

**1237 Imaging system for fecal and ingesta detection on poultry carcasses.** K. C. Lawrence, B. Park, W. R. Windham, and D. P. Smith\*, *USDA, ARS.*

A hyperspectral imaging system was developed to detect surface contaminants on poultry carcasses. The system consists of a transportable stand, two quartz halogen line lights, a prism-grating-prism imaging spectrograph connected to a 1280x1024 pixel silicon CCD camera, and a computer with a frame-grabber card. The imaging system is capable of collecting spectral reflectance information from 430 to 900 nm with 1-nm resolution for every pixel of a carcass image. The resulting three-dimensional image cube is typically reduced to 320x340 pixels of spatial information with 512 pixels of spectral information for each spatial pixel. This paper reports the results of this system for the detection of ingesta from the crop or gizzard and feces from the duodenum, ceca, and colon on the surface of a poultry carcass. Sixteen six-week old male birds on a corn/soybean diet were meal-fed, subjected to an 8-hour feed withdrawal, cooped, slaughtered, hard scalded (57.5 C for 2 min.), picked, eviscerated, and feces and ingesta was collected. Uncontaminated carcasses were then immediately imaged. Next, feces and ingesta were applied to varying locations on the breast, wings, and legs of the carcass, typically three spots per contaminant, 12 spots per bird, for a total of 190 contaminant spots, and second images were taken. Results are presented for several data analysis techniques including principal component analysis, which includes all measured wavelengths, and the wavelength ratio of the 565-nm image divided by the 517-nm image. The wavelength-ratio images were further processed with background masking, thresholding, and histogram stretching. The threshold and histogram stretching values were fixed for all birds. Linear and square root histogram stretches were performed. Results indicated that 97.3 and 100 percent of the contaminants were detected with the linear and square-root histogram stretches, respectively. The research shows the feasibility of a real-time system for fecal and ingesta detection from two wavelength images at typical poultry processing line speeds.

**Key Words:** Feces, Imaging, Food safety

**1238 Effects of post-mortem deboning time and L-value classification of raw fillets on color and texture characteristics of cooked broiler breast meat.** B. G. Lyon\*<sup>1</sup>, E. T. Moran<sup>2</sup>, C. E. Lyon<sup>1</sup>, and E. M. Savage<sup>1</sup>, <sup>1</sup>*USDA, ARS, Russell Research Center, Athens, GA*, <sup>2</sup>*Auburn University, Auburn, AL.*

Color of raw broiler meat has consumer implications and may also be indicative of functional properties of the meat. In this study, broiler breast samples from two deboning times were sorted by instrumental color (Minolta); then evaluated for cooked color, aroma and texture. Samples were from 8 wk old male broilers (Ross X Hubbard HiY) subjected to common live production and normal pre-slaughter handling. After processing, *pectoralis majors* were removed from chilled carcasses either at 4-6 h (early, ED) or 24-30 h (late, LD) post-mortem. Fresh fillet light reflectance was measured 48 h after slaughter. Each fillet was

IQF, held at 0C, and sorted by L-value (lightness) into two groups, low (<46, LL) or high (>52, HL) from the total having a grand mean of 49. Thawed samples were cooked individually in heat-and-seal bags immersed in 85C water to internal temperature of 78C, and evaluated for aroma, shear force, and light reflectance measurements of outside surface, inside cut surfaces and decanted cook fluid. Thawed raw weight and cooked yield were not significantly different. Cooked L-values for outside and inside cut surface of EDHL were significantly higher than the other three groups. Significant differences in cooked liquid color measurements were found (EDLL < EDHL). Shear force values were significantly higher for EDHL breast meat. Aroma of EDLL was the least brothy, most chickeny and most bloody/serumy; EDHL samples were most metallic. These results agree with other reports that cooking reduces color variation. However, early deboning may present more color differences than late deboning. Further work is needed to elucidate factors that can allow prediction and control of breast meat color and quality.

