

AAAP/AVMA Scientific Program



Honolulu, Hawaii
July 15-19, 2006



BOARD OF DIRECTORS

President	Robert Owen
President-Elect	Patricia Wakenell
Secretary-Treasurer	Charles Hofacre
Director-Northeast	Patty Dunn
Director-South	Louise Dufour-Zavala
Director-Central	Ching Ching Wu
Director-Western	Stewart Ritchie
Director-at-Large	Mick Fulton
Director-at-Large	Rick Sharpton
Past President	Richard P. Chin

FOUNDATION BOARD

Robert Owen, Chair	Patty Dunn
Bob O'Conner, Vice Chair	Robert Eckroade
Mark Bland	Yan Ghazikhanian
Bruce Calnek	Charles Hofacre
Richard P. Chin	Stewart Ritchie
Mick Fulton	Rick Sharpton
John Donahoe	Pat Wakenell
Louise Dufour-Zavala	

American Association of Avian Pathologists

2006

Athens, GA 30602-4875

Phone: (706) 542-5645

Fax: (706) 542-0249

Email: aaap@uga.edu

Website: <http://www.aaap.info>

Table of Contents

Special Presentations	7
Schedule of Events – AAAP/AVMA Meeting	8
AAAP Scientific Program	9
Poster Session	15
Abstracts -	
Monday, July 17, 2006	
Session A	27
Session B	36
Tuesday, July 18, 2006	
Session A	45
Session B	55
Wednesday, July 19, 2006	
Session A	65
Session B	74
Posters	84



aaapTM
AMERICAN ASSOCIATION OF AVIAN PATHOLOGISTS

A Sincere Thank You to
Intervet, Inc.

for sponsoring the
AAAP Welcome Reception
at the
Sheraton Waikiki
on Saturday evening,
July 15, 2006

Please be sure to thank Intervet
and all of our sponsors

This year the AAAP Awards Luncheon is being sponsored by Fort Dodge Animal Health.

AAAP appreciates their generous support. Please remember to thank the folks from Fort Dodge.

People who requested their tickets can pick them up in the Poster Room on Sunday, July 16th.

**SHERATON WAIKIKI
LANA'I BALLROOM**

**Monday, July 17th, 2006
11:30 – 2:00 PM**



THANK YOU!

MANY THANKS TO THE 2006 CONTRIBUTORS TO THE AAAP/AVMA
ANNUAL MEETING IN HONOLULU, HAWAII

Platinum (>\$5,000)

Fort Dodge Animal Health
Intervet, Inc.

Gold (1,000-\$4,999)

Alltech
Alpharma Animal Health
Aviagen, Inc.
Charles River Spafas
Elanco Animal Health
Fieldale Farms
Foster Farms
Huvepharma, Inc.
Hy-Line International
IDEXX Laboratories
Lohmann Animal Health
Merial Select
Novus International, Inc.
Schering-Plough Animal Health

Silver (\$500-\$999)

Biomune, Inc.
Gold Kist
Hy-Vac
Jones-Hamilton Company
Perdue Farms
Sanderson Farms
Sunrise Farms

Bronze (<\$500)

Cagle-Keystone Foods LLC
Colorado Quality Research, Inc.
Mountaire Farms



Poster Presenters – VERY IMPORTANT -

Posters for Session 1 must be set up before 7:00 AM on Sunday, July 16th and removed promptly at 3:00 PM on Monday, July 17th.

Posters for Session 2 must be set up before 7:00 AM on Tuesday, July 18th and removed promptly at 3:00 PM on Wednesday, July 19th.

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

SPECIAL PRESENTATIONS

Monday, July 17, 2006

7:00 AM

Keynote Speaker: David Swayne

“The Changing Role of Avian Influenza on Global Avian H Health”

10:00 AM

Reed Rumsey Award Presentation: Kellye S. Joiner

“Pathogenesis of Infectious Bronchitis Virus in Chickens Expressing Different MHC B Complex Genotypes”

Tuesday, July 18, 2006

10:00 AM

Lasher History Lecture: Bruce Calnek

“Avian Diseases: The Creation and Evolution of P. Philip Levine’s Enduring Gift”

1:00 PM

Richard Rimler Memorial Paper: Michele N. Maughan

“Detection, Subtyping, and Characterization of Avian Influenza via cDNA Microarray”



AAAP Schedule of Events -- Sheraton Waikiki (Honolulu, Hawaii)

	Meeting	Room Assignments
Friday, July 14, 2006		
6:00 am – 3:00 pm	AAAP Board of Directors Meeting	Ni'hau
Saturday, July 15, 2006		
6:00 am – 12:00 pm	AAAP Board of Directors Meeting	Ni'hau
1:00 pm – 3:00 pm	AAAP Foundation Board Meeting	Ni'hau
7:00 am – 7:00 pm	ACPV Exam	Kohala/Kona
7:00 am – 7:00 pm	ACPV Exam # 2	Puna
7:00 am – 7:00 pm	ACPV Exam # 3 – (Exam Grading Room)	Hilo
8:00 am – 5:00 pm	Association of Veterinarians in Broiler Practice 5:30 am – 6:30 am Breakfast / O'ahu	Honolulu/Kahuku 11:30 – 1:00 pm Lunch/ O'ahu
1:00 pm – 5:00 pm	Association of Veterinarians in Turkey Production	Waialua
5:00 pm – 9:00 pm	AAAP Opening Reception sponsored by Intervet, Inc.	Lana'i Ballroom
Sunday, July 16, 2006		
6:00 am – 7:00 am	AAAP Awards Committee	Waimea Canyon
6:30 am – 7:30 am	Georgia MAM Alumni Breakfast	Honolulu/Kahuku/O'ahu
6:30 am – 8:30 am	AAAP Animal Welfare Committee	Lana'i Ballroom
12:00 pm – 1:30 pm	California Poultry Medicine Alumni	Honolulu
11:30 am – 1:00 pm	Association of Primary Poultry Breeders Veterinarians Luncheon	Ni'ihau
12:00 pm – 1:30 pm	AAAP Biologics Committee	Kohala
12:00 pm – 4:00 pm	ACPV Board Meeting	Puna
7:30 pm – 10:00 pm	NC State University Poultry Health Management	Ni'hau
Monday, July 17, 2006		
5:00 am – 7:00 am	Association of Veterinarians in Egg Production	Ni'hau
6:00 am – 7:00 am	AAAP History Committee	Hilo
6:00 am – 9:00 am	AAAP Avian Histopathology Manual Committee	Kohala
6:30 am – 7:00 am	AAAP Toxic, Infectious, Miscellaneous & Emerging Diseases (TIME) Committee	Puna
7:00 am – 8:00 am	AAAP Enteric Diseases of Poultry Committee	O'ahu/Waialua
9:15 am – 4:30 pm	Prevention & Control of Avian Influenza in the U.S. (AICAP)	Honolulu/Kahuku
12:00 pm – 2:00 pm	AAAP Awards Luncheon sponsored by Fort Dodge Animal Health	Lana'i Ballroom
3:00 pm – 5:00 pm	AAAP Poultry Research Priorities Committee	Ewa
3:30 pm – 5:30 pm	AAAP Avian Diseases Editorial Board Meeting	O'ahu/Waialua
5:00 pm – 7:00 pm	Association of Primary Poultry Breeders Veterinarians	Kona
5:30 pm – 6:30 pm	AAAP Education Committee	Puna
Tuesday, July 18, 2006		
6:00 am – 7:00 am	AAAP Electronic Information Committee	Akaka Falls
6:30 am – 7:30 am	AAAP Biotechnology Committee	Kohala/Kona
6:00 am – 8:00 am	ACPV Reception / Annual Meeting	Lana'i Ballroom
10:30 am – Noon	AAAP Business Meeting (All members come)	Convention Center Room 316A
11:00 am – Noon	AAAP Epidemiology Committee	Waimea Canyon
12:00 pm – 1:00 pm	AAAP Drugs & Therapeutic Committee	Akaka Falls
3:15 pm – 4:45 pm	AAAP Respiratory Diseases Committee	Ni'hau
6:00 pm – 7:00 pm	AAAP Tumor Virus Committee	Koko Crater
Wednesday, July 19, 2006		
6:00 am – Noon	AAAP Board of Directors Meeting	Ni'hau



American Association of Avian Pathologists
July 15-19, 2006
Hawaii Convention Center
Honolulu, Hawaii



