**171    Genetic characterization of commercial broiler lines experimentally infected with Subgroup J Avian Leukosis Virus (ALV-J).** M Karaca*, J. K. Rosenberger, and S. S. Cloud, *University of Delaware, Newark,DE* .

This study was initiated to determine the genetic basis for weight gain or immune response in chickens infected with ALV-J. Embryos from 4 genetic lines of chickens were inoculated with the UD-4 isolate of ALV-J by the yolk sac route at 3 days of incubation. An equal number of embryos were sham inoculated. At hatch chicks were vaccinated with NDV and IBV and during grow-out exposed to infectious bursal disease virus, reovirus and chicken anemia virus to simulate commercial conditions. Body weights were measured every 2 weeks. At 5 weeks, chickens were assessed for their ability to clear *E.coli* following intravenous inoculation. This was done because ALV-J is believed to compromise resistance to bacterial infections. Biweekly body weights and *E.coli* blood clearance assays were analyzed using the GLM procedure of SAS. A total of 129 (A=42, B=40, C=47) chickens were genotyped using 33 microsatellite markers. The association of genotype with the phenotypic measurements of body weight and *E.coli* clearance were determined for sham vs ALV-J inoculated chickens. The heterogeneity of each locus was calculated within lines. The average observed heterogeneity of line A, B and C over 33 loci was 30.65 %, 33.31 % and 31.29 % with 3.24, 2.97 and 3.45 alleles per locus, respectively. The average allele sharing between lines A and B was 72.23 %, between lines A and C it was 66.07 % and 61.91 % between lines B and C. Overall, the lines shared 55.38 % of their alleles. The genetic distance between line A and B or C was found to be 25.82 or 27.41 % respectively. Both suggestive (P<0.10) and significant (P<0.05) associations of marker genotypes with quantitative trait loci (QTL) affecting biweekly body weights and clearance of *E.coli* were detected. Two marker loci were found to be significantly (P<0.05) associated with body weight at 2 and 4 weeks of age for line A. One marker genotype had a significant effect (P<0.05-P<0.10) on clearing pathogenic *E.coli*. Further characterization of the genetic lines is continuing by genotyping progeny tested sires with 100 microsatellite markers.

**Key Words:** Genotyping, Subgroup J Avian Leukosis Virus (ALV-J), Disease resistance

**172    Relationships between skeletal growth and body weight in Japanese quail selected for 4 week body weight.** J. M. Reddish*, A. El-Keredy, K. E. Nestor, and M. S. Lilburn, *Dept of Animal Science, The Ohio State University, Ohio Agricultural Research and Development Center*.

A weight selected strain of quail(HW) selected for increased 4 week body weight was compared with a randombred control to investigate the relationship of skeletal growth and selection for body weight. The heavy weight (HW) strain was derived from the randombred control strain (RBC). Body weight differences (HW>RBC) between strains were consistent throughout the experiment. The objective was to investigate the growth of the tibia and femur and determine if selection for resulted in differences between the HW and RBC strains. Measurements were taken of body weight, length and width of both the tibia and femur at weekly intervals from 1 to 35 days of age. Body weight difference (50%) was consistent throughout the experiment (HW>RBC) and growth patterns were similar. Differences in skeletal measures and body weight were only 15% for both RBC and HW (p<.01). Correlations for skeletal traits and body weight were positive, and significant(p<.01) for RBC thru day 28 but HW showed disruption in this pattern. The results suggest that selection for body weight and skeletal growth are symmetrical in the RBC, but different in HW. The HW strain has proportional growth but does not have symmetry between skeletal traits and body weight as noticed in the RBC strain.

