. Odhiambo, J.P. Thaxton, Y.V. Vizzier-Thaxton, and W.L. Dodson, *Mississippi State University.*

Single Comb White Leghorn birds that had been in egg production for 36 wk were used. Hens were housed in commercial type cages and fed a standard layer ration and water ad libitum. Two separate experiments were conducted. In Experiment 1, 128 hens were used. One half of them (64) received ACTH 8 IU/kg BW/d for 7d via osmotic pumps, and controls received saline. Sixteen hens on each of the treatments were bled then killed at 2, 4, 6, and 8d post-pump implantation. Experiment 2 was a replicate of Experiment 1 with two exceptions. Six ACTH and 6 control hens were sacrificed at each time interval, and these intervals were 7, 14, and 21d post-implantation. Blood levels of corticosterone (CS), glucose (GLU), cholesterol (CHOL), triglycerides (TRI), and high-density lipoproteins (HDL) were determined as well as liver (LW) and spleen (SW) weights. ACTH increased levels of CS, GLU, CHOL, TRI, HDL and LW, while SP was decreased. This is the first physiological profile of adult birds and the stress changes agree with those of broilers.

Key Words: Layer, Stress, Adrenocorticotropin, ACTH

41 Immune Response of Chickens to BSA Following Drinking Water Administration. K.A. Ameiss^{*1}, A. Barri¹, A.P. McElroy², H.D. Danforth³, J.L. McReynolds¹, and D.J. Caldwell¹, ¹Texas A&M University, College Station, TX, ²Virginia Tech, Blacksburg, VA, ³USDA/ARS/LPSI/PBEL, Beltsville, MD.

Oral administration of protein antigen in solution leads to tolerance in most mammals but has been shown to be immunogenic in the chicken. Previous studies, including several experiments conducted by our laboratory, have shown that oral administration of BSA for six consecutive days is fully immunogenic in the chicken. While these studies administered antigen in a single daily bolus by gavage, the present investigation was conducted to determine immune responsiveness in SWCL chicks when BSA was administered ad libitum in the drinking water as compared to IP injection or oral gavage. IP immunization consisted of administering 0.5 ml injections of 10 ug/ml BSA-10mg alum to chicks on days 1 and 7. IP control groups received 0.5 ml injections of 10 mg alum only. Oral gavage consisted of administering a daily per os bolus of 1 ml of 25 mg BSA/ml to chicks between 12 and 18 days of age. Drinking water administration consisted of *ad libitum* access to drinking water containing 0.14 mg BSA/ml between days 12 and 18. Antibody production was measured by antigen specific indirect ELISA in serum, distal ileum, cecal tonsil, and distal ceca samples 14 days after the first immunization. Mean absorbance values (Abs) for serum IgG revealed that the response to BSA administered *ad libitum* by drinking water (Abs = .618) was significantly higher ($P < .05$) than BSA administered by oral gavage (Abs = .220) or controls (Abs = .163), but not IP administration (Abs = .480). Similar results were seen in intestinal antibody secretion in the distal ileum and cecal tonsils, where drinking water administration (Abs = .129 and Abs = .124) differed ($P < .05$) from gavage (Abs = .032 and Abs = .047) and controls (Abs = .019 and Abs = .032), but not IP (Abs = .179 and Abs = .117). These data indicate that chickens respond to antigens administered orally in a manner distinct from mammals and support drinking water as a route for immunization or vaccine administration.

Key Words: Chicken, BSA, Oral Immunization, Antibody

42 The effects of storage time on yolk protein banding patterns and interior egg quality. A. J. Kelley* and S. G. Birkhold, *Texas A&M University, College Station, Texas.*

Vitelline membrane strength plays a major role in regulating contamination of albumen by yolk during separation and is important to food safety. Two trials were carried out to determine if a relationship exists between protein banding patterns and interior egg quality. In the first trial, 21 eggs were gathered on day 0 and stored at refrigeration temperature (4C). Three eggs per storage time were used on day 0, 7, 14, 21, 28, 35, and 42. The vitelline membrane from each egg was isolated, and then rolled on a wet paper towel to remove adhering albumen and aid in separating inner and outer membranes. The whole membrane was

placed into water and divided into two sections. The first section was a whole membrane sample and the other was separated by forceps into inner and outer membrane samples. The three sections were then dissolved in 1% SDS/70 mM Tris/HCl, pH 6.8. Protein concentration was determined using Lowry's and proteins separated on 4-20% gel gradient by SDS-PAGE. Protein banding patterns were analyzed using the Bio-Rad Multi-Analyst Densitometer. Disappearances in the protein bands occurred between 116 kDa and 45 kDa. Interior egg quality was evaluated based on Haugh unit, yolk index, albumen height, albumen pH, yolk pH, and vitelline membrane strength using a compression anvil. One hundred forty eggs were gathered on day 0 and stored at refrigeration temperature (4C). Twenty eggs per storage time were evaluated on day 0, 7, 14, 21, 28, 35, and 42. Two different treatments were used on the yolk when evaluating rupture strength: 10 egg yolks with inner thin albumen layer, and 10 egg yolks rolled on wet paper towel to remove inner thin albumen layer. A decline in interior egg quality and vitelline membrane strength was seen over storage time.

Key Words: Vitelline membrane, Interior egg quality, yolk rupture strength

43 A comparison of bone mineral density in live broilers and White Leghorns. M. A. Schreiwes^{*1}, J. I. Orban², M. C. Ledur³, D. E. Moody¹, and P. Y. Hester¹, ¹Purdue University, W. Lafayette, IN, ²Southern University, Shreveport, LA, ³Embrapa Swine and Poultry Research Center, Concordia, SC, Brazil.

Densitometry is being used in our laboratory as a noninvasive tool to evaluate bone integrity of live birds. The objective of the current study was to monitor bone mineral density (BMD) among White Leghorns during the life cycle in comparison to bone integrity of broilers at the same ages. Densitometric scans were conducted on live, unanesthetized females from 15 to 65 wk of age at 10 wk intervals using a Norland pDexa X-ray bone densitometer. The BMD of the left leg (tibia and fibula) was determined in White Leghorn (n = 31 to 35) and broiler females (n = 19 to 47). Similar measurements were conducted on the tibia of male broilers at 15 and 25 wk of age. Using the mixed model procedure of SAS, an analysis of covariance (ANCOVA) with repeated measurements (15 to 65 wk of age) was conducted to compare the female strains (Cobb x Cobb broiler vs. White Leghorn), and another ANCOVA with repeated measurements at 15 and 25 wk of age was used to compare the tibial BMD of male (n = 34 to 44) and female broilers. Body weight was used as a covariate in both analyses. The BMD of the tibia of the female birds increased from 15 to 65 wk of age with the BMD of the broiler tibia increasing at a greater rate than the Leghorn tibia (strain by age interaction, $P < 0.0001$). There were no statistical differences in the BMD of the tibia of the male and female broilers at 15 and 25 wk of age. Coefficients of variation for BMD were greater than 10% after 25 wk of age for both the Leghorn and the broiler females. This variability among hens in BMD suggests that there is genetic potential to improve skeletal integrity of birds through the use of densitometry. In addition, there appears to be adequate divergence in BMD between the broiler and egg strains to create a resource population for quantitative trait loci (QTL) studies on skeletal integrity. Research supported by NRI Competitive Grants Program No. 2001-02426 and SCRP-USDA No. PL 95-113.

Key Words: Bone Mineral Density, Broiler, White Leghorn

44 Site-specific Defects in Electron Transport in Duodenal Mitochondria is Associated with Low Feed Efficiency in Broiler Breeder Males. C. Ojano-Dirain^{*1}, M. Iqbal¹, D. Cawthon¹, S. Swonger¹, T. Wing², M. Cooper², and W. Bottje¹, ¹Department of Poultry Science, Center of Excellence for Poultry Science, University of Arkansas, ²Cobb-Vantress Inc., Siloam Springs, AR 72761.

This study was conducted to assess relationships between feed efficiency (FE, gain to feed) and duodenal mitochondrial function and oxygen radical production in broiler breeder males. At 7 wk, FE was determined and birds with high FE (0.79 ± 0.01 , n=9) and low FE (0.63 ± 0.02 , n=9) were selected. Duodenal mucosal mitochondria were isolated to measure function and assess site-specific defects in electron transport. The respiratory control ratio (RCR, an index of respiratory chain coupling) was greater after sequential additions of ADP in high FE duodenal mitochondria provided succinate (FADH-linked energy substrate), but there were no differences in the ADP to oxygen ratio (ADP:O; an index of oxidative phosphorylation) between groups. With malate (NADH-linked

substrate), there were no differences in RCR between groups and the ADP:O ratio was improved by sequential additions of ADP in low FE mitochondria. Sites of electron leak were assessed by measuring H₂O₂ production by dichlorofluorescein fluorescence following site-specific inhibition of electron transport. Basal electron leak was greater in low FE Mito provided either energy substrate. Electron leak in low FE mitochondria provided succinate was elevated by Complex II inhibition and marginally increased ($P < 0.10$) by Complex I inhibition in low FE Mito provided malate. These results indicate that differences in function in low and high FE mitochondria were substrate dependent, and that oxidative phosphorylation was equal or superior in low FE compared to high FE mitochondria. Importantly, basal electron leak was greater in low FE duodenal mitochondria apparently due to site-specific defects in electron transport at Complex I and II. Thus, low FE duodenal cells may experience greater oxidative stress from mitochondrial oxygen radical production that could oxidize critical cellular components and may be important in the phenotypic expression of FE in broilers. Supported in part by USDA-NRI #2001-03443 to W. Bottje

Key Words: Broilers, Feed efficiency, Duodenum mitochondria, Electron leak, RCR

45 Estrogen receptor alpha (ER α) and beta (ER β) in chicken skeletal muscle: Mediators of myoprotection? V. E. Cooke*, A. J. Carlisle, M. A. Mitchell, R. T. Talbot, and T. Boswell, Roslin Institute, Roslin, UK.

Genetic selection for improved production traits in broiler chickens has resulted in increased susceptibility to spontaneous and stress induced myopathies. In female birds the pathology is decreased at sexual maturity. Estradiol administration in immature birds mimics these effects and the action is inhibited by administration of tamoxifen, an estrogen receptor (ER) modulator. It is therefore proposed that the myoprotective effect of estradiol in birds, unlike mammals involves ER occupation. The present study was undertaken to determine if ER α and ER β isoforms are expressed in chicken skeletal muscle. Studies were performed on 30 week old female ISA Brown hens. The birds had been grown on a 14h light:10h dark regime and 21°C/55%RH and fed a commercial layer ration. Oligonucleotide primers were designed from complementary DNA (cDNA) sequences of chicken ER α (accession number XO3805) and ER β (accession number AB036414) using current DNA databases. 100 mg of liver (control tissue), and breast (Pectoralis major) and leg (Iliotibialis lateralis) muscle samples were homogenised and digested. Total RNA was isolated and content was assessed by absorbance at 260nm and purity by A260/A280 ratios. Messenger RNA was reverse transcribed to cDNA at 37°C and amplified using the polymerase chain reaction (PCR). PCR products were visualised on a 3% agarose gel containing ethidium bromide. Amplified fragments were ligated into pBluescript II SK+ plasmid, transformed into E. coli XL1-Blue strain, and sequenced. The deciphered sequences were analysed by the Staden package to check homology. The primers produced fragments of the size expected for ER α (260 bases) and ER β (350 bases) in Pectoralis major and Iliotibialis lateralis. Sequencing of PCR products indicated 100% homology for ER α and ER β against the recognised 260 and 350 base characters respectively. This is the first demonstration of ER α and ER β expression in chicken skeletal muscle. The full functional significance of these receptors awaits further elucidation but it is proposed that the apparent myoprotective actions of estrogens in birds may uniquely involve receptor mediation and regulation of gene expression, in contrast to mammalian models.

Key Words: Myopathy, Estrogen receptor, Estrogen

46 Stress and Reproduction in Laying Hens. J. P. Thaxton*, J. Odiambo, Y. Vizzier-Thaxton, and W.L. Dodson, Mississippi State University.

Laying hens housed in commercial type cages were used in two separate experiments. In Experiment 1, 128 hens were randomized into two groups of equal numbers. One group received ACTH (8 IU/kg BW/d for 7d) and the other group was controls that received an equivalent amount of distilled water. ACTH and water were delivered continuously via osmotic pumps that were surgically implanted on each hen. Sixteen hens from each of the treatments were killed at 2, 4, 6 and 8 d post-pump implantations. Experiment 2 was a replicate of Experiment 1 with two exceptions. Six ACTH and 6 control hens were killed at each time interval and these intervals were 7, 14 and 21d post-implantation. Body, liver, spleen, heart, oviduct, fat pad, and complete intestine weights were

determined on each hen of both experiments at each time interval. ACTH treatment increased BW and concomitantly decreased spleen and oviduct weights. Weights of the spleen, heart, fat pad and complete intestine were unaffected by ACTH.

Key Words: Stress, ACTH, Layers, Body Weight, Organ Weight

47 Age of F-strain *Mycoplasma gallisepticum* inoculation affects the lipoprotein profiles of commercial layer hens housed in a caged layer facility. M. R. Burnham*¹, E. D. Peebles², R. L. Walzem³, S. L. Branton⁴, and P. D. Gerard⁵, ¹USDA-ARS, SPARC, Food and Feed Safety Research Unit, College Station, Texas 77845, ²Department of Poultry Science, Mississippi State University, Mississippi State, Mississippi 39762, ³Department of Animal Science, Texas A&M University, College Station, Texas 77843, ⁴USDA-ARS, SCPRL, Mississippi State, Mississippi 39762, ⁵Department of Ag. Info. Science, Mississippi State University, Mississippi State, Mississippi 39762.

Age of inoculation with F-strain *Mycoplasma gallisepticum* (FMG) and its subsequent effects on serum lipoprotein profiles in commercial egg laying hens were investigated during a 60 wk trial. At 12 wk of age, sixty layer hens were randomly assigned to individual cages in each of two enclosed ends of a temperature regulated caged layer facility and were either FMG (treated)- or sham (control)- inoculated. At 22 wk of age an additional sixty layer hens were randomly assigned to individual cages in each enclosed end of the same facility and were likewise FMG- or sham-inoculated. Each inoculation period group on each side of the house (treatment) were replicated 6 times, with ten hens in each replication. Blood samples were collected at 44 and 58 wk from 2 tagged birds within each replication. Variables determined included serum VLDL particle diameter distributions (10th, 50th, and 90th population percentiles), mean VLDL particle diameter (MA), and percentage of serum cholesterol (SCH) recovered in VLDL, LDL, and HDL. Particle diameter within each population percentile and MA were significantly increased in 12 wk treated birds when compared to 12 wk control birds, and also were significantly decreased in 22 wk treated birds when compared to 22 wk control birds. These same variables were significantly decreased in birds inoculated with FMG at 22 wk when compared to those inoculated with FMG at 12 wk. At 58 wk of age, percentage of SCH recovered in VLDL was significantly decreased in birds inoculated with FMG at 12 wk as compared to 12 wk sham-inoculated controls; however, percentage of SCH recovered from LDL was significantly increased in birds inoculated with FMG at 12 wk as compared to 12 wk sham-inoculated controls. These data suggest that FMG inoculation (12 and 22 wk) and timing of an FMG-inoculation affect yolk formation through liver lipoprotein metabolism with subsequent effects on egg production. Previous reports indicated that the characteristics of lipoproteins from birds housed in fiberglass isolation units were not significantly affected by FMG-inoculation. However, higher degrees of physiological stress experienced by hens in a caged layer facility may exacerbate the effects of FMG-inoculation and lead to changes in lipoprotein synthesis.

Key Words: Layer, Lipoproteins, *Mycoplasma gallisepticum*, VLDL, LDL, HDL

48 Associations Among Liver Glycogen Content and Various Physiological Parameters in Broiler Chicks Subjected to Delayed Brooding Placement. E.D. Peebles*, R.W. Keirs, L.W. Bennett, P.D. Gerard, and S.K. Whitmarsh, Mississippi State University, Mississippi State, MS .

The time dependent changes of liver glycogen content and its correlation with other physiological parameters between 6 and 72 h posthatch in broiler chicks from young (29-wk-old) breeders were assessed. Other physiological parameters determined included BW, rectal temperature (RT), hematocrit, serum refractive index (RI), serum glucose, relative liver weight, and kidney pathology and body fat loss scores. After hatch (0 h), chicks were held for 6 h in a commercial hatchery prior to a 2 h transport to a laboratory brooding facility. Therefore, chicks were deprived of feed and water through 8 h posthatch, but from that time through 72 h were brooded and provided feed and water *ad libitum*. Liver glycogen content increased through 48 h, with a subsequent decrease between 48 and 72 h. At 6 h posthatch, there were no significant correlations between any parameters. Liver glycogen content was not correlated with any other parameter between 24 and 72 h posthatch. However, between 24 and 72 h, BW was positively correlated with RT

and both were negatively correlated with hematocrit and RI. Body fat loss score was also negatively correlated with RT and serum glucose level. Furthermore, hematocrit was negatively correlated with serum glucose, but positively correlated with body fat loss score. Feed and water deprivation through 8 h posthatch had no immediate effect on liver glycogen content. However, liver glycogen decreased by 72 h posthatch while feed and water were being provided. Nevertheless, increased hematocrit, RI, and body fat loss coincided with slower growth and decreased RT and serum glucose levels between 24 and 72 h posthatch.

Key Words: Broiler, Brooding, Chick, Glycogen, Liver

49 Association of mitochondrial dysfunction with feed efficiency in male broilers within single genetic line: A proteomics approach. M. Iqbal^{*1}, N. Pumford¹, K. Lassiter¹, Z. X. Tang¹, T. Wing², M. Cooper², and W. G. Bottje¹, ¹*Department of Poultry Science, University of Arkansas, Fayetteville, 72701*, ²*Cobb-Vantress, Inc., Siloam Spring, AR 72761*.

Earlier studies have shown an increased production of H₂O₂ at Complex I and III (Bottje et al., *Poult. Sci.* 81:546-555, 2002) and protein carbonyl levels, a marker of protein oxidations, (Pumford et al., *Poult. Sci.* 81 (1): 79, 2002) in breast muscle mitochondria of broiler breeder males with lower feed efficiency. The objectives of this study were to determine the effects of low or high feed efficiency (LFE, HFE) on; a) the activities of various electron transport chain (ETC) complexes, b) ubiquitin conjugated proteins, and c) various ETC protein subunits encoded by mitochondrial (mt-) and nuclear (n-) DNA in male broilers within a single genetic line. Tissue homogenate or mitochondria were isolated from breast muscle or liver of broilers with HFE (0.80 ± 0.01, n = 7-8) and LFE (0.62 ± 0.02, n = 5-8). The activities of the ETC complexes were measured spectrophotometrically, while the levels of ubiquitin and immunoreactive proteins were analyzed using western blots. Activities of all complexes (I, II, III & IV) were higher in HFE compared to LFE broilers for both breast muscle and liver mitochondria. The levels of ubiquitin conjugated proteins were higher in LFE compared to HFE broilers breast muscle suggestive of enhanced protein turnover in LFE mitochondria. While the levels of the ETC immunoreactive proteins were higher in LFE muscle mitochondria for core I, cyt c1, cyt b (Complex III proteins) and COII (one of 8 proteins of cytochrome oxidase), but the levels of subunits NAD3, NAD4, NAD5, NAD6, NAD7 (from Complex I), 70S (Complex II), core II, ISP (Complex III), COX II (Complex IV), α-ATPase (complex V) were not different between the groups. We hypothesize that increased expression of certain ETC proteins (core I, cyt c1[n-encoded], cyt b and CO II [mt-encoded]) might be a compensatory response to lower ETC complex activity possibly to overcome the increased protein oxidation in LFE birds. Furthermore, inherent differences in ETC protein expression might lead to electron leak and increased protein oxidation, and

lower function of mitochondria. Further studies are warranted to determine mechanisms responsible for differences in ETC protein expression in broilers with low and high FE.

Funded in part by USDA-NRI #2001-03443 to W. Bottje.

Key Words: broiler, feed efficiency, mitochondria, protein subunits, respiratory chain complexes

50 Expression of mRNA encoding BMP-2, -4, -6, -7 and BMPR-IA, -IB and -II in the ovarian follicles of the chicken. O.M. Onagbesan^{*}, P. Van As, V. Bruggeman, and E. Decuypere, *Catholic University, Leuven, Belgium*.

Expression of mRNA encoding BMP-2, -4, -6, -7 and BMPR-IA, -IB and -II in the ovarian follicles of the chicken.

Onagbesan OM, Van As P, Bruggeman V, Decuypere E. Dept of Animal Science, Catholic University, Leuven, Belgium.

BMPs and their receptors (BMPRs) have recently been implicated in the regulation of mammalian ovarian function. In birds, BMPs are implicated in osteogenesis but any relevance to ovarian function has not been reported. The mRNA encoding BMPs and BMPRs have been localized in mammalian follicles where they regulate development in an autocrine/paracrine fashion. Whether such mechanism exists in the avian ovary is still unknown. This study examined the gene expression of BMP-2, -4, -6, -7 and BMPR-IA, IB and II in the growing follicles of the domestic hen. The F1, F2, F3 and F4 follicles were excised from Labelle hen ovaries and the theca and granulosa layers were separated. Tissues were pooled from four chickens and snap-frozen in liquid nitrogen. Total RNA was extracted, quantified and purity certified. Using RT-PCR, the mRNA encoding BMPs and BMPRs were detected in both tissues. With a quantitative method utilizing GAPDH as standard to verify and correct for RNA loading, mRNA levels were quantified on an image analysis system. Data were analysed from three separate repeat experiments. Results showed that mRNA for the peptides and receptors are expressed in the theca and granulosa. The mRNA levels of BMP-4 and #7 were 2-3 fold higher than BMP-2 and #6 in both granulosa and theca tissues of all follicles. However, BMP-4 levels were higher in granulosa than in theca but BMP-6 was higher in theca. Levels of BMP-2 and #7 were similar in both tissues. All BMP levels decreased significantly with transition of F4 to F3 followed by gradual increase up to F1 stage. BMPR-IA and -IB tended to be higher than BMPR-II in the granulosa of most follicle sizes. In the theca, the trend of mRNA for receptors were BMPR-IA > BMPR-IA > BMPR-II and receptors levels tended to increase with follicle enlargement. The presence of BMPs and receptors in the ovary suggest a role in avian ovarian function. The differential expression levels between tissues and follicle sizes may indicate preferential regulatory roles during development.

Key Words: Chicken ovary, BMPs, mRNA

**Monday, January 20, 2003
Environment & Management
Room: B314**

51 Factors Affecting Broiler Mortality During Live Haul. C. W. Ritz^{*}, A. B. Webster, and M. Czarick III, *The University of Georgia, Athens, Georgia USA*.

DOA (dead on arrival) broilers represent unrecoverable cost and loss of salable product for poultry processing companies. Mortality as a result of live haul indicate that the process is stressful to broilers. Flock health status, physical injury, and thermal stress all contribute to the incidence of DOA. Determining the leading cause of broiler DOA during the live haul process can reveal points where appropriate measures might be taken to reduce DOA numbers. Randomly sampled DOA from 4 flocks was characterized as part of an on-going thermal stress evaluation study. The mortality was necropsied from flocks caught in mid-afternoon during hot weather conditions. Results correspond to the existing weather conditions and not a yearly mean. Of the 270 birds necropsied, 30% were categorized as farm-related, 62% as live haul-related, and 8% as plant-related. Mean weight was 5.52 lb (SD 0.94), 72% of posted birds were male, and 97% of the total were deemed fit for processing had they not been DOA. Disease incidence and physical trauma are factors that impacted DOA the most during the live haul process under the existing conditions, implicated in 30 and 41% of the DOA, respectively. Thermal stress was attributed to 12% of the DOA. Disease control on the farm

and minimizing live haul stress are key areas for poultry companies to target DOA reduction efforts. Continued evaluation of DOA causative factors will shed light on procedures that can assist poultry companies to reduce DOA losses during all seasons.

Key Words: DOA, broiler, live haul

52 Weight loss in transit: an important issue in broiler transportation. M. A. Mitchell^{*1}, A. J. Carlisle¹, R. R. Hunter¹, and P. J. Kettlewell², ¹*Roslin Institute, Roslin, UK*, ²*Silsoe Research Institute, Silsoe, UK*.

Previous studies have demonstrated that thermal stress in transit compromises both welfare and productivity of broiler chickens during transport from farm to processing plant. Increased stress, mortality and reduced welfare have been identified in association with poor ventilation regimes and thermal challenge on commercial transport vehicles. No studies have examined in detail weight loss in transit and how this relates to transport conditions. The present study has addressed these issues in complementary laboratory and commercial field trials. Three

separate but related experiments quantified water loss from broiler chickens (2 kg body weight) under a range of thermal environments. In the first respiratory evaporative water loss was estimated gravimetrically for individual broiler chickens in the laboratory over a range of environmental temperatures from 24-36°C. In the second lab study total weight loss was estimated in groups of broilers in commercial transport crates over a similar range of micro-environments. In the final phase weight change and evaporative water losses of broilers on a commercial transporter were measured by means of pre and post-journey weighing of the load and determination of inlet and outlet water vapour density on a mechanically ventilated vehicle throughout the journey. Respiratory evaporative water loss increased from $0.5 \pm 0.1 \text{ g kg}^{-1} \text{ hr}^{-1}$ at 24°C to $2.9 \pm 0.4 \text{ g kg}^{-1} \text{ hr}^{-1}$ at 36°C ($p < 0.001$) in individual birds which would equate to weight losses of 20-111 kg on a typical UK broiler transporter carrying 6500 bird on a 3 hour journey. Measurements of total weight of broilers over a range of temperatures from 20-30°C on similar simulated journeys indicate losses of $3.2\text{-}8.2 \text{ g kg}^{-1} \text{ hr}^{-1}$ ($p < 0.001$) which would result in weight reductions at the factory of 125-320 kg per load. Subsequent studies on commercial vehicles revealed actual weight losses of $2.7\text{-}4.7 \text{ g kg}^{-1} \text{ hr}^{-1}$ (mean 144 kg per load) under varying thermoneutral conditions chiefly attributable to the measured water vapour heat exchange. The results illustrate the importance of understanding the factors influencing the physiological status of animals in transit and how the on-board environment can influence both product quality and welfare.

Key Words: Broiler, Transportation, Weight loss

53 Using recycled motor oil to heat broiler houses.

B. D. Fairchild* and M. Czarick, *University of Georgia, Athens, GA*.

Poultry house heating costs represent a significant expense for broiler growers during cold weather brooding. Identifying alternative fuels to produce heat may help growers to lower winter heating expenses typically incurred by high propane gas prices. The objective of this experiment was to compare fuel savings and environmental conditions in a broiler house using a recycled motor oil furnace versus a house using traditional propane forced air furnaces. A single indirect fire recycled motor oil unit, located outside the house, pulled air from the house, ran it through a heat exchanger, then the heated air was exhausted into a duct that ran the length of the brooding area. House air temperatures, humidity, fuel usage, flock mortality, water consumption, gaseous CO₂, O₂ and CO were recorded and compared to the standard propane-heated house. No differences in 10-day mortality or water consumption were observed. A 40% cost savings was found using the oil heat but cannot be considered a true savings since the environmental conditions within the two houses were different. The oil furnace system as installed was unable to deliver sufficient BTUs to maintain the proper house temperatures when outside temperature dropped below 40 degrees F. Furthermore, the delivery system was insufficient because the heat was not distributed uniformly and did not provide proper litter temperatures as compared to the house with forced air furnaces. Floor temperatures near the sidewalls were 10 degrees cooler as compared to a 2-3 degree difference in the house with forced air furnaces. Floor temperatures were 5-10 degrees cooler at the end of the house farthest from the unit. Consequently, the system was not used as the sole heat source through the entire brood period but was supplemented with force air furnaces. As the design is improved and other heat sources are developed it is likely that growers will have a means to use alternative heat sources to offset expenses due to high propane prices.

Key Words: Broilers, Heat source, Brooding, Temperature, House environment

54 Long-term effects of alum-treated poultry litter, untreated litter and ammonium nitrate on soil chemical properties. P.A. Moore¹, T.C. Daniel², and D.R. Edwards³, ¹USDA/ARS Poultry Production and Product Safety Research Unit, ²University of Arkansas, ³University of Kentucky.

Aluminum sulfate (alum) additions to poultry litter have been shown to greatly reduce phosphorus (P) runoff and ammonia emissions from poultry litter. A 20 year experiment was initiated in April of 1995 to determine the effects of poultry litter, alum-treated poultry litter and ammonium nitrate on soil chemistry, P runoff and forage yields. Fifty two small plots were established on a Captina silt loam soil at the University of Arkansas Main Agricultural Experiment Station. There were 13 treatments; four rates of normal poultry litter, four rates of alum-treated litter, four rates of ammonium nitrate and one unfertilized control. Litter

application rates were 1, 2, 3 and 4 tons/acre (2.24, 4.49, 6.73 and 8.98 Mg/ha). Ammonium nitrate rates were 65, 130, 195 and 260 kg N/ha, which were the same N rates as alum-treated litter. Soil samples (0-5 cm) were taken from each plot (10 cores/plot) periodically during the past 7 years. Soil chemical parameters that were evaluated included Mehlich III P, water soluble P, pH (with KCl), and exchangeable aluminum (KCl extractable). At year 7 depth profiles (0-5, 5-10, 10-20, 20-30, 30-40 and 40-50 cm) were also taken to evaluate P leaching. Water soluble P levels in soils fertilized with 8.98 Mg litter/ha were 45.6, 31.1, and 16.4 mg P/kg soil for plots fertilized with normal litter and 20.4, 10.2 and 2.9 mg P/kg soil for plots fertilized with alum-treated litter for the 0-5, 5-10, and 10-20 cm depths, respectively. Similar results were found for the different rates of litter, indicating that alum applications to poultry litter greatly reduces P leaching in soils. Both untreated poultry litter and alum-treated litter had a net liming effect on the soil, with pHs increasing over time, particularly at the higher rates. Ammonium nitrate applications acidified the soil. Whereas exchangeable aluminum levels were very low in soils amended with alum-treated or normal litter, soils fertilized with ammonium nitrate had elevated Al levels (up to 100 mg Al/kg), due to acidic soil conditions, resulting in poor forage growth. Yields have consistently been higher with alum-treated litter. These results indicate that fertilizing with alum-treated litter is more sustainable than fertilizing with normal poultry litter or ammonium nitrate.

Key Words: Phosphorus, Alum, Leaching, Eutrophication, BMPs

55 Development of a model system to study impacts of manure management practices on microbiological water quality. M.F.A Bal'a¹, G.E. Brink¹, A Adeli¹, and D.E Rowe¹, ¹USDA-ARS-WMFRU, Mississippi State, MS 39762.

Time-based comparative microbiological analysis is critical to understanding the impacts manure management practices have on environmental water quality. Field studies rarely lend themselves to well controlled experiments in view that time course studies are not readily implemented due to climatic, topographic and hydrological factors. This is particularly true of runoff studies, where several factors, such as study plots properties, time of rain, sample collection and transport methods, elapsed time between collection and analysis as well as other factors can influence microbiological analysis. This work describes the development of an experimental model to measure the impacts of manure management practices on water quality. The system consists of soil troughs, a rain simulator, and aseptic collectors for surface and sub-surface runoff. This study has focused on time-based parameters not readily measured under field conditions to include: 1) elapsed time between collection and analysis, 2) time of manure application to time of rain, 3) elapsed time between repeated rain events, 4) frequency, intensity and length of a rain event, and 5) impact of repeated rain events on the microbiological quality of runoff water from control and manure amended troughs. Parameters critical to manure management practices and their impacts on water quality are readily investigated and modeled with the developed system.

Key Words: Method Development, Runoff and Water Quality, Manure Management Practices, Microbiological Analysis, Mathematical Modeling

56 Water Flow Rates in Commercial Broiler Houses. B. D. Lott¹, W. A. Dozier², J. D. Simmons³, and W. B. Roush³, ¹Mississippi State University, ²University of Georgia, Tifton, Ga., ³USDA-ARS Poultry Research Unit, Mississippi State, Ms.

Water consumption and feed intake of broilers are highly correlated ($R^2 = 0.98$). In a thermoneutral environment, broilers consume approximately 1.6 times as much water compared to feed on a weight basis. Inadequate nipple water flow rates may limit water consumption translating to reduced feed consumption and growth rate. Measurements of flow rates of nipple drinkers were made in commercial broiler houses using a commercially available water measuring stick. Nipple flow rates were measured on a farm with birds weighing approximately 3,200g. The average flow rates among houses on this farm varied from 145 ml/min to 45 ml/min. At processing, average bird weight on the high flow rate was 3,700g and the low flow rate average weight was 3,450g. On another four-house farm with large birds the flow rate was approximately 40 ml/min; the average processing weight was 3,475g. Research has confirmed that reducing flow rates will decrease body weights and water usage. It has been recommended that water flow be 7 mls per week of age plus 20 ml. This

would equate water usage of a 5 wk old broiler to approximate 55 ml/min at a density of 10 to 12 birds per nipple. However, if there are more birds per nipple, the flow rate may need to be increased to achieve maximum growth rate. These data indicate that water flow rates of nipple drinkers can be highly variable among houses as well as among farms. Low water flow rates can limit final live weight at the end of the growout.

Key Words: Broiler, Nipple, Water consumption, nipple flow rate

57 Water requirements and supply flow rates for broiler production. D.M. Miles¹, J.D. Simmons*¹, and B.D. Lott², ¹USDA-ARS Poultry Research Unit, ²Mississippi State University.

Surveys conducted recently revealed that a wide variety of nipple flow rates are being routinely used in broiler houses throughout the southern broiler producing states. A study was conducted to determine if the variety of nipple water flow rates found in industry has any effect on broiler performance. Three trials were performed and flow rates of 25, 50, and 75 ml/min were made available to broilers reared in environmental chambers. Temperatures of 21 and 27 degrees C were maintained and feed and water (under the restrictions of the treatments) were provided ad libitum. The broilers were reared from day old to seven weeks of age. Results indicated that during the most productive week (fifth week) birds supplied 75 ml/min out gained birds supplied 50 ml/min by 20g but out gained birds being supplied 25 ml/min by over 100g. At the end of the trial the birds being supplied 75 ml/min were the heaviest by 30g over birds supplied 50ml/min and were again over 100g heavier than birds at the lowest water rate. Feed to gain ratios were lowest with the highest flow rate at 2.05 and highest with the lowest flow rate at 2.3.

Key Words: water supply, water flow rate, broilers

58 Field Trial Examining the Effect of Broiler Strain on Processing Yield. T. N. Chamblee*¹, M. T. Kidd¹, S. J. Barber¹, and D. W. Chamblee², ¹Poultry Science Department, Mississippi State University, ²USDA-ARS South Central Poultry Research Laboratory.

Two trials were conducted to determine the processing yield of two widely used broiler strain crosses grown in commercial broiler houses. In Trial 1, four hundred eighty 47 d old broiler chickens were obtained randomly from a broiler integrator (strains A and B). Sixty male and sixty female broilers were obtained from each of four houses (2 houses/strain and 2 replications/strain). Trial 2 followed the design of Trial 1, and four hundred broilers were obtained at 60 d of age. In each trial, all birds were from the same broiler farm and were subjected to the same nutritional regime. Body weight, hot carcass weight, leaf fat weight, and breast meat weight were obtained from each bird in both trials. Broiler house was the experimental unit for all parameters in both trials. Body weight was obtained after birds had received an 8 h feed withdrawal and had been transported to the pilot processing plant. In Trial 1, BW and hot carcass weight were affected by strain and gender. Breast and leg yield were affected by gender in Trial 1. In addition, there was a significant strain x gender interaction for hot carcass yield in Trial 1. Body weight, hot carcass weight, and leg yield were affected by gender in Trial 2.

Key Words: Strain, Broiler, Field trial, Processing yield

59 Influence of stocking density and pellet quality on summer broiler performance from 6 to 8 weeks of age and resultant carcass quality. Jaume Galobart*¹ and Edwin T. Moran, Jr.¹, ¹Poultry Science Dept. Auburn University.

Hot weather significantly burdens broiler production prior to marketing at heavy weight and can adversely affect carcass quality. Present work experimentation studied the effect of stocking density (6.7 vs. 3.1 birds/m²) on Ross x Ross 308 male broilers from 6 to 8 weeks of age while being fed either a whole pelleted finishing feed (18% CP & 3.15 kcal ME/g) stabilized with 0.5% gelatine or when crumbed. A total of 656 males at 6 weeks of age were randomly distributed to 8 replicate pens respective of the 4 treatments. All pens were in an open-side house and received existing summer conditions (25 ± 4 C and 71 ± 11 % RH) through to 8 weeks of age. Feed form and stocking density had effects that were independent. Feed designated as whole pellet had 83% particulates greater than 2.36mm diameter (vs. 17% in crumbed feed) and led to improved body weight gain without altering feed conversion, however, additional mortality largely due to SDS was prominent. Males fed pellets also had chilled carcasses having an increased percentage of abdominal fat that adversely affected yield when removed. Weights of fillets and

tenders deboned from the carcass 24 hours after processing were similar with birds receiving pellets and crumbs. Reducing the number of males from 6.7 to 3.1 birds/m² improved body weight gain, carcass weight, and breast fillet and tender weight. Feed conversion and mortality were not affected by stocking density. Light reflectance of fillets 24 hours after deboning revealed that fillets from birds that had received the crumbed feed had higher L* and lower a* than those receiving pellets, whereas stocking density was uninformative. Reducing the work of eating and decreasing stocking density to improve heat dissipation can enhance broiler performance while altering carcass quality but does so in a manner that is independent and complementary.

Key Words: Broiler, carcass quality, environment, pellet quality, stocking densities

60 The impact of outdoor coop housing and forage based diets vs. cage housing and mash diets on hen performance, egg composition and quality. H. D. Karsten, G.L. Crews, R.C. Stout, and P.H. Patterson*, *The Pennsylvania State University, University Park, PA, USA.*

Brown egg pullets were assigned at 17 wk of age to commercial cages (48 birds) or one of 3 outdoor coops (25 birds/coop) with pasture for three 2-wk periods during July-Aug 2002 in PA. In a modified Latin square design the 3 coops of birds rotated between alfalfa, clover and grass pastures, while the caged birds remained under commercial management. Caged hens (387cm²/bird) were provided commercial hen mash (16.9% CP, 2863kcal/kg) and water ad libitum. Pasture hens received 70g/hen/day mash, and calcium, water and their respective forage treatment ad libitum. Hen performance, egg quality and egg composition were summarized in each 2-wk period. Body weight of the pastured birds while not significantly different at 17 wk (1571g), was influenced by the period of lay. At the end of Period-1, birds were significantly heavier (1691g) than at the end of Periods 2 or 3 (1579 and 1570g, respectively). At the end of the study caged hens weighed more than the pastured birds (1821 vs. 1570g, P<0.05). Hen day egg production was influenced by both treatment and period (P<0.05). Caged hens laid at 85.0% over the three 2-wk periods, and pasture birds laid 73.4^b, 74.4^b and 69.1^b% for the alfalfa, clover and grass, respectively. Egg production averages for Periods 1, 2, and 3 were 56.2^c, 89.9^a and 80.2^b%. Livability at the end was 100, and 97.9% for the pastured and caged hens. Egg yolk total fat was not influenced by treatment or period and averaged 30.5%. Yolk fat omega-3 fatty acids were significantly impacted by treatment (alfalfa 3.77^a; clover 3.54^a; grass 3.15^b; caged 1.31^c%) and the period x treatment interaction. Yolk levels of vitamin A and E were significantly increased by pasture treatments compared to caged hens (vitamin A, 2932 vs. 2075IU/100g; vitamin E, 10.41 vs. 4.97IU/100g). Yolk vitamins from pastured hens at the end of the study were increased from continuous foraging, but did not change in caged hens.

Key Words: Egg, Yolk, Pasture, Omega-3, Vitamins

61 Mycoplasma gallisepticum Vaccination of Commercial Layer Chickens: Impact of Vaccine Mixing. S.L. Branton*¹, S.D. Collier¹, J.D. Evans¹, and G.T. Pharr², ¹USDA-ARS Poultry Research Unit, ²Mississippi State University.

Vaccination of commercial layer chickens with live Mycoplasma gallisepticum (MG) vaccine is usually performed at 8 to 10 weeks of age. The vaccine is typically administered via eyedrop or aerosol spray. Proper vaccine mixing is necessary to insure proper and uniform dosing of all chickens within a house otherwise, "spotty takes" may result which can ultimately negatively impact egg production over the subsequent laying cycle. This research demonstrates spotty takes resulting from improper vaccine mixing.

Key Words: disease, eggs, layers

62 A Cooperative Learning Environment is Desired in a Challenging Undergraduate Task. Mickey Latour*¹ and Abigail Cox¹, ¹Purdue University.

The purpose of this study was to investigate how cooperative learning among peers may influence examination outcome. For this study, students within a Freshman Class-Introduction to Animal Sciences were randomly divided into the following treatments: Group A-individuals; Group B-pairs; Group C represented three students, and Group D was composed of four students. Within a 50 min period, each group had to

complete a 100 question multiple choice exam. Students were allowed to use the textbook as well as interact verbally within their respective group. For comparison purposes, class averages for individuals and pooled averages within a group were compared to the 100 question exam score. Prior to the 100 question exam, overall averages for groups were statistically similar; however, following the 100 question exam there was a significant difference noted in the outcome for this exam. Similar scores following the exam compared to group averages prior to the exam were observed for groups C and D. Both groups A and B were significantly lower than groups C and D with Group A being significantly lowest of all groups. In

fact, Group A scored on average 22 points lower than the class average for individuals. Following a survey, students in Group A strongly wanted a partner and found the exam to be overwhelming. Groups B, C and D did not find the task overwhelming, enjoyed working within a group and felt the work load was shared equally among the students. These data would suggest effective partner numbers to be three within a group for there was no difference in outcome measures. Also, students seem eager to cooperate even though the partners are unknown.

Key Words: Teaching, Cooperative, Learning, Undergraduate, University

Monday, January 20, 2003 Nutrition Room: B313

63 The effects of distiller's dried grains plus solubles fed to laying hens. B.S. Lumpkins*, A.B. Batal, and N.M. Dale, *University of Georgia.*

An experiment was designed to examine the effects of 'new generation' distiller's dried grains plus solubles (DDGS) in commercial laying hen diets. Four experimental diets were fed to eight replications of 16 Hy-line W36 laying hens per treatment from 21 to 43 weeks of age. The four dietary treatments consisted of a commercial grade diet (18.5% CP, 2870 Kcal ME/Kg, 4.0% Ca and 0.42% available P) with 0 or 15 percent DDGS and a low density diet (17% CP, 2800 Kcal ME/Kg, 4.0% Ca and 0.40% available P) also with 0 or 15 percent DDGS. Hen body weights and feed consumption were measured periodically throughout the 22 week period. Several tests were performed throughout the experiment to explore all parameters of egg characteristics that may be affected by DDGS. These tests included egg production, egg weight, specific gravity, Haugh units, yolk color, and shell breaking strength. No significant differences were observed ($P > 0.05$) in egg production between the two commercial grade diets (0 and 15% DDGS) and the low density diet with 0% DDGS. However, there was a slight, but significant, depression ($P < 0.05$) in egg production with the low density diet containing 15% DDGS as compared to the other three dietary treatments. No significant differences ($P > 0.05$) in egg weights were observed among the four dietary treatments. There was also no significant difference observed ($P > 0.05$) for any of the egg characteristics measured. One might expect to see an increase in feed consumption when hens are fed the low density diet yet no significant differences in feed consumption were observed. Summer temperatures may provide some explanation for the lack of differences in feed consumption. Distiller's dried grains plus solubles proved to be an acceptable feed ingredient when used up to 15% in commercial laying hen diets.

Key Words: Laying hens, Distiller's dried grains plus solubles, Low density, Feed ingredients, DDGS

64 Effect of heat on the metabolizable energy value of cassava meal (*Manihot esculenta*). H. Romero*¹, D. Mora², A. Gallego¹, L. Quintanilla¹, and J. Brake³, ¹*Universidad de Antioquia, Medellin, Antioquia, Colombia*, ²*Universidad Nacional de Colombia, Medellin, Antioquia, Colombia*, ³*North Carolina State University, Raleigh, NC USA.*

Many tropical countries import corn as their main source of dietary metabolizable energy (ME) for poultry although cassava has higher ME potential depending upon its origin and processing. Two experiments were conducted in Colombia to evaluate the effect of processing temperature and origin on the nutritional value of cassava root meal (CRM) for laying hens. In the first experiment, two samples of CRM of different origin with one sample of CRM subjected to additional cooking at 115 C and 15 psi were evaluated. Samples were substituted in a basal diet at levels of 15, 30 and 45% for AMEN determination. The TMEN values were determined by Sibbald's fasted cockerel method. In the second experiment, seven diets were evaluated in a one-way anova using seven replicates of nine hens each. Levels of 0, 10, 20 and 30% of either CRM and cooked CRM were included in iso-energetic (2.79 kcal/g) and iso-protein (16% CP) diets. Samples averaged (DM basis): 13.7% moisture, 4.2% CP and 65.4% starch. Predicted AMEN values were lower than calculated, but the difference was less for the cooked CRM. There were differences ($P \leq 0.05$) for origin and process in AMEN and TMEN. Cooked CRM showed higher AMEN (2.5 kcal/g) and TMEN (3.2 kcal/g) values than uncooked CRM (AMEN 2.4 kcal/g and TMEN 3.13 kcal/g). Higher ME

values were associated with a 60% decrease in cyanogenic compounds and possibly with improved gelatinization of carbohydrates. This was in agreement with the fact that higher ME values are assumed when CRM diets are subjected to the heat and pressure of pelleting or extrusion. In the second experiment, there were no differences in egg production, yolk color, albumen or eggshell quality. Feed intake was decreased ($P \leq 0.05$) by CRM at all levels and by cooked CRM at 30%. Although egg weight decreased with CRM at the 30% level, no differences were observed in egg production, egg mass or feed conversion. CRM can be included in laying hen diets at levels of up to 20% without difficulty.

Key Words: Cassava, Energy, Prediction equations, Commercial layers

65 Peanut Meal: an Excellent Ingredient for Laying Hen Diets. Gene Pesti¹, Remzi Bakalli¹, John Driver*¹, Kimberly Sterling¹, Erin Bell¹, and Lena Hall¹, ¹*University of Georgia Department of Poultry Science.*

Peanut protein is severely limiting in threonine and has been used to create threonine deficiency in animals. The availability of purified threonine at low cost, raises the possibility of economically using peanut meal and threonine combinations in poultry diets. An experiment was conducted to compare corn and peanut meal (PNM) basal diets to corn and soybean meal (SBM) based diets at three protein levels (16.0, 18.5 and 21%) in diets for 22 to 34 week old commercial leghorns. Birds were housed 2 per cage with 4 cages per replicate and 6 replicates per treatment. Feed consumption, egg production and feed per dozen eggs were practically identical for PNM (93.8g/hen/day, 92.2 eggs per 100 hens/day, and 1.22 kg/dozen) and SBM (93.7g/hen/day, 92.2 eggs per 100 hens/day, and 1.22 kg/dozen). Dietary protein level had no consistent effect on any of these parameters but did significantly improve body weight gains and egg weights (1.2 to 2.5g /egg). PNM-fed hens laid slightly smaller eggs during the first 6 weeks ($P < 0.05$) but there were no significant egg size differences during the last 6 weeks ($P > 0.14$). PNM-fed hens laid eggs with better interior quality at 26 and 30, but not 34 weeks of age ($P < 0.05$). After 2 weeks of storage, Haugh units remained better for eggs from hens fed PNM than SBM when kept refrigerated (5C; $P < 0.05$) and at room temperature (20 C; $P < 0.10$). It is concluded that PNM is an excellent ingredient for laying hen diets.

