

AAAP/AVMA Scientific Program



Washington, D.C.
July 14-18, 2007



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Continued from page 4.

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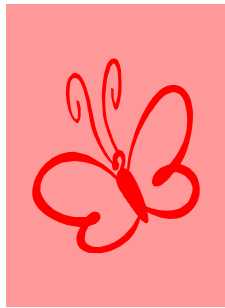
Tuesday, July 17, 2007 – 5:00 til 11:00 PM

AAAP's 50th Anniversary & Awards Banquet

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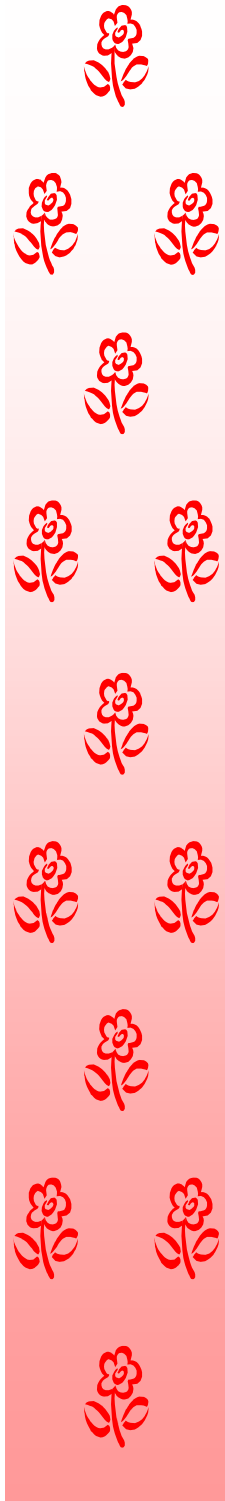
for sponsoring the

**AAAP Welcome Reception
at the**

**Grand Hyatt Hotel
Independence A Room**

**on Saturday evening,
July 14, 2007
5:00 – 11:00 pm**

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Poster Presenters – VERY IMPORTANT -

Posters must be set up before **7:00 AM on Sunday, July 15th** and removed promptly at **12:00 PM on Wednesday, July 18th.**

Presenters should be available each morning and during scheduled breaks in the scientific program to discuss their posters.

SPECIAL PRESENTATIONS

Monday, July 16, 2007

8:00 AM

Keynote Speaker: John Glisson

“The Future of Veterinary Medicine and Poultry Production”

Tuesday, July 17, 2007

10:00 AM

Lasher History Lecture: Richard Witter

“Origin and Evolution of the American Association of Avian Pathologists: 50 Years of Dedication and Accomplishment”

1:00 PM

Reed Rumsey Award Presentation: M. F. Abdul-Careem

“Cellular and Cytokine Responses Associated with Dinitrofluorobenzene-induced Contact Hypersensitivity in Chicken”

Tuesday, July 18, 2007

10:00 AM

Richard Rimler Memorial Paper: H. M. Yassine

“Interspecies Transmission: Host Range Determinants of Swine H3N2 Influenza A Virus Transmission from Pigs to Turkeys”



AAAP Schedule of Events – Grand Hyatt (Washington, D.C.)

Date / Time	Meeting Name	Room Assignments
Friday, July 13, 2007		
6:45 am – 5:00 pm	AAAP Board of Directors Meeting	Latrobe
Saturday, July 14, 2007		
6:45 am – 12:00 pm	AAAP Board of Directors Meeting	Latrobe
1:00 pm – 5:00 pm	AAAP Foundation Board Meeting	Latrobe
7:00 am – 8:00 pm	ACPV Exam	Roosevelt/Wilson
7:00 am – 8:00 pm	ACPV Exam # 2	Cabin John
7:00 am – 10:00 pm	ACPV Exam # 3 – (Exam Grading Room)	Arlington
8:00 am – 5:00 pm	Association of Veterinarians in Broiler Practice	(Breakfast-Renwick) Constitution C (Lunch-Renwick)
12:00 pm – 5:00 pm	Association of Veterinarians in Turkey Production	Arlington
1:00 pm – 4:00 pm	AAAP-CAST	Constitution E
2:00 pm – 4:00 pm	AAAP Membership Committee	Potomac
2:30 pm – 5:00 pm	AAAP Histopathology/ Case Report Interest Group	Conference Theater
3:00 pm – 4:00 pm	AAAP Education Committee	Bulfinch
3:00 pm – 4:30 pm	AAAP Biologics Committee	Washington Boardroom
3:00 pm – 5:00 pm	AAAP Poultry Research Priorities Committee	Renwick
3:30 pm – 5:30 pm	AAAP Avian Diseases Editorial Board Meeting	Constitution A
4:00 pm – 5:00 pm	AAAP Electronic Information Committee	Potomac
6:00 pm – 10:00 pm	AAAP – Intervet Reception	Independence A
Sunday, July 15, 2007		
7:00 am – 8:00 am	AAAP Awards Committee	Potomac
7:00 am – 8:30 am	Georgia MAM Alumni Breakfast	Constitution CD
11:30 am – 1:30 pm	Association of Primary Poultry Breeder Veterinarians Luncheon	Arlington/Cabin John
12:00 pm – 1:00 pm	AAAP Drugs & Therapeutic Committee	Renwick
12:00 pm – 1:30 pm	California Poultry Medicine Alumni	Constitution E
2:00 pm – 6:00 pm	ACPV Board Meeting	Renwick
3:00 pm – 4:00 pm	AAAP-AVMA Liaison Committee	Arlington
3:00 pm – 4:00 pm	AAAP Epidemiology Committee	Washington Boardroom
3:00 pm – 4:30 pm	AAAP Food Safety Committee	Cabin John
3:15 pm – 4:30 pm	AAAP History Committee	Potomac
3:15 pm – 5:15 pm	AAAP Animal Welfare Committee	Burnham
3:15 pm – 5:15 pm	AAAP Respiratory Diseases Committee	Constitution A
4:00 pm – 5:00 pm	AAAP Biotechnology Committee	Constitution E
4:00 pm – 5:00 pm	AAAP Diseases of Poultry Committee	Arlington
4:00 pm – 5:30 pm	AAAP Enteric Diseases of Poultry Committee	Roosevelt
4:00 pm – 5:00 pm	AAAP Toxic, Infectious, Miscellaneous & Emerging Diseases (TIME) Committee	Washington Boardroom
4:00 pm – 5:00 pm	AAAP Tumor Virus Committee	Wilson
7:30 pm – 11:00 pm	NC State University Poultry Health Management	Constitution CD
Monday, July 16, 2007		
7:00 am – 8:00 am	AAAP Eskelund Preceptorship Committee	Arlington
7:00 am – 9:00 am	Association of Veterinarians in Egg Production	Burnham
3:00 pm – 4:00 pm	AAAP Task Force on Future of AAAP	Washington Boardroom
Tuesday, July 17, 2007		
7:00 am – 9:00 am	ACPV Reception / Annual Meeting	Constitution AB
10:30 am – Noon	AAAP Business Meeting	(Convention Center) Room 207
5:00 pm – 11:00 pm	AAAP 50th Anniversary & Awards Banquet	Independence A
Wednesday, July 18, 2007		
6:45 am – Noon	AAAP Board of Directors Meeting	Farragut Square



American Association of Avian Pathologists
July 14-18, 2007
Washington DC Convention Center
Washington, D.C.



SCIENTIFIC PROGRAM

Monday, July 16, 2007 – Morning Program		
	Moderator: A. G. Rosales	
8:00 AM	Keynote Speaker: John R. Glisson Room 207A <p style="text-align: center; color: red;">The Future of Veterinary Medicine and Poultry Production</p>	
	Moderator: Stewart Ritchie Room 207A	Moderator: Kenton Kreager Room 207B
8:30 AM	Poultry House of the Future Mark A. Dekich and Jeannine M. Harter-Dennis	Use of Molecularly Cloned Avian Leukosis Virus to Study Antigenic Variation Following Infection of Meat-Type Chickens Aly M. Fadly, Jody K. Mays, R. F. Silva, and H. D. Hunt
8:45 AM	Welfare Audits: Present and Future Needs of the Poultry Industry Ken Opengart	Diphtheritic Tumorous Growths Caused by Fowlpox Virus Containing REV Provirus Deoki N. Tripathy
9:00 AM	Annual Antibiotic Consumption in a Commercial Turkey Operation Eric Gonder and Becky Tilley	Molecular Characterization of Three Isolates of Avian Leukosis Virus Isolated From Contaminated Marek's Diseases Vaccines Taylor Barbosa, Guillermo Zavala, and Sunny Cheng
9:15 AM	Use of 4-H Project Turkeys as Serological Sentinels in North Carolina David V. Rives and Summer M. Russell	Control of Contemporary Marek's Disease Isolates Robin W. Morgan, Grace Isaacs, Mark Parcells, Milos Markis, Amy S. Anderson, Erin L. Bernberg, Sandra Cloud Rosenberger, and John K. Rosenberger
	Break 9:30 AM – 10:00AM	Break 9:30 AM – 10:00AM
	Moderator: Ken Opengart	Moderator: Susan Williams
10:00 AM	Teaching Poultry Necropsy Techniques to Afghan Veterinary Students Richard M. Fulton, Ezatullah Jaheed, and Robert M. Smith	Pathological and Viral Characterization of a Case of Splenomegaly in F Line Turkeys: Is HVT Able to Produce Disease? Devorah A. Marks, Aneg Lucia Cortes, John Barnes, Oscar J. Fletcher, and Isabel M. Gimeno
10:15 AM	Pre-Peak Lay Broiler Breeders with Impaired Mobility and High Mortality Associated with Myopathy and Hypocalcemia Valerie Rochelle Bullock, Michael P. Martin, Michael J. Wineland, Oscar J. Fletcher, and H. John Barnes	The Effect of Vaccination on Marek's Disease Virus Shedding Robert F. Silva, Abd A. El-Galil El-Gohary, and John Dunn
10:30 AM	Use of the I-Stat Serum Chemistry Analyzer for Evaluation of Flock Health in Pre-Peak Lay Broiler Breeders Michael P. Martin, Valerie R. Bullock, Michael J. Wineland, Oscar J. Fletcher, and H. John Barnes	A Comparison Between Marek's Disease Virus Production in Feather Pulp Versus Feather Follicle Epithelium (FFE) John R. Dunn, James B. Finlay, Richard L. Witter, and Robert F. Silva

Monday, July 16, 2007 – Morning Program		
	Room 207A	Room 207B
10.45 AM	Incubation Conditions and Leg Health in Large High Yield Broilers Edgar O. Oviedo-Rondón, Mike Wineland, Sarah Funderburk, Heather Cutchin, John Small, and Mike Mann	Effect of Vaccination on the Development of Marek's Disease Virus-Induced Eye Lesions Arun K.R. Pandiri, Aneq Lucia Cortes, and Isabel M. Gimeno
11:00 AM	Vertebral Canal Stenosis as a Cause of Lameness in Broilers: Additional Findings Kelli Jones, H. John Barnes, Kenneth C. Powell, and Suzanne Y. Stamey	Relationship Between Load of Marek's Disease Vaccine DNA in Peripheral Blood and Level of Protection Aneq Lucia Cortes and Isabel M. Gimeno
11:15 AM	State Quarantines of Respiratory Disease Found at Auction: Lessons Learned Donna J. Kelly	Pathogenesis of Serotype 1 Marek's Disease Vaccines in the Lung Isabel M. Gimeno and Aneq L. Cortes
11:30 AM	Chromium Associated Discoloration of Egg Yolks in a Backyard Flock H. M. Opitz, Tiffany A. Wilson, Bruce R. Hoskins, Dawna J. Beane, and Xi Wang	Recombinant Marek' disease virus lacking the oncogene Meq as a candidate for future control of Marek's disease in chickens Lucy F. Lee, Sanjay Reddy and Kenton Kreager
11:45 AM	Differential Diagnosis of Liver Hemorrhage in Laying Hens Craig Riddell	Functions of Marek's Disease Virus MicroRNAs Robin W. Morgan, Amy S. Anderson, Erin L. Bernberg, Emily Huang, Grace Isaacs, Milos Markis, and Joan Burnside
LUNCH NOON – 1:00 pm		
Monday, July 16, 2007 – Afternoon Program		
	Moderator: Hector Cervantes	Moderator: Richard Fulton
1:00 PM	Field Experiences in Commercial Broilers with an <i>In Ovo</i> Coccidiosis Vaccine Andrea J. Sinclair, David Kelly, Rebecca Poston, Cherilyn Heggen-Peay, Larry Charniga, Lynn Murray, and Vivian Doelling	Evaluation of Different Hemagglutinin-Neuraminidase (HN) Chimeras Using a Newly Developed Reverse Genetic System Based on the Mesogenic Anhinga Newcastle Disease Virus Strain Carlos Estevez, Qingzhong Yu, Daniel King, and Patti Miller
1:15 PM	Eimeria Shedding Patterns Under Various Environmental and Coccidiosis Management Programs Linnea J. Newman and Greg F. Mathis	Newcastle Disease Virus Isolates from US Waterfowl Reveal Novel Genomic Subgroups with Diverse and Evolving Genetics L. Mia Kim, Daniel J. King, David E. Swayne, David Stallknecht, Richard D. Slemons, Janice C. Pedersen, Dennis A. Senne, Kevin Winker, and Claudio L. Afonso
1:30 PM	Comparison of <i>Eimeria</i> Species Composition Between High-Performance and Low-Performance Broiler Operations Mark C. Jenkins, Spangler Klopp, Donald Ritter, and Katrzyna Miska	Presence of Newcastle Disease Virus and Avian Influenza Virus in Wild Birds at the Peruvian Coast Armando Gonzalez, Eliana Icochea, Rosa Gonzalez, Bruno Gherzi, and David Blazes
1:45 PM	Analysis of Gene Expression in the Intracellular Stages of the Poultry Parasite, <i>Eimeria acervulina</i> Katarzyna B. Miska and Raymond H. Fetterer	Isolation of Exotic Newcastle Disease Virus (ENDV) from Field Collected Flies and Experimental ENDV Infections of Three Arthropod Species Daniel J. King, Seemanti Chakrabarti, Claudio L. Afonso, Alec C. Geery, Carol J. Cardona, and David E. Swayne

Monday, July 16, 2007 – Afternoon Program		
	Room 207A	Room 207B
2:00 PM	Characterization of Novel Lytic Peptide Secreted by Intestinal Intraepithelial Lymphocytes Infected with Coccidia Hyun S. Lillehoj, Sung Hyun Lee, Doug Bannerman, and Yeong H. Hong	The Macroscopic and Microscopic Pathology Associated with Outbreaks of Exotic Newcastle Disease in Commercial Poultry, and Pigeon Paramyxovirus Infection in Laughing Doves (<i>Streptopelia Senegalensis</i>) in South Africa Neil M. Duncan and Emily P. Lane
2:15 PM	The Use of Chlorine Dioxide as a Disinfectant in a Broiler Hatchery Mark Burlleson, Phil Stayer, John Smith, and Joel Tenney	A “Poult Enteritis Mortality Syndrome” in Guinea fowl: a Pathological and Etiological Study Jean-Luc Guerin, Bertrand Grenier, Cyril Boissieu, and Caroline Lacroux
	Break 2:30 PM – 3:00PM	Break 2:30 PM – 3:00PM
	Moderator: Suzanne Young Stamey	Moderator: Kate Barger
3:00 PM	Enumerating Salmonella in Poultry Samples using a DNA Sequence Capture Combined with Q-PCR Randall S. Singer and Janet M. Anderson	Newcastle Disease Virus Vaccine Potency Determination Karen A. Lijebjelke, Darrell R. Kapczynski, and Daniel J. King
3:15 PM	Is Salmonella Enterica Kentucky a Newly Emergent, Poultry Adapted Strain? John J. Maurer, Adriana Pedroso, Margie D. Lee, Holly Sellers, Erich Linnemann, and Katherine Zamperini	Avian Adeno-Associated Virus-Based Expression of the Newcastle Disease Virus HN Protein for Poultry Vaccination Francisco Perozo, Pedro Villegas, Carlos Estevez, Ivan Alvarado, and Linda Purvis
3:30 PM	Discrimination Between Strains of <i>Salmonella</i> Enteritidis PT13 and PT13a by Comparative Genome Sequencing of the Virulence Plasmid Jean Guard-Bouldin, Cesar Morales, Adam B. Olson, and Matthew W. Gilmour	Development of a Quantitative Light Cycler Real-Time PCR for the Detection of Fowl Adenoviruses Éva Nagy, Nadya Romanova, and Juan Carlos Corredor
3:45 PM	Crop Isolated Lymphoid Follicles and Ileal Peyer’s Patches in Egg-Layer Hens Challenged with <i>Salmonella</i> Enterica Enteritidis Lara E. Vaughn, Peter S. Holt, Randle W. Moore, and Richard K. Gast	Multiple Expressions of Inclusion Body Hepatitis P. A. Stayer, C. Gabriel Senties, and John El-Attrache
4:00 PM	Oligodeoxynucleotides Containing CpG Motifs (CpG-ODN) Confer Protection Against a Lethal Challenge of <i>Salmonella typhimurium</i> Septicemia in Neonatal Broilers Susantha Gomis, Azita Taghavi, Lorne Babiuk, Andrew Potter, Brenda Allan, George Mutwiri, and Andrew Van Kessel	Physiochemical and Molecular Characterization of Adenovirus-Like Virus (Isolate R11/3), The Putative Etiology of Transmissible Viral Proventriculitis James S. Guy, John Barnes, and Fred Fuller
4:15 PM	Construction and Evaluation of Recombinant <i>Salmonella</i> Vaccines Expressing <i>Eimeria</i> Antigen SO7 Vjollca Konjufca, Maria-Dolores Rodrigues, Mark Jenkins, Shifeng Wang, and Roy Curtiss III	Histopathology of Transmissible Viral Proventriculus Caused by Adenovirus-Like Virus (Isolate R11/3) H. John Barnes and James S. Guy
4:30 PM	Effects of RofenAid® Impact on Salmonella Enumeration in the Intestine and Litter and on Performance of Male Broiler Chickens after a Challenge with Salmonella and a Mixed Culture of Coccidia Sharon Heins Miller, Charles Hofacre, and Greg F. Mathis	Effect of Maternal Antibodies on Vaccine-Induced Protection of Turkey Poults Against Virulent Avian Metapneumovirus Subtype C Challenge Binu Velayudhan, Sally Noll, Sagar Goyal, David Halvorson, and Kakambi Nagaraja
4:45 PM	ADJOURN	ADJOURN

Tuesday, July 17, 2007 – Morning Program		
	Room 207A	Room 207B
	Moderator: Timothy Cummings	Moderator: Alejandro Banda
8:00 AM	Molecular Characterization of Reoviruses Isolated from Broilers with Runting and Stunting Syndrome Reveals a Lack of Homogeneity with Current U.S. Vaccine Viruses Holly S. Sellers, Veronica Walker, Erich G. Linnemann, and Guillermo Zavala	IFN-A Regulates Infectious Bursal Disease Virus Induced Macrophage Activation Mahesh Khatri and Jagdev M. Sharma
8:15 AM	Extensive Analysis of Pathogenicity Island PAI 1_{APEC O1} of Avian Pathogenic <i>Escherichia coli</i> Subhashinie Kariyawasam, Lisa K. Nolan, and Timothy J. Johnson	Cell-Mediated Immunity Induced by a Macrophage-Adapted Infectious Bursal Disease Virus Jagdev M. Sharma and Mahesh Khatri
8:30 AM	Analysis of the Complete Genome Sequence of an Avian Pathogenic <i>Escherichia coli</i> Timothy J. Johnson, Subhashinie Kariyawasam, Yvonne Wannemuehler, Paul Mangiamele, Sara J. Johnson, Curt Koetkott, Jerod A. Skyberg, Aaron M. Lynne, James E. Johnson, and Lisa K. Nolan	Identification of Infectious Bursal Diseases Viruses from Several Countries Pedro Villegas, Linda Purvis, Francisco Perozo, and Taylor Barbosa
8:45 AM	Evaluation of Differentially Expressed Proteins Following Serum Exposure in Avian Pathogenic <i>Escherichia coli</i> Steven L. Foley, Cynthia Tyler, and Cheryl Lichti	Impact of IBD Vaccines on the Bursa and Other Immune Tissues of Commercial Broilers Enrique Montiel, Nikki Pritchard, and Julio Cruz-Coy
9:00 AM	Role of Large Plasmids in the Virulence of Avian Pathogenic <i>Escherichia coli</i> Melha Mellata and Roy Curtiss III	Challenge Studies with Infectious Bursal Disease and Chicken Anemia Virus Field Strains Linda B. Purvis, Pedro Villegas, John Smith, and Francisco Perozo
9:15 AM	<i>E. coli</i> Challenge Study in Commercial Broilers by Either Respiratory or Skin Route of Exposure and the Effect of Prior Vaccination With a Live Attenuated (aro-A) <i>E. coli</i> Kalen Cookson and Steve Davis	Efficacy of a Vectorized Commercial Vaccine Against Infectious Bursal Disease in Layers Eliana Icochea, Blanca Talavera, Elmer Dávila, Rosa Gonzalez, John Guzman, and Hermelinda Rivera
	Break 9:30 AM – 10:00AM	Break 9:30 AM – 10:00AM
	Moderator: Richard Chin	
	Lasher History Lecture: 10:00 – 10:30 AM Room 207A SPEAKER: Richard Witter Origin and Evolution of the American Association of Avian Pathologists: 50 Years of Dedication and Accomplishment	
	AAAP Business Meeting 10:30 – 12:00 noon Room 207A	
	Lunch 12:00 noon – 1:00 PM	

Tuesday, July 17, 2007 – Afternoon Program		
	Room 207A	Room 207B
	Moderator: David Ley	Moderator: Peter Woolcock
1:00 PM	A Challenging Case of Suspected <i>Mycoplasma gallisepticum</i> Infection in Broiler Breeders Frederic J. Hoerr, Lanqing Li, Samuel Christenberry, and C. Stephen Roney	REED RUMSEY AWARD: Cellular and Cytokine Responses Associated with Dinitrofluorobenzene-Induced Contact Hypersensitivity in Chicken M.F. Abdul-Careem, D. Bruce Hunter, Melissa D. Lambourne, Hamid R. Haghighi, Niroshan Thanthrige-Don, and Shayan Sharif
1:15 PM	Epidemiology of <i>Mycoplasma synoviae</i> Isolates in Georgia from 2000-Present V. A. Laibinis, S. H. Kleven, Louise Dufour-Zavala, and Guillermo Zavala	Serum and Yolk Antibody Titers in Late Stage Broiler Embryos Alan Avakian, Donald Link, and John Dickson
1:30 PM	The Evaluation of <i>Mycoplasma gallisepticum</i> Challenge Routes Naola Ferguson-Noel, Victoria A. Laibinis, Ziv Raviv, Ruth S. Wooten, and Stanley H. Kleven	Validation of a Multiplexed Fluorometric Immunoassay™ (MFIA™) for Health Monitoring of Specific Pathogen Free Chickens Joe H. Simmons, Elena Seletskaja, Theodore Girshik, Rajeev K. Dhawan, and William R. Shek
1:45 PM	Intraspecific Differentiating Real-Time PCR for <i>Mycoplasma gallisepticum</i> Live Vaccine Evaluation Stanley H. Kleven, Ziv Raviv, Scott A. Callison, and N. Ferguson-Noel	Bursal Disease-Marek's Disease Vaccine, Live Marek's Disease Virus Vector, Serotype 3, (VAXXITEK™) Vaccination Effect on NDV Vaccination Schedules in Commercial Broiler Type Birds Rafael Fernandez, Mike McCabe, Francisco Rojo, and Julio S. Cruz-Coy
2:00 PM	Effects of TS-11 Strain <i>Mycoplasma Gallisepticum</i> Vaccination in Broiler Breeders Infection and Immunity on the Breeders and Its Progeny Francisco J. Rojo, Alejandro Rojas, and Rafael J. Fernandez	Antibody-Antigen Complexes Protect Against Chicken Infectious Anemia Virus Infection Karel A. Schat, Priscilla H. O'Connell, and Michael S. Piepenbrink
2:15 PM	Effect of Selected Water Temperatures Used in <i>Mycoplasma gallisepticum</i> Vaccine Reconstitution/Dilution on Color Change Units at Selected Time Intervals Scott L. Branton, Spencer A. Leigh, William B. Roush, Jody L. Purswell, Hammed O. Olanrewaju, and Stephanie D. Collier	The Effect of Vaccination Against Chicken Anemia Virus on Colonization, Shedding and Susceptibility to Infection with <i>Salmonella typhimurium</i> in Broilers Franz Sommer and Carol J. Cardona
2:30 PM	Use of the Microdilution Method for the Detection of the In Vitro Antimicrobial Susceptibility Against Avian Mycoplasmas Ariel M. Ortiz, Ernesto P. Soto, and Clemente F. Lemus	A Study of the Viral Shed of Laryngotracheitis in Cornish Birds at the Processing Plant During a Regional Outbreak and Area Wide LT Vaccination Bret Rings and Steve Breeding
2:45 PM	Antibiotic Resistance in <i>Mycoplasma gallisepticum</i> and <i>Mycoplasma synoviae</i> Isolates from Meat-Type Turkeys in Israel Sharon Levisohn, Irina Gerchman, and Shimon Perk	Challenge Study to Evaluate Vaccine Protection against Infectious Laryngotracheitis Virus (ILTV) Andrés Rodríguez and Maricarmen García
3:00 PM	ADJOURN	ADJOURN

Scientific program will adjourn at 3:00 PM on Tuesday in order to allow time for participants to attend the

AAAP 50th Celebration Banquet and the AAAP Awards Dinner at:

Grand Hyatt Hotel – Independence A Room -- 5:00 PM

Wednesday, July 18, 2007 – Morning Program		
	Room 207A	Room 207B
	Moderator: Kalen Cookson	Moderator: Erica Spackman
8:00 AM	Vertically Transmitted Viral Arthritis (Reovirus) in Commercial Broilers: A Field Case Report Suzanne Young Stamey, Calvin Anthony, Kevin Crider, and Todd Cartwright	Characteristics of a Novel Infectious Bronchitis Virus Isolates from Delmarva Broiler Chickens Jack Gelb
8:15 AM	An Outbreak Of Tenosynovitis in Broilers Caused by Vertically-Transmitted Reovirus J. J. Courtney, M. M. E. Andersen, G. Zavala, H. S. Sellers, and S. M. Williams	Infectious Bronchitis Viruses Isolated in California 2004-2006 Peter R. Woolcock and Carol J. Cardona
8:30 AM	Unusual Case: Hemochromatosis Due to Lead Poisoning in a Wild Mallard Duck Scott D. Fitzgerald, Jessica S. Hoane, and Thomas M. Cooley	Pathogenesis of Infectious Bronchitis Virus in the Fully Functional Oviduct of Unvaccinated Laying Hens Kapil Chousalkar and Juliet R. Roberts
8:45 AM	Vitamin E Deficiency Induced Cardiomyopathy in Commercial Pekin Ducks Richard M. Fulton	The Dynamics of Spray Vaccination for IBV in Commercial Broilers Mark W. Jackwood, Deborah A. Hilt, and Enid T. Mckinley
9:00 AM	Field Report: Acute Sodium Toxicosis in Turkey Poults Charles Corsiglia, Rocio Crespo, Richard Chin, H. L. Shivaprasad, Murugan Subbiah, Birgit Puschner, and Arthur Bickford	Rapid Selection in Chickens of a Subpopulation Within an Attenuated Infectious Bronchitis Virus Vaccine Vicky L. Van Santen, Haroldo Toro, and Kellye S. Joiner
9:15 AM	Effect of Novel Intestinal Anaerobes on Early Development of the Broiler Intestine Margie D. Lee, Brett Lumpkins, Youngiae Cho, and Amy Batal	Risk Factors Associated with the Prevalence of Airsacculitis in Broiler Chickens in Quebec, Canada Rachid Ankouche, Diane Brodeur, Martine Boulianne, and Jean-Pierre Vaillancourt,
	Break 9:30 AM – 10:00AM	Break 9:30 AM – 10:00AM
	Moderator: Scott Fitzgerald	Moderator: Nathaniel Tablante
10:00 AM	<i>Clostridium perfringens</i> and <i>Clostridium septicum</i> in Commercial Turkeys – Recent Findings Daniel Karunakaran	RICHARD RIMLER PAPER AWARD: Interspecies Transmission: Host Range Determinants of Swine H3N2 Influenza A Virus Transmission from Pigs to Turkeys H. M. Yassine, C-W. Lee, and Y. M. Saif
10:15 AM	Comparison of Level of Necrotic Enteritis in Broilers Vaccinated with Either an Attenuated or Non-Attenuated Live Coccidial Vaccine and Challenged with <i>Clostridium perfringens</i> Greg F. Mathis, Charles Hofacre, Jerry Chapman, and Elleen Katigbak	RCA-Free Recombinant Adenovirus-Vectored Vaccine for Mass Immunization of Poultry Against Avian Influenza H. Toro, D. C. Tang, F. Van Ginkel, and Z. Shi
10:30 AM	Innate Immune Response to <i>Clostridium perfringens</i> and <i>Eimeria maxima</i> in necrotic enteritis Model Hyun S. Lillehoj, Soon S. Park, Patricia C. Allen, Dong Woon Park, Steve Fitz-Coy, and Daniel A. Bautista	Differential Growth of Avian Influenza Virus in Chicken and Duck Cells Luciana Sarmiento, Kristin Zaffuto, Mary Pantin-Jackwood, and Claudio Afonso

Wednesday, July 18, 2007 – Morning Program		
10:45 AM	Attempts to Develop an Oral Challenge Model for Gangrenous Dermatitis using <i>Eimeria maxima</i> and <i>Clostridium septicemia</i> Michelle M. Early-Andersen, Jeffery J. Courtney, and Steve R. Collett	Efficacy of Three Inactivated Vaccines Against Challenge with HPAI H5N1 Vietnam/05 Viruses in Ducks Mary J. Pantin-Jackwood, David L. Suarez, Jennifer Pfeiffer, and Luciana Sarmento
11:00 AM	Development of an Experimental Challenge Model for Cellulitis in Turkeys Anil J. Thachil, Binu T. Velayudhan, David A. Halvorson, and Kakambi V. Nagaraja	Comparison of In Vivo Innate Immune Responses in Lung and Spleen Tissue Following Infection with Asian H5N1 Avian Influenza Viruses in Ducks and Chickens Darrell R. Kapczynski and Mary Pantin-Jackwood
11:15 AM	Field Investigation of a Mycoplasma Outbreak in the Southeastern United States Guillermo Zavala, Michelle Andersen, Louise Dufour-Zavala, Victoria Leiting, and Stan Kleven	Surveillance, Testing and Epidemiology of Avian Influenza in the New York Live Bird Market System Susan C. Trock, Michelle Gaeta, Lisa Weisse, Tara Howard, Kimberley Tropea, Adam Holloway, Sung Kim, J. Beeby, Jan Pederson, Dennis Senne, and Edward Dubovi
11:30 AM	Correlation of Experimentally Induced Colisepticemia with Septox Condemnations in Broilers Timothy S. Cummings, Marty Ewing, Floyd Wilson, and Mark Burleson	Using Resident, Wild Mallards as Sentinels for Detecting Low Pathogenic H3, H4, H5, H6, And H7 Type A Influenza Viruses Circulating in Resident, Wild Birds Richard Slemmons, Jacqueline Nolting, Lloyd Alexander, and Dennis Senne
11:45 AM	Antibiotic Resistance Plasmid Eliminated from Poultry Bacteria: New Scientific Evidence to Counter the Expert Critics and Spin-Factories Steven R. Clark and Jeremy J. Mathers	Results of Avian Influenza Monitoring in Wild Birds in Maryland: 2005-2006 Cindy P. Driscoll, Larry J. Hindman, Richard Slemmons, and Dennis Senne
12:00	Adjourn	Adjourn





INSTRUCTIONS FOR POULTRY POSTER PRESENTATION

The following is specific information about the 2007AVMA Poultry Poster Presentation:

Location of Poultry Poster Presentation:
Convention Center

Please refer to the Poultry Poster Program for your session number.