**Key Words:** selection, quail, growth

**173    The effect of selection for increased egg production in turkeys on incubation characteristics of embryos.** A. L. Antonelli*, K. E. Nestor, and M. S. Lilburn, *Department of Animal Sciences, Ohio State University/OARDC, Wooster, OH.*

In strains of turkeys selected for egg production (E; Nestor et al., 1980, 1995), genetic increases in egg production are proportional to decreases in the weight of the albumen and yolk, as well as the total egg weight when comparisons are made to a randombred control line (RBC1). In the present study, E line hens were mated to E line toms or RBC1 toms, and RBC1 hens were mated to RBC1 toms. Egg weight at set was 67.2 ± .39g for E/E eggs, 65.6 ± .32g for E/RBC1 eggs, and 87.16 ± .68g for RBC1/RBC1. At 21 days of incubation, E/E yolk-free embryo weight was 5% less than in E/RBC1 embryos, yet at 25 days there was no differences. At hatch, E/E yolk-free embryo weight was 5% greater than that of E/RBC1 embryos. Overall, the E/E line embryos had a 64% increase in yolk-free weight between 21 and 25 days with overall weight gain of 95%. E/RBC1 had only a 56% increase in yolk-free weight between 21 and 25 days with overall weight gain of 70%. RBC1/RBC1 embryosl had an 80% increase in yolk-free weight between days 21 and 25 with an overall weight increase of 118%. At 21 days of incubation, RBC1/RBC1 yolk-free embryo weight was 11% heavier than the E/E line and these differences increased to 25% and 26% at 25 d and hatch (28 d), respectively. The data suggest that genetic differences in embryonic development are maximally expressed from 21 to 25 days of incubation, concomitant with the period of maximal yolk lipid transfer to the development embryo.

**Key Words:** Turkey, Incubation, Embryo

**174    Germ-line transmission of a *lacZ* gene in chickens using an avian Spleen Necrosis Virus-based vector.** S. Borwornpinyo*, D.W. McCoy, P.E. Mozdziak, and J.N. Petitte, [1]*North Carolina State University.*

Replication-defective retroviral vectors based on the avian spleen necrosis virus (SNV) have been successfully used to produce transgenic chickens. One particular vector, SNTZ, expresses nuclear-directed beta-galactosidase. The SNTZ vector has been used as a cell lineage marker for the analysis of early embryonic development of the chick. The purpose of the present study was to produce transgenic chickens using SNTZ. High titers ($2.0 \times 10^7$ virion/ml) of SNTZ were injected into the subgerminal cavity of 66 stage X white Leghorn embryos followed by *ex ovo* embryo culture. Of the 66 infected embryos, 16 hatched (24%). To date, of the 16 hatched chicks, 12 have reached sexual maturity. Genomic DNA from blood and semen samples from these $G_0$ birds were tested for the presence of *lacZ* sequences using the polymerase chain reaction in combination with primers specific for *lacZ*. Out of 7 hens, 2 birds had detectable *lacZ* sequences in their blood. Of the 5 cockerels, 3 were carrying the bacterial *lacZ* gene in the semen. To date, test mating of males and females yielded germ-line transmission from one male at a frequency of 1 out of 20 offspring. No correlation was observed between the presence of *lacZ* sequences in circulating red blood cells and semen. The results of this study suggest that the SNTZ vector can be used to develop a line of transgenic chickens that expresses nuclear-directed bacterial beta-galactosidase. Support was provided through funds under projects NC05293 (PEM), NC06590 (PEM), and NC0168 (JNP).

**Key Words:** retroviral vectors, germ-line transmission, transgenic

**175    Molecular characterization of the genomic chicken prolactin receptor (cPRLR) gene from a native Chinese chicken (Wai Chow strain).** Angela Hui* and Frederick Leung, *University of Hong Kong.*

The aim of this study is to clone and characterize the genomic cPRLR gene from a native Chinese chicken (Wai Chow strain). Prolactin receptor (PRLR) participates in a number of physiological functions in birds including reproduction, maternal behavior and osmoregulation. It is widely distributed in many tissues including the brain, the testis and, particularly abundant, in hypothalamus and the anterior pituitary gland. The cDNA of PRLR has been previously shown to consist of an extracellular ligand-binding domain, a single transmembrane and an intracellular domain involved in signal transduction. However, the genomic molecular structure of the cPRLR gene is still lacking. We used the Polymerase Chain Reaction (PCR) trapping method. By designing different pairs of primers flanking the possible intron splice sites, fragments of introns can subsequently be amplified for subcloning and further sequencing analysis. Three introns have been successfully trapped, and the estimated sizes are 1.4kb, 1.5kb and 1.7kb. These DNA fragments were subcloned and further confirmation by DNA sequencing. Consequently, these sequences will be analysed in the databank for any

significance discovered, which helps further understanding of the gene expression pattern and to reveal important pieces of evidences on the gene diversity. Tissue culture will also be carried out to further characterize the promoter region using deletion expression approach.