Key Words: Leghorns, Peanut Meal, Soybean Meal

66 Effect of Dietary Zinc Source on Reproductive Performance and Immune Status of Broiler Breeder Hens. B. P. Hudson*¹, W. A. Dozier, III¹, J. L. Wilson¹, J. E. Sander², and T. L. Ward³, ¹*Department of Poultry Science, The University of Georgia*, ²*Department of Avian Medicine, The University of Georgia*, ³*Zinpro Corp., Eden Prairie, MN.*

Zinc amino acid complex has been shown to have increased bioavailability when compared to inorganic sources of zinc. Furthermore, short-term studies with broiler breeders have reported that dietary zinc amino acid complex affects reproduction and immune status. This experiment was conducted due to limited data regarding the long-term effects of zinc source on reproduction. Cobb 500 slow-feathering hens were given one of three diets from 0 to 65 wk. All experimental diets consisted of 160 ppm supplemental Zn from ZnSO₄, Availa[®] Zn zinc amino acid complex (ZnAA), or a mixture of ZnSO₄ and ZnAA (ZnSO₄+ZnAA, 80 ppm Zn from each). At 20 wk, all birds were caged (2 per cage) and 16 hr light per day was provided. Egg weights and specific gravity were measured every

four wk starting at 26 wk. Eggshell breaking strength was measured at 50, 58 and 66 wk. Egg production, percentage cracked and hatching eggs were determined weekly. Hens were artificially inseminated, and eggs were incubated and hatched weekly (25-35 wk), biweekly (37-45 wk) or every fourth wk (49-65 wk). To assess immune status, hemagglutination inhibition titers to Newcastle disease virus (NDV), phytohemagglutinin (PHA)-induced wattle swelling and footpad scores were determined. Hens fed ZnSO₄+ZnAA expressed the lowest incidence of cracked eggs (ZnSO₄=8.0%, ZnAA=7.3%, ZnSO₄+ZnAA=6.6%, P<0.05), highest hen-day egg production (ZnSO₄=56.6%, ZnAA=56.7%, ZnSO₄+ZnAA=57.3%, P<0.05), greatest hatching egg production per hen housed (ZnSO₄=137, ZnAA=138, ZnSO₄+ZnAA=140, P<0.05) and highest chick production (ZnSO₄=106, ZnAA=105, ZnSO₄+ZnAA=109, P<0.05). Hens fed ZnSO₄ had the lowest specific gravity (ZnSO₄=1.079, ZnAA=1.080, ZnSO₄+ZnAA=1.080, P<0.05). Cellular immune response to PHA injection was enhanced as dietary ZnAA increased (P<0.05). Antibody titers to NDV were greatest for the ZnAA treatment (P<0.05). Hen-housed egg production, mortality, egg weight, eggshell strength, fertility, hatchability and footpad score were not influenced by zinc source. Data from this experiment indicate that additions of Availa[®]Zn to broiler breeder diets enhanced immune response, shell quality and hatching egg production; and hens fed ZnSO₄+ Availa[®]Zn generally had superior performance.

Key Words: Dietary zinc source, Broiler breeder hen performance, Immune response, Eggshell quality

67 Effect of dietary nitrogen on chick hepatic malic enzyme and histidase mRNA expression. T. P. Chendrimada* and A. J. Davis, *University of Georgia, Athens, GA USA.*

Previously, we reported that feeding broiler chicks a high (40%) protein diet compared to a basal (22%) protein diet decreased chick hepatic malic enzyme mRNA expression, and increased hepatic histidase mRNA expression. Additionally, chicks fed the basal diet supplemented with either a mixture of essential or non-essential amino acids to equal the amount found in the high protein diet had malic enzyme and histidase mRNA expression levels which were intermediate to those of chicks fed the basal or high protein diets. These results suggested that the expression of these enzymes was controlled by a specific mixture of essential and non-essential amino acids or simply by an increase in dietary nitrogen intake. To investigate if increasing dietary nitrogen levels alters mRNA expression of these enzymes, a series of experiments were conducted. One wk old broilers were fed the basal diet for a period of 5d. The birds were then split into 3 groups of 6 replicate pens of 2 birds each. One group was fed the basal diet, while another group was fed the high protein diet. The third group was fed the basal diet supplemented with 9.5% glutamic acid (Exp.1), 6% alanine (Exp.2), 5% glycine (Exp.3) or 7.5% diammonium citrate (Exp.4). Each of the supplements contributed only half of the difference in the total amount of dietary nitrogen between the basal and high protein diets. After feeding the diets for 24h, liver samples were collected for RNA isolation and subsequent Northern Analysis. Birds fed the supplemented diets had significantly lower hepatic malic enzyme levels compared to birds fed the basal diet. The decrease in malic enzyme expression in chicks fed the supplemented diets tended not to be as great as those fed the high protein diet. Histidase mRNA expression, was not affected by any of the dietary nitrogen supplements. The results suggest that hepatic malic enzyme expression may be regulated by dietary nitrogen intake while histidase expression is regulated by intake of specific dietary amino acids. The malic enzyme mRNA expression results also suggest that supplementing protein adequate broiler diets with any source of dietary nitrogen might reduce malic enzyme activity, and thus lower body fat levels, since the activity of malic enzyme in poultry is positively correlated with body fat content.

Key Words: Malic enzyme, Histidase, Nitrogen

68 Effect of Multiphase Feeding on Broiler Performance and Nitrogen Excretion. N. Surbakti*¹, A. Haq¹, J. Carey¹, and C. A. Bailey¹, ¹Texas A&M University System.

Two experiments were conducted simultaneously to evaluate the effects of multi-phase feeding programs on broiler performance and nitrogen excretion. In Experiment 1, sixty day-old male broiler chicks (Ross x Ross) were randomly placed in 30 separate battery brooder pens with three

treatments (10 replications per treatment). Broilers fed Treatment 1 consumed four-phase industry type broiler diets. Treatment 2 and 3 were multiphase feeding programs in which the diets were changed every three days. Diets for treatment 2 were linear blends of treatment 1 and treatment 3 was calculated by EFG Software (Natal)#. Feces were collected every three days and frozen for later nitrogen analysis. After seven weeks of feeding the birds were killed, ground whole and analyzed for nitrogen retention. Experiment 2 was similar to Experiment 1 with the exception that 540 day-old male broilers were randomly placed in 36 floor pens (12 replications). In both experiments multiphase feeding improved body weight gain and feed gain ratio in week 4 (Experiment 1) and week 6 (Experiment 2); but not in week 7. Feed consumption, nitrogen excretion and nitrogen retention were unaffected by the treatments. Economic analysis indicated that both multiphase feeding programs could potentially lower production costs per kg of gain.

Key Words: multiphase feeding, broiler, performance, nitrogen excretion

69 Studies on the source of unidentified D₃ activity in some broiler chick experiments. A. Atencio*, R. Shirley, H. Edwards, Jr., and G. Pesti, *University of Georgia, Athens, GA/USA.*

Studies utilizing a corn-soybean meal diet adequate in all other nutrients when fed to chicks in an ultraviolet light-free environment from 1 to 16 days produces chicks having 22 to 28% bone ash (Poultry Science 81:664-669). In two experiments conducted in November 2001 and March 2002 the bone ash values obtained from chicks fed the basal diet were 35.0% and 34.6%, respectively, suggesting some unidentified source of vitamin D activity in these experiments. Poultry fat was identified as one of the most likely sources of this unidentified D activity which would be equivalent to 5,670 IU of D/kg of poultry fat. A subsequent experiment (April 2002) where corn oil was used as a fat source, instead of poultry fat, produced chicks with an average bone ash value of 24.2%. Two experiments have subsequently been conducted to evaluate vitamin D activity in three sources of poultry fat. The same diet was utilized in these studies and standard response curves to D₃ levels were obtained for the criteria of 16-d body weight gain, gain:feed, bone ash and incidence of rickets. In Experiment 1, a poultry fat of unknown origin was fed at 5, 2.5, and 1.25% of the diet at the expense of vegetable oil. In Experiment 2, poultry fat (1) from a plant where chickens have been fed 25-hydroxycholecalciferol and (2) from an unknown origin were tested. The bone ash data from the D₃ standard curves yielded regression equations that were used to evaluate the bone ash responses to the poultry fats. Using these equations, the poultry fat in experiment 1 contained 754, 423 and 0 IU/kg of D activity, corresponding to the three fat levels of 5, 2.5 and 1.25%, respectively. In the second experiment, the poultry fat from 25 hydroxycholecalciferol fed chickens contained 28 and 1,226 IU/kg of D activity, when 2.5 and 5% poultry fat were fed, respectively, while the other poultry fat did not show any D activity in improving the bone ash. These studies establish that there is vitamin D activity in some samples of poultry fat, and the activity may be related to the amounts and source of D₃-fed birds from which the fat was made.

Key Words: Broilers, Vitamin D₃, Poultry fat, 25-hydroxycholecalciferol

70 Broiler performance to 21 days as affected by cumulative broiler breeder pullet nutrition during rearing. J. Brake, B. A. Lenfestey, and P. W. Plumstead*, *North Carolina State University, Raleigh, NC USA.*

Cumulative nutrient intake of broiler breeder pullets to 21 wk of age has been shown to affect subsequent performance. It was hypothesized that a higher plane of rearing nutrition may allow early lay broiler breeders to produce better performing broilers. To evaluate vertical effects of cumulative nutrition during the breeder rearing period on performance of broiler offspring, four broiler trials were conducted using chicks hatched from breeders reared on a range of cumulative nutrition in three consecutive trials. Breeder Trials 1 and 2 each used three graded levels of cumulative crude protein (CP) and metabolizable energy (ME) intakes (High (27,788 kcal ME and 1485 g CP), Medium (26,020 kcal ME and 1391 g CP), Low (24,242 kcal ME and 1296 g CP)) to 21 wk. Breeder Trial 3 used only the High and Low levels. A single breeder diet and identical management practices were applied to birds in all three trials. Identity of breeder treatments was preserved during incubation to evaluate vertical effects on broiler offspring. All broilers were fed starter crumbles to 21 d with breeder cumulative nutrition and broiler sex as

main effects. Broiler Trial 1 used chicks hatched from Breeder Trial 1 at 27 wk of age. Broiler Trials 2 and 3 evaluated chicks hatched from Breeder Trial 2 at 28 and 39 wk of age, respectively and Broiler Trial 4, chicks hatched from Breeder Trial 3 at 33 wk of age. The high plane of cumulative breeder nutrition increased 21 d broiler male BW in Broiler Trials 1 ($P \leq 0.05$), 2 ($P \leq 0.06$), and 4 ($P \leq 0.08$). There was no effect in Broiler Trial 3 when the breeders were 39 wk of age. Female BW was affected by plane of breeder nutrition in Broiler Trial 2 ($P \leq 0.05$) only and the effect appeared to be less consistent for female offspring. Broiler breeder pullet nutrition during rearing appeared to affect both subsequent female breeder and male broiler performance from early lay broiler breeders.

Key Words: Broiler breeders, Cumulative nutrition, Broiler performance, Maternal influences

71 Impact of Feed Processing on Broiler Performance. S. M. Hill^{*1}, S. J. Barber¹, E. H. Robinson¹, M. D. Holifield², and M. T. Kidd¹, ¹Mississippi State University, Mississippi State, MS, ²Delta Western, Indianola, MS.

Two floor pen experiments were conducted to evaluate the impact of feed processing on male broiler performance. In experiment 1, live performance was measured from Days 1-18. Live performance and processing traits were measured in Experiment 2 from Days 1-42. Treatments consisted of common diets that were prepared as mash, steam pelleting, or extrusion (starter processed feed was crumbled). Treatments were administered to 54 pens containing 45 birds per pen each in Experiment 1 (18 replications/treatment) and 36 pens of 40 birds each (12 replications/treatment) in Experiment 2. Experiment 1 was conducted in a conventional house and Experiment 2 was conducted in a tunnel-ventilated house and light intensity was reduced to mimic current industry practices. Measurements consisted of BW gain, feed intake, and mortality in both experiments and live BW, hot carcass weight, fat pad weight, and total breast meat weight from five birds per pen in Experiment 2. In Experiment 1, BW gain was improved ($P < 0.05$) in birds fed pellet/crumble feed over birds fed mash feed. But birds fed extruded/crumbled feed had better ($P < 0.05$) BW gain than birds fed pellet/crumbles. Feed conversion followed a similar pattern to that of BW gain and differences in mortality did not occur. In Experiment 2, BW gain responses were improved ($P < 0.05$) in all time periods in birds fed the pelleted or extruded feed versus birds fed mash feed. However, overall livability was reduced ($P < 0.05$) in birds fed the pelleted and extruded feed over birds fed the mash feed. Similar to BW gain, carcass and breast meat weights, but not yields, were increased ($P < 0.05$) in birds fed pelleted and extruded feed over birds fed mash feed. Birds fed extruded versus pelleted feed had improved early growth in Experiment 1, but not Experiment 2. Although finishing growth was improved in birds fed either type of processed feed in Experiment 2, processed feed increased mortality over birds fed mash feed.

Key Words: Broiler, Pelleted feed, Extruded feed

72 Effects of Soybean Meal Particle Size on Phosphorus Utilization in Turkey Poults. R. A. Charbeneau* and K. D. Roberson, Michigan State University, East Lansing, MI.

A 4-wk experiment was conducted to determine the effects of soybean meal (SBM) particle size on body weight (BW), rickets incidence (RI), tibia ash, gizzard and intestine weight, serum phosphorus and gizzard content pH. One hundred and ninety-two 1-day old Hybrid hens were randomly allocated into groups of eight per pen (24 pens). A corn-soybean meal based diet was formulated to contain calcium at 1.20% and non-phosphate phosphorus at 0.48%. Three treatments (trts) were fed, varying only by SBM particle size. SBM from a commercial source (trt 3), was then passed through a 1.75 mm (trt 1), or 6.35 mm (trt 2) hammer mill screen. Geometric mean diameter of SBM (microns) was determined using a sieve shaker (trt 1= 501, trt 2= 550, trt 3=856). Corn particle size stayed constant throughout the three trts, having a geometric mean diameter of 853 microns, after being ground through a 9.53 mm hammer mill screen. At 14 days of age, 24 poults from each treatment group (72 total) were placed into a grower unit. Four replications with six poults per pen were used for each treatment. Body weight was measured weekly, while RI, tibia ash, gizzard weight, and gizzard pH were measured at 14 and 28 days. Serum phosphorus concentrations were determined at 14 days of age, and intestine weights were determined at 28 days of age. As particle size increased, 14-day gizzard weight ($p=0.006$), 28-day gizzard weight ($p=0.0709$), and 28-day gizzard content pH ($p=0.0886$) increased

linearly, while 14-day gizzard content pH ($p=0.0290$) decreased linearly. Tibia ash on days 14 and 28 were not affected ($p=0.3347$, $p=0.4199$), nor was 14-day RI ($p=0.6767$). Serum phosphorus, intestine weight, body weight, and 28-day RI were also not affected. These results demonstrate that grinding SBM to a finer particle size has no effect on phosphorus utilization.

Key Words: Phosphorus utilization, SBM, Particle size, Poultry

73 Vitamin C restores the loss of pigmentation in brown eggshells caused by vanadium. A. Z. Odabasi¹, R. D. Miles^{*1}, M. O. Balaban¹, V. N. Sampath¹, and C. W. Comer¹, ¹University of Florida.

The detrimental effects of vanadium (V) on albumen quality and eggshell pigmentation has been documented. Restoration of albumen quality by supplementation of Vitamin C to the diet has been demonstrated in several research studies with white Leghorns and brown egg-type laying hens. No research data have been published on the effects of supplementing various compounds to diets in an attempt to restore pigmentation in eggshells of eggs from hens fed V. An experiment was conducted with Hyline brown layers to determine if supplementing diets containing 15 and 30 ppm V with 100 ppm Vitamin C (VC), 100 IU Vitamin E (VE), or 100 ppm beta-carotene (BC) would restore eggshell pigmentation. From a population of 480 hens, 60 were selected that had eggshells with the highest pigment concentration (darkest shells). Birds were randomly assigned to three treatment groups. Each treatment consisted of four pens of 5 individually caged hens eating from the same feeder. A corn-soybean meal diet served as the control (0 ppm added V). Two other diets were supplemented with sodium metavanadate at concentrations to provide 15 and 30 ppm V, respectively. These 3 diets were fed continuously for 9 days. After feeding V for only 3 days, the significant detrimental "bleaching" effect was observed in hens fed 30 ppm. By day 6, a significant "bleaching" effect in shells was observed in hens fed 15 ppm V. On day 9, each group of 5 hens within each V group (0, 15, 30 ppm V) were fed diets containing either VC, VE or BC throughout the entire feeding period. Supplementing VC resulted in recovery of pigmentation in eggs of hens fed 15 and 30 ppm V. Egg pigmentation did not recover from feeding VE or BC. These data show that feeding up to 30 ppm V, a common contaminant of poor quality feed-grade phosphate sources, does have a detrimental bleaching effect on brown eggshells and that the pigmentation can be restored by feeding 100 ppm VC in the diet of brown egg birds.

Key Words: Vanadium, Pigmentation, Eggshells, Vitamin C

74 Effect of dietary electrolyte balance on the immune response (Newcastle disease virus antibody titers) of broiler chickens at various ages following vaccination and during heat stress. E. Santin¹, S. A. Borges¹, A. V. Fischer da Silva², D. M. Hooge^{*3}, and K. R. Cummings, ¹UNESP - FCAV, Jaboticabal, Brazil, ²Universidade Federal do Parana, Brazil, ³Hooge Consulting Service, Inc., Eagle Mountain, UT, ⁴Church & Dwight Co., Inc., Princeton, NJ.

The objective of this research was to determine Newcastle titers of broiler chickens at different ages following vaccination and under heat stress. A total of 240 broiler chickens were utilized. Corn-soy and soy oil based diets were nutritionally balanced. Four dietary electrolyte balances (DEB; 40, 140, 240, or 340 mEq Na + K - Cl/kg feed) were developed by using NaCl, NaHCO₃, and NH₄Cl supplements. Each treatment consisted of four replicate groups of 15 chicks each. At 7 and 21 d of age, the broilers were vaccinated by eye with LaSota strain of Newcastle virus, and blood was taken at 25, 35, and 42 days of age for serum hemagglutination inhibition (HI) testing to obtain means of antibody titers (log₂). From 14 to 42 d of age, chickens were exposed to heat stress of 35° C for ten hours daily. A significant ($P < 0.05$) linear increase in immune response as judged by Newcastle titers with increasing DEB occurred at 35 d of age (Table 1). An equation was found to describe this relationship (HI log₂ = 1.6249 + 0.0010 base excess). Blood bicarbonate was calculated from pCO₂ as follows: log HCO₃ = pH + log pCO₂. Base excess was calculated as follows: base excess = HCO₃⁻ - 24.8 + 16.2 (pH - 7.4). Serum HI test antibody titers (log₂) to Newcastle vaccine (LaSota) by DEB levels of 40, 140, 240, and 340 were: 25 d - 2.07, 2.14, 2.16, 2.07 ($P > 0.05$); 35 d - 1.69, 1.99, 1.95, 2.37 (linear; $P < 0.05$); and 42 d - 1.48, 1.62, 1.73, and 1.80 ($P > 0.05$), respectively. By age, overall mean titers were: 25 d, 2.11; 35 d, 2.00; and 42 d, 1.66. It was concluded that under heat

stress broiler chickens exhibited increasing antibody titers to Newcastle virus with increasing levels of DEB from 40 to 340 mEq/kg at 35 d of

age. Following vaccination at 7 and 21 d, antibody titers declined from 25 to 42 d under conditions of this study.

Key Words: Broiler, Electrolyte balance, Immunity, Newcastle disease, Sodium bicarbonate

Monday, January 20, 2003 Processing & Products Room: B316

75 Effect of Immersion Chilling on Incidence of *Salmonella* on Post-Chill Carcasses During Commercial Turkey Processing. J.L. Cavitt^{*1}, A.P. McElroy², J.A. Byrd³, D.J. Nisbet³, and D.J. Caldwell¹, ¹Texas A&M University, College Station, TX, ²Virginia Tech, Blacksburg, VA, ³USDA/ARS/SPARC, College Station, TX.

Our laboratories recently conducted a multi-state survey of six commercial turkey-processing facilities to determine the impact of immersion chilling on incidence of *Salmonella* on post-chill turkey carcasses. Within each facility, on each of two consecutive days of sampling, 100 pre- and 100 post-chill carcasses were sampled by carcass rinsing to recover *Salmonella*. Collected rinsates were pre-enriched in buffered peptone water prior to subsequent selective enrichment in RV broth and plating onto DMLIA plates. Chiller microbial intervention strategies within individual plants consisted of administering chlorine gas (Cl₂), sodium hypochlorite (NaOCl), potassium hypochlorite (KOCl), calcium hypochlorite (Ca(OCl)₂), or chlorine dioxide (ClO₂). Plants 2, 3, 4, and 6 all achieved significant reductions (P<.001) in post-chill carcass *Salmonella* incidence on both days of sampling. Sampling of Plant 2 revealed reductions in *Salmonella* incidence of 78.2% and 90% on days 1&2, respectively, while Plant 3 data revealed reductions of 70.2% on day 1 and 94.1% on day 2. Similarly, sampling of Plant 4 was associated with post-chill *Salmonella* reductions of 79.2% and 94.2% on days 1 and 2, respectively, while Plant 6 achieved post-chill reductions of 86.2% and 61.3% on days 1 and 2, respectively. Due to a low overall pre-chill incidence, sampling of Plant 1 revealed no significant differences between pre- and post-chill *Salmonella* incidence (P>.05). In Plant 5, a statistical difference (P>.05) was not observed in *Salmonella* incidence on day 1 of sampling, however, day 2 sampling revealed a 107.1% increase in *Salmonella* on post-chill carcasses (P<.001). These observations suggest immersion chilling of whole turkey carcasses, when coupled with an appropriate microbial intervention strategy, is effective for the reduction of *Salmonella* incidence levels on commercially processed turkey carcasses.

Key Words: *Salmonella*, Carcass Rinsing, Turkey, Chill Immersion

76 Association of immersion chiller intervention strategies with post-chill *Salmonella* and *Campylobacter* incidence in commercial turkey processing. A.P. McElroy^{*1}, J.L. Cavitt², J.A. Byrd³, D.J. Nisbet³, and D.J. Caldwell², ¹Virginia Tech, ²Texas A&M University, ³USDA/ARS/SPARC.

Microbiological sampling of turkey carcasses and measurement of immersion chiller environmental parameters were conducted in a multi-state survey of six commercial turkey-processing facilities. The study objective was to compare *Salmonella* and *Campylobacter* incidence data from carcasses with chiller environment measurements to identify effective intervention strategies for pathogen reduction. On two consecutive sampling days in each facility, 100 pre- and 100 post-chill carcasses were sampled by carcass rinse procedures. Chiller measurements included pH, type of microbial intervention, and total and free chlorine concentration. Chiller intervention strategies consisted of chlorine gas (Cl₂), sodium hypochlorite (NaOCl), potassium hypochlorite (KOCl), calcium hypochlorite (Ca(OCl)₂), or chlorine dioxide (ClO₂). Plant 1 maintained total Cl⁻ within a range of 2.6-44ppm, free Cl⁻ from 0.01-1.44ppm, and pH from 6.2-6.9 with *Salmonella* (32%) and *Campylobacter* (6.1%) reduction. Plant 2 indicated a 0.04-1.89ppm range of ClO₂ and pH of 7.3-8.45 with reductions in *Salmonella* (84.4%) and *Campylobacter* (48.1%). Plants 3 (0.04-21.1ppm) and 4 (0.01-23.2ppm) had higher free Cl⁻ on average with total Cl⁻ ranging from 9.7-59.6ppm and 0.94-24.4ppm, respectively. Ranges of pH were 6.2-7.4 in Plant 3 and higher (8.1-8.7) for Plant 4. *Salmonella* and *Campylobacter* levels were reduced in Plant 3 (82% and 89.9%) with Plant 4 having an 86.2% reduction and 35% increase, respectively. Plant 5 had the lowest range of total (0-3.4ppm) and free (0-1.94ppm) Cl⁻ and pH of 6.8-8.0 with an associated 32% increase

in *Salmonella* and 79.4% reduction in *Campylobacter*. Plant 6 consistently reduced both *Salmonella* (74.5%) and *Campylobacter* (71.5%) with ClO₂ of 0.05-0.91ppm and pH of 8.0-8.8. Collectively, these data provide basis for developing best management practices for chiller intervention strategies for reduction of *Salmonella* and *Campylobacter* on commercial turkey carcasses.

Key Words: *Salmonella*, *Campylobacter*, Turkey, Chiller

77 Impact of the Chiller Environment on *Campylobacter* Carcasses During Turkey Processing. J.A. Byrd^{*1}, J.L. Cavitt^{2,3}, A.P. McElroy⁴, D.J. Nisbet¹, and D.J. Caldwell^{2,3}, ¹USDA-ARS, SPARC, College Station, TX, ²Texas A&M University, College Station, TX, ³Texas Agricultural Experiment Station, College Station, TX, ⁴Virginia Tech, Blacksburg, VA.

As a part of a multi-laboratory, multi-state survey of commercial turkey processing to assess the effects of immersion chilling on the incidence of *Campylobacter* recovery from processed turkey carcasses, one hundred pre- and 100 post-chill turkey carcasses were collected from six individual processing facilities on two consecutive days of processing. All carcasses were subjected to a whole carcass rinse using 200 mL of buffered peptone water and the recovered rinsate was enriched in Bolton's Broth at 42C for 24 h. The enriched rinsate was streaked on Campy-ceflex plates and incubated an additional 48h at 42C in a microaerophilic environment for *Campylobacter* detection. Chiller microbial intervention strategies within individual plants consisted of administering chlorine gas (Cl₂), sodium hypochlorite (NaOCl), potassium hypochlorite (KOCl), calcium hypochlorite (Ca(OCl)₂), or chlorine dioxide (ClO₂). Pre- and post-processing chill tank characteristics and management methodologies were recorded for each plant during both sampling days. The incidence of Day1/Day 2 *Campylobacter* reductions for Plants 1-6 are as follows: Plant 1 (7%/4%), Plant 2 (18.9%/76.1%), Plant 3 (88.5%/98.3%), Plant 4 (33.3% increase/50% increase), Plant 5 (69.5%/88.2%), and Plant 6 (67.4%/75%). Significant (P<.025) post-chill *Campylobacter* reductions in incidence were observed during both days of samplings in Plants 2, 3, 5, and 6, indicating the effectiveness of these selected chiller intervention strategies on bacterial remediation. These data suggest that management of the chill immersion environment, including the represented intervention strategies, is effective for reducing the spread of *Campylobacter* on processed turkey carcasses.

Key Words: *Campylobacter*, Chill Immersion, Turkey Processing

78 Overall Contribution of Immersion Chiller Management on Remediating *Salmonella* During Commercial Turkey Processing. D.J. Caldwell^{*1}, J.L. Cavitt¹, J.A. Byrd², D.J. Nisbet², and A.P. McElroy³, ¹Texas A&M University, College Station, TX, ²USDA/ARS/SPARC, College Station, TX, ³Virginia Tech, Blacksburg, VA.

Our laboratories recently completed a multi-state survey of commercial turkey processing to assess the contribution of management of immersion chilling systems in remediating *Salmonella* on processed carcasses. Phase 1 of the study consisted of a preliminary survey of 5 plants between February and June of 2000 and included rinsing 100 pre-chill and 100 post-chill carcasses within each plant using common sampling and culture procedures. Results revealed post-chill *Salmonella* incidence was significantly increased (P<.05) in 3 of 5 plants and significantly decreased (P<.001) in 2 plants. Data and observations during Phase 1 suggested chiller management in both plants with significant reductions in *Salmonella* incidence contributed to improved microbiological quality of post-chill carcasses. Phase 2 of the study was a survey of six plants between March and September of 2002. Four of the 6 plants sampled also participated in Phase 1 and, based upon our initial observations, each implemented

changes in chiller management prior to the start of the second survey. For Phase 2, on two consecutive days of processing, direct chiller measurements were obtained to assess management practices, and extensive microbiological sampling of processed carcasses was performed using similar sampling and culture procedures. Significant reductions ($P < .001$) in post-chill *Salmonella* incidence were achieved in 4 of 6 plants, with a significant increase ($P < .05$) observed in only 1 plant. One plant was associated with low overall incidence levels that were not statistically different ($P > .05$). Importantly, assessment of chiller management within each plant was associated with observed microbiologic data. Conclusions from the samplings indicate that properly managed immersion chilling systems with effective microbial intervention strategies consistently remediate *Salmonella* from processed carcasses during commercial turkey processing.

Key Words: Turkeys, Immersion Chilling, *Salmonella*, Management

79 Persistence of *Salmonella* applied to breast skin one hour prior to scalding and picking of feathered and featherless (scaleless) broilers. R. J. Buhr*¹, A. Jr. Hinton¹, K. D. Ingram¹, D. V. Bourassa², and D. L. Fletcher², ¹Russell Research Center USDA-ARS, ²University of Georgia.

A series of processing trials was conducted to determine the relative persistence of *Salmonella* applied to breast skin of feathered and "featherless" (scaleless) broilers that were scalded and picked 1 h after application. The breast skin surface of feathered and featherless broilers with matched body weights were inoculated with 1 mL of 10^7 *Salmonella typhimurium* suspended in the cecal contents and returned to coops. Control broilers were not inoculated. Feathered or featherless broilers were processed three at a time in eight consecutive batches; inoculated followed by non-inoculated feathered or featherless broilers. The breast skin was aseptically removed from the carcass and stomached in peptone water. *Salmonella* was enumerated in an aliquot of rinsate from each skin sample, and another portion of each rinsate was enriched for *Salmonella* to detect the presence of low levels of the human pathogen. At the beginning of each processing day broilers that were not scalded and picked were sampled without inoculation (0 of 12 *Salmonella* positive) and 1 h after inoculation (recovery at \log_{10} 5.7 cfu / g of breast skin for feathered and \log_{10} 3.8 for featherless broilers). Enrichment procedures detected *Salmonella* in 19 of 22 samples in which the number of *Salmonella* was too low to be detected through enumeration by direct plating. The prevalence of recovery following enrichment from inoculated feathered broilers was 50% (6 / 12 positive) and was similar ($P = 0.6818$) for featherless broilers 58.3% (7 / 12). The prevalence of recovery from the subsequent batch of non-inoculated broilers was 33.3% (4 / 12) for feathered and 41.7% (5 / 12) for featherless broilers ($P = 0.3372$). The presence or absence of feathers and feather follicles on the broiler carcass does not appear to influence the recovery of *Salmonella* distributed on breast skin 1 h prior to scalding and picking. Furthermore, scalding and picking were not able to eliminate *Salmonella* applied to breast skin prior to processing, and *Salmonella* remained within the picker and was distributed onto subsequent non-inoculated carcasses even though the picker was thoroughly cleaned with hot water (>82.2 C; 180 F) between each batch of broilers.

Key Words: feathered broiler, scaleless, *Salmonella*, scalding, picking

80 Persistence of *Salmonella* applied to breast skin between scalding and picking of feathered and featherless (scaleless) broiler carcasses. R. J. Buhr*¹, A. Jr. Hinton¹, K. D. Ingram¹, D. V. Bourassa², and D. L. Fletcher², ¹Russell Research Center USDA-ARS, ²University of Georgia.

Two processing trials were conducted to determine the relative persistence of *Salmonella* applied to breast skin of feathered and "featherless" (scaleless) broiler carcasses between the scalding and picking stages of processing. The first and third carcass in each batch of 4 carcasses were inoculated with 1 mL 10^7 *Salmonella typhimurium* suspended in the cecal contents, while the second and fourth carcasses were not inoculated. The *Salmonella* suspension was spread over the breast skin area after carcasses exited the scald. The breast skin was aseptically removed from the carcass after picking and stomached in peptone water. *Salmonella* was enumerated in an aliquot of rinsate from each skin sample, and another portion of each rinsate was enriched for *Salmonella* to detect the presence of low levels of the human pathogen. Inoculated carcasses that were scalded but not picked were sampled at the beginning and end of

processing to confirm the *Salmonella* level of the inoculum. The level recovered from these nonpicked carcasses was \log_{10} 5.9 cfu / g of breast skin for carcasses sampled at the beginning and \log_{10} 5.5 cfu / g for those sampled 2 h later. Enrichment procedures detected *Salmonella* in 44 of 50 samples (88 %) in which the number of *Salmonella* was too low to be detected through enumeration by direct plating. The prevalence of *Salmonella* recovery from the inoculated carcasses (first and third) was 81.2% (13/16 positive) for feathered and 62% (10/16) for featherless carcasses, and the difference approached significance ($P = 0.0672$). However, the prevalence of recovery from the subsequent non-inoculated carcasses (second and fourth), although significantly higher ($P = 0.0150$) at 88% (14/16), was identical for feathered and featherless carcasses. The presence of cecal contents containing *Salmonella* on breast skin prior to picking does not accurately predict the likelihood of *Salmonella* recovery after picking. Furthermore, the magnitude of cross contamination (88%) that occurs during picking obscures determination of the true impact of the presence or absence of feathers and feather follicles in harboring *Salmonella* on breast skin of broilers.

Key Words: feathered broiler, scaleless, *Salmonella*, picking, cross contamination

81 Effect of time on the recovery of *Campylobacter* from feces in transport cages. M. E. Berrang*, J. K. Northcutt, and J. A. Cason, USDA-Agricultural Research Service, Athens GA 30605.

Feces left in transport cages by a *Campylobacter*-positive flock can cause the spread of *Campylobacter* to the next flock placed in the cages. This experiment was designed to determine how long *Campylobacter* can remain viable in feces deposited on the floor of a cage during commercial transport and holding. Commercial farms were tested for *Campylobacter* by culturing feces. A *Campylobacter*-positive house was chosen to provide the birds for each of two replications. Following 4 hours off feed, broilers were caught by commercial catching crews, placed into three new commercial cages and transported to the holding shed at a commercial processing plant. Broilers were allowed to remain in the cages for eight hours before being unloaded, resulting in a 12 h total feed withdrawal. Following removal of the broilers, cages were held on a trailer under a shed. The cages were sampled at time intervals for the presence of viable *Campylobacter*. Sampling was done by scraping all the feces out of a different randomly assigned compartment in each cage at: 30 min, 2 h, 4 h, 6 h, 8 h, 24 h, and 48 h after unloading. Mean number of *Campylobacter* cfu recovered from feces was calculated from the three cages at each sample time for each replication. No decrease in *Campylobacter* numbers was noted through 8 h after unloading. In both replications, *Campylobacter* was detected in only 2 of 3 compartments by direct plating and detected in the third by enrichment 24 h after unloading. After 48 h of cage holding, *Campylobacter* was detected in one replication only by enrichment, and was not detected in the second replication at all. Contaminated feces left to dry in a cage for 24 h has recoverable numbers of *Campylobacter*. Some *Campylobacter* may remain viable in feces allowed to dry for 48 h in a cage. Allowing a transport cage to dry for 24 to 48 h between uses may lower the numbers of *Campylobacter* the next flock is exposed to, but cannot be expected to eliminate that exposure altogether.

Key Words: *Campylobacter*, Transport, Cage, Feces, Broiler

82 Pressurized marination of broiler breast fillets. D. P. Smith* and L. L. Young, USDA, ARS, Russell Research Center.

An experiment was conducted to determine the feasibility of pressurized marination for broiler breast fillets. In each of two replicate trials, two replications of two treatments (pressure tumble marination, PRESS, or vacuum tumble marination, VAC) were assigned ten fillets each ($n=80$). Postchill broiler carcasses from a commercial plant were obtained and held 4 h postmortem, fillets deboned, weighed, individually tagged, and marinated in a 15% (added weight) solution (92% water, 6% salt, 2% STPP) in both treatments. PRESS tumble settings were 15 PSI at 14 RPM for 15 min.; VAC tumble settings were 15 mm Hg at 14 RPM for 15 min. PRESS and VAC tumble chambers were similar size and shape, with interior baffles installed. Fillets were then weighed, cooked, reweighed, and duplicate slices of each sheared by the Warner-Bratzler method. Percent pickup, drip loss, cook loss, and overall yield were calculated. There was no significant difference ($P < 0.05$) between PRESS and VAC for pickup (11.4 vs. 12.1%, respectively) or shear (5.4 vs. 4.9 kg, respectively). PRESS was significantly lower than VAC for drip loss (0.1 vs. 2.2%, respectively) and cook loss (6.9 vs. 8.8%, respectively).

PRESS yield (93.1%) was significantly higher than VAC yield (91.2%). Marination of breast fillets with pressure tumbling may be an acceptable alternative to vacuum tumbling, improving overall yield without increasing toughness.

Key Words: pressure marination, breast fillets, yield, shear, vacuum tumbling

83 Efficacy of DBDMH (1,3-Dibromo-5,5-Dimethylhydantoin) as a disinfectant on chill tank water and carcass bacteria populations. James L. McNaughton*¹, Michael S. Roberts¹, and Robert W. Kuhlmeier², ¹*Solution BioSciences, Inc.*, ²*Albemarle Corporation*.

A critical step in poultry processing HACCP food safety plans is reduction of bacteria during the chilling process. Although oxidizing biocides based on Cl₂ & Br₂ are recognized as effective microbiological controls, Br₂ is more effective in wide pH ranges and more stable in high organic loads than Cl₂. Trials were conducted to determine the efficacy of DBDMH (containing only bromine or Br₂ and zero Cl₂), when used in the chill tank during processing, on controlling poultry chill tank water and carcass bacteria (*E. coli* or EC, *Campylobacter* or CAM & *Salmonella* or SAL). For each treatment, six reps of 5 birds each (1950g), were ice-chilled for 80 min under typical commercial processing conditions. After each trial, water and whole carcass bacteria, moisture uptake, and any adverse carcass quality factors were noted. In Trial 1, prior to disinfectant addition, chill tank water was inoculated with 10⁷ of each bacteria (EC, CAM, & SAL) source per mL of water. Post-processing water and whole carcass EC, CAM, & SAL were significantly (P<0.05) reduced by 3.5-4.5 log₁₀ and 99.8937-99.9981% bacteria reduction with 34-56 ppm Br₂ from DBDMH as compared to the control (no disinfectant). Similar reductions were found across all bacteria strains tested. In Trial 2, 10⁹ per carcass of gene-marked *SAL Agona & Kentucky* was spotted and dried for 30 min in order to simulate pre-chill fecal bacteria contamination. Post-processing whole carcass SAL was significantly (P<0.05) reduced by 7.5 log₁₀ and 99.9999% bacteria reduction with 34 ppm Br₂ (15 ppm Cl₂ equivalence) from DBDMH and by 5.0 log₁₀ and 99.9985% bacteria reduction with 15 ppm Cl₂ from NaOCl as compared to the control (no disinfectant). The conclusions are made that DBDMH is an effective biocide in significantly reducing bacteria loads in chill tanks and that bromine chemistry is more effective than chlorine chemistry. No carcass quality effects were found.

Key Words: 1,3-Dibromo-5,5-Dimethylhydantoin, Bromine, *E. coli*, *Campylobacter*, *Salmonella*

84 Development of Automated Devices to Sort Poultry Eggs By Gender. S. Neuman*, S. Bryan, A. Chalker, W. Ferrell, M. Schnupper, P. Strayer, E. Gross, and P. Phelps, *Embrex Inc., Research Triangle Park, NC*.

Although manual sex separation of chicks at hatch is an important aspect of the poultry industry, it is a laborious process which significantly increases handling stress to the chick. Thus, an automated system to sort eggs by gender is under development. The system consists of three independent gender sort devices linked by a data transaction network. The first module, or sampling device, removes allantoic fluid (AF) from live embryonated Day 16 eggs. The AF is dispensed into individual microwells on a bar-coded assay template. Sampled eggs are then placed in bar-coded flats and put back in the incubator(s) until *in ovo* vaccination on Day 18 of incubation. The second device is an automated assay module which distinguishes each AF sample as male or female after dispense of a live cell-based sensor, which consists of a genetically modified yeast transformed with a yeast expression vector for the human estrogen receptor. Coupled with a colorimetric substrate to yield a yellow signal, pixel counts of each well are read with a camera system after a timed incubation. The signal increases in intensity in the presence of estrogenic compounds in female AF, while little or no signal is produced from male AF samples. A computer correlates each male and female AF sample on a template to the appropriate eggs in bar-coded flats. On Day 18 of incubation, sampled eggs are removed from the incubator and placed on a third device which segregates, vaccinates, and transfers eggs according to gender. Each bar-coded flat is automatically scanned and male and female eggs are placed via vacuum transfer onto separate male and female conveyors in order to undergo sex-separate vaccination and

transfer into hatching baskets. This commercial prototype system is currently undergoing testing at a commercial hatchery. Preliminary results of these trials will be discussed.

Key Words: Gender, Sort, Sexing, Allantoic fluid, Embryo

85 Use of Statistical Process Control to assure finished product standards for chicken paws during processing. S.F. Bilgili*¹, D. Zelenka², and J.E. Marion³, ¹*Auburn University*, ²*Tyson Foods, Inc.*, ³*National Chicken Council*.

Processed chicken feet (paws) have been a unique and economically important export item for the broiler industry in the US. Hong-Kong and China are the two major destinations for chicken paws, with China recently requiring USDA mark of inspection on all imported paws. Although USDA provides sanitation supervision during harvesting, chilling, and packaging processes for un-inspected paws, inspection mark requires post-mortem inspection. Paw inspection is based on condemnation disposition criteria that includes signs of systemic diseases (septicemia and toxemia, leukosis complex) and localized conditions with evidence of systemic involvement (wounds and ulcers). All inspected paws must meet USDA Finished Product Standards (FPS). Statistical Process Control (SPC) is a widely accepted process control tool that can be used to meet FPS (both trimmable defects and processing errors) and to assure the safety and wholesomeness of paws. In 18 volunteer broiler processing plants (10 producing un-inspected paws based on Hong Kong and 8 producing based on new China paw specifications), 100 paws were collected (50 post-chiller and 50 post sorting) for ten days. Each paw was then scored for localized trimmable defects (calluses, bruises, and fractures) and processing errors (remnants of cuticle, mutilations, and extraneous material). Data was grouped by defect, specification (size of the defects and errors) and sampling location. Data from individual plants were then plotted as an np control chart (n=50) and compared to control limits set on upper 80th percentile for the entire data set. The data collected represented average process quality, as paws were scored prior to plant grading. The analysis of SPC charts indicates that 80th percentile value represents a reasonable quality expectation. Furthermore, adoption of a SPC based system would allow individual plants to improve process capability.

Key Words: Broiler, Paws, SPC

86 Microbiological quality of broiler chicken feet (paws). S.F. Bilgili*¹, D. Zelenka², and J.E. Marion³, ¹*Auburn University*, ²*Tyson Foods, Inc.*, ³*National Chicken Council*.

Poultry feet (paws) are considered edible and desirable product as human food in many parts of the world. Until recently, many processing plants in the US have produced and exported paws under the sanitation supervision of USDA, but without the USDA inspection certification. To receive the mark of USDA inspection, a recent requirement for paws exported to the People's Republic of China, the feet must be inspected post-mortem. Paws from carcasses with systemic disease conditions and those with localized lesions with systemic involvement are considered unwholesome and condemned. In addition to meeting sanitation requirements, plants producing inspected paws must address food safety hazards likely to occur as part of their HACCP plans. The purpose of this study was to compare the microbiological quality of un-inspected and inspected paws, under commercial conditions. Seventeen broiler processing plants (nine producing un-inspected and eight producing inspected paws) participated in this field study. In each plant, five paws were sampled per day for ten days and microbiologically evaluated for Aerobic Plate Counts (APC) and *Escherichia coli* counts and *Salmonella* incidence. Paws were selected post-chill and after sorting, placed in sterile bags containing 25 ml of buffered peptone, and shaken for 60 sec. Count data (CFU/ml) were Log₁₀ transformed prior to analysis. Incidence data was analyzed by Chi-square test. Although individual plants varied in paw APC and *E. coli* counts, there were no statistically significant differences (P>0.05) due to the inspection process. Average *E. coli* levels were well below one CFU/ml of rinse for both un-inspected and inspected paws. Similarly, *Salmonella* incidence did not vary (P>0.05), averaging 1.56 and 1.51% for un-inspected and inspected paws, respectively. It is clear from the results of this study that paw microbial counts are extremely low, regardless of post-mortem inspection, due to the nature of the paw production process.

Key Words: Broilers, Paws, Microbiology

87 Effect of thickness and fat content on surface heating rate of ready to eat sliced meat bologna during in package pasteurization. S. Mangalassary*¹, I.Y. Han¹, and P.L. Dawson¹, ¹Clemson University.

In package pasteurization of ready to eat meat products is used as an effective method to reduce post processing surface contamination. But often there is lack of attainment of required surface temperature in a short time due to heat sinking effect of meat. While numerous studies report on the effect of meat thickness and composition on heat penetration, there are limited studies on their effect on surface heating rates.

The objective of the study was to evaluate the effect of thickness and fat content of the meat on the surface heating rate during in package pasteurization of sliced meat bologna.

Bologna containing 2 different fat levels(13 and 18%), 3 different thickness levels(4, 12 and 20 mm)corresponding to 1, 3 and 5 slices of bologna and 4 different pasteurization temperatures(60, 70, 80 and 90°C)were used in this study. Teflon insulated thermocouples were placed on the surface of samples and then each was vacuum packaged in separate LLDPE pouches. Packaged samples were kept at refrigeration temperature overnight and then directly immersed in a water bath at set temperature for pasteurization. The time and surface temperature were monitored using thermocouples and a data logger for a period of 10 minutes. The surface heating rate was slower with increased thickness levels and also slower in bologna with higher fat content. In bologna with higher fat content (18%), the 4mm thickness sample took an average time of 2.5 minutes to reach the surface temperature of within a degree of set pasteurization temperature of 60 °C and the 12 mm thickness sample took an average time of 8 minutes. The thickest sample (20 mm) could not attain the set pasteurization temperature at the surface within the 10 minutes. Three thickness levels from the bologna with lower fat content (13%) took less time to attain the set pasteurization temperature on the surface compared to the higher fat content one. The trend was similar in other three pasteurization temperatures used in the study.

Key Words: meat bologna, in package pasteurization, surface heating rate, meat thickness, fat content

88 Real composition of poultry exudates versus current fluids used for testing; impact on food pad testing and performance in the field. G. Bland*¹, ¹Lysac Technologies Inc..

Packaging appearance and safety are very important for consumers, and poultry processors are always looking for tools to improve these elements. Lysac's R&D team has searched for causes and solutions to leakage from poultry packaging.

Extensive research showed that analysis based on real poultry exudates produced different results compared to the fluids currently used by the industry to test food pads (water or saline solution). Some of the consequences are explored here.

The composition of poultry exudates from packages containing various brands and pieces of poultry were subjected to chemical and physical analysis. Viscosity was measured, as were levels of water, proteins, minerals, carbohydrates and lipids. The results were compared to water and to saline solution.

Poultry processors demand increasing levels of absorbency in food pads. To meet their needs, food pad manufacturers are introducing superabsorbents in pad structures. Since the performance of superabsorbents is affected by the composition of liquids, their absorption capacity was analysed with real poultry exudates. Theoretical analysis of the effect of exudates on the absorption capacity of superabsorbents used in food pads was verified by comparing the absorption capacity of various types of superabsorbents (SNAPs and SAPs) in different food pad structures. The study concluded that absorption levels vary between water or saline solution and exudates as well as from one food pad structure to another, with SAPs or SNAPs. The current industry standard specifying food pad performance in 0.2% saline solution leads to differences between the expected performance of food pads and real performance in the field. In order to perform accurate tests and avoid major gaps between expected performance and real performance, Lysac developed a distinctive synthetic exudate formulation that replicates real exudates. This unique formulation is useful to poultry processors in guaranteeing the integrity of food packaging that reaches food store shelves.

