291P Initial assembly of the turkey whole genome sequence. R. A. Dalloul*[1], O. Folkert[1], E. J. Smith[1], K. M. Reed[2], O. Crasta[1], A. P. McElroy[1], R. A. Coulombe[3], E. A. Wong[1], J. B. Dodgson[4], and D. W. Burt[5], [1]*Virginia Polytechnic Institute and State University*, *Blacksburg*, [2]*University of Minnesota*, *St. Paul*, [3]*Utah State University*, *Logan*, [4]*Michigan State University*, *East Lansing*, [5]*Roslin Institute*, *Midlothian, United Kingdom*.

A community-driven consortium has initiated sequencing the genome of the domesticated turkey, *Meleagris gallopavo*. Turkey meat is currently the fourth protein choice for American consumers with estimated US production of 271 million birds raised in 2008. In addition to the economic impact, the turkey serves as a model organism for a number of metabolic and medical diseases. The Turkey Genome Sequencing Consortium comprised of US and international scientists has used the latest 454 sequencing technology during this initial phase, with other sequencing platforms to be employed soon. The DNA for sequencing was isolated from a female turkey (NT-WF06-2002-E0010, referred to as “Nici” (Nicholas inbred)). Nici is from an inbred sub-line (sibmating for nine generations) originally derived from a commercially significant breeding line, with 88% monomorphism based on SNP genotyping. Roche/454 GS-Titanium sequencing at Virginia Tech has already produced more than 5x random and paired-end genome coverage (> 8 billion bases sequenced). The latest sequence assembly contains ~880 million base pairs in 428,910 large contigs, with average size of ~2 kb. The expected outcome of the fully sequenced (99% coverage) turkey genome is a rich genomic resource suitable for future academic and industrial, basic and applied poultry research. It will also provide solid foundation for the development of species-specific SNP panels for genome-based selection and improvement, and comparative genomics in poultry and other avian species.

Key Words: turkey, genome, *Meleagris gallopavo*, 454 sequencing

Poster Session: Immunology

292P WITHDRAWN.

293P Immunobiological effects of three phytonutrients, carvacrol, cinnamaldehyde, and capsicum oleoresin on chicken cells cultured *In vitro*. D. K. Kim*[1], H. S. Lillehoj[1], S. H. Lee[1], S. I. Jang[1], C. Ionescu[2], and D. Bravo[2], [1]*Animal Parasitic Diseases Laboratory, Animal and Natural Resources Institute, Beltsville Agricultural Research Center, United States Department of Agriculture, Agricultural Research Service*, *Beltsville, MD*, [2]*Pancosma S. A., Research Department/ Nutrition & Technology, Voie des Traz 6*, *Le Grand-Saconnex, Switzerland*.

The present study was conducted to investigate the effects of three different plant-derived phytonutrients, carvacrol, cinnamaldehyde and capsicum oleoresin, on innate immune responses and tumor cell growth. To evaluate their effects, lymphocyte proliferation and the growth rate of tumor cell were accessed using a non-radioactive CCK-8 assay, and nitric oxide production was also measured using an *in vitro* culture treated with the three phytonutrients (carvacrol, cinnamaldehyde and capsicum oleoresin). Quantitative real-time RT-PCR was performed to measure the transcriptional expression of cytokine genes in macrophages in response to treatment with each phytonutrient. Results showed that each phytonutrient induced significant proliferation of spleen lymphocytes compared with the untreated control, and all stimulated robust nitric oxide production to the levels similar to that induced by recombinant chicken interferon-γ. All phytonutrients inhibited the growth of chicken tumor cells in a dose-dependent manner. The levels of mRNAs encoding IL-15 and IL-18 were enhanced when macrophages were treated with cinnamaldehyde. Capsicum oleoresin provoked the high expression of IL-18 and TNFSF15. The genes of IFN-α, IL-1b, IL-6 and IL-12 were not significantly influenced by the treatment of cinnamaldehyde or capsicum. These results suggest that these three phytonutrients, carvacrol, cinnamaldehyde and capsicum oleoresin, enhance host innate immune system in chicken.