**Key Words:** Chicken, Prolactin receptor, Intron

**176    Molecular characterization of the chicken prolactin (PRL) gene:genomic gene structure, its polymorphism and promoter analysis.** Florence Au* and Frederick Leung, *University of Hong Kong.*

Prolactin (PRL) is a polypeptide hormone of the anterior pituitary gland and has been shown to have a diverse spectrum of biological activities and functions in all vertebrates. Characterization of the chicken PRL gene and its polymorphism is the first step in establishing the genotypes and traits association. Polymerase chain reaction (PCR), cloning and sequencing were used to obtain the four intron sequences of the chicken PRL genomic gene. PCR products indicated that the sizes of the four introns are of 1.5 kb, 0.5 kb, 1.3 kb and 2.0 kb respectively. Sequence analysis of the four PRL introns of the chicken PRL gene revealed that they share high homology with that of the turkey PRL gene and the other strains of chicken PRL gene. In addition, PCR-restriction fragment length polymorphism (PCR-RFLP) was used to identify polymorphic sites within the four introns. An Ava II enzyme cut site in the first intron was found to be polymorphic in Chinese native (Guangdong Xing Hua) chickens. PCR-RFLP was also applied to other chicken strains such as broiler, layer and other native Chinese strain (Shek kai). However, No polymorphism was identified. Present results enable extended study in the PRL gene diversity. In addition, an approximated 2 kb 5' flanking of the cPRL was also obtained by PCR and confirmed by DNA sequencing. Deletion analysis and luciferase reporter gene assay will be used to characterize and define the promoter region in PRL gene expression.

**Key Words:** Chicken, Prolactin, Polymorphism

**177    Detection of a single nucleotide polymorphism in exon 10 of the chicken growth hormone receptor gene.** Joanna Lau* and Frederick Leung, *University of Hong Kong.*

Interaction of Growth Hormone (GH) with its receptor (GHR) is required for normal growth in both mammalian and avian species. Defect in GHR gene functions caused by mutation is one of the major causes of genetic disorder, eg. Laron Syndrome and Dwarfism in human and chicken respectively. Recent studies suggest that polymorphisms on both the GH and GHR genes in chicken might serve as genetic markers for phenotypes of commercial values. In this study, a new restriction fragment length polymorphism (RFLP) at exon 10 of the GHR gene has been identified in native Chinese strain chicken, using polymerase chain reaction (PCR) and Alu I restriction enzyme digestion. The same study has been extended to broiler and layers strains and no RFLP was detected. The PCR-RFLP results were further verified by direct sequencing analysis and confined to a single nucleotide polymorphism (SNP) silent mutation at position 924th nucleotide (GA) counting from the translational start site on the published cDNA sequence, resulting in no change in the amino acid. Future studies will be set up to examine whether such mutation has any association with phenotypic traits.

**Key Words:** Chicken growth hormone receptor, Restriction fragment length polymorphism (RFLP), Single nucleotide polymorphism (SNP)

**178    Candidate genes and reproductive traits in a commercial broiler breeder population, an association study.** I C Dunn*[1], Y-W Miao[1], A Morris[2], M N Romanov[1], D Waddington[1], P W Wilson[1], and P J Sharp[1], [1]*Roslin Institute, Roslin, Midlothian EH25 9PS, Scotland,* [2]*The Cobb Breeding Company, East Hanningfield, Essex, CM3 8BY, England.*