**Key Words:** Broiler breast meat color, Instrumental L-values, Deboning time

**1239 Lipid and Fatty Acid Composition of Some Specialty Eggs.** Gita Cherian\*, Troy B. Holsonbake, and Mary P. Goeger, *Oregon State University, Corvallis, Oregon, USA.*

Many specially fed and raised chicken eggs are available in the United States with labels such as 'vegetarian', 'cage-free', 'organic', 'non-medicated', 'naturally-nested' or 'free-range'. The objective of the present study was to compare the egg components, total fat and nutritionally important fatty acids in eggs with special labels or claims. A total of six different brands of eggs with labels such as 'vegetarian high n-3', 'free-range', 'organic', 'uncaged-non-medicated', 'vegetarian-cage-free' or 'cage-free-naturally-nested' were collected and analyzed. A significant (P <.05) difference was observed in the egg components and fatty acid content in different brands. The percent yolk was lower (P <.05) in 'organic' and 'cage-free-vegetarian eggs' with a concomitant increase (P <.05) in the percent white. The percent shell was lower (P <.05) in 'cage-free-vegetarian' and 'naturally-nested cage-free' eggs. No difference (P >.05) was observed in the total edible portion. The total lipids were lower (P <.05) in 'cage-free-vegetarian' eggs. However, this was not noticed in 'free-range', 'vegetarian' or 'naturally-nested-cage-free' eggs. The content of C16:0, C18:0 and total saturated fatty acids were lower (P <.05) in 'vegetarian high n-3' eggs. No difference was observed in the content of C16:1, C18:1 or total monounsaturated fatty acids. The content of n-3 fatty acids were lower (P <.05) in 'organic', 'vegetarian free-range' and 'cage-free' eggs. The ratio of total n-6:n-3 polyunsaturated fatty acids ranged from 39.2 for 'cage-free' to 11.5 for 'vegetarian high n-3' eggs (P <.05). No difference was observed in the total polyunsaturated fatty acid content of eggs (P >.05).

**Key Words:** Specialty Eggs, Lipids, Fatty Acids

## PSA Immunology

**1240 Enhanced macrophage function in broilers fed diets supplemented with *E. coli* bacterial cell powder.** G. F. Erf\*<sup>1</sup>, T. K. Bersi<sup>1</sup>, and Y. Toride<sup>2</sup>, <sup>1</sup>*University of Arkansas, Fayetteville, AR, USA*, <sup>2</sup>*Ajinomoto Co., Inc., Tokyo, Japan.*

Bacteria are important members of the gut flora and are known to play a role in innate immunity. Additionally, bacterial cell wall products such as lipopolysaccharide (LPS) and peptidoglycan are known to have immunopotentiating effects. In chickens, little information is available on immunopotentiating effects of orally administered bacterial cell wall products. This study was designed to examine and compare the effects of dietary administration of purified *E. coli* LPS and of *E. coli* bacterial cell powder (BCP) on macrophage function in broilers. Newly-hatched male broiler chicks were assigned to 8 treatment groups consisting of standard diet (control), standard diet supplemented with 1, 10, 100, or 1000 ppm *E. coli* BCP, or standard diet supplemented with 0.034, 0.34, or 3.4 ppm *E. coli* LPS. When the broilers were 3 to 4 weeks of age, Sephadex-elicited abdominal exudate cells (macrophages) were collected. Macrophages from at least 10 birds per diet were then cultured with or without *in vitro* LPS stimulation to assess oxidative radical production (oxidation of 2',7'-dichlorofluorescein diacetate), nitric oxide

production (nitrite assay), and tumoricidal activity (% killing of RP9 tumor cells). Dietary *E. coli* BCP administration did not affect oxidative radical production by macrophages, but did increase macrophage nitric oxide production and tumoricidal activity compared to controls. Similarly, addition of purified *E. coli* LPS to the diet had no effect on oxidative radical production. Macrophages from broilers fed 0.034 ppm *E. coli* LPS exhibited enhanced nitric oxide production and tumoricidal activity, however, these enhancing effects of dietary *E. coli* LPS were masked when macrophages were further stimulated with LPS in culture. Although both *E. coli* products enhanced macrophage function in young broilers, the immunopotentiating effects of *E. coli* BCP were more consistent than those of pure *E. coli* LPS.

**Key Words:** Broiler macrophage, Lipopolysaccharide, *E. coli* bacterial cell powder

**1241 Enhanced macrophage function in broilers fed diets supplemented with digested bacterial cell powder prepared from *Brevibacterium lactofermentum*.** T. K. Bersi\*<sup>1</sup>, B. B. Madison<sup>1</sup>, M. K. Redhorse<sup>1</sup>, Y. Toride<sup>2</sup>, and G. F. Erf<sup>1</sup>, <sup>1</sup>University of Arkansas, Fayetteville, AR, USA, <sup>2</sup>Ajinomoto Co., Inc., Tokyo, Japan.