SCIENTIFIC PROGRAM

Monday, July 17, 2006 – Morning Program		
	Room 316A	Room 314
	Moderator: John Glisson	
7:00 AM	Keynote Speaker: David Swayne Room 316A The Changing Role of Avian Influenza on Global Avian Health	
	Moderator: Danny Magee	Moderator: Fred Hoerr
7:30 AM	Interesting Cases from the Poultry Diagnostic Laboratory, Part II Linares, Jose A.	Influence of Embryonic Incubation Temperature on the Immune Response and Performance of Broiler Chickens McElroy, Audrey P., David J. Caldwell, Michael Hulet, Richard Kerr, and Robert M. Gogal
7:45 AM	Four Practical Feed Interventions that Positively Affect the General Health Status of Broilers Allard, Jean Paul	Bacteriophage: An Alternative to Antibiotics in Avian Medicine and Poultry Production Huff, William E., Geraldine R. Huff, Narayan C. Rath, and Annie M. Donoghue
8:00 AM	The Role of Soybean Meal Trypsin Inhibitors in Field Outbreaks of Feed Passage in Broilers Ruiz, N. and F. de Belalcázar	In Vitro Sensitivity Testing of Field Isolates of <i>Mycoplasma gallisepticum</i> Levisohn, Sharon, Irina Gerchman, and Shimon Perk
8:15 AM	Free Range Chicken = Healthy? Lopez, Juan Carlos, Robin McFarlane, and Adriana Rodriguez	<i>Mycoplasma gallisepticum</i> in Commercial Layers Davison, Sherrill, Stanley Kleven, Eric Gingerich, Perry Habecker, Maricarmen Garcia, Susan Casavant, and Robert J. Eckroade
8:30 AM	Wind Speed Effects in "Green-leg" Condemnations in Broilers Cummings, Timothy S., Scott L. Branton, Philip A. Stayer, and Danny L. Magee	Effect of Overlaying F Strain <i>Mycoplasma gallisepticum</i> onto Commercial Layer Hens Previously Vaccinated with 6/85 Strain <i>Mycoplasma gallisepticum</i> Branton, Scott L., Jeff D. Evans, Spencer A. Leigh, Stephanie D. Collier, William A. Dozier, William R. Roush, and Joseph R. Olenrewaju
8:45 AM	Experiences with Peritonitis in a Commercial Table Egg Layer Complex Medina, Hugo A.	Role of <i>Mycoplasma synoviae</i> with or without <i>Escherichia coli</i> Infection in Egg Production and Peritonitis in Commercial Layers Raviv, Ziv, Naola M. Ferguson-Noel, Victoria A. Leiting, Ruth S. Wooten, and Stanley H. Kleven
9:00 AM	Soft Egg Shell Problem and Mortality: absence of Vit D3 in Vitamin Mineral Premix Dhillon, A. Singh and Curt Nelson	A Comparison of the Efficacy of <i>Mycoplasma gallisepticum</i> Vaccines Ferguson-Noel, Naola, Kalen Cookson, Ziv Raviv, Victoria A. Leiting, Ruth S. Wooten, and Stanley H. Kleven
9:15 AM	Calcium Tetany and Early Lay Mortality in Broiler Breeders Martin, Michael, Harold J. Barnes, and Mike J. Wineland	Delayed Serological Response to <i>Mycoplasma synoviae</i> Kleven, Stanley H., Naola Ferguson-Noel, Ziv Raviv, and Victoria A. Leiting
	Break 9:30 AM – 10:00AM	Break 9:30 AM – 10:00AM

Monday, July 17, 2006 – Morning Program		
	Room 316A	Room 314
	Moderator: Suzanne Young-Stamey	Moderator: Hector Cervantes
10:00 AM	Multiple Vitamin Deficiencies in Commercial Turkeys Crespo, Rocio, H.L. Shivaprasad, Richard P. Chin, Portia L. Cortes, and Robert Poppenga	REED RUMSEY AWARD: Pathogenesis of Infectious Bronchitis Virus in Chickens expressing Different MHC B Complex Genotypes Joiner, Kellye S., Frederic J. Hoerr, Sandra J. Ewald, Vicky L. van Santen, James C. Wright, and Haroldo Toro
10:15 AM	Can Poults Survive and Thrive without Hatchery Antibiotics? Hermes, David	A New Aid in the Prevention of Avian Colibacillosis Maiers, Jerry D., K. Cookson, J. Tian, and M. Kumar
10:30 AM	Effect of Electrolytes in Feed and Water on Health, Growth and Biochemical Properties of Chickens Fed an All Vegetable Diet Venne, Daniel, Younes Chourfi, Amer N. Silim, and Sylvain Gingras	Possible Transmission of Campylobacter between Hog and Turkey Production Systems Carver, Donna K., Sophia Kathariou, Sandra Wright, and Robin Siletzky
10:45 AM	The Role of Various Variables on Broiler Performance Parameters when Comparing Two Production Complexes Waldrip, Don	Crop Immune Response Post-Salmonella Enteritidis Challenge in Eight Commercial Layer Breed-Strains and Specific-Pathogen-Free (SPF) White Leghorns Vaughn, Lara E., Peter S. Holt, Randle W. Moore, and Richard K. Gast
11:00 AM	Temperature Programs for Rearing Heavy Broilers Owen, Robert L. and Karen Christensen	Tracking Salmonella thru the Poultry Production Pyramid Maurer, John J., Dana Cole, Charles L. Hofacre, and Michael P. Doyle
11:15 AM	Identification and Correction of Wing Breakage in Broiler Production Williams, Robert M.	Why has Salmonella Serovar Kentucky become Widespread in Poultry? Cole, Dana, Katherine Zamperini, Charles L. Hofacre, and John J. Maurer
AAAP Awards Luncheon Sponsored by Fort Dodge Animal Health 11:30 – 2:00 PM Lana'i Ballroom – Sheraton Waikiki		
	Moderator: Karen Burns Grogan	Moderator: Eric Jensen
2:00 PM	Hurricane Katrina vs. the Mississippi Poultry Industry: Part 1 - The Storm Magee, Danny L., Sue A. Hubbard, Philip A. Stayer, and Marshall R. Putnam	Salmonella Bio-Mapping for Salmonella through the Poultry Processing Plant Stewart-Brown, Bruce, Will Morris, and Clay Silas
2:15 PM	Hurricane Katrina vs. the Mississippi Poultry Industry: Part II - Short Term Response Stayer, Philip A., Marshall R. Putnam, Sue A. Hubbard, and Danny L. Magee	Development and Evaluation of a Recombinant Strain based on the SEF14 Fimbrial Operon against a Salmonella enteritidis Challenge in Chickens Lopes, V.C., B.T. Velayudhan, D.N. Foster, D.A. Halvorson, and K.V. Nagaraja
2:30 PM	Hurricane Katrina vs. the Mississippi Poultry Industry: Part III - Long Term Response Putnam, Marshall R., Philip A. Stayer, Danny L. Magee, and Sue A. Hubbard	Developing a Predictable Gangrenous dermatitis Model for Broilers: lessons learned Collett, Stephen Richard, Young-Jae Cho, John R. Glisson, Charles L. Hofacre, and Margie D. Lee
2:45 PM	Hurricane Katrina vs. the Mississippi Poultry Industry: Part IV - Lessons Learned Hubbard, Sue Ann, Danny L. Magee, Marshall R. Putnam, and Philip A. Stayer	The Association of Enteric Infections with Clostridium perfringens in Broiler Field Cases of Gangrenous Dermatitis and the Successful Reproduction of Gangrenous Dermatitis using Oral or Intravenous Inoculation with Clostridium septicum Davis, Stephen W.
3:00 PM	ADJOURN	ADJOURN

Tuesday, July 18, 2005 – Morning Program

	Room 316A	Room 314
	Moderator: Stewart Ritchie	Moderator: Guillermo Zavala
7:00 AM	Findings from Data-Mining Broiler Companies and Correlating Unique Parameters - Disease Indices, Vaccine Programs, Production Practices, and Timeline Trends Keck, Lloyd, John R. Tierce, and Greg Rennie	Evaluation of Roxarsone and/or Bacitracin Methylene Disalicylate on Broiler Performance Using Necrotic enteritis and Salmonella Challenge Models Heins Miller, Sharon, Mark W. LaVorgna, Stephen W. Davis, and Steven Clark
7:15 AM	Prevalence and Antimicrobial Resistance in Organic and Conventional Broilers Hapner, Kyle M. and Teresa Y. Morishita	Passive Immunity to <i>Clostridium perfringens</i> Type A: Practical Efficacy against Necrotic enteritis under US and Canadian Broiler Management Systems Newman, Linnea J., Charles Broussard, and Richard Phillips
7:30 AM	Identification of Heat Stress Risk Factors in Ontario Boiler Industry Sanei, Babak, Harry Huffman, and Lloyd Weber	Evaluation of Elanco Tylan Premix for the Prevention of Necrotic enteritis in Broiler Chickens Brash, Marina L., Randy N. Bragg, Paul C. Dick, Gordon H. Vessie, and Jeff B. Wilson
7:45 AM	Temperature and Oxygen Conditions during the last Four Days of Incubation in Bone Development of Chickens and Turkeys Oviedo, Edgar O., Michael J. Wineland, Vern L. Christensen, Debbie T. Ort, Michael K. Mann, and Sarah L. Funderburk	Organic Acids Effective to Ameliorate the Negative Impact on Broiler Performance due to Necrotic enteritis Quiroz, Marco A., Charles L. Hofacre, Greg F. Mathis, Julia Dibner, and Chris Knight
8:00 AM	Multistage and Single Stage Incubation Comparison: Field Performance Results with Commercial Yield Breed Broilers in a Paired House Trial Hill, Donna L. and Karen Christensen	Molecular Basis for Antimicrobial Growth Promoter Effects in Poultry Lee, Margie D., Jingrang Lu, and Charles L. Hofacre
8:15 AM	A Comprehensive Worm Study in Several Broiler Breeder Pullet Flocks Rings, Bret and Tom Yazwinski	Evaluation of Oral Administration with Live CU Strain <i>Pasteurella multocida</i> Vaccine in Broiler Breeder Pullets Finklin, Marilynn N., Bradley J. Turner, Charles L. Hofacre, and John R. Glisson
8:30 AM	Evaluation of Vertebral Lesions in Broilers with Gait Abnormalities: A Field Study Young-Stamey, Suzanne, John Barnes, and Ken Powell	Performance Differences in Sister Flocks: Is chicken Anemia Virus (CAV) the Culprit? Sommer, Franz, Carol J. Cardona, and Bruce R. Charlton
8:45 AM	Cycling Patterns of <i>Eimeria spp.</i> and its Correlation with Microscopic Lesions, Shedding of Fecal Oocysts, and Johnson and Reid Scores Ruano, Miguel and Douglas Marvil	The Pivotal Role of Feathers in Chicken Infectious Anemia Virus (CAV) Horizontal Spread Davidson, Irit, Irena Shkoda, Emmanuel Loebb, and Karel A. Schat
9:00 AM	Field Problems with <i>Eimeria maxima</i>: Effects of <i>Eimeria acervulina</i> on Concurrent <i>E. maxima</i> Infections Mathis, Greg F.	Chicken Anemia Virus (CAV) Serology Profile of Breeders and their Progeny Cardoso, Beatriz and Lindolfo Rocha
9:15 AM	Uniformity of Infection and Grow-out Performance following <i>in ovo</i> Vaccination with the Coccidiosis Vaccine Inovocox™ Doelling, V.W., R. Posten, and A. Martin	Vaccination of Broilers using a Live Chicken Infectious Anemia Virus Vaccine Hopkins, Brett A.
	Break 9:30 AM – 10:00AM	Break 9:30 AM – 10:00AM
	Moderator: Jagdev Sharma Lasher History Lecture: 10:00 – 10:30 AM Room 316A <b style="text-align: center;">Bruce Calnek <b style="text-align: center;">Avian Diseases: The Creation and Evolution of P. Philip Levine's Enduring Gift	
	AAAP Business Meeting 10:30 – 12:00 noon Room 316A Convention Center	