To take advantage of programmes to identify candidate genes for variation in traits of economic importance, methods to test these genes in selected pedigree populations need to be developed. To this end we have carried out a study of association between candidate genes and reproductive traits in a pedigree line of broiler breeders. Gonadotropin releasing hormone (GnRH), its receptor (GnRHR), growth hormone receptor (GHR) and neuropeptide Y (NPY) were selected for their role in controlling aspects of reproduction. Genetic markers for NPY, GnRHR and GHR alleles were detected using bulk PCR-restriction fragment length polymorphism or BESS-T Scan (Epicentre Technologies). Genotyping of 772 hens from one generation was by PCR-restriction fragment length polymorphism. Total number of eggs, age at first egg (AFE) and number of double yolked eggs (DY) for each hen were recorded. Additive and dominance effects were fitted for the autosomal GnRHR and NPY genes; additive effects were fitted for the sex linked GHR gene. To control for some of the background genetic variation, candidate genes were assessed within heterozygous sire families. A dominance effect of NPY (14 sire families) on AFE and an additive effect of GnRHR (36 sire families) on DY, were found (P<0.02). If the latter effect were true, selection could increase overall flock performance by 0.13 usable eggs per hen. A simplified model, omitting sires, was also fitted. This analysis gave four significant associations (P<0.05), a surprisingly large number. In conclusion it is possible to detect association between economic traits and candidate genes in a population undergoing selection, and test if a candidate gene explains some of the trait variation. However, statistical associations between trait and genes require to be treated with caution and models should account for as many genetic and environmental variables as possible.

**Key Words:** Reproduction, Candidate genes, Association

**179    Mapping QTL Loci Affecting Growth And Disease Resistance to Avian Coccidiosis.** J Zhu*[1], H Lillehoj[1], C Van Tassell[2], M Emara[3], P Allen[1], H Cheng[4], D Pollock[5], M Sadjadi[5], and T Sonstegard[2], [1,2]*U.S.Department of Agriculture, BARC, Beltsville, MD,* [3]*University of Delaware, Newark, DE,* [4]*U.S.Department of Agriculture, ADOL, East Lansing, MI,* [5]*Perdue Farms, Inc., Salisbury, MD.*

Selection of commercial poultry stocks with improved disease resistance using classical genetic breeding techniques has been unsuccessful due to technical difficulties. Although selection based on progeny tests can be used, this is labor-intensive, time consuming, and costly. In order to develop a DNA marker-assisted selection strategy to improve disease resistance against avian coccidiosis in commercial breeder chickens, chicken genes controlling resistance to coccidiosis are being identified. Three hundred and twenty four F2 offspring for mapping quantitative trait loci (QTL) affecting disease resistance were produced from 12 full-sib families of a commercial broiler breeder. The F2 offspring were inoculated with 104 sporulated oocysts of Eimeria maxima at 4 weeks of age. Body weight gain and fecal oocyst shedding were determined as a measure of infection. One hundred and twenty chicken microsatellite markers with an average genome distribution of 20 cM were used for genotyping the F1 and F2 generations. Genotypic data were analyzed with CRIMAP version 2.4 to construct a marker linkage map. A minimum LOD score of 3.00 was used as the statistically significant threshold for declaring linkage. The QTL analysis was conducted using SOLAR on genotypes of the F1 and F2 and the phenotypes of the F2 chickens. A locus on chromosome 1 was identified that was significantly associated with reduced oocyst shedding and 3 potential loci affecting growth were identified on chromosomes 1 and 6. (Supported by Fund for Rural America, Grant No 9704985 and partially by ARS CRIS).

**Key Words:** Coccidiosis, QTL mapping, Disease resistance

**180    The use of molecular markers to associate feather color alleles with tissue pigmentation in broiler chickens.** R Okimoto*, *University of Arkansas.*

Consumers dislike black melanin pigment in the abdominal skin and fascia. It has been demonstrated that certain feather color alleles are associated with this pigmentation. The dominant white allele ($I$) and sex-linked barring allele ($B$) are known to reduce the incidence of abdominal pigmentation when coupled with the extended black allele ($E$) of the $E$ locus. Since most commercial broilers are white feathered the cryptic feather color alleles that are segregating within the population cannot be determined without test mating. Molecular PCR based markers would facilitate selection against unwanted alleles. We have developed PAMSA (PCR amplification of multiple specific alleles) tests that can distinguish the various $E$ locus alleles. These tests detect specific nucleotide substitutions in the melanocortin 1-receptor gene that result in amino acid substitutions in the receptor sequence associated with specific alleles of the $E$ locus. In order to test the efficacy of using these tests in selection against tissue pigmentation a cross between two broiler dam lines was made to create an $F_1$ population that was nearly