Bacterial cell wall products such as lipopolysaccharide (LPS) and peptidoglycan are known to have immunopotentiating effects. In chickens, little information is available on immunopotentiating effects of orally administered bacterial cell wall products. This study was designed to examine and compare the effects of dietary administration of purified muramyl dipeptide (MDP) and digested bacterial cell powder (DBCP) prepared from *B. lactofermentum* on macrophage function in broilers. Newly-hatched male broiler chicks were assigned to six treatment groups consisting of standard diet (control), standard diet supplemented with 10, 100, or 1000 ppm DBCP, or standard diet supplemented with 0.35 or 3.5 ppm purified MDP. When the broilers were 3 to 4 weeks of age, Sephadex-elicited abdominal exudate cells (macrophages) were collected from at least 10 birds per diet. Various aspects of macrophage function were examined *in vitro*, including adherence, phagocytosis of sheep red blood cells (SRBC), and phagocytosis of antibody-opsonized SRBC. Additionally, to assess oxidative radical production (oxidation of DCF-DA), nitric oxide production (nitrite assay), and tumoricidal activity (% killing of RP9 tumor cells), the cells were cultured with (stimulated macrophages) or without (unstimulated macrophages) LPS. Macrophages from broilers fed the DBCP diets exhibited enhanced phagocytosis of antibody-opsonized SRBC compared to controls. Additionally, dietary DBCP resulted in enhanced nitric oxide production and tumoricidal activity by unstimulated macrophages, and enhanced production of oxidative radicals and nitric oxide by stimulated macrophages. These enhancing effects of DBCP on macrophage function were observed when the diets contained 100 or 1000 ppm DBCP. Dietary administration of purified MDP did not significantly affect any of the parameters examined.

**Key Words:** Broiler macrophage, Muramyl dipeptide, Digested bacterial cell powder

**1242 The Effects of Epigallocatechin Gallate on the Avian Macrophage *In Vitro*.** Jennifer Paquette\* and Fred McCorkle, PhD., Central Michigan University, Mt. Pleasant, MI.

Epigallocatechin gallate, EGCG, is the active polyphenol in green tea. EGCG has been shown to inhibit the growth of certain tumors through the inhibition of certain transcription factors. EGCG has also been shown to promote IL-1 production in monocytes and enhance the proliferation of B-cells. In this study, the effects of EGCG on several avian macrophage functions were examined. The macrophage is a major secretory cell within the immune system and aids in the phagocytosis and destruction of invading matter. The MQ-NCSU macrophage cell line was used. The null hypothesis was that EGCG would have no effect upon the avian macrophage function. Macrophage functions tested included the adherence and the phagocytosis. All data was performed in triplicate format and was analyzed using a one-way ANOVA. The cytotoxicity of EGCG to macrophages was performed at concentrations of  $1 \times 10^{-6}$  to  $1 \times 10^{-12}$  M. EGCG had no cytotoxic effects upon the avian macrophage. The remaining assays used concentration levels of  $1 \times 10^{-8}$  to  $1 \times 10^{-12}$  M. EGCG affected the ability of macrophages to adhere to a substrate at all doses tested ( $113.11 \pm 16$  at  $1 \times 10^{-12}$  M vs.  $260 \pm 24.9$  for controls). The phagocytosis assay used sheep red blood cells (SRBC) and *Escherichia coli* (*E. coli*) as substrates. EGCG did not affect the ability of MQ-NCSU macrophages to phagocytize either substrate and did not affect the number of substrate particles taken into a macrophage ( $24.00\% \pm 2.71$  at  $1 \times 10^{-8}$  M vs.  $23.89\% \pm 2.31$  for controls). EGCG does affect the ability of MQ-NCSU macrophages to adhere but is not cytotoxic at the concentrations tested and does not affect phagocytosis of SRBC or *E. coli*.

**Key Words:** Epigallocatechin Gallate, Macrophage

**1243 The *in vitro* effects of Caffeic Acid Phenethyl Ester, the active component of Bee Propolis, on the avian macrophage.** Tricia Anscomb\*<sup>1</sup> and McCorkle Fred<sup>1</sup>, Central Michigan University, Mt. Pleasant, MI.