Tuesday, July 18, 2005 – Afternoon Program		
	Room 316A	Room 314
	Moderator: Greg Mathis	Moderator: Pedro Villegas
1:00 PM	A Field Report on Salinomycin Toxicity in Broiler Breeder Chickens Corsiglia, Charles, Bruce Charlton, Portia Cortes, and H.L. Shivaprasad	RICHARD RIMLER PAPER AWARD: Detection, Subtyping, and Characterization of Avian Influenza via cDNA Microarray Maughan, Michele N., Travis W. Bliss, David L. Suarez, and Calvin L. Keeler, Jr.
1:15 PM	Construction and Evaluation of Recombinant <i>Salmonella</i> Vaccines expressing <i>Eimeria acervulina</i> Sporozoite and Merozoite Antigens Konjufca, Vjolca, Wanda Soo-Young, Mark Jenkins, and Roy Curtiss III	Evidence of Immune System Dysfunction in Poults Infected with Turkey-origin Reoviruses Day, J. Michael, Mary J. Pantin-Jackwood, and Erica Spackman
1:30 PM	Application of Polymerase Chain Reaction (PCR) and Drug-sensitivity Testing to Compare Species Composition and Anti-coccidial Drug Resistance in <i>Eimeria</i> Isolated from Vaccine- and Coccidiostat-utilizing Poultry Operation Jenkins, M.C., S. Klopp, G. Wilkins, and K. Miska	Investigation on the Pathogenicity of Avian Reoviruses Hafez, Hafez M., Olivai Gooß, Christine Prusas, and Doerte Lueschow
1:45 PM	Characterization of Viral Agents Associated with Runting and Stunting Syndrome in Young Broilers Sellers, Holly S., Erich G. Linnemann, Steven Bell, Susan M. Williams, and Guillermo Zavala	Evaluation of Infectious Bursal Disease and Chicken Anemia Viruses as Key Factors in the Reemergence of Gangrenous Dermatitis in Broilers Purvis, Linda B., Pedro Villegas, Ivan Alvarado, and Francisco Perozo
2:00 PM	The Pathogenesis of Agents Associated with Runting-Stunting Syndrome of Broilers in SPF Turkeys Spackman, Erica, Mary J. Pantin-Jackwood, and J. Michael Day	Field Safety and Efficacy Study of Bursal Disease-Marek's Disease Vaccine, Serotype 3, Live Marek's Disease Vector administered to one-day-old Commercial Birds or 18-day-old Embryonated Commercial Eggs Fernandez, Rafael J., Clovis Oliveira, and Jeovane Pereira
2:15 PM	Molecular Characterization of Enteric Viruses Circulating in the United States Pantin-Jackwood, Mary J., Erica Spackman, and James M. Day	Influence of Maternal Antibodies and Differences in Genetic Background on Infectious Bursal Disease Pathogenesis Rautenschlein, Silke, Jung Arne, Rebeski Dierk, and Scharr Heike
2:30 PM	Comparison of Enteric Virus RT-PCR Results with Production Data from Commercial Turkey Hens Rives, David V. and Mary Pantin-Jackwood	Protection against Infectious Bursal Disease by <i>in ovo</i> Vaccination with a Recombinant Avian Adeno-associated Virus Expressing the VP2 Gene Alvarado, Ivan, Pedro Villegas, Carlos Estevez, Francisco Perozo, and Linda Purvis
2:45 PM	Experimental Reproduction of Transmissible Viral Proventriculitis by Inoculation of Chickens with a Novel Adenovirus-Like Virus (Isolate R11/3) Guy, James S., John Barnes, Lynda Smith, and Maria Evans	Is There a Perfect Timing for Vaccination against Infectious Bursal Disease? Experiences from Field Studies in Broilers Block, Herman, Karen Meyer-Block, Rebeski Dierk, Scharr Keike, De Wit Sjaak, and Silke Rautenschlein
3:00 PM	Adjourn	Adjourn

Wednesday, July 19, 2006 – Morning Program		
	Room 316A	Room 314
	Moderator: Mark Jackwood	Moderator: David Swayne
7:00 AM	Case Report: LT in Broiler Breeder Flock Vaccinated with LT Vector Vaccine Johnson, Benjamin C.	Biological and Molecular Characterization of Recent Asian H5N1 Strains Suarez, David L., Erica Spackman, May J. Pantin-Jackwood, and David E. Swayne
7:15 AM	New Studies using GIS for Understanding Vaccinal Laryngotracheitis Epidemiology Dufour-Zavala, Louise, Allyson Jason, and Chris Somerjian	Overview of the USDA H5/H7 Low Pathogenicity Avian Influenza Program for the Live Bird Marketing System Hegngi, Fidelis N., Andrew Rhorer, Patrice Klein, Karen Grogan, Bruce Carter, and Thomas J. Myers
7:30 AM	Evaluation of Heating and Down Time Effects on Poultry Environment Contaminated with VLTV Jones, Kelli Holloway, Maricarmen Garcia, Marta Jaramillo, and Susan M. Williams	Implementation of the NPIP Low Path H5/H7 Monitoring Program in Commercial Poultry Grogan, Karen Burns, Andrew R. Rhorer, and Thomas J. Myers
7:45 AM	The Incidence and Clinical Significance of Hemorrhagic Enteritis Virus (HEV) Infections in Broilers Rosenberger, John K., Sandra Cloud, Nannette Olmeda-Miro, and Conrad Pope	A Field Report on H3N2 Swine Influenza in Minnesota Turkeys: Prevalence, Effect on Breeders in Production, Serology Comparisons, and Vaccination Strategy Lippert, Ron and Dale C. Lauer
8:00 AM	Characterization of Inclusion Body Hepatitis Adenovirus Isolates Cloud, Sandra S., John Rosenberger, Nannette Olmeda-Miro, and Conrad Pope	Can Live Attenuated Avian Influenza Viruses be Prepared for Use in Poultry? Song, Haichen and Daniel R. Perez
8:15 AM	Inclusion Body Hepatitis as a Primary Disease in Broilers in Saskatchewan, Canada Gomis, Susantha, Davor Ojkic, Bob Goodhope, and Phil Wilson	Protective Efficacy of Inactivated Influenza Vaccines against Highly Pathogenic Asian H5N1 Avian Influenza Viruses in Chickens Jadhao, Samadhan J., Chang-Won Lee, and David L. Suarez
8:30 AM	Protection of Broiler Breeder against Inclusion Body Hepatitis by Vaccination of Grant Parent Flocks with a Bivalent Inactivated Fowl Adenovirus Vaccine Villegas, Pedro, Ivan Alvarado, Eric Jensen, and Gregorio Rosales	ELISA Test for the Differentiation of Infected and Vaccinated Animals (DIVA) using a Natural Truncated NS1 Protein of Avian Influenza Virus Avellaneda, Gloria E., Chang-Won Lee, and David L. Suarez
8:45 AM	Pathogenicity and Cross Protection Studies of IBV Field Isolates of Ontario, Canada Grgic, Helena, Bruce D. Hunter, Peter Hunton, and Eva Nagy	Impact of Respiratory Virus Vaccination on Detection of Avian Influenza Virus Infection in Broiler Chickens Gelb, Jr., Jack, Brian S. Ladman, and Conrad Pope
9:00 AM	Experimental TCV and REV Co-infection in Turkeys Turner, Bradley J., Guillermo Zavala, Taylor Barbosa, Sunny Cheng, Mark Jackwood, and Debbie Hilt	In ovo Vaccination with an Adenovirus Replicative-defective Recombinant Vaccine Protects Chickens against Avian Influenza Virus Challenge Toro, H., De-Chu Tang, David Suarez, and Kent van Kampen
9:15 AM	Effects of Experimental Infection with CAV and/or IBDV on IBV Infection and Immune Response van Santen, Vicky L., Haroldo Toro, Kellye S. Joiner, and Frederic J. Hoerr	Swine Flu (H3N2) in Turkey Breeders Tilley, Becky J., Eric C. Gonder, Chad E. Smith, and Sharon J. Jackson
	Break 9:30 AM – 10:00AM	Break 9:30 AM – 10:00AM