fixed for recessive white ($cc$), segregating for sex-linked barring ($B$) and heterozygous for the $I$ and $E$ loci ($IiEe^b$). $F_2$ chicks were hatched and 345 were typed for tissue pigmentation at 15 days of age and 1700 were typed at 5+ weeks of age. All $F_2$ chicks were white feathered due to recessive white ($cc$). Pigmentation ranged from no visible pigment to pigmented birds with a few spots around the navel to large areas of the abdominal region having intense pigmentation. Total incidence of abdominal tissue pigmentation was 27% at 15 days and 23.6% at 5+ weeks. At this time, 79 $F_2$ chicks have been PAMSA genotyped for their $E$ locus alleles, and were found to segregate 1:2:1, as expected, for the $EE$:$Ee^b$:$e^be^b$ genotypes. As expected, the $EE$ genotype was associated with the least abdominal pigmentation (8.7%), while 18.4% of the $Ee^b$ chicks and 44.4% of the $e^be^b$ chicks were pigmented. Results indicate that selection for the $E$ allele in dominant white broiler stocks would decrease the incidence of abdominal tissue pigmentation, and that molecular based tests would facilitate this selection.

**Key Words:** Broiler, $E$ Locus, Pigmentation

### 181    Effect of dietary protein, photoperiod, and genetic background on growth and sexual maturity in Japanese quail. A. El-Karedy, K. Nestor, and M. Lilburn*, *The Ohio State University/OARDC, Wooster, OH*.

Japanese quail are a valuable species for genetic studies in poultry due to their short generation interval and relative ease of maintenance. The current study included two lines of Japanes quail, a large-bodied line (HW)selected for increased BW at 4 wk of age and a randombred control line (R1) from which the HW line was developed. The selection environment includes a period of reduced daylength and this environment results in sexual maturity (age at first egg) at about 53 d in both lines (Anthony et al., 1993), whereas continous lighting can reduce sexual maturity by approximately 10 d (Lilburn et al., 1992). The selection diet contains 28 % CP and, in previous experiments, this diet resulted in a significant line by diet interaction for growth and onset of sexual maturity (Lilburn et al., 1992). In the present study, HW and R1 quail were fed one of three diets (21, 24, or 27 % CP) and exposed daily to either an 8- or 24-h photoperiod from hatch to 28 d of age and 24 h thereafter. The R1 hens reached sexual maturity about 2 d earlier than HW hens (49.8 vs 51.9 d; P≤0.004). The 8-h photoperiod delayed sexual maturity by 12 d (56 vs 44 d; P≤0.0001)and the 27 % CP diet enhanced sexual maturation by 4 d compared with hens fed either 24 or 21 % CP diets (47.3 vs 51.5 d; P≤0.0001). There were no significant two-way interactions. As would be expected, the 24-h photoperiod significantly reduced BW at sexual maturity in both lines but there were no significant diet effects. The 24 and 27 % CP diets increased oviduct weight at sexual maturity in HW hens compared with the 21 % CP diet (10.24 vs 9.3 g) and there was no diet effects in the R1 line (5.6 g), resulting in a significant line by diet interaction (P≤0.031). There was a linear decrease in ovary weight in both lines with each incremental increase in dietary CP but no significant effects due to photoperiod. The data suggest that dietary CP and photoperiod had independent effects on various traits measured at sexual maturity and these effects were not necessarily similar within the two genetic backgrounds.

**Key Words:** Japanese quail, sexual maturity, genetics

### 182    Is improved feed conversion associated with increased lethargy and docility in broiler chickens? D. O. Skinner-Noble*[1], R. B. Jones[2], and R. G. Teeter[1], [1]*Oklahoma State University, Stillwater, OK 74078*, [2]*Roslin Institute (Edinburgh), Midlothian EH25 9PS*.