Interest in nutritional supplements has increased within the past decade. This includes increased interest in products from the common honeybee, *Apis* species. Dose ranges of caffeic acid phenethyl ester (CAPE), the active component of bee propolis, are unclear and few studies have determined the effects of CAPE on the immune system. The macrophage is one of the main cells of the immune system. This study determined the effects of CAPE on the avian macrophage line MQ-NCSU *in vitro*. MQ-NCSU is a well-established and studied macrophage line. The effects of CAPE on cytotoxicity, adherence, and phagocytosis were tested. Using the trypan blue exclusion test, CAPE was not cytotoxic to avian macrophages from  $1 \times 10^{-3}$  to  $1 \times 10^{-12}$  M. The average macrophage viability for controls and CAPE concentrations was 92%. CAPE did not have any effect on macrophage adherence (controls  $128 \pm 10$  adhered cells versus  $1 \times 10^{-7}$  M CAPE  $133 \pm 8$  adhered cells). The effect of CAPE on the ability of macrophages to phagocytize *Escherichia coli* was determined. The number of macrophages undergoing phagocytosis of *E. coli* was not statistically different ( $38 \pm 2$  macrophages). At  $1 \times 10^{-10}$  M CAPE statistically lowered the number of *E. coli* taken up per phagocytizing macrophage ( $1.6 \pm .07$  *E. coli* versus  $2.2 \pm .25$  *E. coli* for controls). All statistics were analyzed using a one-way ANOVA and performed in triplicate for reproducibility. This study suggests that CAPE is not toxic to the avian macrophage and does not inhibit macrophage adherence to substrate or phagocytosis of *E. coli*, at the concentrations tested. The results suggest that human consumption of bee products with CAPE does not harm macrophage functions.

**Key Words:** Caffeic Acid Phenethyl Ester, Macrophage, Immune system

**1244 Pulmonary hypertensive response to endotoxin and immune activity in primed and unprimed broiler chickens.** W. Wang\*, R. F. Wideman, and G. F. Erf, University of Arkansas, Fayetteville, AR, USA.

Pilot studies in broiler chickens indicated that the magnitude of pulmonary hypertensive responses to intravenous endotoxin varied widely among individual birds, which might contribute to their variable susceptibilities to pulmonary hypertension syndrome (PHS, ascites). The purpose of this study was to determine whether immunologically primed broilers have more consistent and enhanced pulmonary hypertensive responses to intravenous endotoxin injections than controls. For this study, birds were primed using a particulate immunological stimulus known to result in an increase of lymphocyte aggregates in the lungs. The pulmonary and peripheral arterial pressures prior to and following intravenous administration of *Salmonella typhimurium* endotoxin were examined in primed and unprimed broilers when the birds were between four and five weeks of age. Additionally, the number of lymphocyte aggregates in the lungs, as well as, the proportions and concentrations of circulating white blood cells were assessed in primed and unprimed broilers. Results showed that the primed group resembled the control group in their pulmonary hypertensive responses to endotoxin in terms of time of onset, magnitude, duration, as well as the variability in response among individual birds. The respiratory rate was higher in primed than in control broilers. The concentration of white blood cells was similar in both groups, whereas the percentage of eosinophils in the blood was higher in primed broilers compared to controls ( $P < 0.05$ ). Lung tissue weight decreased ( $P = 0.056$ ) in the primed birds within 48 hours after priming. The right ventricle to total ventricle weight ratio was similar for the two groups. In conclusion, priming reduced lung weight and increased the density of pulmonary lymphocyte aggregates but did not affect the magnitude or consistency of the subsequent pulmonary hypertensive response to intravenous endotoxin challenge.

**Key Words:** Broiler leukocytes, Endotoxin, Pulmonary hypertension

**1245 Humoral Immunity Against Newcastle Disease Virus in Broilers fed *S. cerevisiae* cell wall and aflatoxin.** Elizabeth Santin\*<sup>1</sup>, A.C Paulillo<sup>1</sup>, E.L. Krabbe<sup>1</sup>, A. Maiorka<sup>1</sup>, and M. Macari<sup>1</sup>, <sup>1</sup>FCAV - Universidade Estadual Paulista.