NOTE: Posters must be set up before 7:00 AM on Sunday, July 15th.

All posters must be removed by 12:00 PM on Wednesday, July 18th.

Dimensions: Size of the Mounting Board for Poster: 4 feet X 8 feet (48 inches x 96 inches). All posters must fit within the outer edges of the board. All boards will be double sided, so another presenter may be mounting a poster on the other side of the board at the same time.

General Information:


1. Your poster presentations must be available for viewing during the hours scheduled by the Poultry Poster Section.
2. Handouts are permitted. *Sale of any material is strictly prohibited.*
3. Poster sessions are intended to serve as informal discussions and not as lectures or paper reading sessions.
4. Pushpins will be available in the poster areas. Please do not write or paint on the poster boards.
5. Projection equipment and electrical outlets will not be provided in the poster session area.
6. One (1) complimentary convention registration will be provided for the primary author of each poster.



Poster Session
Sunday, July 15 – Wednesday, July 18, 2007
Room 206

Avian Influenza

1. **Histopathological Studies of the Respiratory System of Essential Oil-Treated Broilers Against *Mycoplasma gallisepticum* and/or H9N2 Challenges**
Elie K. Barbour, Rindala G. El-Hakim, Danyelle D. Gerges, Pia A. Nehme, Hussam A. Shalb, and Marc S. Kaadi
2. **Evaluation in Chickens of a Killed NS1 Mutant Avian Influenza Virus Vaccine**
Vinyak Brahmakshatriya, Blanca Lupiana, and Sanjay M. Reddy
3. **Comparative Pathology of H5N1 Highly Pathogenic Avian Influenza Virus Infection in Avian Species in the Orders Anseriformes and Charadriiformes**
Justin D. Brown, David E. Stallknecht, and David E. Swayne
4. **Epidemiological Studies on Avian Influenza Infection in the Migrating Wild Bird**
Seong-Hwan Byun, Min-Jeong Kim, Jeong-Nyeo Kim, and In-Pil Mo
5. **Development of Microsphere-Based Assays for the Detection of H5 and H7 Subtype Avian Influenza Virus**
Wonhee Cha, Megan Strother, Mo Saif, and Chang-Won Lee
6. **Avian Influenza Neuraminidase 1 (N1) ELISA Using Baculovirus Expressed Antigen and its Application on DIVA Vaccination Strategy**
Maricarmen García, Yuru Liu, Xiuqin Xia, David E. Swayne, David L. Suarez, Mark W. Jackwood, and Egbert Mundt
7. **Determination of Pathogenicity of Jordanian Local Isolate of Avian influenza (H9N2) in Broiler-Chickens**
Saad Gharaibeh
8. **Efficacy of Inactivated Vaccine Against Low Pathogenic Avian Influenza (H9N2) in SPF and Commercial Chickens**
Bong-Do Ha, Chang-Hee Lee, Jong-Nyeo Kim, and In-Pil Mo
9. **Sequence Analysis and Phylogenetic Study of the Entire Genome of Three Avian Influenza H9N2 Subtypes from South China**
Mazhar I. Khan, Zhixun Xie, Jianbao Dong, and Xiaofei Tong
10. **Zoning as a Measure to Minimize Avian Influenza Spread During an Epidemic**
Heather Labelle, Jean-Pierre Vaillancourt, Michel Bigras-Poulin, and Alex Thompson
11. **Replication of Influenza Viruses in Chicken-Origin (DF-1) and Quail-Origin (QT-6) Cell Lines**
Chang-Won Lee, Keumsuk Hong, and Megan Strother
12. **Development of a Triplex Fluorescence Microsphere Based Immunoassay (FMIA) for the Detection of Antibodies to Avian Influenza Virus Proteins**
Blanca Lupiani, Douglas S. Watson, and Sanjay M. Reddy
13. **Ups and Downs of Implementation of an Eradication Strategy During an Outbreak of HPAI (H5N1) in Iran**
Seyed Mehdi Mirsalimi and Afshin Hedayati
14. **Efficacy and Safety of the Low Pathogenic Avian Influenza Vaccine (H9N3) in SPF Chickens**
In-Pil Mo, Jung-Eun Kim, Jeong-Hwa Shin, Seong-Hwan Byun, and Jong-Nyeo Kim
15. **Validation of a New Avian Influenza Antibody Blocking ELISA**
R. Munoz, K. Velek, A. Rice, B. Packer, S. Michaud, H. Liauw, and V. Leathers

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16. **Genetic and Biological Characterization of the H5N2 Virus Isolated from a Parrot**
Smitha Somanathan-Pillai, David L. Suarez, and Chang-Won Lee
 17. **Evaluation of Poultry Diagnostic Tests for Avian Influenza Virus in Duck Origin Samples**
Erica Spackman And Mary J. Pantin-Jackwood
 18. **The Importance of Educational Programs in the Prevention and Control of Avian Influenza**
Nathaniel L. Tablante
 19. **New Way to Develop Live Influenza Vaccine Candidate Strains**
Leyi Wang, Smitha Somanathan-Pillai, Megan Strother, Keumsuk Hong, Mo Saif, David Suarez, and Chang-Won Lee

Bacteria, Miscellaneous

20. **Case Report: Dactylariosis in Bobwhite Quail**
Joel Cline, J. Scott Helton, and Frederic J. Hoerr
21. **Non-Serotypable *Avibacterium paragallinarum* Infection in Commercial Layers**
A. García, F. Romo, A. M. Ortiz, and P. J. Blackall
22. **The Incidence of *Bordetella avium* in Mississippi Broilers**
Sue Ann Hubbard and Roy D. Montgomery
23. **Serotypes of *Riemerella anatipestifer* Isolated from Commercial Ducks and Vaccination of Ducks with Killed Oil Vaccine in Korea**
Seong-Joon Joh, Pornpen Pathanasophon, Yong-Kuk Kwon, Min-Chul Kim, Min-Jeong Kim, Hyuk-Man Kwon, Jae-Hong Kim, and Jun-Hun Kwon
24. **Pathogenic *Campylobacter* in Turkey Production**
Catherine M. Logue, Mohamed K. Fakhr, Jessica L. Thorsness, and Julie S. Sherwood
25. **Cutaneous Candidiasis of the Comb in a Broiler Breeder Flock**
Claudia Osorio, Oscar J. Fletcher, Michael J. Dykstra, and H. John Barnes
26. **In Vitro Adhesion Assays of *Gallibacterium* to Chicken Tracheal or Oviductal Epithelial Cells**
Saúl Ramírez, Andrea Zepeda, Vicente Vegas, Vladimir Morales, Luis Pérez, Soledad Díaz, Henrik Christensen, Anders Miki Bojesen, and Edgardo V. Soriano
27. **Detection of *Avibacterium paragallinarum* By PCR: Conditions of Nasal Swab Samples Submitted To The Diagnostic Laboratory**
Edgardo V. Soriano, Vladimir Morales, Salvador Lagunas, Simón Martínez, Celene Salgado-Miranda, and Patrick J. Blackall
28. **Peritonitis in Egg-Type Chickens Caused by *Gallibacterium anatis* and *Escherichia coli***
Darrell W. Trampel, Yvonne Wannemuehler, and Lisa K. Nolan
29. **Hemadsorption and Hemagglutination of *Ornithobacterium rhinotracheale***
Vicente Vega, Andrea Zepeda, Saúl Ramírez, Pomposo Fernández, Roberto Montes De Oca, Patrick J. Blackall, and Edgardo V. Soriano
30. **Hemadsorption and Hemagglutination of *Gallibacterium* Reference Strains**
Andrea Zepeda, Saúl Ramírez, Vicente Vega, Vladimir Morales, Martín Talavera, Henrik Christensen, Anders Miki Bojesen, and Edgardo V. Soriano

Chicken Anemia Virus


31. **Peruvian Serological Survey for Chicken Anemia Virus in Broiler Chickens**
Rosa Gonzalez, Eliana Icochea, Branko Alva, Xavier Castro Pozo, and Paola Cruz
32. **Mapping of Epitopes of VP2 Protein of Chicken Anemia Virus Using Monoclonal Antibodies**
Xiaomei Wang, Xiaoyan Wang, Honglei Gao, and Yulong Gao

E. coli

33. **Emergence of Antimicrobial Resistance, Class 1 Integrons, and R Plasmids Among Avian *Escherichia coli***
Lisa K. Nolan, Timothy J. Johnson, Yvonne M. Wannemuehler, Jennifer Scaccianoce, and Catherine M. Logue
34. **Creation of an *Escherichia coli* Plasmid Genome Database**
Paul Mangiamiele, Timothy J. Johnson, Yvonne M. Wannemuehler, and Lisa K. Nolan
35. **Multiplex PCR Can Distinguish Between Virulent And Commensal Avian *Escherichia coli***
Yvonne Wannemuehler, Timothy J. Johnson, Curt Doetkott, Sandy Cloud Rosenberger, and Lisa K. Nolan

General Diseases And Management

36. **Focal Duodenal Necrosis (FDN) an Emerging Disease Affecting the Layer Industry**
Tammy A. Baltzley and Thomas G. Rehberger
37. **A Case of Acute Intoxication with Monensin in Broiler Chickens**
Elisabeta Bianu and Daniela Nica
38. **High Mortality of Newly Hatched Commercial Meat Turkeys**
Richard P. Chin, Rocio Crespo, Charles Corsiglia, H. L. Shivaprasad, Murugan Subbiah, Franz Sommer, and George L. Cooper
39. **Characterization of Broiler Carcasses Condemned at Processing Plants**
Marty Ewing, Timothy S. Cummings, Floyd Wilson, and Mark Burlison
40. **Proliferative Lung Disease in Broiler Breeder Hens**
Oscar J. Fletcher, Michael Martin, and John Barnes
41. **The Use of a Field Data Collection Software to Aid in a More Informed Decision Making Process in a Production Setting**
Luis B. Gomez and Francisco J. Obispo
42. **Addressing the Continuing Educational Needs of Poultry Producers**
Teresa Y. Morishita
43. **Accidental Poisoning of Geese with Zinc Phosphide**
Daniela Nica and Elisabeta Bianu
44. **Agroterrorism – Is the Threat to Agribusiness Real?**
Robert A. Norton and Mark D. Gorwitz
45. **Histologic Evaluation of the Extramedullary Hemopoietic Tissue in Healthy and Yolk Sac Infected Young Broilers**
C. Gabriel Senties-Cué, Floyd D. Wilson, and Danny L. Magee
46. **Early Diagnosis of Fatty Liver-Hemorrhagic Syndrome in Commercial Layers Using Clinical Biochemistry**
Hyun-Hee So, Min-Jeong Kim, Hwan-Hee Kim, and In-Pil Mo

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47. **Biosecurity Multimedia Training: A Follow-Up to the US Poultry & Egg Association Training Program**
Jean-Pierre Vaillancourt, Gene Lambert, Andrew Rhorer, and Charles Beard
 48. **Effect of Breeder Flock and Management System on Sibling Turkeys**
Eduardo J. Vivas, Michael P. Martin, Sophia Kathariou, Robin M. Siletzky, and John Barnes
 49. **Interesting Microscopic Lesions in Cases Submitted to Poultry Diagnostic And Research Center and Georgia Poultry Laboratory Network**
Susan M. Williams
 50. **Routine Histological & Histomorphometric Finding in the Spleen and Bone Marrow of “Small Birds” and Other Carcass Cohorts Condemned at Slaughter Under the Sep-Tox Disposition Category**
Floyd Wilson, Fred Hoerr, Tim Cummings, and Marty Ewing

Immunology, Immunity And Vaccines

51. **Anatomic Histopathology Evaluation of Thymus, Bursa and Spleen from Broilers Chickens Raise in Reuse Litter vs New Litter**
Monica Alba Chinta, Eliana E. Icochea, Pablo S. Reyna, Rosa Gonzalez, Branko Alva, Pilar Vejarano, and Antonio Tambini
52. **Practical Comparison of Spray and Barrel Methods of Vaccine Administration to Broiler Breeder Pullets**
Samuel P. Christenberry, Darryl M. Moore, N. Scott Vanhoy, and Clyde A. Weathers
53. **Development of a Real-Time RT-PCR Assay for Turkey Cytokines**
J. Michael Day and Erica Spackman
54. **US Veterinary Immune Reagents Network**
H. Lillehoj, J. Lunney, C. Baldwin, J. LaBresh, D. Horohov, J. Hansen, N. Miller, E. Bengten, G. Chinchar, M. Wilson, and B. Wagner
55. **Interpreting ELISA Test Results**
Danny L. Magee and Philip A. Stayer
56. **Oligodeoxynucleotides Containing CpG Motifs (CpG-ODN) Induce Predominantly A Th1 Type Immune Response in Neonatal Chickens**
Bhavinibahen Parekh, Susantha Gomis, Suresh Tikoo, Lorne Babiuk, Andrew Potter, Philip Willson, George Mutwiri, and Arshud Dar
57. **Development of Recombinant Antigens for Multiplexed Serologic Monitoring of Specific Pathogen Free Chickens**
Michelle L. Wunderlich, Michael J. Smith, Barry A. Bronson, Rajeev K. Dhawan, and Joe H. Simmons

Infectious Bronchitis Virus

58. **Spike Gene Sequence Analysis of Infectious Bronchitis Virus Vaccine Viruses and Their Genetic Stability In Vivo**
Enid T. Mckinley, Deborah A. Hilt, and Mark W. Jackwood
59. **Cloning of the Pathogenic M41 Strain of Avian Infectious Bronchitis Virus as Bacterial Artificial Chromosome**
Shankar P. Mondal and Elizabeth L. Buckles



Infectious Bursal Disease

- 60. The Use of Gamma Irradiation for the Inactivation of Infectious Bursal Disease Viruses**
Daral J. Jackwood, Susan E. Sommer-Wagner, and Howard Pharo
- 61. Comparison of the VP4 Sequences Among Very Virulent, Pathogenic and Attenuated Infectious Bursal Disease Viruses**
Lester Lefever and Daral J. Jackwood
- 62. DNA Vaccination Conferring Protection of Chickens Against Infectious Bursal Disease by Priming with DNA Vaccine and Boosting with Killed Vaccine**
Tsang Long Lin, Ming Kun Hsieh, and Ching Ching Wu
- 63. Evaluation of the Pathogenicity of Infectious Bursal Disease Viruses from Layer Flocks in the United States**
Daral J. Jackwood, Bollini Sreedevi, Lester J. Lefever, and Susan E. Sommer-Wagner
- 64. Bursal Disease Virus Situation in Mexico: Epidemiology and Phylogenetic Studies**
Maritza Tamayo, Mario Lechuga, and Mauricio Gonzalez
- 65. Infectious Bursal Disease Virus (IBDV) Nonstructural Protein VP5 Contributes to Virulence of Very Virulent IBDV**
Kaori Terasaki, Tsuyoshi Yamaguchi, Kenji Ohya, and Hideto Fukushi
- 66. Molecular Detection and Differentiation of Infectious Bursal Disease Virus: A Review**
Ching Ching Wu, Peter Rubinelli, Ming Kun Hsieh, and Tsang Long Lin
- 67. An Improved Method for Infectious Bursal Disease Virus Rescue using RNA polymerase II System**
Xiaomei Wang, Ziaole Qi, Yulong Gao, and Honglei Gao

Laryngotracheitis

- 68. Differentiation by RFLP and Nucleotide Sequencing of ILTV Strains Isolated During an Outbreak in São Paulo, Brazil**
Jorge Luis Chacón Villanueva, Laura Y. Villarreal, Mario S. Asayag, and Antonio J. Piantino Ferreira
- 69. In Vitro Characterization of Infectious Laryngotracheitis Virus (ILTV) Isolates from United States (US)**
Ivomar Oldoni and Maricarmen García

Miscellaneous Virus

- 70. A Duck Hepatitis Virus Type 1 is Agent Of Pancreatitis and Encephalitis in Muscovy Duckling**
Jean-Luc Guerin, Olivier Albaric, Vincent Noutary, and Cyril Boissieu
- 71. Epidemiological Studies of Adenoviral Infection in the Broiler Farms**
Jong-Nyeo Kim, Seong-Hwan Byun, Min-Jeong Kim, and In-Pil Mo
- 72. Characterization of M-Class Genome Segments of Muscovy Duck Reovirus**
Yun Zhang, Dongchun Guo, and Hongwei Geng



Mycoplasma

73. ***Mycoplasma iowae* Associated with Vertebral Chondrodystrophy in Commercial Turkeys**
David H. Ley, Eduardo J. Vivas, Oscar J. Fletcher, and H. John Barnes

Newcastle

74. **Immunoprotection of a Commercial Vaccine (VG/GA - Strain) Against Newcastle Disease in Broilers Chicken**
Eliana Icochea, Rosa Gonzalez, Pablo Reyna, Jhon Guzman, and Blanca Talavera
75. **Molecular Analysis of Velogenic Newcastle Disease Virus in Avian Species From Venezuela**
Hugo Moscoso, Mariela Brett, Rafael Fernandez, and Charles L. Hofacre
76. **Development of an Immunochromatographic Kit for the Detection of Newcastle Disease Virus**
Haan-Woo Sung, Hyuk-Moo Kwon, JG Choi, JS Oh, and In-Pil Mo

Parasitic Diseases

77. **Effect of Coccidiosis on Serum Alkaline Phosphatase Levels in Chickens**
Chris R. Buckley
78. **Changes in Immune-Related Chicken Cytokine and Chemokine Gene Expression Following *Eimeria* Infection**
Hyun S. Lillehoj and Yeong Ho Hong
79. **Kinetic Analysis of Local Gene Expression Of Duodenum Intraepithelial Lymphocytes Following Primary and Secondary *E. acervulina* Infections**
Hyun S. Lillehoj, Chul Hong Kim, Yeong Ho Hong, Dong W. Park, and Calvin L. Keeler, Jr.
80. **Cytokine Gene Expression Profile in Fayoumi Chicken After *Eimeria maxima* Infection**
Hyun S. Lillehoj, Kyung Kim Duk, Yeong Ho Hong, Dong Woon Park, and Susan J. Lamont
81. **Immunomodulatory Effect of Dietary Safflower Leaf on Coccidiosis**
Hyun S. Lillehoj, Sung-Hyen Lee, Soo-Muk Park, Dong-Woon Park, Yeong-Ho Hong, Duk-Hyung Kim, Hye-Kyung Chun, and Hong-Ju Park
82. **Comparative Coccidial Oocyst Shedding Patterns of Turkeys Given Coccivac-T**
Greg F. Mathis, Charles Broussard, and Linnea J. Newman

Pneumovirus

83. **Molecular Characterization and Phylogenetic Analysis of Subtype B Avian Metapneumovirus Detected from Brazilian Commercial Flocks**
Jorge Luis Chacón Villanueva, Antonio C. Pedroso, Laura Y. Villarreal, Paulo E. Brandão, Antonio J. Piantino Ferreira
84. **Immunization of Broiler Chickens Against Avian Metapneumovirus Infection**
Eliana Icochea, Mercedes Sialer, Raúl Guillermo, Rosa Ríos, Rosa Gonzalez, Pablo Reyna, and Jhon Guzman



Salmonella

- 85. CAUTION – Diagnostic Laboratory Data on Salmonella Testing Being Presented**
Bruce R. Charlton, Hailu Kinde, and Mark Bland
- 86. Plasmid Mediated Antibiotic Resistance in *Salmonella enterica* Serovar Heidelberg from Turkeys**
Aaron M. Lynne and Steven L. Foley
- 87. Development and Preliminary Evaluation of a Live Recombinant Vaccine Based on the *sefABCD* Fimbrial Operon to Elicit Immune Response in Chickens Against *Salmonella enterica* Serotype Enteritidis**
K. V. Nagaraja, Vanessa Burkas, Binu Velayudhan, and David Halvorson
- 88. Organic Acid Water Treatment Reduced Salmonella Horizontal Transmission in Broiler Chickens**
Marco A. Quiroz, Charles L. Hofacre, Greg F. Mathis, Julia Dibner, and Chris Knight
- 89. Functions Exerted by the Type Three Secretion Systems (TTSS) During *Salmonella enteritidis* Infection of Chicken Reproductive Epithelial Cells and Macrophages**
Shuping Zhang, Shuhui Li, Mike Zhang, Hyun S. Lillehoj, Lanny Pace, and Danny Magee

Tumor Viruses

- 90. *In Vitro* Transformation Properties of the Meq Protein of MDV**
Dharani K. Ajithdoss, Blanca Lupiani, Lucy Lee, and Sanjay M. Reddy
- 91. Effect of Adding an Antibiotic to a Marek's Disease Vaccine**
Celina Buscaglia

Session A, Monday, July 16, 2007

Room 207A

Moderator: Gregorio Rosales

8:00-8:30 AM “The Future of Veterinary Medicine and Poultry Production”

Dr. John R. Glisson, Keynote Speaker

The University of Georgia, College of Veterinary Medicine,
Poultry Diagnostic and Research Center, Dept. of Population Health

Session A, Monday, July 16, 2007

Moderator: Stewart Ritchie

8:30-8:45 AM

Poultry House of the Future

Mark A. Dekich and Jeannie M. Harter-Dennis

AviTech LLC.
510 Naylor Mill Road
Salisbury, MD 21801

Department of Agriculture
University of Maryland Eastern Shore
Princess Anne, MD 21853

Ammonia production by commercial poultry houses has become a major environmental concern. Disposal of poultry litter in the volume produced today is also problematic for environmental concerns. A novel concept has been engineered to address these issues in the poultry house. The engineered concept includes a plenum in the floor (no wood shavings are required), a unique ventilation system, and F.A.P.P. nursery.

Poultry flock performance is improved, better maximizing genetic potential. Poultry pathogens are minimized resulting in better health status and lowering carcass contamination to the processing plant.

8:45-9:00 AM

Welfare Audits: Present and Future Needs of the Poultry Industry

Ken Opengart

Keystone Foods
6767 Old Madison Pike
Huntsville AL 35806

The poultry industry has and will continue to conduct animal welfare audits to assure its customers, and ultimately the consumers, that products that enter the food chain are produced using accepted animal welfare standards. The primary mechanism that the industry has developed to evaluate compliance with currently accepted animal welfare parameters is the audit. Standardization of audits and training of individuals employed by the third party auditing companies is sometimes lacking and can result in inaccurate evaluations. A mechanism for training auditors now exists with the formation of the Professional Animal Auditing Certification Organization (PAACO). Audit criteria, however, are often subjectively assigned without the use of any objective and quantitative metrics. Specifically, criteria for measuring broken wings, gait scoring and foot pad scoring is particularly variable between auditing companies. To further improve the audits themselves, an organization/system needs to be established that would draw on available literature or commission controlled studies to develop objective, easily measurable and scientifically-based scoring criteria.



Session A, Monday, July 16, 2007

9:00–9:15 AM

Annual Antibiotic Consumption in a Commercial Turkey Operation

Eric Gonder and Becky Tilley

Goldsboro Milling Company

P. O. Drawer 10009

Goldsboro, NC 27532

egonder@gmcom.net

Annual consumption of antibiotics in the poultry industry is an issue of continuing debate between agriculturists and anti-agriculture activists. Policy-makers have virtually no actual antimicrobial consumption information with which to conduct policy discussions. Information on antimicrobial consumption retrieved from feed consumption and water treatment records from a typical production year(s) in the recent past for a market turkey production company will be presented.

9:15–9:30 AM

Use of 4-H Project Turkeys as Serological Sentinels in North Carolina

David V. Rives and Summer M. Russell

Prestage Farms, Inc.

4651 Taylors Bridge Highway

Clinton, NC 28328

Poults hatched on June 6, 2006 were distributed among 192 youth from twenty-six North Carolina counties. One hundred sixty-five turkeys were registered on October 12th to be shown at the North Carolina State Fair. A blood sample was obtained from each bird, along with body weight and other pertinent information. Serum from each sample was tested by ELISA for antibodies against avian influenza virus, Newcastle disease virus, turkey coronavirus, *Pasteurella multocida*, *Mycoplasma gallisepticum*, *Mycoplasma synoviae*, and *Mycoplasma meleagridis*. Assay results will be discussed as they relate to the poultry density of the areas in which the turkeys were raised.

9:30–10:00 AM

BREAK

**Session A, Monday, July 16, 2007
10:00–10:15 AM**

Moderator: Ken Opengart

Teaching Poultry Necropsy Techniques to Afghan Veterinary Students

Richard M. Fulton, Ezatullah Jaheed, and Robert M. Smith

Diagnostic Center for Population and Animal Health

and

Pathobiology and Diagnostic Investigation

Michigan State University

4125 Beaumont Road

Lansing, Michigan 48910-8104

Fifth year veterinary students at the Faculty of Veterinary Sciences, Kabul University, Kabul, Afghanistan were taught poultry necropsy techniques as part of the effort to reestablish a national catastrophic disease surveillance system for Afghanistan. Due to the lack of funding prior to multinational occupation of Afghanistan, necropsy technique was not taught to veterinary students for over 20 years. After first training graduate veterinarians, the effort focused on retraining an Afghan veterinary pathologist, helping him develop computerized lectures with images, allowing him to lecture to his students, and then providing him with the animals and equipment to train his students. It was the first time in over 15 years that the veterinary pathologist was able to project an image while he lectured.

10:15–10:30 AM

Pre-Peak Lay Broiler Breeders with Impaired Mobility and High Mortality Associated with Myopathy and Hypocalcemia

**Valerie R. Bullock, Michael P. Martin, Michael J. Wineland, Oscar J. Fletcher,
and H. John Barnes**

Poultry Health Management Team

Population Health & Pathobiology Department

College of Veterinary Medicine, NC State University

4700 Hillsborough Street

Raleigh, NC 27606-1499

A broiler breeder flock that had hens with impaired mobility and increased mortality was compared with a similar unaffected flock. Gross necropsy, histopathology, and clinical chemistries were performed on birds with impaired mobility, fresh mortality, and control birds at ages 28, 29, and 30 weeks. Affected birds typically showed lesions of myopathy in the adductor and breast muscles. Ionized calcium, determined with an I-Stat analyzer, was lower in hens with impaired mobility than normal hens. These findings indicate muscle disease and low ionized blood calcium characterize a metabolic disease that affects young broiler breeders prior to peak egg production.

Session A, Monday, July 16, 2007
10:30–10:45 AM

**Use of the I-Stat Serum Chemistry Analyzer for Evaluation of Flock Health in Pre-Peak Lay
Broiler Breeders**

**Michael P. Martin, Valerie R. Bullock, Michael J. Wineland, Oscar J. Fletcher,
and H. John Barnes**

Poultry Health Management Team
Population Health & Pathobiology Department
College of Veterinary Medicine, NC State University
4700 Hillsborough Street
Raleigh, NC 27606-1499

Hens with impaired mobility in flocks with elevated mortality often remain undiagnosed. Investigations of ‘calcium tetany’ as a possible etiology suggest multiple metabolic abnormalities as probable causes for early-lay impaired mobility and elevated mortality.

The I-Stat serum analyzer has been used in our investigations to help evaluate hens with impaired mobility in flocks with increased mortality. Since this is the first use of the I-Stat analyzer in commercial poultry, normal baseline data and standardized procedures had to be established. Electrolyte imbalances including hypocalcemia, hypoglycemia, and pH changes are among the abnormalities that have been identified. Blood parameters of clinically affected hens have been compared with unaffected hens and correlated with other diagnostics.

10:45–11:00 AM


Incubation Conditions and Leg Health in Large High Yield Broilers

**Edgar O. Oviedo-Rondón, Mike Wineland, Sarah Funderburk, Heather Cutchin,
John Small, and Mike Mann**

North Carolina State University, Department of Poultry Science, Raleigh, NC. 27695 -7608

Leg health is one of the most prevalent causes of late mortality for large broilers and has major impact on welfare audits. Our previous research has shown that incubation conditions can affect thyroid hormones, development of bones and other organs that influence leg health in broilers and turkeys. This project evaluated the effects of incubation profiles on leg health of broilers at eight weeks of age under commercial conditions. Eggs from the same breeder flocks were incubated in either single stage or multi stage machines. Hatchlings were placed in paired houses on the same farms. At 56 days of age, 200 chickens per house, in each of the four farms, were sexed, weighed and legs inspected for crooked toes, valgus/varus deformities, hock burns, foot pad dermatitis, and gait scores. Walking ability was divided into six categories of gait scores ranging from completely normal (score 0) to immobile (score 5).

Results indicated that incubation profiles influenced ($P < 0.05$) body weights at 56 days of age. Broilers hatched in single stage incubation machines were heavier than those hatched in multi stage machines at 56 d of age. Male broilers had higher incidence ($P < 0.001$) of crooked toes, valgus, gait scores 1 and 2 than females. Higher percentage of birds with crooked toes (0.8 vs 0.1%), and gait scores 1, 2 and 3 were observed ($P < 0.001$) in broilers hatched in multistage stage machines, while broilers hatched in single stage machines had higher percentage of birds with gait score 0 (63.4 vs 53.2%). Valgus deformation, foot pad dermatitis and hock burns were mainly ($P < 0.05$) due to farm management conditions. At the moment of evaluation, farm litter conditions were more humid and caked where foot pad dermatitis and hock burn incidence was higher. Varus deformation, twisted legs or severe lameness (gait scores 4 and 5) were rarely observed and were not significantly affected ($P > 0.05$) by any of the variables evaluated. No



significant interactions between incubation profile and farm conditions or sex were observed, indicating that the effect of incubation is similar for both genders and across farms.