Key Words: carvacrol, cinnamaldehyde, capsicum oleoresin, innate immunity, chicken

294P Ethanol-induced changes in oxidative stress and immunological parameters of the chicken, *Gallus gallus*. H. Deng*[1], X. Guan[1], K. B. Gyenai[1], J. Xu[1], R. Dalloul[1], R. M. Gogal[2], R. E. Pearson[3], and E. J. Smith[1], [1]*Virginia Polytechnic Institute and State University, Department of Animal and Poultry Science*, *Blacksburg*, [2]*Virginia Polytechnic Institute and State University, Department of Biomedical Science and Pathobiology*, *Blacksburg*, [3]*Virginia Polytechnic Institute and State University, Department of Dairy Science*, *Blacksburg*.

Oxidative stress is believed to be responsible for many diseases and physiological abnormalities in animals. However, the potential effect of oxidative stress on the immune system of chicken has not been investigated. Using ethanol as an inducer of oxidative stress, the main objective of this study was to examine the possible association between oxidative stress and immunosuppression in chickens. To evaluate these relationships, 4-week-old White Leghorn chickens were randomly divided into 4 groups of 24 birds each, and provided ad libitum starter diet and drinking water containing 0, 2, 6, and 10% ethanol for 2 weeks. As oxidative stress increased, plasma IgG but not IgM decreased. Similarly, the weights of the major immune organs, thymus, spleen and bursa, were inversely correlated with oxidative stress. The data presented here suggest that supplementation of drinking water with ethanol at 2% level is optimum for enhancing immunocompetence in chickens, while higher levels of ethanol increased oxidative stress and reduced immunocompetence.

Key Words: chicken, ethanol, oxidative stress, antibody response, organ weight

295P Effects of dietary β-glucan on immune-related gene expression of broiler chicks. C. M. Cox*[1], L. H. Stuard[1], S. Kim[1], A. P. McElroy[1], M. Bedford[2], and R. A. Dalloul[1], [1]*Virginia Polytechnic Institute and State University*, *Blacksburg*, [2]*AB Vista Feed Ingredients*, *Marlborough, United Kingdom*.

During the first week post hatch, the avian immune system is immature and inefficient at protecting chicks from invading pathogens. Among immunomodulators, β-glucans are known as biological response modifiers due to their ability to activate the immune system. Current research suggests that β-glucans may enhance avian immunity; however, very little is known about their influence on regulation of immune function. A pilot study was performed to evaluate the effects of dietary β-glucan on growth performance and immune-related gene expression in the intestine. Day old chicks were fed a diet containing 0, 0.02, or 0.1% yeast β-glucan. On days 1, 7 and 14 post-hatch, body weight was measured and small intestinal sections were collected to evaluate gene expression by quantitative real-time PCR. Supplementation of β-glucan did not significantly impact body weight gains. Compared to controls, expression of interleukin (IL)-8 was decreased in the duodenum of both β-glucan treated groups on d7 and d14 and in the jejunum of the 0.1% β-glucan group on d7 and the 0.02% β-glucan group on d14. On d14, 0.1% β-glucan inclusion led to increased iNOS expression in the duodenum and ileum. On d7, IL-18 expression was upregulated in the jejunum, but decreased on d14 in the duodenum of the 0.02% β-glucan birds. IL-18 expression also decreased on d14 in the ileum of both β-glucan groups when compared to control. Interestingly, though there was variation in IL-18 expression, intestinal interferon-γ expression decreased in the 0.1% β-glucan group on d7. These results suggest that β-glucans are capable of altering cytokine/chemokine levels, particularly those of the T helper type-1 cells. However, since varying levels of β-glucan result in different gene expression profiles, more research is needed to determine optimal dosage for immune modulation in various disease situations.

Key Words: β-glucan, chicken, immunity, cytokines

296P Evaluation of the inflammatory response in broilers following intra-abdominal administration of *Clostridium perfringens*. C. R. Waneck*[2,1], J. L. McReynolds[1], C. L. Swaggerty[1], K. J. Genovese[1], D. J. Nisbet[1], S. E. Duke[1], and M. H. Kogut[1], [1]*USDA-ARS-SPARC-FFSRU*, *College Station, TX*, [2]*Texas A&M University, Department of Poultry Science*, *College Station*.