Key Words: Food/meat soaker pads, Test methods, Poultry, Exudates, Superabsorbent

Monday, January 20, 2003

SCAD/Avian Diseases

Room:B312

Moderator(s):Maricarmen Garcia & Adrea Miles

89 Spread of blackhead disease within flocks of chickens or turkeys without the aid of cecal worm carriers. J. Hu¹ and L. McDougald*¹, ¹Department of Poultry Science, University of Georgia.

Experiments were conducted to determine whether direct bird-to-bird transmission of *Histomonas meleagridis* in the absence of cecal worms or other carriers was important in the spread of outbreaks through a flock. The turkey poults were placed in floor pens on clean litter and exposed to infection by introduction of turkeys infected cloacally with cultured *Histomonas meleagridis*. All birds in the high exposure group (25 percent inf.) became infected and died of blackhead within 23 days PE. Birds in the low exposure group (10 percent inf.) became infected and died or had severe lesions of blackhead by day 31 PE. Unexposed birds had no signs of infection at the end of the experiment. A similar floor pen experiment with chickens showed that broiler-type birds subjected to feed restriction (skip-a-day) were more likely than full-fed birds to contract histomoniasis, although infections were relatively mild. Water acidification reversed the effect of feed restriction in a battery experiment. These results suggest that *H. meleagridis* is more contagious within flocks of turkeys and chickens than previously thought, and that transmission is possible between birds without the presence of cecal worms or other carriers. Cecal worms (*Heterakis gallinarum*) are the primary reservoir of infection, but need not be present for the infection to spread through the flock. The possibility that feed management contributes to outbreaks of blackhead disease in broiler breeder pullets needs further investigation.

Key Words: *Histomonas meleagridis*, Blackhead disease, Transmission

90 Evaluation of the salmonella vaccine PoulVac ST in commercial broilers. M. D. Sims*¹, J. D. Maiers², K.C. Cookson², and D.M. Hooge³, ¹Virginia Scientific Research, Inc., Harrisonburg, VA, ²Fort Dodge Animal Health, Fort Dodge, IA, ³Hooge Consulting Service, Inc., Eagle Mountain, UT.

A 49-d floor pen study was conducted with 1,920 Ross x Ross broiler chicks positive for salmonella to compare the performance of PoulVac ST vaccinated broilers to broilers not vaccinated for salmonella, with or without a BMD/Stafac growth promotant feed shuttle program. There were four treatment groups: 1) Non-vaccinated without growth promotants (NVU); 2) PoulVac ST Vaccinates without growth promotants (PVU); 3) Non-vaccinates with growth promotants (NVG); 4) PoulVac ST Vaccinates with growth promotants (PVG). There were eight replicate pens of 60 birds each per treatment at an initial stocking density of 0.7 ft² per bird. Broilers were fed corn/soy diets. Data collected were live weights, feed conversion, mortality, costs, carcass rinse for salmonella, and pen drag swabs for salmonella. Mortality and flock homogeneity were not significantly ($P \geq 0.05$) affected by the use of the salmonella vaccine. The 21-d feed conversion ratio for NVG birds was lower ($P < 0.05$) than NVU broilers. Final (49-d) average live weights were heavier for the groups receiving PoulVac ST in each paired comparison as well as in the pooled comparison (main effect). Final feed conversion ratios of the PVU birds were significantly lower ($P < 0.05$) than those of the NVU group. Pooled final feed conversions were significantly lower ($P < 0.05$) for the Poul-Vac ST treated birds than the Negative Controls (non-vaccinated). Feed, feed additive and vaccine input costs/bird of the four groups were not different ($P \geq 0.05$) while Revenue/Bird of the PVU group was greater ($P < 0.05$) than that of the NVU group. Pooled Revenue/Bird of the PoulVac ST groups was greater ($P < 0.05$) than that of the pooled Negative Controls. Pen drag swabs were positive for salmonella in all treatment groups. Carcass rinse samples collected at processing indicated that the PVU group

had approximately 75% fewer salmonella positive carcasses ($P < 0.05$) than the other 3 treatment groups. It was concluded that PoulVac ST vaccine administered at the hatchery followed by a booster at 14 days provided substantial cost effective protection to broilers allowing performance to be minimally affected by salmonella.

Key Words: Broilers, Floor Pen, Salmonella, Vaccine

91 Development of Recombinant Flagellar Antigens for Serological Detection of *Salmonella* in Poultry. Joseph Minicozzi*, Margie Lee, Charles Hofacre, Peter Holt, and John Maurer, *University of Georgia, Athens Georgia.*

A biological hazard can be introduced at any point during food production, from the farm to the fork. *Salmonella* contamination is a constant concern in the poultry industry. By quickly and accurately identifying *Salmonella* contaminations of poultry flocks, appropriate course of action can be taken during processing. Using current recombinant DNA, protein-fusion technology, *Salmonella* flagellar antigens representative of the four major pathogenic serotypes: *S. typhimurium*, *S. enteritidis*, *S. heidelberg*, and *S. hadar*, have been developed and produced for a serological test, free of cross-reactive antigens for screening poultry for these specific serotypes. Phase 1 flagellar antigen, *fljC*, and phase 2, *fljB*, were fused to a biotinylated protein and purified using a streptavidin column. Protein fusions were visualized via SDS-PAGE and Western blot using Spicer-Edwards flagellar antisera. Sera collected from serotype-known challenged birds were tested for cross-reactivity between protein fusions to determine specificity and sensitivity.

Key Words: Salmonella, Recombinant, Serology

92 A Histological and Immunological Comparison of the Upper Gastrointestinal Tract from Normal and *Salmonella enteritidis*-Challenged Hens. Lara Vaughn*, Peter Holt, and Henry Stone, *USDA, ARS, Southeast Poultry Research Laboratory, Athens, GA.*

The focus of the study was to assess the crop and various other tissues within the upper gastrointestinal tract for a localized immunological response. Tissue regions from both normal and SE-infected hens were observed for the presence or absence of lymphocytic foci/GALT. The upper gastrointestinal tract tissues for comparison consisted of: oropharynx, esophagus, crop (ingluvies) and proventriculus. Seventy-week-old SPF White-leghorn spent hens were orally challenged with *Salmonella enteritidis*, nalidixic-acid resistant, Phage-type 13 strain at 4×10^9 SE cfu/mL. Hens were SE-free before challenge and then the progression of infection was monitored at days 4, 10, 17 and 24 post-SE challenge. Throughout the experiment, routine fecal and tissue cultures were performed to confirm positive SE-infection. Crop lavage, gut flush and serum samples were assessed for antibody titers specific for SE via ELISA. On day 24 post-SE infection, hens positive for SE and normal control hens were sacrificed. The upper gastrointestinal tracts from both groups were harvested into 10% buffered formalin. After proper fixation, the appropriate tissue regions were sectioned and submitted for routine H&E staining. The upper gastrointestinal regions from normal and SE-infected hens were then observed for the presence or absence of lymphoid tissue by light microscopy. The upper GI tract histopathology interpretations were then compared to the ELISA SE-specific antibody titer results from day 24 post-SE crop lavage, serum and gut flush samples. The ELISA results revealed progressively elevated SE-specific antibody titers in all samples. There was a finding of increased lymphoid tissue (GALT) presence within the upper gastrointestinal tract at day 24 post-SE infection.

Key Words: Salmonella enteritidis, Gastrointestinal tract, Mucosal Immunology, Food Safety

93 In Vivo Transfer of Antibiotic Resistance Genes between Resident Intestinal Microflora of Broiler Chickens and *Salmonella typhimurium*. Tameka Buffington*¹, John Maurer¹, Charles Hofacre¹, Margie Lee², and Karen Liljebjelke¹, ¹*University of Georgia, College Veterinary Medicine, Dept. of Avian Medicine,* ²*University of Georgia, College Veterinary Medicine.*

An *in vivo* study was conducted in order to examine the transmissibility of antibiotic resistance genes from the intestinal microflora of poultry to salmonella in healthy commercial broiler chickens. Newly hatched specific-pathogen free (SPF) broiler chicks were orally inoculated with two *Salmonella typhimurium* nalidixic acid-rifampicin resistant isolates

(107 cells/ml). The isolates are genetically identical, except that one carries a Class I integron containing no gene cassettes. Chicks were subsequently administered litter microflora containing resistant enteric bacteria. At 1 week of age, the birds were administered tetracycline or streptomycin per os. For 6 weeks, the typical maturation period of commercial poultry, resistance transfer to *Salmonella* was monitored weekly by plating tetrathionate enrichments of litter/ drag swabs onto XLT4 selective media containing antibiotics: ampicillin, chloramphenicol, gentamicin, kanamycin, streptomycin, or tetracycline. At the end of the six-week period, ceca were harvested and resistant *Salmonella* were enumerated using a modified three-tube MPN procedure. Transfer of antibiotic resistance genes from the gut flora of chickens to *Salmonella* occurred. The highest prevalence of resistance transferred was to ampicillin, followed by tetracycline, gentamicin, and kanamycin. There was no transfer of chloramphenicol resistance detected. However, prior to antibiotic treatment with tetracycline or streptomycin, resistant *Salmonella* were isolated. This suggests that antibiotic selective pressure is not required for resistance to emerge.

Key Words: Salmonella, Broiler, In vivo, antibiotics, resistance

94 The influence of early *Campylobacter sp.* cecal colonization on the growth and mortality of turkeys during the brooding period. NDT Reimers*¹, HJ Barnes¹, S Kathariou², JP Vaillancourt¹, S Kozlowicz¹, and J Hartsell¹, ¹*College of Veterinary Medicine, NC State University,* ²*College of Agriculture and Life Sciences. NC State University.*

A prospective epidemiologic study was performed in which 19 commercial turkey flocks were followed for growth, mortality, and *Campylobacter sp.* incidence through 5 weeks of age. All nineteen flocks had *Campylobacter sp.* isolated from ceca during the brooding period. Most flocks had reasonable brooding period performance in spite of the colonization, however six of the flocks experienced mortality severe enough to meet the clinical definition of Poult Early Mortality Syndrome (PEMS). Two of those six had a high incidence of *Campylobacter sp.* recovered from the ceca in the first week coupled with extremely high mortality in the second week.

Key Words: PEMS, *Campylobacter*, Stunting, Mortality, Poult

95 The effect of air sacculitis on bird weights, uniformity, fecal contamination, processing errors, and populations of *Campylobacter spp.* and *Escherichia coli*. S.M. Russell*¹, ¹*The University of Georgia.*

The effect of air sacculitis in broiler chickens on yield, fecal contamination, processing errors, and *Campylobacter* and *Escherichia coli* populations was determined. During processing, carcasses from air sacculitis positive (AS) and air sacculitis negative (ASN) flocks were collected, weighed, evaluated for cut or torn areas on the digestive tracts, and assessed for *Campylobacter* and *Escherichia coli* counts. Additionally, fecal contamination (FC) was monitored. AS reduced carcass weight averages in 2 of 5 reps. The net loss for AS birds equated to a loss of 32,379 lbs/house/year. AS carcasses had significantly higher FC in 4 of 5 reps. The number of digestive tract cuts or tears were much higher on AS carcasses at 42, 49, 37, 60, and 59 % as compared to 14, 12, 17, 24, and 16 % for ASN carcasses in Reps 1 to 5, respectively. In three of the five replications, the presence of air sacculitis in the flocks significantly increased the number of *Campylobacter* recovered from broiler carcasses. Hence, there appears to be a relationship between the presence of air sacculitis and *Campylobacter* positive carcasses. *Escherichia coli* counts for AS flocks were significantly higher than ASN flocks in Reps 1 and 3. In Rep 5, *E. coli* numbers were significantly lower for the AS flock. Previous unpublished research showed that *E. coli* counts for AS flocks are significantly higher than ASN flocks. The difference may be because, in the pilot studies, visibly infected carcasses were sampled and in this study, healthy birds that had passed inspection were sampled within an AS flock. Because flocks of chickens showing signs of air sacculitis had lower weights, more FC, more processing errors, and higher levels of *Campylobacter spp.*, broiler companies should emphasize control of air sacculitis as a means of preventing subsequent food-borne bacterial infection.

Key Words: Air Sacculitis, Processing Errors, *Campylobacter*

96 Clonal relationship among Romanian avian *Escherichia coli* isolates determined by random amplified polymorphic DNA technique - RAPD. Virgilia Popa*¹, Monica Vanghele¹, Daniela Botus¹, Andreea Stanica¹, A. Popovici¹, Doina Sofei¹, and Doina Danes², ¹N.S. Pasteur Institute S.A., ²University of Agronomic Sciences and Veterinary Medicine.

The genetically diversity of 40 avian *Escherichia coli* (and 1 *E. fergusonii*) strains, 8 isolates with Congo Red binding (CRb+) phenotype (2 clinical and 5 commensal respectively), 14 clinical isolates (2 CRb+) and 25 commensal isolates (6 CRb+) from three large scale farms was evaluated by comparing the RAPD patterns obtained with 4 commercial primers (P1, P2, P3 and P4) characterized by different Simpson indices (0.9670, 0.9219, 0.9268 and 0.8790 respectively). With the P1 primer, 60.0% of the strains fell in 8 of 23 genotypes (RAPD Types or RT), the P2 primer grouped 70.5% of the strains in 7 RTs of 19, the P3 primer generated 20 RTs, 3 of which included 45.3% of the strains while the P4 primer discriminated 8 RTs, 3 of which included 53% of the strains. Although they were not prevalent, exclusive RTs for the three isolates classes were recorded with the P4 primer. No geographically or host restriction were noticed in the RTs distribution. The biochemical and antibioresistance characteristics were randomly associated with various RTs.

Key Words: poultry, *Escherichia coli*, RAPD

97 Treating an *Escherichia coli* Respiratory Infection with Bacteriophage Administered as an Aerosol Spray or Intramuscular Injection. W. E. Huff*, G. R. Huff, N. C. Rath, J. M. Balog, and A. M. Donoghue, USDA/ARS/PPPSRU, Fayetteville, AR.

Two studies were conducted to determine the efficacy of either aerosol or intramuscular (im) injection of bacteriophage to treat an *E. coli* respiratory infection in broiler chickens. In Study 1 the birds were challenged with *E. coli* at 7 d of age followed by spraying the birds with either heat killed phage or active phage at 2, 24, or 48 h after challenge. In Study 2 the birds were challenged with *E. coli* at 7 d of age followed by injection of either heat killed or active phage immediately after challenge, or 24, or 48 h after challenge. In both studies the *E. coli* challenge consisted of injecting 10⁴ cfu into the thoracic air sac. There were 3 replicate pens of 10 birds per pen per treatment. Two studies were conducted to enumerate the phage in the blood of birds 1, 2, 3, 4, 5, 6, 24 and 48 h after either being sprayed or injected with phage. There were 5 birds at each time period in this study. Treating this severe *E. coli* infection with the phage aerosol spray significantly reduced mortality from 50 to 20% when given immediately after the challenge, but had little treatment efficacy when administered 24 or 48 h after challenge. The im injection of phage significantly reduced mortality from 53 to 17%, 46 to 10%, and 44 to 20% when given immediately, 24, or 48 h after challenge, respectively. Only a few birds sprayed with phage had detectable phage in their blood with an average of 956 pfu per mL 1 h after phage administration and no phage was present 24 or 48 h after phage administration. All the birds injected im with phage had detectable levels of phage in their blood at levels of 10⁴ pfu per mL of blood up to 6 h after phage administration, and 4 of the 5 birds had detectable phage in their blood at an average level of 70 pfu per mL 24 h after phage administration. The relative inefficiency of the spray treatment to the im injection treatment may be due to the inability to get phage in the blood at high concentrations when the birds are sprayed versus the consistent high titers achieved with the im injection of phage. These data suggest that phage may be an effective alternative to antibiotics in animal production when they are administered in a way that delivers high titers of the phage to the critical site of the infection.

Key Words: Bacteriophage, *Escherichia coli*, Chickens

98 Examinations of Reasons for Field Problems with *Eimeria maxima*: Part 1: *Eimeria acervulina* versus *E. maxima*. Greg M*¹, ¹Southern Poultry Research, Inc..

The three most commonly occurring species of *Eimeria* infecting chickens are *Eimeria acervulina*, *E. tenella*, and *E. maxima*. Even though *E. maxima* is very immunogenic, lesions are often observed in the field late in a growout. A survey of 50 coccidial field isolates showed that 36 were predominately *E. acervulina*, 4 *E. maxima*, and 10 *E. tenella*. All of the *E. maxima* isolates came from farms where the broilers were over 28 days old. Most of the *E. acervulina* isolates were from broilers that were 18

to 28 days old. The daily oocyst shedding pattern for a commercial coccidial vaccine was examined in floorpen birds. Birds vaccinated for coccidiosis at the hatchery were placed into pens on new pine shaving. The shedding of *E. acervulina* type oocysts peaked around 18 days. A small peak of *E. maxima* was observed around 28 days. A battery cage study was conducted to examine whether *E. acervulina* could be interfering with *E. maxima* development. Birds were challenged at 14 days of age with *E. acervulina* and/ or *E. maxima*. The oocyst per bird challenge levels were none (Trt. 1), *E. acervulina* 100,000 (Trt. 2), *E. acervulina* 100,000 plus *E. maxima* 5,000 (Trt. 3), *E. acervulina* 50,000 plus *E. maxima* 5,000 (Trt. 4), *E. acervulina* 25,000 plus *E. maxima* 5,000 (Trt. 5), and *E. maxima* 5,000 (Trt. 6). Each treatment consisted of 3 replications in a complete randomized block design. *E. maxima* alone caused 21 % weight reduction and 2.75 lesion score. The 100,000 and 50,000 *E. acervulina* oocyst level reduced *E. maxima* lesions to 1.33. The 25,000 *E. acervulina* oocyst level only slightly reduced *E. maxima* lesions to 2.25. The *E. maxima* did not interfere with any of the *E. acervulina* infections. This study suggests that *E. acervulina* interferes with colonization or development of *E. maxima* development. As birds become more immune to *E. acervulina* then *E. maxima* has more of an opportunity to develop.

Key Words: *E. acervulina*, *E. maxima*, Coccidiosis, Broiler, Lesions

99 Comparison of several challenge models and isolate characteristics for studies in avian colibacillosis. P. S. Gibbs*¹ and R. E. Wooley², ¹North Dakota State University, Fargo, ND, ²University of Georgia, Athens, GA.

Demonstrating that the embryo lethality assay is a challenge model that produces results similar to chicken challenge studies could ease studies involving virulence in avian *Escherichia coli*. Twenty avian *E. coli* isolates used extensively in previous studies were used in both intravenous (IV) and cellulitis challenge studies for the purpose of comparing the results of each with the previous embryo challenge studies. Results indicate that the combination of mortality and morbidity resulting from IV challenge of 3-week old broiler chickens correlates well with the two previous embryo challenge studies ($r_2 = .861$, $r_2 = .830$ for embryo studies 1 and 2, respectively); both had resulting p-values <0.0001. The correlation of the mortality and morbidity resulting from the cellulitis challenge and the two previous embryo lethality studies were not as high ($r_2 = .792$, $r_2 = .738$ for studies 1 and 2, respectively); these resulting p-values were <0.0001 and 0.0002, respectively. Interestingly, several isolates varied in their ability to cause mortality in the IV and cellulitis challenge models. The most significant isolate traits correlating with mortality were the presence of Colicin V production and the increased serum survival gene; presence of the temperature sensitive hemagglutinin (tsh) was not significantly correlated with mortality in the IV challenge; however, tsh was significantly correlated with combined mortality and morbidity results from IV challenge and the mortality of the cellulitis challenge.

Key Words: Avian Colibacillosis, Colibacillosis Challenge Models, Embryo Lethality Assay

100 Evaluation of Coccidiosis Vaccines by Different Methods of Administration. S Thompson*, T Cherry, W Weatherford, J Brumley, J Bray, J David, S Antle, C Roberts, and E Oviedo, Stephen F. Austin State University.

The purpose of this experiment was to evaluate a coccidiosis vaccine by different methods of administration as compared to commonly used coccidiostat programs. The evaluation was based upon weight gain, feed conversion, lesion scoring, oocysts shedding, and mortality. The trial consisted of 7 treatments (Trt). They consisted of: Trt 1 - 60 grams/ton of Salinomycin in starter and grower feed; Trt 2 # 90 grams/ton of Moneinsin in starter and grower feed; Trt 3 - 0.2 ml gavage of coccidiosis vaccine; Trt 4 # 21 ml coccidiosis vaccine spray; Trt 5 # 14 ml coccidiosis vaccine spray; Trt 6 # 25 ml coccidiosis vaccine spray; and treatment Trt 7 # coccidiosis vaccine on Oasis; The study was conducted in a cool cell tunnel ventilated floor pen facility. There were 3528 birds randomly assigned to pens with 8 replications per treatment containing 63 birds each. All treatments included 50 gm/ton of Bacitracin in starter, grower, and withdrawal feed. With regard to 45 day body weight and adjusted feed conversion, there was no statistical difference between the vaccines and the coccidiostats.

Key Words: Coccidiosis, Coccidiostat, Coccidiosis Vaccine, Poultry Diseases, Coccidiosis Experiments

101 Comparative Anticoccidial Efficacy and Performance of Arsenical Feed Additives in Broiler Chickens. Greg Mathis*¹, Phil Pearson², and George Fleming², ¹*Southern Poultry Research, Inc.*, ²*Fleming Laboratories, Inc.*

Studies were conducted comparing the anticoccidial efficacy and performance Arsanilic Acid and Roxarsone. A coccidiosis challenge battery study was conducted using 5 treatments with 3 replications in a randomized block design. The treatments were Nonmedicated, Uninfected (NMU), Nonmedicated, Infected (NMI), Salinomycin 60 g/t, Infected, Salinomycin (SAL) 60 g/t plus Arsanilic Acid (AA) 90 g/t, Infected, and Salinomycin 60 g/t plus Roxarsone (ROX) 45.4 g/t, Infected. At 12 days of age, 10 birds were weighed and issued treatment feed. Two days later birds were orally inoculated with 50,000 oocysts of *E. tenella*. Six days post inoculation all birds were weighed and cecal lesion scored. Measurements were weight gain, feed conversion, cecal lesion score, and coccidiosis mortality. The NMI birds had significantly lower performance, higher lesion scores, and more coccidiosis mortality than the other treatments.

SAL alone was only significantly better than NMI. AA and ROX had very similar performance and anticoccidial efficacy with reduced lesion scores to 0.5 or less. For both treatments, feed conversions and weight gain were not significantly different from NMU. A built up litter floor pen study was conducted to simulate summer time conditions using 3 treatments with 8 replications a randomized block design. The treatments were SAL 60 g/t, SAL 60 g/t plus AA 90 g/t, and SAL 60 g/t plus ROX 45.4 g/t. The parameters measured were adjusted feed conversions and average live weight on Day 42 and 52. AA and ROX birds had significantly better performance than SAL alone. AA had significantly heavier male and average live weights than ROX fed birds. Total hot weather related mortalities showed SAL alone had the most, followed by ROX, and then AA. The results of these studies confirm that Arsanilic Acid 90 g/t has anticoccidial efficacy equal to Roxarsone 45.4 g/t, that both will improve performance over Salinomycin alone, and that Arsanilic Acid 90 g/t can safely be used in summer time conditions.

Key Words: Arsanilic Acid, Roxarsone, Chicken, *E. tenella*, Coccidiosis

**Tuesday, January 21, 2003
Environment & Management
Room:B314**

102 BioScience Air Sampler vs. Settling Plates for Microbiological Air Samples. Y Vizzier-Thaxton*, T. E. Arbourgh, H.S. Renschler, and D. Wilbourn, *Mississippi State University, Mississippi State, MS.*

Research into the epidemiology of Salmonella and other human pathogens within poultry the U.S. poultry industry has made it clear that control must be maintained within the commercial hatchery. In order to verify control it is necessary to have a method of testing. For many years the industry has relied on settling plates to determine the microbiological quality of the air through out the hatchery. The method is easy to perform and relatively inexpensive. However, the sampling process is time consuming and subject to many variables. The BioScience International SAS Super 180 Microbial Air Sampler was tested as a way to reduce the time of sampling, control the variables and increase accuracy. Tests were run comparing 3 different volumes of air through the sampler to 3 exposure times for settling plates. The results indicate that the sampler is a superior method of estimating microbial air quality and is quicker to use than settling plates.

Key Words: Air Sampling, Air Quality, Microbiology, Sanitation

103 Translocation of *Campylobacter*, *Salmonella* and *Clostridium perfringens* to serveral lymphoid organs following oral or intracloacal inoculation of broiler chicks. N.A. Cox*¹, C.L. Hofacre², J.S. Bailey¹, R.J. Buhr¹, J.L. Wilson³, D.E. Cosby¹, M.T. Musgrove¹, L.J. Richardson¹, J.D. Tankson¹, Y.L. Vizzier⁴, P.F. Cray¹, K.L. Hiatt¹, L.E. Vaughn¹, P.S. Holt¹, and D.V. Bourassa³, ¹*Russell Research Center, USDA-ARS*, ²*Department of Avian Medicine, University of Georgia*, ³*Department of Poultry Science, University of Georgia*, ⁴*Department of Poultry Science, Mississippi State University.*

Day old broiler chicks were either orally or intracloacally inoculated with a 100ul suspension containing 10⁶-10⁹ cells of one of three marker strains of either *Campylobacter jejuni*, *Salmonella spp.* or *Clostridium perfringens*. At one hour, one day and one week following inoculation, five birds from each group were euthanized by cervical dislocation. The bursa of Fabricius, ceca, liver/gallbladder, spleen and thymus were aseptically removed and individually cultured for the appropriate bacterial species. Each of the inoculation routes for the three bacteria was replicated twice. Regardless of route of inoculation, after one hour *Salmonella* and *Clostridium* were isolated from all allocated sample sites, while *Campylobacter* were only isolated from the ceca, bursa and spleen. By 24 hours, all three bacteria were isolated from all five-sample sites. Seven days post inoculation *Clostridium perfringens* were no longer isolated from any samples and the presence of *Campylobacter* was approximately the same as it was after 24 hours. However, *Salmonella* was isolated from a majority (75%) of all tissues. Results indicate that *Salmonella* translocated more often via the intracloacal route, while *Campylobacter* were more likely to appear in the liver, spleen or thymus via the oral route. No differences between routes were observed for *Clostridium perfringens*. This study demonstrated that *Salmonella*, *Campylobacter* and *Clostridium*

perfringens could rapidly invade and colonize lymphoid organs of the baby chick. Whether or not these bacteria develop long-term reservoirs in breeders and ultimately contribute to broiler flock contamination is not yet known.

Key Words: Broiler, *Campylobacter jejuni*, *Salmonella spp.*, *Clostridium perfringens*, Intraocloaca

104 *Campylobacter jejuni* and *Salmonella* contamination of broiler breeders fed cottonseed versus soybean based feed. N. A. Cox*¹, J. S. Baily¹, J. L. Wilson², N. M. Dale², R. J. Buhr¹, M. T. Musgrove¹, D. E. Cosby¹, and C. L. Hofacre³, ¹*USDA, ARS, Russell Research Center, Athens, Ga.*, ²*Department of Poultry Science, UGA, Athens, Ga.*, ³*Department of Avian Diseases, UGA, Athens, Ga.*

Two studies were conducted to document the effect of dietary cottonseed meal on the incidence of *Campylobacter jejuni* and *Salmonella* colonization of broiler breeders. In the first study we were identifying experimental breeder flocks that were free of *Campylobacter jejuni* (Cj) and *Salmonella* (Sal) in order to inoculate their reproductive tracts with Cj and Sal and follow persistence and fate. Composited fresh cecal droppings (10-15) from each of the four 30 bird pens were analyzed for Cj and Sal. During the pullet rearing period, three of the four pens were fed a corn-based cottonseed meal diet and the fourth a corn soybean meal diet. At 28 weeks of age, the droppings from all four pens were Sal negative. However, the three cottonseed meal pens were negative for Cj, while the soybean meal flock was Cj positive. In the second study, two separate rooms of breeders were used. One received a corn-cottonseed meal diet from week 2-18, while the other a corn-soybean meal diet from week 2-18. From week 18-28 both rooms were fed a corn-soybean meal diet. At week 28, nine composited cecal dropping samples (10 droppings per sample) from each treatment were analyzed for Cj and Sal. Once again there was no difference in the presence of Sal in birds from either room regardless of feed type. However, 3 of the 9 samples from the soybean group were positive for Cj, while none of the 9 was Cj positive from the cottonseed room. Additional studies are needed to determine: a) if these results would be consistent b) if cottonseed fed birds are more resistant to artificial challenges of Cj and c) if this approach is feasible as a means of purging breeder flocks of Cj.

Key Words: Broiler breeder, *Campylobacter jejuni*, *Salmonella*, Cottonseed meal, Soybean meal

105 Evaluation of an experimental chlorate product as a feed supplement to reduce *Salmonella*. M. R. Burnham*, J. A. Byrd, J. L. McReynolds, R. C. Anderson, L. F. Kubena, K. M. Bischoff, T. R. Callaway, T. L. Crippen, K. J. Genovese, and D. J. Nisbet, *USDA-ARS, SPARC, Food and Feed Safety Research Unit, College Station, Texas 77845.*

The objectives of this study were to evaluate the effectiveness of feeding an Experimental Chlorate Product (ECP) for 7 d prior to slaughter on the reduction of *Salmonella* in the crop and ceca of market-age broilers, and its subsequent effects on their performance. At 6 wk of age, one hundred and sixty broilers were randomly assigned to eight groups of twenty birds and placed in floor pens containing pine litter. Prior to placement, each bird was orally challenged with 1×10^9 *Salmonella* Typhimurium (ST). Groups 1 through 6 were fed a normal broiler ration supplemented with 0, 0.5, 1.0, 5.0, 10.0, or 18.0 g/bird/d of a feed grade ECP, respectively. Group 7 was fed a normal diet supplemented with 25.0 g/bird/d of the experimental carrier alone (containing no chlorate) and group 8 was fed a normal broiler ration with 1x ECP (containing 15mM chlorate ion equivalent) added to the drinking water. Performance variables investigated were BW, feed consumption, feed conversion, and mortality. Crop and cecal contents were aseptically collected and spread on Brilliant Green and MacConkey agar plates to enumerate ST and wildtype *Escherichia coli* (EC), respectively. No dietary effects on BW, feed consumption, feed conversion, or mortality were observed. However, litter samples from the pens of birds exposed to ECP had significantly higher moisture content than birds not exposed to ECP. Litter moisture significantly increased as the percentage of ECP in the diet increased. ECP in the diet or in the drinking water significantly reduced the incidence of birds positive for ST without affecting the number of birds positive for EC in both the crop and ceca. However, birds fed higher ECP concentrations had 1-2 log reductions of ST and EC in the crop and ceca. These results indicate that ECP supplementation of feed or water prior to slaughter effectively reduces *Salmonella* and *Escherichia* species in broilers, and may potentially reduce the risk of contaminating poultry products.

Key Words: Broilers, *Escherichia coli*, Experimental Chlorate Product, *Salmonella* Typhimurium, Performance

106 Attempts to isolate naturally occurring *Campylobacter*, *Salmonella*, and *Clostridium perfringens* from the ductus deferens, testes, and ceca of commercial broiler breeder roosters. N. A. Cox*¹, C. L. Hofacre², R. J. Buhr¹, J. L. Wilson³, J. S. Bailey¹, D. E. Cosby¹, M. T. Musgrove¹, K. L. Hiatt¹, and S. M. Russell³, ¹USDA, ARS, Russell Research Center, Athens, Ga, ²Department of Avian Diseases, UGA, Athens, Ga, ³Department of Poultry Science, UGA, Athens, Ga.

Recent studies have shown a significant presence of *Campylobacter* in the semen of mid-life and late-life roosters. The present study was done to determine if several foodborne pathogens (*Campylobacter*, *Salmonella*, *Clostridium perfringens*) could be isolated from the ductus deferens, testes and ceca of 45-65 week old commercial broiler breeder roosters. Five roosters from each of three separate commercial breeder farms were transported to the laboratory. Limited necropsy to remove ductus deferens, testes and ceca without contamination from blood and other tissues was carried out. All samples were analyzed for each of the three previously mentioned bacteria and for total aerobic bacteria, and Enterobacteriaceae. None of the three foodborne pathogens were isolated from the testes of any of the 15 commercial roosters. *Clostridium perfringens* was isolated from one of the 15 ductus deferens, while no *Campylobacter* or *Salmonella* were isolated from this tissue. *Campylobacter* was cultured from the ceca of all 15 roosters, *Clostridium perfringens* from 4/15, and *Salmonella* from 2/15. A quarter of all commercial broiler breeder's semen samples were found to be contaminated with *Campylobacter* in a previous study, however this organism along with *Salmonella* and *Clostridium perfringens* were not found in intra-abdominal tissues. *Escherichia coli* and *Enterococcus faecium* were occasionally cultured from the ductus deferens. The data suggest that the contamination of semen by these foodborne pathogens is a result of sources (cecal/fecal contamination) external to the testes and ductus deferens.

Key Words: Broiler breeder rooster, *Campylobacter*, *Salmonella*, *Clostridium perfringens*, Testes

107 Live Production and Meat Yield Responses of Male Broilers Provided Feeds Subjected to Post-Grinding During a 41-Day Production Period. W.A. Dozier, III*¹, J.M. Long², and R.C. Lacy¹, ¹University of Georgia, ²Georgia Institute of Technology.

This study examined the effects of post-grinding on pelleting parameters, broiler live performance, and processing yields. A total of 1,080 Ross x Ross 308 day-old male broiler chicks was randomly distributed into 24 floor pens (45 chicks/pen; 0.95 ft²/bird). All birds were given a three-phase feeding program, which consisted of starter (0-17 d), grower (18-32 d), and finisher (33-41 d) feeds. Two treatments (12 reps/trt) were employed: 1) control feed that was manufactured under standard procedures and 2) feed subjected to post-grinding prior to the conditioning/pelleting process. Both treatments were included in the starter, grower, and finisher feeds. All feeds were manufactured in a commercial mill. The feed manufacturing parameters on all feeds consisted of a 16-ton batch, conditioning temperature of 82 C, and the pelleting production rate was 50 tons/hr. The corn of the control feeds was ground in a hammer mill screen hole size of 3.2 mm and mixed and pelleted as standard practice. The corn of the post-grind feeds was cracked in a hammer mill screen hole size of 9.5 mm and it was mixed with the macro and micro ingredients. Then, the complete feed was ground in a hammer mill screen hole size of 3.2 mm and pelleted. Grower feed subjected to post-grinding had a higher percentage of pellets ($P < 0.002$) and increased pellet durability index percentage over the control feed ($P < 0.01$). No treatment differences were observed for live performance from 0 to 17 d. Birds consuming feed subjected to post-grinding had improved feed conversion ratio (1.50 vs. 1.54; $P < 0.01$) over the control group from 18 to 32 d. From 33 to 41 d, broilers provided post-grinding feed had reduced feed consumption (1.72 vs. 1.77 kg; $P < 0.01$) and decreased feed conversion (1.64 vs. 1.68; $P < 0.01$). Cumulative feed consumption (3.80 vs. 3.87 kg; $P < 0.01$) and feed conversion (1.51 vs. 1.55; $P < 0.001$) were reduced with the birds fed post-grinding feeds over the control feeds. The absolute and relative amounts of the whole carcass were similar for the two treatments as was the amount of abdominal fat yield. These results indicated that post-grinding improved pellet quality, reduced feed disappearance, and decreased feed conversion ratio with broilers.

108 Evaluating the Efficacy of Dietary Mannan-oligosaccharides to Modify the Toxic Expression of Aflatoxicosis in Broiler Breeder Hens. V.G. Stanley*¹, T. Ogunleye¹, C. Gray¹, M. Daley¹, and A.E. Sefton², ¹Prairie View A&M University, ²Alltech, Guelph, Canada.

An earlier study demonstrated that live yeast culture neutralized the toxic expression of aflatoxicosis in the performance of broiler breeder hens. This study was designed to compare the efficacy of a yeast cell wall preparation (MOS) with the live yeast culture on similar response variables. Broiler breeder males and females of a Cobb strain at 36 wk of age were fed for 21 d a fully fortified broiler breeder diet supplemented with MOS (1 kg/ton) and aflatoxin (AF) at 3 mg/kg of the diet. Egg production, fresh egg weight, percent fertility, early and late embryonic mortality, percent hatchability and chick weight at hatch were examined. After 21 d of exposure to AF, compared to control, egg production and hatchability decreased by 20 and 18%, respectively. Fertility, egg size and chick weight at hatch were not negatively affected by AF. The addition of MOS alone and in the presence of AF significantly ($P < .05$) improved egg production, hatchability and reduced late embryonic mortality to the level of the control. Egg production and hatchability increased by 19 and 15%, respectively, with the inclusion of MOS, despite the presence of AF. Aflatoxin negatively impacted significantly late embryonic mortality. The data presented demonstrated that MOS is comparable to live yeast culture in modifying the toxic expression of AF and improving the performance of broiler breeder hens.

Key Words: Mannan, Yeast, Aflatoxin, Hatchability

109 The effects of hen feeding calls on chick feeding behavior and productivity. M.B. Woodcock*¹, M.A. Latour¹, and E.A. Pajor¹, ¹Purdue University.

A serious poultry welfare and production issue is ensuring that chicks locate food and water quickly after hatch. Studies have shown that hens use vocalizations and visual cues to aid chicks in finding food. Few have investigated the impact of such vocalizations on production traits through market weight of broilers. In this experiment, 1000 broiler chicks were divided equally into sixteen pens. For the first 9 d, 8 pens received 3 min of

feeding calls every hour from speakers near the feeder, the other 8 pens received no sound. Feed was provided *ad libitum*. Feed and chicks were weighed every other day and behavior recorded continuously through 10 d of age. On Day 1 more sound treated chicks than control chicks were found within 9.44 cm of the speaker ($P < 0.05$). During the first 9 d, sound treated chicks had lower feed conversion than the control group ($P < 0.005$), but food consumption did not differ. On Day 9, sound chicks weighed more (139.12 ± 1.52 g) than control chicks (133.17 ± 1.59 g, $P < 0.01$). After the sound was removed on Day 10, differences between treatment groups disappeared by Day 42 (market weight). In summary, playing feeding calls attracted chicks to feeders and improved measures of production and welfare. In addition to attracting chicks to feeders, the calls of adult birds may have a calming effect on chicks and these vocalizations could be used to minimize stress under various production practices.

Key Words: Broilers, Vocalizations, Feeding, Production, Stress

110 Supplementation of Corn-Soybean Diets That Contain Adequate Amino Acids With Versazyme™ Improves Broiler Growth Performance Through to Market Age. N. H. Odetallah¹, J. J. Wang¹, J. C. H. Shih², and J. D. Garlich², ¹Bio Resource International, Inc., Raleigh, NC, ²North Carolina State University, Raleigh, NC.

The objective of this study was to determine the efficacy of Versazyme™ (VZ) upon supplementation to corn-soy broiler diets containing optimum levels of amino acids. Day-old broiler chicks were randomly assigned to 32 floor pens with 25 birds per pen and received one of 4 treatments in a 2 X 2 factorial arrangement. There were 8 replicate pens per treatment. Birds were raised to 42 d of age and fed one of two diets supplemented with or without VZ (0.1% w/w). The diets were formulated to meet either 100% (C) or 110% (HP) of the 1994 NRC recommendation for amino acids. Versazyme™, a fermentation preparation of *Bacillus licheniformis*, contained approximately 600,000 keratinase activity units/g and was added on top of the diets. Results of growth performance showed positive ($P < 0.05$) main effects of diet and VZ supplementation on BW at all periods and FCR at 21 and 42 d of age with no interaction effects ($P > 0.05$). Birds receiving the HP diet had significantly ($P < 0.05$) higher BW at 21, 35 and 42 d than those receiving the C diet (767, 1968, and 2703g vs. 738, 1890, and 2603 for HP vs. C, respectively). Supplementing both diets with VZ gave significantly ($P < 0.05$) higher BW than that of the non supplemented diets at 21, 35 and 42 d of age (821, 2027 and 2716g vs. 738, 1890 and 2603g for the C+VZ vs. C diets; 848, 2055 and 2829g vs. 767, 1968 and 2703g for HP+VZ vs. HP, respectively). Birds fed diets supplemented with VZ had better overall FCR than those fed corresponding diets without VZ supplementation (1.83 vs. 1.86 and 1.76 vs. 1.84 for the C+VZ vs. C and HP+VZ vs. HP, respectively). The results indicated that growth performance and market weight of broiler chickens receiving adequately formulated corn-soybean meal diets may be improved by dietary supplementation with Versazyme™.

Key Words: Broilers, Keratinase, Feed Additives, Versazyme, Growth Performance

111 Grinding and Pelleting Responses of Pearl Millet-Based Diets. W. A. Dozier, III¹, W. Hanna², and K. Behnke³, ¹University of Georgia, ²USDA/ARS, ³Kansas State University.

Previous research has demonstrated that pearl millet is an alternative feed ingredient for broilers. This study examined the effects of grinding and pelleting on pearl millet diets. Four treatments, consisting of either grinding corn through a hammer mill hole screen size of 4.0 mm or pearl millet ground through either a 4.0 mm, a 3.2 mm, or a 2.4 mm hammer mill hole screen size, were employed in the grinding phase. The grain was ground using a Jacobson 30 HP Hammer Mill. Each treatment was replicated four times. In the pelleting phase, a grower broiler diet (20% CP) was manufactured. The treatment structure was a 2 x 3 factorial with a corn-soy control, 2 levels of millet and 3 grind sizes of millet. The main factors of the factorial consisted of two concentrations of millet in the diet at either 25 or 50% and the grinding sizes of the pearl millet were 4.0, 3.2, or 2.4 mm. All grain used during pelleting was derived from the grinding study. Feeds were manufactured with a Model 1000 Master HD Model California Pellet Mill having a 38.1 mm die with 4.0 mm diameter holes. Electrical consumption of grinding was greater for the corn vs. the pearl millet-based diets (8.45 vs. 5.09 Kwh/ton; $P <$

0.001). Reducing the hammer mill hole screen size, increased the electrical usage (4.0 mm = 4.73, 3.2 mm = 4.60, and 2.4 mm = 5.95 Kwh/ton; quadratic $P < 0.001$). Particle size of the pearl millet was decreased linearly as the hammer mill hole screen size was reduced (4.0 mm = 628, 3.2 mm = 611, and 2.4 mm = 492 microns, respectively; $P < 0.001$), but particle size was not affected by grain source. For the pelleting study, pellet durability index, percent fines, production rate, electrical energy, and temperature change were similar for the corn control compared with the pearl millet diets at all inclusion rates. Decreasing the grind size, resulted in a linearly improvement in pellet durability index ($P < 0.04$) and percent fines ($P < 0.03$). These data indicate that pearl millet-based diets have acceptable grinding and pelleting performance compared with a typical corn-soybean meal diet.

Key Words: Grinding, Particle size, Pearl millet, Pellet quality

112 Evaluation of distiller's dried grains with solubles as a feed ingredient for broilers. B. S. Lumpkins, A. B. Batal, and N. M. Dale*, University of Georgia.

Distiller's dried grains with solubles (DDGS), a by-product of the ethanol industry, is becoming available to feed producers in large quantities. The material used in these studies is "new generation" DDGS, derived completely from corn, the solubles fraction being free of by-product streams from other processes, and using drying conditions that are less harsh than in the past. Two experiments were conducted to evaluate the use of DDGS in practical broiler diets. Experiment 1, a 2 x 2 factorial design with diets containing two levels of DDGS (0 and 15%) and two diet densities (high and low). The high density diets were formulated to contain 23% CP and 3200 Kcal ME/kg and the low density diets contained 20% CP and 3000 Kcal ME/kg. Eight pens of six chicks each were fed an experimental diet from 0 to 18 d of age. Chicks were weighed and feed consumption was measured on 7, 14 and 18 d of age. At 18 d of age, body weight and feed efficiency of chicks receiving the high density diet was significantly ($P < 0.05$) better than the chicks fed the low density diet. However, within the two density levels, there was no difference ($P > 0.05$) between chicks receiving diets with 0 or 15 percent DDGS. Experiment 2, a floor pen study ran from 0 to 42 d of age, which was randomized into 6 replications of 50 chicks fed one of four dietary treatments. The dietary treatments were formulated to be isocaloric and isonitrogenous and contained 0, 6, 12 or 18% DDGS. There was no observable difference ($P > 0.05$) in productive performance between treatments except for a slight, but significant, depression in body weight gain at the 18% DDGS. Ten birds from each pen were processed to observe carcass yield. No differences were noted between treatments. These studies indicate that the "new generation" DDGS evaluated is a highly acceptable feed ingredient for broiler chickens.

Key Words: Distiller's dried grains with solubles, Broilers, DDGS, Carcass yield, Feed ingredients

113 Nutrient Composition of Feed-Grade and Pet Food-Grade Poultry By-Product Meal. W. A. Dozier, III*, N. Dale, and R. Dove, University of Georgia.

Poultry-by product meal (PBM) is a popular protein source for poultry feeds. However, in recent years, the pet food industry has placed an increased demand for higher protein PBM. Some pet food companies have specified to renders the need for the ingredient to be manufactured without offal (head, feathers, feet) leading to a higher protein product than conventional feed-grade poultry meal. As a result, the practicing poultry nutritionist has been faced with extreme variation in nutrient content among PBM sources. An experiment was conducted to document the nutrient content of currently available PBM samples. A total of 36 samples was obtained from feed mills in AL, DE, GA, NC, TN, and VA. Twenty-six of the samples were feed-grade sources and 10 of the samples were described as pet-food grade. Sub-samples were analyzed for proximate composition and mineral content. Amino acid digestibility was also estimated using an immobilized digestive enzyme assay (Novus, IDEA® system). Average proximate composition and macro mineral content of the feed-grade sources were: protein = 58.07% \pm 3.23 STD, moisture = 4.18% \pm 1.27 STD, ether extract = 14.40% \pm 3.06 STD, and ash = 17.08% \pm 2.35 STD, calcium = 5.17% \pm 1.10 STD, phosphorus = 2.48% \pm 0.44 STD, whereas, the pet-food grade product had a proximate composition and macro mineral content of: protein = 66.06% \pm 1.89 STD, moisture = 4.09% \pm 1.43 STD, ether extract = 12.63% \pm 1.55 STD, and ash = 15.14% \pm 3.07 STD, calcium = 4.61% \pm 1.39 STD, phosphorus

= 2.59% ± 0.59 STD. The average digestibilities of lysine, methionine, and threonine were 62.9% ± 11.75 STD, 72.2% ± 7.56 STD, and 70.0% ± 7.04 STD, respectively, for the feed-grade source. The average digestibility for lysine, methionine, and threonine of the pet-food grade were 85.2% ± 6.46 STD, 87.8% ± 5.36 STD, 84.7% ± 5.13 STD, respectively. The higher protein and improved amino acid digestibility of the pet-food grade source, was thus confirmed. These data also indicate that nutrient variability is more pronounced with feed-grade than pet-food grade PBM.