These results indicated that incubation conditions have an effect on leg health of broiler populations. Our prior results indicated that cold conditions during the first days of incubation, and elevated incubation temperatures and hypoxia during the plateau stage of incubation (last four days) affect long bone development and increases leg fluctuating asymmetry. Proper incubations conditions to improve hatchability, chick quality and broiler performance may also improve leg health.

Session A, Monday, July 16, 2007
11:00–11:15 AM

Vertebral Canal Stenosis as a Cause of Lameness in Broilers: Additional Findings

Kelli H. Jones^A, H. John Barnes^B, Kenneth C. Powell^C, Suzanne Y. Stamey^D

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A case study on the importance of vertebral canal stenosis as a cause of lameness in broilers was conducted. The objective of the study was to evaluate gross and histopathologic lesions of the spinal column to determine the etiology of paresis/paralysis. Study candidates included meat type broilers over 35 days with clinically acute lameness, and included birds that had received antibiotics and growth promotants, and those that had not. Vertebral samples were collected, and various abnormal lesions (scoliosis, osteomyelitis, fractures, spondylolisthesis, and rickets) were identified. Incidences of these lesions, as well as bacterial agents involved are discussed.

11:15–11:30 AM

State Quarantines of Respiratory Disease Found at Auction: Lessons Learned

Donna J. Kelly

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In the spring of 2006, the Georgia Department of Agriculture began to quarantine birds sold at auction exhibiting signs of respiratory disease, as a result of complaints from purchasers that the birds were either sick or dying after they returned to home from the sale. Lots were returned, birds were submitted to diagnostic laboratories, and owners needed to comply with treatment and husbandry recommendations. Infectious Coryza and Mycoplasma gallisepticum infection were the leading diagnoses. Other findings will also be summarized. The economic, social, and bird health ramifications will be discussed. Flock follow-ups will be presented.

Session A, Monday, July 16, 2007
11:30–11:45 AM

Chromium Associated Discoloration of Egg Yolks in a Backyard Flock

**H. M. Opitz, DVM, T. Wilson, Ph.D., B. Hoskins, Ph.D, S. Perron, Ph.D., Xi Wang^A, Ph.D.
and D. Beane, B.A.**

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and ^ADepartment of Genetics and Biochemistry, Clemson University

Persistent green discoloration of egg yolks was observed in 10 to 60% of the eggs laid by a ranged backyard flock of 18 New Hampshire Red hens. Flock health and fertility were not adversely affected.

The green color in the yolk was layered in concentric rings. Green discolored egg yolks contained between 0.6 and 23 PPM chromium, compared to yellow egg yolk that contained less than 0.13 PPM chromium. Chromium levels were also elevated in the bile, liver, blood and kidney but not in the albumen, fat or muscle tissues. A preliminary study also showed elevated cobalt levels in the yolk. Gossypol was not detected in yolk or feed.

Discoloration of yolk disappeared and chromium levels in yolks and tissues returned to non-detectable levels within 3- 5 weeks after removal of the birds from the range and keeping them indoors. The highest chromium level found in soil samples was 39 PPM. Chromium was not detected in well or surface water. The mechanism of chromium absorption remains undetermined. Tannery waste is the suspected source of the soil contamination that caused this problem. Chromium in commercial feed at 1 PPM was not detected in yolk and is excreted in the feces.

The total chromium content in green discolored egg yolks was up to 300 times greater than the recommended dietary chromium levels and 37 times greater than the safe treatment level of diabetes II for humans.

11:45–12:00 PM

Differential Diagnosis of Liver Hemorrhage in Laying Hens

Craig Riddell
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Furdale SK Canada S7T1B1

Increased mortality in caged leghorn hens, characterized by sudden death and pale heads, is due primarily to two distinct syndromes called Fatty Liver-Hemorrhagic Syndrome (FLHS) and Hepatitis-Splenomegaly Syndrome (HSS). These syndromes can be differentiated by abdominal blood clots and large pale livers in FHLS while abdominal unclotted blood is associated with large mottled livers and splenomegaly in HSS. Differences in liver microscopic pathology include very fatty hepatocytes and scattered eosinophilic masses in FHLS compared with areas of necrosis, massive cellular infiltration and diffuse eosinophilic material separating hepatocytes in HSS. FLHS is considered to be a metabolic disease while recent research has demonstrated that HSS is a viral disease.

12:00–1:00 PM

LUNCH

**Session A, Monday, July 16, 2007
1:00–1:15 PM**

Moderator: Hector Cervantes

Field Experiences in Commercial Broilers with an *In Ovo* Coccidiosis Vaccine

**Andrea Sinclair, David Kelly, Rebecca Poston, Cherilyn Heggen-Peay, Larry Charniga,
Lynn Murray and Vivian Doelling**
Embrex, Inc.
P. O. Box 13989
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Inovocox™ is a USDA licensed, sporulated oocyst vaccine composed of two strains of *Eimeria maxima* and one strain each of *Eimeria acervulina* and *Eimeria tenella*. Field trials were conducted in 2006 at various commercial poultry integrators. The vaccine was administered *in ovo* at the time of transfer, with embryo age ranging from 18-19 days. It was injected into the embryonated egg in a volume of 0.050 ml Marek's vaccine diluent using the Embrex Inovoject® system, and was co-administered with Marek's disease and infectious bursal disease vaccines in the same *in ovo* injection. Hatchability, livability, clinical appearance, weight gain, feed conversion ratio, and condemnations were compared with broilers receiving the integrator's conventional coccidiosis control program.

1:15–1:30 PM

***Eimeria* Shedding Patterns Under Various Environmental and Coccidiosis Management Programs**

Linnea J. Newman
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Summit, NJ 07901
Greg F. Mathis
Southern Poultry Research
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Eimeria spp. in infected birds produce higher fecal oocyst counts with each sequential life cycle until the infection is curbed by the immunological response. The timing and the peak oocyst numbers produced in a poultry flock depend upon the anticoccidial program and the environmental management of the birds. Fecal samples were collected every 3rd day from U.S. and Canadian broilers reared on clean and reused litter, from low-density broiler breeder pullets, from breeder turkeys and commercial turkey flocks. Samples represented conventional anticoccidial programs as well as live coccidiosis vaccination programs. The overall oocyst shedding pattern, as well as patterns by species were evaluated. As the portfolio of anticoccidial drugs ages and live coccidiosis vaccination increases, it is important to understand the impact of environmental management programs on both coccidiosis control methods.



Session A, Monday, July 16, 2007
1:30–1:45 PM

Comparison of *Eimeria* Species Composition Between High-Performance and Low-Performance Broiler Operations.

Mark Jenkins¹, Spangler Klopp², Donald Ritter³, and Katarzyna Miska¹

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The level and species composition of *Eimeria* oocysts in litter were determined for high performance and low performance poultry farms in three different U.S. broiler regions. The technique of ITS1 rDNA PCR was used to ascertain the relative level of *Eimeria* species present. Although oocyst concentrations in litter were similar between all poultry operations, higher numbers of *E. maxima* were often associated with low performance farms. Also, *E. acervulina* and *E. praecox* were often found at high concentrations irrespective of broiler operation type. *E. praecox* oocysts were isolated and pathogenicity studies were conducted on this infrequently identified *Eimeria* species.

1:45–2:00 PM

Analysis of gene expression in the intracellular stages of the poultry parasite, *Eimeria acervulina*

Katarzyna B. Miska and Raymond H. Fetterer

USDA/ARS

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Coccidiosis in chickens is caused by seven species belonging to the genus *Eimeria*. Even though coccidiosis is a complex disease that can be caused by any combination of these species most of the molecular research concerning chicken *Eimeria* has been limited to *Eimeria tenella*. This study describes the first large-scale analysis of over 1000 genes expressed by the second stage merozoites and schizonts of *E. acervulina*. Expression profiles appear to be drastically different than those of *E. tenella* merozoites. One of the drastic differences is the lack of transcripts that may encode microneme proteins.



**Session A, Monday, July 16, 2007
2:00–2:15 PM**

**Characterization of Novel Lytic Peptide Secreted by Intestinal Intraepithelial Lymphocytes
Infected with Coccidia**

Hyun Lillehoj¹, Sung Lee¹, Doug Bannerman² and Yeong H. Hong¹

¹Animal Parasitic Diseases Laboratory and ²Bovine Functional Genomics Laboratory, Beltsville
Agricultural Research Center, USDA-ARS, Beltsville, MD. 20705

The inflammatory response to parasites is mediated by multiple host factors. In this report, we present molecular and functional characterizations of a novel immune mediator whose gene expression increased following infection with *Eimeria*. NK-lysin is an anti-microbial and anti-tumor protein expressed by NK cells and T lymphocytes. Full-length clone encoding chicken NK-lysin was isolated from intestinal intraepithelial lymphocytes (IELs) cDNA library. NK-lysin is consisted of an 868 bp DNA sequence with an open reading frame of 140 amino acids and a predicted molecular mass of 15.2 kDa. Comparison of its deduced amino acid sequence showed less than 20% identity to mammalian NK-lysins. The tissue distribution of NK-lysin mRNA revealed highest levels in intestinal IELs, intermediate levels in splenic, peripheral blood lymphocytes and lowest levels in thymic, bursa lymphocytes. The kinetics of NK-lysin mRNA expression indicated that, whereas infection with *E. acervulina* induced maximum expression only at 7-8 days post-infection, *E. maxima* and *E. tenella* elicited biphasic responses at 3-4 and 7-8 days post-infection. Finally, recombinant chicken NK-lysin expressed in COS7 cells exhibited anti-tumor cell activity against LSCC-RP9. NK lysin did not show any cytolytic activity against *E. coli*. We conclude that chicken NK-lysin plays important roles during anti-microbial and anti-tumor defenses.

2:15–2:30 PM

The Use of Chlorine Dioxide as a Disinfectant in a Broiler Hatchery

**Mark Burleson, DVM, MS¹, Phil Stayer, DVM, MS, ACPV¹, John Smith, DVM, MS, MAM,
ACPV², and Joel Tenney³**

¹Sanderson Farms, Inc., ²Fieldale Farms, ³ICA Trinova

Formaldehyde is a commonly used disinfectant in broiler hatcheries, despite human health concerns with its use. Chlorine dioxide gas may provide similar disinfecting properties, while using a different safety profile than formaldehyde. Initial trials tested the viability and efficacy of chlorine dioxide and demonstrated less bacteriocidal and fungicidal effects when compared to formaldehyde. No deleterious effects were seen in chicks from hatcheries treated with chlorine dioxide. Numerous other trials were performed using increasing doses of ClO₂ and no disinfectant as a control. Final results will be presented at the 2007 AVMA meeting.

2:30–3:00 PM

BREAK



**Session A, Monday, July 16, 2007
3:00–3:15 PM**

Moderator: Suzanne Young Stamey

**Enumerating *Salmonella* in Poultry Samples using a DNA Sequence Capture
Combined with Q-PCR**

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St. Paul, MN 55108

Quantitative measurements of pathogens such as *Salmonella* are needed because prevalence estimates are not directly related to risk. Standard Most Probable Number (MPN) methods are laborious and expensive. The purpose of this study was to develop a method to quantify *Salmonella* in a diversity of poultry samples without the need for enrichment. Biotinylated capture probes were designed using the *invA* gene and hybridized with DNA extracted from the poultry sample. Streptavidin-coated magnetic beads separated the target DNA from the remainder of the community DNA and any residual inhibitors. Gene quantity was assayed by SYBR Green qPCR. This approach is especially useful when *Salmonella* loads are low, for example less than 1000 cfu / g of sample.

3:15–3:30 PM

Is *Salmonella enterica* Kentucky a Newly Emergent, Poultry Adapted Strain?

**John J. Maurer, Adriana Pedroso, Margie D. Lee, Holly Sellers, Erich Linnemann,
and Katherine Zamperini.**

Dept. of Population Health, The University of Georgia, Athens, GA 30602

There has been a significant upsurge in the isolation of *Salmonella enterica* Kentucky from poultry. We have identified a major *S. Kentucky* clone in poultry that lacks several, important virulence genes. Is *S. Kentucky* prevalence due to its competitive advantage over other serovars in poultry? We performed in vivo competition experiments in broiler chickens between *S. Kentucky* and *S. Typhimurium*. We did not observe any significant differences in fecal shedding, but did observe differences in the serovars' compartmentalization within tissues of infected animals. These differences were not attributed to defects in cell invasion, but absence of the virulence plasmid.

Session A, Monday, July 16, 2007
3:30–3:45 PM

Discrimination between Strains of *Salmonella* Enteritidis PT13 and PT13a by Comparative Genome Sequencing of the Virulence Plasmid

Jean Guard-Bouldin^{*1}, Cesar Morales¹, Adam B. Olson² and Matthew W. Gilmour²

¹U. S. Dept. of Agriculture, Agricultural Research Service, 950 College Station Road

²National Microbiology Laboratory, Public Health Agency of Canada, Winnipeg, MB, Canada

Salmonella Enteritidis (SE) is an important pathogen of poultry products and it appears to be increasing within broiler flocks. Subtyping of SE is commonly done by phage typing, plasmid profiling, genomic microarrays, pulsed field gel electrophoresis and other restriction-enzyme based fingerprinting methods. A series of subtyping methods applied to SE PT13 strains isolated from disparate geographical and sources within Canada did not achieve adequate discrimination. However, comparative genomic sequencing detected putative single nucleotide polymorphisms (SNPs) in the large virulence plasmid that discriminated PT13 isolates obtained from humans and from mung bean sprouts from PT13a phenotypic variants and from the reference PT4 sequence. When compared to genomic sequence, the SE virulence plasmid appears to have a lower evolutionary rate of change. Three virulence plasmid SNPs were reliable for distinguishing PT13 lineage strains, which have the Fels2 phage, from the PT4 lineage, which has ST64b.

3:45–4:00 PM

Crop Isolated Lymphoid Follicles and Ileal Peyer's patches in Egg-Layer Hens Challenged with *Salmonella enterica* Enteritidis

Lara E. Vaughn¹, Peter S. Holt², Randle W. Moore² and Richard K. Gast²

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²United States Department of Agriculture (USDA),

Agricultural Research Service (ARS),

Egg Safety and Quality Research Unit (ESQRU),

Russell Research Center, 950 College Station Road, Athens, Georgia 30605 USA

The enteric pathogen *Salmonella enterica* Enteritidis (SE) can be harbored within the upper and lower gastrointestinal (GI) tract of chickens. Induction of immune response by SE at various alimentary tract regions may perhaps be attributable to local gut-associated lymphoid tissue (GALT). The study investigated the mucosal immune response and GALT within crop and ileum of specific-pathogen-free White Leghorn (WL) hens and four commercial layer-hen breed-strains following *per os* SE challenge. Crop contents and feces were cultured weekly to monitor SE infection. Crop lavage and lower GI tract gut-flush samples were analyzed for SE lipopolysaccharide (LPS)-specific IgA humoral response by enzyme-linked immunosorbent assay. Gross and microscopic evaluation of crop and ileum determined presence/absence of lymphoid tissue/GALT. Morphometric analysis of lymphoid tissue sites was conducted. Results showed elevated SE-LPS-specific IgA response in crop lavage and gut-flush samples. Well-defined isolated lymphoid follicles were observed in hematoxylin & eosin stained tissue sections from mid-body region of crop. Peyer's patches of proximal and distal ileum were identified at two consistent locations in eosin-Y and modified-crystal violet stained fresh tissue segments. Finding crop isolated lymphoid follicles and ileal Peyer's patches in SPF WL hens and commercial layer-hen breed-strains appears to support the idea that crop and ileum of the chicken have the capacity to generate a local mucosal immune response against SE.

Session A, Monday, July 16, 2007

4:00–4:15 PM

Oligodeoxynucleotides containing CpG motifs (CpG-ODN) confer protection against a lethal challenge of *Salmonella typhimurium* septicemia in neonatal broilers.

Azita Taghavi¹, Brenda Allan², George Mutwiri², Andrew Van Kessel³, Lorne Babiuk², Andrew Potter², and Susantha Gomis¹

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³Dept. of Animal and Poultry Science, College of Agriculture, University of Saskatchewan, Saskatoon, SK, S7N 5A8.

Oligodeoxynucleotides (ODN) containing CpG motifs (CpG-ODN) have been shown to stimulate the innate immune system against a variety of viral, bacterial and protozoan infections in a variety of vertebrate species. The objective of this study was to investigate the immunostimulatory effect of CpG-ODN in neonatal broilers against *Salmonella typhimurium* septicemia. Day old broiler chicks, or embryonated eggs that had been incubated for 18 days, received 50 µg of CpG-ODN, 50 µg of control non-CpG-ODN, or saline. Four days after exposure to CpG-ODN, 1×10^6 or 1×10^7 cfu's of a virulent isolate of *S. typhimurium* was inoculated by the subcutaneous route in the neck. Clinical signs, pathology, bacterial isolations from the air sacs, and mortality were observed for ten days following challenge with *S. typhimurium*. The survival rate of birds in groups receiving either non-CpG-ODN or saline following *S. typhimurium* infection was 40-45%. In contrast, birds receiving CpG-ODN had significantly higher survival rate of 80-85% ($P < 0.0001$). Bacterial loads and pathology were low in groups treated with CpG-ODN compared to the groups receiving saline or non-CpG-ODN. This is the first time that CpG-ODN has been demonstrated to have an immunoprotective effect against an intracellular bacterial infection in neonatal broiler chickens following *in ovo* delivery.

4:15–4:30 PM

Construction and evaluation of recombinant *Salmonella* vaccines expressing *Eimeria* antigen SO7.

Vjollca Konjufca*¹, Maria Dolores Juarez-Rodrigues¹, Mark Jenkins², Shifeng Wang¹ and Roy Curtiss III¹.

¹Center for Infectious Diseases and Vaccinology

Biodesign Institute, Arizona State University, Tempe, AZ 85287

²Animal Parasitic Diseases Laboratory, ARS, Beltsville, MD 20705

Our work focuses on constructing a recombinant vaccine against *Eimeria* sp.. We are using bacterial Type Three Secretion System to deliver SO7 antigen into the cytoplasm of the immunized host cells and induce antigen-specific CTL responses. For delivery, SO7 was fused to the *Salmonella* gene *sopE*. In the host strain $\chi 9242$ ($\Delta phoP233 \Delta asdA16 \Delta araBAD23 relA198::araCP_{BAD} lacI TT$), SO7 is fused to the signal sequence of the *bla* gene and its expression is controlled by *lacI* repressor gene, whose expression in turn depends on free arabinose. The lack of arabinose in the tissues increases the expression of SO7 antigen *in vivo*.



**Session A, Monday, July 16, 2007
4:30–4:45 PM**

Effects of RofenAid® Impact on Salmonella Enumeration in the Intestine and Litter and on Performance of Male Broiler Chickens After a Challenge with Salmonella and a Mixed Culture of Coccidia

Sharon Heins Miller¹, Charles Hofacre², and Gregory F. Mathis³

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This study provides data on the effects of RofenAid® as used in the starter (Days 0-10 or 0-21) and/or grower (days 21-28) programs on *Salmonella* heidelberg enumeration and the rate of gain and feed conversion of broiler chickens grown to 42 days of age after a challenge with a mixed culture of coccidian (*Eimeria acervulina*, *E. maxima*, and *E. tenella*). The results show the weight gain at 41 days was statistically significant for the program of RofenAid® used from 0-21 days and trending to significance for the other programs. *Salmonella* reduction was statistically significant for the RofenAid® program used from 0-10 and 21-28 days.

4:45 PM

ADJOURN

Session B, Monday, July 16, 2007

Room 207A

Moderator: Gregorio Rosales

8:00-8:30 AM "The Future of Veterinary Medicine and Poultry Production"

Dr. John R. Glisson, Keynote Speaker

The University of Georgia, College of Veterinary Medicine,
Poultry Diagnostic and Research Center, Dept. of Population Health

Session B, Monday, July 16, 2007

Moderator: Kenton Kreager

8:30-8:45 AM

Use of molecularly cloned avian leukosis virus to study antigenic variation following infection of meat-type chickens

Aly M. Fadly, Jody K. Mays, R. F. Silva, and H. D. Hunt

USDA-Agricultural Research Service
Avian Disease and Oncology Laboratory
East Lansing, Michigan

A molecularly cloned strain of subgroup J avian leukosis virus (ALV-J) termed R5-4 was used to study antigenic variation following infection of meat-type chickens. Chickens were inoculated with R5-4 virus at either 8 days of embryonation or at 1 week of age. Each chicken was housed in a separate isolator; chickens were sampled for ALV and antibody at 4 week intervals up to 20 weeks of age. Inoculation of R5-4 virus at 8 days of embryonation resulted in tolerant infection, as all chickens were persistently viremic with no detectable antibody. However, infection at 1 week of age induced the following infection profiles by 8 weeks post-infection: 17/30 (57%) were virus positive, antibody negative (V^+A^-), 5/30 (16%) were virus positive, antibody positive (V^+A^+), and 7/30 (23%) were virus negative, antibody positive (V^-A^+). Viruses isolated from the 5 chickens classed as V^+A^+ were not neutralized by antibodies against originally inoculated virus, R5-4, suggesting development of antibody-escape mutants. These isolates are being sequenced to determine molecular differences from originally inoculated virus.

8:45-9:00 AM

Diphtheritic tumorous growths caused by fowlpox virus containing REV provirus

Deeki N. Tripathy
Department of Pathobiology
University of Illinois, Urbana, Illinois

A fowlpox virus strain isolated from a layer flock at the University of Illinois (UI strain) is pathogenic for susceptible chickens. Chickens inoculated orally with this virus developed acute respiratory signs and tumorous diphtheritic lesions. The appearance of such lesions could not be associated with fowlpox virus until integration of reticuloendotheliosis virus (REV) in its genome was demonstrated by evaluation of nucleotide sequences of cloned genomic fragments. Unlike vaccine strains of fowlpox virus, majority of field isolates of fowlpox virus show integration of full length REV in their genome, while vaccine strains have only long terminal repeats (LTR) of REV.



Session B, Monday, July 16, 2007
9:00–9:15 AM

Molecular Characterization of Three Isolates of Avian Leukosis Virus Isolated From Contaminated Marek's Diseases Vaccines

Taylor Barbosa, Guillermo Zavala and Sunny Cheng
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Avian Leukosis viruses (ALV) were isolated from Marek's Disease vaccines. The viruses were replicated in DF-1 cells and their genomes were amplified by PCR and cloned. Direct sequencing was done using the dideoxynucleotide method. The sequences of all three genomes were analyzed and compared with known ALV sequences. The envelope region in all three viruses was identical in all three viruses and highly similar to ALV-A. The LTR region of the three ALVs is highly similar to the LTR of endogenous ALVs. The genomic sequence similarities of the three viruses examined suggest that all viruses had the same chimeric ancestor.

9:15–9:30 AM

Control of Contemporary Marek's Disease Isolates

Robin W. Morgan, Grace Isaacs, Mark Parcels, Milos Markis, Amy S. Anderson, Erin L. Bernberg, Sandra Cloud Rosenberger, and John K. Rosenberger

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Marek's disease (MD) has been controlled for nearly a half-century by vaccination with live vaccines. During this period, challenge strains have evolved toward increased virulence, and currently under some field conditions, vaccination with trivalent vaccines is required. We continue to design new MD vaccines with increased protective potential, but to date have not developed vaccines that exceed the protective indices of the best commercial vaccines that are currently available. Given the likelihood of continued evolution of MD challenge strains, we have invested considerable effort into optimizing use of available vaccines and vaccine cocktails using a shedder challenge system that mimics field conditions. These efforts have included examining variables associated with in ovo technology, the effect of vaccine dose, and the influence of embryo age at vaccination. In addition, we have characterized recent field isolates, and these results will be discussed.

9:30–10:00 AM

BREAK

**Session B, Monday, July 16, 2007
10:00–10:15 AM**

Moderator: Susan Williams

Pathological and viral characterization of a case of splenomegaly in F line turkeys: Is HVT able to produce disease?

Devorah A. Marks, A.L. Cortes, J. Barnes, O.J. Fletcher, and I.M. Gimeno

Population Health and Pathobiology
College of Veterinary Medicine
North Carolina State University

A severe case of splenomegaly in 65 week old F line turkeys with lymphoproliferative lesions in the spleen is reported. Immunohistochemistry displayed a moderate expansion of CD8 cells and a severe depletion of CD4 cells in the spleen. The samples were negative for REV and ALV by virus isolation and for serotype 1 MDV by real time PCR. All spleens had high load of herpes virus of turkey (HVT) DNA by real time PCR. These results suggest that the splenic lesions are probably of hyperplastic than neoplastic origin. The most likely cause for the splenomegaly is HVT and our results support previous studies showing the high susceptibility of Line F turkeys to infectious diseases.

10:15–10:30 AM

The Effect of Vaccination on Marek's Disease Virus Shedding

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Current Marek's Disease vaccines are efficient at preventing disease. However, vaccination can reduce but not completely eliminate virus shedding. Our hypothesis is that an efficient vaccine will result in fewer viruses being shed. To test this hypothesis, we developed a real-time PCR to measure Marek's disease virus (MDV) in dander shed by infected chickens. We will report on our findings comparing the amount of MDV shed in the dander of birds vaccinated with a good vaccine, to the amount of virus shed in birds vaccinated with a poor vaccine.



Session B, Monday, July 16, 2007

10:30–10:45 AM

A comparison between Marek's disease virus production in feather pulp versus feather follicle epithelium (FFE)

John R. Dunn¹, James B. Finlay^{1,2}, Richard L. Witter¹ and Robert F. Silva¹

1. USDA, ARS, Avian Disease and Oncology Laboratory, East Lansing, MI 48823

2. Michigan State University College of Veterinary Medicine (Class of 2009)

Although Marek's disease viral load in chickens can be assessed by quantifying the viral DNA in homogenized feather tips, this value may not correlate to the viral load being shed. This study was designed to determine what portion of the feather tip tissue is most reflective of virus load in the shed dander by comparing feather follicle epithelium (FFE) cells, adhered to the exterior of the shaft, to interior cells within the feather pulp. Using multiplex quantitative PCR, the FFE cells from infected birds had significantly higher viral copy number (VCN) per host cell than from feather pulp, and were more comparable to the VCN measured from feather dander. These results suggest that DNA isolated from FFE is more reflective of actual shed virus than VCN measured from the feather pulp.

10:45–11:00 AM

Effect of vaccination on the development of Marek's disease virus-induced eye lesions

Arun K.R. Pandiri, Aneg Lucia Cortes, Isabel M Gimeno

Population Health and Pathobiology

College of Veterinary Medicine

North Carolina State University

We have recently demonstrated that Marek's disease virus (MDV) is able to induce an acute retinopathy few days after inoculation. In this study we are presenting chronological lesions induced by a very virulent (Md5) and a very virulent plus (648A) strains of MDV. In addition, the effect of vaccination on the development of MDV-induced eye lesions was evaluated. Development of eye lesions induced by both Md5 and 648A correlated with the lesions in the brain. Severe lymphocytic choroiditis was present in all non-vaccinated chickens as early as 11 days post challenge. Vaccine viruses (HVT and CVI988) did not induce any lesion in the eye at any time point. The protective efficacy of different combinations of vaccine and challenge virus will be discussed.



Session B, Monday, July 16, 2007
11:00–11:15 AM

Relationship between load of Marek's disease vaccine DNA in peripheral blood and level of protection

Aneg Lucia Cortes, Isabel M Gimeno
Population Health and Pathobiology
College of Veterinary Medicine
North Carolina State University

Sporadic outbreaks of Marek's disease still occur in spite of the efficacy of Marek's disease (MD) vaccines. MD virus (MDV) evolution towards more virulence is a major factor for the decreasing efficacy of MD vaccines. Unfortunately, there is not a method to monitor the level of protection against MDV challenge in a vaccinated flock. The aim of this study is to develop a method to assess vaccine protection under field conditions. In this study, we have used a combination of vaccines and challenge viruses of different virulence to create groups with different level of protection. Load of MDV DNA vaccines in whole peripheral blood and buffy coats was chronologically monitored by real time PCR (3, 5 and 15 weeks post challenge). Evaluation of MDV-induced lesions was done at the termination of the study. Association between load of vaccine DNA and develop of MD lesions will be discussed.

11:15–11:30 AM

Pathogenesis of serotype 1 Marek's disease vaccines in the lung

Isabel M Gimeno, Aneg L Cortes
Population Health and Pathobiology
College of Veterinary Medicine
North Carolina State University

The role of pulmonary immune response in Marek's disease (MD) vaccine- induce protection is poorly understood. Since natural route of MDV infection is through the respiratory tract, the local immune response in the lungs might greatly contribute to the efficiency of vaccines. In this study we have evaluated the replication of various serotype 1 MD vaccines, of different efficacy, in the lung. Our results showed that serotype 1 MD vaccines replicates in the lung as early as 6 days. Highly protective vaccines replicate more in the lung than lower protective vaccines. The effect of the route of vaccination and the dose of vaccine on the lung replication is discussed.



Session B, Monday, July 16, 2007
11:30–11:45 AM

Recombinant Marek' disease virus lacking the oncogene Meq as a candidate for future control of Marek's disease in chickens

Lucy F. Lee, Sanjay Reddy and Kenton Kreager

¹U.S. Department of Agriculture, Agricultural Research Service, Avian Disease and Oncology Laboratory, East Lansing, MI, 48823. ²Department of Veterinary Pathobiology, College of Veterinary Medicine, Texas A & M University, College Station, TX, 7783 ³Hy-Line International, PO Box 310, Dallas Center, IA 50063-0310

Vaccination has dramatically reduced the incidence of Marek' disease, but more virulent viruses are emerging and developing new control strategies is needed. We have used Marek's disease virus (rMd5) cosmid clones to generate recombinant viruses for gene function studies. The Meq gene is a unique gene, which resembles oncogene jun/fos, transformed DF-1 cells in culture and induction of MD tumors. The Meq gene is located in the A6 cosmid, its deletion was by the RecA-assisted restriction endonuclease (RARE) cleavage method. The Meq deletion A6 cosmid together with SN5, P89, SN16, and B40 cosmids were transfected into duck embryo fibroblasts to generate a recombinant rMd5 viruses lacking the oncogene Meq (rMd5ΔMeq). We have studied the recombinant rMd5ΔMeq virus both in the laboratory and commercial setting and found it to be excellent candidate virus for protection against Marek's disease.

11:45–12:00 PM

Functions of Marek's Disease Virus MicroRNAs

Robin W. Morgan, Amy S. Anderson, Erin L. Bernberg, Emily Huang, Grace Isaacs, Milos Markis, and Joan Burnside

Dept. of Animal and Food Sciences
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MicroRNAs (miRs) are important regulatory small RNAs that control gene expression at the post-transcriptional level by targeting specific mRNAs for translational repression or for degradation. MicroRNAs are believed to function in important cellular pathways, including developmental pathways and oncogenesis. In the case of herpesvirus infections, they appear to function in viral latency, inhibition of apoptosis, and evasion of host immune responses. We have discovered eighteen miRs encoded by the Marek's disease virus (MDV) genome. Eight MDV miRs flank the meq oncogene, seven map to the LAT region of the genome and are anti-sense to the ICP4 gene, and one maps to the ribonucleotide reductase gene. Information on the roles of MDV miRs in regulating viral replication and latency will be presented. Their possible contribution to MDV oncogenicity and their potential as disease control reagents will be discussed.

