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**BREEDING AND GENETICS:  
APPLICATIONS AND METHODS IN  
ANIMAL BREEDING – POULTRY**

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**0951 (T049) Regulation of microRNAs in necrotic enteritis infected two genetically disparate chicken lines.** Y. H. Hong\*, *Chung-Ang University, Anseong-si, South Korea.*

Necrotic enteritis (NE) is an acute clostridial disease causing weight depression, loss of appetite, and sudden death. MicroRNAs (miRNA) play a critical role in post-transcriptional regulation by influencing the 3'-UTR of target genes. Using two inbred White Leghorn chicken lines, line 6.3 and line 7.2 showing Marek's disease-resistant and-susceptible phenotypes, respectively, we used small RNA high-throughput sequencing to investigate whether miRNAs are differently expressed in these two chicken lines after inducing necrotic enteritis (NE). The 12 miRNAs, selected from the most downregulated or up-regulated miRNAs following NE induction, were confirmed by their expressions in real-time PCR. Among these miRNAs, miR-215, miR-217, miR-194, miR-200a, miR-200b, miR-216a, miR-216b, and miR-429 were highly expressed in the intestine derived from line 7.2, whereas, miR-1782 and miR-499 were downregulated ( $P < 0.05$ ). In spleen, miR-34b and miR-1684 were the most up-regulated miRNAs in line 6.3 ( $P < 0.05$ ). Notably, five out of six target genes, CXCR5, BCL2, GJA1, TCF12, and TAB3 were differentially expressed between line 6.3 and line 7.2 ( $P < 0.05$ ), and showed suppression in the MD-susceptible chicken line. Their expression levels were conversely correlated with those of miRNA obtained from both HTS and quantitative real-time PCR.

These results suggest that some miRNAs are differentially altered in response to NE, and they modulate the expression of their target genes in the two inbred lines. Collectively, high-throughput analysis of intestinal miRNAs from NE-afflicted inbred chickens showing different disease phenotypes led to the identification of host immunity genes regulated by miRNA. Future studies of the function of these miRNAs and their target genes in the host will lead to enhanced understanding of molecular mechanisms controlling host-pathogen interaction in NE.

**Key Words:** necrotic enteritis, miRNA, chicken

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**0952 (T050) Changes in variance of top SNP windows over generations under selection for three traits in broiler chicken.** B. D. Fragomeni<sup>\*1</sup>, I. Misztal<sup>1</sup>, D. Lourenco<sup>1</sup>, I. Aguilar<sup>2</sup>, and R. Hawken<sup>3</sup>,  
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The purpose of this study was to determine whether the top SNP windows that explain the most variance are stable over multiple generations of selection in a GWAS analysis using single-step GBLUP. Phenotypes were available for five generations of a pure line of broiler chicken for body weight, breast meat, and leg score. Pedigrees included 297,017 animals, of which 294,632 had phenotypic records over five generations. Genotypes of 57,635 SNP were available for 4922 animals. After quality checks, 41,036 SNP and 4866 animals remained in the genomic file. SNP effects were calculated by a GWAS type analysis using single-step GBLUP approach. In each run, the generations were grouped from 1–3, 2–4, 3–5, and 1–5. The evaluation model included sex and contemporary group as fixed effects, animal additive and maternal permanent environmental as random. In GWAS by single-step GBLUP, genomic breeding values (GEBV) are converted to SNP effects. Variances of SNP effects were derived iteratively in three iterations without re-estimation of GEBV. As individual SNP explained very small portion of the total genetic variation, variances were then calculated for windows of 20 SNP and interpreted as the percentage of the total genetic variance. Ten windows for each trait were identified that explained the largest fraction of the variance in any combination of generations. All the top 10 windows explained at least 0.5% (but less than 2%) of the total genetic variance in all the traits. The variance explained by each window varied greatly among the combinations of generations. In several cases, a window identified as top for one combination of generations explained less than 0.1% variance in the remaining combinations. Top windows of SNP variance in the broiler population are unstable and unsuitable for direct selection. Results in this study could be influenced by many generations of intensive selection in broiler chicken and by a small number of genotypes.