This study aimed to evaluate the effect of aflatoxin in the diet, in the presence or absence of cellular wall of *Saccharomyces cerevisiae* (CWSC) on the humoral immune response of broilers vaccinated and challenged

against Newcastle disease virus (NDV). The exposure of 320 broilers to 1 ppm of aflatoxin reduced their humoral immune response in broiler vaccinated against NDV ( $P < 0.05$ ), and the CWSC ameliorated this parameter but did not significant ( $P > 0.05$ ). All groups reacted to antigenic stimulation to NDV at 11 days of age, but the best hemoagglutination inhibiting (HI) antibodies titers was obtained in birds not exposed to aflatoxin (GMT 7.75, 7.50, 6.37 and 5.75 to the birds fed without aflatoxin and CWSC; birds fed CWSC; birds fed aflatoxin and CWSC and birds fed only aflatoxin, respectively). When these birds were challenged with the velogenic strains of VDN, all the birds exposed at aflatoxin died, but in the birds fed with aflatoxin plus CWSC it was observed 50% of protection against VDN, demonstrating the deleterious effects of this mycotoxin in the immune response of the broilers vaccinated against Newcastle disease, and although the CWSC did not show significant improvement in HI antibodies titers, the cell wall of *S. cerevisiae* ameliorated the immune response of the birds challenged with VDN. FAPESP:Proc. n. 99/12952-7

**Key Words:** Aflatoxin, Immunity, *S. cerevisiae*

**1246 *In Vitro* or *In Vivo* Effects of Recombinant Turkey Interferon Gamma (rtIFN $\gamma$ ) on *Eimeria* Invasion or Infection.** R Beltran<sup>\*1</sup>, P Augustine<sup>2</sup>, M El Halawani<sup>3</sup>, H Danforth<sup>2</sup>, A McElroy<sup>4</sup>, and D Caldwell<sup>1</sup>, <sup>1</sup>Texas A&M University, College Station, TX, <sup>2</sup>USDA/ARS/LPSI/PBEL, Beltsville, MD, <sup>3</sup>University of Minnesota, St. Paul, MN, <sup>4</sup>Virginia Tech, Blacksburg, VA.

IFN $\gamma$  is integral to many parameters of cellular immunity and promotes the barrier nature of mucosal membranes to enteric pathogens in mammals. Our interest in gut immunity in commercial poultry led to our investigation of the *in vitro* and *in vivo* effects of rtIFN $\gamma$  on pathophysiology and immunity to avian coccidia. *In vitro* experiments consisted of exposing baby hamster kidney cells (BHK) to increasing concentrations of rtIFN $\gamma$  and measuring subsequent *E. tenella* (ET) sporozoite invasion in short-term culture. When BHK were pretreated with 0, 2.5, 25, 250, 500, or 1000 ng rtIFN $\gamma$  per ml of cell culture medium 45 min prior to the addition of the sporozoites to confluent monolayers, ET invasion was significantly reduced ( $P < .05$ ) in BHK exposed to concentrations of 25 ng rtIFN $\gamma$  and higher. In a subsequent experiment, when BHK were pretreated with the same concentrations of rtIFN $\gamma$  for 48 hours prior to sporozoite addition, significant differences in cellular invasion were not observed. *In vivo* investigation consisted of administering rtIFN $\gamma$  to day-of-hatch turkey poults by intraperitoneal injection 30 min prior to *per os* challenge with *E. adenoeides* (EA) and measuring effects on cecal lesion development and body weight gain 6 days post EA challenge. When poults received rtIFN $\gamma$  at concentrations of 0.25, 2.5, or 12.5  $\mu\text{g/poult}$ , cecal lesions associated with EA challenge were significantly reduced ( $P < .05$ ). Body weight gains of poults receiving rtIFN $\gamma$  and EA were not statistically different from that of poults administered EA alone. Further, administration of rtIFN $\gamma$  at 12.5  $\mu\text{g/poult}$  and EA resulted in body weight gains not statistically different from that of poults receiving only rtIFN $\gamma$ . These data, while preliminary, suggest the potential involvement of IFN $\gamma$  in the development of immunity to avian coccidia in commercial poultry.

**Key Words:** IFN $\gamma$ , Coccidial Immunity

**1248 Croos reactivity determination for Salmonella enteritidis biovar issatschenko and Salmonella gallinarum using LT antibodies in immunoblot technique.** O. Urquiza<sup>\*1</sup>, G. Tellez<sup>1</sup>, L. Paasch<sup>1</sup>, G. Ruiz-Palacios<sup>2</sup>, and B. Diaz<sup>2</sup>, <sup>1</sup>Departamento de Produccion Animal Aves, FMVZ, UNAM, <sup>2</sup>Departamento de Infectologia e Investigacion del instituto nacional de nutricion (INNSZ).