12:00–1:00 PM

LUNCH

**Session B, Monday, July 16, 2007
1:00–1:15 PM**

Moderator: Richard Fulton

Evaluation of different hemagglutinin-neuraminidase (HN) chimeras using a newly developed reverse genetic system based on the mesogenic anhinga Newcastle disease virus strain

Carlos Estevez, Qingzhong Yu, Daniel King, and Patti Miller
USDA-ARS-Southeast Poultry Research Laboratory
Athens, GA 30605

A major factor in the pathogenicity of Newcastle disease virus (NDV) is the amino acid sequence of the fusion protein cleavage site, but the role of other viral genes that contribute to different clinical forms of the disease remain undefined. To assess the role of other NDV genes in virus pathogenicity, a reverse genetics system was established with the NDV anhinga strain, a virus of moderate virulence, to provide a backbone for generating gene mutations or gene exchanges in attempts to enhance or attenuate virulence of that virus. Chimeras of that backbone created by exchange of the anhinga hemagglutinin-neuraminidase (HN) gene with HN genes of neurotropic and viscerotropic velogens produced no significant change in virus pathogenicity as assessed by conducting the mean death time and intracerebral pathogenicity index assays and by inoculation of susceptible day-old SPF birds. A HN gene exchange alone within the context of the NDV anhinga backbone failed to increase virus virulence and suggests a multigenic role for NDV pathogenicity.

1:15–1:30 PM

Newcastle disease virus isolates from US waterfowl reveal novel genomic subgroups with diverse and evolving genetics

**LM Kim¹, DJ King¹, DE Swayne¹, D Stallknecht², RD Slemons³, JC Pedersen⁴,
DA Senne⁴, K Winker⁵, CL Afonso¹**

¹USDA ARS Southeast Poultry Research Laboratory, Athens, GA 30605; ²The University of Georgia, Department of Population Health, Athens, GA 30602; ³The University of Ohio, Department of Veterinary Preventive Medicine, Columbus, OH 43210; ⁴USDA APHIS Veterinary Services National Veterinary Services Laboratories, Ames, IA 50010; ⁵University of Alaska Museum, Fairbanks, Alaska 99775

Distribution of genetic groups (genogroups) of Newcastle disease virus (NDV) isolated during 1986 to 2005 in the US from 209 waterfowl and shorebirds (W&S) and 17 live bird market (LBM) samples was investigated. Waterfowl and shorebirds viruses were distinct from vaccine viruses and from the virulent genotypes that cause severe disease in poultry. Most genogroups were widely distributed across the US and present in multiple bird species with evidence of rapid genomic changes. The majority of Class I viruses found in US LBM in 2005-06 (15/17) belong to the same genogroup that infected mallard samples from the same region in 2004.

Session B, Monday, July 16, 2007

1:30–1:45 PM

Presence of Newcastle Disease virus and Avian Influenza virus in wild birds at the Peruvian coast

Armando Gonzalez¹, Rosa Gonzalez¹, Bruno Gherzi¹, David Blazes² and Eliana Icochea¹

¹College of Veterinary Medicine, University of San Marcos, Lima-PERU.

²US Naval Medical Research Center Detachment, Lima-PERU

The objective of the present study is to evaluate the presence of Newcastle Disease virus and Avian Influenza virus by collecting, at least, 298 fecal samples of migratory and resident bird of the Peruvian coast during the migratory season of the year 2006-2007. The samples will be inoculated in 10 days old SPF embryonating chicken eggs, by allantoic route.

1:45–2:00 PM

Isolation of Exotic Newcastle Disease Virus (ENDV) From Field Collected Flies and Experimental ENDV Infections of Three Arthropod Species

Daniel J. King¹, Seemanti Chakrabarti², Claudio L Afonso¹, Alec C. Gerry², Carol J. Cardona³, and David E Swayne¹

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Veterinary Medicine Extension, Davis, CA 95616

During the 2002 Exotic Newcastle Disease (END) outbreak in California arthropods were collected from two quarantined backyard poultry premises after removal of END virus infected birds. The END virus (ENDV) isolated from field collected pools of three fly species was found to have >98% homology by nucleotide sequence analysis and identical monoclonal antibody binding profiles as viruses recovered from poultry during the 2002-03 outbreak. Two fly species and beetles commonly associated with poultry operations were provided ENDV in feed and then sampled daily for up to 12 days post-infection. In the experimental flies, the virus was recovered through days 4-5 in *Musca domestica* and through days 7-8 in *Fannia canicularis*. We failed to recover virus in similar studies conducted with the Lesser Mealworm, *Alphitobius diaperinus*. These findings suggest biosecurity measures should include an aggressive vector control program on and near infected premises to minimize possible contact between flies from contaminated poultry premises and susceptible poultry.



Session B, Monday, July 16, 2007
2:00–2:15 PM

The Macroscopic and Microscopic Pathology Associated with Outbreaks of Exotic Newcastle Disease in Commercial Poultry, and Pigeon Paramyxovirus Infection in Laughing Doves (*Streptopelia Senegalensis*) in South Africa

Neil M Duncan Diplomate ACVP M Med Vet (Aves) BVSc¹
Emily P Lane Diplomate ACVP M.Phil BVSc¹

¹Section of Pathology, Faculty of Veterinary Science, University of Pretoria, South Africa

Exotic Newcastle disease is endemic in South Africa and outbreaks occur in commercial poultry flocks when bio-security and immunization programmes are not stringently adhered to. Poorly immunized broiler flocks report mortalities of over 80% while pullet rearing operations record mortality figures of between 20 and 30%. Well immunized broiler flocks show increased mortality due to secondary E coli of up to 10% while layer flocks show a 10% drop in egg production and 0.5% increase in mortality. Viral isolation and typing revealed a goose origin paramyxovirus.

Macroscopic lesions:

Cyanosis
Pericardial fat and proventriculus: multifocal hemorrhage
Trachea: catarrhal to hemorrhagic tracheitis
GALT: necrosis and hemorrhage

Microscopic lesions:

Trachea: cilia loss, mucous gland necrosis, epithelial repair and lymphocyte infiltration
GALT: varying degrees of necrosis and hemorrhage

Deaths in groups of Laughing Doves were also reported at the same time.
PCR confirmed velogenic pigeon paramyxovirus

Macroscopic lesions:

Dehydration,
Haemoconcentration

Microscopic lesions:

Liver and pancreas: acute multifocal necrosis
Kidney: acute segmental tubular necrosis

Session B, Monday, July 16, 2007

2:15–2:30 PM

A “Poult Enteritis Mortality Syndrom” in Guinea fowl: a pathological and etiological study

Jean-Luc Guerin, Bertrand Grenier, Cyril Boissieu, and Caroline Lacroux

National Veterinary School of Toulouse - UMR INRA, ENVT 1225 .

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An emerging digestive disease has been identified in Guinea fowl farms in France. The clinical and pathological pictures of this disease are suggestive of PEMS (Poult Enteritis Mortality Syndrom).

The inoculation of 1-day-old guinea chicks with semi-purified 0.22 μm filtrate reproduced both prostration and diarrhoea, associated with a low mortality rate.

A molecular analysis was performed, using degenerate primers targeted to conserved sequences of known avian astrovirus: all “PEMS-like” birds were detected positive by astrovirus RT-PCR, in contrast to apparently healthy birds. This result was confirmed by electron microscopy.

Sequence analysis of both ORF 1b and ORF 2 demonstrated that so-called “Guinea fowl astrovirus” (GFAsV) is actually closely related to but distinct from TAsV2, one of the putative agents of PEMS.

Altogether, these results suggest that guinea fowl may be affected by a PEMS-like syndrome, associated with a specific Avian Astrovirus. Further epidemiological and pathological investigations are on the way to assess the causative involvement of the astrovirus in this condition.

2:30–3:00 PM

BREAK

Session B, Monday, July 16, 2007

3:00–3:15 PM

Moderator: Kate Barger

Newcastle Disease Virus Vaccine Potency Determination

K. Liljebjelke, D.J. King, D.R. Kapczynski

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Potency of inactivated Newcastle disease virus (NDV) vaccines is determined using vaccination and challenge. If the minimum killed viral antigen necessary for clinical protection can be determined, vaccines meeting or exceeding this dose might be considered of adequate potency. In these studies, correlation between hemagglutinin units per dose, protection from clinical disease and antibody response was determined. The PD_{50} of an in-house vaccine, and a commercially licensed vaccine were determined using the OIE approved method for testing vaccine potency. Results provide guidelines for minimum HA content for inactivated NDV vaccines necessary to ensure potency.



Session B, Monday, July 16, 2007

3:15–3:30 PM

Avian Adeno-associated virus-based expression of the Newcastle disease virus HN protein for poultry vaccination.

Francisco Perozo, Pedro Villegas, Carlos Estevez, Ivan Alvarado and Linda Purvis

Poultry Diagnostic and Research Center
University of Georgia
953 College Station Road, Athens, GA 30602

The avian Adeno-associated virus (AAAV) is a replication defective non-pathogenic virus member of the family *Parvoviridae* that has been successfully used for gene delivery. The generation of recombinant AAAV virions expressing the immunogenic hemagglutinin-neuraminidase (HN) of Newcastle disease virus (rAAAV-HN) was demonstrated by immunohistochemistry and electron microscopy. Serological evidence of systemic immune response was detected after *in ovo* or intramuscular inoculation of rAAAV-HN in specific pathogen free chickens using a commercial enzyme-linked immunosorbent assay and the hemagglutinin inhibition test. Newcastle disease virus neutralizing antibodies were demonstrated by the virus neutralization test performed in embryonating eggs.

3:30–3:45 PM

Development of a Quantitative Light Cycler Real–Time PCR for the Detection of Fowl Adenoviruses

Éva Nagy, Nadya Romanova and Juan Carlos Corredor

Dept. of Pathobiology, Ontario Veterinary College, University of Guelph
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A highly sensitive real–time PCR assay was developed and optimized to detect and quantify fowl adenoviruses (FAdV). The assay had a dynamic range of 6 logs and minimum detection limit of 20 copies of the FAdV-9 genome when plasmid DNA was used as a template. The SYBR Green real-time PCR assay exhibited high specificity and sensitivity. The assay was validated using virus DNA preparations derived from tissues of chickens infected with FAdV-9. Fowl adenovirus was detected in livers, bursa of Fabricius and cecal tonsils and the copy number ranged from 10^3 to 10^6 copies per 100 μ g of total DNA.

Session B, Monday, July 16, 2007

3:45–4:00 PM

Multiple Expressions of Inclusion Body Hepatitis

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Inclusion Body Hepatitis (IBH) diagnoses are increasing in U. S. commercial chicken flocks. Over the past five years, one integrator documented three different manifestations of IBH: there were differences in type of chicken, flock age, clinical signs and gross lesions. One case occurred in pullets, twice on the same farm. The second case occurred in broilers in during the first year of production in a new complex. The third case occurred in one house of broilers in an established complex. Clinical presentation and laboratory findings will be presented on each case.

4:00–4:15 PM

Physiochemical and molecular characterization of adenovirus-like virus (isolate R11/3), the putative etiology of transmissible viral proventriculitis

James S. Guy, John Barnes, and Fred Fuller

NCSU College of Veterinary Medicine, 4700 Hillsborough St., Raleigh, NC 27606

A novel adenovirus-like virus (AdLV [R11/3]) was identified as the likely etiology of transmissible viral proventriculitis based on isolation of the virus from diseased proventriculi and experimental challenge studies. AdLV (R11/3) was determined to have characteristics consistent with adenoviruses (intranuclear morphogenesis, icosahedral, ~70-nm diameter, non-enveloped, density ~1.32 in CsCl); however, immunohistochemical and PCR procedures using antisera and oligonucleotide primers specific for avian adenoviruses indicate that this virus is distinct from known avian adenoviruses. Identification of this virus is dependent upon additional studies particularly characterization of viral DNA and DNA sequencing. These studies are in progress.

Session B, Monday, July 16, 2007

4:15–4:30 PM

Histopathology of Transmissible Viral Proventriculus Caused by Adenovirus-like Virus (Isolate R11/3)

H. John Barnes and James S. Guy

Poultry Health Management Team

Department of Population Health & Pathobiology

College of Veterinary Medicine, North Carolina State University

Raleigh NC 27606-1499

Microscopic lesions in the proventriculus of 2-wk-old SPF layers and commercial broilers were determined during a 3-week period following inoculation with adenovirus-like virus (isolate R11/3). Degeneration and hyperplasia of ductal epithelium, necrosis of intermediate and glandular epithelium, increased interstitial lymphoid tissue, and replacement of lost glandular epithelium by ductal epithelium characterized changes caused by the virus. Lesions were present at day 3 and were maximal at day 10 in SPF layers and day 14 in commercial broilers. Lesions tended to be more severe and persist longer in broilers compared to layers. By day 21 lesions consisted of multiple lymphoid follicles, decreased glandular tissue, and increased ductal epithelium.

4:30–4:45 PM

Effect of maternal antibodies on vaccine-induced protection of turkey poults against virulent avian metapneumovirus subtype C challenge

Binu Velayudhan^{1*}, Sally Noll², Sagar Goyal³, David Halvorson¹, and Kakambi Nagaraja¹

¹Department of Veterinary and Biomedical Sciences, ²Department of Animal Science, and ³Department of Veterinary Diagnostic Medicine, University of Minnesota, Saint Paul, MN 55108.

The effect of maternal antibodies on vaccination of turkeys to protect against avian metapneumovirus (aMPV) challenge was examined. Maternal antibody positive and negative birds were further subdivided into three subgroups each. Birds in the first subgroup were kept as unvaccinated controls. Birds in the second and third subgroups were vaccinated by eyedrop and spray cabinet methods, respectively. We evaluated immune response, reduction in clinical disease and virus shedding post-challenge. Unvaccinated birds in the maternal antibody positive as well as maternal antibody negative groups showed clinical signs upon challenge. Birds in both maternal antibody positive and negative groups vaccinated by either eyedrop or spray cabinet vaccine did not show any clinical signs upon challenge. The results of RT-PCR, immunohistochemistry, and aMPV-ELISA will be discussed.

4:45 PM

ADJOURN



Session A, Tuesday, July 17, 2007
8:00–8:15 AM

Moderator: Timothy Cummings

Molecular Characterization of Reoviruses Isolated from Broilers with Runting and Stunting Syndrome Reveals a Lack of Homogeneity with Current U.S. Vaccine Viruses

Holly S. Sellers, Veronica Walker, Erich G. Linnemann, and Guillermo Zavala

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Avian reoviruses continue to contribute to economic losses even though reovirus vaccination is widespread throughout the U.S. Numerous reoviruses were isolated from severe cases of runting stunting syndrome in young broilers in the southeast. Following virus isolation, the sigma C protein (S1 gene) was sequenced and analysed. Comparison of the predicted amino acid sequences of the newly isolated reovirus field isolates with other U.S. reovirus vaccine and field isolates revealed less than 60% percent similarity. However, sequence comparisons with previously reported malabsorption isolates from Europe and Asia revealed a greater than 80% similarity within the sigma C protein. This is the first report of reovirus isolates of Eurasian lineage in the U.S.

8:15–8:30 AM

Extensive Analysis of Pathogenicity Island PAI 1_{APEC O1} of Avian Pathogenic *Escherichia coli*

Subhashinie Kariyawasam, Timothy J. Johnson, and Lisa K. Nolan

Department of Veterinary Microbiology and Preventive Medicine, College of Veterinary Medicine, Iowa State University, 1802 Elwood Drive, VMRI #2, Iowa State University, Ames, IA 50011

Colibacillosis caused by avian pathogenic *Escherichia coli* (APEC) is an economically important disease to the poultry industry worldwide. Recently, our laboratory identified several pathogenicity islands (PAIs) of APEC including PAI 1_{APEC O1} of an APEC O1:K1:H7 strain. We constructed mutants of this strain by deletion of PAI 1_{APEC-O1} or some of its genes. The virulence of these mutants was then tested in 12-day-old-chick embryos and 1-day-old and 2-week-old chickens to ascertain the contributions of PAI 1_{APEC O1} to the virulence of APEC.

Session A, Tuesday, July 17, 2007
8:30–8:45 AM

Analysis of the Complete Genome Sequence of an Avian Pathogenic *Escherichia coli*

Timothy J. Johnson^{A*}, Subhashinie Kariyawasam^A, Yvonne Wannemuehler^A, Paul Mangiamele^A, Sara J. Johnson^A, Curt Doetkott^B, Jerod A. Skyberg^A, Aaron M. Lynne^A, James R. Johnson^C, and Lisa K. Nolan^A

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*Presenting author, [email: timjohns@iastate.edu](mailto:timjohns@iastate.edu)

Avian pathogenic *E. coli* (APEC) are a form of extraintestinal pathogenic *E. coli* (ExPEC) causing colibacillosis in production birds. Colibacillosis continues to be problematic for the poultry industry, resulting in multi-million dollar annual losses in the United States alone. Here, we present the first genome sequence of a representative APEC strain, APEC O1 (O1 :K1 :H7). Analysis of this strain reveals 43 genomic islands (GIs), larger than 5 kb, which are not present in the *E. coli* K-1 2 backbone. Of these 43 GIs, most are present in all of the avian and human ExPEC thus far sequenced. We have recently begun a systematic analysis of these GIs for their occurrence among avian *E. coli* populations and their contributions to APEC's ability to cause disease. From these analyses, we have identified several GIs which occur frequently among APEC populations and contribute to APEC virulence. This presentation will provide a snapshot view of the APEC O1 genome and a detailed look at several of the GIs of APEC O1.

8:45–9:00 AM

Evaluation of Differentially Expressed Proteins following Serum Exposure in Avian Pathogenic *Escherichia coli*

Cynthia D. Tyler¹, Cheryl Lichti², and Steven L. Foley¹

¹National Farm Medicine Center

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Colibacillosis is an extraintestinal disease that causes great economic loss to the poultry industry. We used a proteomic approach to evaluate avian pathogenic *Escherichia coli* (APEC) proteins that were differentially expressed following exposure to chicken serum in hopes of identifying specific proteins that may be involved in serum resistance of APEC isolates. Proteins were isolated and separated by two-dimensional gel electrophoresis and protein spots corresponding to differentially expressed proteins were identified using electrospray ionization tandem mass spectrometry. Significantly, outer membrane protein A (OmpA) was among those proteins identified, indicating that differential regulation of OmpA may be involved in serum resistance.



Session A, Tuesday, July 17, 2007
9:00–9:15 AM

Role of large plasmids in the virulence of avian pathogenic *Escherichia coli*

Melha Mellata and Roy Curtiss III

Biodesign Institute (CIDV)
Arizona State University
1001 S. McAllister Avenue
Tempe, AZ, 85287-5401

Elucidating the role of large plasmids in virulence of avian pathogenic *E. coli* (APEC) is an important step to find the best way to eradicate avian colibacillosis. APEC strain χ 7122 (O78:K80) was cured of its large plasmids (pAPEC-1, pAPEC-2, and pAPEC-3). Despite the absence of selector markers in these plasmids, they were respectively transferred by conjugation into *E. coli* and *Salmonella typhimurium* recipient strains. Both *E. coli* and *Salmonella* transconjugants with pAPEC-1 produced Colicine V. Sequencing of pAPEC-1 (100kb) revealed that this IncFI plasmid harbors virulence factors involved in systemic infection of bacteria, including iron acquisition systems, *iss*, and *tsh*.

9:15–9:30 AM

***E. coli* challenge study in commercial broilers by either respiratory or skin route of exposure and the effect of prior vaccination with a live attenuated (aro-A) *E. coli*.**

Kalen Cookson¹ and Steve Davis²

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Wellington, CO 80549

E. coli infections in broilers can result in either cellulitis or respiratory disease/colibacillosis. This challenge study attempts to emulate these two conditions. Commercial broilers were housed in floor pens and half received a live *E. coli* vaccine by day-of-age spray. At 5 weeks of age all birds were challenged with a virulent *E. coli*, serotype O78, by either intratracheal or subcutaneous inoculation. Birds were posted at weekly intervals after challenge until the study was terminated at 49 days of age. The incidence of *E. coli* lesions, mortality and performance in the various groups will be discussed.

9:30–10:00 AM

BREAK

Session A, Tuesday, July 17, 2007
10:00–10:30 AM

Moderator: Richard Chin

LASHER HISTORY LECTURE:

**“Origin and Evolution of the American Association of Avian Pathologists:
50 Years of Dedication and Accomplishment”**

Dr. Richard Witter
Michigan State University

10:30–12:00 PM

AAAP BUSINESS MEETING

ROOM 207A

12:00–1:00 PM

LUNCH

1:00–1:15 PM

Moderator: David Ley

A Challenging Case of Suspected *Mycoplasma gallisepticum* Infection in Broiler Breeders

Frederic Hoerr, Lanqing Li, Samuel Christenberry, and C. Stephen Roney

Thompson Bishop Sparks State Diagnostic Laboratory
P.O. Box 2209, 890 Simms Road
Auburn, Alabama 36831-2209

Our work focuses on constructing a recombinant vaccine against *Eimeria* sp.. We are using bacterial Type Three Secretion System to deliver SO7 antigen into the cytoplasm of the immunized host cells and induce antigen-specific CTL responses. For delivery, SO7 was fused to the *Salmonella* gene *sopE*. In the host strain $\chi 9242$ ($\Delta phoP233 \Delta asdA16 \Delta araBAD23 relA198::araCP_{BAD} lacI TT$), SO7 is fused to the signal sequence of the *bla* gene and its expression is controlled by *lacI* repressor gene, whose expression in turn depends on free arabinose. The lack of arabinose in the tissues increases the expression of SO7 antigen *in vivo*.



**Session A, Tuesday, July 17, 2007
1:15–1:30 PM**

Epidemiology of *Mycoplasma synoviae* isolates in Georgia from 2000-present

V.A. Laibinis¹, S.H. Kleven¹, Louise Dufour-Zavala², Guillermo Zavala¹

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Polymerase chain reaction (PCR) of the *vlhA* gene is an important diagnostic tool for the determination of *Mycoplasma synoviae* (MS) infection. The advantage of using this specific PCR is that the product may be sequenced, giving information regarding relatedness of MS strains among outbreaks. Since the disease is transmitted both horizontally and vertically, sequencing may help to clarify the origin of the disease. Sequencing will establish relationships among organisms important in the epidemiology of disease. Phylogenetic trees will be presented.

1:30–1:45 PM

The Evaluation of *Mycoplasma gallisepticum* Challenge Routes

N.M. Ferguson-Noel, Z. Raviv, V.A. Leiting, R. Wooten, and S.H. Kleven^A

Department of Avian Medicine,
University of Georgia, Athens, GA 30602-4875

It is often difficult to closely replicate field challenges under experimental conditions. In this study we attempt to evaluate different routes of challenging chickens with *Mycoplasma gallisepticum* (MG). The routes evaluated included aerosol challenge, aerosol challenge with a diluted culture, intraocular challenge and intratracheal challenge. Our goal was to identify a consistent challenge method that allows the accurate evaluation of the virulence of MG strains and the safety and efficacy of MG vaccines.



Session A, Tuesday, July 17, 2007
1:45–2:00 PM

Intraspecific Differentiating Real-Time PCR for *Mycoplasma gallisepticum* Live Vaccine Evaluation

Stanley H. Kleven, Ziv Raviv, Scott A. Callison, N. Ferguson-Noel
Department of Population Health, Poultry Diagnostic and Research Center,
The University of Georgia, 953 College Station Rd., Athens, GA 30602-4875.

Mycoplasma gallisepticum causes respiratory disease and production losses in poultry. Vaccination of poultry with *Mycoplasma gallisepticum* live vaccines was demonstrated to be an efficacious approach to reduce susceptibility to infection and to prevent the economic losses. The development and evaluation of live vaccines usually requires the involvement of several vaccine and challenge strains in the same experimental setup. Our goal was to develop a tool to allow the absolute differentiation between a set of known *Mycoplasma gallisepticum* strains in a quantitative manner. We developed 5 real-time PCR assays that absolutely differentiated between one of the five commercial and laboratory vaccine strains: F, ts-11, 6/85, K5831, K5054, and the standard challenge strain R_{low} when tested on *in vitro* cultures. The assay K5831 vs. R_{low} was also tested on specimens from live birds that were vaccinated with K5831 and challenged with R_{low}, and successfully differentiated between the vaccine and the challenge strains in a quantitative manner. This preliminary *in vivo* application of the method also shed light on possible protection mechanisms for the *Mycoplasma gallisepticum* K5831 vaccine strain.

2:00–2:15 PM

Effects of TS-11 Strain *Mycoplasma gallisepticum* Vaccination in Broiler Breeders Infection and Immunity on the Breeders and its Progeny

Francisco J. Rojo¹, Alejandro Rojas², Rafael J. Fernandez¹
¹Merial Avian Global
²Merial Mexico

Breeder pullets were vaccinated at 28 days of age with the *Mycoplasma gallisepticum* TS-11 vaccine in the rearing farm. At 22 weeks the birds were moved to a multi-age-layer production farm. Birds were monitored by serology using rapid slide agglutination (RSA) and hemagglutination inhibition test (HI). With the use of FTA cards, tracheal swabs were taken from breeders at different ages to determine the presence of the vaccinal strain. In the progeny birds were sampled at the hatchery. Tracheas from day-old-chicks were pooled to determine the presence of *Mycoplasma* by PCR test.



**Session A, Tuesday, July 17, 2007
2:15–2:30 PM**

**Effect of Selected Water Temperatures Used in *Mycoplasma gallisepticum* Vaccine
Reconstitution/Dilution on Color Change Units at Selected Time Intervals**

**S. L. Branton, S. A. Leigh, J. D. Evans, W. B. Roush, J. L. Purswell,
H. A. Olanrewaju, and S. D. Collier**
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Numerous methods are currently utilized throughout the poultry industry for the administration of vaccines. Each uses water for vaccine reconstitution and/or administration, including two of the three commercially available live *Mycoplasma gallisepticum* (MG) vaccines. Selected water temperatures were used to reconstitute and/or dilute the three commercially available live MG vaccines. Water temperatures included 4 C, 22 C (room temp.), and 32 C. Results showed significant differences among the three water temperatures on vaccine viability as determined by color change units (CCU) for F strain MG and for ts-11 while no significant difference was noted for 6/85 MG.

2:30–2:45 PM

Use of the microdilution method for the detection of the *in vitro* antimicrobial susceptibility against avian mycoplasmas.

Ariel Ortiz, Ernesto Soto and Clemente Lemus.
Universidad Autónoma de Nayarit, Posgrado en Ciencias Biológico Agropecuarias.
Carretera Tepic-Compostela. Km 9 Xalisco, Nayarit. Mexico CP 63780.

In vitro mycoplasma sensitivity tests to different antimicrobials were performed. The methodology suggested by the National Committee for Clinical Laboratory Standards (NCCLS) was used and adapted for the micro-dilution test. Tests were carried out using *Mycoplasma synoviae*, WVU 1853 strain, and five antibiotics.

Results indicate that tylosin tartrate shows the lowest MIC (0.156 µg/ml), which is statistically different to Lincomycin HCL (0.781 µg/ml) that shows the highest MIC, but not to Enrofloxacin HCL (0.25 µg/ml) or to Chlortetracycline HCL (0.25 µg/ml). It was not possible to determine the MIC for Erythromycin estolate because no anti-mycoplasmic activity was detected at any dilution tested.



Session A, Tuesday, July 17, 2007
2:45–3:00 PM

Antibiotic resistance in *Mycoplasma gallisepticum* and *Mycoplasma synoviae* isolates from meat-type turkeys in Israel

Sharon Levisohn*, Irina Gerchman and Shimon Perk
Division of Avian and Aquatic Diseases,
Kimron Veterinary Institute, Bet Dagan Israel

Little is known about antibiotic susceptibility patterns of *Mycoplasma gallisepticum* (MG) and *M. synoviae* (MS) field isolates. Our study compared isolates from meat-type turkey flocks in 2005-06 with archived and reference strains. Minimal inhibitory concentration (MIC) values for six antibiotics (fluoroquinolones, macrolides and tetracyclines) were determined by three methods. Compared to archived strains, MICs of fluoroquinolones in recent MG isolates (5 out of 6 flocks tested) were above the therapeutic range whereas, except for one flock, MS isolates were sensitive. Resistance to macrolides was detected in several archived MG isolates and in one recent flock, but not at all in MS.

3:00 PM

ADJOURN

5:00–11:00 PM

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**Session B, Tuesday, July 17, 2007
8:00–8:15 AM**

Moderator: Alejandro Banda

IFN- α regulates infectious bursal disease virus induced macrophage activation.

Mahesh Khatri and Jagdev M. Sharma

Department of Veterinary and Biomedical Sciences, College of Veterinary Medicine, University of Minnesota, 1971 Commonwealth Avenue, St. Paul, MN 55108, USA

Infectious bursal disease virus (IBDV) causes an acute, highly contagious and immuno-suppressive disease in chickens. The virus infects and destroys actively dividing IgM-bearing B cells in the bursa. We have shown previously that the virus also infects and replicates in macrophages. Infection with IBDV causes production of proinflammatory mediators and cytokines in the macrophages. Toll-like receptors (TLRs) mediate innate immune responses to microbes. Here we have analyzed TLRs mRNA expression in chicken spleen macrophages infected with IBDV. Viral infection enhanced TLR3 and melanoma differentiation-associated gene 5 (mda-5) mRNAs expression. TLR3 and mda-5 are receptors for dsRNA. Neutralizing anti-IFN- α/β antibodies down-regulated gene expression of TLR3 and mda-5. The data suggests that IFN- α plays a role in the activation of innate immunity.

8:15–8:30 AM

Cell-mediated immunity induced by a macrophage-adapted infectious bursal disease virus

Jagdev M. Sharma and Mahesh Khatri

Department of Veterinary and Biomedical Sciences, College of Veterinary Medicine, University of Minnesota, 1971 Commonwealth Avenue
St. Paul, MN 55108, USA

We have shown previously that cell-mediated immunity (CMI) is important for defense against virulent IBDV. We adapted a virulent, highly immunogenic strain of IBDV to a chicken macrophage cell line, MQ-NCSU. Macrophage-adapted virus induced minimal bursal lesions, high antibody levels and up-regulated the expression of IFN- γ mRNA and protected chickens against challenge with virulent IBDV. The IFN- γ upregulation was more pronounced in birds exposed to macrophage-adapted virus than in those exposed to chick embryo fibroblast-adapted virus. These data indicated that macrophage-adapted IBDV induced a strong cell mediated protective immune response in chickens.