Currently, not much is known about *Clostridium perfringens* (CP) and its interactions with the avian immune system. Etiologically, CP is an anaerobic Gram-positive spore-forming bacillus that is an opportunistic pathogen and causes several poultry diseases. The chicken abdominal cavity is a good model to study inflammatory cell recruitment and the immune response to bacterial pathogens *in vivo* . Typically, heterophils are the first leukocyte to migrate from the peripheral blood to the site of infection, in this case the abdominal cavity. Birds are dependent on the recruitment of heterophils to fight off infection and foreign invaders. In the present investigation (3 replicate studies) a combination of four different field isolates of CP (type A) were administered intra-abdominally at 1×10^5 CFU/0.2mL/bird for CP treated birds and a 0.2mL/bird of physiologic saline was administered to the controls birds. Standard abdominal washes were performed at 0, 1, 2, 3, 4 h to evaluate total leukocyte influx into the abdominal cavity of the bird. When evaluating the total leukocytes from the CP injected and saline controls there was a significant ($P \leq 0.05$) increase in leukocytes in the abdominal cavity of CP injected birds (1.2×10^5) at 3 h compared to saline control (2.83×10^4). Differentials were determined by counting monocytes, lymphocytes and heterophils. When evaluating the differential counts, there were significant ($P \leq 0.05$) increases between CP and saline treated birds involving heterophil recruitment at 3 h. These results suggest there is a complex immune interaction occurring in the abdominal cavity of the birds to protect them from CP involving heterophil recruitment to the site of infection. Understanding the birds innate immune response to Clostridia will help develop future research to elucidate host/CP immune interactions.

Key Words: chickens, leukocytes, heterophils, *Clostridium perfringens*

297P Determination of the effect genetic polymorphisms on subcellular localization of the chicken Mx protein. W. L. Richardson*, R. A. Ali, C. M. Ashwell, and M. D. Koci, *North Carolina State University*, *Raleigh*.

Acute viral infections are one of the leading causes of illness and death in humans and animals worldwide. Identifying and understanding how host proteins are involved in protecting cells from viral infection is critical to our ability to develop novel antiviral therapies. One host protein known to be directly involved in inhibiting viral replication is known as Mx; however the mechanism by which it blocks virus replication is not well characterized. In mammals there are typically two Mx genes, with the product of one gene found in the nucleus and the other in the cytoplasm. In mice the nuclear localized Mx (MxA) has been demonstrated to inhibit influenza virus. Further analysis of MxA demonstrated that differences in mouse strain susceptibility to influenza virus were associated with specific Mx alleles. Mx genes have also been discovered in non-mammalian species. Recent reports have identified numerous Mx alleles in various lines of chickens and related these to differences in their antiviral activity. However, how the different polymorphisms in Mx affects its function is not currently known. To begin to characterize how sequence changes affect Mx biology, we examined the effect of amino acid changes on cellular localization of the Mx protein. One allele of the chicken Mx gene was cloned into the pEGFP-N1 expression system to generate a construct which produces an Mx-enhanced green fluorescent protein (EGFP) fusion protein. This initial plasmid was then subjected to a series of site directed mutagenesis reactions to generate subsequent expression constructs encoding other Mx alleles and transfected into chicken embryo fibroblast (CEF) cells. Because localization of the Mx protein can be determined using UV-microscopy, the recombinant Mx-EGFP construct will allow us to begin to understand how various alleles of chicken Mx is related to its structure/function in cells, and how these differences may be related antiviral activity.

Key Words: Mx, antiviral

298P Effect of dietary arginine on peripheral blood mononuclear cell populations and cytokine profiles during the acute phase response in broiler chicks. J. L. D'Amato* and B. D. Humphrey, *California Polytechnic State University*, *San Luis Obispo*.

The effect of dietary arginine (ARG) levels on peripheral blood mononuclear cell (PBMC) populations and T helper (T_H) subset cytokine mRNA abundance was determined during the acute phase response (APR) in broilers. Cobb hatchlings were fed one of three diets formulated to meet or exceed NRC requirements, except for ARG. Diets contained low (1.1%), adequate (1.3%), or high (1.5%) ARG. On d19 posthatch, chicks from four replicate pens per treatment were not injected (control) or injected subcutaneously with *Salmonella* typhimurium lipopolysaccharide (LPS; 1 mg/kg BW). At 2 and 24 h post-injection, peripheral blood and spleen from one chick per pen were collected. PBMC were isolated by density centrifugation and CD4+ T cell, CD8+ T cell, B cell and monocyte populations were identified by flow cytometry. Spleens were frozen in liquid nitrogen prior to total RNA isolation, reverse transcription and quantification of T_H1-3 cytokine mRNA abundance by real-time PCR. Chicks fed 1.3% and 1.5% ARG had greater BW than chicks fed 1.1% ARG ($P<0.05$). From 0-2 h, LPS decreased feed intake by 40% compared to controls ($P<0.05$), but feed intake was similar to controls from 2-24 h post-injection ($P>0.05$). The concentration of viable PBMC was lower in chicks injected with LPS compared to controls at 2 h ($P<0.05$), but not at 24 h ($P>0.05$). At 24 h post-injection, the concentration of dead PBMC was higher in LPS injected chicks ($P<0.05$). The percentage of CD4+ T cells, CD8+ T cells, B cells and monocytes in peripheral blood did not differ due to dietary ARG ($P>0.05$). LPS decreased peripheral blood CD4+ T cells, CD8+ T cells, B cells and monocytes by 46, 52, 38, and 82%, respectively, at 2 h post-injection ($P<0.05$). The APR reduces food intake and T cell, B cell and monocyte populations in peripheral blood by 2 h post-infection, irrespective of dietary ARG levels.