An experiment was conducted to test the hypothesis that broilers classified as good rather than poor feed converters (high versus low gain-to-feed ratios, respectively) would show decreased activity (increased lethargy) and reduced fear of human caretakers. The birds (450 large broiler-type) were reared straight run to 36 d of age when they were sexed. Of the 227 males that were then weighed and subjectively evaluated for breast conformation and leg defects, 192 were placed into individual-bird cages (46 x 60 cm) at 40 d of age and their feed conversion ratio (FCR) was measured over the next 7 days. During this time the birds were subjected to three behavioral tests. Firstly, we scored their approach / avoidance responses to a novel object placed in the feeder at a time when feeding would be expected. Secondly, in an assessment of the general behavior patterns of individual birds we recorded feeding, drinking, walking, pecking, preening, standing and

resting. Thirdly, the 20 birds showing the best FCR and the 20 showing the worst were induced into tonic immmobility (TI); this test of underlying fearfulness is an anti-predator reaction to manual restraint. We found that feed conversion was not related to either the novel object or the TI measures of fearfulness. Neither was FCR related to feeding, drinking, walking or pecking. However, gain-to-feed ratio was positively correlated with both standing and preening, and negatively correlated with resting behavior. Therefore, contrary to expectations, the present results suggest that broilers that showed good feed conversion ratios were actually less lethargic and no more fearful than those that showed poor FCR values.

**Key Words:** Feed conversion, Lethargy, Fearfulness

### 183    Level and pattern of DNA sequence variation in the chicken genome. Edward Smith*, *Virginia Tech*.

Variation among chickens for metabolic and physiological factors are significant indicators of differences in performance and health. Individual variation in physiological and metabolic factors can be influenced by differences in candidate genes. Identification of mutations in these candidate genes as well as the determination of the distribution and structure of the variants in different populations are essential in properly defining their roles. Here results of a DNA sequence-variation screen of five metabolically important chicken genes including type X collagen (col10), aggrecan (agc1), cyclin D3 (cycD3), ovalbumin A (ovbA) and chicken PepT 1 (cpept1) are described. For each gene, a panel of 48 unrelated birds from six commercial layer-type breeder lines were used without regard to their health or performance status for resequencing of PCR-amplified diploid products. Variable DNA sites or single nucleotide polymorphisms were identified in iterative stages that involved alignment of multiple sequence traces and the identification of heterozygous loci based on context-specific patterns of the individual sequences. Each variable site was re-sequenced at least once and most were aligned in more than one contig. As expected and have been observed in human and mouse, there was significant variation among genes. For example, within the 800-bp sequence evaluated from the 3d end of cpept1, a total of 16 variable sites were detected and confirmed. From the cycD3 gene, however, no variable sites were detected within the coding sequences. Within a total of 40 kb of DNA sequence screened, an average of 1/150 bp of variable sites were detected. The value of this information is at several levels: First, SNP discovery within these functionally important genes will now make it easier to carryout association studies designed to identify genes that influence disease and performance traits. Second, the high frequency of variable sites suggest that biallelic markers may be useful for the construction of a third generation chicken genetic map that can be used for quantitative trait locus identification. (Acknowledgment: Hong Chen, Yuan Pan, Kenny Webb and Eric Wong are gratefully acknowledged for sharing the PepT1 sequence with me prior to publication).

**Key Words:** chicken, DNA sequence variation, linkage disequilibrium

### 184    Novel Randomly Amplified Polymorphic DNA Markers for the Turkey Genome. Amy Spellerberg* and Edward Smith, *Virginia Polytechnic Institute and State University, Blacksburg Virginia/USA*.

Though it is a commercially important meat animal species, our knowledge of the turkey genome, relative to the chicken and other species, remains limited. To address this limitation, a publicly available and widely disseminated turkey genome map is necessary. The utility of such a map includes gene discovery and quantitative trait loci identification for economically important traits. To support the development of a turkey genome map, randomly amplified polymorphic DNA (RAPD) markers may be a useful resource. Here we describe a novel set of RAPD markers developed by screening parental samples from members of a turkey DNA reference panel. Using conventional PCR techniques, ninety-six RAPD primers were used in the screening. Amplified products were separated on agarose gels and analyzed for the presence of informative fragments based on Mendelian inheritance. A total of thirty-two primers, or 33.3% of the primers used for screening, revealed polymorphic fragments that showed Mendelian inheritance. These fragments ranged in size from 400 bp to 3 Kb and cumulatively represented the analysis of a 46.8 Kb

portion of the turkey genome. This study establishes that RAPD markers may be a reliable and efficient resource for constructing a genomic linkage map of the turkey genome.