Session B, Tuesday, July 17, 2007
8:30–8:45 AM

Identification of Infectious Bursal Diseases Viruses from several countries

Pedro Villegas, Linda Purvis, Francisco Perozo and Taylor Barbosa

University of Georgia
Poultry Diagnostic & Research Center
953 College Station Rd.
Athens, Georgia 30602

Identification of infectious bursal disease viruses from different countries was performed using bursal impressions on FTA paper or bursas fixed in formalin and embedded in paraffin. All samples were analyzed using either the real time RT-PCR or the normal RT-PCR tests. Very virulent strains of infectious bursal disease virus (vvIBDV) were detected in numerous countries, indicating that the presence of this virus in the world's poultry industry is increasing, causing mortalities and severe immunosuppression if affected flocks. The presence of variant strains of the virus was also detected. Vaccination programs to control the vvIBDV varied significantly in the different countries.

8:45–9:00 AM

Impact of IBD vaccines on the bursa and other immune tissues of commercial broilers

Enrique Montiel, Nikki Pritchard and Julio Cruz-Coy

Merial- Select Inc
1112 Airport Parkway
Gainesville GA 30501

The impact of live Infectious Bursal disease (IBD) vaccines on the chicken immune system is an important factor for the protection the birds may develop later, not only against IBD but also against other diseases. Egg from a single broiler breeder flock with no IBD maternal antibodies were either vaccinated at 18 days of embryonation or hatched and vaccinated at various ages with commercially available and experimental IBD vaccines. Histopathological evaluation of bursa, spleen and thymus and protection against Newcastle disease were used to measure the immunosuppressive potential of the IBD vaccines used during the trial.



Session B, Tuesday, July 17, 2007
9:00–9:15 AM

Challenge studies with Infectious Bursal Disease and Chicken Anemia Virus field strains

Linda B. Purvis, Pedro Villegas, Francisco Perozo and John Smith
Department of Population Health, University of Georgia
Athens, GA 30602

Farms experiencing recurrent problems with immunosuppression and gangrenous dermatitis were molecularly surveyed for Infectious Bursal Diseases Virus (IBDV) and Chicken Anemia Virus (CAV). Some molecular changes for both IBDV and CAV were found on a farm with a severe history of immunosuppression problems. This isolate (MM strain) was passed in birds, and titrated for use in a challenge study to determine the effects this strain had on immunosuppression and production.

Broilers and SPF birds were used in the study. Birds were inoculated with the IBDV and CAV MM strains at different time intervals and later vaccinated with a Newcastle disease vaccine. Antibody responses and histopathological evaluations will be presented.

9:15–9:30 AM

Efficacy of a vectorized commercial vaccine against Infectious Bursal Disease in Layers.

**Eliana Icochea¹, Blanca Talavera², Elmer Davila², Rosa Gonzalez¹, John Guzman¹,
Hermelinda Rivera¹.**

¹College of Veterinary Medicine, University of San Marcos, Lima-PERU.

²Inversiones Veterinaria S.A

The objective of this trial was to evaluate the efficacy and the immune response against Infectious Bursal Disease using a vectorized commercial vaccine. Four hundred Isa Brown pullets at day old were divided in four groups according to the following vaccination programs: group A (vectorized vaccine), group B (vectorized vaccine plus SB1 Marek vaccine strain), group C (traditional vaccination program plus HVT Marek vaccine strain) and group D (control). The birds were challenged at 35 days old with the F52/70 challenge strain. Mortality, gross and microscopic lesions, bursal index, clinical signs and immune response (ELISA and VN) were evaluated at 3, 6, 10 and 20 days after challenged. Lymphoid tissues were collected at 16 and 20 weeks of age for histological study. The results will be statistically analyzed.

9:30–10:00 AM

BREAK

Session B, Tuesday, July 17, 2007

Moderator: Rich Chin

10:00–10:30 AM

LASHER HISTORY LECTURE:

**“Origin and Evolution of the American Association of Avian Pathologists:
50 Years of Dedication and Accomplishment”**

Dr. Richard Witter
Michigan State University

10:30–12:00 PM

AAAP BUSINESS MEETING

ROOM 207A

12:00–1:00 PM

LUNCH

1:00–1:15 PM

Moderator: Peter Woolcock

REED RUMSEY AWARD

**Cellular and Cytokine Responses Associated with Dinitrofluorobenzene-Induced Contact
Hypersensitivity in Chicken**

**Mohamed F. Abdul-Careem (M.V.M., B.V.Sc), D. Bruce Hunter, Melissa D. Lambourne, Hamid R.
Haghighi, Niroshan Thantrige-Don, Shayan Sharif**

Department of Pathobiology, Ontario Veterinary College, University of Guelph, Canada

The objective of the study was to determine the cellular and cytokine responses associated with dinitrofluorobenzene (DNFB)-induced skin contact hypersensitivity, which is a form of cell-mediated immune response, in chickens. Chickens were first sensitized by topical administration of DNFB on the breast. Seven days later, DNFB was topically administered to the left foot webs of sensitized chickens, whereas the right foot webs were treated with the vehicle. Double-fold thickness of the left foot webs was increased by 6 hrs post-induction (pi), peaked by 24 hrs and then declined gradually by 48 and 72 hours pi. The foot web skin was sampled at 6, 12, 24, 48 and 72 hrs pi for histology, immunofluorescence and real-time RT-PCR. In foot webs treated with DNFB, eosinophil infiltration was observed. The tissue infiltration by eosinophils peaked by 6 hrs pi and then subsided by 24 hrs pi. Mononuclear cells were first observed at 6 hrs pi and gradually increased by 48 hrs pi. Among mononuclear cells, CD4+ and CD8+ T cells were the dominant subsets. The expression of cytokines in DNFB-induced foot web skin samples was compared to untreated foot webs. The peak of interleukin (IL)-4, IL-6, IL-10 and interferon (IFN)- γ gene expression was at 6 hrs pi, which all declined gradually, except for IL-10 which remained unchanged until 24 hrs pi and then declined. In conclusion, DNFB-induced skin hypersensitivity in chickens was associated with infiltration of eosinophils followed by CD4+ and CD8+ T cells. The event was also associated with early up regulation of cytokines genes.



**Session B, Tuesday, July 17, 2007
1:15–1:30 PM**

Serum and yolk antibody titers in late stage broiler embryos

Alan P. Avakian, Donald Link and John Dickson

Embrex, Inc.
PO Box 13989, RTP, NC, 27709-3989

Maternal antibody is known to interfere with post hatch vaccinations in broilers. Less is known about maternal antibody levels and its impact on vaccines delivered *in ovo*. Yolk and blood of broiler embryos were sampled on days 17.0 to 20.0 of incubation and percent yolk absorbed was scored. Serum was collected post hatch. Yolk and serum antibody titers to infectious bursal disease and infectious bronchitis were determined by ELISA. Embryonic serum antibody titers were significantly lower on days 17-19 of incubation than on day of hatch. Yolk titer, percent yolk absorbed and embryonic age were significant predictors of embryonic serum antibody titer.

1:30–1:45 PM

Validation of a Multiplexed Fluorometric ImmunoAssay™ (MFIA™) for Health Monitoring of Specific Pathogen Free Chickens

Joe H. Simmons, Elena Seletskaja, Theodore Girshik, Rajeev K. Dhawan and William R. Shek

Charles River Laboratories
Research Animal Diagnostic Services
251 Ballardvale St.
Willmington, MA 01887

Early detection of adventitious infections in specific-pathogen-free (SPF) chicken flocks supplying embryonated eggs for vaccine production and research is critical. To meet this challenge, Charles River Laboratories Avian Division (SPAFAS) performs weekly health monitoring on its SPF chicken flocks for an extended panel of adventitious infectious agents resulting in over 3 million individual serology assays each year. To facilitate and expedite this high-volume testing, we have developed an avian Multiplexed Fluorometric ImmunoAssay™ (MFIA™) for a comprehensive panel of adventitious infectious agents, using Luminex's Multi-Analyte Profile (xMAP®) Technology. These assays have been validated by comparison of common assay performance characteristics (analytical sensitivity and specificity, and diagnostic sensitivity and specificity) versus traditional serology techniques including ELISA, IFA, and AGP using sera from a large number known negative and known positive birds.



**Session B, Tuesday, July 17, 2007
1:45–2:00 PM**

**Bursal Disease-Marek's Disease Vaccine, Live Marek's Disease Virus Vector, Serotype 3,
(VAXXITEK™) vaccination effect on NDV vaccination schedules
in commercial broiler type birds.**

**Rafael Fernandez, Mike McCabe, Francisco Rojo, Hector Garcia,
and Julio S. Cruz-Coy**

1Merial Select, Inc., 1168 Airport Parkway, Gainesville, GA 30501

One-day-old commercial broilers were SQ vaccinated with a Bursal Disease-Marek's Disease Vaccine, Live Marek's Disease Virus Vector, Serotype 3, (VAXXITEK™) or with a combination of HVT and a highly virulent IBDV vaccine. Seven days after the initial vaccinations the birds in all the groups were vaccinated with an inactivated NDV vaccine alone or in combination with a live NDV B1 type, LaSota strain vaccine. Four weeks after the NDV vaccinations, the birds in the vaccinated groups and a non-vaccinated control group were challenged with the NDV GB Texas strain, and observed for 14 days. The purpose of the IBDV vaccination with a highly virulent strain was to mimic field immunosuppression conditions and the way immunosuppressed birds would respond to NDV vaccination and subsequent challenge. Results will be presented.

2:00–2:15 PM

Antibody-antigen Complexes Protect Against Chicken Infectious Anemia Virus Infection

Karel A. Schat, Priscilla O'Connell and M. Piepenbrink

Dept. of Microbiology and Immunology

College of Veterinary Medicine

Cornell University, Ithaca NY 14853

Chicken infectious anemia virus (CIAV) causes subclinical immunosuppression when birds become infected after 3 weeks of age. Maternal antibodies protect against virus infection and may interfere with replication of vaccine virus. We investigated the possible use of CIAV-antibody complexes for the induction of protective immunity. Optimal levels of virus and antibodies were established and used to vaccinate maternal antibody-positive and -negative one-day-old chickens. Virus replication levels and antibody development was determined. Vaccinated birds were challenged with a second CIAV. Protection was measured by real-time PCR using primer sets, which differentiate between the two viruses. Results of vaccination will be discussed.



**Session B, Tuesday, July 18, 2007
2:15–2:30 PM**

**The effect of vaccination against chicken anemia virus on colonization,
shedding and susceptibility to infection with Salmonella typhimurium in broilers**

Franz Sommer, Carol J. Cardona

PHR - UC Davis
One Shields Avenue
Davis, Ca 95616

Infections with chicken anemia virus (CAV) are common in commercial broilers, and are, despite their negative effect on the immune system, widely thought to be of lesser importance. However, infections of broilers with Salmonellae are considered to be important because of a possible intake into the human food chain.

To gain knowledge about the interactions between CAV and Salmonella typhimurium under different vaccinations regimens, we vaccinated groups of birds with live and killed CAV vaccines, and challenged parts of these groups with S. tm. to compare the results to those of unvaccinated and challenged birds. The results of these experiments will be presented.

2:30–2:45 PM

**A Study of the Viral Shed of Laryngotracheitis in Cornish Birds at the Processing Plant during a
Regional Outbreak and Area Wide LT Vaccination**

Bret Rings¹ and Steve Breeding

¹Tyson Foods, Inc., Springdale AR 72702
University of Arkansas Diagnostic Lab, Springdale, AR 72706

During an outbreak of laryngotracheitis (LT) in northwest Arkansas and northeast Oklahoma, all broilers in the region were vaccinated for LT with the exception of a 2.3 pound Cornish class bird. Because broilers can shed the virus before showing clinical signs, there was concern that Cornish birds going to slaughter could be shedding LT virus and exposing birds on other farms during live haul. Two to four Cornish flocks were sampled over a 3 month period to determine the LT prevalence in this class of birds at the processing plant. Complete tracheas were removed at the plant and placed in whirl-pak bags and submitted to the University of Arkansas Diagnostic Lab in Springdale, Arkansas for LT PCR. The results from this study will be presented and discussed.

**Session B, Tuesday, July 17, 2007
2:45–3:00 PM**

**Challenge Study to Evaluate Vaccine Protection against
Infectious Laryngotracheitis Virus (ILTV)**

Andrés Rodríguez-Avila and Maricarmen García

Poultry Diagnostic and Research Center, Department of Population Health,
College of Veterinary Medicine, The University of Georgia,
953 College Station Road, Athens, GA. 30602.

A challenge study was conducted to evaluate the protection elicited by a chicken embryo origin (CEO) vaccine against a currently circulating ILTV field strain. Protection was evaluated by scoring clinical signs (morbidity), mortality, and weight of vaccinated chickens as compared to non-vaccinated/challenge and contact-exposed chickens. Shedding and transmission of the challenge virus to contact-exposed chickens was evaluated by virus isolation and real-time PCR. Briefly, a total of 60 four week-old specific pathogen free (SPF) chickens were placed in negative pressure HEPA filtered isolator units. At 4 weeks of age ten chickens were vaccinated via eye-drop with CEO vaccine with the recommended full dose and kept separated from the other chickens. At 6 week of age ten vaccinated chickens and ten non-vaccinated chickens were inoculated intra-ocularly and intra-tracheally with 200 μ l of 3.0 log₁₀ TCID₅₀ of the challenge virus, and placed in separate units to commingle with ten non-vaccinated/non-challenge contact-exposed chickens. A third group of twenty chickens was kept uninoculated as negative control. Eye conjunctiva, trachea, and cloacal swabs were collected from two contact-exposed chickens from day 2 to 10 post-challenge. Differences in morbidity mortality and weight were observed between vaccinated, non-vaccinated/challenged, and contact-exposed chickens. Viral DNA was detected and virus isolated only from contact-exposed chickens in the non-vaccinated/challenged group, but not in contact-exposed chickens in the vaccinated/challenged group. Results showed that the utilized vaccine protected against morbidity, mortality, and viral shedding.

3:00 PM

ADJOURN

5:00–11:00 PM

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**Session A, Wednesday, July 18, 2007
8:00–8:15 AM**

Moderator: Kalen Cookson

Vertically Transmitted Viral Arthritis (Reovirus) in Commercial Broilers: A Field Case Report

Suzanne Young Stamey, Calvin Anthony, Kevin Crider and Todd Cartwright

Pilgrims Pride, Inc.
110 S. Texas Street
Pittsburg, Texas 75686

Commercial broilers at 8-11 days of age were reported to have gait deficits (wing walking) and huddling around feeders. Within an affected flock anywhere from a 15 - 75% of the flock was reported to show clinical signs when approaching the end of grow out (~38d). Initially gross lesions included reddened hock joints and femoral head necrosis; however lesions progressed to non-purulent synovitis and thickened gastrocnemius tendon 10-15 days after first showing signs. This presentation will review the clinical and gross signs and epidemiology of the outbreak. The diagnostics and challenge studies will be discussed in another presentation.

8:15–8:30 AM

**An Outbreak of Tenosynovitis in Broilers Caused by
Vertically-Transmitted Reovirus**

J.J. Courtney, M.M.E. Andersen, G. Zavala, H.S. Sellers, S.M. Williams

The University of Georgia, Poultry Diagnostic and Research Center
Dept. of Population Health, Athens, GA 30602

Birds from multiple broiler flocks were submitted for necropsy to the Poultry Diagnostic and Research Center (PDRC) at the University of Georgia with a history of swollen hocks and inability to walk. Two young breeder flocks were identified as suspect reovirus shedders based upon hatchery and field records. Serology demonstrated rapid seroconversion in the breeder flocks identified as suspects, which continued to shed virus for at least twelve weeks. Reovirus was isolated from tendons, visceral organs, and intestinal contents. All virus isolates were identical to each other and were genotypically different from conventional vaccine isolates. The viral isolates were inoculated into SPF broilers and caused severe tenosynovitis and mortality.



**Session A, Wednesday, July 18, 2007
8:30–8:45 AM**

**Unusual Case Report: Hemochromatosis due to Lead Poisoning
in a Wild Mallard Duck**

Scott D. Fitzgerald,¹ Jessica S. Hoane,¹ and Thomas M. Cooley²

¹Diagnostic Center for Population & Animal Health, and
²Wildlife Laboratory, MI Department of Natural Resources,
4125 Beaumont Road, Lansing, MI 48910-8104

A wild mallard duck was found in a pond located at the local zoo, exhibiting weakness, and neurologic signs including head bobbing; it was captured and euthanized. Gross findings were limited to emaciation. Diseases of greatest concern included West Nile virus, Newcastle disease, and avian influenza. Virologic tests were negative, and histopathology failed to demonstrate inflammation in any tissue. However, both the liver and kidneys contained large amounts of brown pigment which stained positively for iron. Causes of intravascular hemolysis in ducks include leucocytozoonosis and lead poisoning. The liver demonstrated toxic levels of both iron and lead on mineral analysis.

8:45–9:00 AM

Vitamin E Deficiency Induced Cardiomyopathy in Commercial Pekin Ducks

Richard M. Fulton

Diagnostic Center for Population and Animal Health
and
Pathobiology and Diagnostic Investigation
Michigan State University
4125 Beaumont Road
Lansing, Michigan 48910-8104

Multiple 13-day-old commercial Pekin ducks were submitted for diagnostic investigation due to a recent rise in daily mortality. Gross examination revealed a hydropericardium, an enlarged dilated heart with marked pallor of the ventricular myocardium. Microscopically, there was myodegeneration and myonecrosis with heterophils and macrophages within the necrotic cardiac myofibers and within the interstitium. Analysis of liver Vitamin E levels revealed levels below normal.



Session A, Wednesday, July 18, 2007
9:00–9:15 AM

Field Report: Acute Sodium Toxicosis in Turkey Poults

Charles Corsiglia, Rocio Crespo, Richard Chin, H. L. Shivaprasad, Murugan Subbiah, Birgit Puschner, Arthur Bickford
Foster Farms, Delhi, CA

This field report will describe a case of suspected sodium toxicosis in young commercial turkey poults. A review of the history, diagnostic results, pathology, and potential cause of acute sodium toxicosis will be presented.

9:15–9:30 AM

Effect of novel intestinal anaerobes on early development of the broiler intestine.

Margie D. Lee, Brett Lumpkins, Youngjae Cho, and Amy Batal
Depts. of Population Health and Poultry Science, The University of Georgia, Athens, GA 30602

To evaluate the effects of novel intestinal anaerobes on the development of the intestine, Cobb male chicks were orally inoculated with novel species of *Bacteroidaceae*, *Clostridiaceae*, or a combination of the two. Throughout the experiment birds were fed a standard corn-soy ration. The performance parameters were similar among the 4 treatments however, the birds inoculated with the combination of *Bacteroidaceae* and *Clostridiaceae* were observed to have increased villi height and goblet cell concentration during the first three days of age. These data indicate that some bacteria can act to stimulate intestinal development however a complex bacterial community may be necessary for optimal early development.

9:30–10:00 AM

BREAK

Session A, Wednesday, July 18, 2007
10:00–10:15 AM

Moderator: Scott Fitzgerald

***Clostridium perfringens* and *Clostridium Septicum* in Commercial Poultry – Recent findings**

Dr Daniel Karunakaran
Agtech Products, Inc.
W227 N752 Westmound Drive
Waukesha, WI 53186

There has been an increase in *Clostridial* diseases in the US poultry in the recent years. The industry has been challenged with necrotic enteritis, gangrenous dermatitis in broilers and cellulitis in turkeys. Over the past several years Agtech Products, Inc. and poultry production companies have collaborated on several research projects focusing on developing a better understanding of *Clostridial* disease challenge in commercial poultry in the US. During this time several hundreds of *Clostridial* isolates have been characterized using molecular techniques. The characterization data from the trials will be presented. Research data on the mode of *Clostridial* diseases also will be discussed.

10:15–10:30 AM

Comparison of level of Necrotic Enteritis in broilers vaccinated with either an attenuated or non-attenuated live coccidial vaccine and challenged with *Clostridium perfringens*

G.F. Mathis
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2011 Brock Road, Athens, GA 30607

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University of Georgia,

J. Chapman and E. Katigbak
Merial Select
1112 Airport Parkway
Gainesville, GA 30503

The objective was to compare the level of Necrotic Enteritis in *Clostridium perfringens* challenged broilers vaccinated with either an attenuated or non-attenuated live coccidial vaccine. The treatments were non-vaccinated, non-challenged or challenged, coccidian vaccinated with attenuated or non-attenuated vaccine and challenged. Vaccination was performed on day of hatch, prior to placement. Challenged birds received *Clostridium perfringens*. Presence of litter oocysts in all vaccinated pens confirmed viability of both vaccines. The non-attenuated vaccinated birds had higher percent NE mortality and NE lesion scores and lower performance at Days 22 and 42 compared to attenuated vaccinated birds. The results demonstrate that the severity of Necrotic Enteritis was not as great in birds challenged with attenuated strains of coccidia than the non-attenuated coccidial strains.

Session A, Wednesday, July 18, 2007
10:30–10:45 AM

Innate immune response to *Clostridium perfringens* and *Eimeria maxima* in necrotic enteritis model

**Hyun S. Lillehoj¹, Soon S. Park¹, Patricia C. Allen¹, Dong Woon Park¹
Steve FitzCoy², Daniel A. Bautista³,**

¹Animal Parasitic Diseases Laboratory, ANRI, BARC, USDA-ARS, Beltsville, MD20705

²Schering-Plough Animal Health, Millsboro, DE 19966 ³Lasher Poultry Diagnostic Laboratory,
University of Delaware, Georgetown, DE19947

The incidence of necrotic enteritis (NE) due to *Clostridium perfringens* (CP) infection in commercial poultry has been increasing at an alarming rate. While pre-exposure of chickens to coccidia infections is believed to be one of the major risk factors leading to NE, the underlying mechanisms of CP virulence remain undefined. The objectives of this study were to utilize an experimental model of NE produced by *Eimeria maxima* (EM) and CP coinfection to investigate the pathological and immunological parameters of the disease. Broilers coinfecting with EM plus CP exhibited more severe gut pathology compared with animals given EM or CP alone. Additionally, EM/CP coinfection increased the numbers of intestinal CP bacteria compared with chickens exposed to an identical challenge of CP alone. Coinfection with EM and CP repressed nitric oxide synthase gene expression that was induced by EM alone, leading to lower plasma NO levels. Intestinal expression of a panel of cytokine and chemokine genes following EM/CP coinfection showed a mixed response depending on the transcript analyzed and the time following infection. In general, IFN- α , IFN- γ , IL-1 β , IL-2, IL-12, IL-13, IL-17, and TGF- β 4 were repressed, while IL-8, IL-10, IL-15, and LITAF were increased during coinfection compared with challenge by EM or CP alone. These results are discussed in the context of EM and CP to act synergistically to create a more severe disease phenotype leading to an altered cytokine/chemokine response than that produced by infection with the individual pathogens.

10:45–11:00 AM

Attempts to Develop an Oral Challenge Model for Gangrenous Dermatitis using *Eimeria maxima* and *Clostridium septicum*

M.M.E. Andersen, S.R. Collett, J.J. Courtney

Poultry Diagnostic and Research Center

University of Georgia

953 College Station Road

Athens, GA 30602

Gangrenous dermatitis (GD) can be reproduced by challenge with the etiologic agents through the subcutaneous, intravenous, or oral route. While subcutaneous challenge most consistently reproduces clinical disease, it only allows for the study of factors affecting the consequence of infection. The oral model is more versatile in that it allows for examination of predisposing factors affecting both the development of disease and the consequences of infection. Several attempts were made to improve the predictability of an oral challenge model using a concomitant challenge of *Eimeria maxima* and *Clostridium septicum*.



**Session A, Wednesday, July 18, 2007
11:00–11:15 AM**

Development of an experimental challenge model for Cellulitis in turkeys.

Anil J. Thachil., Binu T. Velayudhan., David A. Halvorson, and Kakambi V. Nagaraja,
Department of Veterinary and Biomedical Sciences,
University of Minnesota, 1971 Commonwealth Ave, St. Paul, MN 55108.

Cellulitis in turkeys has been identified as an emerging infectious disease in adult turkeys. Cellulitis lesions in turkeys occur on the breast and ventral abdomen with apparent absence of any external wound. The infectious agents isolated are *Clostridium perfringens* and *Clostridium septicum*. The objective of our study was to develop a challenge model for subsequent development and evaluation of a vaccine preparation in turkeys. We selected isolates from cellulitis in field turkeys. The standardized preparation was evaluated experimentally for reproduction of cellulitis and mortality in 6 and 8-week-old turkeys. The results of this experimental challenge model will be presented.

11:15–11:30 AM

**Field investigation of Mycoplasma infection outbreaks in
The Southeastern United States**

**Guillermo Zavala¹, M. Early-Andersen¹, L. Dufour-Zavala²,
V. Leiting¹, and S. H. Kleven¹**

¹Department of Population Health, University of Georgia

²Georgia Poultry Lab, Oakwood, Georgia

A field investigation was conducted on recent outbreaks of *Mycoplasma gallisepticum* (MG) and *M. synoviae* (MS). The *mgc2* and *vlhA* genes of MG and MS, respectively, are routinely used for molecular detection of MG and MS. During a recent field investigation, the *mgc2* gene was unable to differentiate between wild and vaccine-related strains of MG. Instead, the highly conserved intergenic spacer region (IGSR) DNA sequence efficiently differentiated wild from vaccine-related MG strains in field outbreaks and was successfully used to link epidemiologically related outbreaks in commercial poultry. The *vlhA* gene of MS was a suitable target gene for molecular epidemiology investigations of MS outbreaks.



**Session A, Wednesday, July 18, 2007
11:30–11:45 AM**

Correlation of Experimentally Induced Colisepticemia with Septox Condemnations in Broilers

Drs. Timothy S. Cummings, Marty Ewing, Floyd Wilson, Mark Burluson
Mississippi State University, Mississippi State, MS 39762

Broilers were challenged with the most common bacterial cause of sepsis in poultry (*E. coli*) in order to help document true gross and microscopic septicemic changes over time. Samples and pictures were taken from the challenged birds over a period of three weeks which were then compared with tissues and digital images from actual carcasses which had been condemned for septox. Gross and/or microscopic lesions of sepsis were not consistently appreciated in birds that were condemned in the septox category. These findings further elucidated the pathological spectrum of carcasses comprising the septox disposition and implies that the septox category needs to be critically reevaluated.

11:45–12:00 PM

Antibiotic Resistance Plasmid Eliminated from Poultry Bacteria: New Scientific Evidence to Counter the Expert Critics and Spin-Factories

Steven R. Clark, Jeremy J. Mathers
Alpharma, Inc. Animal Health Division
Bridgewater, New Jersey

On-going research is demonstrating, contrary to most public media, that antimicrobials can have a positive benefit to the issue of antibiotic resistance by actually reducing or eliminating R-plasmids – in addition to proven safety, efficacy, environmental and economic benefits. Compounds are evaluated in the presence of plasmid-bearing microorganisms and are monitored via differential plating methodologies for the evidence of loss or maintenance of the R-plasmid. Recent *in vitro* studies evaluated resistant microorganisms collected from poultry diagnostic cases and demonstrated the ability of common animal antimicrobial compounds to inhibit the transfer or totally eliminate R-plasmids. New data will be presented.

12:00 PM

ADJOURN

**Session B, Wednesday, July 18, 2007
8:00–8:15 AM**

Moderator: Erica Spackman

Characteristics of a Novel Infectious Bronchitis Virus Isolates from Delmarva Broiler Chickens

Jack Gelb, Jr., Brian S. Ladman, Conrad Pope, and Michelle Wood

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Avian Biosciences Center
531 South College Ave.; O44 Townsend Hall
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Newark DE 19716-2150

A novel S1 genotype of avian infectious bronchitis virus was isolated in 2006 from commercial broiler chickens raised on the Delmarva peninsula. The virus was associated with respiratory disease in field cases.

The S1 sequence of the isolates will be determined and compared to sequences in the public databases. The pathogenicity of the isolates for broiler chickens will be determined.

8:15–8:30 AM

Infectious bronchitis viruses isolated in California 2004-2006

Peter R Woolcock & Carol J Cardona.

CAHFS, UC-Davis, Fresno, CA 93725

Since 2004 infectious bronchitis virus has been isolated from chicken broiler and layer flocks in California. These isolates have been further characterized with monoclonal antibodies and by sequence analysis of the hypervariable region of the S1 gene. Data presented will be analysed not only by molecular and monoclonal antibody typing but also by the age of the flock and the reason for submission.


8:30–8:45 AM

Pathogenesis of Infectious bronchitis virus in the fully functional oviduct of unvaccinated laying hens

Chousalkar, K.K. and Roberts, J.R.

Animal Science
University of New England,
Armidale, 2351
NSW, Australia

The ultrastructural interaction of Infectious Bronchitis Virus (IBV) in different parts of the oviduct was studied in unvaccinated hens exposed to T and N1/88 strains of IBV. The infundibulum and magnum were more negatively affected as compared to the shell-forming regions. Eggs with watery whites during infection could be attributed to reduced synthesis of albumen proteins resulting from multiplication of virus in the reticular endoplasmic reticulum and Golgi complex of surface epithelial and tubular gland cells of the magnum. Egg shell quality was affected only by changes in shell colour and shape index. Cessation of egg formation could be due to disturbance in the physiology of cell organelles. The virus in the oviduct was further detected by single tube RT-PCR at different days post infection.



**Session B, Wednesday, July 18, 2007
8:45–9:00 AM**

The Dynamics of Spray Vaccination for IBV in Commercial Broilers

Mark W. Jackwood, Deborah A. Hilt, and Enid T. McKinley

Department of Population Health
College of Veterinary Medicine
953 College Station Road
University of Georgia
Athens, GA 30602

The Arkansas type of infectious bronchitis virus (IBV) is the most frequently isolated type of IBV in the field. Herein, we examined spray vaccination in commercial broilers in the field to determine if that practice is contributing to the frequency of Arkansas IBV isolations. Vaccine titer, vaccine coverage, and antibody response were examined. In addition, we used real-time RT-PCR to determine the level of vaccine viruses recovered from the trachea. The data shows maximum vaccine coverage at 14 days post-vaccination and a minimal but positive antibody response at 28 days post vaccination. The quantitative data showing the dynamics of IBV vaccine replication will be discussed with regard to the frequency of Arkansas type virus isolation.

9:00–9:15 AM

**Rapid Selection in Chickens of a Subpopulation Within an Attenuated
Infectious Bronchitis Virus Vaccine**

Vicky L. van Santen, PhD; Haroldo Toro, PhD, DVM; Kellye S. Joiner, PhD, DVM.