Wednesday, July 19, 2006 – Morning Program		
	Room 316A	Room 314
	Moderator: Kenton Kreager	Moderator: Gregorio Rosales
10:00 AM	<i>In vivo</i> and <i>in vitro</i> characterization of an oncogenic reticuloendotheliosis virus Barbosa, Taylor, Guillermo Zavala, Sunny Cheng, and Pedro Villegas	Duration of Immunity to the 2002-03 California Virulent Newcastle Disease Virus (vNDV) following a Single Newcastle Disease Vaccination of SPF Chickens Kapczynski, Darrell R. and Daniel J. King
10:15 AM	Current Epidemiology of RNA Tumor Viruses Zavala, Guillermo and Sunny Cheng	Killed Newcastle Disease Vaccination resulting in Significant Mortality Reduction in the Presence of Endemic Newcastle Disease Virus Olson, Terry R. and Mark Cox
10:30 AM	Efficacy of MDV and the Duration of Immunity against NDV and LT of the Recombinants rHVT/F and fHVT/LT Hein, Rudolf G., Gwen F. Slacum, and Lillian F. Melson	Incorporation of Realtime Reverse Transcriptase Polymerase Chain Reaction (rtRT-PCR) in Development of a Mucosal Challenge Model for a Lentogenic Strain of Newcastle Disease Virus (NDV) Inman, Melissa, Martin Ficken, and Timothy Miller
10:45 AM	Feather PCR Diagnostic Testing to Optimize Marek's Disease Vaccination Gustafson, Cheryl R., R. Currie, H. LeGalludec, and S. Baigent	Chicken Mucosal Immunity against VG/GA, LaSota and B1 Strains of Newcastle Disease Virus Perozo, Francisco, Pedro Villegas, Ivan Alvarado, and Linda Purvis
11:00 AM	The Role of the Marek's Disease Virus UL13 Gene in Generating Cell-Free Virus Silva, Robert F. and Isabel Gimeno	Role of the Attachment Glycoprotein in Avian Metapneumovirus Virulence and Pathogenesis Govindarajan, Dhanasekaran
11:15 AM	Propagation and Molecular Characterization of a Slow Growing Subgroup A Avian Leukosis Virus that was Originally Isolated from Commercial Marek's Disease Vaccines Fadly, Aly M., Robert F. Silva, and Scott P. Taylor	Development of a Vaccine-Challenge Model for Avian Metapneumovirus Infection in Turkeys: Evaluation of Turkey Turbinate Virus Preparations Halvorson, David, Binu T. Velayudhan, Anil J. Thachil, Sally L. Noll, Daniel P. Shaw, Sagar M. Goyal, and Kakambi V. Nagaraja
11:30 AM	Emergence of Subgroup J Avian Leukosis Virus Neutralizing Antibody Escape Variants in Meat-type Chickens Infected with Virus at Hatch Pandiri, Arun K.R., Willie M. Reed, Robert F. Silva, and Aly M. Fadly	Elimination of APV from Commercial Turkey Farm while Concurrently Stopping Vaccination Garcia, Marion
11:45 AM	Methods to Distinguish Selected Serotype 1 MDV in Dually Infected Chickens Dunn, John R., Shari B. Gross, Richard L. Witter, Robert F. Silva, and Lucy F. Lee	Comparative Evaluation of Avian Metapneumovirus Spray and Eyedrop Vaccination Protocols for their Efficacy to Protect against Challenge Velayudhan, Binu, Anil J. Thachil, Igor Radovic, Sally Noll, David A. Halvorson, Daniel Shaw, Sagar M. Goyal, and Kakambi V. Nagaraja
12:00	Adjourn	Adjourn

Poster Session
Sunday, July 16 – Wednesday, July 19, 2006
Room 315

SESSION 1 **Sunday, July 16 – 7:00 AM – 3:00 PM**
Monday, July 17 – 7:00 AM – 3:00 PM

Avian Influenza

1. **Isolation and Characterization of Type A Avian Influenza Viruses (H9N2) from Poultry Flocks in Jordan**
Al-Natour, Mohammad Q., Nadim M. Amarin, Hisham M. Al-Maaltah, and Ilaria Capua
2. **Transmissibility of H9N2-Avian Influenza Infection among Poultry Farms, Swine, and Farmers of Lebanon**
Barbour, Elie K., Vatche Cagherian, Henssam Sheib, Samar Dankar, Mohammed Farrah
3. **Laboratory Evaluation of the Survivability of H7N2 Avian Influenza and Newcastle Disease Virus to Commercial Disinfectants**
Benson, Eric R., Robert L. Alphin, Brian S. Ladman, George W. Malone, Michael D. Dawson, and Megan E. Lombardi
4. **Sensitivity of Dry Swabs Utilized in an Avian Influenza Surveillance Program**
Charlton, Bruce R., David H. Willoughby, Beate M. Crossley, Sharon K. Hietala
5. **PCR Screening during the First Outbreak of Avian Influenza in Romania**
Coste, Handan, Mihaela Zaulet, Monica Vanghele, and Mihai Turcitu
6. **Analysis and Utilization of CpG-Motif Oligonucleotides as Potential Immunostimulatory Agents against Avian Influenza Challenge**
El-Attrache, John, Adam Jester, Ping Cui, and Blanca Lupiani
7. **Evaluation on Safety and Efficacy of the Killed Vaccine against Low-Pathogenic Avian Influenza in Commercial Layer**
Ha, Bong-Do, In-Pil Mo, Hyun-hee So, Ho-gun Won
8. **Development of a Multiplex RT-PCR for Type A Influenza Virus and Avian H5, H7, and H9 Hemagglutinin Subtypes**
Khan, Mazhar I., Zhixun Xie, Yao-shan Pang, and Jianhua Sun
9. **Production of Nucleoprotein (NP) of Avian Influenza Virus (AIV) from Codon-optimized Synthetic NP Gene**
Kwon, Hyuk-Joon, Tae-Eun Kim, Sun-Hee Cho, and Sun-Joong Kim
10. **Immunopathogenesis of H9N2 Low Pathogenic Avian Influenza Virus Infection in Immunosuppressed SPF Chickens by Cyclosporine A (CsA) Treatment**
Kwon, Ji-sun, Hyun-jeong Lee, Yong-kuk Kwon, Youn-Jeong Lee, Chang-seon Song
11. **Evaluation of New Rapid Methods for the Surveillance of Avian Influenza**
Lamichhane, Chinta M.
12. **Are There Better Ways to Determine the Potential Pathogenicity of Avian Influenza Virus?**
Lee, Chang-Won
13. **Development and Evaluation of H5 Subtype-Specific Monoclonal Antibodies for Avian Influenza Diagnostic Tests**
Lu, Huaguang, Lin Lin, and Bill Scheuchenzuber