**Key Words:** genomic selection, genome-wide association study, ssGBLUP

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**0953 (T051) Relationship between laying frequency and egg sizes in quail.** O. T. Abanikannda\*, O. N. Ottun, and A. O. Leigh, *Lagos State University, Ojo-Lagos, Nigeria.*

The number of eggs laid by hens during a laying cycle is one of the desirable traits in commercial egg production, and the size of eggs produced by hens determine the quality, grade, marketability, and acceptability of eggs by the consumers. Several factors such as breed, age, weight, and management

**Table 0953.** Mean  $\pm$  SE of measured variables

Laying Group	N	HenWt (g)	FeedWt (g)	EggWt (g)	EggLt (mm)	EggWd (mm)	EggSSA (mm <sup>2</sup> )	EggVol (mm <sup>3</sup> )	EggDens
Low	4	166.40 $\pm$ 1.51	30.13 $\pm$ 0.84 <sup>ab</sup>	10.08 $\pm$ 0.11 <sup>a</sup>	30.39 $\pm$ 0.19	24.36 $\pm$ 0.14	22.40 $\pm$ 0.24	10.19 $\pm$ 0.16	1.0525 $\pm$ 0.0006 <sup>a</sup>
Medium	3	168.99 $\pm$ 0.92	27.30 $\pm$ 0.72 <sup>b</sup>	9.64 $\pm$ 0.11 <sup>b</sup>	29.92 $\pm$ 0.16	24.09 $\pm$ 0.14	21.81 $\pm$ 0.18	9.79 $\pm$ 0.12	1.0522 $\pm$ 0.0006 <sup>b</sup>
High	10	169.06 $\pm$ 0.78	32.64 $\pm$ 0.49 <sup>a</sup>	9.96 $\pm$ 0.04 <sup>a</sup>	30.01 $\pm$ 0.12	24.19 $\pm$ 0.10	22.01 $\pm$ 0.14	9.95 $\pm$ 0.09	1.0524 $\pm$ 0.0005 <sup>a</sup>

practices have been reported to influence quantity and quality of eggs produced by quail hens. This study investigates the relationship between quantity and quality of eggs as depicted by the frequency of lay and measurements taken on eggs. After an initial stabilization period, a total of 435 eggs were collected from 17 quail hens over a continuous laying period of 34 d. The hens were classified into three nominal groups as Low (17 to 21), medium (22 to 26) and High (26 to 31) depending on the number of days of lay during the study period. Majority (58.82%) of the hens were in the High laying group, while the Medium and Low laying groups accounted for 17.65 and 23.53%, respectively. Consequently, about two-thirds (66.44%) of the eggs studied were from hens in the High laying group. Parameters studied include Hen Weight (HenWt), Feed Consumed (FeedWt), Egg Weight (EggWt), Egg Length (EggLt), Egg Width (EggWd), Shape Index (ShpInd), Egg Surface Area (EggSSA), Egg Volume (EggVol), Egg Density (EggDens), and Surface Area to Volume Ratio (SSAVol). All statistical analyses (Descriptive, Correlation, Regression, and ANOVA where  $Y_{ij} = \mu + \alpha_i + e_{ij}$ ) were done using Minitab 16 software. A one way analysis of variance (ANOVA) on each of the parameter revealed that only three of the parameters (EggWt, FeedWt and EggDens) were significantly ( $P < 0.05$ ) affected by laying group (Table 0953). There was a significantly ( $P < 0.001$ ) moderate correlation ( $r = 0.5$ ) between ChickWt, FeedWt and EggWt and a highly significant ( $P < 0.001$ ) correlation ( $r = 0.99$ ) between EggWt and EggDens. Hens with lower frequency of lay consumed less feed and had higher egg weight, whereas hens in the middle laying group consumed least and had the least egg weight. Small-sized quail hens consumed moderately, laid least eggs laid but had the heaviest eggs.

**Key Words:** laying frequency, quail, egg weight

**0954 (T052) Phenetic classification of six bird species based on the proximate and mineral composition of their eggs.** O. T. Abanikannda\*, O. N. Ottun, and A. O. Leigh, *Lagos State University, Ojo-Lagos, Nigeria.*

Classification is a systematic grouping of organisms into categories on the basis of evolutionary or structural relationships between them, based on their biological similarities and differences. Phenetic classification is the quantification and statistical assessment of characters based on overall or observable similarities rather than on phylogenetic or evolutionary relationships, with an orderly arrangement of organisms in hierarchical series. A total of 240 eggs comprising 40 eggs

from each of six species (chicken, duck, guinea fowl, pigeon, quail, and turkey) were sampled. Proximate composition (moisture content, dry matter, total ash, crude protein, crude fat, and carbohydrate) of the eggs along with mineral analyses (calcium, magnesium, manganese, iron, zinc, and cobalt) were conducted using standard laboratory procedures for proximate and mineral assay. All statistical analyses which included descriptive, analysis of variance and multivariate cluster analyses were done with Minitab Statistical software. Species was a highly significant ( $P < 0.001$ ) source of variation in all variables measured except for crude fat, Mg and Zn, which were not affected ( $P > 0.05$ ) by specie. The complete linkage method, with squared Euclidean distance and three clusters specified as final partition was used on the standardized variables. Three main clusters were identified with duck, turkey, and quail forming a cluster and joining with chicken and guinea fowl as a second cluster, while pigeon was in the third cluster. This clustering is close to the phylogenetic classification in traditional taxonomy of the three classes (anseriforms, galliforms and columbiforms), and provides a good basis for comparative classification of the six different species of birds.