Department of Pathobiology, College of Veterinary Medicine, Auburn University

The sequence of the S1 gene of a live-attenuated commercial Ark-serotype vaccine exhibited heterogeneity in 12 codons, with minor components comprising approximately 10- 30% of the total at each heterogeneous position. Each nucleotide difference observed resulted in an amino acid difference. Sequence analysis of cloned S1 cDNA confirmed the heterogeneity and identified at least 15 distinct predicted S1 amino acid sequences among 25 cDNA clones. Surprisingly, during a single passage in chickens, a single IBV S1 coding sequence was found in 18 of 20 trachea, Harderian gland, or tear samples from individual chickens 3 to 14 days after ocular inoculation in two independent experiments. This sequence differed from the vaccine consensus sequence, but was identical to that of one of the cDNA clones of the vaccine stock, suggesting efficient selection in chickens of a minor component of the vaccine. The other two of the 20 samples (Harderian glands collected 3 days after inoculation) contained the same S1 sequence selected in the other chickens, but also contained an additional S1 sequence matching a different one of the cDNA clones of the vaccine stock. The predicted S1 amino acid sequence of the IBV selected in chickens differed from the majority S1 amino acid sequence of the vaccine strain at 6 positions and was identical to that of the original (non-attenuated) ArkDPI parent strain at 5 of those positions. The implications of these findings relative to the epidemiology of Ark-type IBV will be discussed.

Session B, Wednesday, July 18, 2007

9:15–9:30 AM

Risk Factors Associated With the Prevalence of Airsacculitis in Broilers Chicken in Quebec, Canada

Rachid Ankouche, Diane Brodeur, Martine Boulianne Jean-Pierre Vaillancourt

College of Veterinary Medicine, University of Montreal, CP 5000,
St-Hyacinthe, Quebec, Canada J2S 7C6

In 2004, an unusually high prevalence of broiler chicken carcasses condemned for airsacculitis was observed in Quebec slaughter plants. A paired case-control study was conducted between May 2005 and February 2006 in order to identify environmental and management risk factors associated with airsacculitis among broiler chickens in this province. Information was collected at the slaughter plants as well as on the 58 farms (29 pairs) selected for this study. The mean prevalence of airsacculitis condemnations (including trimming) was 8.01% for case flocks and 0.68% for controls. The final logistic regression analysis indicates that changing boots or plastic overboots between poultry houses and having an active darkling beetles control program were the main factors associated with a reduction in airsacculitis at slaughter.

9:30–10:00 AM

BREAK

10:00–10:15 AM

Moderator: Nathaniel Tablante

RICHARD RIMLER PAPER

**Interspecies Transmission: Host Range Determinants of Swine H3N2 Influenza A Virus
Transmission from Pigs to Turkeys**

H.M. Yassine¹, C-W Lee, Y.M. Saif

¹Ph.D. Candidate, Food Animal Health Research Program/OARDC/OSU
Wooster, OH 44691

In 1998, H3N2 influenza A subtype viruses emerged and rapidly spread in the swine population in the United States (US). Molecular analysis revealed that these viruses are triple reassortants, having genes from human, swine and avian viruses. Similar viruses were then isolated from turkey flocks around the US. Isolated viruses were also shown to be triple reassortants similar to those isolated from the swine populations. These findings prompted us to initiate a study on the interspecies transmission of H3N2 influenza viruses between swine and turkey in an attempt to understand the genetic bases of this phenomenon.

Three viruses of turkey origin (TK/IL/04, TK/OH/04, TK/NC/03) and one virus of swine origin (SW/NC/03; vaccine strain) were tested for their transmissibility between swine and turkeys. All viruses were shown to replicate in both species (using real-time PCR on swab samples and HI-test on blood serum collected at day 15 post infection), however only the Ohio strain was shown to transmit both ways between the two animal species. The Hemagglutinin (HA) and Neuraminidase (NA) genes of the Ohio strain isolated from pigs and turkeys in the transmission experiments will be sequenced and compared using DNA-STAR program to detect any changes that occurs upon the replication of the virus in different host systems.



Session B, Wednesday, July 18, 2007
10:15–10:30 AM

**RCA-Free Recombinant Adenovirus-Vectored Vaccine for Mass Immunization of Poultry
Against Avian Influenza**

Haroldo Toro,¹ De-chu Tang,² Frits van Ginkel,¹ Z. Shi,²
¹Department of Pathobiology, Auburn University; ²Vaxin Inc.

Protective immunity against avian influenza (AI) virus can be elicited in chickens vaccinated *in ovo* with replication competent adenovirus (RCA)-free recombinant adenovirus encoding the H5 gene of AI (AdH5). We evaluated antibody kinetics in chickens vaccinated *in ovo* with increasing doses of AdH5. Vaccination with 300 μ l of AdH5 containing 10⁹ ifu/ml induced significantly higher titers in more than 85% of the vaccinees. For the purpose of using this vaccine in existing chicken populations we evaluated the ocular route for vaccine delivery. Chickens primed and boosted ocularly with AdH5 developed both systemic and local antibodies. Finally, we produced a new recombinant construct encoding an H7 transgene (AdH7). Chickens vaccinated *in ovo* or intramuscularly with the AdH7 vaccine developed high specific antibody against the H7 glycoprotein

10:30–10:45 AM

Differential Growth of Avian influenza Virus in Chicken and Duck Cells

Luciana Sarmiento , Kristin Zaffuto, Mary Pantin-Jackwood, Claudio Afonso
Southeast Poultry Research Laboratory-USDA/ARS
934 College Station Road
Athens-GA 30605

Ducks and chickens infected with AI viruses display clear differences in disease manifestation. To understand the mechanisms responsible for these differences we have determined the ability of several AI isolates to replicate in primary tracheal epithelial cells and fibroblasts from both species. Results suggest differences in the capacity of the host species to support viral replication, and a varied ability of different avian influenza viruses to infect identical cell types. Transcriptional host responses were analyzed using a microarray of the complete chicken genome.



**Session B, Wednesday, July 18, 2007
10:45–11:00 AM**

Efficacy of three inactivated vaccines against challenge with HPAI H5N1 Vietnam/05 viruses in ducks

**Mary J. Pantin-Jackwood, David L. Suarez, Jennifer Pfeiffer,
and Luciana Sarmento**

Southeast Poultry Research Laboratory, USDA-ARS
934 College Station Rd. Athens, GA 30605

The objective of the study was to compare the efficacy of inactivated vaccines containing a European isolate (A/turkey/England/73, H5N2 Chinese commercial vaccine), an American isolate (A/chicken/Hidalgo/94, H5N2 Mexican commercial vaccine) or a recombinant virus (RE-1, H5N1 recombinant Chinese vaccine) in protecting ducks against a HPAI H5N1 challenge. One week-old Pekin ducks were vaccinated with one of the three mentioned vaccines and then challenged two weeks later with a lethal dose of either A/duck/Vietnam/218/05 or A/duck/Vietnam/203/05. All unvaccinated ducks died within 5 days, whereas 95% of ducks vaccinated with A/turkey/England/73 and 100% of ducks vaccinated with either A/chicken/Hidalgo/94 or RE-1 survived. However, virus shedding was detected from all vaccinated ducks. All three vaccines provided good protection from clinical disease, but differences in antibody titers and duration of viral shedding were observed among the different groups.

11:00–11:15 AM

Comparison of *in vivo* innate immune responses in lung and spleen tissue following infection with Asian H5N1 avian influenza viruses in ducks and chickens.

Darrell R. Kapczynski and Mary Pantin-Jackwood

Exotic & Emerging Avian Viral Disease Research Unit, Southeast Poultry Research Laboratory,
ARS, USDA, Athens, Georgia, USA.

The immune system can be divided into two functional components, the innate and adaptive, that differ in their mechanism of pathogen recognition and response. The innate immune response is responsible for detecting invading microorganisms during the initial stages of infection, which is a crucial determinant of disease resistance or susceptibility. Because chickens normally succumb to disease within 3-4 days after infection with highly pathogenic Asian H5N1 avian influenza (AI), the adaptive immune response likely contributes little to defense against disease. On the other hand, waterfowl, including ducks, are considered natural reservoirs for AI and rarely display clinical signs of disease. The reasons for the differences in susceptibility and pathogenicity of different avian species to different pathotypes of AI are unclear and ill defined. These studies were designed to examine the response of the innate immune response by measuring cytokine expression with RRT-PCR immediately following infection. The results indicate differential cytokine expression *in vivo* between chickens and ducks following exposure to AI. Ducks resistant to disease generally displayed increased cytokine expression, while chickens susceptible to disease tended to exhibit suppressed cytokine expression.



Session B, Wednesday, July 18, 2007

11:15–11:30 AM

Surveillance, testing and epidemiology of avian influenza in the New York live bird market system

Susan C. Trock¹, Michelle Gaeta², Lisa Weisse², Tara Howard², Kimberley Tropea³, Adam Holloway³, Sung Kim¹, J. Beeby¹, Jan Pederson⁴, Dennis Senne⁴, and Edward Dubovi¹

1 = Cornell University, 2 = New York State Agriculture and Markets, 3 = USDA, APHIS, VS, and
4 = USDA, NVSL

An estimated 18-23 million birds annually move through the East Coast live bird market system. Recently the incidence of low pathogenic H5 or H7 subtype viruses has decreased in the New York markets, with no positive isolations for four months in 2006. At this same time sampling has increased. Use of the RRT-PCR test has shortened the time to detecting positive markets, allowing for a more rapid trace back response. In addition to summary market findings, discussion of the voluntary market control measures, selected trace back findings, and use of preliminary RRT-PCR findings for regulatory action will be presented.

11:30–11:45 AM

Using Resident, Wild Mallards as Sentinels for Detecting Low Pathogenic H3, H4, H5, H6, and H7 Type A Influenza Viruses Circulating in Resident, Wild Ducks

Richard Slemons, Jacqueline Nolting, Lloyd Alexander, and Dennis Senne

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The Ohio State University

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During the summer type A influenza virus surveillance efforts in local wild, free ranging waterfowl populations is generally dependent upon the availability of wildlife biology studies engaged in trapping live birds and early hunting seasons in September. To circumvent these limitations and to permit sequential sampling in a defined population, sentinel captive ducks were first used for AIV surveillance in Minnesota in the early 1980s (Halvorson, et al.). This study reports on the use of resident, wild mallards as sentinels for detection of type A influenza viruses circulating in local wild bird populations before the fall return of the northern migrants. The source flock for the resident, wild, mallards monitored in this study tested negative for the presence of type A influenza viruses five times from May to early September. This strategy was effective in detecting low pathogenic waterfowl-origin H5 AIVs in early August, H7 and H4 AIVs in early September, and H3, H4 and H6 AIVs in early October. The August isolates suggest AIVs were circulating in local, wild birds as sampling was conducted before the arrival of the earliest fall migrants. The source of the September isolates could include local wild birds as well as early arriving shore birds and blue-winged teal. These findings indicate that the natural history of AIVs is very dynamic and alerts the commercial poultry industry to the fact that the risk of interspecies transmission of AIVs is not limited to the arrival of large populations of migratory birds in the fall.



**Session B, Wednesday, July 18, 2007
11:45–12:00 PM**

Results of Avian Influenza Virus Monitoring in Wild Birds in Maryland: 2005-2006

Cindy P. Driscoll, D.V.M., Larry J. Hindman Maryland Department of Natural Resources
Richard D. Slemons, D.V.M., Ph.D., Department of Veterinary Preventive Medicine, Ohio State
University, Columbus, Ohio
Dennis A. Senne USDA National Veterinary Services Laboratory, Ames, Iowa

Avian influenza (AI) viruses were first recovered from wild waterfowl sampled on the Delmarva Peninsula in 1973. It is assumed AI virus infections have been common in the ensuing years. Delmarva has one of the highest concentrations of commercial broiler poultry farms in the United States and the Chesapeake Bay region has the highest concentration of wintering migratory waterfowl on the East Coast. Therefore, this area is important for widely different reasons, but it is critical to both wild birds and commercial poultry. Another concern is the interface between the commercial poultry and live bird marketing systems. This interface was associated with the LP H7N2 AI outbreaks in Delaware and Maryland in 2004. To the credit of poultry health experts and government agencies, the viruses causing these LP AI outbreaks were quickly and efficiently eradicated. Unfortunately, AIVs are not predictable and new developments in Eurasia and Africa and HPAI outbreaks during the last few years in Canada and Chile and possibly Texas have raised concern the poultry industry in the Western Hemisphere is at increased risk due to HPAIVs. Therefore interest in proactive AIV surveillance in domestic and wild birds has increased.

In the summer of 2005, the Maryland Department of Natural Resources in collaboration with The Ohio State University and the USDA APHIS NVSL initiated a proactive, expanded sampling effort in wild, free-ranging waterfowl and opportunistic sampling of other wild bird species throughout the region. Over 1250 samples were collected between July 7, 2005 and May 1, 2007. To date, a total of 15 non-H5 and non-H7 low pathogenic AI isolates were recovered from species of great interest including Snow Geese, Long-tailed ducks, Common Scoters, White-wing Scoters. These isolates appear to represent the first recoveries of AI viruses from Common Scoters and Snow Geese in the world and the first report of recoveries from White-winged Scoters and Long-tailed Ducks in the Western Hemisphere. These species have been identified as species of special interest by the USDOJ since they breed above the Arctic Circle and have increased potential for contact with wild birds from the Eastern Hemisphere.

12:00 PM

ADJOURN



INSTRUCTIONS FOR POULTRY POSTER PRESENTATION

The following is specific information about the 2007AVMA Poultry Poster Presentation:

Location of Poultry Poster Presentation:
Convention Center

Please refer to the Poultry Poster Program for your session number.

NOTE: Posters must be set up after 6:00 AM on Sunday, July 15th and no later than 7:00 AM on Monday, July 16th.

All posters must be removed by 12:00 PM on Wednesday, July 18th.

Dimensions: Size of the Mounting Board for Poster: 4 feet X 8 feet (48 inches x 96 inches). All posters must fit within the outer edges of the board. All boards will be double sided, so another presenter may be mounting a poster on the other side of the board at the same time.

General Information:

7. Your poster presentations must be available for viewing during the hours scheduled by the Poultry Poster Section.
8. Handouts are permitted. *Sale of any material is strictly prohibited.*
9. Poster sessions are intended to serve as informal discussions and not as lectures or paper reading sessions.
10. Pushpins will be available in the poster areas. Please do not write or paint on the poster boards.
11. Projection equipment and electrical outlets will not be provided in the poster session area.
12. One (1) complimentary convention registration will be provided for the primary author of each poster.

AAAP POSTERS

AVIAN INFLUENZA

1.

Histopathological studies of the respiratory system of essential oil-treated broilers against *Mycoplasma gallisepticum* and/or H9N2 challenges

Elie K. Barbour, Rindala G. El-Hakim, Danyelle D. Gerges, Pia A. Nehme, Hussam A. Shaib and Marc S. Kaadi

Dept. of Animal Sciences
Faculty of Agricultural and Food Sciences
American University of Beirut
Beirut, Lebanon

The objective of this work is to evaluate the impact of eucalyptus and peppermint essential oils (Mentofin®) in the protection of the respiratory system of broilers against controlled challenges by *Mycoplasma gallisepticum* (MG) and/or avian influenza virus H9N2. Seventy 1-day-old broilers were reared in 7 groups (10 birds/group) up to 1 week of age. Group 1 was the control (nontreated with Mentofin and unchallenged); challenged groups were Group 2 (non-treated with Mentofin and MG challenged), Group 3 (Mentofin treated and MG challenged), Group 4 (non-treated with Mentofin and H9N2 challenged), Group 5 (Mentofin treated and H9N2 challenged), Group 6 (non-treated with Mentofin and MG/H9N2 challenged), and Group 7 (Mentofin treated and MG/H9N2 challenged). At 1 week of age, an intratracheal challenge of the birds with MG (2 hemagglutination units/0.5 mL/bird) and/or H9N2 (2 hemagglutination units/0.5 mL/bird) was given to specific groups mentioned previously. Essential oils of Mentofin were administered for 6 days, effective 1 day post-challenge. Histopathological observations were concluded at 6 days post-challenge and revealed a significant reduction ($P < 0.05$) in microscopic tissue lesions of birds treated with Mentofin in comparison to birds deprived from this treatment but challenged similarly. The significant ($P < 0.05$) reduction in microscopic lesions included a decrease in tracheal deciliation in MG- and MG/H9N2-challenged birds, a decrease in mucosal hypertrophy in MG-, H9N2-, and MG/H9N2-challenged birds, a decrease in goblet cell degeneration in MG and MG/H9N2-challenged birds, a decrease in mucus accumulation in MG-challenged birds, and a decrease in heterophil infiltration in MG/H9N2-challenged birds.

2.

Evaluation in Chickens of a Killed NS1 Mutant Avian Influenza Virus Vaccine

V. Brahmakshatriya² B. Lupiani^{1,2} and S.M. Reddy^{1,2}

¹Department of Veterinary Pathobiology; ²Department of Poultry Sciences, Texas A&M University, College Station, TX 77843

Low pathogenic avian influenza (LPAI) H5 and H7 virus subtypes have the potential to mutate to highly pathogenic AI (HPAI) strains. We investigated the role of an NS1 mutant virus as a potential DIVA vaccine. NS1 protein of influenza virus plays a major role in blocking the host's antiviral response. Using reverse genetics, we recovered parental (rH5N3) and NS1 mutant (rH5N3/NS1-143) influenza viruses. The growth properties of rH5N3 and rH5N3/NS1-143 were compared in cell culture and in different age, embryonated chicken eggs. Vaccine protection in chickens will be based on antibody titers, lesions observed and virus re-isolation.

3.

Comparative pathology of H5N1 highly pathogenic avian influenza virus infection in avian species in the Orders Anseriformes and Charadriiformes

Justin D. Brown¹, David E. Stallknecht¹, and David E. Swayne²

¹Southeastern Cooperative Wildlife Disease Study, Department of Population Health, Wildlife Health Building, College of Veterinary Medicine, The University of Georgia, Athens, GA, USA, 30602

²United States Department of Agriculture, Agricultural Research Service, Southeast Poultry Research Laboratory, Athens, GA, USA, 30605

Thirteen species of ducks, geese, swans and gulls present in the North American wild bird populations were inoculated intranasally with A/Whooper Swan/Mongolia/244/05 (H5N1) avian influenza virus (AIV) to evaluate the range of viral shedding and pathology within these two avian orders. Based on mortality and the distribution of virus, these species were separated into four categories: 1) 100% mortality with viral antigen disseminated in the vasculature and parenchyma of numerous visceral organs, 2) high mortality with antigen primarily distributed in the brain, pancreas, adrenal glands and heart, 3) moderate mortality with antigen primarily located in the brain, and 4) No mortality and no viral antigen was detected.

4.

Epidemiological studies on Avian Influenza Infection in the Migrating Wild Bird

Byun, Seong-Hwan, Kim, Min-Jeong, Kim, Jeong-Nyeo, Mo, In-Pil

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In Korea, outbreaks of highly pathogenic avian influenza (HPAI) has been occurred in 2003 and this year and caused severe economic losses in poultry industry. Although we tried to find out the source of infection, we failed. Migrating water fowl is one of the sources for introduction of avian influenza virus in the domestic fowls. We attempted to isolate AI virus from both wild birds and domestic birds near the wild bird habitats. In this study, 200 samples were assessed from domestic chickens and 1138 samples were assessed from migrating wild birds. From these samples fifty nine viruses were identified and 38 viruses were confirmed as avian influenza (AI) virus among them. Most AI viruses isolated from domestic chickens were H9 subtype and other isolates from wild birds were either H2, H5 or H7 subtypes. To find out the source of the infection and relation between isolates using molecular epidemiology further study such as sequencing the major gene of the AI isolates has been conducted.

5.

Development of microsphere-based assays for the detection of H5 and H7 subtype avian influenza virus

Wonhee Cha, Megan Strother, Y. Mo Saif, Chang-Won Lee

The Ohio State University, Food Animal Health Research Program, Wooster, OH 44691

Real-Time RT-PCR (RRT-PCR) provides a rapid and feasible alternative to virus isolation in embryonating chicken eggs and subtyping by hemagglutination inhibition test and has emerged as an invaluable screening tool for influenza virus. However, the main disadvantages of using RRT-PCR are its high assay cost and the limitation in multiplexing which further increases the cost as well as the assay time. The microsphere-based array system is a newly emerging technology that provides the multiplexing of up to 100 different assays within a single sample. In this study, we utilized this system coupled with branched DNA (bDNA) signal amplification technology (a sandwich nucleic acid hybridization assay) to detect and subtype H5 and H7 influenza virus. In our 3-plex assay, we were able to detect different HA subtype of influenza virus and differentiate H5 and H7 HA subtype at the same time based on capture probes specific for the M, H5, and H7 gene. In addition to multiplex capacity, this system does not require an RNA extraction step and samples can simply be treated with lysis buffer for the assay.

6.

Avian Influenza Neuraminidase 1 (N1) ELISA Using Baculovirus Expressed Antigen and its Application on DIVA Vaccination Strategy

**Maricarmen García¹, Yuru Liu¹, Xiuqin Xia², David E. Swayne², David L. Suarez²,
Mark W. Jackwood¹ and Egbert Mundt¹**

¹Poultry Diagnostics and Research Center, Department of Population Health, College of Veterinary Medicine, University of Georgia. ²Southeast Poultry Research Laboratory, Agricultural Research Service, United States Department of Agriculture.

An ELISA was developed using baculovirus express N1 protein from the A/Chicken/Indonesia/11/03 (H5N1) virus. The objective of this study was to evaluate the specificity and sensitivity of the N1-ELISA. The specificity of the ELISA was tested with a chicken anti-sera panel raised against N1 to N9 virus subtypes. To evaluate the reactivity range of the N1-ELISA sera samples raised against N1 virus from the North America lineage were tested. The N1-ELISA was specific for the detection of N1 antibodies. The N1-ELISA was capable to detect N1 antibodies in chicken serum samples raised against North America H1N1 viruses (CK/NY/94, TK/SD/80, TK/NC/88). Antibodies against other neuraminidase subtypes (N2 to N9) did not cross-reacted with the N1 antigen. To evaluate the sensitivity of the N1-ELISA to detect N1 antibodies in vaccinated/infected chickens, serum samples collected from chickens vaccinated with H5N9, H5N2, and challenge with Asian H5N1 viruses, and samples from chickens vaccinated with H7 Pox vaccine and challenge with H7N1 will be tested by N1-ELISA. The N1-ELISA responses will be compared to hemagglutination inhibition (HI) titers and agar-gel precipitation (AGP).

7.

Determination of Pathogenicity of Jordanian Local Isolate of Avian influenza (H9N2) in Broiler-Chickens

Saad Gharaibeh, DVM, PhD, Dip ACPV

Dept of Pathology and Animal Health, Faculty of Veterinary Medicine, Jordan University of Science and Technology, Irbid 22110, Jordan.

Jordanian local isolates of H9N2 avian influenza has been classified as a low pathogenic virus that almost causes no clinical signs or pathology in chickens. Field observations suggested higher virulence of this virus causing serious respiratory disease especially in broilers. This study was designed to determine the pathogenicity of this virus when natural route of infection is used. Twenty broiler-chickens were divided into two groups at one day of age and reared in a control environment. At 25 days of age, one group was intranasally challenged with a local isolate of avian influenza (H9N2). Group two remained unchallenged and served as a negative control. At the end of the experiment at 41 days of age, serology, body weight, gross, and histopathologic lesions were evaluated between groups. The infected group had a titer of $2^{8.2}$ on HI test while the control group remained negative. The infected group had a decreased average body weight of 230 g compared to the control group ($p = 0.004$). Lymphocytic inflammation was present in the sinuses, trachea, lung, and airsacs of infected group and the most severe lesions were in the tracheas and airsacs. This study confirms the pathologic nature of the H9N2 local viruses and its effect on the local chicken industry in Jordan.

8.

Efficacy of Inactivated Vaccine against Low Pathogenic Avian Influenza (H9N2) in SPF and Commercial Chickens

Bong-Do Ha, Chang-Hee Lee, Jong-Nyeo Kim, In-Pil Mo

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Low pathogenic avian influenza (LPAI) is widely distributed and cause significant economic loss, especially in the egg production. However, drop of egg production is not easy to reproduce in the laboratory condition because of limited the number of birds used in the experiments. We inoculate the inactivated vaccine (H9N2) in the both SPF and commercial chickens and evaluated the efficacy after challenge with field LPAI virus. In the SPF study, the vaccine developed antibody titer enough for protection against challenge of field AI virus and reduced significant amount of the virus in the swab and tissue samples when comparing with those of non-vaccinated control group. In the commercial layer study, egg production rate of control group dramatically declined 5 days after challenge and it took 4 weeks, almost at the end of the test to return to normal pace. Such egg drop is similar to the clinical signs observed in the field infection of AI. On the other hand, there were minute changes in the egg production rate of the treatment group. Regarding all of the above test results, the safety and efficacy of the vaccine (ADL0401) is appropriate enough to provide protection against AI field infection (H9N2) that is epidemic in Korea.

9.

Sequence analysis and phylogenetic study of the entire genome of three avian influenza H9N2 subtypes from south China

**Mazhar I Khan^b, Zhixun Xie^a, Jianbao Dong^a, Xiaofei Tang^a, Jiabo Liu^a, Yaoshan Pang^a,
Xianwen Deng^a, and Zhiqin Xie^a**

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^b Department of Pathobiology Science, University of Connecticut,

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CT 06269-3089

The complete nucleotide sequences of three subtypes of H9N2 of avian influenza viruses from Guangxi, China were performed. The sequence analysis of each of the eight segments of genes showed higher homologies among the Guangxi isolates and two isolates of H9N2 subtypes from Guangdong and Jiangsu provinces of China. This suggested that Guangxi H9N2 isolates have come from the same source. However on phylogenetic analysis the eight segment genes of these three isolates were not in the same sublineages in the phylogenetic trees, which showed that they were products of natural re-assortment between H9N2 avian influenza viruses from different sublineages. The nine nucleotides ACAGAGATA which according to the amino acids threonine (T), glutamic acid (E), and isoleucine (I) were lost in these H9N2 isolates between nucleotides 187 and 195 in the open reading frame of neuraminidase (*na*) genes.

10.

Zoning as a measure to minimize avian influenza spread during an epidemic

Heather Labelle, Jean-Pierre Vaillancourt, Michel Bigras-Poulin, Alex Thompson

College of Veterinary Medicine, University of Montreal, CP 5000, St-Hyacinthe,
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Zoning is the regionalization of poultry related activities in order to minimize spread of highly contagious diseases such as avian influenza. The project has for main objectives to identify all parameters needed in establishing zones, as well as to determine the geographical density distribution of poultry farms and related businesses in Ontario and Quebec. After consultation with local poultry industries, zones will be established. Using disease dispersion parameters obtained from recent avian influenza epidemics, we will assess the potential value of zoning for these provinces.

11.

Replication of influenza viruses in chicken-origin (DF-1) and quail-origin (QT-6) cell lines

Chang-Won Lee, Keumsuk Hong, Megan Strother

The Ohio State University, Food Animal Health Research Program, Wooster, OH 44691

The primary goal of the study was to evaluate the characteristics of two avian cell lines, DF-1 (chicken-origin) and QT-6 (quail-origin) and their abilities to support the growth of influenza viruses from different species in order to identify a cell substrate with a broad viral susceptibility range for use as an alternative to primary chicken embryo fibroblast (CEF) cells. We evaluated the replication efficiency of 14 strains of avian influenza viruses of 9 hemagglutinin subtypes in QT-6 and DF-1 cell lines. The results were compared side by side with those obtained using primary CEF cells and Madin-Darby canine kidney (MDCK) cells. The infectivity titers of most viruses in QT-6 and DF-1 cells were comparable to those in MDCK or CEF cells. However, human and swine-lineage viruses preferentially replicated well in MDCK cells. We will continue to test the replication of more diverse influenza viruses in these cell lines. Furthermore, receptor distribution of each cell line and the receptor specificity of the individual viruses will be analyzed and compared with the virus replication efficiency.

12.

Development of a triplex fluorescence microsphere based immunoassay (FMIA) for the detection of antibodies to avian influenza virus proteins.

Douglas S Watson, Sanjay Reddy and Blanca Lupiani*

Department of Veterinary pathobiology, Texas A&M University, College Station, TX.

Using baculovirus expressed proteins we have developed a fluorescence microsphere based immunoassay (FMIA) for the detection of antibodies to individual avian influenza virus proteins. Our data indicates that individual chickens react differently to all three viral proteins and that M1 and NS1 induce an immune response that peaks between weeks 1 and 3 post-infection and declines significantly by week 4. On the other hand, the levels of NP antibodies seem to remain stable even at 4 weeks post-inoculation. Therefore, NP should be the antigen of choice for developing single antibody detection immunoassays.

13.

Ups and Downs of Implementation of an Eradication Strategy During an Outbreak of HPAI (H5N1) in Iran

Seyed Mehdi Mirsalimi and Afshin Hedayati

Intervet Iran

During the fall of 2005 an outbreak of HPAI H5N1 was reported in Turkey which was very close to Iranian boarder. Later, due to the outbreak in other neighboring countries, and confirmed report of dead swans due to H5N1 inside Iran and some suspected cases of the poultry farms, the Iranian veterinary organization (IVO) decided to cull all kind of backyard poultry in all villages within the 10 kilometer of the boarder and birds around the suspected areas inside the Iranian territory.

High movement from various areas including suspected areas caused some problem for the eradication plans.

Although the IVO was quite successful in prevention and eradication of HPAI by full compensation to the villagers but due to the lack of plan for replacement of the cull birds, this strategy remained questionable. Regarding the culled poultry farms, the eradication strategy was more successful because of the implementation of an insurance policy for full compensation. More details of implementation and problems during the process and the outcome will be discussed.



14.

Efficacy and Safety of the Low Pathogenic Avian Influenza Vaccine (H9N3) in SPF chickens

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Low pathogenic avian influenza (LPAI) has been recognized as one of the most important disease in the poultry industry. To reduce the economic impact and control the disease, vaccination with inactivated vaccine has been considered in this country. However, the vaccination cause problem in the surveillance of AI outbreak because the antibody induced by inactivated vaccine could not be differentiated from the antibody induced by field infection. Therefore, we tried to develop inactivated vaccine with reassorted H9N3 AI virus which has different type of neuraminidase (NA) compare to those of field AI virus. After limiting dilution, we choose RV7CE4 as a vaccine candidate. In the vaccine study, once or twice vaccination was performed and challenged with H9N2 virus (01310) which is the most pathogenic among the viruses recently isolated in Korea. When we inoculated this vaccine to SPF chickens, the vaccine has no adverse effect on birds and formed good immune capability which reduce viral shedding in the birds challenged with 01310. Based on the above result, we developed reassorted H9N3 vaccine which will efficiently prevent the low pathogenic AIV (H9N2) infection in the poultry farms.

15.