This study was carried out to determine by Immunoblot, cross reactivity among supernatant proteins (SP) and periplasmic proteins (PP) of *Salmonella enteritidis* biovar. Issatschenko against CT and LT antibodies and *Salmonella enteritidis* biovar. Issatschenko SP and PP antibodies produced in rabbit and *Salmonella gallinarum* total antibodies produced in chicken. Results obtained showed that *Salmonella enteritidis* biovar. Issatschenko after a 24 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  $\times$  Biotin and Peroxidasa conjugates, reveled homologous and cross reactivity more intensive with anti LT, anti SP and anti PP of

*Salmonella enteritidis* biovar. Issatschenko antibodies and anti Sg of *S. gallinarum* against *Salmonella enteritidis* biovar. Issatschenko SP and PP and PS and PP of the same *Salmonella* but produced a year ago. 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, utilizing 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 reveled 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 *Salmonella gallinarum* PP could discriminate different protein bands by Immunoblot whom are visible by the use of total Sg antibodies. That discrimination could be used as a tool for differentiation among some salmonelas.

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

**1249 The Interleukin-1 $\beta$  sequence of Japanese quail (*Coturnix coturnix japonica*) and Mallard ducks (*Anas platyrhynchos*).** B.D. Humphrey<sup>\*</sup>, E.A. Koutsos, and K.C. Klasing, University of California, Davis, Davis, CA.

The Interleukin-1 $\beta$  (IL-1 $\beta$ ) DNA sequence of Japanese quail and Mallard ducks was determined. This cytokine is produced in response to inflammatory agents such as gram-negative bacteria, and induces localized responses including tissue destruction and activation of lymphocytes and vascular endothelial cells. In addition, systemic effects of IL-1 $\beta$  include induction of fever and the hepatic acute phase response. To stimulate production of IL-1 $\beta$ , Japanese quail were injected with 7.5 mg lipopolysaccharide (LPS; isolated from *Salmonella typhimurium*)/kg body weight, and ducks were injected with 10<sup>6</sup> fixed *E. coli* (wild type K12). Two hours after inoculation, spleen samples were taken from both quail and duck and analyzed for IL-1 $\beta$  mRNA levels using RT-PCR procedure. PCR conditions for IL-1 $\beta$  were based on previous work in our lab, and the PCR primer encompassed nucleotides 37 through 828 from the published chicken IL-1 $\beta$  sequence (GenBank #Y15006). PCR products were isolated, purified, and the nucleotide sequence was determined. The quail and duck nucleotide sequences were analyzed for sequence homology using a protein-nucleotide database search program (BLAST). The isolated nucleotide sequence (735 nucleotides) from the spleen of Japanese quail was determined to be 93% homologous to chicken IL-1 $\beta$ . The nucleotide sequence isolated from the spleen of Mallard ducks (637 nucleotides) was found to be 85% homologous to chicken IL-1 $\beta$ . Isolation of the complete IL-1 $\beta$  sequence is required to assess phylogenetic differences between chicken, quail and duck IL-1 $\beta$  structure. However, it seems that the pro-inflammatory cytokine cascade functions in a similar manner in these avian species, as IL-1 $\beta$  mRNA was induced and a systemic acute phase response occurred in Japanese quail and Mallard ducks as in chickens.

**Key Words:** IL-1 $\beta$ , Japanese quail, Mallard duck

**1250 Initiation of Humoral Immunity: The Role of Cytokines and Hormones in the Initiation of Humoral Immunity Using T-Independent and T-Dependent Antigens.** A.E. Gehad<sup>1</sup>, H.S. Lillehoj<sup>2</sup>, G.L. Hendricks III<sup>3</sup>, and M.M. Mashaly<sup>\*3</sup>, <sup>1</sup>Virginia Commonwealth University, Richmond, VA/USA, <sup>2</sup>USDA-ARS, Beltsville, MD/USA, <sup>3</sup>The Pennsylvania State University, University Park, PA/USA.

The purpose of the present study was to investigate the role of different cytokines and hormones in the initiation of humoral immunity. Immature Cornell K-strain male chickens were injected i.v. with 8mg/kg BW of lipopolysaccharide (LPS) from *Escherichia coli*, a T-independent antigen or with 40mg/kg BW BSA, a T-dependent antigen. Control birds were injected with 0.9% saline. Blood and spleen were collected 0, 1, 3, 6, and 24 h following the injections. The blood was centrifuged and the plasma was collected. Plasma samples were assayed for Interleukin-1 (IL-1) like activity using the thymocyte co-mitogenesis assay following the precipitation of inhibitory factors with polyethylene glycol. Tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) like activity was measured as cytotoxic activity against the L929 mouse fibroblast cell line. The plasma was also assayed for corticosterone and T<sub>3</sub> by RIA. Interleukin-2 (IL-2) activity was measured in the conditioned medium from splenic lymphocytes following their stimulation with the T-cell mitogen Concanavalin-A (Con-A) for 24 h. Interleukin-1 activity was increased significantly at 1, 3,