**Key Words:** Turkey genome linkage map, RAPD markers, polymorphic loci

**185    Use of AFLP DNA markers to evaluate genomic diversity and genetic distances in Japanese quail lines divergently selected for stress responsiveness.** F. M. Odeh*[1] and G. G. Cadd[1], [1]*Department of Poultry Science, Louisiana State University Agricultural Center, Baton Rouge, LA 70803.*

The secretion of corticosterone is a classic endocrine response to stress in avians. In this study, the genomic diversity and genetic distances in Japanese quail lines divergently selected for stress responsiveness were evaluated using an amplified fragment length polymorphism (AFLP) technique to generate DNA markers. DNA from individuals of extreme performance within each selected line was extracted and pooled for analysis. DNA from five different individuals was pooled for each replicate. Four replicates from each selected line and two replicates from a randomly bred (RB) line were screened by DNA fingerprinting. The enzyme combination *EcoRI/TaqI* was used for a double digestion of the DNA pools. Fragments with *EcoRI/TaqI*-end were radioactively labeled, separated by molecular weight by gel electrophoresis, and apposed to X-ray film for visualization. Ninety-seven *EcoRI/TaqI* primer combinations generated 20,416 bands with 5,089 of them being polymorphic. The AFLP procedure amplifies a high number of restriction fragments (52 bands, on average, in a single primer combination) and produces markers that are scored as the presence or absence of a band. The average band sharing (BS) values produced from AFLP fingerprinting within lines were 0.590, 0.631, and 0.581 for HS, RB, LS lines, respectively, whereas the average BS values between the lines were 0.573, 0.550, and 0.572 for HS-RB, HS-LS, and LS-RB respectively. The BS values within each given line and between the lines were, however, very sporadic, regardless of primer-combination used and the line analyzed. In this study, 5.5% of the genomic variation was attributed to mutation rate, 0.57% to genetic drift, and 0.057% to bidirectional selection. There were insignificant differences in average genetic distances between the selected lines. Although AFLP failed to resolve genetic diversity between the selected lines, it clearly identified fragments of DNA that may help determine the major genes involved in the stress responsiveness phenotype.

**Key Words:** Stress responsiveness, AFLP, Genetic distance, Japanese quail

# PSA Nutrition: Amino Acids

**186    Impact of phase-feeding on growth performance of broilers fed diets adjusted every other day for decreased amino acid content.** H.R. Pope*, J.A. Townsend, and J.L. Emmert, *University of Arkansas*.

An experiment was conducted during the finisher period (42 to 56 d) to determine the effects of decreasing dietary amino acid levels as often as every other day using the phase-feeding (PF) approach. Experimental diets were formulated using either NRC recommendations for lysine (Lys), sulfur amino acid (SAA) and threonine (Thr) or recommendations from linear regression equations generated from estimates of Lys, SAA and Thr requirements. To facilitate implementation of PF, two diets were prepared that contained Lys, SAA and Thr levels matching the predicted requirements for birds at 42 d (high nutrient density) and 56 d (low nutrient density). After being properly mixed and pelleted, the high and low nutrient density diets were blended in variable quantities to produce rations containing amino acid levels that matched the predicted PF requirements over two day intervals (42-44 d, 44-46 d, 46-48 d, 48-50 d, 50-52 d, 52-54 d, 54-56 d). Birds were fed either a single NRC diet from 42 to 56 d or a series of PF diets that were switched every other day to supply a gradual decrease in Lys, SAA and Thr. Weight gain, feed intake, and feed efficiency of birds fed the PF regimen were unaffected (P > 0.05) relative to birds fed the NRC diet. An evaluation of digestible Lys, SAA, and Thr intake revealed a significant decrease (P > 0.05) in Thr intake for those birds given the PF diets, however no differences (P > 0.05) in digestible Lys or SAA intake were observed between the NRC and PF diets. These data indicate that dietary Lys, SAA, and Thr content maybe reduced as often as every other day under a PF program in which high and low nutrient density diets are blended without adversely affecting growth performance. Moreover, because dietary Lys, SAA, and Thr levels are decreased under the PF system substantial dietary cost savings may be available with such a program.