Validation of a New Avian Influenza Antibody Blocking ELISA

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Well characterized serum samples from different avian species (commercial and non-commercial chickens, turkeys, ducks, geese, quail and ostriches) were analyzed with a new blocking enzyme-linked immunosorbent assay (ELISA). The results showed detection of at least 14 different avian influenza virus subtypes, excellent correlation with other traditional test methods, such as agar gel immunodiffusion (AGID) and hemagglutination inhibition (HI) tests, and improved specificity and sensitivity when compared to indirect ELISAs.



16.

**Genetic and Biological Characterization of the H5N2 Virus
Isolated from a Parrot**

Smitha Somanathan-Pillai¹, David L. Suarez², Chang-Won Lee¹

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In 2004, H5N2 influenza A virus was identified in a psittacine bird for the first time in the U.S. Complete genomic characterization of the virus was performed and demonstrated high similarity in all 8 genes to the Mexican-lineage H5N2 viruses. Antigenic analysis done by the hemagglutinin inhibition (HI) test further confirmed the close relatedness of this virus to Mexican-lineage viruses with the highest cross reactivity to A/CK/Guatemala/194573/02 antibody. The virus replicated well in chickens and high titer of virus was detected from tracheal swab samples ($10^{5.0}$ - $10^{6.0}$ EID₅₀/0.2ml) collected at 3 days post intranasal infection. Biological characterization will be further conducted in turkeys and ducks to assess the potential threat of this virus to the U.S. poultry.

17.

Evaluation of Poultry Diagnostic Tests for Avian Influenza Virus in Duck Origin Samples

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Diagnostic tests for the detection and identification of avian influenza virus (AIV) infection in chickens and turkeys have been well established and commercial test kits are available for some applications. However few, if any diagnostic tests have been validated or evaluated for use with specimens from domestic ducks, which represent a smaller, but critical sector of the commercial poultry industry for AIV monitoring. In this study, we report the evaluation of routine virus detection methods including real-time RT-PCR and commercial antigen immunoassay kits for the detection of virus in experimentally exposed ducks. Virus isolation was used as a virus detection reference standard.



18.

The Importance of Educational Programs in the Prevention and Control of Avian Influenza

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The impact of Avian Influenza outbreaks on both poultry and human health can be devastating. Control measures that are implemented in response to outbreaks may be adequate but are nonetheless reactive “salvage” operations that are often costly and time-consuming. A more proactive approach to the prevention of Avian Influenza involves basic research, biosecurity, monitoring and surveillance, vaccination, and education. Academia, industry, and government experts can easily get caught up in a flurry of scientific and regulatory activities and may not put enough emphasis on educational programs which are necessary because understanding the nature of the disease and how it spreads are vital to its successful prevention and control.

19.

New Way to Develop Live Influenza Vaccine Candidate Strains

**Leyi Wang¹, Smitha Somanathan-Pillai¹, Megan Strother¹, Keumsuk Hong¹,
Y. Mo Saif¹, Mary Pantin-Jackwood², David Suarez², Chang-Won Lee¹**

¹The Ohio State University, Food Animal Health Research Program, Wooster, OH 44691; ²Southeast Poultry Research Laboratory, USDA ARS, Athens, GA 30605

The NS1 protein of influenza virus functions as an interferon antagonist and is thus directly associated with the pathogenicity of the virus. From the TK/OR/71-del (H7N3) virus, we previously found that several variants with different sizes of the NS gene can be generated by serial passage of the virus in embryonating chicken eggs. In this study, we have further pursued the identification of different NS genes and have found 20 different NS genes that have unique deletions in different regions of the NS gene. To date, we were able to biologically purify 3 different variants that have different truncated NS genes by plaque purification in chicken embryo fibroblast (CEF) cells. We expect that these naturally selected NS1 deletion variants will be useful in the development of live influenza vaccines both in their current state and with further modification of the NS1 protein. Furthermore, a deletion in the NS1 protein can also be useful as a negative marker for the DIVA (Differentiating Infected from Vaccinated Animals) approach.

BACTERIA, MISCELLANEOUS

20.

Case Report: Dactylariosis in Bobwhite Quail

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Eleven-day-old bobwhite quail were presented to the diagnostic laboratory with a complaint of high mortality and ataxia. Clinical signs were consistent with a neurologic disease. Confluent foci of malacia were seen in the cerebrum and cerebellum. Histologic examination and cultures demonstrated intralesional organisms and fungal colonies morphologically consistent with *Dactylaria sp.* The birds were housed on pine bark litter in a deer carcass composter that had been converted to quail production.

21.

Non-Serotypable *Avibacterium paragallinarum* Infection in Commercial Layers

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We report on an outbreak of coryza in layers (starting at 17 weeks of age) by *Avibacterium paragallinarum* that could not be assigned to one of the three recognized Page serovars. The flock showed the typical clinical signs of coryza and egg production was always at least 3% below the standard production till 26 wks of age.

The affected flock received two doses of a commercial bacterin (that included the three serovars A, B, and C) prior to the outbreak. This suggests that there is little immunological relationship between this *Av. paragallinarum* non-typable strain and the three recognized Page serovars.

22.

The Incidence of *Bordetella avium* in Mississippi Broilers

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Various pathogens such as viruses, bacteria and fungi may cause respiratory disease in chickens. Many of these pathogens may be primary invaders or secondary invaders. *Bordetella avium* is historically thought of as a primary pathogen of turkeys, and a secondary pathogen in chickens with mild to no affect. In recent history, however, the number of isolations from broilers experiencing respiratory problems in the Mississippi poultry industry has brought this bacterium under focus. Field experience has shown that if *B. avium* is isolated on a farm with unexplainable respiratory problems and then removed or reduced in the environment, the respiratory problems can be alleviated. This poster will discuss the incidence of *B. avium* in Mississippi broilers, the clinical signs and lesions of affected birds, and the treatments utilized to control this pathogen.

23.

Serotypes of *Riemerella anatipestifer* Isolated from Commercial Ducks and Vaccination of Ducks with Killed Oil Vaccine in Korea

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Hyuk-Man Kwon, Jae-Hong Kim¹ and Jun-Hun Kwon¹**

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Riemerella anatipestifer (RA) causes a serious septicemic disease and economic loss in ducklings in Korea. Confirmation of RA serotypes prevailing in duck farms is vital to vaccine development. Among thirty-two isolates, twenty-two isolates were serotype 7, seven isolates were serotype I and the others were serotype 4 and 16. We prepared the killed oil vaccine with 0.3% formalin and ISA70 from serotype 7 and 1, and tested the protection efficacy of the vaccine against field isolates of pathogenic RA. We confirmed the protective capacity of the homologous strain vaccination against RA and the ineffectiveness of heterologous preparations.

24.

Pathogenic *Campylobacter* in Turkey Production

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Poultry have been implicated as one of the primary sources of foodborne *Campylobacter* infections in humans. In this study we have investigated the prevalence of key *Campylobacter* virulence factors in *C. coli* and *C. jejuni* recovered from production turkey flocks during brooding, and grow-out phases. Isolates were assessed for the possession of a range of key virulence factors associated with pathogenic *Campylobacter* at different phases of growth and through different flock rotations from three different brooder facilities using molecular techniques and polymerase chain reaction (PCR). Genes evaluated include those associated with invasion, toxin production, plasmid-borne virulence genes and secretion systems.

25.

Comb Candidiasis Affecting Roosters in a Broiler Breeder Flock

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A cutaneous mycosis caused by *Candida albicans* that involved the combs and less frequently the wattles, facial skin, ear lobes, and neck of male broiler breeders is described. Roosters were 35 wks old and housed with hens in two conventional broiler breeder houses on a farm in western North Carolina. Morbidity was approximately 10% in one house and less than 2% in the other house. Mortality and flock fertility were not affected. Three birds from the most affected house were examined. All birds had white adherent material on their combs that presented as crusty patches or lighter diffuse areas. Often lesions were roughly circular or had a defined margin. Small black scabs were present in a few lesions. Similar but less extensive lesions were located on the wattles, facial skin, ear lobes, and rictus. In one bird, lesions extended down the neck and were accompanied by hyperemia and feather loss. Hyperkeratosis with little to no inflammation and intralesional fungi occurring as yeasts and pseudohyphae was seen microscopically. High numbers of *C. albicans* were isolated and identified from the lesions.

26.

In Vitro Adhesion Assays of *Gallibacterium* to Chicken Tracheal or Oviductal Epithelial Cells.

Saúl Ramírez, Andrea Zepeda, Vicente Vega, Vladimir Morales, Luis Pérez, Soledad Díaz, Henrik Christensen, Anders Miki Bojesen, and Edgardo V. Soriano

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Facultad de Medicina Veterinaria y Zootecnia,
Universidad Autónoma del Estado de México.

Adhesion of six *Gallibacterium* reference strains to chicken tracheal or oviductal cells was investigated. In vitro assays were conducted with epithelial cells and bacteria adjusted at 1:100, respectively. *Avibacterium paragallinarum* and *Gallibacterium* genomospecies 2 (CCM 5976) showed the highest number of adhered bacteria to tracheal epithelial cells. Similarly, this *Gallibacterium* strain showed the highest number of adhered bacteria to oviductal epithelial cells. Obtained results showed the ability of some *Gallibacterium* strains to adhere to tracheal epithelial cells, indicating the capability to establish an infection at the upper respiratory tract of the chicken. Similarly, adhesion of *Gallibacterium* to oviductal epithelial cells could be involved in the pathogenesis of salpingitis and other reproductive lesions observed at the field. The nature and receptors of adhesins in this bacterium are unknown.

27.

Detection of *Avibacterium paragallinarum* by PCR: Conditions of Nasal Swab Samples Submitted to the Diagnostic Laboratory

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A HP-2 PCR test that is specific for *Avibacterium paragallinarum*, rapid and able to detect all known variants is available. Sinus swabs stored for up to 180 days at 4° C or -20° C are positive in the PCR. In the present study, conditions of nasal exudate swab samples for submission to the diagnostic laboratory were investigated. Samples were storage at 4° C or into an ice container up to 96 hours. Positive results were observed up to 48 h hours of storage in both conditions. These results indicate that dry swabs taken from nasal exudates and submitted up to 48 hours after, are suitable samples for testing in PCR. Evaluation of dry swabs from nasal exudates kept at room temperature for PCR testing is being performed.

28.

**Peritonitis in Egg-type Chickens Caused by *Gallibacterium anatis*
and *Escherichia coli***

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Peritonitis is a major disease problem of laying hens in commercial table egg operations. Two bacterial species, avian pathogenic *Escherichia coli* (APEC) and *Gallibacterium anatis*, are routinely isolated from cases of avian peritonitis. The role played by *G. anatis* in the pathogenesis of this disease has only recently been recognized. Danish researchers have reported that *G. anatis* is frequently associated with peritonitis lesions. In addition, peritonitis in table egg chickens has been associated with ascending bacterial infections of the oviduct.

In this study, adult leghorn chickens were inoculated via the oviduct with *E. coli* alone, *G. anatis* alone, or with *G. anatis* and *E. coli* together. *E. coli* alone produced no mortality or lesions. *G. anatis* caused 4 or 10 chickens to die but exudate associated with field cases of peritonitis was missing. *G. anatis* and *E. coli* resulted in the death of 6 of 10 hens with typical lesions of peritonitis in the body cavity.

Preliminary data suggest that to artificially recreate the prototypic lesions of peritonitis in laying hens, chickens must receive concurrent challenge with APEC and *G. anatis*. This research demonstrates that the pathogenesis of peritonitis might be quite complex and involve an interplay of host and polymicrobial factors.

29.

Hemadsorption and Hemagglutination of *Ornithobacterium Rhinotracheale*

**Vicente Vega, Andrea Zepeda, Saúl Ramírez, Pomposo Fernández, Roberto Montes de Oca,
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The hemagglutinating properties of nine (serovars A to I) reference strains of *Ornithobacterium rhinotracheale*, which were tested by using fresh erythrocytes from various animal species, were examined. In general, hemagglutinating activity by the strain BAC 96-0334 #MINN 18 was not observed. Similarly, despite the hemagglutination titer of strains O-95029 no. 12229 and ORV 94084 K858 ORT were adjusted (hemagglutinating titer of 8 and 2, respectively) by using glutaraldehyde-fixed erythrocytes, hemagglutinating activity of fresh erythrocytes of the different animal species was not observed. Obtained results showed differences in the hemagglutinating activity of the reference strains of *O. rhinotracheale*. It is unknown if these differences are involved in pathogenicity and virulence of this bacterium. Further studies are focused on the role of hemagglutinins in the antigenicity, pathogenicity, and immunogenicity of *O. rhinotracheale*.

30.

Hemadsorption and Hemagglutination of *Gallibacterium* Reference Strains

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Hemagglutinating activity and hemadsorption ability by six *Gallibacterium* reference strains were investigated. Differences in the agglutination of fresh erythrocytes of broiler chickens, layer hens, quail, rabbits, and pigs were observed. Fresh erythrocytes of turkeys, pigeons, ducks, Harris hawks, cows, sheep, horses, dogs, or human (types ABO) were not agglutinated by any of the *Gallibacterium* strains. Also, differences in the hemadsorption ability of *Gallibacterium* strains were recorded. It is unknown if differences in the hemagglutination and hemadsorption of *Gallibacterium* strains included in the study could be related with differences in both pathogenicity and virulence. Further studies on the antigenicity, pathogenicity and immunogenicity of the hemagglutinins of *Gallibacterium* will be performed. Obtained results indicate that hemagglutinins may act as a putative virulence factor among *Gallibacterium* strains as similar in other *Pasteurellaceae* members.

CHICKEN ANEMIA VIRUS

31.

Peruvian serological survey for Chicken Anemia Virus in broiler chickens

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The objective of this study was to evaluate the serological profile of CAV in chickens from commercial broilers farms located in the three most populated poultry areas in Peru (Lima, Trujillo and Arequipa). A survey comprised 36 farms and was done from February through March of 2006. Were analyzed 789 broilers serum samples, 325 from one-day old and 464 from market age, was used the competitive Idexx ELISA test at 1:100 dilution. Were obtained information about 'good', 'regular' or 'bad' productive performance in order to make a correlation with final serology titers. We found a higher number of negative samples to CAV antibodies at day old, may be related to type of CAV vaccination program of heavy breeders and to their age. Were observed strong correlation between bad productive performance and CAV seroconversion. The results are being analyzed by Student and Duncan test.

32.

Mapping of epitopes of VP2 protein of chicken anemia virus using monoclonal antibodies

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To map the epitopes of VP2 protein of chicken anemia virus (CAV), VP2 was expressed as a fusion protein in *Escherichia coli* BL21 (DE3). The Western blot demonstrated that recombinant protein could be recognized by sera of chickens infected with CAV. Female BALB/c mice were immunized with purified VP2 produced in *E. coli* BL21 (DE3) and seven VP2-specific MAbs were developed. The results of Western blot showed that all the seven MAbs recognized the VP2 protein expressed in the Baculovirus and reacted with MDCC-MSB1 cells infected with CAV by indirect immunofluorescence assay. The VP2 protein was dissected into 21 overlapping fragments, expressed as fusion peptides in *Escherichia coli* and used for epitope mapping by pepscan analysis. The linear immunodominant epitope of VP2 was located in amino acid residues 111-136.

E. COLI

33.

Emergence of Antimicrobial Resistance, Class 1 Integrons, and R Plasmids Among Avian *Escherichia coli*

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Recent research has revealed that class 1 integrons, containing multiple gene cassettes, may be linked to large transmissible plasmids in avian pathogenic *Escherichia coli* (APEC). In addition to class 1 integrons, these R plasmids may harbor determinants of resistance to heavy metal compounds, such as those containing arsenic, mercury, silver, and copper. Class 1 integrons themselves typically encode resistance to quaternary ammonium disinfectants, sulfonamide compounds, and such antimicrobials, as streptomycin, etc. Therefore, use of multiple agents in the production environment could selectively kill sensitive *E. coli* strains, leaving the resistant, R plasmid-containing strains to emerge. Here, 722 APEC and 179 *E. coli* strains from the feces of apparently healthy birds, isolated over the past 28 years, were assessed for resistance to 15 antimicrobial agents and possession of class 1 integron- and plasmid-associated resistance genes. Results indicate that APEC and avian *E. coli* commensals, characterized by possession of class 1 integrons, certain plasmid replicon types, and several resistances, are emergent. Analyses of the results suggest that these changes are R plasmid associated.



34.

Creation of an *Escherichia coli* Plasmid Genome Database

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While plasmid genome databases exist, they are uninformative, difficult to use, and of limited utility. This becomes problematic for researchers studying organisms such as APEC in which plasmids are abundant. Here we describe the creation of a database that focuses on *E. coli* plasmids. This database allows the user to visualize plasmid maps, perform a cross-database search for genes and proteins, perform alignments between plasmids, and perform BLAST searches of sequences within the database. A description of its use for the study of APEC will be described.

35.

Multiplex PCR can Distinguish between Virulent and Commensal Avian *Escherichia coli*.

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In the present study, we used extensive genotyping data to create a simple multiplex polymerase chain reaction (PCR) protocol for distinguishing avian pathogenic *E. coli* (APEC) from commensal *E. coli* found in the production industry. The genes used included *cvaC*, *iroN*, *hlyF*, *etsA*, *iss*, *aerJ*, *ireA*, and *papC*. To validate our protocol, 129 APEC samples, previously characterized for lethality towards 1-day-old chicks, were compared to 106 avian commensal *E. coli*. Based on the presence of the 8 genes tested, we developed a schema to estimate the likelihood of an isolate's virulence capabilities. Results and implications of this study are discussed.



GENERAL DISEASES AND MANAGEMENT

36.

Focal Duodenal Necrosis (FDN) an Emerging Disease Affecting the Layer Industry

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Focal Duodenal Necrosis (FDN) is an emerging disease affecting the layer industry and is poorly understood. To better recognize what occurs in the duodenum prior to, during, and after FDN, six layer hen gastrointestinal tract (GIT) samples were collected weekly over 11 months. GIT samples were analyzed using DGGE and TRFLP to identify changes in GIT microflora that are associated with disease as well as characteristics that are associated with health. We were able to identify a pathogenic organism highly correlated with FDN lesions. We also identified a beneficial bacterium that appears to be crucial in this complex disease process.

37.

A case of acute intoxication with monensin in broiler chickens

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An acute intoxication affecting a total of 253 chickens occurred in a poultry farm because of an accidental inclusion of monensin sodium in excessive dose in the feed. Mortality rate was about 30 %. The signs of toxicosis included anorexia, weakness, paralysis in which the legs were extended backward and death despite intensive supportive therapy. Few gross lesions were identifiable post mortem: emaciation, generalized congestion and hydropericardium.

Confirmatory diagnosis was realized by laboratory assay. A liquid chromatographic / mass spectrometric (LC/MS) electrospray confirmation method has been used to determine the monensin levels in the suspected feed.



38.

High Mortality of Newly Hatched Commercial Meat Turkeys

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A commercial hatchery experienced up to 15% mortality of newly hatched turkeys over a 3 month period. Birds died in the hatcher trays, and during sexing, servicing, and transferring to the ranches. No changes in management were reported. Numerous breeder-sources were involved. Necropsy of poults found opaque, whitish ascites fluid, and subcutaneous and pulmonary edema in many of the carcasses. Various bacteria were isolated, from the hatchery and the poults, but no links were identified. Thorough cleaning and disinfection, along with changes in temperature and humidity of the hatcher were tried, but to no avail. The cause of the mortality is currently unknown.

39.

Characterization of Broiler Carcasses Condemned at Processing Plants

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Over half of all the whole birds condemned by USDA, FSIS inspectors are for Septicemia/toxemia (Septox) under 9 CFR 381.83. Investigations were conducted during correlations at several processing facilities comparing carcasses that were condemned for septicemia/toxemia to carcasses of birds in the field know to be exhibiting signs of acute sepsis. Digital imaging and histologic samples were submitted. Findings will be reported, and an updated abstract will be submitted to reflect the conclusions.

40.

Proliferative Lung Disease in Broiler Breeder Hens

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Examination of the lungs obtained from thirty broiler breeder hens not yet in peak production (29 weeks old) revealed histologic lesions of a proliferative disease. Characteristics of this condition are hypertrophy of smooth muscle in the walls of parabronchi with hyperplasia and metaplasia of epithelium (normally simple squamous) lining parabronchi, atria, and infundibulae. This proliferative process results in increased cellularity in the respiratory lobules and blocks the air passages from the parabronchi into the air capillary network. In some cases hens had evidence of bacterial (3 hens) or mycotic infection (3 hens), but most cases had no evidence of infection. The term avian proliferative pulmonary disease is suggested because exudative inflammation (fibrin, heterophils, and lymphocytes) is absent or minimal. These proliferative lesions were found in 21 of the 30 hen lungs examined. Fifteen hens with locomotory disturbances were examined and 14 of these had proliferative lesions. Seven normal (no clinical signs) hens were examined and 6 had proliferative lesions. Eight fresh dead hens were examined with only one having proliferative lesions. Fixation of the lungs *in-situ* provides better preservation of lung histology and improves interpretation of proliferative lesions.

41.

The use of a field data collection software to aid in a more informed decision making process in a production setting

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A field data collection database was designed and arranged into a piece of software to help gather information from different areas of a poultry production company in order to compile the data and create information for a more accurate decision making process at an executive level. The modules designed go from breeders all the way to processing. The standard databases help in the collection of breeder mortality surveys, hatchery residue breakouts, chick quality surveys, broiler health surveys, DOA surveys and condemnation correlation sessions. The user will be able to create or modify the databases according to its own needs.



42.

Addressing the Continuing Educational Needs of Poultry Producers

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Recent events of emerging diseases has emphasized the needs of educating poultry producers on avian disease topics, This current study examines the disease concerns of poultry producers over a 12-year period. Questions were obtained from poultry producers and categorized into different categories of disease concerns in order to determine trends in educational needs that should be provided to poultry producers. These results will provide valuable information to educators in university, governmental, and industry by enhancing their educational outreach efforts to poultry producers.

43.

Accidental poisoning of geese with zinc phosphide

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Intentional poisoning of wild birds by farmers is common to reduce the damages produced to the crops.

Every morning the geese from an individual farm were released to go to a lake situated nearby. In an evening the owner observed that the birds didn't come back and he decided to look for them. He found all his geese dead and near the bodies a lot of corn grains covered with a black-grey powder. The pathological exam showed congestion and hemorrhage in liver, congestion in kidney and pulmonary edema. The results of toxicological investigations were positive for phosphide both in corn grains and gizzard contents.



44.

Agroterrorism – Is the Threat To Agribusiness Real?

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Recent events have caused many to realize the potential for disaster when critical infrastructures are damaged or destroyed. The animal production system is a critical infrastructure for U.S. national security. Questions have been raised after the 9-11 tragedy and subsequent anthrax attacks as to whether animals are truly being targeted by adversaries. Foreign nations are conducting biological weapons research. Other adversaries including al Qaeda have explored methods to attack the U.S. with biological weapons. For a safe and secure food supply to remain intact, the federal government must accurately identify the vulnerabilities and potential threat groups.

45.

Histologic evaluation of the extramedullary hemopoietic tissue in healthy and yolk sac infected young broilers.

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It is already known that bone marrow hyperplasia results from inflammatory disease stimuli. However, the response of the extramedullary hemopoietic tissue to infections in young chicks is still unknown. In this study, a histological evaluation of the extramedullary hemopoietic tissue in the heart, yolk sac, liver, spleen, and bone marrow from healthy and yolk sac infected young broilers was made. The results will be discussed.

46.

**Early Diagnosis of Fatty Liver-Hemorrhagic Syndrome
in Commercial Layers Using Clinical Biochemistry**

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Fatty liver-hemorrhagic syndrome (FLHS) is a common nutritional disease in the commercial layers and breeders. The most important clinical sign is a sudden drop in egg production with occasionally increased mortality which cause significant economic loss in poultry industry. The definitive diagnosis of the FLHS is usually made from the findings at necropsy. However, the diagnosis based on the necropsy is usually not help to reduce the economic loss. Therefore, we need early diagnosis using live birds before damage of FLHS in the field. In this study we tried to find early diagnostic method using clinical biochemistry for FLHS.

47.

**Biosecurity multimedia training: a follow-up to the US Poultry & Egg Association training
program**

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This second version of the multimedia biosecurity training program originally produced by US Poultry and Egg Association will bring together key professionals from numerous states and provinces to develop the material required to foster a paradigm shift relative to biosecurity at the individual farm and regional levels. Emphasis will be on program development to improve compliance. Elements of the new program will include a website, a developers' forum for professionals involved in biosecurity program development, "how-to" videos, audit programs, and a scenario generator designed to address regional biosecurity issues. This effort is supported by a grant obtained via the National Poultry Improvement Plan.

48.

Effect of Breeder Flock and Management System on Sibling Turkeys

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In this study the same breed of turkeys was obtained from two different sources (A, B). Poults from each source were divided into two sibling flocks and raised under different management systems (COM, TAU). Turkeys were weighed, dead and cull birds necropsied, and samples taken for diagnostics. Growth rates differed according to poult source and management system. Mortality was affected by management system. *Salmonella* was isolated from all samples until approximately 6 weeks. *Campylobacter* was isolated from the COM flocks, but not the TAU flocks. Chondrodystrophy associated with *Mycoplasma iowae* was related to poult source but not management system.

49.

Interesting Microscopic Lesions in Cases Submitted to the Poultry Diagnostic and Research Center and Georgia Poultry Laboratory Network.

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A review of interesting microscopic lesions from diagnostic cases submitted to the Poultry Diagnostic and Research Center at UGA and the Georgia Poultry Laboratory Network will be discussed. Cases range from commercial birds to game birds and back yard flocks. Microscopic lesions and special stains employed to make a diagnosis will be highlighted. Morphological diagnoses and etiologies will be given when possible.

50.

Routine Histological & Histomorphometric Finding in the Spleen and Bone Marrow of “Small Birds” and Other Carcass Cohorts Condemned at Slaughter Under the Sep-Tox Disposition Category

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2. Auburn University College of Veterinary Medicine
3. Sanderson Farms, Laurel, MS

Routine subjective histological and quantitative histomorphometric evaluations were performed on the spleen and bone marrow of broilers condemned under the septicemia-toxemia disposition category. Spleen follicular areas were enlarged in most carcasses and the bone marrow cellularity increased in a subset of the carcasses. The spleen follicles demonstrated expansion of cells with histiocytic morphology with a relative depletion of lymphocytes. Bone marrow changes involved increases in mature heterophils with variable elevations in primitive forms. While the spleen changes appeared more “sensitive” being present in a greater number of carcasses, bone marrow heterophil hyperplasia may be more “specifically” related to septic conditions.

IMMUNOLOGY, IMMUNITY AND VACCINES

51.

Anatomic histopathology evaluation of thymus, bursa and spleen from broilers chickens raised in reused litter vs new litter

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A total of 250 a day old male commercial broiler chickens from Ross 308 breeders were reared in two pens. Each pen with 125 broiler chickens placed in one pen with new litter and the other with reused litter during 5 flocks. We study the morphogenesis index of the Bursa, Thymus and Spleen (Index Rbo, Rti and Rba) every week. We also saw bursa diameter and the relation between bursa and spleen of both groups. Correlation coefficient was considering between body weight, weight of Lymph nodes and weight-diameter of the Bursa. Lymphoid tissues were evaluated by histopathology. We measure antibody titers using enzyme-linked immunosorbent assays (ELISA) to antibody specific against Gumboro disease (IBDV), Newcastle disease (NCV), Infectious Bronchitis virus (IBV), Chicken Anemia (CAV) and Reovirus (REO) from the beginning of the trial to the end (49 days).

52.

**Practical Comparison of Spray and Barrel Methods
of Vaccine Administration to Broiler Breeder Pullets**

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The barrel method is frequently employed to mass administer vaccine to broiler breeder pullets. This method requires withholding water from the flock prior to vaccine administration, a chlorine and biofilm free drinking system, verification of uniform distribution of vaccine throughout the drinker system and verification that the water is turned back on prior to departure. These are not relevant concerns with spray application techniques resulting in significant time and economic savings.

Field vaccination by the spray and barrel methods were compared by ELISA serology. Percent CV and GMT were monitored for ten paired broiler breeder pullet flocks following administration of Newcastle Disease Virus, Chicken Anemia Virus, Infectious Bronchitis Virus, Infectious Bursal Disease Virus and REO virus vaccines.

53.

Development of a real-time RT-PCR assay for turkey cytokines

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Recent pathogenesis studies with turkey-origin reoviruses (TRVs) in specific pathogen free (SPF) poults have revealed evidence of immunosuppression associated with TRV infection. The ability to quantitatively measure the expression and/or relative amounts of cytokine mRNA in poult immune system tissue (i.e., bursa, spleen) would greatly enhance our ability to understand the immunosuppression caused by the TRVs and other disease agents. To this end, a real-time RT-PCR assay was developed for the turkey cytokines IL-1 β , IL-2, IL-18, and INF- γ . Initial data on the development of this assay will be presented.

54.

U. S. Veterinary Immune Reagents Network

Lillehoj¹, H., Lunney, J., Baldwin, C., LaBresh, J., Horohov, D., Hansen, J., Miller, N., Bengton, E., Chinchar, G., Wilson, M., Wagner, B.

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A major obstacle to advances in veterinary immunology and disease control is the lack of sufficient immunological reagents specific for ruminants, swine, poultry, equine and aquaculture species". Sets of reagents, i.e., monoclonal (mAb) and polyclonal antibodies, that can identify the major leukocyte subsets (T and B lymphocytes, NK cells, macrophages, dendritic cells, neutrophils) are needed to evaluate changes during disease and following vaccination and to give scientists the ability to manipulate these cell populations in order to evaluate their roles in protective immunity as well as in immunopathology. This poster will describe a new CSREES NRI-funded project to develop above reagents which represents a broad community plan to begin to systematically address the immunological reagent gap for the US veterinary immunology research community including for the following groups: ruminants (concentrating on cattle), swine, poultry (primarily chickens with some evaluation of reagents on turkey cells), horses and aquaculture species (concentrating on channel catfish and salmonid trout, two of the principal economically important species) with a goal of 20 reagents per species group.

55.

Interpreting ELISA Test Results

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Enzyme Linked Immunosorbent Assay (ELISA) tests are utilized by poultry veterinarians as an aid in the diagnosis of various diseases. Many times ELISA tests incriminate the correct pathogen, but in other cases the results may lead to an incorrect conclusion. Two case reports demonstrate the two potential outcomes of ELISA testing. In the first case, ELISA tests indicated that airsacculitis was secondary to Newcastle disease virus infection. In the second case, ELISA test results suggested a diagnosis of reoviral proventriculitis. The steps taken to finalize each diagnosis will be presented.

56.