Key Words: arginine, cytokine, leukocyte, lipopolysaccharide, monocyte

299P In vitro effects of plant and mushroom extracts on immunological function of lymphocytes, macrophages, and tumor cells. S-H. Lee*[1], H. Lillehoj[1], Y. Hong[1,3], D. Bravo[2], and L. Mazuranok[2], [1]*Animal and Natural Resources Institute, Agricultural Research Service-U.S. Department of Agriculture*, *Beltsville, MD*, [2]*Pancosma S.A.*, *Geneva, Switzerland*, [3]*Chung-Ang University*, *Anseong, Korea*.

The present study was conducted to examine the ability of various plants extracts (turmeric, milk thistle) and mushrooms extracts (Shiitake, Reishi) to stimulate innate immunity and kill tumor cells. Innate immunity was measured by lymphocyte proliferation and nitric oxide production. Ability of extracts to inhibit tumor cell growth was assessed using a non-radioactive CCK-8 assay. In vitro culture of chicken spleen lymphocytes with extracts of turmeric (*Curcuma longa*), milk thistle (*Silybum marianum*), shiitake (*Lentinus edodes*) and reishi (*Ganoderma lucidum*) mushrooms, induced significantly higher cell proliferation compared with the untreated control cells. Stimulation of macrophages with extracts of milk thistle, shiitake and reishi mushrooms, but not turmeric, stimulated robust nitric oxide production to levels similar to that induced by recombinant chicken interferon-γ. All extracts uniformly inhibited the growth of chicken tumor cells in vitro. Finally, the levels of mRNAs encoding IL-1β, IL-6, IL-18, and TNFSF15 were enhanced when macrophages were treated with the extracts of turmeric or shiitake mushroom compared with the untreated control. These results show that the extracts examined (turmeric, milk thistle, shiitake and reishi) enhanced innate immunity in chickens.

Key Words: plant, mushroom, lymphocytes, macrophages, tumor cells

300P Gene expression profiling difference between resistant and susceptible broilers responding to *Campylobacter jejuni* infection. X. Li[1], C. Swaggerty[2], M. Kogut[2], H. Chiang[1], Y. Wang[1], K. Genovese[2], H. He[2], I. Pevzner[3], and H. Zhou*[1], [1]*Texas A&M University*, *College Station*, [2]*United States Department of Agriculture, Agricultural Research Service*, *College Station, TX*, [3]*Cobb-Vantress, Inc*, *Siloam Springs, AR*.

Campylobacter jejuni (*C. jejuni*) is one of the most common pathogens causing human gastroenteritis worldwide. Chickens, as a natural reservoir of *C. jejuni*, are a good animal model to study the molecular mechanism of a hosts' response to *C. jejuni* infection. Previous studies showed there was a significant difference in *C. jejuni* colonization between two distinct parental lines (A and B) and that there was considerable variation within line. To elucidate the molecular mechanisms of genetic resistance to *C. jejuni* colonization, the Agilent chicken 44K microarray was used to profile gene expression. Day-old chickens from lines A and B were challenged orally with *C. jejuni*. Cecal content and the cecal tonsil were collected for colonization quantification and total RNA isolation 7 days post-challenge, respectively. Twenty samples with the highest (S) or lowest bacterial number (R) in cecal content within each line were randomly pooled into four biological replicates. The pair comparisons between the two groups (R and S) within each line (A and B) were analyzed. The signal intensity of each gene was normalized using LOWESS method. A mixed model was used to identify differentially expressed genes by SAS ($P < 0.01$). There were 564 and 6105 genes differentially expressed between groups in the comparison of AS/AR and BS/BR, respectively, and 1565 and 2488 genes between the two lines (AS/BS, AR/BR, respectively). However, there were 18 and 62 immune-related genes differentially expressed in AS/AR and BS/BR, respectively, while only two immune-related genes were shared between AS/AR and BS/BR. The results showed that a significantly different response to *C. jejuni* infection occurred not only between different genetic lines but also between resistant and susceptible chickens within line, and the individual contribution should be considered when evaluating genetic resistance.