and 6 h post-LPS injection, whereas there was no change in IL-1 activity at any time postinjection in the BSA-injected birds. TNF- $\alpha$  activity significantly increased at 6 and 24 h post-LPS injection, whereas there was no difference in the TNF- $\alpha$  activity at any time postinjection in the BSA-injected birds. Interleukin-2 activity was decreased significantly at 3 h post-LPS injection compared to base line levels at 0 h. However, IL-2 activity increased at 3 h post-BSA injection compared to 3 h post saline injection. Corticosterone levels significantly increased 1, 3, and 6 h post-LPS injection, whereas there was no change in corticosterone levels at any time postinjection in the BSA-injected birds compared to saline injected birds. Tri-iodothyronine levels significantly decreased 3, 6, and 24 h post-LPS injection, whereas there was no change in T<sub>3</sub> levels at any time postinjection in the BSA-injected birds compared to saline injected birds. The results indicate that although LPS and BSA injection can induce a humoral antibody response in chickens, the mechanism of the initiation of this antibody response involving the activation of the cytokine network and the neuroendocrine system are different for each antigen.

**Key Words:** Chickens, Antigen, Cytokines and Hormones

**1251 Control of coccidiosis in chicken by trickle immunisation.** Srinivasan K<sup>1</sup>, Dilipkumar Garikipati\*<sup>2</sup>, and Venkaram A<sup>2</sup>, <sup>1</sup>Madras Vety College, <sup>2</sup>College of Vety Sci, Tirupati.

Immunisation control of coccidiosis caused by *Eimeria tenella* was carried out by trickle immunisation (TI) method. Immunizing chickens daily with 100 number of oocysts at 1-7 days of age (experiment I), 1-14 days (experiment II), and 8-14 days (experiment III) of age. The corresponding doses were also administered at 1 and 8 days as double dose and at 8 days as single dose in experiment II and III, respectively. Chickens were challenged with 50,000 oocysts at 28 days of age and were sacrificed 7 days after injection. Highly significant increase in ceecal length was observed in experiment I and II, whereas no significant difference was noted in experiment III when compared to control. Trickle-immunized chickens showed lesser lesion score, decreased faecal oocyst output, and ceecal oocyst contents when compared to control. Further trickle immunized groups showed higher immunizing response than their corresponding single or double dosed groups against challenge infection. The results suggested that trickle immunization of chickens continuously

for 1-14 days of age conferred better protection than trickle immunization for 1-7 days or 8-14 days of age.

**Key Words:** coccidia, chicken, trickle immunisation

**1252 gamma-Interferon and IL-2 activities in supernatant of lymphocytes on chicken splenocytes stimulated with concanavalin A.** G. Gomez\*<sup>1</sup>, G. Tellez<sup>1</sup>, A. Isibasi<sup>3</sup>, and V. Ortiz<sup>2</sup>, <sup>1</sup>Departamento de Produccion Animal Aves, FMVZ, UNAM, <sup>2</sup>Departamento de Biomedicina Molecular del CINVESTAV del IPN, <sup>3</sup>Unidad de Investigacion Medica en Inmunoquimica del hosp. de especialidades del centro medico nacion.

Previous studies have shown a protection for the experimental challenge against infections resulting from *Salmonella gallinarum* and *Salmonella enteritidis* in chickens, through the manipulation of birds' immunocompetent system by the use of lymphokines (ILK) raw extract obtained from T lymphocytes of hyper-immunized birds with *Salmonella gallinarum* (ILK-Sg) or *Salmonella enteritidis* (ILK-Se), and re-stimulated with concanavalin A. In order to identify interleukines present in the ILK, the presence of interleukine-2 (IL-2) and Interferon was assessed. The results obtained show the presence of both interleukines in ILKSe. The chicken IL-2, as in other mammals, participates in the proliferation of T lymphocytes; however, a significant difference compared to other species is that there was a species barrier in its biological activity. The INF induced an increase in the expression of class I and class II molecules of the Major Histocompatibility Complex (MHC). The presence of INF in the supernatant, suggests that this interleukine could be participating in the protection effect which is conferred by the administration of ILK, encouraging macrophages and heterophils activation, which could be responsible for the bacteria elimination. Besides, these studies offer more evidences indicating that the observed immunity in the in vivo studies against *Salmonella gallinarum* and *Salmonella enteritidis* is probably induced by these important lymphokines. Studies are in progress to purify and identify the effector lymphokines genes in chickens, as well as a systemic research regarding the mechanism by means of which these lymphokines potentialized their protective effects.