14. **Evaluation of the Immune Response of Chickens to Individual Avian Influenza Proteins**
Lupiani, Blanca, Vinayak Brahmakshatriya, Chinta Laminchane, and Sanjay M. Reddy
15. **Comparison of Sensitivity for the Detection of H5N1 using Two Different Serology Techniques**
Munoz, Ricardo and Miao DeYuang
16. **Avian Influenza in Romania – Apparition, Diagnostic and Epidemiology**
Nicolae, Stefan, Eugen Olaru, Juliana Onita, and Handan Coste
17. **Molecular Epidemiology and Surveillance of Avian Influenza Virus in Wild and Domestic Birds**
Pascua, Annabelle M., Nathaniel L. Tablante, and Daniel R. Perez
18. **Validation of H5 and Matrix Real-Time Reverse Transcriptase Polymerase Chain Reaction (rRT-PCR) Bead Reagents for the Detection of H5 Avian Influenza Virus**
Pedersen, Janice C., David L. Suarez, Amaresh Das, Dennis A. Senne, and B. Panigraphy
19. **Prevention and Control of Avian Influenza: Advances and Perspectives**
Avian Influenza Coordinated Agricultural Project (PI: Daniel R. Perez, Co-PI: Richard Slemmons)
Perez, Daniel R.
20. **Use of the Hemagglutination Inhibition Test to Assess Antigenic and Genetic Relatedness of Avian Influenza H5 Proteins**
Pfeiffer, Jennifer and David L. Suarez
21. **Pathogenesis of Avian Influenza Virus with Mutation in the NS1 Gene**
Reddy, Sanjay M., Vinayak Brahmakshatriya, and Blanca Lupiani
22. **Efficacy of a Vectored Fowl Pox-Avian Influenza Vaccine Administered Subcutaneously to One-day-old Broiler Chickens and Challenged with Fowl Pox Virus at Five Weeks Post-Vaccination**
Rojo Barrañón, Francisco J., Rafael Fernandez, Enrique Montiel, Héctor Garcia
23. **Efficacy of a Vectored Fowl Pox-Avian Influenza Vaccine Administered Subcutaneously to One-day-old Broiler Chickens and Challenged with Fowl Pox Virus Local Strain at Five Weeks Post-Vaccination**
Rojo Barrañón, Francisco J., Rafael Fernandez, Enrique Montiel, Héctor Garcia
24. **Avian Influenza Virus Surveillance in Wild Birds in Lower 48 during 2005**
Members of USDA CSREES AIV Surveillance in Wild Birds Network (Richard Slemmons, presenter)
Slemmons, Richard D., Carol J. Cardona, David A. Halvorson, Joseph J. Giambrone, John El-Attrache, and Daniel R. Perez
25. **Characterization of H9N2 Avian Influenza Virus Isolated from Korea**
Sung, Haan-Woo, Youn-Jeong, Lee, Jun-Gu Choi, Eun-Kyoung Lee, Jae-Hong Kim, Hyuk-Moo Choi
26. **H5N1 HPAI Virus in Wild Birds of Mongolia**
Swayne, David E. and David L. Suarez
27. **Vaccination with Alphavirus-derived Neuraminidase Partially Protects Chickens against Heterologous Avian Influenza Challenge**
Sylte, Matthew J., David L. Suarez, and Bolyn Hubby
28. **Responding to an Avian Influenza Outbreak: A Quick Reference Guide**
Tablante, Nathaniel L.
29. **Properties of an Avian Influenza Virus (H4N8) isolated from Adult Tom Turkeys**
Woolcock, Peter R., Jinling Li, Nichole Anchell, George Cooper, and Carol J. Cardona
30. **Quail carry Sialic Acid Receptors Compatible to bind Avian and Human Influenza Viruses**
Wan, Hongguan and Daniel R. Perez
31. **Antigenic and Genetic Studies on H3N2 Influenza A Viruses Isolated from Swine and Turkeys**
Yassine, Hadi
32. **Differentiation of Highly Pathogenic Strain H5N1 of AIV by Real Time PCR in the First Outbreak in Romania**
Zaulet, Mihaela, Handan Coste, Mihai Turcitu, and Monica Vanghele

Bacteria, Miscellaneous

33. **Campylobacter Susceptibility to Ciprofloxacin and Corresponding Fluoroquinolone Concentrations within the Gastrointestinal Tracts of Chickens**
Blore, P.J., I. Reyes-Herrera, K. Cole, and D.J. Donoghue
34. **Chronic Fowl Cholera with Osteomyelitis of the Skull in Broiler Breeders**
Bruzual, J.J., Matilde Alfonso, Guillermo Zavala, and John Smith
35. **The Efficacy of NETVAX[®] Necrotic Enteritis Vaccine in Protecting Cobb 500 Broiler Progeny from Vaccinated Breeders when Artificially Challenged with *Clostridium perfringens***
Davis, Stephen W., Charles Broussard, and Richard A. Phillips
36. **Serotyping and Antibiotic Susceptibility of *Riemerella anatipestifer* Isolates Obtained from Commercial Pekin Duck Flocks in Northeastern United States**
Galloway-Haskins, Rakijah, Alejandro Banda, and Tirath Sandhu
37. **Hemagglutinin Antibody Levels and Protection in Chickens given Infectious Coryza Vaccines**
Garcia F., Alejandro, Ariel M. Ortiz, Patrick J. Blackall, and Fernando Romo
38. **Development of Competitive ELISA to Determine Antibody Titer in Birds Vaccinated with *C. perfringens* Type A Alpha Toxoid (NetVax[™])**
Jayappa, Huchappa, Suzan Dimmick, Lindy Dierks, Joan Schrader, and Terri Wasmoen
39. **Molecular Characterization of *L. monocytogenes* Isolated from Poultry Meat**
Kurkure, Nitin V., Dewanand R. Kalorey, Pramod K. Jaliewar, Prashant S. Gunjal, Arun G. Bhandarkar, and Sukhadeve B. Barbuddhe
40. **A Serotypic Survey of poultry *Pasteurella multocida* Isolates from 1996 to 2005**
Lobsinger, Christine M., Charles L. Hofacre, and Stephan G. Thayer
41. **Virulence and Resistance Genotyping of *Campylobacter* from Production Turkeys in the Midwest**
Logue, Catherine M., Mohamed K. Fakhr, Shana R. Petermann, Ellen Lutgen Johnson, Julie S. Sherwood
42. **Prevalence of Bacterial Pathogens and their Antimicrobial Resistance in Backyard Poultry Flocks**
Moherman, Kimberly L. and J. David Latshaw
43. **A Bacteriologic Survey of Wing Web-applied Vaccine Reservoirs**
McRee, Andy
44. **Bacterial Orchitis and Epididymo-orchitis in Broiler Breeders**
Monleon, Rafael and Harold John Barnes
45. **Virulence and Immunogenicity of a *Riemerella anatipestifer* Serotype 3 Strain**
Sandu, Tirath
46. **M13 and ERIC 1R PCR Fingerprinting of *Ornithobacterium rhinotracheale***
Thachil, Anil J., Binu T. Velayudhan, Vanessa Lopes, Kakambi V. Nagaraja
47. **Histopathologic Lesions of Experimentally Induced Gangrenous Dermatitis in Commercial layers**
Williams, Susan M. and Stephen Collett
48. **Macrolide-resistance in *Campylobacter*: Emerging Frequency and Resistance Mechanisms**
Zhang, Qijing, Meiguan Yan, Sonia Pereira, and Orhan Sahin

E. coli

49. **Role of Iron Acquisition Systems in Virulence of an Avian Pathogenic *Escherichia coli* Strain**
Caza, Melissa, Roy Curtiss III, and Charles M. Dozois
50. **Bacterial Adhesins and Colonization of Respiratory Tissues by Avian Pathogenic *Escherichia coli* (APEC)**
Dozois, Charles M., Maria H. Lymberopoulos, Maryvonne Moulin-Schouleur, Roy Curtiss III
51. **Effects of a Dietary Yeast Extract on the Response to Transport Stress of Turkey Poults Previously Challenged with *Escherichia coli***
Huff, Geraldine, William E. Huff, Narayan C. Rath, Morgan B. Farnell, Fausto Solis de los Santos, and Anne M. Donoghue
52. **Distribution of Plasmids among Avian Pathogenic *Escherichia coli***
Johnson, Sara J., Timothy J. Johnson, and Lisa K. Nolan
53. **Exploring the Evolution of APEC Plasmids through Comparative Genomics**
Johnson, Timothy J. and Lisa K. Nolan
54. **A Novel Virulence Marker of Avian Pathogenic *Escherichia coli***
Kariyawasam, Subhashinie, Timothy J. Johnson, and Lisa K. Nolan
55. **Molecular Characterization of Avian Pathogenic *E. coli* (APEC) in Korea**
Kim, Sun-Joong, Tae-Eun Kim, Sun-Hee Cho, and Hyuk-Joon Kwon
56. **Detection of Iss Protein on the Outer Membrane of an Avian Pathogenic *E. coli* Isolate**
Lynne, Aaron M., Catherine M. Logue, and Lisa K. Nolan
57. **Analysis of the Avian Pathogenic *Escherichia coli* O78:K80:H9 Virulon**
Mukhopadhyay, Suman and Chris D. Herren
58. **Emergence of Virulent *Escherichia coli* Isolates in Poultry Production**
Nolan, Lisa K., Timothy J. Johnson, and Yvonne J. Wannemuehler
59. **Relationship between Host Age and Virulence Capacity of Avian Pathogenic *Escherichia coli* (APEC)**
Scaccianoce, Jennifer A., Yvonne M. Wannemuehler, Timothy J. Johnson, and Lisa K. Nolan
60. **Characterization of *E. coli* Isolates from Peritonitis Lesions in Commercial Laying Hens**
Tampel, Darrell W., Lisa K. Nolan, and Yvonne Wannemuehler
61. **Distribution of Plasmid-mediated Resistance and Resistance Genes among Avian *Escherichia coli***
Wannemuehler, Yvonne, Timothy J. Johnson, and Lisa K. Nolan
62. **Histopathological Effects of Intravenous Injections of *E. coli* on the Bone Marrow in Broilers including Quantitative Histomorphometric Changes**
Wilson, Floyd D., Timothy S. Cummings, and Mark A. Burleson

General Diseases

63. **Performance of Broiler Chickens Raise in Reuse Litter vs. New Litter**
Alba Chinch, Monica, Maria E. Icochea, Pablo S. Reyna, Rosa I. Gonzalez, Branko B. Alva, and Maria P. Vejarano
64. **Utility of Molecular Diagnostic Tests in Timely and Accurate Avian Disease Diagnosis**
Bautista, Daniel A.
65. **Fire Fighting Foam as an Emergency Mass Euthanasia Methodology for Floor-Reared Meat-Type Poultry**
Benson, Eric R., Michael D. Dawson, George W. Malone, Robert L. Alphin, Inma Estevez, and Garrett L. Van Wicklen
66. **Hurricane Katrina's Effect on the Poultry Industry in Mississippi**
Burleson, Mark A., Danny Magee, Sue Ann Hubbard, and Phil Stayer