Key Words: broiler, *C. jejuni*, microarray, gene expression

301P Innate immune response to *Campylobacter jejuni* infection in the broiler bursa. J. Hilley[1], X. Li[1], C. Swaggerty[2], H. Chiang[1], Y. Wang[1], K. Genovese[2], H. He[2], V. DiRita[3], I. Pevzner[4], and H. Zhou*[1], [1]*Texas A&M University*, *College Station*, [2]*United States Department of Agriculture, Agricultural Research Service*, *College Station, TX*, [3]*University of Michigan*, *Ann Arbor*, [4]*Cobb-Vantress, Inc.*, *Siloam Springs, AR*.

Campylobacter jejuni (*C. jejuni*) is one of most common causes of food-borne illness worldwide. Chickens are a major reservoir for *C. jejuni*. To identify the differentially expressed genes between chickens infected with wild-type (wt) *C. jejuni* and non-infected chickens, a broiler line susceptible to *C. jejuni* infection was challenged with wt *C. jejuni* or PBS at the day of hatch. Total RNA was isolated from bursa harvested at 1 and 4 hours post-inoculation. Eight biological replicates were used in infected and non-infected groups at each time point. The signal intensity of each gene was normalized using LOWESS method. A mixed model was used to identify differentially expressed genes by SAS program ($P < 0.01$). There were 1016 and 275 genes significantly differentially expressed between infected and non-infected chickens at 1 and 4 hours post-challenge, respectively. The finding of more differentially expressed genes at 1 hour than 4 hours post-challenge between infected and non-infected chickens was compared to previous results observed in a line more resistant to *C. jejuni*. There were 2591 and 2936 genes significantly changed between 1 and 4 hours post-challenge in infected and non-infected chickens, respectively. The results show there was a strong early host response to wt *C. jejuni* in this susceptible chicken line. To further characterize the molecular mechanisms of the host innate immune response to *C. jejuni* infection in broilers, gene expression profiling at additional time-points post-challenge is underway in our laboratory.

Key Words: broiler, *C. jejuni*, innate immune response, microarray, bursa

302P Downregulation of liver expressed antimicrobial peptide (LEAP-2) mRNA following coccidiosis challenge. S. Casterlow*[1], H. Li[1], A. P. McElory[1], R. A. Dalloul[1], D. A. Emmerson[2], and E. A. Wong[1], [1]*Virginia Polytechnic Institute and State University*, *Blacksburg*, [2]*Aviagen Inc.*, *Huntsville, AL*.

Avian coccidiosis is a major disease of poultry caused by the intestinal protozoa *Eimeria*, which results in reduced feed efficiency and body weight gain. The objective of this study was to compare gene expression in Aviagen line A and line B birds that show differential susceptibility to *Eimeria* infection. Line B chicks showed higher lesion scores and higher mortality compared to line A chicks when orally challenged with 1 x 10^4 *Eimeria maxima* oocysts/bird. In the challenged line A chicks, there appeared to be two populations: one group with lesion scores of 1 (on a scale of 0-4) similar to controls and a second group with high lesion scores (3-4). DNA microarray analysis was performed on jejunal tissues from Line A and B control chicks and low (0-1) and high (2-4) lesion score challenged chicks. Liver expressed antimicrobial peptide 2 (LEAP-2) was downregulated by 20-fold in challenged A chicks (scores 3-4) vs. control A, by 11 to 17-fold in challenged B chicks (scores 2-3) vs. control B, and by 37 to 71-fold in challenged B chicks (score 4) vs. control B. LEAP-2 is a cationic antimicrobial peptide that is part of the innate immune system and provides an essential role in defense against infection. DNA Microarray results of LEAP-2 expression was verified using real time PCR and a similar pattern of downregulation was observed. LEAP-2 was downregulated greater in line B chicks compared to line A chicks. Also, expression of LEAP-2 also appeared to differ between males and females. Despite individual variation, there was an association between lesion score and magnitude of LEAP-2 downregulation, with the greatest downregulation observed in chicks with lesion scores of 4. These results suggest that levels of LEAP-2 expression may be used as an indicator for identifying chicks with resistance or susceptibility to coccidiosis.