Key Words: Amino acid, Feedstuff, Poultry by-product meal

114 Effect of Zinc and Manganese Amino Acid Complexes (Availa®Z/M) on Tom Turkey Performance and Leg Quality. S.W. Davis¹, B.A. George¹, C.L. Quarles¹, T.L. Ward^{*2}, C.J. Rapp², and T.M. Fakler², ¹Colorado Quality Research, Wellington, CO, ²Zinpro Corporation, Eden Prairie, MN.

Five hundred twenty-eight toms (Nicholas, 0 to 19 wk of age) were used to determine the effect of dietary Zn and Mn source and level on performance, carcass quality and leg quality. Toms were allocated randomly to one of four dietary treatments, with 12 replicates per treatment. Diets were fed from 0 to 19 wk of age. The experiment was designed and data

analyzed as a 2 x 2 factorial arrangement of treatments with main effects of level (low and high) and source (inorganic or inorganic combined with complexed Zn and Mn). Levels of Zn and Mn were 65 ppm Zn and 60 ppm Mn in the low level supplementation and 110 ppm Zn and 110 ppm Mn in the high level supplementation treatments. Source effects were determined by using an all sulfate inorganic treatment or a complexed treatment with 40 ppm Zn and 40 ppm Mn replacement by complexed sources (Availa®Zn and Availa®Mn, Zinpro Corp.). There were no interactions (P > 0.10), so only main effect means are presented. Source and level of Zn and Mn did not affect (P > 0.10) breast meat weight, skin tears, skin scratches, breast blisters or mortality. Replacement of 40 ppm Zn and 40 ppm Mn with complexed sources improved (P < 0.05) live weight (16.51 vs. 16.08 kg) chill weight (13.5 vs. 13.2 kg) and adjusted feed conversion (2.33 vs. 2.37 kg feed/kg gain). At 16 wk of age, all birds were walked and leg quality observed. The number of birds with mechanical, infectious, shaky, tibial dyschondroplasia, osteomyelitis or varus/vulgus leg problems were noted and recorded. Replacement of 40 ppm Zn and 40 ppm Mn with complexed sources decreased (P = 0.05) the percentage of toms with leg problems (9.3 vs. 15.2%). These data indicate that addition of complexed Zn and Mn to tom turkey diets improves performance and leg quality.

Key Words: Turkey Production, Zinc, Manganese, Leg Quality

Tuesday, January 21, 2003 Nutrition Room:B313

115 Estimation of relative effectiveness of different methionine sources in broiler chickens using non-linear regression. D. Hoehler^{*1}, S.K. Jensen², and A. Lemme¹, ¹Degussa Corporation, Kennesaw, GA, ²Danish Institute of Agricultural Sciences, Dep. of Animal Nutrition and Physiology, Tjele, Denmark.

There is an ongoing discussion regarding the relative effectiveness of the hydroxy analogue of methionine (liquid MHA-FA) relative to DL-methionine. The aim of the present experiment was to determine the relative effectiveness of liquid MHA-FA, and to test if multi-exponential regression is the proper mathematical model for estimation of bio-effectiveness. A total of 2880 day-old male Ross 208 broilers were equally assigned to 16 dietary treatments for 42 days. Treatments comprised a basal diet and 3 series of diets containing graded levels (0.04/0.08/0.12/0.16/0.20%) of either DL-Met, diluted DL-Met (65%), or liquid MHA-FA. Basal starter and grower diets were formulated to be deficient in Met but adequate in all other nutrients and energy. DL-Met (65%) treatments can be regarded as an internal standard for the validation of the multi-exponential regression analysis. Dilution to 65% Met for the DL-Met (65%) product was chosen because a recent comprehensive literature survey revealed a relative effectiveness of 65% for liquid MHA-FA compared to DL-Met (99%). Birds were housed in 96 floor pens with 30 birds each. Data were analysed by ANOVA and bio-effectiveness was analysed by multi-exponential regression. Broilers performed well and body weight was almost doubled by incremental dose levels up to 0.20%. Simultaneously, feed conversion decreased by 29 points on average. These statistically significant responses clearly showed the Met deficiency of the basal diets. Exponential regression revealed a relative effectiveness of liquid MHA-FA of 64% for weight gain and 67% for feed conversion. Both figures are significantly lower than 88%, which is the content of active substance in liquid MHA-FA. Relative effectiveness estimates of diluted DL-Met (65%) were in the expected range with 67% for weight gain and 59% for feed conversion, confirming that simultaneous regression analysis represents the proper mathematical model for comparative nutritional purposes.

Key Words: Broiler, Methionine Sources, Exponential Regression

116 Isoleucine Needs of Ross Male Broilers. M. T. Kidd^{*1}, S. J. Barber¹, C. D. Zumwalt¹, D. J. Burnham², and B. J. Kerr³, ¹Mississippi State University, Mississippi State, MS, ²Ajinomoto Heartland, Chicago, IL, ³United States Department of Agriculture, Ames, IA.

Isoleucine (Ile) needs of growing and finishing Ross x Ross 308 male broilers were measured by evaluating growth performance and processing attributes in six floor pen experiments. The degree of deficiency of the mash Ile test diet (corn, soybean meal, and blood cell based) and the

ability of it to support growth equal to that of a mash diet based on corn and soybean meal containing surfeit Ile was measured in Experiments 1, 2, and 3 from Days 18 to 30, 30 to 42, and 42 to 56, respectively. In all time periods, the Ile deficient test diets (31% lower than the 1994 NRC Ile recommendations) reduced (P < 0.05) growth and carcass weights of birds and the test diet containing surfeit Ile restored growth and carcass weights (P > 0.05) equal to that of birds receiving the corn and soybean meal diets. Because the Ile response was validated, the Ile need (95% of the asymptote from the quadratic model) was measured for growth and carcass needs from Days 18 to 30, 30 to 42, and 42 to 56 in Experiments 4, 5, and 6, respectively. Most parameters measured yielded significant (P < 0.05) quadratic regression models. Recommended needs for Ile are expressed as total percentage of diet and the digestible Ile coefficient is 90.4%. The Ile needs from Days 18 to 30 for BW gain and feed:gain were 0.68 and 0.71%, respectively. From Days 30 to 42, BW gain and feed:gain Ile needs were 0.64 and 0.66%, respectively. Quadratic carcass responses for this period were not significant. From Days 42 to 56, Ile needs for BW gain, breast meat weight, and breast meat yield were 0.55, 0.60, and 0.63% of diet, respectively. Dietary Ile needs for live performance in Ross male broilers were well below published values (NRC, 1994). Meeting Ile needs for breast meat weight and yield in the 42 to 56 d period required more Ile than did growth. The Ile minimums can be used in feed formulation to reduce protein while taking advantage of commercially available feed grade amino acids and minimizing dietary amino acid excesses.

Key Words: Isoleucine, Amino acid, Broiler

117 Effects of varying dietary ideal amino acid levels in the starter diet on performance of male and female broilers. M. L. Locatelli^{*1}, D. Hoehler¹, A. Lemme¹, P. J. A. Wjitten², and D. J. Langhout², ¹Degussa Corporation, Kennesaw, GA, ²Provimi B. V., Rotterdam, The Netherlands.

Previous studies have shown that the amino acid (AA) response of broilers in the grower and finisher period to a truly optimized essential AA profile on weight gain and feed conversion is significantly higher than that reported in the literature. In the present study, possible effects of graded ideal protein (IP) levels in the starter diet on overall performance from 1 to 37 d of age were investigated. The apparent fecal digestible AA profile, expressed as ratio to lysine in all diets was: Lys (100), Met + Cys (75), Thr (63), Trp (19), Arg (112), Val (81) and Ile (71). Four different IP levels were fed in the starter period (100, 110, 120 and 130%, d 1-14). During the grower (100% IP, d 15-30), and finisher period (100% IP, d 31-37) one diet was provided to all treatment groups. The 100% IP levels for the starter/grower/finisher diets represent apparent faecal digestible lysine levels of 1.05/1.02/0.99%, respectively. All diets were isocaloric and had identical AA ratios. Each treatment was assigned to

12 battery cages, six cages with 20 male and six cages with 20 female Ross 308 birds. Pelleted feed and water were freely available. Increasing the dietary IP level improved weight gain during the starter period and over the entire experimental period linearly, resulting in +30 and +64g, respectively, for the 130% IP treatment. Feed conversion improved linearly (7.9%, 130% IP) during the starter period, and significantly at 130% IP over the entire experimental period by 2.6%. Moreover, breast yield significantly improved (1.9%) at the 130% IP level. The effects found were not affected by sex of the broilers. It can be concluded that the IP requirements of broilers in the starter period are higher than reported in literature for obtaining maximum performance. In addition, the increases found for weight gain at 14 days of age are even more pronounced at slaughtering. However, breast yield and abdominal fat yield are not consistently affected by dietary IP levels in the starter period.

Key Words: Broiler, Amino Acid, Ideal protein

118 Effect of 2-Hydroxy-4(methylthio) butanoic acid (HMB) and DL-methionine in sorghum-soybean meal diets on broiler performance. M Vazquez-Anon^{*1}, E Saleh¹, S Peak¹, T Hampton¹, C Lopez-Coello², J Firman³, and C Knight¹, ¹Novus International, Inc. St. Louis, MO, ²Universidad Nacional Autonoma (UNA), Mexico, ³University of Missouri-Columbia (UMC), MO.

Two floor pen trials in 2 locations (Study 1 at UNA; Study 2 at UMC) evaluated the efficacy of an 88% solution of HMB (Alimet Feed Supplement, Novus International, Inc.) and DL-methionine, dry 99% (DLM; Degussa Hulls). Diets were formulated to be deficient in methionine and cystine (M+C) but adequate in all other nutrients. Study 1 diets included a starter (21d) and grower (21d) with ME (Mcal/kg), CP (%), and total M+C (%) levels of 3.1, 3.2; 22.5, 19.9; 0.72, 0.68, respectively and Study 2 diets included a starter (17d), grower (18d), and finisher (13d) containing 3.1, 3.15, 3.2; 21.2, 19.5, 16.4; 0.68, 0.63, 0.54, respectively. Three supplemental levels of the two M sources were added on an equimolar activity basis for each diet and study. Data from the two studies were analyzed separately as 2 x 3 factorial with a basal treatment. BW data were regressed over additional M intake using linear and nonlinear regression to estimate relative efficacy of the two M sources. In both studies, BW and feed conversion (FC) were improved with M addition at all ages (P .05). In Study 1, HMB birds were heavier at 42 d (1.996 vs. 1.963 kg; P .05) and had better FC (1.95 vs. 1.98; P .05) resulting in a relative efficacy of HMB vs DLM of 115% 40 for linear and 218% 140 for nonlinear regression. In Study 2, HMB fed birds were heavier at 17d (0.64 vs. 0.60; (P .05) with better FC (1.41 vs. 1.50; P .05). Average performance for HMB was better than DLM in later time periods but not significant. The resulting efficacy for HMB relative to DLM at 49 d was of 125% 64 for linear regression. The nonlinear regression failed to converge (SAS 1996). These results confirm broilers fed HMB performed better than DLM, which questions the validity of using DLM as a standard in the slope ratio analysis when evaluating two M sources. $\leq \leq \leq \pm \pm \leq \leq \pm$

Key Words: 2-hydroxy-4-methylthio butanoic acid, methionine, broilers, sorghum

119 Effects of B-Mannanase(Hemicell[®]) on Performance and Body Weight Uniformity in Commercial Turkey Hens with Two Feeding Programs Varying in Nutrient Density. M.E. Jackson^{*1}, R.L. James¹, H. Hsiao¹, and G.F. Mathis², ¹ChemGen Corporation, ²Southern Poultry Research .

Four dietary treatments consisting of two commercial feeding programs with and without a target -mannanase at 100 MU/ton were provided to 10 replicate pens with 19 Nicholas female turkeys per pen. All diets were assayed for proximate composition and -mannanase activity. The feeding programs included a 6-feed moderate nutrient density regime (average C fat = 7.98%, average CP = 21.97%) and a 5-feed high nutrient density regime (average C fat = 8.75%, average CP = 25.02%). Liquid enzyme or water was applied to all pelleted diets. Individual body weight was determined at 0, 6, 12, and 14 weeks of age and feed consumption was determined between these ages. In the moderate density program, -mannanase improved feed conversion from 0-14 weeks by 2.9% or 6.2 points (2.010 vs. 2.163, P<.05), increased 14-week weight by 4.1% (9.600 vs. 9.219 kg, P<.001), and reduced live weight coefficient of variation (CV) at 14 weeks of age (6.73 vs. 9.25%, P<.005). Live performance differences were similar at 6 and 12 weeks of age. In the high density program, -mannanase increased 14-week weight by 1.9 % (P<.05). -mannanase

numerically improved feed conversion from 0-14 weeks by 0.4 % and reduced CV at 14 weeks of age (6.67 vs. 7.99%, NS). CV differences at 12 weeks of age were approaching significance (P<.06). The experiment revealed that largest responses to -mannanase in terms of gain and feed conversion are realized in moderate density feeding programs demonstrating that the enzyme improves nutrient utilization. Its ability to improve flock uniformity appears consistent across nutrient densities.

Key Words: -mannanase , Body Weight, Feed Conversion, Coefficient of Variation, Nutrient Density

120 Effects of B-Mannanase (Hemicell[®]) on broiler performance and flock uniformity fed normal and low energy diets with and without antibiotic. X. Piao^{*1}, C. Wang¹, D. Li¹, L. Gong¹, G. Xu², X. Kang², F.L. Jin³, H. Hsiao³, and M.E. Jackson³, ¹China Agricultural University, Ministry of Agriculture Feed Industry Centre, ²China Agricultural University, Ministry of Agriculture Poultry Research Centre, ³ChemGen Corporation.

Day-old mixed-sex commercial Avian broilers were placed in floor pens and provided with corn-soy based mash diets for 54 days. The birds were randomly assigned to 24 pens with 18 birds per pen and six replications of four dietary treatments. Treatments consisted of: 1. normal energy + chlortetracycline 50 mg/kg; 2. low energy, no additives; 3. low energy + -Mannanase 500 mg/kg; 4. low energy + -Mannanase 500 mg/kg + CTC 50 mg/kg. Three feeding phases were employed from 0-21, 21-42, and 42-54d. (Phase I: ME = 2950 vs 2850 Kcal/kg, Lysine=1.15%; Phase II: ME=3000 vs 2900 Kcal/kg, Lysine=1.08; Phase III: ME=3050 vs 2950 Kcal/kg, Lysine=0.92%). Individual body weights were determined at 0, 3, 6 and 8 weeks of age and pen CVs were determined. Feed consumption was determined between these ages. Gain from 0 to 21 days of age was approximately 10% lower in T2 than in the other treatments (P<.001), birds fed T3 had similar performance compared to those fed T1. Similar trends in weight gain among treatments were observed during the 21 to 54 day period although differences were not as pronounced. From 0-54 days, there were no significant performance differences between Treatments 1, 3, and 4. Body weight % CV was significantly higher in T2 than T3 (11.58 vs 9.17%, P<0.05). The experiment demonstrated that -Mannanase improved live performance similar to a 100 Kcal/Kg increase in ME and also significantly improved flock uniformity, an indicator of flock health status. Addition of CTC resulted in no further improvements in performance. Under the conditions of this experiment, data suggests that -Mannanase may be used as an alternative to CTC in circumstances where antibiotics are not desired.

Key Words: Hemicell, B-Mannanase, Chlortetracycline, Broiler, Body weight uniformity

121 CRINA[®] poultry blend reduces colonization and proliferation of Clostridium perfringens in the intestine and feces of broiler chickens. P. G. Williams¹, P. Mitsch², B. Kohler³, C. Gabler², and R. Losa^{*4}, ¹Akzo Nobel / CRINA SA, Davis, CA, ²University of Veterinary Medicine, Vienna, Austria, ³Office for Consumer Protection and Agriculture, Potsdam, Germany, ⁴CRINA S.A., Gland, Switzerland.

Clostridium perfringens (Cp) is the main causative agent of necrotic enteritis in commercial broiler chickens. A ban on zinc bacitracin and virginiamycin in the European Union, consumer demands for antibiotic-free chicken, and an increasing number of antibiotic-resistant strains of Cp require new methods of control. Plant essential oil (EO; e.g., thyme oil) compounds (e.g., thymol) are known to stimulate intestinal secretion of digestive enzymes and to have *in vitro* antimicrobial activity against many bacteria including Cp. These effects may lead to better regulation and stabilization of the broiler's gut microflora. A field trial involving six sets of paired houses was designed to assess the effect of CRINA[®] Poultry, a specific patented blend of EO compounds and spice extracts, on colonization and proliferation of Cp in the intestine and feces of chickens. Each set of paired houses received either corn-soy based negative control diets or CRINA[®] 100 ppm diets. All feeds contained monensin sodium as a coccidiostat. Intestinal digesta and fecal samples were collected at 14, 21, and 30 days of age. The Cp bacteria were identified via typical hemolysis on blood agar plate, stormy-clot-reaction in litmus milk medium, and ELISA tests. CRINA[®] significantly (P < 0.05) reduced fecal Cp at each age and lowered Cp counts in jejunum and ceca on days 14 and 21, and in the rectum on day 14. In the CRINA[®] group there were fewer Cp contaminated intestinal and fecal samples, fewer samples with

over one million Cp per g, and more samples with counts below 100 per g. No necrotic enteritis was apparent in the CRINA[®] houses, but two of six negative control houses had gross necrotic lesions and increased mortality in week three. The specific blend of EO compounds controlled Cp colonization and proliferation in the gut of broilers, and therefore may be helpful in preventing problems with Cp and necrotic enteritis.

Key Words: Broiler, *Clostridium perfringens*, CRINA[®], Necrotic enteritis, Essential oils

122 Dietary crude protein and metabolizable energy requirement of French guinea keet broilers. S. N. Nahashon*, N. A. Adefope, A. A. Amenyenu, and D. Wright, *Cooperative Agricultural Research Program, Tennessee State University.*

The potential of guinea fowl (*Numida meleagris*) production for food and profit appears to be promising. However, very little is known of their nutrient requirements. In a 3 x 3 factorial arrangement of dietary treatments, three hundred day-old French guinea keets were assessed for their dietary crude protein (CP) and metabolizable energy (ME) requirements from hatch to eight weeks of age (WOA). In three replicates, experimental diets comprising 3050, 3100 and 3150 ME kcal/kg diet each in combination with 21, 23 and 25% CP were fed from hatch to four (0-4) WOA. At 5-8 WOA, dietary ME and CP levels were adjusted to 3100, 3150 and 3200 kcal/kg diet and 19, 21 and 23%, respectively. Feed and water were provided at free choice. At each ME level, a 2% increase in dietary CP was associated with 3-5 and 5-10% increase in feed consumption (FC) at 0-4 and 5-8 WOA, respectively. The body weight gain (BWG) of keets fed 3100 and 3150 ME kcal/kg diet were not different, but were 6% higher ($P < 0.05$) than those of keets fed 3050 ME kcal/kg diet at 0-4 WOA. Keets on 3200 ME kcal/kg diets also exhibited 6% higher BWG and lower feed conversion ratios (FCR) than those on 3100 and 3150 ME kcal/kg diet at 5-8 WOA. The FCR of keets on 3150 and 3050 ME kcal/kg diet and 21, 23 and 25% CP diets were 1.74 and 1.87, and 1.86, 1.74, and 1.80, respectively, at 0-4 WOA. The FCR of keets on 19, 21 and 23% CP diets was 2.64, 2.50 and 2.60, respectively, at 5-8 WOA. French guinea keet broilers will therefore utilize more efficiently diets containing 3100-3150 ME kcal/kg diet and 23% CP at 0-4 WOA, and 3200 ME kcal/kg diet and 21% CP at 5-8 WOA.

Key Words: Guinea keet broilers, Metabolizable energy, Crude protein

123 Evaluation of the nutrient matrix values for phytase in chicks. J. L. Shelton*, L. L. Southern, and L. A. Gaston, *Louisiana State University Agricultural Center.*

Two experiments (EXP) were conducted to evaluate the nutrient matrix values for phytase (Natuphos[®]) in Ross x Ross broilers. The EXP lasted 14 d (EXP 1, 5 to 19 d posthatching, initial and final BW were 72 and 574 g) or 42 d (EXP 2, 0 to 42 d posthatching, initial and final BW were 44 and 2,203 g). There were seven reps (four male and three female) with six chicks each in EXP 1, and 10 reps (five male and five female) with 50 (male) or 55 (female) chicks per rep. On d-43 of EXP 2, six broilers per pen were killed for determination of carcass traits and meat quality. Treatment diets for EXP 1 were 1) C-SBM, 2) C-SBM deficient in AA, 3) as Diet 2 + 600 FTU phytase/kg using the matrix values for Ca, aP, and AA, 4) as Diet 3 without phytase, 5) C-SBM deficient in ME, 6) as Diet 5 with 600 FTU phytase/kg using the matrix values for Ca, aP, and ME, 7) as Diet 6 without phytase. Digestible Lys concentration was 0.82% in the diet deficient in AA, and ME was 2,937 kcal/kg in the diet deficient in ME. Treatment diets for EXP 2 were 1) C-SBM, 2) C-SBM + 600 FTU phytase/kg and using the nutrient matrix values for ME, Ca, and aP, 3) C-SBM + 600 FTU phytase/kg and using the nutrient matrix values for ME, AA, Ca, and aP. For the AA and ME data in EXP 1, ADG and GF were decreased ($P < 0.04$) in chicks fed the diets deficient in AA or ME relative to chicks fed the other diets. In EXP 2, growth performance, mortality, and tibia ash were not affected ($P > 0.05$) in chicks fed diets with phytase relative to those fed the control diet. Chicks fed the diets with phytase using the matrix values for ME, Ca, and aP had decreased ($P < 0.03$) breast weight as a percentage of chill weight compared with those fed the diet with phytase using the matrix values for ME, AA, Ca, and aP. Total P, total soluble P, and inorganic soluble P were decreased ($P < 0.03$) in litter of chicks fed diets with phytase relative to those fed the control diet. These data indicate that the nutrient matrix values for Natuphos[®] 1200 can be used in formulation programs with no loss in growth performance or carcass yield.

Key Words: Chick, Nutrient, Phytase

124 Comparative Efficacy of two Microbial Phytases for improving performance in Broilers Fed Low-P Diets. J. S. Sands*¹, R. Stilborn², J. Berg², and R. E. Salmon², ¹Danisco Animal Nutrition, ²Lakeside Research Center.

The comparative efficacy of two microbial phytases that initiate hydrolysis at position 6 (6-phytase) and position 3 (3-phytase) of the inositol-6-phosphate molecule was compared in a 42-d broiler study. A total of 2016 d-old male broilers were used to assess growth and bone ash response. The study was arranged as a RCBD, with 6 blocks consisting of 8 pens each, 42 chicks per pen and 8 replicate pens per diet. Eight corn/soybean meal-based diets were fed in the starter (0-21 d) and grower (22-42 d) phases and consisted of a positive control (PC), basal (1.2 g/kg reduction in non-phytate P compared to the PC in the 0-21 d and 22-42 d phase), basal plus 6-phytase at 250, 500, 750, and 1000 FTU/kg, and basal plus 3-phytase at 500 or 750 FTU/kg. Feed intake (FI) and body weight gain (BWG) were measured and feed conversion efficiency (FCE) calculated from 0-21 and 22-42 d. Tibia samples were collected on d-42. Mortality was not affected by dietary treatment. The basal diet significantly reduced performance in comparison to the PC diet ($P < 0.05$). The addition of graded levels of 6-phytase up to 1000 FTU/kg resulted in significant increases in BWG ($P < 0.001$) and FCE ($P < 0.003$) from 0-21 and 22-42 d. The 6-phytase at >250 FTU/kg resulted in significantly greater ($P < 0.01$) FI, BWG and FCE than 500 FTU/kg of the 3-phytase from 0-21 d. Overall, there was a tendency ($P < 0.10$) towards better BWG and FI and significantly improved FCE ($P < 0.01$) with 250 FTU/kg of the 6-phytase compared to 500 FTU/kg 3-phytase. Bone ash was significantly increased with the addition of the 6-phytase compared to the low-P basal diet and was similar between phytases. In conclusion, broilers fed diets supplemented with 250 FTU/kg of 6-phytase achieved similar BWG and FCE as those fed a PC diet whereas, 750 FTU/kg of the 3-phytase was required to achieve a similar response.

Key Words: Microbial Phytase, Efficacy, Growth, Bone ash, Broilers

125 Dose response of a novel phytase. M.R. Bedford*¹, E. Koepf², P.F.S. Street¹, and J.S. Tuan¹, ¹Zymetrics, ²Syngenta Biotechnology Inc.

The principal use of phytase in animal nutrition has been the replacement of inorganic phosphate. The benefits include reduced phosphorus (P) excretion (i.e. pollution) coupled with reduced feed cost due to favourable pricing of the enzyme against inorganic phosphate. Recent publications have suggested that the benefits of phytase may extend beyond P replacement. Our research with a novel, thermo-tolerant phytase has provided evidence to suggest that additional nutrient claims with this particular enzyme may be justified. 6 replicate pens of 42 male broiler chickens were fed either a positive control (0.4% available phosphorus, AvP in both starter # 0-21d, and finisher 22-42d) or negative control (0.21% AvP in starter, 0.20% AvP in finisher) wheat, soy and canola based ration supplemented with 0, 250, 500, 750, 1000, 2000 or 4000 U (AOAC, 2001) of a novel phytase per kg of feed. All diets were pelleted at a min 80C conditioning temperature and were formulated to meet NRC requirements for all nutrients with the exception of AvP. As expected, birds fed the negative control returned poorer intakes, gains and FCR at 42d compared with the positive control. Whereas no level of supplementation of phytase restored intake to the level of the positive control, gain and FCR achieved or exceeded such a level of performance between 250 and 500 U/kg. Moreover, incremental levels of phytase continued to deliver benefits in FCR such that inclusion levels of 750 U/kg and greater resulted in better performance than that of the positive control ($p < 0.05$). In terms of intake, gain and FCR, the relationship between dose of enzyme and response was best described by a log dose relationship, as predicted by Rosen (2001). These data suggest that the main benefits of this phytase are two-fold, 1) recovery of animal performance in low AvP rations to that of normal AvP diets at dosages similar to current recommendations, and 2) delivery of additional performance benefits beyond those expected from increased AvP alone when used at higher rates of supplementation. The dose response observed may provide additional flexibility for producers to further optimise their economic return through differential dosing of this novel phytase.

Key Words: Phytase, Thermolerant

126 Improved Performance of Brown Shell Laying Hens Fed Low-Phosphorus Diets with Supplements of 25-Hydroxy-Vitamin D and Phytase. A. H. Cantor*, T. Ao, A. J. Pescatore, and M. J. Ford, *University of Kentucky, Lexington, KY, USA.*

The influence of adding 25-hydroxy-cholecalciferol (25-OH-D₃) and phytase to a low-P layer diet on production performance and eggshell quality was studied using 432 brown shell laying hens (Hy-Line). Twelve replicate groups of six individually caged hens, 20 wk of age, were assigned to each of six treatments. Treatments consisted of feeding the following diets: 1) low-P, corn-soy basal layer diet (0.12% available P); 2) basal + 25-OH-D₃ (69 g/kg diet, HY-D, Roche Vitamins Inc., Parsippany, NJ); 3) basal + phytase (Ronozyme P, 234 units/kg diet, Roche Vitamins Inc.); 4) basal + 25-OH-D₃ + phytase; 5) normal-P layer diet (0.30% available P); and 6) normal-P diet + 25-OH-D₃. All diets were supplemented with cholecalciferol (2120 IU/kg diet) and contained 3.75% Ca. During 48 weeks of feeding, the addition of 25-OH-D₃ and phytase to the low-P basal or feeding a higher level of P significantly ($P < 0.05$) increased egg production (80% for Treatment 1 vs. 85-87% for Treatments 2-6) and BW gain. Use of phytase, alone or in combination with 25-OH-D₃, and the normal-P diets significantly increased feed intake. Average egg weight was greater for hens in Treatments 4-6 (66 g) than for those in Treatments 1 and 2 (64 g). Manure P concentration for Treatments 1, 3, 4, and 6 (1.01-1.27%, DM basis) was significantly lower than that for Treatment 5 (1.85%). Key words: phytase, 25-OH-cholecalciferol, phosphorus. The results indicate that both 25-OH-D₃ and phytase can be useful in correcting production performance of hens fed a low-P diet.

Key Words: phytase, 25-OH-cholecalciferol, egg production

127 The effect of phytase and phytic acid on endogenous losses from broiler chickens. A. J. Cowieson*¹, T. Acamovic¹, and M. R. Bedford², ¹*Scottish Agricultural College, Ayr, Scotland, UK*, ²*Zymetrics Inc., Marlborough, England, UK.*

The effect of phytase and myo-inositol hexakis phosphate (IP6) ingestion on the excretion of amino acids, minerals, phytate phosphorus and sialic acid from broiler chickens, was investigated.

Approximately 70% of phosphorus in plant material is in the form of phytate phosphorus. However, phytate phosphorus is largely unavailable to poultry as they do not possess effective endogenous enzymes for the hydrolysis of phytate. The poor utilisation of plant P by poultry leads to ecological problems associated with the disposal of phosphorus-rich manures. In addition, phytate reduces the availability of amino acids, carbohydrates and many mineral ions. The addition of exogenous phytases to poultry diets can improve the digestibility of minerals and amino acids. An important aspect of the beneficial effects of exogenous phytase addition to poultry diets may be a reduction in the inimical effects of phytate on endogenous losses.

A total of thirty-two Ross broilers with a mean body weight of approximately 2.5kg were used in a 2x2 factorial precision feeding study (n=8). The birds were deprived of feed for 48 hours prior to gavage of the treatment material. The four treatments used were glucose, glucose with 1000 FTU, glucose with 1g of IP6, and glucose with 1g IP6 and 1000 FTU.

After intubation of the treatment material the excreta from each bird was quantitatively collected over a 48-hour period. The total excretion of amino acids, minerals, phytate phosphorus and sialic acid (as an indicator of mucin production) was subsequently determined.

The addition of 1g of IP6 to the glucose solution resulted in an increase in the excretion of sialic acid, many dispensable and indispensable amino acids and also iron, sodium and sulphur compared to birds fed glucose ($P < 0.05$). Phytase treatment of the IP6 solution reduced the excretion of some amino acids, minerals, sialic acid and phytate-P compared to birds fed IP6 with no enzyme treatment ($P < 0.05$).

It can be concluded that IP6 increases the loss of endogenous material from broiler chickens and that some of the beneficial effects of phytase may be explained by a reduction in these endogenous losses.

Key Words: Phytase, Phytate, Chickens

128 Effects of broiler breeder age and egg storage on egg quality, hatchability, chick quality and relative growth to slaughter weight at 42 days. K. Tona, O.M. Onagbesan*, V. Bruggeman, B. De Ketelaere, and E. Decuyper, *Catholic University, Leuven, Belgium.*

Effects of broiler breeder age and egg storage on egg quality, hatchability, chick quality and relative growth to slaughter weight at 42 days.

Tona K, Onagbesan OM, Bruggeman V, De Katelaere B, Decuyper E. Dept. of Animal Science, Catholic University, Leuven, Belgium.

The age of breeders and egg storage are fundamental factors that affect poultry production parameters. Apart from increasing incubation duration, the extent of the effects of the interaction between breeder age and egg storage on hatchability, chick quality and their subsequent growth to slaughter are still not fully understood. This study examined the effects of prior storage of incubating eggs (fresh vs 7d storage), age of broiler breeder (33 wk vs 43wk) and their interactions on egg albumen Haugh Units (HU), hatchability, day-old chick weight, chick quality and chick relative growth (RG) to 42d. Four replicates of 150 eggs/group were incubated at 37.6C/29C (dry bulb/wet bulb temp) in a forced-draft incubator until hatch. Albumen HU was determined in samples before incubation. After hatching, hatchability, chick weight and chick quality were determined and 20 high score chicks/group/replicate were reared on floor pens for 42d. Body weights were recorded at 7, 14, 21, 28, 35 and 42d. Results: Storage depressed HU and storage interacted with increased breeder age to further depress HU. Storage depressed hatchability and chick quality only in older breeders. Day-old weight was higher in older breeders with no effect of storage. Storage depressed RG only in young breeders, but overall, RG was lower in older breeders. Day-old body weights had no correlation with slaughter weights at 42d but there was correlation of body weight from 7d onwards. Correlation coefficient increased with age. The effect of storage on body weight gains of broilers was apparent only after 14d of age whereas the effect of age manifested in the first 14d of age. It is concluded that both storage and age may have interacted to alter different egg quality components that manifest in the performance of the chicks at different periods post-hatch. Key words: broiler breeders, age, storage, chick quality, relative growth

Key Words: broiler breeders, age, storage, chick quality, relative growth

Tuesday, January 21, 2003
SCAD/Avian Disease
Room:B312
Moderator(s): Joe Giambrone & John Smith

129 Development of an antibody complex vaccine against avian reovirus. ZY. Guo* and J.J. Giambrone, *Auburn University, Auburn, AL.*

Vaccination is an important means of controlling diseases caused by avian reoviruses in commercial broilers. *In ovo* (in the egg) administration of MD and IBV vaccines is commonplace in commercial broilers due to increased speed and lower labor costs. Reovirus vaccines are not often given by *in ovo*, since they may cause embryo pathology. In this study, a commercial reovirus vaccine complexed with various amounts of specific antibody was inoculated into 18-day-old specific-pathogen-free (SPF) broiler embryos at 0.1 of the recommended dose by the manufacture. At 3,6,9,12 and 15 days post *in ovo* vaccination (PIOV), serum was collected and analyzed for antibody against reovirus by ELISA. At

the same time, spleens were collected and examined for vaccine virus by inoculation of chicken embryo fibroblasts. At 16 days of age, chickens were challenged in the foot-pad with a standard S1133 virulent reovirus isolate. At 22 days of age, birds were euthanized, examined for gross lesions, and weighed. Efficacy of the vaccination program was based on safety (absence of morbidity and mortality), and efficacy (antibody responses and percent protection based on gross lesions in the foot-pad). Chickens in the group given only vaccine virus were the first to show an antibody response and vaccine virus was isolated from their spleens on the day they hatched. Chickens vaccinated with the vaccine virus-antibody complex had a delayed antibody reaction and vaccine virus was not isolated from them until 6 days PIOV. Chickens in groups 2 and 3, which received vaccine virus-complexed with a 1/4 or 1/8 dilution of antibody, respectively, had the best percent protection. Results showed

the safety and efficacy of an *in ovo* administered, experimental vaccine virus-antibody complex, similar to what is marketed for IBDV.

Key Words: In ovo, Avian reovirus vaccine, Complex vaccine

130 Evaluation of Marek's disease quantitative trait loci in a commercial broiler population. AA Gabr^{*1}, H Kim¹, and MG Emar¹, ¹*University of Delaware Department of Animal and Food Science.*

Marek's disease (MD) continues to be a growing nuisance to the poultry industry, costing millions of dollars due to loss of egg production, carcass condemnations, and increased mortality. Intensive rearing and forced selection pressure on the virus over the past two decades has caused new strains to emerge. Currently, control of MD is solely dependent on vaccines and better bio-security. These methods have proven to be efficient in the short term. Genetics possesses the key to future disease control leading to a path of improved production and better cost efficiency. With current technologies, DNA marker-assisted selection has become a feasible compliment to vaccination for disease control. This study represents the first report on characterizing microsatellites associated with MD in broiler chickens. Two broiler pure lines varying in their susceptibility to MD were line-crossed to generate an F1 population. The F1 parents consisted of 8 sires and 32 dams, generating 742 F2 progeny. Chicks were placed in 4 hatches, challenged with 3000 PFU of RB1B MDV intraabdominally at 5 days of age, were observed over a period of 8 weeks, and documented for MD-related clinical signs. Blood sampling, DNA extractions, and necropsies were performed on ALL birds. Twelve microsatellites (located on chromosomes 1,2,4,7,8, & E16) that were previously associated with MD quantitative trait loci (QTL) in experimental and layer chickens were amplified by PCR and later genotyped in the broiler F1 parents and their F2 progeny. Samples were evaluated using an automated gel sequencer and genotyping programs (Genescan and Genotyper). Several broiler families were informative for microsatellite loci, associations of microsatellite genotype and MD phenotype were identified using chi-square analysis. We conclude that a limited number of regions across the chicken genome are pivotal for MD resistance. Identification of markers for genotyping in valuable breeding populations will become a valuable tool providing both priceless and essential data needed by primary breeders to aid in the selection against MD susceptibility for the future.

Key Words: QTL, Marek's, Broiler, Genotyping, Microsatellites

131 Comparison of the clinical effects of a Marek's disease challenge between two broiler MHC haplotypes originating from a commercial population. AA Gabr^{*1}, H Kim¹, SJ Ewald², and MG Emar¹, ¹*University of Delaware Department of Animal and Food Science,* ²*Auburn University Department of Poultry Science.*

Marek's disease (MD) is caused by a herpesvirus that is both highly contagious and environmentally ubiquitous worldwide. High mortality, neurological signs, and visceral tumors accompany birds affected with MD, which consequently leads to severe, egg loss in layers and carcass condemnations in broilers. With the emergence of highly virulent strains, vaccination may no longer be sufficient to control MDV. An alternative approach for breeders is to genetically select for MD resistance. The MHC haplotype is an important genetic component that influences MD resistance. Hence, the current study was conducted to assess two broiler MHC haplotypes for resistance or susceptibility to MD. Five sire families from a commercial source were established and they were segregating for two MHC haplotypes, BA4 and BA5 that are similar to standard B21 and B13, respectively. The progeny (n=186) consisted of the two homozygotes (BA4 BA4 and BA5 BA5) and heterozygotes (BA4BA5). All broiler chicks were randomly placed in colony houses at one day of age, and each chick was challenged intraabdominally with 3000 plaque-forming-units of the MDV strain RB1B at five days of age. Chicks were observed for clinical signs, mortality, and body weight until 8 weeks post-challenge. Blood samples for MHC genotyping were collected at 3 weeks post-challenge. All birds were necropsied and gross lesions were assessed. The results indicated that both sire family and MHC type had significant effects on MD-associated mortality, tumors, paralysis and total MD. Homozygous BA4 BA4 individuals were more resistant to MD tumor development than homozygous BA5 BA5 individuals. An interesting finding was that two of the families challenged displayed severe stunting. This observation confirms an earlier conclusion that there is a

genetic explanation for birds that stop growing between 3 and 4 weeks post-MD challenge, never recovering from this wasting syndrome. In addition, the data paves the way for future research in gene expression analyses during an MD insult to identify candidate genes associated with the clinical effects of MD.

Key Words: MHC, Broiler, Marek's, Stunting

132 Evaluation of apoptosis in infected tissues and MSB-1 infected cell cultures for the presence of chicken infectious anemia virus (CIAV) using fluorescence. Sumanth Putta^{*1}, David Cross¹, Fred Clark¹, Billy Hargis¹, and Lisa Newberry¹, ¹*Center of Excellence for Poultry Science, University of Arkansas.*

One day-old specific pathogen free (SPF) chicks were inoculated intramuscularly with a 1:4 dilution of a primary chicken infectious anemia virus (CIAV) isolate. At 14 and 28 days post inoculation, ten birds from each of the two groups were weighed, bled and sacrificed. Samples of liver, thymus, spleen, bone marrow and bursa were taken for both histopathology and virus isolation. At necropsy no clinical signs of CIAV infection were observed in any of the experimental groups. Histopathological examination of tissues revealed atrophy of the thymus with thin, disorganized cortices and hemorrhagic medullas. There was mild reticuloendothelial hyperplasia in the spleens, but no specific changes occurred in the livers and bursas. At 28 days post inoculation, lesions were diminished in the thymus, but no changes were seen in bursas, spleens and livers. Hematocrit values were decreased but not significantly different from the negative controls at both 14 and 28 days post inoculation. Additionally, MSB-1 cells at 5×10^5 cells/mL were infected with various strains of CIAV and viral presence was determined at 24 and 48 hours post infection utilizing Mab R-25. Cellular pathology associated with apoptosis was determined using TUNEL (terminal deoxy nucleotidyl transferase-mediated dUTP nick end-labeling) and VYBRANTTM assays. Virus was present in the tissues harvested from SPF challenged chicks, but apoptosis associated pathology could not be confirmed. However, apoptosis and virus presence were evident in MSB-1 infected cell cultures.

Key Words: Chicken infectious anemia virus, TUNEL, Apoptosis, Pathology

133 The use of the single-strand conformational polymorphism analysis (SSCP) for genotyping isolates of the infectious bursal disease virus (IBDV). A. Banda^{*1}, P. Villegas², J. El-Attrache², and N. Ledesma¹, ¹*Universidad Nacional Autonoma de Mexico, Mexico City, Mexico,* ²*The University of Georgia, Athens, GA. U.S.A..*

Single-strand DNA molecules can fold into complex three-dimensional structures as a result of intrastrand base pairing. Changes on the nucleotide sequence of the DNA molecule can reshape the secondary structure with consequent changes in electrophoretic mobilities through acrilamide gels. Single-strand, conformational polymorphism (SSCP) analysis is a technique that detects mutations based on this principle. In this study, 390-bp RT-PCR amplicons encompassing the hypervariable region of the VP2 gene of different infectious bursal disease virus (IBDV) strains were obtained and further analyzed by SSCP. It was possible to detect variations in the migration patterns between Delaware variant, standard, and very virulent strains of IBDV using SSCP. According these results, SSCP, resulted an effective method to genotype IBDV strains.

Key Words: Infectious bursal disease virus, Genotyping, Single-strand conformational polymorphism analysis, Field strains, Diagnosis

134 Expression of Infectious Bursal Virus VP2 gene in a Transgenic Plant. Hongzhan Wu^{*1}, Narendra Singh², Locy Robert², Karyn Gunn³, and Joseph Giambrone¹, ¹*Department of Poultry Science, Auburn University,* ²*Department of Biological Sciences, Auburn University,* ³*Biomedical Research Training Program, Alabama State University.*

Edible plants for the production and continual delivery of immunogenic proteins to poultry flocks could provide an alternative to field boosting by drinking water or course spray. We report the development of transgenic arabidopsis expressing the major host-protective immunogen of infectious bursal disease virus (IBDV). The VP2 gene of a variant E IBDV was isolated and amplified by a reverse transcriptase-polymerase chain reaction (RT-PCR), and introduced into a vector pE1857 with specific

super-promoter, designated as rpE1857. Arabidopsis, a common weed used in the laboratory studies, received the VP2 gene by Agro-bacterium tumefaciens mediated transformation. The presence of the transgene in plants was confirmed by PCR and southern blotting. Specific transcription and expression were demonstrated by RT-PCR, western blotting, and an antigen capture ELISA. Immunogenic feeding trials in chickens with the transgenic plant are currently under way. It is hoped that this work will lead to the development of an edible vaccine in transgenic alfalfa to prevent IBD in chickens.

Key Words: Infectious Bursal Disease Virus (IBDV), VP2 gene, transgenic arabidopsis, edible plant vaccines

135 Monitoring of breeder flock immunity against infectious bursal disease virus using progeny challenge. J. J. Giambrone^{*1}, T. V. Dormitorio¹, and K. Takeshita², ¹Auburn University, Auburn, AL., USA, ²Lohmann Animal Health, Gainesville, Ga., USA.

Two studies were done to monitor the success of vaccination of commercial broiler breeders in providing passive protection to the progeny against infectious bursal disease viruses. The challenge viruses were APHIS, Var E, and Var Ark. All belonged to different molecular groups. The newly isolated Var Ark virus was placed in molecular group 6 and can cause a high incidence of atrophy of the bursae of Fabricius and proventriculitis in broilers. Proventriculitis has seen increased occurrence in broilers in the SE USA, resulting in decreased weight gain and processing plant contamination. The studies consisted of 2 broiler progeny challenge experiments with identical materials and methods. Day-old chicks were taken from the commercial hatchery and placed in isolation units at Auburn. The chickens were derived from various commercial broiler breeders flocks in the SE USA, that had been vaccinated several times as pullets with both live and killed vaccines containing both standard and variant IBDVs. The chicks from each breeder flock were placed into 4 separate groups. At two weeks of age, one group of 20 chicks from each of six breeder flocks was challenged with each of the 3 viruses and one group of 10 birds served as a non-challenged control. At 7 days post challenge, the birds were killed, a post-mortem exam done, and bursa weight to body weight ratios determined. Results from the 2 studies were similar. Using atrophy of bursae as an indication of protection, results showed that chicks were adequately protected against all viruses. Using gross lesions in the proventriculus as an indication of protection, results showed that chicks were well protected against all viruses. Percent protection was best against the APHIS virus, followed by Var E, and then Var Ark. Current IBDV vaccines are effective in immunizing breeders and their progeny against these 3 IBDVs. However, vaccine manufacturers should continue to perform efficacy testing with their vaccines against new antigenic variant IBDVs, especially group 6 viruses, which are becoming increasing more prevalent in the field.

Key Words: IBDV, Vaccination, Proventriculitis

136 Effect of *in ovo* infectious bursal disease vaccination on early IBD protection in commercial broilers. K.C. Cookson^{*1}, J.J. Giambrone², and J.H. Rodenberg¹, ¹Fort Dodge Animal Health, Overland Park, Kansas, ²Department of Poultry Science, Auburn University, Alabama.

Infectious bursal disease virus (IBDV) infections by 2 weeks of age can cause profound and long-lasting immune suppression in chickens. Although passive immunity is the most efficient way to prevent early field infections, some flocks still become challenged by two weeks of age. The objective of this study was to see if adding IBD vaccine to Marek's given *in ovo* would be both safe and increase early IBDV protection in a commercial broiler flock. Broilers came from a 39-week old breeder flock that received two bursa-derived shots prior to lay. Day-of-age broiler GMT was 3,448 on Idexx XR. Broiler flock was divided into 3 groups, according to the following *in ovo* vaccination programs: Marek's serotype2/3, and Marek's plus IBD vaccine #1 or #2. IBD vaccine #1 is intermediate-type and vaccine #2 is mild-type. Challenge groups received 3.0 logs of IBD virus 1174, a Jackwood Group-6 isolate, at either 8 or 11 days of age. Bursal protection was evaluated at 15 days for both challenge groups using bursa to body weight ratios (B:BW) and a cut-off set two standard deviations below the mean of the Marek's-only controls. Hatchability (89% in Marek's-only vs. 88% and 92%) and livability (94% vs. 92%) were not affected by IBD vaccination. In the non-challenge groups there were no significant B:BW differences between Marek's-alone and

Marek's/IBD vaccinates at 8 and 21 days, but both IBD vaccine groups were lower at 15 days, suggesting both a vaccine "take" and subsequent bursal regeneration. Comparing the challenge groups at 15 days, IBD vaccines #1 and #2 both significantly enhanced protection from an 8-day IBDV challenge compared to the Marek's-alone group (64% and 68% vs. 12% protection 7dpc.). However, only the Marek's plus IBD vaccine #1 combo significantly enhanced protection from 11-day IBDV challenge (100% vs. 30% and 40% protection 4dpc., respectively). In this study both IBD vaccines were safe and significantly enhanced early IBDV immunity when given *in ovo* with Marek's vaccine.

Key Words: IBDV, Vaccination, In ovo, Broiler, Marek's

137 What is your diagnosis for lost hatch? P Stayer^{*1}, ¹Sanderson Farms, Inc..

This is a case presentation of a sudden loss of hatchability. The pathognomonic signs were sudden drop in hatchability, late dead embryos and embryos stuck to egg shells. Diagnostic techniques and results are presented in a logical progression to lead the audience to a conclusive diagnosis for this situation of lost hatch. Experienced hatchery personnel will quickly uncover the cause for the catastrophic hatch. Poultry workers of all stations should become familiar with this situation and its' characteristic markers.