67. **A Serological Study of One Company's Vaccination Program**
Burluson, Mark A., Phil Stayer, and Tim Cummings
68. **Incidence of Subclinical Diseases and Pathological Conditions in Clinically Normal Broilers from 3 Production Complexes Sorted by Sex and Age**
Cervantes, Hector M.
69. **Mixed Tumor in the Right Oviduct of a White Leghorn**
Cortes, Portia L., Rocio M. Crespo, H.J. Barnes, and H.L. Shivaprasad
70. **Histologic Lesions in the Proventriculus of Broilers and Broiler Breeders**
Fletcher, Oscar J., James S. Guy, and H. John Barnes
71. **Teaching Poultry Disease and the Importance of Poultry in Afghanistan**
Fulton, Richard M. and Robert M. Smith
72. **Use of Composting for Disposing Dead Poultry in Venezuela in Normal and Emergency Situations**
Gomez, Luis B. and Sonia Puche
73. **Efficiency of an Aluminosilicato in the Ammonia Control of Litter of Broiler Chickens**
Icochea, Eliana, Pablo S. Reyna, Jhon H. Guzman, Monica C. Alba, Rosa Gonzalez, and Anthony Berrios
74. **The Effect of Two Broiler Catching Techniques on Wing and Leg Damage**
Jones, Kelli Holloway, Suanne D. Young-Stamey, Stephen R. Collett, Susan M. Williams, and Marilynn Finklin
75. **Pigeon Disease in Georgia 1996-2006**
Kelly, Donna K.
76. **Intestinal Intussusception in 11 wk-old Broiler Breeder Pullets**
Linares, Jose A.
77. **Acidification of Drinking Water: An Overview**
Marsh Johnson, Trisha
78. **Therapeutic and Prophylactic Anticoccidial Sensitivity of Coccivac-B**
Mathis, Greg F.
79. **Intestinal Changes Associated with Feed Deprivation and Recovery from Feed Deprivation in Laying Hens**
Moore, Randle W., Peter S. Holt, and Deana R. Jones
80. **Addressing Poultry Health Needs of Private Veterinary Practitioners**
Morishita, Teresa Y. and Meredith F. Davis
81. **Interfering Effect of Multiple Poultry Agents on Same FTA Cards on the Detection of Poultry Pathogens**
Moscoso, Hugo, Gwen Brown, and Charles L. Hofacre
82. **Prevalence of Antimicrobial Resistance in Migratory Passerines**
Decker, Crystal D. and Teresa Y. Morishita
83. **A Case of Acute Intoxication with Carbofuran in Ducks**
Nica, Daniela and Elisabeta G. Bianu
84. **Biomechanical Factors that Influence Femoral Spiral Fractures of Turkeys**
Oviedo, Edgar O., Peter R. Ferket, H. John Barnes, Diego V. Bohórquez, and Jesse L. Grimes
85. **Litter Impaction of the Lower Intestinal Tract of Broiler-Breeders**
Roza, Kristen, Michael P. Martin, and H. John Barnes
86. **Dermal Squamous Cell Carcinoma in a Six-month-old New Hampshire Red Chicken**
Sarver, Craig F. and Teresa Y. Morishita
87. **Resident Canada Geese: Vectors of Disease**
Schaul, Jordan, Lori Martin, Amna El Tayeb, Teresa Morishita, Peter Kobalka, and Walt Threlfall

88. **Development of Multiplexed Fluorometric Immunoassay for Poultry Diagnostics**
Seletskaja, Elena, Joe H. Simmons, Rajeev Dhawan, William R. Shek
89. **Severe Neuropathy in Broiler Breeder Pullets Associated with High Levels of Dietary Salt**
Senties-Cué, C. Gabriel, Danny L. Magee, Floyd D. Wilson, Philip A. Stayer, and William R. Maslin
90. **The Use of Oxidative Reduction Potential (ORP) as a Measure of the Effect of Water Sanitisers on Gumboro Vaccine**
Silim, Amer, Daniel Venne, Younes Chorfi, and Dianne Frenette
91. **Serological Evidence of Five Poultry Pathogens in Free-Ranging Chickens in Grenada**
Sharma, Ravindra N., Mohamed Iqbal Bhaiyat, Snehal Tawde, and Calum Macpherson
92. **A Comparison of Two Customer Requested and One Industry Derived Lighting Program**
Stayer, P.A., J. Paul Thaxton, Martha L. Ewing, and John Rice
93. **Lactic Acid Fermentation of Poultry Carcasses Prior to Rendering**
Tamim, Nada M., Rami A. Dalloul, Timothy A. Shellem, and John A. Doerr
94. **The Harderian Gland of a Mucosal Effector Site for Viral Infections**
van Ginkel, Frederik W., Vicky L. van Santen, and Haroldo E. Toro
95. **Cutaneous Aspergillosis in Broiler Breeder Pullets**
Van Sambeek, Francene Sophia, Fred J. Hoerr, and Susan Lockaby
96. **Some Answers to the Causes of 'Femoral Head Necrosis'**
Wilson, Floyd D., Philip A. Stayer, Lanny W. Pace, and Fred Muhammad

Immunology, Immunity and Vaccines

97. **Development of Microsphere Assays for the Detection of Antibodies against Avian Pathogens**
Callison, Scott A., Tye O. Boynton, Deborah A. Hilt, and Mark W. Jackwood
98. **Investigating the Avian Macrophage Responses to Different *Eimeria* Species using cDNA Microarray**
Dalloul, Rami A., Hyun S. Lillehoj, Travis W. Bliss, Yeong H. Hong, Dong W. Park, and Calvin L. Keeler, Jr.
99. **Development and Use of an Avian Innate Immunity Microarray (AIMM)**
Keeler, Jr., Calvin L., Hyun Lillehoj, Michael Kogut, Susan Lamont, and Travis Bliss

Infectious Bronchitis Virus

100. **siRNA Inhibition of Infectious Bronchitis Virus Replication**
Jackwood, Mark W. and Ralph A. Tripp
101. **Construction and Evaluation of DNA Vaccines coding for S1 and N of Infectious Bronchitis Virus**
Kwon, Hyuk Moo, Ena Kim, Son Il Pak, and Haan Woo Sung

Infectious Bursal Disease

102. **Detection by RT-PCR and Nucleotide Sequence Analysis of a Duck Circovirus Detected in Pekin Ducks in the United States**
Banda, Alejandro, Rakijah Galloway-Haskins, Tirath Sandhu, and Karel Schat
103. **Efficacy of a Turkey Herpesvirus (HVT-MDV serotype 3)-Infectious Bursal Disease (IBD) Vaccine, Live HVT Vector, IBD-VP2, Administered *in ovo* and to One-day-old SPF Chickens**
Cruz-Coy, Julio S., Clovis Oliveira, Jeovane Pereira, Fernanda Ambrosino, Airton Gaudenci, Francois-Xavier Le-gros, and Nikki Pritchard
104. **Construction and Evaluation of Turkey Herpesvirus Vected Infectious Bursal Disease Vaccine**
Esaki, Motoyuki, Kristi M. Moore, Takanori Sato, Shuji Saitoh, Mayumi Kubomura, Atsushi Yasuda, and Joan Leonard
105. **Serological Response with the Use of Three Different Bivalent Killed Vaccines against IBD and Reovirus in Broiler Breeders**
Gonzalez, Mauricio, Miguel M. Melchoir, Odette Urquiza, and Mario G. Lechuga
106. **Profiling of Infectious Bursal Disease Virus (IBDV) in the U.S.A. and Some Foreign Countries Based on Sequencing Genomic Material during 2003-2005**
Hamoud, Mohamed and Pedro Villegas
107. **Efficacy of the Vaccination against Infectious Bursal Disease in Broiler using an Immune Complex Vaccine at one-day-old**
Icochea, Eliana, Pablo S. Reyna, Monica C. Alba, Rosa I. Gonzalez, Jhon H. Guzman, Karina Vidal, Walter Paredes, and Paola Cruz
108. **Phylogenic Analysis of Very Virulent Infectious Bursal Disease Viruses**
Jackwood, Daral J. and Susan E. Sommer-Wagner
109. **Mechanisms of Cell Destruction by Infectious Bursal Disease Virus**
Khatri, Mahesh and Jagdev M. Sharma
110. **The Role of 4 Different Infectious Bursal Disease Vaccines in the Control of Field Infectious Bursal Disease Virus (IBDV) in a Broiler Ranch in California**
Mazaheri, A., B.R. Charlton, M.C. Bland, G.L. Cooper, A.A. Bickford, F. Sommer, and D.J. Jackwood

111. **Lymphocyte Depletion, Vaccine Virus Detection and Bursal Lesions in Commercial Broilers vaccinated with Commercially Available and Experimental Infectious Bursal Disease Vaccines**
Montiel, Enrique, Nikki Pritchard, Amy Holderfield, Julio Cruz, Robert Smith, and Kari Pack
112. **Infectious Bursal Disease Virus (IBDV) Surveillance**
Olmeda-Miro, Nannette, Sandra Cloud, and John Rosenberger
113. **Identification of Very Virulent Infection Bursal Disease (VVIBD) Virus in Colombia: A Five Year Study**
Tamayo, Maritza, Sergio Velez, and Kalen Cookson
114. **Infecting chickens and Inducing Immune Response by a VP3-deleted Infectious Bursal Disease Virus Expressing the GFP Reporter Gene**
Wu, Ching Ching, Michelle Peters, and Tsang Long Lin
115. **The Use of Live IBD Boosts in Breeders**
McCarty, John E.