Oligodeoxynucleotides Containing CpG Motifs (CpG-ODN) Induce Predominantly a Th1 Type Immune Response in Neonatal Chickens

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Earlier, we demonstrated that intramuscular administration of oligodeoxynucleotides containing CpG motifs (CpG-ODN) induces protection in neonatal chickens against a lethal challenge of *Escherichia coli*. However the mechanism of induction of the protection was not clear. The objective of this study was to determine the mechanism of induced immunoprotection by determining the expression profile of various cytokines in the spleen and bursa of Fabricius of neonatal chickens following intramuscular administration of CpG-ODN prior to the challenge with *Escherichia coli*. SyBr green real-time quantitative reverse transcription polymerase chain reaction analysis of the RNA demonstrated increased mRNA levels of IL-1 β , IL-6, IL-8, IL-10, IL-18, IFN- γ and MIP-3 α in the spleen of CpG-ODN treated birds, which were significantly different than the mRNA levels detected in the spleen of control birds. However, there was no significant difference in the mRNA levels of IL-2, IL-4, IL-12 β and IFN- α in CpG-ODN treated birds and control birds. Moreover, there was no significant difference in the mRNA levels of any cytokine in the bursa of Fabricius of CpG-ODN treated birds and control birds, with the exception IFN- α mRNA, which was downregulated in CpG-ODN treated birds. These results suggest that administration of CpG-ODN induces a Th1 biased immune response in neonatal chickens, (upregulation of IL-18 and IFN- γ but not of IL-4).

57.

Development of Recombinant Antigens for Multiplexed Serologic Monitoring of Specific Pathogen Free Chickens

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Rapid and accurate detection of adventitious infectious agents or vaccine seroconversion in poultry is critical. A wide variety of avian serology assays commonly use lysates or extracts from conventionally grown microbes as an antigen source for antibody detection; however, some microbes do not grow well or do not grow at all *in vitro* making antigen production and assay development challenging. This poster will describe the production of recombinant antigens for the detection of Marek's disease virus, chicken infectious anemia virus and avian leukosis virus strain J in baculovirus - insect cell or *E. coli* antigen expression systems. Further, it will describe the development and validation of a Luminex xMAP® based Multiplexed Fluorometric ImmunoAssay™ (MFIA™) for these agents.



INFECTIOUS BRONCHITIS VIRUS

58.

Spike gene sequence analysis of Infectious Bronchitis Virus vaccine viruses and their genetic stability *in vivo*

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Although molecular based IBV diagnostic tests have become routine, no IBV typing test currently available distinguishes between vaccine and field viruses. It is important to distinguish vaccine from field viruses so appropriate control measures can be implemented. The spike gene, which can be used to identify different serotypes, was sequenced from IBV Ark, Conn, GA98, DE072, and Mass obtained directly from vaccine vials from the different manufacturers. In addition, the spike gene from vaccine viruses recovered from vaccinated and contact exposed 1-week-old chicks were also sequenced. The sequences from different vaccine vials were compared to determine uniformity, and genetic stability was examined by comparing the vaccine vial sequences with the same vaccine viruses recovered from vaccinated and contact exposed chicks.

59.

Cloning of the pathogenic M41 strain of avian infectious bronchitis virus as bacterial artificial chromosome

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The recent advance in the coronavirus reverse genetics system has increased our understanding of the function of individual gene(s) in viral pathogenesis. Existing reverse genetic systems of avian infectious bronchitis virus (IBV) nonpathogenic Beaudette strain have limited use studying viral pathogenesis in chickens. The objective of our study is to construct infectious clone of a pathogenic IBV as bacterial artificial chromosome (BAC) and characterize its biology in chickens. To accomplish this, we have already amplified and sequenced the entire genome of IBV pathogenic M41 strain, and created two different cDNA libraries, one in BAC vector and another in Topo vector.

INFECTIOUS BURSAL DISEASE

60.

The Use Of Gamma Irradiation For The Inactivation Of Infectious Bursal Disease Viruses.

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The maximum dosage of gamma irradiation approved by the U.S. Food and Drug Administration (FDA) for poultry is 3.0 kilo Grays (kGy). This treatment is designed to reduce bacterial contamination on uncooked poultry carcasses and meat products. The possible presence of infectious bursal disease virus (IBDV) on poultry post-harvest has prompted some countries to study the risk associated with introducing non-native strains of the virus from imported commodities. The goal of this study was to determine if this risk could be reduced using gamma irradiation to inactivate IBDV. At the dosage approved by the FDA, the titers of IBDV vaccine strains were reduced between 0 and 1 log₁₀. Titers of the pathogenic IBDV strains tested were not reduced after the 3.0 kGy exposure. Furthermore, titers of pathogenic viral strains were not reduced following exposure up to 5.0 kGy. As the exposure to gamma irradiation increased the titers of the vaccine strains decreased. At the maximum dosage tested (10 kGy), the 89/03 variant virus vaccine was completely inactivated. Titers of the three classic IBDV vaccine strains were reduced between 1.6 – 2.0 logs after the 10 kGy exposure, however these viruses remained viable after this treatment.

61.

Comparison of the VP4 Sequences Among Very Virulent, Pathogenic and Attenuated Infectious Bursal Disease Viruses

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The viral encoded protease (VP4) of infectious bursal disease virus (IBDV) is involved cleavage of the viral polyprotein (NH₂-pVP2-VP4-VP4-COOH). This protease is essential for the replication of the virus. It has a catalytic serine/lysine dyad in its active site and is similar to LexA and Lon proteases. Because of its role in the self cleavage of the viral polyprotein and subsequent processing of pVP2 to VP2, this protease may be important in the pathogenicity of IBDV strains. In this study, we determined and compared the nucleotide and predicted amino acid sequences of the entire VP4 gene from very virulent, pathogenic classic, pathogenic variant and attenuated strains of IBDV. Phylogenetic analysis revealed two distinct clades, one containing the very virulent strains and the other containing the pathogenic classic, pathogenic variant and attenuated strains.

62.

DNA vaccination conferring protection of chickens against infectious bursal disease by priming with DNA vaccine and boosting with killed vaccine

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One-day-old chickens were intramuscularly injected with DNA plasmid coding for large segment gene of infectious bursal disease (IBD) virus (IBDV) strain variant E (VE) (P/VP243/E) and followed by one or two weekly intramuscular injection of killed IBD vaccine containing both standard and variant IBDV. Chickens were orally challenged with IBDV at 3 weeks old. Chickens primed with P/VP243/E at one day old and boosted with killed IBD vaccine at 1 or 2 weeks old had 80 to 100% protection against challenge by IBDV strain VE or 71 to 100% protection against strain STC. Prior to challenge, chickens in the groups primed with P/VP243/E and boosted with killed IBD vaccine had significantly higher ($P < 0.05$) ELISA and VN titers to IBDV than chickens in the other groups. The results indicated that priming with DNA vaccine and boosting with killed IBD vaccine provides protection of chickens against IBD.

63.

Evaluation of the Pathogenicity of Infectious Bursal Disease Viruses from Layer Flocks in the United States

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Twenty bursal samples were obtained from four vaccinated layer flocks experiencing problems with infectious bursal disease (IBD). All the samples were found to be positive for infectious bursal disease virus (IBDV) by reverse transcriptase-polymerase chain reaction (RT-PCR). The VP2 hypervariable sequence and phylogenetic analysis of samples GA-1, H-30 and CS-2-35 indicated they were nearly identical to the D-78 vaccine strain whereas the HPR-2 isolate was identical to the pathogenic STC classical strain. The deduced amino acid sequence of these isolates revealed few amino acid substitutions in comparison with the sequences of the IBDV classical reference strains. Although three viruses were similar to vaccine strains, all were pathogenic in specific-pathogen-free chickens. There was a significant difference in the bursa-body weight ratios of birds infected with wild-type viruses compared to the D-78 vaccine and uninoculated control groups. Histopathology of the bursas from the wild-type infected groups showed different degrees of follicular depletion and necrosis. These bursas contained IBDV as determined by RT-PCR and the nucleotide sequences of these viruses were identical to the original samples.

64.

Bursal Disease Virus Situation in Mexico: Epidemiology and Phylogenetic studies

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Since 1997 a virulent form of the IBDV virus has been present in the continent, beginning the problem in Brazil, this virulent form has spread in different countries causing major economic losses, due to mortality and complications due to immune suppression.

It is important to have a correct diagnosis on Infectious Bursal Disease Virus (IBDV) In order to establish a correct vaccination scheme and have a better understanding of the Infectious Bursal Disease (IBD) situation, in a given country. Imaging Analysis, PCR RFLP and sequencing are some of the studies that have been carried out in order to have a better understanding on the epidemiology of this virus.

65.

Infectious bursal disease virus (IBDV) nonstructural protein VP5 contributes to virulence of very virulent IBDV.

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To elucidate the molecular mechanism of lethality of IBDV, cytotoxicity of nonstructural protein VP5 derived from different pathotypes of IBDV were compared. VP5 gene of classical virulent and very virulent (vv) IBDV was expressed independently with GFP in CEF cells. When the cells were cultured with brefeldin A which inhibits transportation of VP5 to plasma membrane, no significant differences were found in GFP fluorescence. However, after washing out the brefeldin A, GFP fluorescence intensity was significantly decreased only in vvIBDV VP5 expressing cells. These results indicate that cytotoxicity of VP5 is different between the pathotypes and contributes to virulence of vvIBDV.

66.

Molecular detection and differentiation of infectious bursal disease virus: A review

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Conventional reverse transcription-polymerase chain reaction (RT-PCR) has been useful in detection of IBDV serotypes and to a lesser extent, differentiation of serotype 1 IBDV subtypes. Use of restriction enzymes to find unique patterns of restriction fragments in VP2 protein was successful in differentiating very virulent IBDV from classical and variant subtypes. Current development in real-time RT-PCR has provided a promising means for molecular diagnosis of IBDV infection. Further studies focused on utilizing real-time RT-PCR with melting curve analysis and different regions of the IBDV genome will allow for rapid and accurate detection and differentiation of IBDV subtypes.

67.

An improved method for infectious bursal disease virus rescue using RNA polymerase II system

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Reverse genetics system is an excellent platform to research the construction and function of viruses. The genome modification such as gene recombination, mosaicism, and mutation may interfere with replication, assembly and release of viruses. An efficient, convenient and economical method of rescue virus is undoubtedly required for elevating the efficiency of rescue cripple virus. In this study, we developed a method to rescue infectious bursal disease virus (IBDV) using RNA polymerase II. The genome of IBDV Gt strain, flanked by hammerhead ribozyme and hepatitis delta ribozyme sequences, was cloned downstream of the cytomegalovirus enhancer and the beta chicken actin promoter of the vector pCAGGS. Through direct transfection in various cell lines, IBDV could be rescued efficiently. The RNA polymerase II-based reverse genetics system is efficient, stable, convenient, and fit to various cells. The system not only provides the basis of the gene function research of IBDV, but also is beneficial to the reverse genetics research of other *Birnaviridae* viruses.

LARYNGOTRACHEITIS

68.

Differentiation by RFLP and nucleotide sequencing of ILTV strains isolated during an outbreak in São Paulo, Brazil

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At the end of 2002, an ILT outbreak characterized by respiratory signs and increased mortality in commercial layer flocks from Bastos region, São Paulo State, was observed. The purpose of this study was to determine differences between field ILTV strains isolated during outbreak using RFLP and DNA sequencing techniques. Twelve ILTV-positive samples by PCR were included. RFLP results showed two patterns, six samples showed the A pattern, and one sample have the B pattern. DNA sequencing showed differences between both patterns. The nucleotide sequence of the two Brazilian strains of ILTV showed 99.3% identity between both. The Brazilian strains had between 98.5% to 99.5% identity with published sequences in Genbank.

69.

***In vitro* characterization of Infectious laryngotracheitis virus (ILTV)
isolates from United States (US)**

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Infectious laryngotracheitis virus (ILTV) is an acute respiratory disease of chickens that affects poultry worldwide. Genotyping characterization showed that different types of ILTV isolates are circulating in commercial poultry in USA. The aim of this study is to evaluate the ability of genotypically different US isolates to form plaques hepatoma cell line (LMH) and to evaluate their growth kinetics. Plaque size and growth kinetic of US ILTV isolates will be compared to the USDA reference strain, as well as tissue culture origin and (TCO) and chicken culture origin (CEO) vaccine strains.

MISCELLANEOUS VIRUS

70.

A Duck Hepatitis Virus type 1 is Agent of Pancreatitis and Encephalitis in Muscovy Duckling

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A disease so-called "Necrotizing Pancreatitis of Muscovy Duckling" has been described in France. The main clinical signs are mortality of ducklings of less than 3 weeks, eventually associated with nervous signs and pancreatitis. The necropsic findings were limited to a hypertrophied yellowish pancreas. The histological study showed significant lesions, mainly focused on pancreas and brain. The inoculation of 1-day-old Muscovy ducklings reproduced nervous signs and moderate mortality. Furthermore, the inoculation of 1-day-old Pekin ducklings resulted in mortality within 2 to 3 days p.i., with typical signs of Duck hepatitis virus infection. Serology, viral and molecular approaches demonstrated that this disease of Muscovy duck is actually associated with a DHV-1 isolate.



71.

Epidemiological Studies of Adenoviral Infection in the Broiler farms

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Hydropericardium syndrome (HPS) caused by adenovirus has been occurred in broiler industry and caused severe economy loss because of high mortality and weight loss. The causative agent has been isolated from the broiler farms throughout country. We will describe the epidemiological status of HPS in broiler industry and the biological characteristics of the virus based on virulence, serotype and genomic relationships.

72.

Characterization of M-class genome segments of muscovy duck reovirus

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Despite of many common properties with the ARV, several notable differences has been demonstrated between DRV and ARV. To establish the phylogenetic relationships between DRV and other *Orthoreoviruses* and broad our understanding the *Reoviridae* family, we first determined the genomic sequences of the entire complement of M-class genome segments from DRV S14 and expressed M1 gene segments with *Escherichia coli* and eukaryotic cells. Based on results of the phylogenetic analyses of M1, M2, M3, and S-class genome segments [Kuntz-Simon et al., 2002; Zhang et al., 2006], we speculate that a common ancestral reovirus could have separated into DRV and ARV at recent evolutionary stage with host dependent evolution manner and natural genome reassortment between DRV and ARV did not contribute to the recent evolution of DRV. Expressed in DEF or Vero cells, μ A protein could be detected in the cytoplasm and nuclear.

MYCOPLASMA

73.

***Mycoplasma Iowae* Associated with Vertebral Chondrodystrophy in Commercial Turkeys**

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Turkey eggs of the same primary breed were obtained from two different multiplier breeder sources. Hens were 43 and 48 weeks-of-age at breeder sources A and B, respectively. Eggs from both breeder sources were incubated and hatched at the same time by a commercial meat turkey company. At day of hatch, female turkeys from both breeder sources were transported to two different production facilities. Each 'brood and grow' production facility received poults from both breeder sources. One production facility was a typical commercial turkey farm with separate houses for brooding and growing. The other facility was a small commercial-style house used for both brooding and growing. At both production facilities, turkeys from breeder sources A and B were physically separated and/or individually identifiable so that the breeder source could be recognized throughout. The general purpose of this trial was to compare the production performance of poults from breeder sources A and B, using two facilities and management systems. In this report, we describe an unanticipated and interesting finding, the association of *Mycoplasma iowae* (MI) with vertebral chondrodystrophy. Swollen hock joints (arthritis) and vertebral chondrodystrophy occurred at very low incidences in turkeys from both breeder sources. MI was identified by culture/immunofluorescence and MI-specific PCR from some of the vertebral lesions. This is the first report of MI associated with vertebral chondrodystrophy in turkeys, which should now be considered in the differential diagnosis of this lesion.

NEWCASTLE

74.

Immunoprotection of a Commercial vaccine (VG/GA- strain) against Newcastle disease in Broilers chicken

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Inversiones Veterinaria S.A

In this study there was evaluated the immunoprotection and serologic response of a vaccine containing VG/GA strain of Newcastle disease virus in broilers. Were used 240 Cobb Vantress broilers chickens at day old -, The birds were divided in 6 groups according to different vaccination programs and they were challenged with a pathogenic strain Newcastle disease virus at 25 days old. Mortality, clinical signs, gross lesions and serological response by ELISA an HI test were evaluated at 3,10, 25, 35 and 45 days of age. Antibody titers by HI test using VG/GA antigen were compared to those contain La Sota strain.

75.

Molecular Analysis of Velogenic Newcastle Disease Virus in Avian Species from Venezuela.

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Newcastle Disease Virus (NDV) was isolated in 32 clinical cases of several avian species from Venezuela such as broilers, commercial layers, backyard birds, fighting gamecocks, and psittacine. FTA[®] filter cards were used for sampling and transporting allantoic fluid containing NDV in order to characterize the virus. Nucleotide analysis of the amplified F0 gene product showed that 100% of the isolates possessed a virulent fusion cleavage site (112RRQKR/F117) belonging to a velogenic pathotype with Intracerebral Pathogenicity Index (ICPI) ranging from 1.63 to 1.84. Five non-pathogenic isolates used as controls showed a lentogenic motif (112 RRQGR/L117) with an ICPI of less than 0.3. Sequence homology in a portion of the F gene suggests that in some cases wild birds may be responsible for the transmission of highly pathogenic NDV to commercial and backyard birds.

76.

Development of an Immunochromatographic Kit for the Detection of Newcastle Disease Virus

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In order to develop a rapid, simple immunodiagnostic assay for Newcastle disease (ND), several monoclonal antibodies (MoAbs) to velogenic ND virus (NDV) were produced and used for immunochromatography assay. An immunochromatographic kit using the MoAbs can detect velogenic NDV with high specificity. The minimum hemagglutination (HA) unit of NDV detected was 1 HA unit. These results indicated that the immunochromatographic kit developed in this experiment could be used for detection of NDV in fecal samples with high specificity within 10 minutes.



PARASITIC DISEASES

77.

Effect of coccidiosis on serum alkaline phosphatase levels in chickens

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There is anecdotal evidence that serum alkaline phosphatase (ALP) decreases in coccidia infected poultry. Serum ALP concentration is elevated in hepatobiliary disease, however Marek's disease causes a decrease in ALP despite causing lymphoid tumors in the liver. To study the effect of coccidiosis on ALP concentration, commercial broilers and / or specific pathogen free chickens were challenged orally with *E. tenella* on day 21 of age. The control group and challenge group each consist of 30 chickens. Each group was housed separately in isolation rooms and fed commercial poultry feed free of coccidiostats. Serum ALP will be reported prior to and post challenge. Clinical signs of coccidiosis will also be reported with corresponding serum ALP changes.

78.

Changes in Immune-Related Chicken Cytokine and Chemokine Gene Expression Following *Eimeria* Infection

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The expression levels of mRNAs encoding a panel of 28 chicken cytokines and chemokines were quantified in intestinal lymphocytes following *E. acervulina*, *E. tenella* and *E. maxima* primary and secondary infections. Transcripts of the pro-inflammatory, Th1 and Th1 regulatory cytokines IFN- γ , IL-1 β , IL-6, IL-12, IL-15, IL-17, and IL-18 were uniformly increased after *E. acervulina* and *E. maxima* primary infection, but either unchanged (IL-15, IL-16, IL-18), increased (IFN- γ , IL-10, IL-12), or decreased (IL-2) following *E. tenella* primary infection. Following secondary infection, Th1 cytokines mRNA levels were relatively unchanged. Similarly, mRNA levels of the Th2 or Th2 regulatory cytokines IL-3 and GM-CSF were increased following primary or secondary infection with three parasites. The chemokines IL-8, lymphotactin, and MIP-1 β revealed significant increase following primary infection, but not secondary infection. We conclude that coccidiosis induces a diverse and robust primary cytokine/chemokine response, but a more subdued secondary response.

79.

Kinetic Analysis of Local Gene Expression of Duodenum Intraepithelial Lymphocytes Following Primary and Secondary *E. acervulina* Infections

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Most pathogens enter the host through the mucosal surfaces of the respiratory, digestive and reproductive tracts. The intestinal mucosal surfaces are defended against enteric pathogens by gut-associated lymphoid tissues where intraepithelial lymphocytes (IELs) represent the primary effector cells playing a critical role in protective immune response to mucosal pathogens. *Eimeria*, the etiologic agent of avian coccidiosis, cause significant damage to the gut and reduce the body weight of birds infected with coccidiosis. A library of genes expressed by intestinal IELs of *Eimeria*-infected chickens was developed using the expressed sequence tag (EST) strategy and used as the basis for constructing a 10 K chicken intestinal IEL cDNA array (CIELA). The microarray was constructed with 9,845 IEL ESTs selected from 14,409 IEL cDNA clones as well as 6 control genes and 1 exogenous gene. All elements were spotted in duplicate, fabricating 19,764 spots on the microarray. The first application of this tissue-specific intestinal cDNA microarray was used to examine the transcriptional response of avian IELs to *Eimeria maxima* infection. More than 1,000 elements on CIELA exhibited greater than two-fold changes in gene expression during primary and secondary *E. maxima* infections. Of these, 344 changed significantly (greater than two-fold, $P < 0.05$). This microarray provides a valuable resource for profiling global gene expression to study local host-pathogen interaction against mucosal pathogens.

80.

Cytokine Gene Expression Profile in Fayoumi Chicken after *Eimeria Maxima* Infection

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Coccidiosis is a major parasitic disease of poultry causing substantial economic losses to poultry industry worldwide. Although drugs and live vaccines are commonly used to control coccidiosis at present, understanding underlying genetic mechanisms of disease resistance against coccidiosis will lead to the development of new control strategies against avian coccidiosis. This study was conducted to investigate the immunological basis for the genetically determined coccidiosis disease susceptibility in two inbred lines of Fayoumi chickens. Two inbred lines of Fayoumi chickens which show different levels of coccidiosis susceptibility were evaluated for the expression of 9 cytokine genes: IFN- γ , IFN-1 β , IL-10, IL-15, IL-17, iNOS, LITAF, NK-lysin and TLFSF15. Local gene expression was measured in intestinal intraepithelial lymphocytes (IEL) and splenocytes by real-time RT-PCR at 0, 3, 4 and 5 days after *Eimeria maxima* infection. Significant differences in the kinetics of cytokine gene expression were evident very early after *E. maxima* infection which may influence the type of host immune response against coccidiosis.

81.

Immunomodulatory effects of dietary Safflower leaf on coccidiosis

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This study was conducted to evaluate the effects of dietary safflower leaf on protective immunity against coccidiosis, the most economically important parasitic disease in poultry. White Leghorn chicks were fed a standard diet without or with safflower leaf at 0.1% and 0.5% (w/w) for 3 weeks, the animals were uninfected or orally infected with 5,000 sporulated oocysts of *Eimeria acervulina* at day 12 post-hatch, and protective immunity assessed by body weight gain, fecal oocyst shedding, splenocyte proliferation, T lymphocyte subpopulations, and proinflammatory cytokine gene expression. Dietary supplementation with 0.1% of safflower leaf reversed body weight loss and reduced fecal oocyst shedding compared with animals given a nonsupplemented standard diet. Furthermore, increased splenocyte proliferation and a greater percentage of CD4⁺ T cells, but decreased CD8⁺ cells, were observed in animals fed 0.1% safflower-supplemented diet. Finally, the levels of mRNAs for IFN- γ , IL-8, IL-15, and IL-17 in the 0.1% safflower-supplemented group were increased compared with the nonsupplemented controls. These results indicate that safflower leaf possesses immune enhancing properties and improves protective immunity against experimental coccidiosis when given as a dietary supplement. The effect of safflower on coccidiosis was dependant on the dose of supplement used in this study.

82.

Comparative coccidial oocyst shedding patterns of turkeys given Coccivac-T

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The objective of the study was to compare the coccidial oocyst shedding patterns of turkeys sprayed or orally dosed with Coccivac-T. Vaccination was performed on day of hatch. During the study fresh faecal droppings were collected from each pen every two days and oocyst numbers and ratios of species were determined. Oocysts were detected as early as five days post vaccination. Peak oocyst output was 16-18 days for spray vaccinated birds. Peak oocyst output was 7-8 days with a smaller secondary peak 16-18 days for orally vaccinated birds. This epidemiological information can be used as a reference for field levels of oocysts in vaccinated turkey houses.

PNEUMOVIRUS

83.

Molecular Characterization and Phylogenetic Analysis of subtype B Avian Metapneumovirus detected from Brazilian commercial flocks

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Until 2005, all reported avian metapneumovirus (aMPV) strains detected in Brazil belonged to the subtype A. Subtype B aMPV was isolated and detected by RT-PCR from Brazilians commercial flocks without history of vaccination against aMPV and presenting respiratory signs. RT-PCR results revealed that the isolates were subtype B. Sequence analysis of the G glycoprotein gene fragment confirmed that the isolated strains belonged to subtype B and were not vaccine type. Comparison of the nucleotide and amino acid sequences of G gene of the Brazilians aMPV and subtype B isolates from other countries revealed 95.1 to 96.1% identity. Nucleotide sequences showed 100% identity among the Brazilians subtype B.

84.

Immunization of Broiler Chickens Against Avian Metapneumovirus Infection

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In this study we evaluated the effectiveness of a vaccination program against Avian Metapneumovirus infection in broiler chickens. The trial was done in two commercial farms located in a place where the Swollen Head Syndrome (SHS) is a frequent problem. A total of 185.593 Cobb Vantress broilers chickens at day old (same weight and age of heavy breeders) were used. The birds were divided in two groups, Group A (vaccinated against aMPV by aspersion at the hatchery) and group B (Unvaccinated control). Mortality, clinical signs, lesions and immune response by ELISA test were evaluated. Also were registered body weight, food consumption and feed conversion. Higher mortality in males (0.6 %) and females (0.41%) were obtained in group B than A. However these results indicate that vaccination against aMPV in broilers increase the presentation of SHS in males causing more severe clinical signs, respiratory lesions and colibacillosis. Also better productive parameters were obtained in group B than A, obtaining more body weight (110 and 70g) more gain weight/ bird /day (2,55 and 1,86g) and better feed conversion (27 and 20 points less) in males and females respectively. Both groups showed antibodies titers but for short time. The results are being statistically analyzed.



SALMONELLA

85.

CAUTION – Diagnostic Laboratory Data on Salmonella Testing being Presented

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Presentations on numbers of positive Salmonella results obtained from laboratories need to be viewed with caution. Sampling frequency, intensity and re-sampling can give the wrong impression as to the prevalence of a specific Salmonella serotype. The possible introduction of Salmonella Enteritidis (SE) infected chicks resulted in the contamination of a few premises. The apparent overall prevalence of SE in the state can be distorted by in-depth sampling and re-sampling until a premises tests negative.

86.

Plasmid Mediated Antibiotic Resistance in *Salmonella enterica* serovar Heidelberg from Turkeys

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Salmonella infections are a major public health concern in the United States with approximately 95% of cases acquired by the consumption of contaminated food products. There has been an increase in the percentage of *Salmonella* that are resistant to multiple antibiotics in the last half century. A resistance plasmid from a turkey-associated multidrug resistant *Salmonella enterica* serovar Heidelberg was cloned and sequenced to identify the genes present and examine the regulatory mechanisms that influence the expression of resistance, which could aid in the efforts to improve the selection and use of antibiotics and disinfectants in turkey production and processing environments.

87.

Development and preliminary evaluation of a live recombinant vaccine based on the *sef*ABCD fimbrial operon to elicit immune response in chickens against *Salmonella enterica* serotype Enteritidis

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Salmonella enterica serotype Enteritidis (*S. Enteritidis*) foodborne infections have been frequently associated with contaminated eggs. In this study, we exploited the *sef*ABCD operon to examine its protection potential in a live vaccine. *sef*ABCD encodes all four proteins of the SEF14 fimbria of *S. Enteritidis* and has a role in bacterial pathogenesis. The lethal balance (or $\Delta asd/asd^+$) system was used to obtain plasmid stability in a non-selective environment. Oral treatment of chickens with *sef*ABCD recombinant strain induced a SEF14-specific Ig G and Ig A antibodies in blood and bile, respectively. Treated and control chickens were later challenged with *S. Enteritidis*. Results showed a 100-fold reduction in re-isolation of *S. Enteritidis* from ceca of treated chickens when compared to control birds at 7 days post-challenge.

88.

Organic acid water treatment reduced *Salmonella* horizontal transmission in broiler chickens

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The purpose of the present study was to determine whether an organic acid water treatment could reduce the spread of *Salmonella* (SAL) to naïve birds when infected birds were part of the population. A total of one thousand eighty (1080), day-old Cobb X Cobb male chicks were allocated 60/pen to each of 18 pens by blocks and divided into three treatment groups: T1, unmedicated control; T2, 0.04%; and T3, 0.08% of an organic acid blend (OAB; ACTIVATE[®] US WD MAX). The OAB was added to water from 0-14 days and 42-49 days. Half of the birds in each pen were orally dosed with Naladixic acid resistant *S. heidelberg* on Day 0 and housed with the remaining uninfected birds. The results demonstrated that the OAB treatment significantly reduced horizontal spread of SAL to uninfected birds and reduced environmental SAL contamination.

89.

Functions Exerted by the Type Three Secretion Systems (TTSS) During Salmonella Enteritidis Infection of Chicken Reproductive Epithelial Cells and Macrophages

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Salmonella enterica serovar Enteritidis (SE) is responsible for a large percentage of all cases of salmonellosis. To determine the role of TTSS-1/-2 in reproductive tract colonization of chickens by SE, cultured chicken tubular epithelial cells (CTECI) and macrophages (HD11) were infected with wild type and TTSS-1/-2 mutant SE strains. The intracellular bacterial load and cytokine/chemokine levels at various times post infection were assessed by plate counts and real time reverse transcription PCR, respectively. Our results indicated that TTSS-1 and TTSS-2 are required by SE to colonize CTECI and modulate cytokine expression in both CTECI and macrophages.

TUMOR VIRUSES

90.

***In vitro* transformation properties of the Meq protein of MDV**

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Marek's disease virus (MDV) type 1 is the causative agent of Marek's disease a T-cell lymphoma in chickens. In our laboratory, two MDV strains, Md5, and CVI988 are being investigated for mechanisms of tumor formation. Though both the strains express meq, a MDV oncogene, only Md5 but not CVI988 virus causes T cell lymphoma. We cloned meqs (Md5 meq, CVI988 meq & CVI988 long meq) in a retroviral system to study their *in vitro* transformation properties. All three Meq proteins induced morphological changes, focus formation, and colonies in soft agar in both rat-2 and NIH 3T3 cells.



91.

Effect of Adding an Antibiotic to a Marek's Disease Vaccine

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In vitro and in vivo studies were conducted to evaluate the effect of adding Ceftiofur from two different laboratories to a Marek's Disease vaccine. Commercial vaccines with the strain FC 126 Herpes Turkey Virus were tested in vitro to establish the average of PFU before and after different periods of incubation with the addition of the antibiotic. One day old SPF P-2a chickens were vaccinated with that vaccine as well. The results will be discussed.

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