Key Words: chicken, LEAP-2, coccidiosis, *Eimeria maxima*, microarray

303P Development of a CXCL8 (IL-8) bioassay to determine its role in turkey poult viral enteritis. R. A. Ali* and M. D. Koci, *North Carolina State University*, *Raleigh*.

Turkey astrovirus type-2 (TAstV-2) was originally identified and associated with poult enteritis and mortality syndrome (PEMS), but has since been recognized to cause severe enteritis in turkey poults throughout the Americas and Europe. In spite of its broad distribution and impact on turkey production we know little about TAstV-2 pathogenesis or the host immune response to it. Studies done in our laboratory have demonstrated TAstV-2 infection induces severe watery diarrhea and growth suppression. Analysis of infected intestines demonstrated no histological change or signs of inflammation despite increased gene expression of the pro-inflammatory chemokine CXCL8 (interleukin-8, IL-8). CXCL8 is recognized as a potent chemoattractant signal for heterophils and is often recognized as one of the first innate immune molecules secreted by infected tissues to elicit cells of the innate immune system to initiate the host response. However, following TAstV-2-induced CXCL8 expression, there is no influx of immune cells to the site of TAstV-2 replication. In addition to its chemotaxis properties, it has been suggested that CXCL8 have angiogenic properties and may play a role in inducing proliferation of intestinal epithelia cells. To better understand the role of CXCL8 following TAstV-2 infection, our laboratory developed a CXCL8 bioassay using the mouse pre-B cell line L1.2 expressing a recombinant chicken CXCR1 and recombinant turkey CXCL8 as a positive control. This new assay will be an important tool in our ability to determine the various functions CXCL8 expression in the intestine and specifically its role in the innate immune response to virally induced poult enteritis.

Key Words: TAstV-2, PEMS, IL-8, CXCL8, CXCR1

304P Mass spectrometric identification and measurement of avian beta defensin 2. N. C. Rath*[1], L. Kannan[1,2], R. Liyanage[3], and J. O. Lay, Jr.[3], [1]*USDA/ARS*, *Fayetteville, AR*, [2]*University of Arkansas, Department Poultry Science*, *Fayetteville*, [3]*University of Arkansas, Statewide Mass Spectrometry Lab.*, *Fayetteville*.

Beta defensins are small, cationic peptides that bind to microbial membranes and disrupt their integrity thereby exerting the antimicrobial effects. These peptides play important role in innate immunity

defending against microbial pathogens. Using matrix assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF MS), we found the chicken heterophils teemed with avian beta defensin 2 (AvBD2) isoform. The heterophils play a crucial role in innate immunity defending against microbial pathogens. Because heterophils undergo activation through their membrane bound "Toll-like receptors (TLR)" that recognize "pathogen associated molecules," we wanted to find whether TLR ligands stimulate AvBD2 production. We cultured chicken heterophils with different TLR ligands representing both bacterial and viral motifs that included lipopolysacharides, peptidoglycans, flagellin, loxoribine, CpG-ODN, and poly I: C for 6 hours, and extracted the conditioned media for measurement of AvBD2. To measure the changes in AvBD2 production, aliquots of each extract containing AvBD2 (*m/z* 3916) was reduced and alkylated, resulting in the carbamidomethylated 6 cysteine residues and a mass shift to *m/z* 4264. Considering that the chemical modification and treatments likely affect spectral intensities, correction factors (CF) for each treatment groups were calculated using the intensity ratios of AvBD2 in 1:1 mixture of nonreduced (*nr*) and reduced (*r*) aliquots of each sample by MALDI-TOF MS. The reduced extracts from individual treatments (*Tr*) were then mixed with non reduced control aliquots (C*nr*) at 1:1 ratio (with respect to protein content) and then subjected to MALDI-TOF MS as above. The T*r*/C*nr* ratio was multiplied by an appropriate correction factor and compared with similarly calculated control (C*r*/C*nr**CF). The results showed that most pathogen mimetics (except CpG-ODN) were potent inducers of AvBD2. In conclusion, these results show that MALDI-TOF MS is able to identify mature AvBD peptides and is also able to to take advantage of their chemistry to modify and measure changes in their production in response to different stimulants.