Laryngotracheitis

116. **An Outbreak of Infectious Laryngotracheitis in Meat Chickens**
Crespo, Rocio
117. **Vaccination of Broilers using a Recombinant Fowl Pox-Infectious Laryngotracheitis Vaccine Inoculated in ovo**
Hopkins, Brett A.

Miscellaneous Virus

118. **Cross Virus-Neutralization Studies on Turkey Coronavirus**
Boynton, Tye O., Mark W. Jackwood, Scott A. Callison, and Deborah A. Hilt
119. **Sequence Comparison of the Right End of Fowl Adenovirus Genomes Representing Each Viral Species**
Garceac, Amalia and Eva Nagy
120. **Genetic Diversity in Turkey Coronavirus Viral RNA following Passage in Embryonating Eggs**
Hilt, Deborah A., Mark W. Jackwood, and Tye O. Boynton
121. **Selecting the Optimal Age to Vaccinate Turkeys against Hemorrhagic Enteritis Virus**
Hopkins, Brett A.
122. **Dynamics of the Chicken Anemia Virus at Commercial Broiler Farms**
Lechuga, Mario, Elena Salazar, Susano Medina, and Maritza Tamayo
123. **Detection and Sequencing of Avian Astrovirus from Broilers using RT-PCR**
Li, Lanqing, Frederic J. Hoerr, Michael J. Luther, and Emily M. Handley
124. **Spike Protein Gene-based Genetic Analysis of Turkey Coronavirus Isolates from Different Geographic Locations of the U.S.**
Lin, Tsang Long, Chien Chang Loa, Ching Ching Lu, Thomas Bryan, Thomas Hooper, and Donna Schrader
125. **Gene Function Studies of Fowl Adenovirus Type 9 Genome**
Nagy, Eva and Juan Carlos Corredor
126. **Genetic and Serologic Evidence of Reticuloendotheliosis Virus (REV) Integrated Avian Poxvirus (APV-REV) Infection of Wild and Exotic Birds**
Tadese, Theodros, Elvet A. Potter, and Willie M. Reed
127. **Rapid Diagnosis and Differentiation of Avianpox Viruses by Amplification of Specific Gene Fragments**
Tripathy, Deoki N. and Trina Westerman
128. **Production of Recombinant VP1 Protein and Specific Antibodies of Avian Polyomavirus**
Tsai, Hsiang-Jung, Chih-Ming Hsu, and Ing-Cerng Guo

129. **A Reproducible Model for Runting and Stunting Syndrome (RSS)**
Zavala, Guillermo, Taylor Barbosa, and Holly S. Sellers

Mycoplasma

130. **Is F Strain *Mycoplasma gallisepticum* (MG) Vaccine as Contagious and as Easily Spread as Thought?**
Davis, Stephen W., Charles Broussard, and Richard A. Phillips
131. ***Mycoplasma gallisepticum* Vaccine Strain 6/85 Penetrates into Chick Embryo Fibroblasts in High Numbers**
Dohms, John E. and Cynthia M. Boettger
132. **Field Evaluation of TS®-11 and Vectormune® FP-MG+AE Vaccines in Controlling MG Outbreak in Commercial Cage Layer Operation in Arkansas and Its Effects on Flock Performance**
Ghori, Hashim M., Lisa A. Newberry, and Stanley H. Kleven
133. ***Mycoplasma gallisepticum* Detection and Genotyping in Vaccinated Layer Flocks**
Godoy, Alecia, Maricarmen Garcia, Kristi Moore, Motoyuki Esaki, and Joan Leonard
134. **A Case Study of a *Mycoplasma* Problem Breeder Farm**
Kelly, Tami F., Fred Hoerr, Marshall Putnam, and Frank Baker
135. **Modulation of Gene Expression in Chicken Macrophages by *Mycoplasma synoviae***
Lavrič, Miha, Travis W. Bliss, John E. Dohms, Dušan Bencina, Mojca Narat, and Calvin L. Keeler, Jr.
136. **Comparison of Culture and PCR for the Detection of *M. iowae* in Turkey Embryos**
Leiting, V.A. and S.H. Kleven
137. **Further Western Spread of *Mycoplasma gallisepticum* Infection of House Finches**
Ley, David H., Deborah S. Sheaffer, and André A. Dhondt
138. **Mycoplasmosis in Free Living Water Fowls**
Moussa, Salah A.
139. **Natural Co-infection of *Avibacterium paragallinarum* and *A. gallinarum* in *Mycoplasma* spp. Seropositive Game Chickens**
Soriano V., Edgardo, Vladimir E. Morales, Vicente H. Vega, Andrea P. Zepeda, Nydia R. Reyes, Saul A. Ramirez, and Salvador B. Lagunas
140. **Use of *M. cloacale* Bacterin in Control of Infectious Sinusitis in Pheasants and Partridges Negative for *M. gallisepticum*, *M. synoviae*, and *M. meleagridis***
Spasojevic, Radivoje

Newcastle

141. **Phylogenetic Characterization of Endemic Newcastle Disease Viruses Isolated from Wild Birds during 2000 to 2004 in Delaware, Maryland, and New Jersey**
Afonso, C.L., Daniel J. King, David J. Stallknecht, and Richard Slemmons
142. **Exotic Newcastle Disease Virus: A Histopathologic Characterization in Encephalic Regions of Specific Pathogen Free Chickens**
Barri, Adriana, Nestor Ledesma, Guillermo Tellez, and John El-Attrache
143. **The Interaction between Newcastle Disease Virus and *E. coli* in Chickens**
Eltayeb, Amna B. and Robert P. Hanson
144. **Construction and Evaluation of Turkey Herpesvirus Vectored Newcastle Disease Vaccine**
Esaki, Motoyuki, Kristi M. Moore, Takanori Sato, Shuji Saitoh, Sakiko Saeki, Ayumi Fujisawa, Atsushi Yasuda, and Joan Leonard
145. **Real Time RT-PCR Developed for a Phylogenetically Divergent Group of Newcastle Viruses not Detected by Current Tests**
Kim, L. Mia, Claudio L. Afonso, David L. Suarez, and D. Jack King

146. **Stability of Selected Newcastle Disease Virus (NDV) Strains at Environmental Temperatures that Range from Hot to Cold**
King, Daniel J.
147. **Protection against CA02ENDV Challenge of Chickens Vaccinated with Inactivated Vaccines of Newcastle Disease Virus (NDV) from Different Genetic Lineages**
Miller, Patti J., Daniel J. King, and David L. Suarez
148. **The Role of Intergenic Sequences in Pathogenesis of Newcastle Disease Virus**
Yan, Yongqi, Daniel J. King, and Siba K. Samal

Parasitic Diseases

149. **Tiamulin and Semduramicin: Effects of Simultaneous Administration on Performance and Health of Growing Broiler Chickens**
Bafundo, Kenneth W., Annette Schuhmacher, and Jurgen Gropp
150. **The Characterization and Localization of the Protective Antigen SO7 in Developmental Stages of *Eimeria tenella***
Fetterer, Raymond H., Mark C. Jenkins, and Katarzyna B. Miska
151. **Biological Characteristics of the Lesser Species of Chicken *Eimeria***
Fitz-Coy, Steve
152. **Toxoplasmosis in Aviary Zebra Finches**
Fitzgerald, Scott D., Matti Kiupel, and Heather Teater
153. **Investigation of the Efficacy of Essential Oil Product on *Histomonas meleagridis* *in-vitro* and *in-vivo***
Hafez, Hafez M. and Ruediger Hauck
154. **Necrotic Enteritis Association with *Eimeria acervulina* and *E. maxima***
Mathis, Greg F. and Charles L. Hofacre
155. **Poultry Coccidiosis Control Programs: Live Vaccines**
Mathis, Greg F.
156. **Molecular Analysis of the Content and Diversity of *Eimeria* Species Present in Litter of Local Poultry Facilities using Ribosomal DNA (rDNA) Sequencing**
Miska, Katarzyna B., Mark C. Jenkins, and Spangler Klopp
157. **Chemical Constituents and Preliminary Antiparasitic Activity of *Ficus Platyphylla* (DEL)**
Mousa, Rehab S. and Salah A. Moussa
158. **Cytological Diagnosis of Coccidia Infection in Turkeys**
Osorio, Claudia, Steven Clark, Michael Martin, and H. John Barnes
159. ***Histomonas meleagridis*: Genotyping of Isolates using a Novel Technique: C-profiling**
van der Heijden, Harold M.J.F., Wil J.M. Landman, Sophie Greve, and Ron Peek