**Key Words:** taxonomy, phenetic classification, cluster analysis

**Table 0954.** Clustering of the six species based on proximate and mineral composition analyses

Step	Number of Clusters	Similarity Level	Distance Level	Clusters Joined	New Cluster	Number of Observation in New Cluster
1	5	97.5304	1.5258	4	3	2
2	4	96.1613	2.3717	2	1	2
3	3	82.2077	10.9930	5	1	3
4	2	63.7853	22.3753	3	1	5
5	1	0.0000	61.7852	6	1	6

**0955 (T053) Effect of shell thickness on quail chick pip-out at hatching.** O. T. Abanikannda\*, A. O. Leigh, and O. N. Ottun, *Lagos State University, Ojo-Lagos, Nigeria.*

Economic losses incurred by farmers when chicks could not pip out at hatching is a major consideration in commercial hatchery operations. The inability of chicks to come out of an egg shell unassisted has its attendant consequences on the survival and livability of quail chicks. Aside from genetic effect, age and nutritional status of the hen, egg shell thickness is also a major factor. This study was conducted to determine

the effect of egg shell thickness on the on the ability of quail chicks to pip out of the shell at hatching. A total of 593 fertile eggs collected from a farm located in Jos, Plateau State, in the Savannah region of Nigeria, were incubated and hatched. The eggs were weighed and linear measures were taken using digital weighing scale and caliper. The measured variables include, egg weight, egg length, egg width, shell thickness, vertical and horizontal circumference, while shape index, egg density, egg volume, egg surface area, and surface area to volume ratio were computed. All statistical analyses were done using JMP statistical software for the descriptive statistics, correlation, model fitting, and logistic regression. After incubation, 570 eggs hatched, and eggs that were not hatched ( $n = 23$ ) were opened up to determine if the chicks fully developed but could not pip out unassisted. The result revealed that there was no statistical difference ( $P > 0.05$ ) in the mean of all variables studied. A binary logistic regression of the shell thickness on hatching status was not statistically significant ( $P > 0.05$ ), indicating that despite the slight numerical difference in shell thickness of the two groups (Hatched, Not Hatched), it was not enough to invoke a statistical significance probably due to unequal subclass numbers in the two groups.

**Key Words:** hatchability, quail, egg shell

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#### 0956 (T054) Weight changes in quail eggs during

**incubation.** O. T. Abanikanda\*, O. N. Ottun, and  
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Commercially, quails are mostly raised for their meat and eggs. The eggs are tastier, low in calories and are good sources of essential vitamins, minerals and amino acid, thereby making it a preferred egg compared to other poultry eggs. Chick weight at hatch positively impacted on subsequent productivity indices of quails thereby making it a primary index for

the future performance traits of the bird. Differences in chick weight at hatch have been largely influenced by the pre-hatch egg weight and weight loss associated with incubation. Chick weight is primarily determined by initial egg weight and is secondarily determined by weight loss during incubation, shell and residue weight, strain, incubation time and conditions, breeder age and chick sex. This study aims at investigating some egg measures and its influence on chick weight at hatch with a view to statistically predict its chick weight. The eggs used in this study were sourced from a semi-intensively managed poultry farm in Jos, Plateau State in the Savannah region of Nigeria. A total of 987 hatching quail eggs were collected and appropriately labeled for identification purposes and set for incubation. Out of the total eggs set, 606 eggs were hatched and were used for the analyses. Egg weight and egg shell weight were measured with a sensitive (0.00 g) digital scale, while egg length and width were measured with a sensitive (0.00 mm) digital Vernier caliper. Other measures included both the vertical and horizontal circumference of the eggs using a flex graduated tape, 14th day incubation weight of egg and chick weight at hatch. Indices such as shape index, egg density, egg surface area, egg volume, and incubation weight difference were computed. With the exception of shell thickness and shape index, all other variables were highly significantly ( $P < 0.01$ ) impacted on chick weight and were used to model. The statistical model for predicting chick weight at hatch using the 11 variables that significantly influenced chick weight at hatch is given by:  $Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_4 X_4 + \beta_5 X_5 + \beta_6 X_6 + \beta_7 X_7 + \beta_8 X_8 + \beta_9 X_9 + \beta_{10} X_{10} + \beta_{11} X_{11}$ . The model accounted for 31% of the variability in chick weight. The study revealed that egg weight, density, volume, and incubation weight loss were the largest influences on chick weight.

**Key Words:** quail eggs, incubation, chick weight