Key Words: Hatchery, Residue, Breakout, Incubation

138 The myths and realities of biosecurity and sanitation. 1. Teaching principles in the classroom. J. Brake^{*}, B. A. Lenfestey, and S. Creech, North Carolina State University, Raleigh, NC USA.

Recent animal disease outbreaks have received much press coverage and have negatively impacted both tourism and trade. The result of this has been an increased interest in biosecurity and sanitation of farms, feed mills, hatcheries, processing plants, and the people and vehicles that move among these facilities. During the recent problems with avian influenza in Virginia, it was noted that the poultry company perceived to have the "poorest biosecurity" actually had the least problems with this disease. This suggests that the commonly accepted practices of "good biosecurity" are at best ineffective, or, possibly, worse than doing nothing at all. Development of effective biosecurity programs depends upon a clear understanding of the basic principles of sanitation. Teaching sanitation principles in a classroom setting requires sampling of common surfaces and objects such as fingers, hats, walls, table tops, shoes, etc. to familiarize students with normal environmental contamination. Demonstrating the effects of cleaning and disinfecting requires that samples be taken during each step of the process so that a visualization of the bacterial population dynamics can be made. Sampling can be done with Rodac plates or sterile drag swabs streaked onto nutrient agar plates. It is important that the agar contain letheen to neutralize any residual disinfectant that is on the surface sampled. One type of visual demonstration is to swab samples from a tabletop. A sample should be taken from the dry table initially, then when the cleaner is sprayed on the table (5, 10, and 20 minutes post), and again after the disinfectant is sprayed (5, 10, and 20 minutes post). The final sample should be taken when the table is completely dry. This can be done on many surfaces including the bottom of a shoe, which can represent the situation where a boot pan is used. These demonstrations will provide a visual display showing that the application of a disinfectant onto a wet, soiled surface will increase the bacterial population and that cleaning is the major means to reduce bacterial populations.

Key Words: Biosecurity, Sanitation, Disinfectants, Cleaners

139 The myths and realities of biosecurity and sanitation. 2. Teaching principles in a field scenario. J. Brake^{*}, B. A. Lenfestey, and S. Creech, North Carolina State University, Raleigh, NC USA.

Development of effective biosecurity programs depend upon a clear understanding of the basic principles of sanitation. Teaching sanitation principles in a commercial field scenario requires bacterial sampling of common surfaces and objects to familiarize clients with normal environmental contamination. Clients have to see the bacterial population increase and decrease to understand what is really needed to develop a good biosecurity program. Good examples can be made by sampling

boots, hats, tires, fingers, floors of vehicle passenger compartments, feed truck tanks, boot pans, etc. Demonstrating the effects of cleaning and disinfecting requires that samples be taken during each step of the process so that a visualization of the bacterial population dynamics can be made. Sampling can be done with Rodac plates or sterile drag swabs streaked onto nutrient agar plates. It is actually preferable to do both, as this gives a more complete and clear picture, especially with tires where the swabs can be used in the tire grooves and the Rodac plates can be used on the tire surfaces. It is important that the agar contain letheen to neutralize any residual disinfectant that is on the surface sampled and allow the existing population of bacteria present at the time of sampling to be visualized and not be killed after plating. A good demonstration is to take samples from a dry tire, a tire immediately, 5, 10, and 20 minutes after being sprayed with disinfectant, a tire immediately, 5, 10,

and 20 minutes after each step of being sprayed with cleaner, water, and disinfectant. These demonstrations will routinely show that application of disinfectant at label use dilution will increase bacterial contamination unless the treated surface has been thoroughly cleaned beforehand. These demonstrations will show that boot pans may be one of the major causes of biosecurity problems by promoting bacterial multiplication as compared to leaving surfaces dry. These demonstrations will show that sanitation remains 99% cleaning and 1% disinfection and that there are no short cuts to time-consuming cleaning and drying before disinfection. If time and resources are not adequate to do a good job of cleaning and disinfecting it may be better to do nothing at all.

Key Words: Biosecurity, Sanitation, Disinfectant, Cleaners

Monday, January 20, 2003

Posters

Room:B308-309

140 Evaluating Biomarkers in Hen Plasma for Medullary Bone Metabolism During an Egg Cycle. W. K. Kim*, B. C. Ford, and R. M. Leach Jr., *The Pennsylvania State University, University Park, PA 16802, U.S.A.*

Medullary bone in laying hens serves as a unique calcium source for eggshell formation and contributes 30 to 40 % of eggshell calcium. When medullary bone is resorbed by osteoclasts during eggshell formation, bone matrix metabolites as well as bone minerals are released into the blood. Thus, these bone matrix metabolites could serve as markers for medullary bone turnover. Potential markers are hydroxyproline (HYPRO) and keratan sulfate (KS). HYPRO is a component of collagen that is an important bone matrix protein. However, it is not specific for medullary bone because collagen is found in cortical bone as well as other tissues. KS is the major glycosaminoglycan in medullary bone. We hypothesized that patterns of these biomarkers in blood would be synchronized with the daily egg cycle. Thus, an experiment was conducted to evaluate these potential biomarkers in hen plasma. Blood was collected by cardiac puncture at 0, 6, and 15 hr post-oviposition from twenty-two Single Comb White Leghorn hens. The plasma was analyzed for calcium, HYPRO, total glycosaminoglycan (GAG), and KS. GAG was measured using the dimethylmethylene blue (DMB) assay, and KS was quantified by calculating the difference in DMB values before and after keratanase digestion. There were no significant differences in plasma Ca and HYPRO levels among the plasma samples. GAG levels at 6 and 15 hr post-oviposition (1304 and 1582 ug/ml, respectively) were significantly increased compared to that at 0 hr post-oviposition (617 ug/ml). KS levels increased as post-ovulation time increased (6.9, 154.5, and 238.5 ug/ml at 0, 6, 15 hr post-oviposition, respectively) and were significantly higher at 15 hr post-oviposition than at 0 hr post-oviposition. These results indicate that KS is a potential marker to monitor medullary bone metabolism.

Key Words: Medullary Bone, Keratan Sulfate, Hydroxyproline, Glycosaminoglycan, Eggshell Calcium

141 Changes in blood pH observed in two strains of laying hens after exposure to two different ambient temperatures. Danilo Franco*¹, Lyle Robeson¹, and Mary Beck¹, ¹*University of Nebraska-Lincoln, Animal Science.*

Ten birds of commercial laying hens (Hyline W-36 and W-98) of approximately 40 weeks of age, were exposed in a temperature controlled chamber at two different ambient temperatures (22C and 38C). Five hens at each time were held in the chamber and allowed to be there for one hour. Blood samples were collected before and after temperature exposure via brachial vein cannulation using heparinized sample tubes. The blood was analyzed in a blood gas analyzer and pH, partial pressure of oxygen (PO₂), partial pressure of carbon dioxide (PCO₂) and bicarbonate concentration (HCO₃) were obtained. The data were analyzed using repeated measures in a factorial 2x2 with proc mixed of SAS Institute Version 8.2 (2002) and a level of significance of 0.05. The results show that at 22C, the W-98 hens had a reduction in blood pH (P=.0456) over the one hour of exposure, along with an increase in PCO₂ (p=.0432). At 38C, there was an increase in blood pH and a reduction in PCO₂ in both strains after one hour exposure (p≤.0005). Also, HCO₃ was reduced at 38C for both strains. There was no difference between the two strains for the changes observed at 38C. These results reinforce the theory that

at higher ambient temperature the birds are susceptible to respiratory alkalosis due to the panting process, which reduces the amount of CO₂ and the amount of HCO₃ in blood, both required by the shell gland to support shell formation. The two strains had the same response in terms of changes in blood pH and PCO₂ when exposed to high ambient temperatures, but when the temperature was sustained at thermoneutral conditions (22C), the W-98 appeared to show a higher stress response that decreased the blood pH and increase PCO₂ in those birds. Because it is unlikely that these changes were temperature-dependent, it may be that the stress response was caused by handling and/or movement to the chamber. The W-98 bird may be more sensitive to various stressors than the W-36.

Key Words: Laying hens, Heat stress, Blood pH

142 Determination of fertility in eggs with staining method. Inkar Castellanos*¹, Jos Barbosa², Guillermo Gaona¹, Jos A. Quintana¹, and Tamas Fehervari¹, ¹*Universidad Nacional Autonoma de Mexico,* ²*Laboratorio Diagnostico Veterinario Cuernavaca.*

The continuous increase of poultry production requires the optimization of all sources though there is an increasing demand for a better estimation of egg fertility before incubation. The method used widely so far as a simple macroscopic inspection of blastodisc with less trustable result sometimes with confusion. With the aim of increase the accuracy of estimation a simple staining method was developed that could give a better result quickly. 250 eggs obtained from different parent stocks were stored no longer than 48 hours after oviposition and sent to the laboratory where they were opened and germinal vesicles were submitted to a staining method using PBS solution for washing away albumen, dimetil sulfoxido (DMSO) for increase membrane permeability and neutral red stain for coloration of germinal discs. The stained germinal discs were observed in stereo microscope with magnifications of 10, 20 and 40x. Fertilized discs showed homogenous staining in the zone pelucida with smooth surface of the disc meanwhile unfertile discs presented vacuoles different in size and position sometimes grouped around the margin of disc. The result was compared with the embryo development in a seven-day long incubation period of eggs proceeded from the same farms and similar result was obtained. Further study is on the way to increase accuracy of the method and to find out the minimum number of eggs needed for analysis enabling to project their result to whole flock.

Key Words: Fertility, Blastodisc, Germinal disc, Zone pelucida

143 Cloacal Gland Development in Male Japanese Quail Selected for Divergent Adrenocortical Responsiveness. R. H. Marin, D. G. Satterlee, T. Lambert, and R. G. Gleiser*, *Department of Animal Sciences, Louisiana State University Agricultural Center, Baton Rouge, LA 70803.*

The present study determined differences in the time course of sexual development in males selected for reduced (LS, low stress) or exaggerated (HS, high stress) plasma corticosterone response to brief restraint. Sexual development was assessed by examination of cloacal gland area (CAREA) and volume (CVOL), the proportion of individuals that produced cloacal gland foam (PICF), and the intensity of cloacal gland foam production (CFP) at 4, 5, 6, 8, 10, 14, and 16 wk of age. These intervals

encompassed ages during which growing, photostimulated quail would be expected to have: 1) no cloacal gland development and foam production (pre-pubescent), 2) partial development of the cloacal gland with some birds producing cloacal gland foam, and 3) full development of the cloacal gland with all birds producing foam (post-pubescent for all birds). Total testes weights (TW) were also measured at 6 and 16 wk of age. Mean CAREA and CVOL were similar in LS and HS quail at 4 wk of age. At 5 wk, LS quail showed a greater ($P < 0.05$) CAREA than HS ones and both cloacal gland size indices were consistently enhanced ($P < 0.05$) in LS quail thereafter. In addition, at 4 and 6 wk of age, a greater ($P < 0.05$) PICF was evident in LS birds. Beyond 8 wk, all birds were in foam production. CFP results generally mimicked those found for CA and CVOL (i.e., LS quail showed higher CFP than HS quail beginning at 6 wk of age), but these line differences were not statistically relevant. LS quail had higher ($P < 0.10$) TW than HS quail at both 6 and 16 wk. The results suggest that selection for reduced adrenocortical responsiveness in *Coturnix* is associated with an accelerated sexual development in males. Further, the enhanced reproductive development in LS males observed in juveniles remained extant at an adult age.

Key Words: Japanese quail puberty, Corticosterone, Stress, Cloacal gland, Testes weight

144 Pubic Spread Development and First Egg Lay in Japanese Quail. D. G. Satterlee*, R. M. Gleiser, and R. H. Marin, *Department of Animal Sciences, Louisiana State University Agricultural Center, Baton Rouge, LA 70803.*

Measurement of the spacing between the pelvic ("pin") bones is used to determine the state of sexual development of female broiler breeders. Herein, we examined pubic spread (PS) at selected intervals before, during and after puberty in *Coturnix*, an avian species often used for extrapolation of data to more valuable poultry stocks. PS was determined by first constructing different sets of wooden applicator sticks joined together to represent fixed "gauges" of increasing widths. The stick sets used an appropriate number of applicators that, when glued together, represented increasing one stick (~2.2 mm) incremental distances from 4 (~8.8 mm) to 13 (~28.6 mm) sticks. These stick gauges were then placed between the pin bones until the best fit was detected. Within a bird ($n = 80$), PS measurements were made at 15 different ages (beginning at 21 and ending at 84 d of age). PS measures were regressed with age at first egg lay (FIRST) to determine the age when measurement of PS best predicts the initiation of oviposition. A cubic relationship ($PS = 22.4 - 0.99d + 0.03d^2 - 0.0002d^3$; $r = 0.98$, $P < 0.0001$) was found to exist between age and PS. The sigmoid best-fit curve of this relationship showed that: 1) little development in PS was evident between 21 to 38 d (prepubescence), 2) rapid and increasing development occurred between 42 to 59 d, and 3) no further development occurred thereafter (from 63 to 84 d of age; postpubescence). Regressions of PS with FIRST showed that the optimum age range to make a PS measurement to predict the onset of egg lay was sometime during 49 to 52 d of age (FIRST (d) = $89.64 - 1.63*(PS, \text{mm})$, $r = .69$, $P < 0.00001$ and FIRST (d) = $97.98 - 1.93*(PS, \text{mm})$, $r = .71$, $P < 0.00001$, respectively). The data demonstrate the usefulness of making pubic measurements in Japanese quail to predict sexual development and the initiation of egg lay.

Key Words: Japanese quail, Pubic spread, Egg lay, Puberty

145 Antiangiogenic factor from TD cartilage. B.K. Urie*¹ and W.D. Berry¹, ¹*Dept. Poultry Science, Auburn University.*

Tibial dyschondroplasia (TD) is a consequence of delayed chondrocyte maturation and inhibition of vascularization of the metaphyseal cartilage. This disease is manifested as a mass of nonmineralized avascular cartilage in the metaphysis of the proximal tibiotarsus and other long bones. Various collagen components of cartilage inhibit vascular invasion. However, antiangiogenic properties vary with collagen type. The 20 kD C-terminal fragment of collagen XVIII, known as endostatin, is a particularly potent antiangiogenic compound. This raises the possibility that abnormal production of an antiangiogenic collagen type contributes to development of the TD lesion. To test this hypothesis, collagen extracts were prepared from lesion cartilages from chickens selected for high and low expression of TD. These extracts were applied to a chick chorioallantoic membrane angiogenesis assay system (CAM assay). The angiogenic factor, basic fibroblast growth factor (FGF), and the antiangiogenic factor, endostatin, were applied as positive and negative controls. Initial results confirmed the angiogenic effect of FGF and the antiangiogenic

effect of endostatin in the CAM assay. Crude extracts of TD cartilage exhibited antiangiogenic effects. Studies are underway to identify the specific antiangiogenic factors.

Key Words: tibial dyschondroplasia, antiangiogenic, endostatin

146 The usage of dimethyl sulphoxide and polyvinylpyrrolidone for cryo preserving cock spermatozoa. JA Herrera¹, Ja Quintana*², M Betancourt¹, MA Lpez³, and R Fierro¹, ¹*Departamento de Ciencias de la Salud, UAM-I*, ²*Departamento de Aves, FMVZ, UNAM*, ³*Departamento de Biología de la Reproducción UAM-I.*

Cryo-preservation allows semen banks creation in order to preserve pure germinal lines. Cryoprotectors are divided in those which cross cell membrane such as glycerol or dimethyl sulphoxide (DMSO) and those which act externally, like polyvinylpyrrolidone (PVP). The objective of this research was to determine and compare indicators of basic sperm evaluation post collection and post conservation with DMSO and PVP, due to lack of recent investigations with the last one. Thirteen sperm pools from five cocks were frozen. BPSE was employed as buffer, at 5 C, and a final dilution of 1:8 and added with DMSO 7 % or PVP 3 %. 200 IU drops were dry-ice frozen to preserve them in cryo-tubes of liquid nitrogen. Average of indicators for fresh seminal evaluation were (plus and minus standard deviation): motility 81.8 % 4; livability 93.3 % 2.79; normal morphology: 97.8 % 1.3; pool volume: 512 43.9 IU; pool concentration: 1680.6 340.5 x 106. Post freezing: motility: 47.3 % 13.3 and 30.7 % 4; livability: 83.5 % 2.7, and 90.2 % 2.2; normal morphology: 94.5 % 2.2, and 95 % 1.7, by using DMSO and PVP, respectively. Statistical difference ($p < 0.01$) between sperm livability and motility was found, but not when comparing normal morphology. About sperm evaluation indicators, a difference was found with the use of both cryoprotectors, although there is previous information where it has been shown that artificial insemination with cryopreserved semen, as an alternative for animal breeding. However, depending on the technique used, the spermatozoon will change, which is reflected on its fertilizing capability.

Key Words: spermatozoon, cock, cryopreservation, DMSO, PVP

147 Encapsulated Poultry Waste Sludge (DAF) into Commercial Poultry Byproducts. John Lee* and John Haid, *ANC Technology LLC, Southwest City, MO.*

Poultry waste sludge from DAF (dissolved air flotation) units is a significant environmental issue for poultry industry. Land spread is often used because it is an easy and inexpensive method. But it causes pollution problems to soil, water and air. The land spread will be affected by future more restricted regulations from governments and more complains from local people. Another method is to cook DAF sludge to about 220 F degree and to centrifuge into wet solid, cloudy water and fat. The cloudy water goes back to DAF unit again. The solid is dried. The fat is sold as low quality fat source at lower price. The fat has high melting point, which needs to be heated into liquid form before use. The processing cost for this method is higher than the value of the DAF solid and fat. The waste sludge has good nutritional value for high level protein and fat. We have developed a new process method to encapsulate the waste sludge into a single encapsulated product at lower processing cost, which includes DAF fat and protein with protein 50 % and fat 42 % on a solid basis. The encapsulated product has no oily physical characteristics even at fat level 42 %, which can be dried at high temperature. The wet solid product has been easily blended with feed-grade poultry meal or feather meal and dried into commercial feed-grade poultry meal and feather meal. The blended feather meal in dry form has crude protein 80 %, fat 10 %, ash 3 %, and moisture 6 %. The blended feed-grade poultry meal in dry form has crude protein 59 %, fat 14 %, and moisture 6 %. This practical process can cover the nutrients in DAF sludge into the usable value-added products for feed applications. The dry processing cost can be reduced significantly because the solid level is increased by mechanical methods. Not only the pollution problem and disposal fee from DAF sludge could be resolved by this new process, but also the product could be converted into millions of dollars. This new process is practical and feasible for commercial production.

Key Words: Waste Sludge, Dissolved Air Flotation, Fat and Protein, Waste Treatment, Byproduct Process

148 Effects of fumonisin B₁ in laying Japanese quail. P. Butkeraitis¹, C.A.F. Oliveira¹, D.R. Ledoux^{*2}, R. Ogido¹, R. Albuquerque¹, J.F. Rosmaninho¹, and G.E. Rottinghaus², ¹Universidade de Sao Paulo, Brazil, ²University of Missouri, Columbia, MO USA.

A 28-d experiment was conducted to evaluate the effects of fumonisin B₁ (FB₁) on egg production and egg quality of young laying Japanese quail fed fumonisin contaminated rations. To this end, 128 7-wk-old birds were randomly distributed into four experimental groups (32 birds per group) and given rations containing 0 (controls), 10, 50 and 250 mg FB₁/kg feed. Each treatment consisted of four replicates of eight quail. Fumonisin B₁ was supplied by *F. moniliforme* culture material that contained 6500 mg FB₁/kg, 2100 mg FB₂/kg, and 680 mg FB₃/kg. Egg production and egg weights were checked daily. Feed consumption and feed conversion were determined weekly. Eggs laid in the last day of each 7-d period were collected and subjected to individual analysis for specific gravity, Haugh units and percentage eggshells. Compared with controls, quail fed ≥ 50 mg FB₁/kg had reduced ($P < 0.05$) feed intake and lower ($P < 0.05$) body weight gain. However, feed conversion was only reduced ($P < 0.05$) in birds fed 250 mg FB₁/kg. Average egg production was significantly lower ($P < 0.05$) in the group fed 250 mg FB₁/kg. Egg weight was also significantly decreased ($P < 0.05$) in birds fed 250 mg FB₁/kg. Average specific gravity and Haugh units were not affected ($P > 0.05$) by FB₁. Eggshell weight was reduced ($P < 0.05$) in birds fed ≥ 50 mg FB₁/kg. However, percentage eggshell was not affected by FB₁. Compared with controls, quail fed ≥ 50 mg FB₁/kg had increased ($P < 0.05$) relative liver weight. Relative kidney weight and relative heart weight were not affected ($P > 0.05$) by FB₁. These results suggest quail are sensitive to the toxic effects of FB₁, and at concentrations which have been reported to occur in feedstuff under field conditions. Data indicate acute exposure to FB₁ at levels ≥ 50 mg/kg could adversely affect quail performance, emphasizing the importance of controlling fumonisin B₁ contamination in quail rations.

Key Words: Fumonisin B₁, Quail, Egg, Quality, Toxicity

149 Effects of Composting Pine Shaving and Sand Litter on Bacterial Counts. K.S. Macklin*, M.A. Davis, J.B. Hess, S.F. Bilgili, M.K. Eckman, J.P. Blake, and R.A. Norton, *Auburn University, Auburn, AL.*

Placing chicks on used litter may lead to unexpected health problems due to the high bacterial counts associated with it. Composting litter between flocks is one possible method of decreasing the bacterial counts. This trial was performed to determine the bacterial counts associated with sand and pine shavings from both composted and non-composted litter. To accomplish this, litter that had been used to rear broilers to 7-weeks of age was utilized. A total of 16 pens were used in which 8 contained pine shavings and the remainder contained sand. Of the 8 pine shavings and sand pens, 4 from each were composted. Composting of the litter was performed by piling the litter in the center of the pen then covering it with a tarp for two weeks. Temperature readings were taken twice daily from the 8-composted pens as well as the ambient temperature. Litter samples were collected both before and at the end of the trial. Collection of litter from the non-composted pens was performed at several random locations, while composted pens had random samples taken from the interior as well as from the exterior. Samples than had water activity, moisture content and bacterial counts measured. Water activity was performed on an AquaLab Water Activity Meter. Litter moisture content was determined by the change in litter weight from samples that had been dried. Bacterial counts were conducted by diluting and plating each pen's litter in triplicate on Plate Count Agar (PCA) and Reduced Blood Agar (RBA). Both media types were incubated at 37C, with PCA being incubated aerobically and RBA anaerobically. After 24 hours bacterial counts were determined. Pine shavings had the greatest initial temperature gain of 55C that lasted for 72 hours, while sand only reached 50C in that time. After the initial temperature gain pine shavings maintained a steady temperature of 35C, while sand maintained it at a steady 40C. Sand had higher water activity than did pine shavings, however sand had substantially lower moisture content than pine shavings. Both the anaerobic and aerobic bacterial counts were lower after composting for both sand and pine shavings versus non-composting. The results of this experiment show that composting is an effective means in reducing the overall bacterial counts for both pine shaving and sand litter.

Key Words: Litter, Composting, Broilers, Bacterial counts, Sand

150 Impact of induced molting on performance and egg quality in spent laying hens reared at various light cycles and feeding regimes. M. Akram^{*1}, Zia-ur-rahman¹, and K. S. Ryu, *Chonbuk National University, Chonju, Korea*, ¹University of Agriculture, Faisalabad, Pakistan.

To investigate the impact of induced molt on post-molt performance and egg quality in spent laying hens reared at various light cycles and feed restriction regimes in pullets, three hundred sixty Bobcock white were individually replaced into cages at the age of 77 weeks. The experiment was completed factorial design with three light cycles (natural, constant, step-down) and four feeding regimes (Ad libitum, 15, 30%, low density feed restriction) in pullets. Diet containing 16% CP and 2800 kcal/kg ME were fed ad libitum after induced molting. Post-molting body weight, egg production, egg weight, feed intake, egg mass, feed conversion and egg quality were fortnightly recorded from 84 to 127 weeks of age. Body weight was significantly higher in the birds raised under natural light and ad libitum feeding ($P < 0.05$). Birds reared under step down light and 15% feed restriction regime in pullets showed significantly higher post-molt egg production than other treatments ($P < 0.05$). However, egg weight was not influenced by light cycle and feed restriction regimes in pullets. Feed intake was the highest in birds reared under natural light, but egg mass gradually decreased, which resulted in poor feed conversion. Eggshell thickness was significantly improved with feed restriction regimes ($P < 0.05$) but not with light cycle. Constant and step down light treatments exhibited superior Haugh unit to that of natural light cycle. It was significantly high in birds reared at 15% feed restriction compared to the other groups ($P < 0.05$). Yolk index was not statistically different by light cycle, but the highest in ad libitum groups. Birds reared with 15 % feed restriction showed lesser blood and meat spots than other treatments. In conclusion, birds reared at step down light and 15% feed restriction in pullets improved the post-molt performance and egg quality in spent laying hens.

Key Words: Induced molting, Spent laying hens, Light cycle, Feed restriction, Post-molt performance

151 Case study: Effect of water treated with KEM SANTM brand liquid acidifier on broiler performance. P.A. Welch*, D. Macklin, M. Burke, D. Sanders, and V. Sewalt, *Kemin Americas, Inc., Des Moines, IA/USA.*

The purpose of this study was to evaluate broiler performance when flocks were supplied water treated with KEM SANTM brand liquid acidifier[KS] continuously throughout the course of one growout, and intermittently during the course of two subsequent growouts. Sixteen broiler houses, representing 496,000 as-hatched Ross x Ross broilers, received water acidified with KS at a concentration of 0.33 ounces per gallon of drinking water [OPGdw] for 40 days in the first trial, and one week applications of 0.33 OPGdw at 14 and 28 days in the second and third trials. Live performance of test flocks was compared to the balance of flocks processed the same week as settled test flocks, and compared to previous flock settlement records. Birds were full-fed and reared on built-up litter consisting of pine shavings. All houses were tunnel ventilated and equipped with evaporative cooling. Birds were initially brooded at 90 F, and bird density was 0.67 ft² per bird. Live body weight (3.83 vs. 3.80 lbs.), feed conversion (1.76 vs. 1.86), livability (96.32 vs. 95.28%), and plant condemnation (0.19 vs. 0.69%) were all improved in test flocks when compared to previous flock settlement records. Under normal conditions broilers consume approximately 5% of their diet as litter, and any form of feed restriction during the growout can cause excessive litter consumption. A variety of poultry pathogens have been isolated from litter, and litter consumption logically can be considered a primary source of microbial contamination of broilers. These results suggest that KS-treated water may indirectly suppress microbial contamination from feed and/or reduce the likelihood of recontamination of the contents of the gastrointestinal tract by pathogenic and saprophytic organisms from litter.

Key Words: KEM SAN, Litter Consumption, Water Sanitation, Total Coliform, Broiler Performance

152 Effect of poultry feed trough delivery devices on hen production and egg quality. P. Tang¹, P.H. Patterson*¹, and V.M. Puri¹, *The Pennsylvania State University, University Park.*

The effect of two different feed trough delivery devices, auger and drag chain, on hen production and egg quality were evaluated in two commercial 83,000 capacity houses. The impact of the 300 m auger and drag chain systems on feed nutrient composition and physical properties were studied. In addition, feed quality might be affected by particles' segregation due to differences in size, and the size reduction due to attrition occurring during the delivery procedure. Hen body weight and egg quality at different locations down the trough was expected to vary with feed quality. The hen parameters included the bodyweight at 21, 39 and 57 wk of age, egg weight, albumen height, Haugh unit, specific gravity, and eggshell weight and thickness at 27 # 63 wk of age. Also a nutrient analysis of feed including protein, amino acids, energy, minerals, and vitamins was performed at age of 39 wk old. Results indicated as hens aged body weight, egg weight and shell weight increased linearly, whereas egg specific gravity, albumen height, and Haugh units decreased ($P < 0.05$). Body weight was lowest at the end of the feeder line (1497g, position-6) and greatest in the middle of the line (1553 and 1551g, positions-3 and 4) with the other positions at intermediate weight. Dietary calories and fat were greatest at the feed hopper (position-1) compared to positions-3, -4 and -6. Other nutrients including CP, total AA, Ash, Ca, P, Na, Zn, vitamins A and D were measured at lower levels at the hopper compared to further down the feed trough. The impact of feed trough chain or auger on egg quality and feed physical properties including size distribution and flowability remains to be determined.

Key Words: Feed trough device, Hen production, Egg quality, Feed nutrients, Particle size segregation

153 Broiler cornea and body weight response to low level ammonia exposure. D.M. Miles*¹, S.L. Branton¹, B.D. Lott², and W.W. Miller³, ¹USDA-ARS Poultry Research Unit, ²Mississippi State University, ³Animal Ophthalmology Clinic.

Recommended ammonia levels in broiler houses are 25 to 50 ppm, the lower of which is set by U. S. regulatory agencies, NIOSH and OSHA, for the human 8-hour exposure limit. Ammonia concentration in a commercial house can easily exceed 150 ppm, especially during the brooding phase. Two trials were conducted where 60 male commercial broilers at 1d of age were placed in each of eight environmentally controlled chambers on 10 cm of fresh, kiln-dried pine shavings. Birds had ad libitum water and food under continuous lighting. Anhydrous ammonia was metered into six of the chambers to maintain 25, 50, and 75 ppm (2 chambers for each level). No ammonia was added to the remaining two chambers (control). At the beginning of each trial, 10 birds were randomly selected from each chamber, permanently identified and had weekly ocular examinations through the remainder of the study. The examiner did not know the chamber origin of each bird. At four weeks, ammonia addition was discontinued and birds were grown to market weights. Broilers in each chamber were weighed weekly. Birds exposed to 25 ppm ammonia demonstrated keratitis at 3 weeks of exposure similar to keratitis observed in birds exposed to 75 ppm ammonia for 1 week. Body weights were most affected by the 75ppm level; this group was 75% of the control at 3 weeks of age, but had recovered to 91% of the control at 7 weeks. Birds exposed to low levels of ammonia demonstrate ocular disease and increasing concentrations of ammonia increase the severity of ocular disease and reduce growth rates.

Key Words: ammonia, broiler, cornea, body weight

154 Effects of Zinc Compounds on the Environment of the Gastrointestinal Tract and Susceptibility of Laying Hens to *Salmonella Enteritidis* During Forced Molt. L. F. Kubena*¹, S. Y. Park², R. W. Moore¹, C. L. Woodward², J. A. Byrd¹, J. L. McReynolds², M. R. Burnham¹, M. E. Hume¹, S. G. Birkhold², and S. C. Ricke¹, ¹USDA/ARS, SPARC, Food & Feed Safety Research Unit, ²Department of Poultry Science, Texas A&M University.

The method most commonly used to induce molting and stimulate multiple egg-laying cycles in laying hens for commercial egg production is feed deprivation. Unfortunately, an increased risk of *Salmonella enteritidis* (SE) may result from the use of this method. Methods to stimulate multiple egg-laying cycles without increasing the risk of SE are needed. In each of three experiments, hens over 50 wk of age were divided into groups of 12 hens and placed in individual laying cages. One wk prior

to dietary changes, hens were put on an 8-h light and 16 h-dark photoperiod that continued for the 9-day experiments. Individual hens in all treatments were challenged orally with 10^4 cfu of SE on the fourth day. Treatments were non-fed hens (molted, M), full fed hens (non-molted, NM), a zinc acetate diet (ZAC), and a zinc propionate diet (ZPR). The zinc diets contained 10,000 mg zinc per Kg of diet. Body weight losses were significantly higher in the M (-24%), ZAC (-11%), and ZPR (-15%) treatments than in the NM (-1%) treatment. When compared with the NM hens, ovary weights were reduced by 67 to 83% in the other treatments. Cecal propionic acid and total volatile fatty acids (VFA) were not significantly altered in any of the treatments; whereas, crop lactic acid was decreased in all the other treatments, when compared with the NM treatment. Crop pH was not consistently altered by treatments but tended to be inversely related to feed consumption. Colonization of SE in the crop and the ceca was higher in the M and ZPR hens. Organ invasion by SE was 7/93 for NM hens, 54/96 for M hens, 15/93 for ZAC hens, and 58/96 for ZPR hens. The use of different concentrations of the zinc-containing compounds may change their efficacy. The ZAC regimen may be a useful tool to induce molting and stimulate multiple laying cycles without increasing the risk of SE.

Key Words: Salmonella Enteritidis, Molting, Laying Hens, Zinc Acetate, Zinc Propionate

155 Evaluation *in vitro* of a vaccine stabilizer using an Infectious Bronchitis and Newcastle Disease combo vaccine administered by the drinking water route. R Merino-Guzman*¹, JA Quintana¹, and J Vidrio², ¹DPA: aves, FMVZ, UNAM, Mexico DF, ²AVIESA, Jal. Mexico.

Vaccination against Infectious Bronchitis (IB), Newcastle Disease (ND) and Infectious Bursal Disease (IBD) by the drinking water route is a common practice in poultry production because it is easy, fast and short time consuming. However, factors such as temperature, hardness and disinfectants in the water can affect the viability of the vaccine virus. Products like skim milk powder are useful to neutralize any chemical contamination in the water. A vaccine stabilizer (BLUE-VAC[®]), which is a concentrate powder of skim milk serum, was tested *in vitro*. Distilled water, mineral water and water from the faucet were used, each kind of water was split in 4: without stabilizer (S-), with stabilizer (S+), with 2 ppm chlorine but without stabilizer (S-Ch+) and with 2 ppm chlorine and stabilizer (S+Ch+). An Infectious Bronchitis (10^5 ID₅₀) # Newcastle Disease (10^7 ID₅₀) combo vaccine was diluted in each kind of water. After 15, 30, 45 and 60 minutes at room temperature, 0.2 ml from each kind of water was inoculated in 5 chicken embryos of 9 days old. After 6 days of incubation, all embryos were sacrificed and evaluated for Bronchitis (gross lesions) and Newcastle (hemagglutination test). Newcastle Disease virus was inactivated in the S-Ch+ water before 45 minutes in distilled water, and before 15 minutes in mineral and faucet water ($p < 0.05$) when compared with S-, S+ and S+Ch+ water. Infectious Bronchitis virus was not neutralized in distilled water in a 60 minutes period, but was inactivated before 15 minutes in both S-Ch+ mineral and faucet water ($p < 0.05$) when compared with S-, S+ and S+Ch+ water. The vaccine stabilizer kept both vaccine viruses active even for 60 minutes in mineral and faucet water with 2 ppm chlorine. This product showed to be useful to preserve the viability of vaccine viruses administered by the drinking water route. It is recommended to carry out the same test with Infectious Bursal Disease virus

Key Words: Vaccine stabilizer, Drinking water vaccination, Infectious Bronchitis-Newcastle Disease

156 Summary of litter analysis for Georgia nutrient management plans. Casey Ritz*, Armando Tasistro, David Kissel, and Parshall Bush, *University of Georgia, Athens, GA USA.*

With the increasing awareness of the potential environmental impacts of land application of manures, Georgia poultry producers are developing voluntary nutrient management plans for poultry litter/manure utilization as fertilizers. From July 2000 to June 2002, 4,154 poultry litter/manure samples were analyzed and the results reported on an "as received" basis. The analysis of data archived in a laboratory database show the following mean nutrient values in % (N, P₂O₅, K₂O) for the various categories of poultry litters (standard deviations in parenthesis): Fresh Broiler Litter (2,903 samples): 3.15(0.60), 2.77(0.81), 2.33(0.62) Stockpiled Broiler Litter (262 samples): 2.78(0.86), 2.84(0.94), 2.29(0.69) Composted Broiler Litter (62 samples): 2.80(0.98), 3.00(1.00), 2.30(0.83) Fresh Layer Litter (209 samples): 2.26(0.83), 3.16(1.34), 2.05(0.81)

Broiler Breeder Litter (325 samples): 2.12(0.79), 3.14(1.17), 1.93(0.63). In the absence of site-specific data these values will provide a good basis for nutrient management planning. The predominant litter category was fresh broiler litter, which includes data for the subset categories "Fresh-Caked" and "Fresh-Full Cleanout". The nutrient concentrations of the two fresh broiler litters are very similar, and are the highest of all litter types. Stockpiled litters contain P₂O₅ and K₂O concentrations similar to fresh litter. Composted broiler litters were about 10% higher in phosphorus and slightly higher in potassium than fresh broiler litter. There is a general (not statistically significant) trend toward increasing N, P₂O₅ and K₂O with increasing number of flock growouts. Litters from layers and breeders are lower in nitrogen, higher in P₂O₅ and similar in potassium and nearly four times higher in calcium concentrations (6.5%) than fresh broiler litters. Application of those litters to crops will help to maintain or raise soil pH.

Key Words: Poultry, Nutrient, Litter

157 Detection by DGGE of modifications in crop and cecal bacterial populations following feed withdrawal and forced molt. M. Hume^{*1}, R. Moore², L. Kubena¹, C. Woodward², S. Park², S. Birkhold², S. Ricke², and D. Nisbet¹, ¹USDA, ARS, SPARC, Food and Feed Safety Research Unit, 2881 F&B Road, College Station, TX 77845, ²Department of Poultry Science, Texas A&M University, College Station, TX 77843.

Spent laying hens often are subjected to forced molt in order to prolong their laying status. One popular method of forced molt involves the removal of access to feed during periods of reduced light. A negative result of total feed withdrawal is increased susceptibility to colonization and infection by enteropathogens. An alternative method to molt induction by total feed withdrawal is to provide hens access to feed containing high levels of zinc, which limits calcium availability. In the current study, a 16S rDNA PCR-based technique, denaturing gradient gel electrophoresis (DGGE), was used to visualize changes in crop and cecal bacterial populations in spent laying hens resulting from total feed withdrawal and the provision in the feed of 10,000 mg/kg of zinc, administered as zinc acetate or zinc propionate. Providing hens with zinc from both sources resulted in crop bacterial DGGE patterns with greater similarity to control patterns than those patterns resulting from complete feed removal. Comparisons of cecal bacterial DGGE patterns indicated that hens provided with dietary zinc acetate or zinc propionate had similarity coefficients 80% or greater. However, DGGE patterns for the hens given dietary zinc shared less than 80% similarity coefficients with bacterial populations from hens given access to feed or subjected to total feed removal. Results from other experiments have shown that hens provided with dietary zinc acetate continue to be resistant to enteropathogen colonization. However, results from the current study indicated that providing hens with dietary zinc from either source resulted in crop and cecal bacterial populations very different from those found in normally fed hens or hens deprived of feed.

Key Words: Molt, DGGE, Zinc, Salmonella enteritidis

158 Early T-maze behavior can predict broiler growth. R. H. Marin^{*1}, D. G. Satterlee¹, S. A. Castille¹, and R. B. Jones², ¹Department of Animal Sciences, Louisiana State University Agricultural Center, Baton Rouge, LA 70803, ²Welfare Biology Group, Roslin Institute (Edinburgh), Roslin, Midlothian, EH25 9PS, Scotland.

In laboratory and on-farm studies in Argentina, broiler chicks that showed a short latency to exit a T-maze (LEX) (HP, high performance) gained more weight than slower ones (LP, low performance). The present study re-examined this relationship and evaluated additional T-maze measures using broilers reared in quasi-intensive, environmentally controlled conditions. A mirror at the end of a corridor in the T-maze stimulates the test chick to leave the start box and to move towards its reflection. Upon reaching the mirror it can see other birds and thereby be stimulated to exit the maze. However, if it hesitates or stops at the mirror its performance categorization may be confounded. Herein, the T-maze performance of 3-d-old broiler chicks was assessed using three measures: 1) latency to exit the start box (LEB), 2) latency to reach the mirror (LRM), and 3) LEX. The fastest (upper 25%) and slowest (lower 25%) of birds within sex and within each T-maze criterion were classified as HP and LP chicks, respectively. Relationships between these measures and body weight within quartiles were examined in 4, 42 and 56-d-old males and females. All HP and LP chicks had similar body weights at 4

d of age but chicks classified as HP according to LEB or LRM measures were heavier than LP ones at 42 and 56 d ($P < 0.02$ for all comparisons). These differences were apparent in both sexes. However, there were no detectable differences in body weight at the latter ages between chicks categorized as HP or LP according to their LEX scores. Our results suggest that: 1) broiler chicks that exit the T-maze start box and reach the mirror quickly gain more weight than the slower birds, and 2) LEB and LRM are better predictors of growth than the LEX value used in previous studies.

Key Words: Breeding program, Body weight, Broiler, T-maze behavior

159 Spatial variability of N and P concentrations within a broiler house. A. S. Tasistro^{*1}, D.E. Kissel¹, and P.B. Bush¹, ¹University of Georgia - Agricultural & Environmental Services Laboratories.

Poultry litter sampling procedures must ensure the taking of representative samples. Exposure of litter to high moisture conditions or waste feed from feeders could influence nutrient concentrations, but the amount of variability is not known. Spatial variability of nutrient concentrations in litter from a commercial broiler house was studied by sampling at 3 positions (waterer, feeder, and center of the house) and at each of 10 sites (5 sites spaced approximately 65 ft. apart on each wing of the house). Results from duplicate determinations of water content and total elemental composition (including water-soluble P forms) showed strong effects of sampling positions and sites. Correlation coefficients between total N (TN) and either Total P (TP), Total Dissolved P (TDP), or Molybdate Reactive P (MRP) were #0.48, -0.72, and #0.71, respectively. Mean TN by the feeder was 4.74%, which was significantly ($p < 0.05$) higher than in the center of the house (3.69%) or by the waterer (3.77%). Mean TN concentrations tended to be lower in the end of the house where chickens were kept at the beginning of their rearing than in the opposite end. Mean TP contents in samples taken by the center of the house and the waterer were comparable (20, 018 and 20, 573 mg P kg⁻¹ on a dry basis), but 21% higher than in those taken by the feeder (16, 712 mg P kg⁻¹ on a dry basis). TDP concentrations varied in a slightly different manner, and were the highest in samples from the waterer (8, 311 mg TDP kg⁻¹ on a dry basis), followed by those sampled by the center of the house (8, 079 mg TDP kg⁻¹ on a dry basis), and with the lowest values in samples taken by the feeder (4, 912 mg TDP kg⁻¹ on a dry basis). As with most other variables, significant interactions between sampling positions and sites were detected, and P contents tended to be higher in the end of the house where chickens were kept during the initial stages of their rearing. These results indicate that care is needed in sampling a broiler house to determine average nutrient concentrations of litter. In order to obtain a representative sample for a broiler house, the numbers of subsamples taken from areas affected by waterers, feeders, and unaffected areas should be proportional to the areas they represent in the house.

Key Words: Poultry litter, Variability, Phosphorus, Nutrient, Sampling

160 An evaluation of meal-time feeding for use in broiler production. K.M. Downs^{*}, Middle Tennessee State University.

A small scale preliminary 48-d investigation was conducted to determine the effects of meal-time feeding of broilers on live performance and carcass characteristics. One hundred fifty straight-run broiler chicks were equally assigned to two treatments with three replicate floor pens (1.49 m², pine shaving litter) per treatment (25 birds per pen; 595 cm² per bird). Treatments included either *ad libitum* feed access from d 0-48 (AL) or *ad libitum* feed access from d 0-7, meal time restricted feeding from d 8-41, and *ad libitum* feed access from d 42-48 (MT). Meal time feeding consisted of one consecutive 4-h feed access period (0730 to 1130) per 24 h (20 h off feed: 4 h on feed). Birds were fed a typical industry program of starter, grower, and withdrawal feeds and managed according to industry guidelines. Body weight, feed consumption, feed conversion, mortality, and shank length were evaluated on d 20, 41, and 48. Ten birds per pen were euthanized on d 48 for subsequent excision of whole, bone-in, skinless breasts and paws for determination of breast and paw yields. Paws were graded based on presence of foot pad lesions and off-pigmentation. On d 20, average body weights were divergent between the AL and MT treatments (AL=794.9 g, MT=521.8 g; $P < .05$), yet adjusted feed conversion was unaffected ($P > .05$) by feeding regime (AL=1.35, MT=1.34). Likewise, early mortality (d 0-20) was similar between treatments ($P > .05$). Measures of shank length on d 20 differed

significantly ($P < .05$) between treatment groups (AL=6.84 cm, MT=5.96 cm). Preliminary conclusions (up to day 20) indicate meal time feeding would be an effective alternative to reduce feed costs. However, the current experimental meal feeding regime would result in excess early growth restriction which may alter market weight and breast meat yield. Further data collection is ongoing.

Key Words: Meal feeding, Broilers, Feed management

161 A comparison of NPIP approved methods for the detection of Salmonella from environmental drag swabs and poultry rinses. Linda Purvis¹, Kelley Intehar*², Charles Young², and Douglas Waltman¹, ¹Georgia Poultry Laboratory, Oakwood, GA, ²IGEN International, Inc., Gaithersburg, MD.

A comparative study was conducted to assess the relative performance of the three NPIP approved methods for detection of Salmonella from drag swabs and poultry rinses. This study evaluated 191 samples, of which 71 were poultry rinse samples and 120 were environmental drag swab samples. All samples were tested by: 1) direct tetrathionate enrichment with delayed secondary enrichment; 2) pre-enrichment followed by selective enrichment; and 3) the PATHIGEN[®] Salmonella Test from the selective enrichment cultures. Both culture-based methods were performed according to 9 CFR 147.12. The presence of Salmonella was confirmed in 42 of the samples tested. Ten of the 42 Salmonella positive samples were identified using direct tetrathionate enrichment with delayed secondary enrichment. Pre-enrichment followed by selective enrichment and the PATHIGEN Salmonella Test identified 36 (86%) of the positive samples. Based on these results, the PATHIGEN Salmonella test was equal to or better than traditional plating methods for the detection of Salmonella in commercial chicken operations and provided results 1-4 days sooner.

Key Words: Salmonella, NPIP, Environmental, detection

162 Risk Factors Associated with the Presence of Salmonella in Post-Chill Broilers. R. W. Wills*¹, R. H. Baily¹, and A. J. Byrd², ¹Mississippi State University, ²USDA, ARS, SPARC, Food and Feed Safety Research Unit.

The objective of this research project was to identify risk factors that are associated with the presence of *Salmonella* in post-chill broiler carcasses. The results of a multivariable analysis of 2007 broilers, sampled in a manner consistent with FSIS *Salmonella* performance standards tests, will be presented. The dependent variable used in the logit model was the presence or absence of *Salmonella* in post-chill broiler carcasses. The independent variables included in the final model were age and weight of birds; days between flocks; number of flocks since litter was changed; litter condition; drinker type; *Salmonella* status of house environment; duration of feed withdrawal; month, time of day and day of week flock was processed; and use of a probiotic in feed, lactic acid in the water at time of feed withdrawal, treatment with a competitive exclusion product, and/or use of a TSP rinse. The independent variables in the model that were found to be associated with *Salmonella* status ($p \leq .10$) included day of the week and month flock was processed, drinker type, days between flocks, *Salmonella* status of house, time of feed withdrawal, use of TSP rinse, use of lactic acid, use of a probiotic in feed, and weight of bird. The study demonstrated that this is a useful approach to identify risk factors that are important in the ecology of *Salmonella* in the production continuum of a poultry complex.

Key Words: Salmonella, Risk factors, Broilers

163 Effects of autoclaving of wild type and low phytate barley on Zn and P utilization by young broilers. L. B. Linares*¹, J. N. Broomhead¹, E. A. Guaiume¹, D. R. Ledoux¹, T. L. Veum¹, and V. Raboy², ¹University of Missouri Columbia, MO USA, ²USDA-ARS Aberdeen, ID USA.