**Key Words:** Broiler, Phase-feeding, Amino Acids

**187    Evaluation of lysine and arginine needs in broiler finisher diets.** E. A. Oviedo-Rondon*, C. A. Fritts, and P. W. Waldroup, *University of Arkansas*.

Two studies were conducted in floor pens to examine response of male broilers to combinations of Lys and Arg in broiler finisher diets. In the first study, a diet was formulated with 0.75% Lys and 1.0% Arg. Aliquots of the diet were supplemented with Lysine HCl and arginine free base to provide a 3 x 4 factorial arrangement with three Arg levels (1.0, 1.2, and 1.4%) in combination with four Lys levels (0.75, 0.85, 0.95, 1.05%). Each treatment was fed to four pens of 50 male Ross 308 broilers from 42 to 63 d. In the second study, three Arg levels (1.0, 1.1, and 1.2%) were fed in combination with four Lys levels (0.75, 0.85, 0.95, and 1.05%) with each treatment fed to four pens of 50 male Cobb 500 broilers from 42 to 56 d. Body weight gain and feed conversion (FC) were determined and birds were processed for dressing percentage (DP), breast yield (BY) and abdominal fat (AF) content. The level of Arg had no significant effect on BW gain, FC, DP, BY, or AF in either study, indicating that the NRC (1994) recommended level of 1.0% is adequate for this age period. No significant interactions occurred between levels of Lys and Arg for any criteria in either study. In the first study, conducted in moderate temperature, Lys had no significant effect on BW, FC, or DP; BY was increased by 0.85% Lys but higher levels gave no further advantage. In the second study, conducted in high temperatures, BW gain, FCR, and BY were significantly improved by increasing Lys from 0.75 to 0.85% but higher levels gave no further advantage. These data suggest that current NRC (1994) levels of Arg and Lys are satisfactory for broilers during the finisher period of 6 to 8 or 9 wks of age under periods of moderate or high temperatures.

**Key Words:** Arginine, Lysine, Broilers

**188    The influence of dietary labile methyl donors on arginine requirement of young broiler chicks using growth and muscle creatine as parameters.** M. Chamruspollert*, G.M. Pesti, and R.I. Bakalli, *Department of Poultry Science, The University of Georgia, Athens, GA 30602-2772.*

Two experiments were conducted to investigate the effect of methyl donors on the arginine requirement of young broiler chicks. One-day-old Ross x Ross chicks were fed corn-whey based diets for 14 days. Four replicate pens of ten chicks each were randomly assigned to each treatment. At 14 d, two birds from each pen were killed and breast muscles were collected for creatine analysis. In Experiment 1, six levels of arginine (ARG) (0.95, 1.05, 1.15, 1.25, 1.35, or 1.45%) were fed with two levels of methionine (MET) (0.45 or 0.65%). Based on body gain data, the ARG requirement was 1.19±0.07% or 1.16±0.06% when chicks were fed the 0.45 or 0.65% MET diet, respectively. The requirement was 1.16±0.04% or 1.19±0.06%, respectively, based on muscle creatine. However, based on FCR, the ARG requirement of chicks fed 0.45% MET was higher (1.25±0.04%), compared with those fed with 0.65% MET (1.16±0.04%;P<0.05). In Experiment 2, two sources of methyl donors, MET or betaine (BET) were compared on an equal percentage basis. Eighteen treatments with 6 levels of ARG (as above) were factorially arranged with no methyl donor supplementation, or 0.2% MET supplementation (0.65% total) or 0.2% BET supplementation (0.2% total). There was no significant interaction (P>0.05) between ARG and methyl donor source on body weight gain and feed intake. The body gain data were pooled to calculate the ARG requirement. Therefore, based on body gain, the ARG requirement was 1.20±0.05%. Based on FCR, we found the interaction between ARG and methyl donor source (P<0.05). Based on FCR, the ARG requirement of chicks fed the unsupplemented diets was 1.29±0.06%, fed diets supplemented MET was 1.24±0.04%,