Key Words: heterophil, beta defensin, mass spectrometry

305P Identification of potential target genes of the novel chicken-specific miRNA, *miR-NC-57*. A. Whisnant*, J. Hicks, and H. C. Liu, *North Carolina State University*, *Raleigh*.

MicroRNA (miRNA), a class of small (~19-24nt) regulatory RNA, function in post-transcriptional regulation of gene expression. It has been suggested that up to 30% of genes are regulated by miRNA. MiRNA function by knocking down the expression of target genes by binding to complementary sites in the targeted mRNA. These target sites often lie within the 3′ untranslated region. MiRNA expression profiles have revealed that hundreds of dynamically expressed miRNA are encoded in the eukaryotic genome. Many miRNAs are conserved across species and it is likely that these conserved miRNAs function similarly. However, it also appears that each genome also encodes a unique set of miRNAs that possess species-specific function(s). We recently used the deep sequencing approach to profile miRNA expressed in the developing chick embryo. This profile identified a group novel and likely chicken-specific miRNA, of which the miRNA *miR-NC-57* is a member. To further understand the involvement of *miR-NC-57* in chick development, the present study was undertaken to identify its potential target genes. Potential *miR-NC-57* target genes were identified using the miRNA target prediction algorithm miRanda. These potential target genes were then validated using a retroviral-based RNA interference (RNAi) strategy. *miR-NC-57* was found to potentially target two members (B-cell translocation gene 2 and B-cell translocation gene 4) of the BTG/Tob gene family. This family consists of structurally related proteins that are involved in regulating cell proliferation by inducing G1 phase arrest during the cell cycle. This suggests that *miR-NC-57* may function to allow increased cell proliferation during chick embryonic development by targeting anti-proliferative genes.

Key Words: microRNA, embryonic development, gene regulation, RCAS, immune system

Poster Session: Metabolism and Nutrition

306P Effect of *Saccharomyces cerevisiae* fermentation product on immune function of broilers challenged with *Eimeria tenella*. J. Gao[1], H. J. Zhang[1], S. G. Wu[1], S. H. Yu[2], I. Yoon[3], D. Moore[3], Y. P. Gao[2], H. J. Yan[1], and G. H. Qi*[1,2], [1]*Feed Research Institute, Chinese Academy of Agricultural Sciences & Key Laboratory of Feed Biotechnology, Ministry of Agriculture*, *Beijing, China*, [2]*College of Animal Science and Technology, Northwest A & F University*, *Yangling, Shaanxi, China*, [3]*Diamond V Mills Inc.*, *Cedar Rapids, IA*.

The purpose of this study was to compare various levels of a *Saccharomyces cerevisiae* fermentation product with and without a coccidian challenge. Three hundred and sixty day-old male Arbor Acres broilers were randomly allotted to 6 groups with 2 X 3 factorial arrangement. Three supplemental levels (0, 0.25 and 0.50%) of *Saccharomyces cerevisiae* fermentation product (Diamond V XP™) were fed to control and *Eimeria tenella* infected broilers. Each treatment consisted of 6 replicates of 10 birds each. Growth performance and immune response criteria were measured after coccidian infection. Broiler ADG and feed conversion were lowered ($P < 0.01$) by coccidian infection. The ADG was improved by XP supplementation ($P < 0.05$) during d 21 to 42 for both coccidia infected and non-infected birds. Supplementation of XP increased CD3+, CD4+, and CD8+ T lymphocyte content ($P < 0.05$) and ratio of CD4+/CD8+ in blood ($P = 0.06$) and spleen ($P = 0.04$) as well as ileum intraepithelial lymphocytes count, cecal tonsil sIgA content, serum lysozyme content ($P < 0.01$), serum IgM content and albumin to globulin ratio ($P = 0.02$). The above immune indexes exhibited a dose response to the XP supplemental level, especially for the coccidian infected birds. When XP supplementation increased in the diet of coccidia-infected broilers, CD3+, CD4+ in blood and spleen, ileum intraepithelial lymphocytes count, cecal tonsil sIgA content, serum lysozyme content increased linearly ($P < 0.01$), and CD8+ quadratically ($P < 0.01$). These results suggest that dietary XP supplementation could improve immune function and growth performance in coccidia-infected broilers.

Key Words: *Saccharomyces cerevisiae* fermentation product, broiler, immune function, coccidiosis