Phytic acid (PA) can form insoluble complexes with minerals like Zn and Ca leading to a decrease in bioavailability of these minerals. Low PA barley mutants (LPB) that show reduction in phytate P (45-90%) with no change in total P have recently been developed. In two recent studies, P and Zn utilization in chicks fed LPB, was higher when compared to those fed wild type barley. Endogenous phytase activity in the barleys may have contributed to the differences in P and Zn utilization. The objective of this study was to evaluate the contribution of endogenous phytase to the observed differences in P and Zn utilization. Two hundred

and forty day-old straight-run broiler chicks were assigned to a 2 x 2 x 3 factorial arrangement of treatments (4 pens of 5 chicks/trt) and fed diets from hatch to day 21. Factors were barley type (wild-type (HC) and LPB mutant (M955), autoclave treatment (non-autoclaved (NAC) or autoclaved (AC; 121°C, 20 kg/cm², 20 minutes), and supplemental Zn (0, 10 or 20 mg/kg of Zn from Zn sulfate.). Barley made up 60% of the diets and was the only source of PA in the diets which contained 1.0% Ca, 0.45% non-phytate P, and 24 mg/kg Zn. Feed conversion, percent toe ash, and percent tibia ash were not affected ($P > .05$) by dietary treatments. Feed intake and body weight gain were greater ($P < .05$) in broilers fed M955 compared to HC. Zinc concentration in toes and tibias were affected ($P < .0001$) by barley type (M955 > HC) and Zn level (20>10>0 mg/kg Zn), and significant barley by Zn interactions were also observed. No main effect of autoclaving was observed for any response variable. Retention of P and Zn were higher ($P < .001$) in broilers fed M955 compared to HC. Zinc retention was also influenced ($P < .0001$) by dietary Zn, and there was a barley type by Zn level interaction ($P < .05$). Chicks fed M955 utilized more dietary Zn and P than those fed HC, and this improved mineral utilization was not due to endogenous phytase activity.

Key Words: Phytic acid, Barley, Zinc, Chicks, Autoclaving

164 Effects of dietary Zn supplementation on broiler growth performance and manure nitrogen retention. W. K. Kim* and P. H. Patterson, The Pennsylvania State University, University Park, Pennsylvania, USA.

An experiment was conducted to evaluate the effects of ZnSO₄ or ZnO supplementation of broiler diets on growth performance and retention of uric acid and total nitrogen in manure. A total of 240 day-old broiler chicks were used for this experiment. Each dietary treatment was replicated three times with 10 birds per replicate. Chicks were fed the control diet for the first 6 d and then treatment diets for the next 12 d. There were eight dietary treatments: the control, CuSO₄-20, ZnSO₄-500, ZnSO₄-1000, ZnSO₄-1500, ZnO-500, ZnO-1000, and ZnO-1500 containing 0, 0, 500, 1,000, 1,500 ppm Zn as ZnSO₄, and 500, 1,000, and 1,500 ppm Zn as ZnO, respectively. All Zn treatments contained an extra 20 ppm of Cu as CuSO₄ to prevent Cu deficiency due to high Zn supplementation. The manure uric acid-N levels of chicks fed the diets containing 1,500 ppm Zn as ZnSO₄ or ZnO were significantly higher than that of the control, whereas there were no significant differences in manure total-N level among the treatments. Weight gain, feed consumption, and feed efficiency of chicks fed the diet containing 1,500 ppm Zn as ZnSO₄ were significantly lower than those of the other treatments, whereas there were no significant differences among the other treatments. Zn levels in manure significantly increased as dietary Zn levels increased. However, there were no significant differences in breast muscle Zn content among the treatments.

Key Words: Zinc, Ammonia, Manure, Nitrogen, Uric acid

165 A forensic ELISA test is suitable for screening and quantifying clenbuterol in tissue and serum samples from chicken. R Merino-Guzman*, A Rojas, and J Molina, Departamento de Produccion Animal: aves, FMVZ, UNAM, Mexico DF.

Clenbuterol is a β_2 agonist used as bronchodilator to treat human asthma and other respiratory conditions, it has marked anabolic effects (increased muscle mass and protein content and decreased fat deposition) leading to abuse by cattle producers to enhance animal performance. In Mexico, clenbuterol use for feeding either cattle, poultry or pork has been banned by law. A forensic commercially available ELISA test was used to screen both negative and clenbuterol spiked samples from chicken (serum, retina, liver, muscle and feather) for human consumption. Feather samples were first washed with 1% SDS and distilled water and then digested with 2M NaOH at 80C for 30 minutes and buffered to pH 6.5 with pure HCl, all other samples were homogenized with distilled water and 2M sodium acetate. Once extracted, samples were centrifuged at 3000 rpm for 5 minutes, supernatant was used for the ELISA test, except serum which was tested without extraction. Spiked samples were inoculated with clenbuterol at 0.31, 0.62, 1.25, 2.5 and 5 ng/ml respectively. All samples were tested in duplicate with the Clenbuterol Quantitative ELISA kit (Neogen Corp., Lansing MI 48912, USA) according with the kit insert. Microwells were read at 450 nm. Clenbuterol concentration were calculated with the Log/Logit software from Neogen Corporation.

Results were for sera: 0, 0.3, 0.8, 1.8, 3.3 and 4.8 ng/ml; for retina: 0, 0.35, 0.7, 1.45, 3.15 and 3.9 ng/ml; for feather: 0.1, 0.65, 1.05, 2.05, 3.45 and 5.8 ng/ml (these results seem to be associated with the 2M NaOH extraction procedure); for muscle: 0, 0.3, 0.7, 1.55, 3 and 4.85 ng/ml; and for liver: 0, 0.4, 0.75, 1.6, 2.45 and 4.25 ng/ml, negative and 0.31, 0.62, 1.25, 2.5 and 5 ng/ml clenbuterol spiked samples respectively. According with these results, this ELISA test is useful for screening and quantifying clenbuterol in several chicken samples, even when this β 2 agonist is not used in Mexico for feeding poultry. It is recommended to carry out the same test with samples from cattle and pigs

Key Words: Clenbuterol, ELISA test, Chicken samples

166 Nutritional Experiments in Broilers Reared in Hot Temperatures: Amino Acids and Sodium. W. S. Virden^{*1}, S. J. Barber¹, C. D. Zumwalt¹, S. L. Branton², D. Hoehler³, and M. T. Kidd¹, ¹Mississippi State University, Mississippi State, MS, ²United States Department of Agriculture, Mississippi State, MS, ³Degussa Corporation, Kennesaw, GA.

Better environmentally controlled broiler houses have resulted in many integrated broiler operations using the same dietary feed formulations in winter and summer months due to minor differences in environmental temperature in the broiler house. Much research has addressed broiler nutrient needs in thermoneutral versus hot temperature conditions. But research evaluating nutrient needs of broilers during moderate temperature conditions (tunnel-ventilated houses during summer months) is lacking. Two experiments were conducted to evaluate the impact of increasing dietary Lys, in addition to other essential amino acids, and the addition of dietary sodium bicarbonate in high and moderate temperature conditions. Test diets in both experiments were based on corn and soybean meal and treatment additions were added at the expense of an inert filler. Experiment 1 was conducted in batteries and evaluated growing broiler (Days 20 to 40) nutritional responses in hot-cyclic temperature conditions (26 to 34 C) and Experiment 2 was conducted in floor pens and evaluated finishing broiler (Days 37 to 49) nutritional responses in environmental temperature ranges that mimic a tunnel-ventilated broiler house during summer months (26 to 31 C). Alternating dietary Lys, Thr, Arg, or sodium bicarbonate did not affect live performance (Experiment 1). Breast meat yield, however, was increased in broilers when dietary Lys was increased ($P < 0.08$) from 100 to 107% of 1994 NRC specifications. In Experiment 2, broilers fed the low crude protein diet in the hot temperature environment had better ($P < 0.06$) livability than broilers fed the high crude protein diet or the low crude protein diet with additional Lys or sodium bicarbonate. Breast meat responses were not affected by Lys in Experiment 2.

Key Words: Lysine, Amino acid, Broiler, Sodium, Heat stress

167 Impact of phase-feeding on CP intake, excretion, and retention of broilers from 21 to 63 d. L. N. Loupe^{*}, H. R. Pope, P. B. Pillai, and J. L. Emmert, University of Arkansas, Fayetteville, AR.

An experiment was conducted to assess the effects of phase-feeding on CP intake, excretion and retention. Individually-housed birds (6 per treatment) were fed diets formulated using recommendations from the NRC or linear regression equations generated from estimates of Lys, SAA and Thr requirements. Two phase-feeding treatments were prepared: standard (PF), and low (PF10), in which predicted Lys, SAA and Thr recommendations were reduced by 10%. For PF and PF10, two diets were prepared that contained Lys, SAA and Thr levels matching predicted requirements for birds at 21 d (high nutrient density) and 63 d (low nutrient density). After being mixed and pelleted, these diets were blended in variable quantities to produce rations matching predicted PF requirements over two-day intervals. Birds were fed a single NRC grower and finisher diet from 21 to 43 and 43 to 63 d, respectively or a series of PF and PF10 diets that were switched every other day. With the exception of weight gain, which was lower ($P < 0.05$) for PF than for birds fed the NRC and PF10 diets, no differences ($P > 0.05$) in growth performance were observed. Crude protein intake (g) of birds fed PF and PF10 diets was numerically reduced during the grower (21-43 d) and total (21-63 d) periods, and significantly reduced during the finisher (43-63 d) period ($P < 0.055$). Regardless of age, CP retention (%) was not impacted by diet ($P > 0.05$), although there was a tendency toward increased CP retention in birds fed the PF10 diet during the finisher period ($P = 0.071$). Excretion of CP (g) during the finisher period (but not the grower period)

was reduced ($P < 0.05$) for birds fed PF and PF10 diets, and total CP excretion was numerically reduced (4.0% and 8.6%, respectively). Coupled with the numerical reduction in total (dry) excreta (4.2 and 4.0% for birds fed PF and PF10 diets, respectively), these data indicate that in addition to economic benefits, phase-feeding may result in environmental benefits.

Key Words: Broiler, Phase-feeding, Crude protein, Amino acids, Crude protein retention

168 Effect of the addition of yeast cell wall material on performance of broilers fed a diet with or without antibiotics. J Arce^{*1}, E Avila², C Lopez², and A Garcia³, ¹FMVZ UMSNH, Mexico, ²Dpa: aves, FMVZ UNAM, Mexico DF, ³Saf Agri, Toluca Mexico.

Two experiments were conducted to evaluate the effect of the addition of yeast cell wall material (YCW) to broiler diets supplemented or not with Flavomycine 50 g/ton as growth promoter (AGP). Live weight (LW), feed consumption (FC), feed conversion ratio (FCR), and mortality (M) were evaluated. For each one of the experiments, 2800 mixed Cobb X Ross broilers of 1 day of age were utilized. In each experiment there was a negative control (NC), without AGP and a positive control (PC) with AGP. In the first experiment, the NC and the PC were compared to the inclusion of 1 kg/ton of feed of YCW and the combination of the YCW + AGP. In the second experiment, 5, 1, and 1.5 kg/ton of YCW were used, and the combination of 1 kg/ton of YCW + AGP. In both experiments, the NC had a lighter ($P < .01$) LW and a higher ($P < .01$) FCR when compared to the PC and the YCW treatments and there were no differences ($P > .05$) for FC and M among treatments. In the second experiment, the broilers supplemented with YCW at 1 and 1.5 kg/ton had a lighter ($P < .01$) LW and a higher ($P < .01$) FCR when compared to the YCW + AGP treatment. However, the group supplemented with YCW at .5 kg/ton had similar ($P > .05$) LW than the YCW + AGP group (2533 vs 2592 g). The YCW + AGP group had a improved ($P < .01$) FCR than the YCW .5 kg/ton (1.88 vs 1.84). In the first experiment, there was a numeric ($P > .05$) advantage in LW for the YCW + AGP group when compared to the PC treatment (2601 vs 2579 g). In the second experiment, these increase in LW and improvement in FCR were statistically different ($P < .01$) in favor of the combination treatment when compared to the PC (2591 vs 2533 g; 1.84 vs 1.89). The inclusion of YCW in the diet of broilers could be an alternative to substitute the AGP or to improve performance in their presence

Key Words: Broilers, *Saccharomyces cerevisiae*, Performance

169 Effect of Dietary Zinc Source on Live Performance of Broilers Resulting from 43-Week Old Hens Fed Various Levels and Sources of Zinc. B. P. Hudson^{*1}, W. A. Dozier, III¹, J. L. Wilson¹, and T. L. Ward², ¹The University of Georgia, Athens, GA, ²Zinpro Corp., Eden Prairie, MN.

Supplementing diets with zinc amino acid complex has improved reproductive performance of broiler breeder hens. Because complexed zinc sources have been associated with increased bioavailability compared to inorganic sources, live performance of broilers may be enhanced by providing complexed zinc to hens and their progeny. This experiment examined the effects of zinc source or level in broiler breeder hen diets on progeny performance. Progeny were also provided different zinc sources to evaluate effects on performance. Slow-feathering, Cobb 500 broiler breeder hens were given one of four experimental diets containing 160 ppm supplemental Zn from ZnSO₄, Availa[®] Zn zinc amino acid complex (ZnAA), a mixture of ZnSO₄ and ZnAA (ZnSO₄+ZnAA, 80 ppm Zn from each), or no supplemental zinc after 20 wk (BASAL). Prior to 20 wk, hens in the BASAL treatment were fed diets containing 160 ppm supplemental zinc. Hatching eggs laid during 43 wk were incubated and hatched. Male chicks were placed in 48 battery cages (7 per cage) and were given feeds containing 140 ppm supplemental zinc from either ZnSO₄ (140 ppm) or ZnSO₄+ZnAA (ZnSO₄, 100 ppm Zn; ZnAA, 40 ppm Zn) during the 17 d production period.

Hen zinc treatment had no influence on progeny BW at hatch or at 17 d. Feed conversion, feed intake, mortality and incidence of leg abnormalities were not influenced by hen zinc treatment. Providing supplemental zinc to hens in the form of ZnSO₄ or ZnAA did not improve progeny performance. Although detrimental effects of zinc deficient hens on chick development have been well documented, zinc levels in the BASAL diet were not low enough to yield negative results on chick performance. Broiler

diets contained sufficient zinc for normal growth and may have negated any effects of low zinc intake by breeder hens. At 17 d, broilers fed $ZnSO_4 + ZnAA$ had lower feed conversion (1.23 vs 1.26, $P < 0.05$) and higher BW (0.59 vs 0.56 kg, $P < 0.05$) than broilers fed $ZnSO_4$. Feed intake, mortality and incidence of leg abnormalities were not affected by broiler zinc treatment. These data indicate that broiler performance may be improved when replacing 40 ppm dietary zinc from $ZnSO_4$ with 40 ppm zinc from Availa[®] Zn zinc amino acid complex.

Key Words: Dietary zinc source, Broiler breeder hen, Broiler performance

170 Lack of effect of phytase supplementation on the metabolizable energy of corn-soybean meal diets. J. S. Talley and W. M. Britton*, *University of Georgia, Athens, GA USA.*

A series of seven experiments was conducted to examine the effect of phytase on the metabolizable energy (ME) of corn-soybean meal based chick starter diets. In the first study, 0, 300, and 600 U/kg of phytase (Natuphos[®]) were supplemented to a diet formulated to contain 0.25% non-phytate phosphorus. As significant improvements from phytase supplementation were recorded in body weight, feed conversion, bone ash, and reduced rickets incidence, it was concluded the sample of phytase employed in these studies was biologically active. The effect of phytase on the ME of a corn-soybean meal based broiler starter diet was evaluated in six experiments, four using a standard AME assay with young broiler chicks, and two with adult SCWL roosters in TME_n assays. In none of the six studies was a significant effect of phytase observed on metabolizable energy. In five studies, phytase supplementation (600 U/kg) was associated with slight numerical decreases in ME, while in the sixth ME values with and without phytase were identical. Results of this study did not confirm the hypothesis that phytase supplementation increases the metabolizable energy of corn-soybean meal diets. This effect, if it does sometimes exist, is apparently of inconsistent magnitude.

Key Words: Phytase, Metabolizable energy, Corn-soybean meal diets

171 A comparison of rapid methods of estimating total and digestible amino acids in protein sources for poultry. C.A. Fritts* and P.W. Waldroup, *University of Arkansas.*

Formulation of poultry diets using digestible rather than total amino acids has been proposed as a means of improving efficiency of protein utilization and reducing nitrogen excretion. In order to accurately formulate poultry diets on a digestible basis it is necessary to have a rapid and accurate means of estimating digestibility of processed protein sources, especially those likely to encounter heat damage during processing. Comparisons were made of different estimates of amino acid digestibility from a common feed sample. Samples of soybean meal and meat meal, the most commonly used protein sources in poultry diets in the United States, were submitted to participating laboratories involved in amino acid analysis to estimate total and digestible amino acids by ion-exchange chromatography, near-infrared reflectance (NIR), and multi-enzyme assay. Variation was observed between estimates of both total and digestible amino acid content of soybean meal and meat meal using the various methods. Greater disagreement was observed among digestible amino acid content of the meat meal sample than for soybean meal. Results of this study indicate that problems in estimating digestibility of processed protein supplements by existing methodology may negate many of the anticipated benefits of formulating diets on a digestible amino acid basis.

Key Words: Amino acids, Digestibility, Bioavailability, Near infrared

172 Field results with broilers fed selenium yeast. F. W. Edens*¹, K. M. Gowdy¹, and A. E. Sefton², ¹*North Carolina State University, Raleigh, NC USA*, ²*Alltech, Inc., Nicholasville, KY USA.*

Our original research, on the use of selenium yeast (Sel-Plex[®], Alltech Inc.) as an aid to improve broiler performance, stimulated research in the USA and around the world. In the USA, some studies indicated that the use of selenium yeast had marginal effects in broiler production, but other studies showed that both BW and FCR were improved significantly along with a decrease in breast fillet drip loss. In the UK, selenium yeast partially replaced sodium selenite in breeder diets, and their progeny were fed diets with a partial replacement of sodium selenite by selenium yeast. The results with several million broilers clearly showed that BW and livability were improved significantly when selenium yeast was added to the diets. In Mexico, several commercial broiler trials with selenium

yeast have been monitored. On broiler farms where selenium yeast was used with grains from the USA, there were significant improvements in BW and FCR. In Australia and Thailand, the use of selenium yeast has resulted in improved BW, FCR and yield of processed carcasses, and in Thailand, the use of selenium yeast singly and in combination with sodium selenite resulted in decreased drip loss from breast fillets. It was of interest that even at 0.1 ppm of selenium yeast in broiler diets in Brazil, there was demonstrable improvement in performance, but even better results were found when the level of selenium yeast was increased to 0.2 and 0.3 ppm while sodium selenite was decreasing. The results from both university and field trial studies have shown that the use of selenium yeast in diets of broilers has the potential to improve performance.

Key Words: Selenium yeast, Broiler, Performance, Livability

173 Liquid L-lysine 50% effectiveness in commercial broilers productive parameters. Valle, V, Krimilda*¹, Arce, M, Jose², Avila, G, Ernesto³, and Lopez, C, Carlos³, ¹*Fermentaciones Mexicanas Mexico, D.F.*, ²*INIFAP Morelia, Mexico*, ³*UNAM Mexico, D.F.*

The effect of a liquid L-lysine 50% source over the broiler productive parameters was evaluated. Two thousand one hundred one day old chicks mixed Ross x Cobb were obtained from a commercial hatchery. The birds were randomly distributed in 7 replicates per treatment (75 birds/replicate). Sorghum meal + soybean meal + sesame meal diets supplemented with two sources of L-Lysine (Liquid 50% and powder 98.5%) respectively. 4 treatments in a 49 feeding period compared: 1) L-lysine HCl (1#49 days); 2) liquid L-lysine 50% (1#49 days); 3) L-lysine HCl (1-21 days) and liquid L-lysine 50% (22-49 days) and 4) liquid L-lysine 50% (1-21 days) and L-Lysine HCl (22-49 days). The results showed that body weight, feed consumption and feed conversion were similar with both L-lysine sources, differences were not significant ($P < 0.05$). Broilers fed with L-liquid lysine 50%, demonstrated similar performance than L-lysine HCl treated in the productive parameter.

Key Words: liquid lysine, broiler chicken, lysine HCl, productive parameters

174 Bone densitometry may be used to predict percent tibia ash in broiler chicks fed varying dietary calcium and phosphorus levels. E. M. Onyango*, P. Y. Hester, and O. Adeola, *Purdue University, West Lafayette, IN.*

The relative sensitivity of tibia bone mineral content and density (BMC, BMD), percent ash and breaking strength as indicators for dietary Ca and P was compared in 3-wk-old broiler chicks. One hundred and eight 7-d old chicks were grouped by weight into 6 blocks. Three corn-soybean meal-based diets were randomly assigned to pens within each block. The diets were low P-, medium P- and adequate P-diet and were formulated to contain 4.0, 5.1, and 7.8 g of total P per kg feed, respectively, and also 5.1, 6.7 and 10.0 g of Ca per kg feed, respectively. Chicks were fed experimental diets for 14 d. On d 22, chicks were killed and tibiae removed from 3 birds/cage. Weight gain, feed intake, feed efficiency, BMC, BMD, shear force and percent ash were determined. The BMC and BMD were determined using dual x-ray absorptiometry. Correlations among the various bone status parameters were measured. Growth performance parameters, BMC, BMD, shear force, and percent ash of tibiae showed a linear increase as dietary concentration of Ca and P increased. Correlation coefficient between percent ash and BMC, BMD, or shear force was 0.84, 0.86, and 0.56, respectively. Correlation coefficient between dietary Ca and BMC, BMD, shear force, or percent ash was 0.76, 0.80, 0.36, or 0.81, respectively, and between dietary P and BMC, BMD, shear force, or percent ash was 0.75, 0.80, 0.35, or 0.81, respectively. The correlation coefficient between BMC and percent ash, or shear force was 0.84 and 0.49, respectively whereas that between BMD and percent ash, or shear force was 0.86 and 0.52, respectively. A regression model for predicting percent ash using BMD was fitted. The model was: bone ash = 26 + (215 × BMD) with an R² of 74 %. It is concluded that bone ash, BMC, and BMD may be more sensitive than shear force as indicators of dietary Ca and P concentrations in broiler chicks and that bone densitometry may be used to predict percent tibia ash.

Key Words: Broiler chick, Bone ash, Bone densitometry, Bone strength

175 A Spreadsheet-Based Instructional Feed Formulation Program for Poultry and Swine (WUFFDA). G. Pesti^{1*}, E. Thomson², R. Bakalli¹, B. Leclercq³, A. Shan⁴, A. Atencio¹, J. Driver¹, C. Zier¹, M. Azain¹, M. Pavlak⁵, D. Vedenov¹, F. van de Vyver⁶, J. F. Menten⁷, J. O. Sorbara⁷, N. Senkoylu⁸, B. Berisha⁹, and R. K. Seon¹⁰, ¹University of Georgia, ²University of New England, ³Recherches Avicoles INRA, ⁴Northwest Agricultural University, ⁵University of Zagreb, Croatia, ⁶University of Pretoria, Pretoria, South Africa, ⁷ESALQ #USP, Piracicaba SP Brasil, ⁸Trakya University, Tekirdag, Turkey, ⁹Technical University of Munich, Freising-Weihenstephan, Germany, ¹⁰Chonbuk National University, Chonbuk, South Korea.

A Microsoft Excel-based workbook has been developed for teaching poultry and swine nutrition. The program uses the Solver feature of Excel to find the least-cost solution to linear programming feed formulation problems. The program is available in Afrikaans, Albanian, Chinese, Croatian, English, French, German, Korean, Portuguese, Russian, Spanish, or Turkish. The complete WUFFDA package consists of 3 files: The WUFFDA Excel workbook itself; a Microsoft PowerPoint file that can be used in lecture or auto-tutorial situations to teach the Excel program; and a Microsoft Word file that explains the Excel Program. All may be downloaded free from <http://www.ces.uga.edu/ES-pubs/WUFFDA.htm>. Problem size is limited to 25 ingredients and 34 nutrients. Several ingredient composition tables are included in the workbook, and new ingredients can easily be copied into the current, or active, problem. Nutrient specifications are included for broiler, laying and breeding chickens, turkeys, quail, ducks, guineas, goslings, and growing, gestating, and lactating swine.

Key Words: Feed Formulation, Spreadsheet, Poultry, Swine, Instruction

176 Effects of Calcium and Protein on the Growth and Performance of Modern Broiler Chickens. J. P. Driver^{*}, G. M. Pesti, R. I. Bakali, and H. M. Edwards, *University of Georgia, Athens, GA, 30605.*

Two experiments were conducted to compare Ca requirements of male and female broilers during the starter and grower phase as well as the effects of two levels of dietary protein on Ca requirements over the grower phase. Experiment 1 was conducted with six levels of dietary Ca (0.325, 0.4, 0.475, 0.55, 0.625 and 0.9% based on NRC ingredient composition tables) in corn-soybean meal diets with 0.45% non-phytin phosphorus and 1100IU/kg vitamin D³ in combination with two levels of dietary protein (17 and 23%). However, dietary calcium analyses using the Technicon auto-analyzer methodology described in Clinical Chemistry (1965) indicates that calculated values from the NRC (1994) tables gave Ca predictions that were between 0.03 and 0.175% too low. Diets were fed to males and females separately in a factorial arrangement from 18 to 42 d in floor pens. A second experiment was conducted using male and female broilers fed the same six levels of Ca as in Experiment 1, but with a single level of crude protein (23%). These diets were fed to birds from 0 to 16 d in battery brooders.

In Experiment 1, neither sex demonstrated a significant body weight gain or feed conversion response to the different levels of Ca. However, higher body weight gains were obtained with birds fed the 23% protein diets (1.97 and 1.66kg for males and females, respectively) vs. the 17% protein diets (1.93 and 1.62kg for males and females, respectively). Feed conversion was also lowest for the birds fed the 23% protein diets (1.82 and 1.88 for males and females respectively) vs. the 17% diets (1.98 and 2.01 for males and females respectively). Percent tibia ash did not respond to increasing Ca levels beyond 0.625% Ca. In Experiment 2, body weight gain increased linearly up to 0.55 and 0.625% dietary Ca for males and females respectively. Feed conversion decreased linearly with increasing Ca up to 0.625% Ca. Tibia ash also increased linearly with maximal levels at 0.9% Ca for both sexes. These results suggest that current broiler starter NRC requirements (1.0%) for Ca are sufficient for maximum bone ash but Ca requirements for grower birds (0.9%) are excessive.

Key Words: Broiler, Calcium, Requirements

177 Effect of reducing cage density on hen performance and economics of second cycle (molted) commercial Leghorns. S. S. Sohail^{*}, M. M. Bryant, and D. A. Roland, *Auburn University, Auburn, Alabama.*

In 2002, the United Egg Producers announced a new certification program that requires a significant reduction in cage density to produce eggs meeting animal welfare guidelines. Because there is little information concerning the influence of cage density on hen performance or increased cost associated with reduced hen numbers, this study was conducted to determine the effect of reducing cage density on hen performance and profits. Hy-Line W-36 hens (n=1120) were molted at 68 wk of age. They were kept in an environmentally controlled house at 3 or 4 per cage (64 or 48 in²/hen). After molting hens were fed a commercial layer corn-soy diet containing 0.92% lysine and 1283 kcal ME/lb feed for 14 weeks. They were housed at a 24 hr average temperature of 78F and were provided a 16 hr light: 8 hr dark photoperiod. Reducing cage density increased 14 wk average feed consumption (85.7 to 89.6g; P ≤ 0.001), egg production (73 to 75%; P ≤ 0.05), and egg weight (61.7 to 62.4g; P ≤ 0.05), but had no effect on egg specific gravity or mortality. Reducing cage density reduced overall feed efficiency from 3.17 to 3.11 lb feed/dozen eggs (P ≤ 0.05), and improved egg mass from 45.0 to 46.8 g/h/d (P ≤ 0.01). Economic analysis of reducing cage density indicated an increase in return (egg price minus feed cost) with the increase dependent upon the price spread between large, extra large and jumbo eggs. The significant improvement in feed efficiency would help offset some of the cost involved in reducing cage density (hen numbers) during summer months. Results could be different for larger strains, and under different environmental conditions.

Key Words: Cage density, Economic analysis, Hen

178 Impact of Feeding *Monascus Purpureus* on Performance of Broiler Chicks and Egg Quality in Laying Hens. K. S. Ryu^{*}, J. H. Park, and M. S. Ryu, *Research Center for Industrial Development of Biofood Materials, Chonbuk National University, Chonju, Korea .*

Two experiments were conducted to investigate the effect of feeding *Monascus Purpureus* (MP) on the performance of broiler chicks and egg quality in laying hens. In Expt 1, MP were supplemented with 0, 0.5, 1.0, 2.0, 4.0% in basal diets. Weight gain, feed intake, feed conversion were examined for five weeks and blood cholesterol, protein were measured at the end of experiment. In Expt 2, diets supplemented with 0, 0.5, 1.0, 2.0, 3.0% MP were fed for four weeks. Egg quality and yolk cholesterol were weekly measured. In Expt 1, Basal diets containing CP 21.5, 19%, ME 3,100, 3,200 kcal/kg were fed *ad libitum* for starting and finishing period, respectively. In Expt 2, laying diet contained CP 16%, ME 2,800 kcal/kg. In Expt 1, weight gain of birds fed 2.0% MP was significantly higher than other treatments (P<0.05) at five weeks of age. Feed efficiency was not statistically different, but tended to improve in 1.0 and 2.0% MP supplemental groups compared with control. Total serum cholesterol tended to be lowered in 2.0% MP supplemental group but were not statistically different. In Expt 2, albumen height and Haugh unit of birds fed 1.0, 2.0% MP were higher than other treatments, but was not statistically different. Yolk cholesterol of hens fed MP showed no significant difference. The present study indicated that 2.0% MP supplemental diets can maximize growth of broiler chicks, improve egg quality.

Key Words: *Monascus Purpureus*, Broiler chicks, Performance, Laying hens, Egg quality

179 PUFA content in poultry meat as affected by dietary unsaturation, α -tocopherol supplementation and cooking process. L. Cortinas, C. Villaverde, M.D. Baucells, J. Galobart^{*}, and A.C. Barroeta, *Department of Animal and Food Science, Facultat de Veterinaria, Universitat Autònoma de Barcelona.*

The present work was carried out to study the effect of dietary polyunsaturated fatty acids (PUFA) and α -tocopherol on the PUFA content in raw and cooked thigh meat and whole chicken. One hundred and ninety-two female broiler chickens were distributed into 16 experimental treatments resulting from the combination of 4 levels of dietary PUFA (15, 34, 45 and 61 g/kg) and 4 levels of supplementation with α -tocopheryl acetate (0, 100, 200 and 400 mg/kg). The unsaturation degree was achieved by replacing linseed and fish oil to a basal diet enriched with 9% tallow. The PUFA content in raw thigh, as well as in the whole animal, increased

linearly as the inclusion of dietary PUFA increased. For raw thigh the following equation was calculated: $y = 2.489x + 17.512$ ($R^2 = 0.855$). The similar response was observed in particular PUFA, like the $\omega 3$ and $\omega 6$ precursors. Thus, a linear increase in linolenic acid (LNA) and linoleic acid (LA) content was observed as the dietary content in these fatty acids increased (LNA: $y = 2.7041x + 2.9268$, $R^2 = 0.9392$; LA: $y = 2.4575x + 7.4791$, $R^2 = 0.6717$). The lineal relation between PUFA in diet and in cooked thighs ($y = 2.5093x + 11.303$, $R^2 = 0.9171$) was parallel to that obtained in raw thigh. Furthermore, the cooked process caused a significant reduction in PUFA content ($P = 0.0124$). The PUFA content in thigh and whole animal was not modified by the dietary α -tocopheryl acetate level. In this study we have assessed the lineal relation between PUFA content in diet and in thigh and whole animal. The PUFA content in tissues studied increases linearly as the inclusion of dietary PUFA independently of α -tocopherol supplemented level. Furthermore, cooking reduces PUFA content in thighs.

Key Words: Polyunsaturated Fatty acids, dietary α -Tocopherol supplementation, raw thighs, cooked thighs, whole Chicken

180 Probiotics do not alter the intestinal microbiota of broiler chickens. Adriana A. Pedroso¹, Marcio R. Lambais¹, Jos F. M. Menten*¹, Flvio A. Longo¹, Jos O. B. Sorbara¹, Aline C. Racanici¹, and Juliano B. Gaiotto¹, ¹USP/ESALQ, Piracicaba, SP/BRAZIL.

The techniques of polymerase chain reaction followed by denaturing gradient gel electrophoresis were used to characterize the changes in intestinal microbiota of broilers as a result of feeding diets supplemented of probiotics growth promoters. At 21 days of age, chicks raised in two experiments conducted in batteries or in floor pens had the small intestine mucosa scraped, the total DNA extracted, the V3 region of 16S rDNA amplified by PCR and analyzed by DGGE. The analysis of amplicons patterns indicated that the supplementation of lyophilized poultry microbiota (MICROBIOTA) in the water, or live yeast (YEAST) in the feed, or the combination of *Lactobacillus reuteri* and *L. johnsonii* in the water plus *Bacillus subtilis* in the feed (LA/BS), or unsupplemented diets (CONTROL) did not differ significantly. The six replicates birds from each treatment did not show significant individual differences. The number of species found in the intestinal microbiota, evaluated by the number of amplicons, was affected by probiotics when the birds were raised in batteries ($P < 0.05$). YEAST resulted in lower number of amplicons than CONTROL ($P < 0.05$). The hierarchical clustering produced from samples obtained in chickens of different treatments raised in batteries or in floor pens indicated that the probiotics did not result in distinct branching. Thus, the intestinal ecosystem of birds was not modified by the probiotics. The amplicons encountered when the commercial products were analyzed were not observed in the samples scraped from gut after probiotic supplementation. Under the experimental conditions adopted, the probiotics did not show the ability to colonize the intestinal epithelium. Samples analyzed considering the two rearing systems showed a pattern of branches specific, for each environment. A newborn chick tested revealed greater similarity with birds raised in batteries than in the pens. In all clusters generated the pure bacterial-based probiotics formed the same branching pattern. These results demonstrate that techniques independent of culture, such PCR followed by DGGE combined to measure of ecological diversity may be useful in monitoring the intestinal microbiota resulting from feeding probiotics. (Supported by FAPESP)

Key Words: Probiotics, Broiler, DGGE, Diversity, Intestinal

181 Feeding non-molted Hy-Line W-36 hens (Phase-4) for maximum profits. S. S. Sohail*, M. M. Bryant, and D. A. Roland, Auburn University, Auburn, Alabama.

ABSTRACT: Molting of hens has been a common practice to improve hen performance and profitability in the layer industry. Because most hens in the past have been molted, limited data is available on feeding modern strains of non-molted hens during Phase-4 (~65 to 80 wk of age). To optimize performance and profitability of non-molted hens, a study was conducted on Hy-Line W-36 hens ($n = 800$; 68 wk old) to determine their optimum nutrient requirement from 68 to 83 wks of age. Replicates of 16 hens each were assigned to five iso-caloric diets (1282 kcal ME/lb feed) containing 15.6, 16.2, 16.8, 17.5, and 18.5% protein. Increasing protein from 15.6 to 18.5% had no influence ($P \geq 0.05$) on egg production (EP), feed consumption (FC), egg weight (EW), egg specific gravity (SG), feed efficiency (FE), or egg mass (EM) from 68 to 73 wk of age.

The same hens were then fed diets containing 14.5, 15.0, 15.6, 16.2, and 16.7% protein from 74 to 83 wk of age. Dietary protein had no significant effect on EP, FC, EW, or SG from 74 to 83 weeks. However, hens fed diet containing 15.6% protein maximized EM ($P \leq 0.05$) at 43.6 g/h/d. Economic evaluation indicated that under trial conditions, the requirement for maximum profits for protein, lysine, methionine plus cystine, and kcal ME/hen/d was 15.16g, 0.77g, 0.57g, and 275 kcal/hen/d respectively.

Key Words: Economical diet, Egg mass, Non-molted hens

182 Econometric Feeding of Commercial Layers: 5. Response Comparison of DL-Methionine and Methionine Hydroxy Analogue with Milo-Soybean Diet Using Different Regression Models. Z. Liu*, A. Bateman, M. M. Bryant, and D. A. Roland, Sr., Auburn University, Auburn, Alabama.

The relative bioefficacy of methionine hydroxy analogue free acid (MHA) was compared to DL-methionine (DLM) on laying hens with a milo-soybean diet, which was formulated to have low methionine and cystine level to increase the sensitivity of methionine deficiency. Five graded supplemental levels (0.02, 0.04, 0.06, 0.08 and 0.10%) of methionine from DLM or MHA were added to the basal diet (containing 0.20% methionine) on equimolar basis. Five different models were used to determine the bioefficacy of MHA compared to DLM. Three of the models were exponential models, with supplemental methionine level based on weight, supplemental methionine intake based on weight and supplemental methionine intake based on molar as the independent variable respectively. Two of the models were slope-ratio models, with supplemental methionine intake based on weight and supplemental methionine intake above basal diet based on weight as the independent variable. Regression analysis showed the average bioefficacy was 122% or 139% (egg production), 109% or 124% (egg mass) and 89% or 101% (egg weight) on weight basis or molar basis respectively. All bioefficacy values were not significantly different from 88% on weight basis or 100% on molar basis ($P \leq 0.05$), but were significantly different from 65% on weight basis or 74% on molar basis ($P \leq 0.05$).

Key Words: DL-methionine, methionine hydroxy analogue, bioefficacy

183 Econometric feeding of commercial layers: 6. Strain comparison (Dekalb and Bovans White) of nutrient requirements for performance and optimum profits (Phase III). A. Bateman*, M. M. Bryant, and D. A. Roland, Sr., Auburn University, Auburn, Alabama.

A study was conducted to compare performance and nutrient requirements of Bovans and Dekalb White hens for maximum profits when kept under warm temperatures (78F average) during Phase III (weeks 54-65). Three diets were used for each of the two strains for a 2×3 factorial arrangement. These diets contained 17.5, 16.2 or 15.1% protein (0.92, 0.83 or 0.75% lysine) and 2,847 kcal ME/kg. The criteria measured were egg production, feed consumption, egg weight and egg specific gravity. Strain of bird significantly affected ($p \leq 0.05$) egg weight, egg specific gravity and feed conversion (g feed/g egg). The Dekalb White hens had higher egg weight (64.2 vs 61.2 g) and egg specific gravity (1.084 vs. 1.079) and lower feed conversion (2.11 vs. 2.19) than the Bovans White hens. Diet had a significant effect on egg production and feed conversion (g feed/g egg). Hens fed the two highest protein diets had the highest egg production and the lower feed conversions, followed by the lowest protein diet. Feed conversion (g feed/g egg) was significantly affected by strain and diet. Feed consumption was not significantly affected by strain or diet. Optimum profits under trial conditions were obtained in both strains using the moderate protein diet. For optimum profits during Phase III, Dekalb White hens required 916 mg lysine, 685 mg TSAA, 17.9 g protein and 314 Kcal ME per hen per day. The Bovans White hens required 932 mg lysine, 696 mg TSAA, 18.2 g protein and 320 Kcal ME per hen per day. The Dekalb White hens required 17.0 mg lysine, 12.7 mg TSAA, 0.33 g protein and 5.84 Kcal ME/g egg, while the Bovans White hens required 17.7 mg lysine, 13.2 mg TSAA, 0.34 g protein and 6.06 Kcal ME/g egg for optimum profits.

Key Words: protein, Bovans White, DeKalb White

184 Broiler chickens are susceptible to extended feeding of grains naturally-contaminated with *Fusarium* mycotoxins. H.V.L.N. Swamy*¹, T.K. Smith¹, E.J. MacDonald², and A.E. Sefton³, ¹University of Guelph, Guelph, Canada, ²University of Kuopio, Kuopio, Finland, ³Alltech Biotechnology Center, Nicholasville, KY.

An experiment was conducted to investigate the effects of feeding grains naturally-contaminated with *Fusarium* mycotoxins on feed intake, weight gain, feed efficiency and brain regional neurochemistry of broiler chickens. Three pens of 30 d-old male broiler chicks per pen were fed one of the four diets, containing graded levels of grains naturally contaminated with *Fusarium* mycotoxins, for 56 d. The diets included (mg/kg of deoxynivalenol, fusaric acid, zearalenone and 15-acetyl deoxynivalenol); control (0.56, 15.2, <0.2, <0.2), low level of contaminated grains (5.9, 19.1, 0.4, 0.3) and high level of contaminated grains (9.5, 21.4, 0.7, 0.5). The fourth diet containing the high level of contaminated grains was supplemented with 0.2% glucomannan polymer (GM polymer) to test its efficacy in preventing *Fusarium* mycotoxicoses. Body weight gains and feed consumption of chickens fed contaminated grains decreased linearly during the grower phase (21 to 42 d, P=0.04). Efficiency of feed utilization, however, was not affected by diet. On d 56 of the experiment, pons, hypothalamus and cortex sections of brain were excised and analyzed for concentrations of neurotransmitters. Inclusion of contaminated grains in the diet linearly increased pons concentrations of 5-hydroxytryptamine (5-HT, serotonin), 5-hydroxyindoleacetic acid (5-HIAA), norpinephrine, 3-methoxy-4-hydroxyphenylethylene glycol, dopamine, and homovanillic acid (P<0.05). Supplementation of GM polymer to the contaminated diet non-specifically increased hypothalamic 5-HIAA to 5-HT ratio. It was concluded that broiler chickens, in contrary to the previous findings, are susceptible to extended feeding of grains naturally-contaminated with *Fusarium* mycotoxins. Altered brain neurotransmitter concentrations might partially explain the *Fusarium* mycotoxin-induced reduction in feed intake and weight gain of broiler chickens.

Key Words: chickens, deoxynivalenol, fusaric acid, *Fusarium* mycotoxins, neurochemistry

185 Broiler performance on enzyme-supplemented defatted rice bran diets. O. Puminn* and M. O. Smith, The University of Tennessee, Knoxville, TN.

Two experiments were conducted to determine the effects of commercially produced phytase and pentosanase enzymes on performance of broilers fed defatted rice bran (DRB) diets. In first experiment, six replicate groups of chicks were assigned to five dietary treatments: 1) basal diet containing 25% corn starch, 2) 10% DRB diet, 3) 10% DRB diet + phytase, 4) 25% DRB diet, and 5) 25% DRB diet + phytase. In experiment 2, five replicate groups of chicks were assigned to dietary treatments similar to first experiment, but using mixed enzymes phytase and pentosanase instead of phytase alone plus an additional basal corn-soy diet. Levels of available phosphorus in all diets containing enzyme(s) were reduced by 0.1%. Birds were raised on floor pens for 21 days posthatch. On day 22, birds from each treatment were transferred to each of two environmental chambers (thermoneutral, 23.9 C and heat stress, 35 C). Data were analyzed using a randomized block design, with factorial treatment arrangement. In both experiments, body weight gain and feed conversion ratio of chickens fed diets containing 25% DRB without enzyme(s) during 0-21 days were significantly better than the other treatments (P<0.0001). Adding enzyme(s) in diets did not improve total weight gain, total feed conversion ratio, or percent carcass (P>.05). Percent thighs and abdominal fat were significantly higher in diets containing 25% DRB. No differences were found in percentage of the other carcass parts or in weight percentage of intestinal parts. There were no treatment differences in dry matter, ash, calcium, and zinc content of tibia. However, percent phosphorus was significantly higher (P<.01) in the diet that contained 25% DRB with phytase in first experiment. In experiment 2, percentit

Key Words: Defatted rice bran, Phytase, Pentosanase, Broiler, Heat stress

186 The effect of *Fusarium* mycotoxins on the efficacy of exogenous phytase in broiler diets. Maher Zaytoun*¹, T. Smith¹, and A. Sefton², ¹University of Guelph, ²Alltech biotechnology centre.

Trichothecene mycotoxins can cause intestinal villi disruption, which can lead to malabsorption and a subsequent decrease in weight gain. Broilers can not utilize phytate phosphorus unless supplemented with phytase

to catalyze hydrolytic cleavage of the phosphate group from phytic acid thereby liberating phosphorus from feedstuffs of plant origin. 2 experiments were conducted to determine if trichothecene mycotoxins can reduce the efficacy of exogenous phytase in broiler diets. In first experiment dietary treatments were 1 Adequate phosphorus 2 Phosphorus deficient 3 Phosphorus deficient+phytase 4 Phosphorus deficient+mycotoxin 5 Phosphorus deficient+phytase+mycotoxin 6 Phosphorus deficient +phytase+mycotoxin+binder. 252 day-old chicks (6 cages of 7 birds per diet) were fed experimental diets with or without naturally-contaminated grain with *Fusarium* mycotoxins and with or without exogenous phytase and mycotoxin binder for 21 days. Supplementation of *Fusarium* mycotoxins to phosphorus-deficient diets with or without phytase significantly improved phosphorus retention compared to controls (p<0.05). The feeding of diets naturally-contaminated diet with *Fusarium* mycotoxins did not reduce phosphorus retention. It was concluded that *Fusarium* fungi and mycotoxins may have phytase-like activities. In second experiment, a total of 540 day-old chicks (3 pens of 30 birds per diet) were fed experimental diets with or without grain naturally-contaminated with *Fusarium* mycotoxins and with or without exogenous phytase and mycotoxin binder for 41 days. Diets naturally-contaminated with *Fusarium* mycotoxins without exogenous phytase significantly reduced weight gain throughout starter and grower periods compared to controls (p<0.05). Diets naturally-contaminated with *Fusarium* mycotoxins significantly reduced plasma creatine kinase and increased plasma lipase activities. Neither supplementation with mycotoxin binder nor phytase affected creatine kinase and lipase activities. It was concluded that trichothecene mycotoxins did not reduce the efficacy of exogenous phytase in broiler diets.

Key Words: Broiler, *Fusarium* mycotoxins, Phytase, Phosphorus retention

187 The effect of two phytase sources on turkey performance and Ca and P retention. D. Ledoux¹, J. Sands², M. Hruby*², J. Remus³, and E. Pierson³, ¹University of Missouri, Columbia, MO, USA, ²Danisco Animal Nutrition, Marlborough, Wiltshire, UK, ³Danisco Animal Nutrition, St. Louis, Missouri, USA.

A 21-day turkey study was conducted to evaluate the comparative efficacy of two microbial phytases (3-phytase and 6-phytase) in reduced non-phytate phosphorus (NPP) and calcium (Ca) corn/soy-based diets. A total of 240 Nicholas Large White turkeys were assigned to eight treatments with six replicates each. A positive control (PC) diet was reduced in NPP and Ca by 0.35% and 0.24%, respectively. The 3-phytase and 6-phytase were supplemented to a negative control (NC) at 0, 250, 500 and 750 units per kg of feed. Turkeys fed NC diet had significantly (P<0.05) lower weight gain (WG), feed intake (FI), tibia ash % (TBA%) and toe ash % (TOA%) compared to PC. Feed conversion (FCR) and calcium retention (CaR) were not affected by reduced NPP and Ca. However, phosphorus retention (PR) was significantly increased in NC compared to PC. All levels of 3-phytase and 6-phytase gave comparable WG and FI to PC. CaR and PR were significantly greater under all additions of 3-phytase and 6-phytase compared to PC. CaR and PR were significantly greater with 6-phytase at 500 units/kg and 250 units/kg, respectively, compared to the same inclusion of 3-phytase.

Key Words: microbial phytase, 3-phytase, 6-phytase, turkey, dose

188 The effect of microbial phytase on performance of broilers fed P and Ca reduced diets. R. Stilborn¹, J. Sands², M. Hruby*², J. Remus³, and E. Pierson³, ¹Lakeside Research, Alberta, Canada, ²Danisco Animal Nutrition, Marlborough, Wiltshire, UK, ³Danisco Animal Nutrition, St. Louis, Missouri, USA.