**Key Words:** interleukines, interferon-gamma, interleukine-2

## ADSA Dairy Foods: Microbiology and Cheese Technology

**1253 Flavor development of cholesterol-reduced Cheddar cheese slurries.** H. S. Kwak\*<sup>1</sup>, C. S. Chung<sup>1</sup>, S. J. Lee<sup>1</sup>, and J. Ahn<sup>1</sup>, <sup>1</sup>Sejong University.

This study was carried out to find the development of flavor in cholesterol-reduced Cheddar cheese slurries by  $\beta$ -cyclodextrin ( $\beta$ -CD). The cheeses were made by 3 different treatments as followings: 1) control (no homogenization, no  $\beta$ -CD treatment), 2) Treatment A (1000psi homogenization, 10% -CD), and 3) Treatment B (cream separation, 10%  $\beta$ -CD). The cheese slurry was aged for 3 wk. The cholesterol removal was 79.30% (Treatment A) and 91.22% (Treatment B). Among 8 volatile flavor compounds, dimethylsulfide, 2-pentanone and 2-heptanone appeared to be insignificantly increased during 3 wk storage, and no difference was found among treatments as expected. The amounts of acetone and ethylacetate were slightly increased in control at 3 wk, however, no difference was found in others. Ethanol production was dramatically increase at 1 wk and decreased thereafter in all treatments. Most volatile flavor compounds were not significantly different among control and treatment A and B as expected. Based on our results, cheese slurry made by cheese milk after cream separation (36% milk fat) and 10%  $\beta$ -CD treatment showed a highest cholesterol removal. Therefore, this study provided a possibility of cholesterol-reduced Cheddar cheese manufacture without any flavor change.

**Key Words:** Low cholesterol Cheddar cheese, Flavor, Beta cyclodextrin

**1254 Dynamic headspace analysis and sensory characteristics of ewes milk La Serena cheese.** Mara Carbonell, Estrella Fernandez-Garcia\*, and Manuel Nunez, Instituto Nacional de Investigacion y Tecnologia Agraria y Alimentaria (INIA).

La Serena cheese is a semi-soft variety, made in Extremadura (Spain) from raw milk of Merino ewes, coagulated with vegetable rennet. The objective of our study was to investigate the composition of the volatile

fraction of La Serena cheese, and its correlation with sensory characteristics. Duplicate batches of La Serena cheese were made at 4 dairies during the 4 seasonal periods. Cheese (60-day-old) samples were homogenized with sodium sulfate and an internal standard. An aliquot was extracted in a purge and trap apparatus. Volatiles were concentrated in a Tenax trap at 30C, and desorbed with He during 1 min at 230C directly into the injection port. Gas chromatography was carried out in a HP-6890 apparatus equipped with a HP Innowax capillary column and a HP 5973 mass spectrometer. Peaks were identified by comparison of retention times and ion spectra with those of real standards and of Wiley 275 library spectra. Quantitation was carried out by sum of characteristic ions abundance. Cheeses were scored by 14 trained panelists for quality and intensity of odor and aroma, on a 7-point scale. Volatile compounds found in the headspace of 60-day-old La Serena cheeses were 13 aldehydes, 8 ketones, 24 alcohols, 24 esters, 10 hydrocarbons, 14 terpenes, 3 sulfur compounds, 3 nitrogen compounds, 7 aromatic compounds and 5 free fatty acids. Most compounds were detected in cheeses made in all seasonal periods, and higher concentrations of most volatiles found in spring cheeses. Some terpenes were only detected in winter and spring cheeses. Analysis of variance showed significant seasonal differences of quality and intensity of cheese odor and aroma. Spring cheeses obtained the best quality scores, whereas winter cheeses had a more intense aroma. Odor intensity correlated positively with 2-butanone and 3-methyl-1-butanol, and negatively with terpenes, ethyl esters and quality of odor and aroma. A high concentration of 2-butanone was responsible for low quality scores, but high levels of esters resulted in improved quality scores even in the presence of high 2-butanone concentrations, as shown by Principal Component Analysis. Stepwise discriminant analysis of selected volatile compounds achieved