A study was conducted to evaluate the efficacy of 6-phytase in reduced non-phytate phosphorus (NPP) and calcium (Ca) broiler corn/soy-based diets. 1764 Arbor Acres x Peterson male broilers were assigned to seven treatments with six replicate pens each. Corn-based mash diets were fed in two phases from 0 to 21 and 22 to 42 days of age. A positive control treatment (PC) was reduced in NPP and Ca by 0.1% and the phytase was supplemented at 0, 250, 500, 1000 and 2000 units per kg of feed. Broilers in a negative treatment (NC) had significantly (P<0.01) lower weight gain (WG) and feed intake (FI) during both feeding phases and bone ash was lower at 42 days of age compared to PC. Phytase inclusions ≥250 units/kg gave a comparable performance (WG, FI, FCR) with PC during 0 to 21 days of age. Additionally, inclusions of phytase at 1000 and 2000 units/kg improved further (P<0.054 and P<0.010)) WG compared to PC. In the second phase, WG and FI in all phytase-supplemented treatments were not significantly different from PC. However, there was

a tendency ($P < 0.106$) for better WG with 2000 units/kg phytase during 22 to 42 days compared to PC.

Key Words: microbial phytase, 6-phytase, broiler, age, dose

189 Guar Meal in Laying Hen Diets. C. Zhang^{*1}, J. Lee¹, N. Surbakti¹, A.L. Cartwright¹, and C.A. Bailey¹, ¹Texas A&M University System.

Hy-line W-36 laying hens (76 weeks old) were divided into 20 blocks of 5, so that egg production for each block averaged 78%. Blocks were randomly divided into 4 treatment groups. For the first period (28 weeks) of experiment, each group was randomly assigned to a 0, 5, 10 or 15% guar meal diet. For the second period, the hens fed 15% guar meal diet were switched to control diet (0% guar meal). Egg production was recorded daily, egg interior quality (egg weight, Haugh unit, yolk color) and eggshell quality (breaking force, shell thickness) were measured weekly. At the end of the experiment, 2 hens in each block were killed and liver, pancreas, gall bladder, spleen, and abdominal fat pad weighted and scored for fatty liver immediately. Results showed that, 5% guar meal in laying hen diets didn't adversely affect laying hen performance. Hens fed 10% guar meal had depressed egg production and feed consumption, and increased body weight loss. Egg production dropped below 12% after only 2 weeks of feeding the 15% guar meal diet suggesting its potential as a molt-inducing diet. Egg production returned to 67% after hens were switched to the control diet during the second period. Egg weight, egg interior quality and shell quality were not affected by guar meal treatment. Guar meal in diets increased liver weight and liver weight/body weight ratio, but didn't affect weight of pancreas, spleen, gall bladder, abdominal fat pad, or fatty liver score.

Key Words: guar meal, laying hens, molt, toxicity

190 Mineral composition of distiller's dried grains with solubles. A. B. Batal^{*} and N. M. Dale, University of Georgia.

Increased emphasis on ethanol production in the United States has and will continue to lead to significant increases in the amount of distiller's dried grains with solubles (DDGS) available to the feed industry. One of the primary concerns with DDGS by poultry nutritionists is the nutrient content variability among ethanol plants. For proper feed formulation accurate information on the nutrient content of DDGS is essential. Twelve samples of commercially prepared DDGS were obtained from the north central United States and analyzed for mineral composition, with primary emphasis placed on the sodium content. The levels of sodium listed by the NRC (1994) for distiller's dried grains (DDG) (0.09%), distiller's dried grains with solubles (0.48%), and dehydrated distiller's solubles (DDS) (0.26%) appear to be inconsistent and illogical. Analysis indicated an enormous range in sodium content of the 12 samples, ranging from 0.09 to 0.57%. The average value is 0.23%; however it is emphasized that use of this value in feed formulation is to be discouraged. The source of the extraordinary variation noted in the sodium content of DDGS samples is not immediately clear. A simple consideration of the sodium content of corn grain and the multiplier effect (3x) resulting from starch fermentation adequately accounts for sodium values of DDGS in the 0.09 to 0.12% range, but not the higher levels which were observed in some samples. The average composition of many of the other minerals (except Ca) agreed with projected values based on a 3 fold increase of the levels found in yellow corn grain, NRC (1994). Nutritionists need to properly characterize the mineral content of DDGS from respective suppliers prior to incorporating into balance rations.

Key Words: Distiller's dried grains with solubles, mineral composition, sodium, feed ingredients, DDGS

191 Corn-soy enzyme product addition to roaster diets with differing metabolizable energy and energy:amino acid ratios. E. T. Moran Jr.¹, E. Pierson^{*2}, M. Hruby³, J. Halley⁴, C. Wyatt⁵, and J. Remus², ¹Auburn University, Alabama, USA, ²Danisco Animal Nutrition, St. Louis, Missouri, USA, ³Danisco Animal Nutrition, Marlborough, Wiltshire, UK, ⁴Wayne Farms LLC, Oakwood, Georgia, USA, ⁵Cargill Feed Applications, Minneapolis, Minnesota, USA.

A 55-day trial was conducted to evaluate the effect of adding amylase, protease and xylanase (Avizyme[®] 1500-Avizyme) on heavy male broiler performance (8 reps/treatment-4 Trt, 50 birds/pen). RossxHI-Y broilers

were fed corn/soy/animal protein diets (industry specifications) with reduced metabolizable energy (RedME) or increased limiting amino acids (IncAA) to support the expected enzyme energy uplift. In the RedME diets, ME was decreased by 3, 4, 5 and 6% in the starter (0-17 days), grower (17-38 days), finisher (38-49 days) and withdrawal (49-55 days) diets, respectively. Relative AAs were increased by 2 to 10% in the IncAA phase diets. Trts were fed with either 0 or 0.1% Avizyme. Dietary changes were achieved through different inclusions of fat, rice mill feed, L-lysine-HCl and DL-methionine. At 55 days, gain was significantly improved only in the Avizyme-IncAA Trt. Cumulative FCR (55 day) was numerically improved in RedME and IncAA diets with Avizyme. A significant improvement in FCR was achieved during the 18 to 38-day period in the Avizyme diets. No significant effect on mortality was observed across Trts. Avizyme reduced fat pad (% carcass) in both RedME and IncAA diets. The advantage of Avizyme supplementation progressed from starter to withdrawal diets, thus, the relative improvement from Avizyme supplementation may occur after the first weeks of use. The absence of enzyme response in RedME diets could be related to the dearth of digestible substrate due to relatively high levels of rice mill feed used to reduce the energy levels. Trial results suggest an opportunity exists to improve broiler performance through optimizing ME:AA balance due to the improved performance observed in the IncAA diets containing 0.1% Avizyme compared to either the IncAA control or RedME diets.

Key Words: Avizyme[®] 1500, corn-soy enzymes, broiler, down specification, ME:AA

192 High Level Dietary Lutein Inclusion for Laying Hens Improves Yolk Color and Increases Lutein Content in Eggs without affecting Production Parameters. C. Troche^{*1}, K. Strahsmeier¹, P. Ruzsler¹, D. Sanders², and C. Novak¹, ¹Department of Animal and Poultry Sciences, Virginia Tech, ²Kemin Industries.

Inclusion of marigold has long been used to increase the xanthophyll content in broiler and layer diets. To determine the feasibility of attaining levels of lutein in eggs as a viable alternative to human dietary supplements, a laying hen study was conducted. In this study a saponified/emulsified marigold extract was supplemented at very high levels followed by analysis of lutein levels in the eggs (at 0, 7, 14 and 28 days of trial) and evaluation of production parameters (in hens at 42 to 50 weeks of age) using a 2 x 2 x 4 treatment design. Two hundred and twenty four commercial laying hens (112 - Hy-Line W98; 112 - Bovans strains), housed at two different densities (54 or 72 in2), were supplied one of four dietary treatments. The treatments consisted of a control diet and control diet with increasing lutein dosages (40, 80, or 130 g/ton diet) from a saponified/emulsified extract of marigold oleoresin. Lutein dosages in the diet significantly increased yolk color and lutein concentration in egg yolk at 7, 14, 21 and 28 days of the trial. All-trans lutein levels at 28 days ranged from 0.8, 1.3 and 1.8 mg per yolk with the 40, 80, or 130 g/ton diets, respectively. Overall, egg production, feed consumption, feed efficiency (g feed/g egg mass) and hen weight were not affected significantly by marigold extract or housing density. Strain differences ($P < 0.05$), however, were present with Bovans having a greater average egg production (B ? 91.1 vs. H ? 82.8%), improved feed efficiency (B ? 1.99 vs. H ? 2.08) and smaller body size (B ? 1.756 vs. H ? 1.966). Bovans hens had a significantly lighter egg compared to Hy-Line W98, but significantly greater percent yolk and shell. The use of a saponified/emulsified marigold extract is a viable way to attain lutein at high levels in eggs without affecting production parameters, thereby providing an alternative source of lutein through eggs.

Key Words: Strain, Egg size, Egg yolk, Marigold extract, Lutein

193 Selenium tissue levels in turkeys supplemented with organic selenium. M.D. Sims¹, D.M. Hooge², and R.E. Weems³, ¹Virginia Scientific Research, Inc., Harrisonburg, VA, ²Hooge Consulting Service, Inc., Eagle Mountain, UT, ³Alltech, Inc., Nicholasville, KY.

Selenium (Se) in turkey rations has traditionally been supplemented from inorganic sources. The feasibility of supplementing rations with an organic source of Se (Sel-Plex, Alltech, Inc., Nicholasville, KY) was evaluated in a recent turkey floor pen trial. Organic Se has proven to be more bio-available than inorganic sources in other animals. Whether or not turkey liver Se levels were within a 1.06 ppm threshold level allowed by the Food and Drug Administration # Center for Veterinary

Medicine (FDA-CVM) was addressed. This study compared two selenium levels, ambient (0.2 ppm) and high (0.5 ppm). One hundred (100) healthy poult of each sex were randomly selected, wingbanded, weighed and placed into one of 4 single sex pens. Each single sex pen was randomly assigned to one of the two Se treatments. A five-phase feeding program was designed to meet NRC requirements and to reflect commercial practices. Hens were grown to 98d, toms to 126d. Final body weights and (feed efficiency) were: 15.56 kg (2.56) and 15.72 (2.68) kg for hens on basal and supplemented rations respectively; 28.74 kg (2.95) and 28.93 kg (2.81) for toms on basal and supplemented rations respectively. After collection of final weights, liver samples were harvested and assayed from ten pre-selected birds from each pen. The Se levels were 0.618 ppm, 0.973 ppm for hens; 0.693 ppm and 1.162 ppm for toms on basal and supplemented rations, respectively. Basal-fed birds and high Se hens were significantly below the critical value, while toms fed the high Se diets exceeded the value by 0.102 ppm ($p \leq 0.0016$). When pooled, the liver Se values for the high Se diet did not differ significantly from the 1.06 ppm critical value ($p \leq 0.7756$). This controlled floor pen study demonstrated that the liver Se levels from turkeys supplemented with organic Se were within the threshold allowed by FDA-CVM. In addition, data suggests that Se from organic sources is more bio-available than Se from inorganic sources in turkeys.

Key Words: selenium, turkeys, organic, liver

194 The relationship between Quantitative Ultrasonometry, bone mechanical properties and production parameters in Single Combed White Leghorn Hens. M.A. Martinez-Cummer* and S. Leeson, *University of Guelph, Guelph, ON, Canada.*

An experiment was conducted to evaluate how humeral speed of sound (S.O.S.) measured with a Sunlight Omnisense 7000s bone sonometer, correlates with bone mechanical properties, egg production and shell quality. A 3X2 factorial trial was conducted using three levels of crude protein (14, 16, and 18% CP) and 2 levels of calcium (3.2 and 4% Ca) fed from 18 to 34 weeks of age. At 18 weeks of age, a total of 192 hens were transferred from a grower to a layer barn, provided with 14 hours of light and 10 hours of darkness and 32 hens per treatment were allocated randomly in each of 8 replicates. In addition, the hens were kept at 26 C, provided with free access to water and fed ad libitum. Hens were randomly selected into two groups; birds in group A were had their humerae scanned in vivo and were subsequently euthanised to harvest and obtain ex vivo ultrasound readings at 34, 50 and 66 weeks of age. The right humerus, tibia and femur were removed, defleshed, surrounded by PBS laden paper towels, double wrapped in plastic and stored at -12 C. For mechanical testing, fracture loads using a three point bending test were measured. Furthermore, all specimens tested for breaking force were fat extracted and dried in a forced-air oven at 90 C until a consistent weight was obtained. The bone ash weight of the bone fragments were obtained after ashing at 600 C for 24 hours. Birds from group B were scanned live at 34, 50, 58, and 66 weeks of age and euthanised at the completion of the trial. Data suggest that the Sunlight Omnisense bone sonometer is able to detect differences in skeletal status using speed of sound as a response criteria ($P < .001$). In addition early nutrition had a significant effect on overall productivity and body weight gain ($P < .0001$).

Key Words: Crude protein, Calcium, Ultrasonometry, Egg shell

195 Effects of feeding grains naturally-contaminated with *Fusarium* mycotoxins on performance and metabolism in laying hens. Shankar Chowdhury*¹ and Trevor Smith¹, ¹*University of Guelph.*

An experiment was conducted to evaluate the effect of feeding grains naturally-contaminated with combination of *Fusarium* mycotoxins to laying hens with respect to performance, hematology and plasma chemistry. One hundred and forty-four, 45-week-old laying hens were fed diets containing control, contaminated grains, or contaminated grains +0.2% polymeric glucomannan mycotoxin adsorbant for a period of 12 wks. The feeding of contaminated grains decreased feed consumption after wk 4, and increased feed consumption after the 8th and 12th wks. Egg production, egg mass, egg weight, yolk weight and eggshell weights were also decreased after the feeding of contaminated diets for varying periods. The feeding of contaminated diets decreased plasma concentrations of calcium, albumin, glucose, and cholesterol but uric acid concentrations were increased after wk 4. The ratio of albumin:globulin, and the

activities of amylase, also decreased after the 4th wk. The feeding of contaminated diets decreased albumin concentrations, albumin:globulin ratio, and hematocrit values but increased uric acid concentrations after the 8th and 12th wks. Kidney weights as a percentage of body weight increased significantly when hens were fed contaminated grains. The feeding of contaminated grains plus glucomannan prevented the elevation of uric acid concentrations, and the reduction of yolk weight, and glucose concentrations after wk 12. It was concluded that the high concentrations of uric acid throughout the experiment and the increased kidney weight after wk 12 indicated an alteration of kidney function. The reduction of laying performance and the alteration of plasma chemistry may have been due to the chronic mycotoxicosis. It was concluded that laying hens were susceptible to chronic exposure to combination of *Fusarium* mycotoxins.

Key Words: *Fusarium* mycotoxins, layer, contaminated grains

196 Effect of dietary putrescine (1, 4-diaminobutane) on growth and intestinal tract development in turkey poults. S. R Girdhar and T. K Smith, *University of Guelph.*

Dietary putrescine (1, 4-diaminobutane) can increase growth rates of neonatal animals under nutritional stress including calves and piglets. Turkey poults often experience a high rate of mortality and this may be due to poor initial feeding behavior and inadequate development of the intestinal tract. Three experiments were conducted, therefore, to determine the potential for dietary putrescine to promote whole body growth and development of the intestinal tract in poults. In the first experiment, a total of 160 day-old poults were fed a corn and soybean meal-based starter diet supplemented with 0.0 (control), 0.2, 0.4 and 0.6% purified putrescine (10 birds per pen, 4 pens per diet) for 21 days. The feeding of 0.2% putrescine increased growth rate ($P < 0.05$) compared to controls. In the second experiment, with a similar experimental design, 320 birds were utilized but there was no significant effect of diet on growth rates. Twenty birds fed each of the experimental diets were subsequently fed practical grower and finisher diets until 12 weeks of age. It was observed that birds fed 0.4% supplemental putrescine in the starter period were heavier than controls at 12 weeks of age ($P < 0.05$). There was no effect of starter diets on the relative weights of duodenum, jejunum + ileum or liver at the end of the three week period. In the third experiment 400 day-old poults (200 males & 200 females) were fed diets supplemented with 0.0 (control), 0.1, 0.2, 0.3, and 0.4% purified putrescine for 3 weeks and from week 4 birds were grown out on non supplemented diets up to 14 weeks of age. There was no significant effect of diet on growth, however a curvilinear response (highest for 0.2%) was observed in the relative weight patterns of duodenum, jejunum+ ileum and breast weights. It was concluded that supplementation of poult starter diets with 0.2-0.4% putrescine may result in increased growth of turkeys in the growing and finishing period

Key Words: Putrescine, Gastrointestinal tract, Turkey

197 Development of the small intestine in broiler chicks fed dietary 1,4-diaminobutane (putrescine). F.A. Santoyo and T.K. Smith, *University of Guelph.*

Putrescine (1,4-diaminobutane, DAB), spermidine and spermine are the mammalian polyamines. Dietary supplements of DAB have been shown to promote intestinal tract development and growth of poultry and other species. An experiment was conducted to compare morphology of the small intestine with a total of 360 five day-old broiler chicks fed corn and soybean meal-based diets containing 0, 0.1, 0.2, and 0.3% supplemental dietary DAB for six weeks. Growth rate and feed consumption were determined weekly. After 3 weeks and at the completion of the experiment, 10 birds fed each diet were killed and intestinal sections, liver, pancreas, spleen and breast and thigh muscles were excised. Intestinal concentrations of soluble protein, DNA, and biogenic amines were determined and morphometric indices were obtained using computer-aided light microscope image analysis of intestinal samples. A linear response to dietary DAB was seen in feed efficiency after weeks two, three and four. Villus height, crypt depth and thickness of muscularis was observed in duodenum, jejunum and ileum and villus width and submucosa was determined in duodenum and ileum. A quadratic response was seen in crypt depth and muscularis in duodenum, jejunum and ileum. A similar response in submucosa was seen in jejunum and ileum. A quadratic response with DAB was seen in villus width only in ileum. It was concluded

that dietary DAB could influence the morphology of the small intestine in broiler chicks.

Key Words: Broiler, Morphometrics, Putrescine

198 High levels of metabolizable energy in broiler breeder diets and their effect in productive parameters. J.A. Quintana¹, E. Avila¹, C. Lopez¹, J.G. Herrera², E. Morales³, and M.J. Gonzalez¹, ¹Depto. de Aves, FMVZ, UNAM, ²Ganaderia, Colegio de Posgraduados, ³CEVAMEX, INIFAP, Mexico, ⁴Depto. Zootecnia, UACH, Mexico.

It has been suggested that high levels of energy in the food increase the hatchability and the productive parameters in broiler breeders. An experiment was conducted to evaluate four levels of metabolizable energy (ME, Kcal/kg). Six hundred Ross X Arbor Acres breeders were allocated in 12 rooms of 15 m² (fifty hens and five cockerels/ room) with similar weight and conformation during twelve weeks, from twenty five weeks old (sexual maturity) to thirty six weeks old. A basal diet sorghum - soybean meal with 16 % of crude protein and 2850 Kcal/kg ME treatment 1 (T1) with 27 kg/ton of soybean oil (SO) was increasing 10 kg/ton of SO to obtain 2940 (T2), 3030 (T3) and 3120 (T4) kcal/kg ME. The results at 36 weeks age indicated no difference (P>0.05) among treatments in live weight (3.8, 3.7, 3.8, 3.8 kg), body gain weight (23.6, 20.6, 22.5, 24.5 %), egg production (59.4, 57.8, 56.3, 57.9 %) egg weight (59.8, 59.8, 59.5, 59.6 g) born weight chick (42.5, 43.3, 42.2, 43 g) male born (50.5, 50.8, 52.3, 49.2 %), double yolk eggs/hen (0.42, 0.38, 0.39, 0.51), infertility (16.6, 10.9, 16.6, 8.2 %), however the lost weight at nineteen days of incubation chick increased (P<0.05) gradually with more SO in the diet (5.8c, 9.9 b , 12.4 a , 10.7 ab). These results indicate that high energy levels did not affect the productive parameters.

Key Words: Metabolizable energy, broiler breeder, soybean oil

199 High levels of metabolizable energy in broiler breeder diets and their effect in total lipids, cholesterol in serum and omega 3-6 fatty acids in eggs. J. Quintana*¹, E. Avila¹, C. Lopez¹, J.G. Herrera², S. Carrillo³, R.M. Castillo³, F. Perez³, J.E. Morales⁴, and M.J. Gonzalez⁵, ¹Depto. de Aves, FMVZ, UNAM, ²Ganaderia, Colegio de Postgraduados, ³Depto. de Nutricion Animal, INCMN Salvador Zubirn, ⁴CEVAMEX, INIFAP, Mexico, ⁵Depto Zootecnia, UACH, Mexico.

It has been suggested that high levels of energy in the food increase the performance in broiler breeders, however is necessary to include more oil and to evaluate egg lipids. An experiment was conducted to evaluate four levels of metabolizable energy (ME, Kcal/kg). Six hundred Ross X Arbor Acres breeders were allocated in 12 rooms of 15 m² (fifty hens and five cockerels/room) with similar weight and conformation during twelve weeks, from twenty five weeks old (sexual maturity) to thirty six weeks old. A basal diet sorghum - soybean meal with 16 % of crude protein and 2850 Kcal/kg ME treatment 1 (T1) with 27 kg/ton of soybean oil (SO) was increasing 10 kg/ton of SO to obtain 2940 (T2), 3030 (T3) and 3120 (T4) kcal/kg ME. Total lipids and cholesterol were measured by optical density on the serum and fatty acids omega 3-6 (ω 3-6) by gas chromatography in eggs. The results at 36 weeks age indicated no difference (P>0.05) in the serum: total lipids (527, 611, 805, 720 mg/dl) and cholesterol (110, 97, 119, 99 mg/dl) and in the egg: total lipids (7.2, 7, 6.7, 7.2 g/100g), linoleic (ω -6)18:2 (810, 798, 992,1024 mg), arachidonic (ω -6) 20:4 (99, 100, 100, 101.4 mg), eicoxapentaenoic (ω -3) 20:5 (1.6, 1.3, 1.4, 1.3 mg), docosapentaenoic (ω -3) 20:5 (5, 5.5, 5.4, 4.9 mg) and docosahexaenoic (ω -3) 22:6 (36.9, 32.3, 42.1, 38.1 mg) were not different (P>0.05) however linolenic (ω -3) 18:3 increased with SO (P<0.10) (37.4b, 37 b, 47.8 ab, 54.4 b mg). These results indicate that high energy levels the ω -3 linolenic in eggs increased as well as SO in the diet.

Key Words: Metabolizable energy, broiler breeder, egg lipids, cholesterol, omega 3-6 fatty acids

200 The effect of source and level of dietary copper on broiler performance. A.J. Pescatore*¹, M.D Ford¹, A.H. Cantor¹, and J.L. Pierce², ¹University of Kentucky, ²Alltech Inc..

The effect of source and level of dietary copper was evaluated using ten dietary treatments. Six birds were allocated to each of nine replicates per dietary treatment based on weight at seven days of age. The birds were housed in cages in an environmental control building. Diets were fed from 7 days to 21 days. The sources of copper were copper sulfate,

chelated copper (Bioplex, Alltech, Nicholasville, KY) and a novel copper proteinate. Copper sources were added to a basal diet (Trt 1): copper sulfate at the levels of 62.5 ppm (Trt 2), 125 ppm (Trt 3), 250 ppm(Trt 4); chelated copper at 25 ppm (Trt 5), 50ppm (Trt 6), 100ppm (Trt7);cooper proteinate at 25 ppm (Trt 8), 50 ppm (Trt 9) and 100 ppm (Trt 10). The copper content of the basal diet was 20 ppm. Feed consumption was significantly depressed by the high level of copper sulfate (Trt 4), resulting in an 18 g reduction in body weight. Body weight and feed consumption was not significantly different for the other treatments. Source or level of cooper did not effect feed conversion. Liver copper levels were elevated with the high level of copper sulfate. Excreta were collected for 14 days from two replicates for each treatment and analyzed for copper. There was a linear increase in copper content of the excreta as dietary copper level increased. Source of copper did not effect the concentration of copper in the excreta.

Key Words: Copper, Copper Sulfate, Bioplex

201 Effects of a European Style Electrical Stimulator for Poultry Processing on Shear Values and Cook Yield of Broiler Breasts Processed under Domestic Conditions. J. A. Dickens*¹, J. A. Cason¹, C. E. Lyon¹, L. L. Young¹, D. P. Smith¹, B. A. Lovingood², and J. M. Walker², ¹Russell Research Center, ²Stork Gamco Inc..

Interest is growing in the U. S. and Europe on the application of pulsed electric current (PEC) to improve poultry meat quality and yield, but European processing and stimulation procedures differ from those used in the U. S. In Europe, carcasses are stimulated after defeathering, contact points being the feet and the sternum and carcasses are air chilled. In the U. S., PEC is applied during bleeding, contact from head to foot and carcasses are water chilled. This study was conducted to test European-style stimulation under domestic processing conditions. Carcasses (120; 3 reps of 10 per treatment) were stunned at 12 V pulsed DC for 10 s, killed by mechanically severing the carotid artery and jugular vein, and bled for 90 s. Carcasses passed through a three stage scald at 56 C for 2 min and were defeathered in a commercial picker for 15 s. Carcasses were then either held for 90 s and transferred to the eviscerating line, or moved to the stimulator for 90 s then to the evisceration line. Eviscerated carcasses were either chilled in a simulated commercial immersion paddle chiller for 1 h and then held for 2 h at 4C prior to deboning or immersion chilled for 3 h then deboned. Fillets were weighed, vacuum bagged, and held at 2 C. The next morning, fillets were heated in a steam jacketed kettle at 80 C to a minimum internal temperature of 78 C. After tempering the fillets were weighed and two 1.9 cm wide strips were cut parallel to the muscle fiber and trimmed to 1.9 cm high. Each strip was sheared twice using a Warner-Bratzler dynamic shear apparatus and peak load was recorded in kg. Mean fillet shear values and cook yield for the 1 h chill followed by 2 h at 4 C were 7.0 kg and 80.6% respectively for the non-stimulated and 3.4 kg and 82% respectively for the stimulated groups. Values for the 3 h chill were 8.5 kg and 78.7% respectively for the non-stimulated and 3.6 kg and 80.7% respectively for the stimulated fillets. Chilling and PEC both had significant effects on cooked yields. The PEC had a significant effect on shear, but the magnitude of the effect varied between replications. PEC under this protocol could increase profits for the processor and a more tender product for the consumer.

Key Words: Broilers, Cook Yield, Electrical Stimulation, Shear

202 Effects of replacement finisher feed and length of feed withdrawal on broiler carcass yield and bacteria recovery. J. K. Northcutt*¹, R. J. Buhr¹, M. E. Berrang¹, and D. L. Fletcher², ¹Russell Research Center, Poultry Processing and Meat Quality Research Unit, USDA-ARS, Athens, GA, ²The University of Georgia, Athens, GA .

The replacement finisher feed (RF) used in the present study is a commercial formulation of a D-glucose polymer (maltodextrin) with added salts and vitamins. This study was conducted to determine the effects of RF on carcass yield and recovery of bacteria from carcasses. Commercial male broilers (41 d of age) were given either RF or traditional starter feed (control) for 8 h, followed by feed withdrawal for 0, 4, 8, or 12 h before processing. During processing, whole carcass rinses (WCR) of pre-eviscerated (missing feathers, feet and heads) and eviscerated carcasses were analyzed for recovery of bacteria. Body weight of broilers at the initiation of feed withdrawal (catch weight) did not differ significantly between types of feed (2.53 to 2.65 kg). Similarly, body weight at

slaughter (dock weight) did not differ significantly between types of feed or with length (0 to 12 h) of feed withdrawal (2.51 to 2.57 kg). At every feed withdrawal time period except 0 h, broilers fed control feed had approximately 0.1% more live shrink per h of feed withdrawal than those broilers fed RF, and this difference was significant for broilers held without feed for 8 and 12 h. Type of feed or length of feed withdrawal did not affect coliform and *E. coli* counts recovered from WCR of pre-eviscerated or eviscerated carcasses. Carcasses representing birds held without feed for 4 h had significantly less *Campylobacter* (\log_{10} 2.2 cfu/mL of rinse) when fed control feed as compared to all other groups (\log_{10} 3.6 cfu/mL of rinse). These data demonstrate that feeding RF to broilers 8 h before initiation of feed withdrawal may reduce live shrink by 0.1% per h without affecting carcass coliform, *E. coli* or *Campylobacter* counts.

Key Words: broilers, replacement feed, live shrink, carcass bacteria, feed withdrawal

203 A survey of common practices in commercial broiler processing facilities and their effects on water use. J. K. Northcutt* and D. R. Jones, *Russell Research Center, Poultry Processing and Meat Quality Research Unit, USDA-ARS, Athens, GA.*

A survey of broiler processing facilities was conducted to determine the relationship between common industry practices and water use. One hundred-thirty written surveys were sent to broiler processing facilities throughout the U.S. with 35% (45/130) responding. The respondents represented a bell distribution of plant size. Data from the surveys were analyzed for significance using Chi-square. Sixty-nine percent of the respondents are on city water ($P < 0.05$), with 60% of the facilities discharging to city sewers. Ninety-six percent of the respondents operate 5 d each week ($P < 0.0001$) and 70% of the respondents run 10 shifts each week ($P < 0.0001$). Thirty-eight percent of the respondents ($P < 0.05$) are located in a southeastern state; however, there is no relationship between location (area of U.S.) and water use or location and water recycling. Average water usage prior to implementation of the Pathogen Reduction, Hazard Analysis and Critical Control Point (HACCP) System final ruling was 21 L/bird compared to current, post-HACCP usage of 27 L/bird. Sixty-seven percent of the respondents reported having low water usage (<15.1 L/bird) before HACCP ($P < 0.0001$). Water is recycled in 38% of the facilities that responded to the survey, but only 27% of the facilities have truck and/or transport coop washing stations ($P < 0.0001$). Of the facilities that recycle water, a significant relationship ($P < 0.05$) was observed between size of facility and amount of water recycled. Large facilities recycle more water than medium or small facilities. Data from this survey may be used to assist companies interested in establishing water conservation programs.

Key Words: broiler processing, water, HACCP, recycling

204 A survey of common practices in shell egg processing facilities and their effects on water use. D. R. Jones*¹ and J. K. Northcutt¹, ¹*Russell Research Center, Poultry Processing and Meat Quality Research Unit, USDA-ARS, Athens, GA.*

Shell egg processing facilities in the US were surveyed for common production practices and water use. Results were compiled and analyzed for frequency and significance via Chi-square. Of the respondents, 62.5% ($P < 0.001$) utilized wells as their primary source of water. Only 22% ($P < 0.01$) of the facilities discharged water to city sewers. Over half of the facilities process 7 days each week ($P < 0.01$) with 8 to 9 hour shifts ($P < 0.05$). There was an even distribution of in-line, off-line and mixed operations represented. Two-thirds of the operations were dual-tank systems with over half being plumbed jointly. The dual tank systems that were plumbed jointly used the least water per case of eggs processed ($P < 0.05$). Over 90% ($P < 0.0001$) of the operations performed daily sanitation. Most facilities ($P < 0.0001$) do not attempt to recycle water from their process. The age of the processing line, number of days processing each week, length of shift, size of facility and type of operation did not have a significant effect on water use.

Key Words: Water, Shell egg, Processing

205 Moisture retention by immersion chilled cut-up broilers. L. L. Young* and D. P. Smith, *Richard B. Russell Agricultural Research Center, Athens, GA USA.*

The objective of this study was to assess chiller water absorption and retention characteristics of broiler chickens through simulated processing,

cutting and storage. Sixty-four broiler chickens were manually slaughtered using conventional techniques. Half the carcasses (controls) were chilled in air (4 C, 2.2 m³ per min), and half (treated) by tumbling in water (4 rpm, 1 C, 30 min). All carcasses were stored overnight and then manually cut into fore- and hindquarters. The quarters from each carcass were subsequently placed in a plastic bag and stored overnight under refrigeration (4 C). Moisture retention of each carcass was evaluated by weight after initial chilling, after overnight storage, immediately after quartering and after overnight refrigerated storage of the quarters. Moisture loss during post-cutting storage by each quarter was assessed by similarly.

Weights of the control carcasses were unaffected by chilling, whereas treated carcasses absorbed 11.7% chiller water. Overnight storage of the intact carcasses resulted in a 0.7% (± 0.01) weight loss by control carcasses and a 7.0% (± 0.41) loss by treated carcasses. Weights of control carcasses did not change significantly ($P \leq 0.05$) during intact storage or cutting averaging 0.6%, (± 0.01), but treated carcasses lost 40.3% (± 0.23) of the absorbed moisture during overnight storage and 8.4% (± 0.29) during cutting. Quarters from control carcasses lost 0.01% (± 0.002) as purge, but treated carcasses lost 18.0% (± 0.26) of the absorbed moisture during overnight post-cutting storage. Forequarter loss during storage was 13.2% (± 0.16) compared to 19.4% (± 0.19) for the hindquarters. Overall added water content of the quarters from water-chilled carcasses was 3.9% (± 0.26).

Under the conditions of this study, the water-chilled parts clearly retained part of the chiller water. This information may be useful as baseline data for developing process monitoring and control methodologies.

Key Words: Moisture Absorption, Chicken, Chill

206 Eggs for Solid Fat Products and Fat Encapsulation. John Lee*, *Rigel Technology Corporation, Shawnee, KS.*

Whole eggs, yolk and albumen are excellent sources of nutrients and, in addition, have valuable functional properties. Liquid whole eggs, which has a solids content about 26 %, is composed of about 36 % yolk and 64 % albumen. Egg products are commercially marketed in three forms: (1) refrigerated, (2) frozen, and (3) spray dried. The egg products from spray drying process are most convenient to be used and handled.

A new process has been developed (US patent pending) to make high fat or high energy egg products with egg albumen or whole egg ingredient and the additional fat ingredients, which are from plant and animal sources. The high fat egg products have high fat or energy and low protein levels such as 62 % fat, 30 % protein, and 6 % moisture. Even at the high fat level, the high fat or high energy products, which can be dried by a spray dryer or other dryers, have no oily physical characteristics. The cholesterol level can be reduced significantly such as 35 % lower when the plant fat ingredient with very low cholesterol level is used for making the encapsulated fat and egg products. Besides as general fat and protein ingredients, which can be pelleted easily, the high fat or high energy egg products can be used specially for the applications such as baby formula, milk replacer, fish diet with adjustable bulk density, and dry fat-protein ingredients for the nutritional needs during young and lactation periods.

Key Words: Eggs, Solid Fat, Fat and Protein, Process, Fat Encapsulation

207 Broiler skin composition and fat characteristics during a 12-month sampling at retail. Judith C. Snow, Rhoda K. Friesen, Woodie P. Williams, Jr., and James C. Acton*, *Clemson University, Clemson, SC.*

Pooled skins from two whole broilers collected biweekly over 12 consecutive months (total n=26) from a local supermarket were analyzed for percents moisture, fat, protein and ash and skin TBA value and fat melting point and viscosity. Fatty acid composition of skin fat in the first 6 months of samples was also determined. Compositional ranges for moisture, fat, protein and ash were 42.9-57.6%, 31.1-51.1%, 7.1-15.6%, and 0.30-0.66%, respectively. TBA values of whole skins averaged 1.25 and the extracted fat had averages for melting point and relative viscosity of 29.8 C and 17.1 centipoise (at 40 C), respectively. Fatty acid analysis yielded 32.9% as saturated (14:0, 16:0, and 18:0) and 67.1% as unsaturated (16:1, 18:1, and 18:2). Variation of fat, protein and ash contents was due to shifts in moisture content, likely related to water retention on chilling. Fat composition and physical fat characteristics remained in a narrow range of values.

Key Words: Broiler, Skin, Composition, Fat Characteristics

208 Tracking *Pseudomonas* spp. in Commercial Chilling and Refrigerated Storage of Poultry Carcasses. A. Hinton, Jr.*¹, J. A. Cason, and K. D. Ingram, *Russell Research Center.*

Four trials were conducted to examine the effect of immersion chilling and refrigerated storage on the population of *Pseudomonas* bacteria in the native microflora of commercially processed broiler carcasses. The whole carcass rinse procedure was used to recover bacteria from fresh prechilled and chilled carcasses and from chilled carcasses that were stored at 4°C for 7, 10, or 14 days. Bacteria in the carcass rinsates were enumerated on Pseudomonas Agar that was incubated for 2 days at room temperature after inoculation with serial dilutions of the rinsates. Bacterial isolates recovered from the carcass rinsates were identified with the MIDI Sherlock Microbial Identification System, and dendrograms of the fatty acid profiles of the *Pseudomonas* bacteria were prepared to determine the degree of relatedness between the isolates. Findings indicated that immersion chilling did not significantly ($P \leq 0.05$) reduce the number of bacteria recovered from the carcasses in 3 of 4 trials. Furthermore, there was a significant ($P \leq 0.05$) increase in the number of bacteria recovered from the carcasses after refrigeration for 7, 10, or 14 days. Dendrograms of the fatty acid profiles of the isolates indicated that the same *Pseudomonas* strain may be isolated from chilled and refrigerated carcasses processed on the same day at the same facility and from carcasses processed on different days at the same facility. *Pseudomonas* species isolated from the carcasses included *Pseudomonas chlororaphis*, *Pseudomonas fluorescens*, *Pseudomonas putida*, and *Pseudomonas syringae*. Experimental results indicated that the some pseudomonads may survive processing and contaminate carcasses as the broilers are passed through the chill tank. These findings may be useful in designing methods to control the spread of spoilage bacteria during poultry processing and extending the shelf life of refrigerated fresh poultry.

Key Words: Pseudomonas, Broilers, Chilling, Poultry Processing, Cross Contamination

209 The effects of diet and feed withdrawal times on the sensory descriptive profile and shear values of broiler breast meat. B. G. Lyon*¹, C. E. Lyon, D. P. Smith, and E. M. Savage, *USDA-ARS, Russell Research Center, Athens, GA.*

The effects of diet and feed withdrawal times on the sensory and physical characteristics of broiler breast meat were determined. Feeds formulated with three dietary carbohydrate sources (corn, milo, wheat) were fed to broilers starting at 28 d of age. Birds (n=192) were processed between 42 and 52 d of age. Feed was withdrawn for either 0 or 8 h prior to processing in the pilot plant under simulated commercial conditions. *Pectoralis major* muscles were removed 4 h post-mortem and frozen until evaluations. Breasts were cooked in heat-seal bags to an internal temperature of 80°C and then cut into sections for sensory and instrumental evaluation. Sensory profiles of 18 flavor and texture attributes were determined by a trained 8-member panel. W-B shear values were also determined from a 1.9-cm wide strip. Feed withdrawal had no effect on the flavor profile whereas diet had a significant effect on two flavor attributes of the breast meat. Breast meat from birds fed the corn diet was significantly ($P \leq 0.05$) more brothy and sweeter than meat from birds fed either wheat or milo. The sensory texture profile was significantly ($P \leq 0.05$) affected by both feed withdrawal and diet. Meat from birds fed wheat was harder, more cohesive, exhibited larger particle size and was more chewy than meat from birds fed either corn or milo. Two texture attributes were significantly affected by feed withdrawal time. Meat from birds processed with 0 withdrawal time was lower in moisture release and higher in toothpick than meat from birds held 8 h without feed. The range of W-B shear values was small but significant for diet. Meat from birds fed corn required significantly less force to shear (6.0 kg) compared to meat from birds fed either milo (6.7 kg) or wheat (7.1 kg). The results indicate that dietary carbohydrate source can have a measurable impact on the flavor and texture of broiler breast meat.

Key Words: Broiler, Diet, Flavor, Texture, Shears

210 Cutaneous recording of electroencephalograms in electrically stunned broiler chickens. A.F. Collier*¹, D. E. Martyn¹, M. K. Adams¹, M. W. Holt¹, D. S. Williams¹, J. S. Burmeister¹, T. L. Foutz¹, D.V. Bourassa¹, D. L. Fletcher¹, and R. J. Buhr², ¹University of Georgia, ²Russell Research Center USDA-ARS.

Methodology was developed to record electroencephalograms (EEGs) from chickens using skin surface contact electrodes and telemetry transmitter and receiving units prior to and immediately after electrical stunning. Optimal location of the three electrodes was determined using scaleless "featherless" chickens. Broilers required plucking of feathers on the neck caudal to the comb (2 x 3 cm) under mild anesthesia the day prior to recording EEGs. The telemetry transmitter was protected from the stunning voltage with a custom-built circuit designed to reduce high amplitude AC and DC voltages to less than 0.8 V. This configuration permitted recording of EEG signals prior to and within 3.5 s after termination of the applied stunning current. EEGs were recorded during two different electrical stunning protocols with the current applied to a standing chicken (wattle + and vent -). The first stun protocol was at 8 mA, 12 V (500 Hz) pulse DC for 11 s immediately followed by 12 V (60 Hz) AC for 4 s. The broilers were given several minutes to recover and then stunned again using the second stun protocol set at 103 mA (60 Hz AC) for 4 s, which was sufficient to induce cardiac arrest. The EEG recordings of the second stun protocol were evaluated to determine wave characteristics and the duration of poststun brain activity. The poststun EEG recordings depicted a brief period of high amplitude spikes, which progressively diminished in amplitude with time. This high amplitude polyspike wave form has been assumed to be analogous to the insensibility period that occurs during epileptic seizures in humans. This poststun data, in both wave form and duration of brain activity (39 s), appears similar to that described in the literature for chickens (32 s). Use of the cutaneous-telemetry system to record brain EEG activity in chickens following electrical stunning may provide the opportunity to quantitatively optimize stunning voltage, current, and frequency. Optimal stun parameters should minimize the time to death, and diminish skeletal muscle contraction and the carcass defects associated with electrical stunning.

Key Words: EEG, broiler chicken, telemetry, electrical stunning, cutaneous electrodes

211 Electroencephalogram recordings following low and high voltage electrical stun-bleeding in broilers. R. J. Buhr*¹, D. V. Bourassa², T. L. Foutz², and D. L. Fletcher², ¹Russell Research Center USDA-ARS, ²University of Georgia.

Electroencephalograms (EEGs) were recorded for 6-week-old commercial broilers before and after stun-bleeding using the BioRadio 110 and skin surface contact electrodes to evaluate low (USA) and high (EU) electrical stunning protocols. Each broiler was suspended inverted in a processing shackle and three cutaneous snap electrodes adhered to the back of the head and neck where feathers were removed the previous day. The electrode wires were attached to a transmitter that was equipped with an in-line custom-made device to open the circuit during stunning to avoid electrical damage. Both stunning protocols delivered the current through the chicken's head (+) and shanks (-) while suspended in a shackle. The first electric stunning protocol (typical for USA) was 15 mA, 23V DC (550 Hz) for 10 s and was immediately followed by 15 V AC (60 Hz) for 5 s. The second electric stun protocol (typical for EU) used a >105 mA AC (60 Hz) for 5 s and was sufficient to induce cardiac arrest. EEG recordings were taken for more than 30 s prior to stunning to acquire a prestun brain activity pattern for each individual broiler. Stunned broilers were immediately bled with a knife and EEGs recorded for 3 min after stunning. The EEG recordings were evaluated to determine wave characteristics and the duration of spontaneous poststun brain activity. The poststun-bleed EEG recordings depicted a brief period of high amplitude spikes, which progressively decreased in amplitude with time. Typically brain activity and EEG amplitude and frequency lessens within the first 30 s after stun-bleeding. Between 30 and 60 s after stun-bleeding, carcass muscular activity sporadically occurs resulting in recording of large spikes on the recordings. After 60 s, broilers appear to be unconscious and rapidly approaching brain death. Comparison among the EEGs recording for broilers stun-bled by either of these two procedures are very similar, and individual recordings would be difficult to distinguish without examination of the stun duration periods.

Key Words: EEG, broiler chicken, electrical stunning, low and high voltage, bleeding

212 Comparison of cytotoxicity of *Campylobacter jejuni* isolated from humans and from chicken carcasses acquired at processing and retail. C. D. Gilbert* and M. Slavik, *University of Arkansas, Fayetteville, AR.*

Campylobacter jejuni has emerged as an important foodborne pathogen. Causing a variety of symptoms, *C. jejuni* has been linked to poultry related foodborne illnesses. Toxin production has been shown to be a primary factor associated with pathogenicity and is the focus of this experiment. In order to compare the cytotoxicity of *C. jejuni* isolated from chicken carcasses taken at processing and at retail to those isolated from humans, 100 *C. jejuni* were isolated, 25 from pre-chilled chicken carcasses, 25 from post-chilled chicken carcasses, 25 from retail chickens, and 25 from humans exhibiting symptoms of campylobacteriosis. Using HeLa cells as the in vitro model, overall toxicity was determined for each isolate within each category. Cytotoxicity levels varied greatly within each group of isolates taken from chicken carcasses but was less variable in *C. jejuni* isolated from humans. Since most *C. jejuni* isolated from humans possessed a high-medium to high level of cytotoxicity, campylobacteriosis appears to be highly dependent upon cytotoxicity of *C. jejuni* isolates.

Key Words: *Campylobacter jejuni*, Cytotoxicity, Pathogenicity, Toxin

213 Effect of Broiler Breeder Age on Performance and Carcass Yield of Broiler Chickens. JA Dalanezi¹, J Moreira¹, ICL Almeida¹, SE Takahashi¹, RP Oliveira¹, and AA Mendes*¹, ¹*Faculty of Veterinary Medicine and Animal Production.*

The experiment was conducted at Faculty of Veterinary Medicine and Animal Production of Sao Paulo State University, Brazil, to evaluate the effect of breeder age on performance and carcass yield of broiler chickens. Eggs of Ross x Ross breeder flocks with 29, 41, 58, 68 and 98 weeks of age were selected and incubated at identical conditions. After hatching, two thousand and four hundred day-old-chicks were randomly assigned to a factorial 5 x 2 design (5 breeder ages and 2 sexes) and reared until 49 days of age. At 35, 42 and 49 days of age, 20 birds per treatment were selected and processed after 8 hours of feed withdrawal period. Broilers from young and old breeder (29 and 98 weeks) presented worst (p<.05) performances than the adult breeders (41, 58 and 68 weeks). Breeder age didn't affect (p>.05) carcass and leg quarter yields but affected (p<.05) breast meat yield at 49 days of age, with best results for birds originated from breeders with 41 and 58 weeks of age. It was concluded that even though breeder age doesn't affect broiler performance, breast meat yield can be reduced when broilers chickens are obtained from very young or very old breeders.

Key Words: breast meat yield, broiler breeder age, carcass yield, performance

214 Efficacy of salinomycin following use of diclazuril and a coccidiosis vaccine in broilers. H. D. Chapman and J. L. McFarland*, *University of Arkansas, Fayetteville, AR/USA.*

A battery cage experiment was conducted to investigate the efficacy of salinomycin (SAL; 66 ppm) against isolates of *Eimeria* species obtained from the litter of floor-pens in which broilers had been reared with different treatments for four successive flocks. Treatments were: a) SAL for four flocks; b) no SAL for four flocks; c) diclazuril (DIC; 1 ppm) in the starter feed and SAL in the grower feed for two flocks followed by spray cabinet vaccination with Coccivac-B for two flocks; d) SAL in the starter feed and DIC in the grower feed for two flocks followed by Coccivac-B for two flocks; e) DIC in both starter and grower feeds for two flocks followed by Coccivac-B for two flocks; f) Coccivac-B for four flocks. Birds treated with SAL were also given roxarsone (ROX; 50 ppm). All birds received bacitracin methylene disalicylate (BMD; 55 ppm). Four replicates of five birds in cages were given SAL + ROX + BMD in the feed and inoculated with 1000 oocysts of the isolates. Oocyst production in the feces from 5 to 8 days later was measured and expressed as a % of that of birds that only received BMD (controls). The oocyst production of isolates from pens where birds had received SAL for four flocks was 38% of controls indicating that the isolates were partially resistant to SAL. Oocyst production of isolates from pens that had not received SAL was 23% of controls indicating that the efficacy of SAL had improved in the absence of medication. Oocyst production of isolates from pens of birds that had received DIC in the starter, grower, or starter and grower for two flocks followed by Coccivac-B for two flocks was 3%, 3% and 1% of controls respectively, indicating that the efficacy of SAL had been fully restored. Oocyst production of isolates from pens of birds that had been

vaccinated for four flocks was 5% of controls also indicating restoration of SAL efficacy. It is concluded that efficacy of SAL may be improved after vaccination of broilers with Coccivac-B or after use of DIC in broiler feeds followed by vaccination.

Key Words: Salinomycin Diclazuril Coccivac-B

215 Comparison of cecal and fecal sampling methods for monitoring *Campylobacter jejuni* in chickens. G. Zhang*, L. Ma, K. Kinard, and M. Doyle, *University of Georgia, Griffin, Georgia.*

The objectives of this study were to (1) determine the effect of time after cecal and fecal droppings are excreted from chickens on *C. jejuni* cell counts; (2) compare *C. jejuni* counts in cecal and fecal droppings with those obtained from cecal and intestinal contents and mucus obtained during necropsy. For Phase I, 5 fresh cecal and fecal droppings were randomly selected and sampled 4 times (0, 1.5, 3, and 4.5h). In another experiment, 6 fresh fecal dropping mixtures were held at 25, 4, and #20°C, respectively, and sampled every day for 4 days. For Phase II, 15 fresh cecal and fecal droppings were randomly selected and enumerated and 15 chicks were dissected from which cecal content, mucus, small and large intestine content were enumerated for *C. jejuni*. Results showed that *C. jejuni* counts from cecal droppings were higher (1.53 log₁₀ CFU/g) and slightly more consistent than those obtained from fecal droppings. There was no significant difference between *C. jejuni* counts of cecal contents from dissected chicks and those of cecal droppings or among *C. jejuni* counts from the two ceca, and among contents of small and large intestines from dissected chicks and fecal droppings. *C. jejuni* counts at the two ends of each cecum were similar. Mucosal *C. jejuni* counts were slightly lower than those of their respective organ contents. There were no substantial differences between *C. jejuni* counts in fresh cecal or fecal droppings and those assayed 4.5h later. Hence, it may not be necessary to obtain cecal contents from freshly dissected chickens to obtain accurate *C. jejuni* cell counts. Fresh cecal droppings may serve as suitable specimens to determine in vivo cecal *C. jejuni* counts. *C. jejuni* was not detected in fecal samples held at 25 and #20°C in the third day sampling and after, but remained constant (6.89 to 6.17 log₁₀ CFU/g) through the 4 days of sampling in fecal droppings held at 4°C. Hence, if fresh fecal samples cannot be enumerated for *C. jejuni* immediately, samples can be held at 4°C for at least 4 days before analysis.

Key Words: Campylobacter, Poultry, Cecal, Fecal dropping

216 Determination of the cross reactivity using LT and CT antibodies by immunoblot technique in *Salmonella enteritidis* biovar Issatschenko and *Salmonella gallinarum*. O Urquiza-Bravo*¹, G Tllez-Isaias¹, L Paasch-Martinez¹, G Ruiz-Palacios², and A Daz-Barroso², ¹*National University of Mexico,* ²*National Nutrition Institute Salvador Zubiran.*

The Immunoblot technique using CT and LT antibodies was used to determine cross reactivity between supernatant proteins (SP) and periplasmic proteins (PP) of *Salmonella enteritidis* biovar Issatschenko and *Salmonella gallinarum* (Sg) total antibodies. The *Salmonella enteritidis* biovar Issatschenko SP and PP antibodies were produced in rabbit and *Salmonella gallinarum* total antibodies were produced in chicken. Results obtained showed that *Salmonella enteritidis* biovar Issatschenko after a 18 hours of growing is able to produce 1.7 mg/mL of SP and 0.303 mg/mL of PP. In 15% SDS- PAGE there were protein bands corresponding to 97, 66.2, 45 y less than 31 kDa with SP and with PP 66, 42, 35, 33, 32 y 24 kDa. The Immunoblot using Avidina#Biotin and Peroxidase conjugates, revealed homologous and cross reactivity more intensive with anti LT, anti SP and anti PP of *Salmonella enteritidis* biovar Issatschenko antibodies and anti Sg of *Salmonella gallinarum* FVA-1 against *Salmonella enteritidis* biovar Issatschenko SP and PP and SP and PP of the same *Salmonella* but of one year old. Any one of the antibodies used were able to detect reactivity against CT, except the homologous antibodies whose were positive controls. In the other hand, using the anti Sg antibody against SP, PP and SP and PP of one year old of *Salmonella enteritidis* biovar Issatschenko and PP of Sg FVA #1, a high number of protein bands were revealed and more bands were observed with fresh *Salmonella enteritidis* biovar Issatschenko PP. The reactivity observed with anti LT against *Salmonella enteritidis* biovar Issatschenko PP and Sg FVA #1 PP could discriminate different protein bands by Immunoblot whom are

visible by the employment of total Sg antibodies. This technique could be used as a tool for differentiation among some salmonelas.

Key Words: Salmonella issatschenko, Exotoxins, Enterotoxic activity, Salmonella gallinarum

217 Analysis of BMPR2 genes and protein expression as related to ascites syndrome in broilers. J. M. Balog*¹, C. R. Cisar¹, N. B. Anthony², G. R. Huff¹, W. E. Huff¹, N. C. Rath¹, and A. M. Donoghue¹, ¹PP&PSR/ARS/USDA, Fayetteville, AR, ²University of Arkansas, Fayetteville, AR.

Mutations in the bone morphogenetic protein receptor type 2 gene (BMPR2) are a major cause of familial primary pulmonary hypertension in humans, which is similar in pathology to ascites in chickens. Studies were conducted to determine if mutations in the BMPR2 genes could be detected in ascitic birds and to determine if BMPR-II protein expression differed with the disease state of the bird. Two replicate trials were conducted utilizing 6 (5 in Trial 2) popular commercial broiler crosses (80 birds per cross). Day-old, straight run broilers were randomly assigned to cages in a hypobaric chamber (simulated 2,900 m above sea level). Heart, lung, and liver tissues were harvested when ascitic birds were identified. The entire coding region, plus most of the 3' and 5' non-translated regions of BMPR2 mRNA were sequenced. No mutations resulting in a change in the BMPR-II protein were identified. However, 13 anonymous single nucleotide polymorphisms were identified. For BMPR-II protein expression analysis, lung samples were collected from commercial broiler crosses and selected ascites resistant and susceptible broiler lines. The ascites susceptible and resistant lines had been selected over 7 generations by researchers at the U of AR and USDA/ARS for ascites susceptibility when reared under simulated high altitude conditions. Using a commercially available antibody to mammalian BMPR-II, differences in tissue protein expression were observed. The protein that is bound by this particular antibody is expressed at much higher levels in ascitic birds, when compared with normal healthy birds. When protein expression is compared between non-ascitic birds from the ascites resistant and susceptible lines, there are no differences. While differential expression is clearly evident based on disease status, there is concern that the protein expressed is not necessarily chicken BMPR-II. Immunopurification and 2D-gel electrophoresis methods have not yet positively identified the protein. This work was supported in part by the U.S. Poultry & Egg Association Project #285 and Cobb-Vantress, Inc.

Key Words: Ascites, Bone Morphogenetic Protein Receptor, Hypobaric, High Altitude, Broilers

218 Isolation of lytic Salmonella bacteriophages. M.R. McLaughlin*¹, M.F. Bal'a¹, D.E. Rowe¹, K.C. Doerner², R. King², and J. Andersland², ¹USDA, ARS, Waste Management and Forage Research Unit, Mississippi State, MS 39762, ²Department of Biology, Western Kentucky University, Bowling Green, KY 42101.

This research was based on the hypothesis that Salmonella bacteriophages (phages) occur naturally in manure and can be isolated for future characterization and potential use as typing reagents, indicators and bio-control agents. The purpose of this research was to test a protocol for isolation of lytic Salmonella phages. Samples collected from lagoons on commercial hog farms in Mississippi were processed as follows. Subsamples were transferred to sterile glass tubes and mixed with chloroform to kill vegetative bacteria. Chloroform and aqueous phases were allowed to separate and samples were passed through 0.22 micron filters to remove chloroform-resistant spores. Samples were pooled for each lagoon. Salmonella phages were selectively enriched by mixing pooled samples with double strength trypticase soy broth inoculated with a Salmonella cocktail, containing *S. typhimurium* (ATCC 43971 and ATCC 14028) and *S. enteritidis* (ATCC 13076). After 17 hr incubation at 37 C chloroform was added and samples were stored at 5 C. Dilutions of enriched samples were prepared in saline and tested by double agar layer plaque assay with individual Salmonella isolates. Test samples were combined with bacterial suspensions in soft trypticase soy agar (TSA) at 45 C. The soft agar mixture was poured over the surface of hardened TSA in a Petri dish and allowed to harden and dry. Plates were incubated inverted at 35 C and plaques counted after 12 hr. Phage titers of 2.9×10^8 to 2.1×10^9 plaque forming units per ml were produced in the enriched samples. Swine lagoon effluent was a good source of Salmonella phages and

the isolation and enrichment protocol produced high titers of phages for further characterization and research.

Key Words: bacteriophage, phage, Salmonella, plaque assay, swine lagoon

219 In vitro Evaluation of the Ability to Force a Salmonella Bacteriophage to Adapt to Related Alternative Hosts. C.D. Sartor*¹, J.P. Higgins¹, S.E. Higgins¹, L.A. Newberry¹, and B.M. Hargis¹, ¹Department of Poultry Science, University of Arkansas, Fayetteville, AR 72701.

The apparent limitation for utilizing bacteriophages as an alternative to traditional anti-microbial treatment for enteric bacterial infections is bacterial host specificity. Presently, 14 passages of progeny phage from phage isolate (PHL-4), were amplified in the original host (*Salmonella enteritidis*, SE) in the presence of high concentrations of the mutagen, Ethidium Bromide (EB). The putative purpose of this repeated exposure to a known mutagen was to increase the mutation frequency of phage during in vitro propagation. Preliminary experiments indicated that incorporation of EB (0.125 mg/mL) did not reduce SE growth rate when included in the growth medium. Experiment One utilized PHL-4 viral stocks that were amplified in the original host bacterium (SE). In addition, different concentrations of this phage stock were co-incubated in soft-agar overlay plates with either SE, or PHL-4-resistant isolates of *Salmonella kentucky*, *Salmonella indiana*, or *Salmonella tennessee*. In experiment two, PHL-4 was propagated in SE as well as SE co-incubated with the resistant isolates of *Salmonella* utilized in experiment one. Following propagation in the presence of the original SE, plaque formation was clearly evident when PHL-4 was plated with SE using soft-agar overlay, but no plaques were observed when PHL-4 was co-incubated with the individual alternative hosts. The absence of plaques in alternative host bacteria in the present studies suggests that *in-vitro* adaptation of some bacteriophages to alternative hosts may be difficult or impossible using the described methodology. Given a high rate of expected phage mutations in nature, these results could be attributed to the need for simultaneous and complementary mutations to allow successful propagation in the alternative hosts, at least for this selected bacteriophage. Alternatively, multiple bacteriophages may be required for initiation of amplification and subsequent plaque formation (known as Multiplicity Of Infection). Thus successful amplification and plaque formation by any capable mutant phage may not be measurable using the described methods.

Key Words: bacteriophage, *Salmonella*, Alternative Hosts, Host Specificity

220 Bacteriophage selection and application for reduction of Salmonella enteritidis in young poultry. S.E. Higgins*¹, K.L. Guenther¹, L.R. Bielke¹, C.D. Sartor¹, L.A. Newberry¹, and B.M. Hargis¹, ¹University of Arkansas.

Wild-type bacteriophages were isolated in our laboratory (PHL 1-71) against *Salmonella enteritidis* PT 13A (SE) using tryptic soy broth media. The objective of this study was to select appropriate bacteriophages that survive in the gastrointestinal tract of neonatal poultry, and utilize those bacteriophages to reduce intestinal colonization of SE in challenged birds. Bacteriophages from our library were administered to broiler chicks, which were utilized as an in vivo biological filter to select bacteriophages capable of surviving the harsh gastrointestinal environment. A mixture of bacteriophage isolates PHL 1-71 was administered orally to three SE challenged chicks for three consecutive days. Each day, phages were recovered from the ileum, ileocecal junction, and cecae for sequential administration the following day. The mixture of bacteriophages recovered was then utilized to treat SE infected turkey poults. In the first experiment, two-day old poults were challenged with 1×10^4 cfu SE and treated 48 h later with 5mM Mg(OH)₂ followed by 2.5×10^9 plaque forming units (pfu) of bacteriophages in the presence of 1mM Mg(OH)₂. This treatment reduced the number of SE in the cecal contents at both 12 and 24 h after treatment as compared to controls. In a second experiment, two-day old poults were challenged with 1.6×10^4 cfu SE and treated with 5mM Mg(OH)₂ followed by 7.5×10^9 pfu phage in the presence of 1mM Mg(OH)₂ 48 h post-challenge. Culture of the cecal contents at 24 hours post treatment demonstrated a numerical reduction of SE from 79,728 cfu/g in the control group to 11,224 cfu/g in the phage treated group. These data suggest that bacteriophages can be preferentially selected in vivo to increase survival in the avian gastrointestinal

tract. However, improved efficacy is required prior to useful application of the approach to reducing *Salmonella* infection in neonatal poultry.

Key Words: bacteriophage, *Salmonella enteritidis*, poultry

221 Performance of broilers vaccinated with Nobilis Cox ATM[®] and challenged with a Mexican coccidial isolates. V. Davila*¹, M.A. Juarez¹, G. Gonzalez², F. Rios², and E. Avila¹, ¹*Departamento de Produccion Animal: Aves FMVZ-UNAM*, ²*Intervet Mexico SA de CV*.

The goal of this study was to evaluate the protection from a coccidial vaccine in broilers challenged at 28 days of age with a Mexican field isolates (CCI) containing 2.0×10^4 *E. acervulina*; 1.0×10^3 *E. maxima* and 1.0×10^4 of *E. tenella*/bird. All diets were isoproteic, isocaloric, 80 ppm yellow pigment and unmedicated. Chicks were immunized at 5 days of age, seven days post-inoculation (PI) corporal weight (CW) and gut lesion score (GLS) were evaluated. Yellow pigment saturation (PS) was measured at 42 and 49 days of age. CW and feed conversion ratio (FCR) was recorded every week. There were 4 experimental groups (40 chicks/group, Ross x Ross): A) Vaccinated whit CCI; B) Vaccinated whitout CCI ; C) Positive control with CCI; D) Negative Control. Vaccine did not affect the CW. At 35 days of age FCR was different between challenged groups (2.18^a) and unchallenged groups (2.07^b). *E. acervulina* GLS was most higher in the A and C groups (1.90.5 and 2.40.5) than B and D groups (0.50.9 y 1.40.2). *E. maxima* GLS was most severe in group C (1.8 0.3) than A,B,D groups (1.161.0, 00, 0.40.8) respectively. *E. tenella* GLS was less in immunized groups A,B,C and D (0.40.8, 00, 2.20.9, 0.50.9) respectively. PS at 42 days of age was better in vaccinated groups (15.73^a) than non vaccinated groups: (12.65^b). PS in the farm at 49 days of age from vaccinated groups was 18.26^a and non vaccinated groups: 15.51^b. In processing plant the PS at 49 days of age from group B was highest (41.55.0^a) (P<0.05) than remainder groups. Nobilis Cox ATM[®] could be help to increase PS, diminish GLS and FCR when the chicks had a CCI, while did not affect the CW from vaccinated groups.

Key Words: Eimeria vaccine, coccidial challenge, pigmentation

222 Arcanobacterium infection in ostrich chicks. Christian Carlin*, José A Quintana, and Tamas Fehervari, *Universidad Nacional Autnoma de Mxico*.

Sudden death amongst ostrich chicks were observed in a full cycle Mexican ostrich farm during a period of March–September. Eggs were incubated without complications and birth rate was normal. Sudden deaths occurred during the first month of post hatching and 58 percentage of chicks died in this period comparing with 25 percentage of death of same period in previous year. On the spot necropsy reveled only severe renal congestion and in some cases friability of the kidneys were also observed. Samples were sent to the laboratory for serology, virology and bacteriology studies. The presence of Newcastle Disease, avian influenza viruses and infection with *E.coli*, *Salmonella* sp were discarded in the laboratory; however *Actinomyces pyogenes* bacteria now classified as *Arcanobacterium pyogenes* were isolated from various organs. Diseases caused by *Arcanobacterium pyogenes* produced cytolytic toxin (pyolisin) were reported in other animal species but not yet in ostriches. Antibiotic therapy was applied successfully on the basis of antibiogram. Case history and the result of bacteriological study suggest toxin can be the primary factor or the origin of sudden death. Increased hygiene measures, the use of tylosin as well as the development of bacterin could be the options for the prevention and treatment of ostrich chicks endangered by the infection with *Arcanobacterium pyogenes* bacteria.

Key Words: Ostrich, Ostrich chicks, *Arcanobacterium pyogenes*, Pyolisin toxin

223 Evaluation of *Salmonella* susceptibility of Chicks Derived from White or Brown Bovan Eggs. C.M. Pixley*¹, C.D. Sartor¹, K.J. Bramwell¹, S.E. Higgins¹, L.A. Newberry¹, and B.M. Hargis¹, ¹*Department of Poultry Science, University of Arkansas*.

Anecdotal evidence has suggested differences in susceptibility to *Salmonella* infection or pathogenicity among Leghorn chicks derived from white or brown eggs. We compared the effect of selected challenge levels of an (antibiotic-marked) isolate of *Salmonella enteritidis*(SE) on cecal colonization and internal organ (liver+spleen) invasion in chicks

derived from white (WEB) or brown egg Bovans (BEB). For each of two experiments, WEB and BEB were simultaneously obtained on the day-of-hatch from a commercial hatchery and were placed in pens on clean softwood shavings and provided age-appropriate temperature as well as water and feed *ad libitum*. In each experiment, 20 chicks (WEB or BEB) were randomly assigned to each of 5 pens for each strain. At placement in Experiment one, each chick/pen was challenged by oral gavage (0.25 ml saline) containing either 0 (control), 5.4, 5.4×10^1 , 5.4×10^2 , or 5.4×10^3 cfu SE. Recovery of SE from either cecal tonsils or internal organ samples was positively associated with increasing challenge levels regardless of chick strain. No significant ($p > .05$) differences in cecal tonsil or internal organ SE recovery incidence was observed except for the group challenged with 5.4 cfu SE (BEB cecal tonsils 65%, WEB 95%; $p < .05$). In Experiment two, challenge groups consisted of 0 (control), 7.1, 7.1×10^1 , 7.1×10^2 , or 7.1×10^3 cfu SE. SE was recovered from a high percentage of internal organs (>85%) or cecal tonsils (>75%) from BEB, regardless of challenge level. This recovery incidence was significantly ($p < .05$) greater in chicks from BEB as compared to chicks from WEB (7.1 or 7.1×10^1 cfu SE challenge). Overall, these data suggest that there is either no difference in susceptibility to SE infection, or a slightly increased susceptibility of chicks derived from BEB. Further experiments may clarify the question of susceptibility differences and further evaluate morbidity and mortality following virulent *Salmonella* challenge (e.g. *S. gallinarum*).

Key Words: *Salmonella*, susceptibility, chickens, brown eggs, white eggs

224 Evaluation of the Safety of Two S1133 Reovirus Vaccines. S. S. Antle*, P. W. Weatherford, E. Oviedo, S. Thompson, and J. Bray, *Stephen F. Austin State University*.

A study was conducted to evaluate two commercial reovirus vaccines. The experiment evaluated the safety of the vaccine A when administered to 1, 6, 12, and 24-day-old chicks as compared to reovirus vaccine B. Both vaccines were given orally and via footpad for each age group. A USDA challenge reovirus was also given both orally and via footpad for each age group. All of the birds given the vaccine via footpad across all ages developed lesions and swelling of the footpad. The oral vaccine groups did not have any adverse reactions. Forty percent of the 1-day-old birds given the USDA challenge virus via footpad, developed pneumonia and died, the remainder developed footpad lesions.

Key Words: Reovirus, Vaccine, Safety, Challenge virus, Lesions

225 Induction of the *Mycoplasma gallisepticum* pMGA 1.2 gene in the chicken tracheal ring organ culture model. Shawn Bearson¹, Stephanie Collier*², Scott Branton², Bradley Bearson¹, and Roy Montgomery³, ¹*USDA, ARS, National Animal Disease Center, Ames, IA*, ²*USDA, ARS, Poultry Research Unit, Mississippi State, MS*, ³*Mississippi State University, Mississippi State, MS*.

Mycoplasma gallisepticum (MG), the causative agent of chronic respiratory disease in poultry, must adhere to tracheal epithelial cells to establish infection. To identify MG genes involved in colonization of the respiratory tract in poultry, an *in vitro* model system was developed utilizing chicken Tracheal Ring Organ Cultures (TROC) and the *lacZ* gene of *E. coli* to serve as a reporter for mycoplasma gene expression. The chromosome of the MG S6 strain was randomly mutagenized following transformation with the plasmid pBSC5 containing the promoterless *lacZ* gene inserted into the distal arm of the Tn4001 transposon; therefore, expression of the *lacZ* gene is dependent on the adjacent mycoplasma chromosomal DNA. The MG mutants were independently screened for expression of the *lacZ* gene in the absence and presence of chicken TROC using the fluorescent substrate, 5-acetylaminofluorescein di- β -D-galactopyranoside. Preliminary data from this screen revealed a mutant of MG that exhibited a 2-fold increase in β -galactosidase production over a 22-hour period in the presence of the TROC. To identify the genetic location of the Tn4001 transposon insertion in the MG chromosome and thereby determine the mycoplasma gene promoter driving the increased expression of the *lacZ* gene in the presence of the TROC, the chromosome of the MG p12G6 mutant was digested with *Bgl* II, cloned into the pBluescript II SK phagemid and transformed into *E. coli* DH5 α with selection for kanamycin resistance. DNA sequence analysis of the clones containing the Tn4001 transposon and flanking mycoplasma DNA revealed that the Tn4001 transposon had inserted into the MG gene pMGA 1.2, a member of a large gene family of cell surface adhesins

that experience frequent phase variation and are associated with hemagglutination. Thus, initial data from this study suggests that a model system using the chicken TROC with a reporter gene expressed from mycoplasma DNA may identify mycoplasma genes involved in adhesion and colonization of the poultry respiratory tract.

Key Words: Mycoplasma gallisepticum, Hemagglutination, Virulence

226 Hemathological findings of a reovirus infection in broilers complicated with *Salmonella* sp. L.M. Charles*, *Departemanto de Produccion Animal: Aves FMVZ-UNAM.*

Hematological findings are valuable tools to understand the nature of most of the physiopathologic conditions of animals. Ten 21-day-old Ross x Ross broilers were presented in our lab with joint inflammation and poor feathering. Gross lesions included fibrinous pericarditis, and breast muscle petechiae. By histopathological examination, multifocal lymphocytic infiltration was observed. *Salmonella* spp was also isolated. The agglutination test was positive for *Salmonella* spp. The antibody mean titer for reovirus detected by ELISA test was 1761. By hematological studies marked leukocytosis ($87.5 \times 10^3/\text{ul}$) with cariolysis, picnosis and toxic granulation were detected in two out of six samples, which is indicative of septic bacterial infection. Microcytic hypochromic anemia with monocytosis was observed as a consequence of the inflammatory process. Severe leucopenia ($11.4 \times 10^3/\text{ul}$) with lymphopenia (18.8%) and monocytosis (17.3%) was present in three out of six samples, suggesting the presence of a viral disease.

Key Words: Hematology, Reovirus, *Salmonella* spp, Broilers

227 Newly hatched turkeys given La Sota by nebulisation is safe and immunogenic. I. Ciglar Grozdanic, Z. Gottstein, E. Prukner-Radovic, S. Curic, and H. Mazija*, *Veterinary Faculty University of Zagreb, Zagreb, Croatia.*

Objectives: Immune response and safety of commercial La Sota vaccine, given by nebulisation to newly hatched turkeys was investigated. **Materials and methods:** Altogether 40 newly hatched turkeys were disposed into three groups A, B, and C. Group A was exposed to the water suspension of La Sota vaccine for 60 and group B for 300 seconds. Control group (C), wasn't treated at all. All the remedies mentioned were applied by the ultrasonic nebulizer SONOVAC 095 which develops particles ranging 2 to 5 microns in diameter. Blood sera were collected every 7 days until 35th day and in challenged subgroups on 38th day (10 days after challenge). Collected sera were analyzed by standard microtiter method of HI test to detect specific antibodies against vaccine virus. Challenge was performed at 28th day on 10 turkeys in A1 and B1 subgroups, and 5 turkeys in C1 subgroups by eye drop giving 106 Herts 33. **Results:** Used vaccine didn't cause any adverse clinical reaction, regardless the time of exposure to aerosol in SONOVAC 095. Developed active immunity to the given La Sota vaccine obviously was influenced by vaccination. Compared to the negative control, turkeys that have received vaccine responded significantly higher. In challenged subgroups level of antibodies against challenge virus on 38th day was about twice as high than on day of challenge (28th day). After the challenge two turkeys in group A died, as well as one in group B, and four in group C. **Discussion:** Turkeys are less sensitive to velogenic strains of ND virus, never the less they suffer of higher mortality. Therefore lentogenic strains, mainly La Sota, are used to protect turkeys. The vaccine is never given to newly hatched turkeys because of neutralizing effect of maternal antibodies. Obviously, nebulisation avoids this effect. The mechanism of developed immunity and early specific resistance to the challenge infection are not known yet. Nebulisation of La Sota strain is a mode of protection against Newcastle diseases in all those areas where turkey production is endangered.

Key Words: nebulisation, La Sota, immunogenicity, newly hatched turkeys

228 Detection of Marek's Disease Virus from frozen chicken blood using a modified Single Tube PCR technique. Michelle Corley¹, Tina Wright¹, Neil Brown¹, and Ingrid Woode*¹, ¹*Tuskegee University.*

A single tube Polymerase Chain Reaction(PCR) technique was developed to enable rapid identification of viral DNA from frozen chicken blood samples. The technique was demonstrated using Marek's Disease (herpes) Virus (MDV) vaccinated chickens and a single tube PCR kit

originally designed to enable the whole procedure from nucleic acid extraction to PCR in a single tube in a short time, from minute amounts of whole blood from rats, mice and humans. The technique involved the use of 10 μ l of frozen whole blood in a single tube subjected to lyses of blood cells, with subsequent addition of a PCR solution from the manufacturer. The PCR cycling parameters provided by the manufacturer were modified and MDV genomic DNA was detected by amplification of the 132 bp tandem repeat(BamH1-H,D) genomic fragments from MDV vaccinated chickens and from a purified MDV strain SB-1 (control). A second round PCR was required for visualization of PCR products after separation on ethidium bromide stained agarose gels from vaccinated chickens, but not from the purified MDV SB-1 strain. This technique allows rapid and easy assessment of frozen chicken blood for the presence of viral nucleic acids.

Key Words: Polymerase Chain Reaction, Marek's Disease Virus, DNA

229 Expression of the recombinant avian reovirus sigma 3 protein in yeast. N. Henry*¹, L. King¹, Y. Williams¹, C. Frett¹, W. Hongzhan², J. Giambone², and K. Scisum Gunn, ¹*Alabama State University, Montgomery, AL, USA,* ²*Auburn University, Auburn, AL, USA.*

Conventional vaccines against poultry viral diseases use subcutaneous (SQ) or *in ovo* injection in the hatchery, and drinking water (DW) or course spray (CS) routes for field boosting. Edible transgenic plants could provide continual delivery of immunogenic proteins as an efficient alternative to DW or CS field boosting. Preliminary studies from this group showed that recombinant poultry viral immunogens retain their antibody (Ab)-stimulating properties after passage through the birds' digestive system. The avian reovirus (ARV) sigma (σ)3 protein is the 34.9 kDa main host-protective immunogenic protein. The early objectives of this study are: (1) to express recombinant (r) ARV σ 3 protein in a yeast system, and (2) to compare the immunogenic properties of (r) σ 3 protein with native ARV σ 3 protein by western blotting and antigen-capture ELISA. The ARV strain S1133 σ 3 gene was isolated and amplified via RT-PCR, and introduced into the *S. pombe* yeast expression vector pESP-1. cDNA sequencing confirmed establishment of the pESP1- σ 3 expression construct. Polyclonal Abs against whole ARV S1133 have been generated in 2-week old specific pathogen free (SPF) white leghorn chicks for western and ELISA analyses. Confirmation of (r) σ 3 expression by western blotting and ELISA are in progress. Continuation studies will involve oral vaccination of SPF chicks with (r) σ 3 protein, *in vitro* virus neutralization assays with (r) σ 3-generated polyclonal Abs, and ARV challenge studies in orally vaccinated SPF birds. The long-term objectives are to develop a transgenic plant system expressing (r) σ 3 protein, and to conduct oral vaccination and challenge studies. These studies will contribute to a better understanding of orally-induced immunity in chickens, and to improved vaccine designs based upon oral vaccination.

Key Words: Avian reovirus (ARV), Viral arthritis, Sigma (σ) 3 protein, Yeast expression systems, Edible transgenic vaccines

230 Effect of water supplementation with soluble vitamin E and sodium salicylate (Uni-Sol)TM on the resistance of turkeys to *Escherichia coli* respiratory infection. G.R. Huff*¹, W.E. Huff¹, J.M. Balog¹, N.C. Rath¹, and R.S. Izard², ¹*USDA/ARS Poultry Production and Product Safety Research, Fayetteville, AR,* ²*Animal Science Products, Inc. Nacogdoches, TX .*

There is a need to evaluate potential antibiotic alternatives for improving disease resistance in high intensity poultry production. Colisepticemia is one of the most costly diseases to affect turkeys and often requires antibiotic treatment. The objective of this study was to determine the prophylactic efficacy of two commercial products, soluble vitamin E (VE) and soluble sodium salicylate (Uni-Sol) in an *Escherichia coli* respiratory challenge. The drinking water of male turkey poults was non-supplemented or supplemented with either VE or Uni-Sol or a combination of both at dosages recommended by the manufacturer. There were 110 birds in each treatment, which were housed in four floor pens. At 5 wk of age, birds in half of the pens were challenged with an air sac inoculation of approximately 10 cfu of *E. coli* or were left unchallenged. Water treatment commenced 5 days before challenge and continued for 2 wk after challenge, when birds were necropsied. The effect on decreased BW due to *E. coli* challenge was ameliorated by all treatments, however VE and Uni-Sol, but not the combination decreased BW in non-challenged controls. Vitamin E and Uni-Sol treatments both significantly decreased

mortality and air sacculitis scores. All treatments protected liver, spleen, and bursa weights relative to BW from the effects of *E. coli* challenge and Uni-Sol alone or VE with Uni-Sol protected relative heart weights from the effect of challenge. Uni-Sol treatment alone and in combination with VE increased total leukocyte counts and the number and percent of lymphocytes. Vitamin E alone increased the heterophil/lymphocyte (H/L) ratio whereas Uni-Sol alone decreased the H/L ratio. All treatments decreased the isolation rates of *E. coli* from the liver. These results suggest that treatment of turkey poults with VE and Uni-Sol, prior to and during the stressful events that can lead to colisepticemia, may decrease disease incidence and mortality.

Key Words: Vitamin E, Sodium salicylate, Turkeys, *Escherichia coli*, Antibiotic alternative

231 Evaluation of a commercial coccidial vaccine for *Eimeria* specific species in broilers after challenge with Mexican field isolates. M.A. Juarez*, N. Ledesma, R. Merino, G. Nava, and G. Tellez, *Departamento de Producción Animal: Aves FMVZ-UNAM.*

The aim of this study was to determine the effect of a commercial coccidial vaccine on body weight (BW) and the presence of specific *Eimeria* species (SS) in broiler chicks vaccinated at one day of age and challenged at 21 days with Mexican field *Eimeria* isolates (CCI) containing 6.0×10^4 *E. acervulina*; 5.0×10^3 *E. maxima*; and 4.0×10^4 *E. acervulina*/bird. All broiler diets (S/S) were isoproteic, isocaloric and without anticoccidial drugs. There were 4 experimental groups (20 chicks/group, Ross x Ross): 1) Negative control; 2) Positive control with CCI; 3) Vaccinated with CCI; 4) Vaccinated without CCI. At days 21 and 28, all birds from each group were weighed and SS found in feces were recorded at 28 days. Body weights at 21 days were: 615.65100.1^a; 622.072.7^a; 544.8105.9^b and 504.988.5^b (P<0.05); for groups 1,2,3, and 4 respectively. At 28 days they were: 923.3126.2^a; 775.0599.05^b; 731.5120.6^b and 726.2130.7^b (P<0.05) for the groups in the same order. Weight gain from 21 to 28 days was: 50.1%; 24.6%; 34.5% and 44.1% for groups 1,2,3, and 4 respectively. No *Eimeria* were found at 21 days of age for groups 1 and 2. The distribution of SS for group 3 was: 80% *E. acervulina*, 8% *E. maxima* and 12% *E. tenella*. For group 4: 83% *E. acervulina*, 2% *E. maxima* and 15% *E. tenella*. At 28 days we observed for group 1: 87% *E. acervulina*, 13% *E. tenella*; for group 2: 54% of *E. acervulina*, 22% of *E. maxima* and 24% of *E. tenella*; for group 3: 21% of *E. acervulina*, 41% of *E. maxima* and 38% of *E. tenella*; and for group 4: 84% of *E. acervulina*, 4% of *E. maxima* and 12% of *E. tenella*. The growth performance in the vaccinated and challenged groups 7 days PI, was very affected. The lack of immune protection against *E. maxima* and *E. tenella* was very important. Molecular characterization of Mexican coccidial field strains and their comparison with the strains present in a commercial vaccine will be necessary to determine possible antigen differences.

Key Words: *E. maxima*, *E. tenella*, challenge, vaccine, chicks

232 Performance improvement with Nobilis Cox ATM[®] and Sacox[®] in broilers challenged with Mexican coccidial isolates. M. A. Juarez*¹, V. Davila¹, F. Martinez¹, G. Gonzalez², F. Rios², and E. Avila¹, ¹*Departamento de Producción Animal: Aves FMVZ-UNAM*, ²*Intervet Mexico SA de CV.*

The objective was to compare the performance of broilers vaccinated at five days of age by drinking water with Nobilis Cox ATM[®] and challenged at 28 days of age with a Mexican field isolates (CCI) containing 2.0×10^4 *E. acervulina*; 1.0×10^3 *E. maxima* and 1.0×10^4 of *E. tenella*/bird. All diets were isoproteic, isocaloric and Salinomycin 60 ppm. Finisher with 80 ppm yellow pigment. A 2 x 2 factorial arrangement were used. The treatments (80 birds Ross x Ross by group) were A) vaccinated with CCI; B) vaccinated without challenge; C) nonvaccinated with CCI; D) nonvaccinated without challenge. Average live weight (AW) and feed conversion ratio (FCR) were recorded weekly. At 35 days of age the gut lesion score (GLS) was evaluated. In farm and processing plant, skin pigmentation degree (SPD) was measured at 49 days of age. AW interactions at 35 days of age were (P<0.043) in A,B,C,D groups (1.740.03^a, 1.750.01^a, 1.720.03^a, 1.680.01^b) respectively. At 42 days of age AW from group D was less than remainder groups. No AW differences were found at 49 days of age. FCR showed interaction, group B (2.05^a) was better than C (2.06^a), A (2.1^a) and D (2.19^b) groups. GLS with *E. acervulina* showed differences between challenged vaccinated groups (1.2^b) and challenged nonvaccinated groups (2.1^a). *E. maxima* GLS interactions were to group A: 0.6^b, group C: 1.16^a; B and D groups not showed GLS. No

E. tenella GLS were found in A, B and D groups; but, C group showed GLS of 0.6. At 42 days of age SPD from group B (19.22^a) was highest; and group D (16.12^b) was lowest. At 49 days of age SPD from groups C (22.7^a) and B (20.0^a) were most better than remainder groups. Broiler performance was not affected from broilers vaccinated with Nobilis Cox ATM[®]. Vaccinated groups with CCI showed the best FCR. Nobilis Cox ATM[®] together Salinomycin supplementation avoid field strain challenge. Nobilis Cox ATM[®] can help to maintain the ionophores until use.

Key Words: Vaccination, Ionophores, *Eimeria spp*

233 Hemotological changes and hemostatics in Leghorn chicks infected with 73688 of infectious bursal disease virus. Araceli Lima, Norma Calderon, Tamas Fehervari*, Teresa Fortoul, and Luz Maria Charles, *Universidad Nacional Autónoma de México.*

Infectious bursal disease virus (IBDV) affects young chickens and highly virulent IBDV causes severe clinical signs, even mortality with lesion in bursa of Fabricius, thymus, spleen and bone marrow. There is only few information about hemorrhagic diathesis caused by IBDV. The objective of this work is to determine if the IBDV causes anemia, leucopenia, thrombocytopenia and prolonged prothrombin time. White Leghorn SPF 2 weeks of age were used. Chickens were kept in isolation units of the Department of Animal Production: Poultry at the Faculty of Veterinary Medicine and Zootecnia, UNAM. Two groups were formed and placed in separate isolation units. Eighteen chickens were infected via oral in the first group with 0.2 ml/birds of 7388 IBDV. Eight chickens in group two were controls, without any treatment. Samples from the first group were collected 24, 36, 48, 60, 72, 84 and 96 hours post infection (hip) and 24, 60 and 96 hip from the control group. Samples were kept with EDTA for the hemogram and thrombocyte count evaluation. Blood samples used for the evaluation of coagulation, were treated with 3.8% sodium citrate of (in proportion 1:9). Prothrombin time (PT) was determined in a coagulometer Trombotimer Organon Teknika. Anemia and leucopenia were found in the infected group from 84 hpi, and no changes were detected in the control group. A light thrombocytopenia was observed at 24 and 36 hpi in the infected group, however, it was not significant (P<0.01). Concerning to the evaluation of PT, the control group kept normal values during all sampling time while the infected group showed significant (P<0.01) prolongation in prothrombin time from 48 to 96 hpi. On the basis of results it can be concluded that IBDV provokes diminution of the different cellular lines and it causes coagulopathy.

Key Words: Infectious bursal disease, Thrombocytopenia, Prothrombin time

234 Hemostatic and hematological alterations in chicks inoculated with H5N2 highly pathogenic avian influenza virus. Guadalupe Ramirez, Norma Calderon, Tamas Fehervari*, Teresa Fortoul, and Luz Mara Charles, *Universidad Nacional Autónoma de México.*

Avian influenza (AI) is characterized by hemorrhages, but its etiology and the description of the development of hematological changes are not discussed in detail in the bibliography. The objective of this study was to determine hemostatic changes in birds infected with AI and detected hematology changes developed after infection. 51 SPF Leghorn birds, 4 weeks of age, of both sexes were used. Animals were divided into 2 groups: 42 birds consisting the first group were inoculated via oral with the H5N2 avian influenza virus (AIV); the second group, with 9 birds were inoculated with physiologic saline and served as controls. Both groups were kept in separated high security units at the Laboratory of National Investigation Institute for Forestry and Agriculture (INIFAP). Six chicks from the first group were sampled at 24, 36, 48, 60, 72, 84, 96 hours of postinoculation (hpi). The control birds were sampled at 24, 60 y 96 hpi. The results show no significant differences (p<0.01) in haemostatic values and in the prothrombin time (PT) didn't in any samples compared with the controls. The number of thrombocytes showed differences (p<0.01) amongst the two groups after the 24 hpi. The thrombocytes in the 24 hpi were 14.22 2.26 (109/L) in the infected group and 96 HPI 7.82 2.01 (109/L) in the control showing leukocytosis characterized by heterophilia and monocytosis at 36 HPI and 60 HPI respectability with a p< 0.01. The thrombocytopenia in the infected animals could be associated with alterations of thrombocytes and its precursors by a direct effect of AIV, as it is reported in case of other viral infections as classic swine fever. Any changes were observed concerning PT and the development of coagulopathy was discarded in these samples. The

hematological changes could be associated to the inflammatory process caused by AIV infection.

Key Words: Influenza, hematology, Haemostatic, Thombocytopenia

235 Detection of enteroinvasive *Escherichia coli* in chickens. C. Rosario^{*1}, C. Eslava², and G. Tllez³, ¹DPA: Aves, FMVZ, UNAM, ²Depto. de Salud Pública, FM, UNAM, ³USDA-ARS-PPPSRU.

Some *Escherichia coli* strains are positive in the Serny test for invasiveness. The ability to invade colonic epithelial cells is mediated by a large plasmid encoding genetics determinants required for invasion. These *E. coli* strains are known as enteroinvasive *E. coli* (EIEC). EIEC strains are biochemically, genetically, and pathogenetically related closely to *Shigella* spp. Both are generally lysine decarboxylase negative, non motile, and lactose negative. The O antigen of *Shigella* and other enteric bacteria is encoded by genes organized as *rfb* operon. In most bacteria this region is located on the chromosome. However, in *Shigella sonnei* is encoded on the invasion plasmid. Embedded within the operon is an *IS630* element. From this element primers HS61 and HS61 were used for PCR. The objective of the present study was to analyze a collection of *E. coli* which previously had been positive to *ipaH* primers in DNA hybridization assay, with different characteristics from classic EIEC. Seventy six *E. coli* strains isolated during the first week from chicks with yolk sac infection were analyzed. Mortality were recorded daily during this week. A PCR using HS61 and HS62 primers was performed. A *S. sonnei* strain were used as positive control and an enteroaggregative *E. coli* were used as negative control. Results show that only 8 strains were positive in this essay. All strains belong to O2NM serotype and were isolated during the third day, when the higher mortality were seen. Two out of the eight positive strains were isolated from the same chick from different organs (yolk sac and liver). This data suggest that invasiveness could be a mechanism in *E. coli* strains which cause yolk sac infection. But the disease in poultry could be caused by different strains than human EIEC because the predominant serotype in this study were not reported as a classic EIEC serotype previously.

Key Words: Yolk sac infection, *Escherichia coli*, invasiveness

236 In vitro screening of nutraceuticals for immuno support in avians. J Wiles^{*1}, R Benyard², and J Grizzle¹, ¹University of Tennessee, Knoxville, TN, ²Albany State University, Albany, GA.

Mycotoxin contamination of feed grains leads to immunosuppression, a reduction in growth rate, feed efficiency, and reproductive performance in avians. In the pet bird industry, many diets and treats are high in fat and moisture making these products susceptible to fungal or mold contamination and oxidative rancidity. Our laboratory is interested in the use of herbal ingredients to act as protective nutraceuticals against mycotoxins and reactive oxygen molecules. Herbal products are garnering attention as over-the-counter supplements for numerous medical conditions, but are most popular as antioxidants. T-2 toxin is an immunosuppressive agent primarily produced by the fungus *Fusarium tricinctum* and can be found on any feed grain. Ginkgo bilboa, curcumin, silymarin (milk thistle), selenium and vitamin E are herbal and vitamin/mineral supplements known for their antioxidant and immunosupportive properties. The objective of this research is to test these antioxidants for their protective properties against insult with an immunosuppressant. At near confluence, HD-11 avian macrophage cells were allowed to adhere to chamber slides overnight. Cells were washed with PBS (phosphate buffered saline), and incubated with PBS, latex beads, T-2 toxin (0, 16 mg/L), and varying concentrations of herb (1, 0.1, 0.01X manufacturer's

recommended dose), vitamin E (0, 0.6, 3.0 mg/L), or selenium (0, 2.0 mg/L). After 2 hours incubation, slides were rinsed, stained, and cells engulfing beads were counted by bright-field microscopy. Phagocytosis of beads was significantly less ($P \leq .01$) among cells incubated with 16 mg/L T-2 toxin versus PBS (30.2% vs 62.5%). Rescue of cells treated with T-2 toxin was facilitated by use of 0.1X ginkgo bilboa (57.3%) or silymarin (62.9%), and was not different ($P \geq .05$) from controls (62.7%). Co-incubation of cells with T-2 toxin and vitamin E or selenium did not restore phagocytosis to control levels ($P \geq .05$). Results from this study suggest that dietary nutraceuticals may be used as immunosupportive agents in avians.

Key Words: Mycotoxins, Avian, Immunosuppressant, Nutraceuticals, Pet bird

237 Human Health Risks from Use of Virginiamycin in Chickens. Louis Anthony Cox, Jr., Ph.D.¹ and Kenneth W. Bafundo, Ph.D.², ¹Cox Associates, 503 Franklin Street, Denver, Colorado 80218, and ²Phibro Animal Health, Fairfield, New Jersey 07004.

The streptogramin antibiotic combination Quinupristin-Dalfopristin (QD) has been used in the U.S. since 1999 to treat patients with vancomycin-resistant *Enterococcus faecium* (VREF) infections. Another streptogramin, virginiamycin (VM), has been used as a growth promoter and therapeutic antimicrobial in farm animals in the U.S. and other countries. Many chickens test positive for QD-resistant *E. faecium*, raising concern that VM use in chicken might compromise QD effectiveness against VREF infections by promoting development of QD-resistant strains that could be transferred to human patients.

Despite the potential importance of this threat to human health, quantifying the risk via traditional farm-to-fork modeling has proved extremely difficult. Enough key data (mainly on microbial loads at each stage) are lacking so that such modeling amounts to little more than choosing a set of assumptions to determine the answer. Yet, regulators in the U.S. and some other countries feel they should act now to protect the efficacy of QD for human patients. Patients prescribed QD are typically severely ill, immunocompromised people for whom other treatment options are not readily available. Thus, there is a pressing need for sound risk assessment methods to guide decision-making for VM/QD using currently available data.

This paper takes a new approach to the QD-VM risk modeling challenge. It recognizes that the usual farm-to-fork approach is unlikely to produce reliable results soon enough to be useful. However, reversing the farm-to-fork process and starting with readily available human data on VREF caseloads and QD resistance rates offers a promising alternative. Combining these data with recent genogroup frequency data for humans, chickens, and other sources (Willems *et al.*, 2000; 2001) allows us to quantify potential human health risks from VM usage in chickens in the United States. The paper presents a risk simulation model, incorporating recent nosocomial transmission and genetic typing data and estimates human QD treatment failures over the next five years with and without continued VM use in chickens. The quantitative estimates and probability distributions are thoroughly grounded in data. They are calculated via a Monte-Carlo uncertainty analysis and simulation model over a 5-year horizon beginning in the first quarter of 2002.

The main results indicate that in the U.S., an immediate ban on VM is predicted to reduce average attributable treatment failures by 1.8 cases in the entire American population over 5 years and expected mortalities by 0.29 cases in the same time period. The model shows that with high confidence, the theoretical statistical human health benefits of a VM ban range from zero to less than one statistical life saved in the entire U.S. population over the next 15 years, and are rapidly decreasing. Sensitivity analysis shows that this conclusion is robust to data gaps and uncertainties, e.g., about the extent of resistance transfer from chickens to people.