Pneumovirus

160. **Comparative Analysis of the Virulence of Early and Recent Isolates of Avian metapneumovirus of Turkey Origin from Minnesota**
Nagaraja, K.V., Binu T. Velayudhan, Sally Noll, and David A. Halvorson
161. **Production and Characterization of Monoclonal Antibodies Produced against Avian metapneumovirus Subtype C which React against the Nucleocapsid Protein**
Yu, Qingzhong, Carlos Estevez, and Darrell R. Kapczynski

Reovirus

162. **Characterization of a Novel Reovirus Isolated from Meat Birds in Central Georgia, USA**
Christenberry, Sam and Holly S. Sellers
163. **Reovirus Progeny Challenge Protection in Broiler Flocks with Differing Maternal Immune Status**
Cookson, Kalen and Joe Giambrone
164. **Case Report: Accidental Vaccination of Baby Chicks with a Virulent Reovirus Vaccine**
Dufour-Zavala, Louise and Guillermo Zavala
165. **Epidemiological Studies of Avian Reovirus Infection in the Broilers**
Kim Jong-man, In-Pil Mo, and Jin-Seok Song
166. **Molecular Investigations of Avian Reoviruses**
Lueschow, Doerte, Olivia Gooß, Christine Prusas, and Hafez M. Hafez

Salmonella

167. **Histopathological, Bacteriological, and Serological Investigation of *S. Enteritidis*-challenged Broilers Fed with Diets Supplemented with Non-immunized Egg Yolk Powder**
Agunos, Agnes C., Umapom Silphaduang, and Yoshinori Mine
168. **Use of an Inactivated Salmonella Vaccine in an Australian Poultry Operation**
Groves, Peter J. and Anthony Pavic
169. **Salmonella Shedding following Necrotic Enteritis Challenge: Efficacy of 2 Feed Additives in this Model**
Hofacre, Charles L., Greg F. Mathis, Sharon Heins Miller, and Mark LaVorgna
170. **Prevalence of *C. Jejuni*, *E. coli*, and *Salmonella* spp. at Public Picnic Areas in Metropolitan Parks**
Kobalka, Peter Joseph, Stephanie S. Gfeller-Samples, Teresa Y. Morishita, and Sara Jankovsky
171. **Critical Review of Egg Quality Improvement Programs in Relation to Suppression of *Salmonella enteritidis***
Shane, Simon M.
172. **A Survey of Salmonella Serotypes from Southeastern United States from 1999-2005**
Thayer, Stephan G., Christine M. Lobsinger, and Charles L. Hofacre
173. **Systemic Review of Intervention Strategies for *Salmonella* in Broiler Production and Processing**
Wills, Robert W., R. Hartford Bailey, and Kristin M. Clements

Toxins

174. **Poisoning of Wild Geese by Organophosphated Pesticides**
Bianu, Elisabeta and Daniela A. Nica
175. **Sodium Hypochlorite Toxicity in 5-day-old Turkey Poults**
Chin, R. P.
176. **A Field Investigation of the Total Tissue Arsenic Content of Broilers Medicated with and without 3-Nitro® (Roxarsone)**
Clark, Steven R., Mark W. LaVorgna, Kurt N. Dobson, and Sharon Heins Miller
177. **Evaluation of Tissue Arsenic Levels Following the Withdrawal of 3-Nitro® (Roxarsone) in the Diet**
Dobson, Kurt N., Mark LaVorgna, Steven Clark, and Sharon Heins Miller
178. **Toxicopathological Effects of Ochratoxin A and Its Interaction with Newcastle Disease in Indian Layer Chicken**
Gounalan, S., C. Balachandran, and B. Murali Manohar

Tumor viruses

179. **Characterization of Various Isolates of a Naturally Occurring Recombinant Avian Leukosis Virus using Biological Assays and Polymerase Chain Reaction**
Banat, Ghida R., Robert F. Silva, Willie M. Reed, and Aly M. Fadly
180. **Sensitivity and Specificity of Subgroup-Specific PCR for Detection of Avian Leukosis Virus**
Cheng, Sunny and Guillermo Zavala
181. **Co-infection and Vertical Transmission of Avian Leukosis Virus Subgroup J and Reticuloendotheliosis Virus in Chicken Flocks in China**
Cui, Zhizhong, Shuhong Sun, and Lucy F. Lee
182. **Dissemination and Spread of a Chimeric Marek's Disease Virus Vaccine**
Gergen, Linda R., Joan S. Schrader, Stephanie M. Cook, and Terri L. Wasmoen
183. **Load of MDV DNA in Peripheral Blood as Criterion for Early Diagnosis of Marek's Disease**
Gimeno, Isabel M. and Robert F. Silva
184. **Skin Leucosis Induced by MDV**
Heidari, Mohammad, Scott D. Fitzgerald, Robert F. Silva, and Huanmin M. Zhang
185. **Immunological Evaluation of Marek's Disease Virus Early Infection on the Pathogenesis of Infectious Bronchitis Virus Infection in Broiler Chickens**
Lee, Hyun-Jeong, Ji-Sun Kwon, Youn-Jeong Lee, Yong-Kuk Kwon, Hyung-Kwan Jang, and Chang-Seon Song
186. **The Role of Marek's Disease Virus Gene pp38 in Transactivation of Promoter Activities**
Lee, Lucy, Zhizhong Cui, and Sanjay Reddy
187. **Immunohistochemical Detection of a B-cell Antigen in Lymphoid Tumors in Chickens**
Lockaby, Susan B. and Frederic J. Hoerr
188. **Polymorphisms in the Genomes of Oncogenic and Attenuated Pathotypes of Marek's Disease Virus Serotype 1**
Spatz, Stephen J. and Venugopal Nair



Session A, Monday, July 17, 2006

Room 316A

Moderator: John Glisson

7:00—7:30 AM **“The Changing Role of Avian Influenza on Global Avian Health”**
Dr. David E. Swayne, Keynote Speaker
USDA, SEPRL, ARS; Athens, GA

Session A, Monday, July 17, 2006
7:30—7:45 AM

Moderator: Danny Magee

Interesting Cases from the Poultry Diagnostic Laboratory, Part II

Jose A. Linares, DVM, ACPV
Texas Veterinary Medical Diagnostic Laboratory
Poultry Diagnostic Laboratory
PO Box 84, 1812 Water St.
Gonzales, TX 78629

Advances in digital photography have made it easier to produce, incorporate and/or exchange images as part of my work as a poultry pathologist. I was pleasantly surprised by the positive feedback I received on last year's presentation on interesting cases from the Poultry Diagnostic Laboratory. This presentation will follow the same format presenting case histories, gross lesions and, after a brief period of discussion, the diagnoses. The format will allow interaction with the audience. Once again the intent is to provide a change of pace from basic research with some “old-fashioned” diagnostics.

7:45—8:00 AM

**Four Practical Feed Interventions that Positively Affect the
General Health Status of Broilers**

Jean Paul Allard
OK Industries, Inc.
PO Box 1119
Fort Smith, AR 72902

While all commercial broiler diets in the US are nutritionally complete, there are practical interventions that should be considered by nutritionists to address bird health issues. Specifically this includes using 125,000 IU/lb of Vitamin E in the Starter for improved skin quality and reduced cellulites. Replacement of choline with betaine will improve general gut health and reduce the optimum level of ionophore coccidiostat. The addition of Hydroxy D3 to improve general bone quality will also positively impact gait scores. The use of Propionic Acid blends should be an integral part of an overall salmonella reduction program.



Session A, Monday, July 17, 2006
8:00—8:15 AM

**The Role Of Soybean Meal Trypsin Inhibitors in
Field Outbreaks of Feed Passage in Broilers**

N. Ruiz¹ and F. de Belalcázar²

¹ContiGroup Companies, Inc. (New York, NY), PO BOX 2989, Suwanee, GA 30024

²Nutrianálisis, Bogotá, Colombia

Feed passage (FP) is defined here as the condition observed in commercial flocks of broiler chickens in which droppings lose their normal shape, do not display the characteristic uric acid cover, contain undigested feed visible to the naked eye, have a yellowish-orange color, and frequently are watery, containing intestinal tissue. As a consequence of a FP outbreak the litter becomes wet and slippery. Birds lack uniformity, pigmentation is poor, and despite mortality is not increased, and birds do not look sick, feed conversion and body weights are considerably affected with the subsequent economic loss. Observations in the field of seven of these outbreaks in South America between 1998-2004 has strongly suggested that among the several antinutritional factors in soybean meal, trypsin inhibitors are correlated with FP.

8:15—8:30 AM

Free Range Chicken = Healthy?

Juan Carlos Lopez, Robin McFarlane, and Adriana Rodriguez

Lincoln University

PO Box 93

New Zealand

The poultry industry has been experiencing a tendency to use more *natural husbandry systems; free range chicken*, where it is more difficult to control environmental factors. The results of studies on the effects of temperature on humoral and cell-mediated immune responses are variable.

In the present study, immune and endocrine systems were monitored in order to understand the role of temperature on broiler welfare. The humoral and cell mediate immune response (antibodies, interferen gamma levels) to IBV and SRBC, the lesions, clinical signs, levels of corticosterone and tonic immobility showed that temperature affect the homeostatis of the birds in antigen-dependent manner.



Session A, Monday, July 17, 2006

8:30—8:45 AM

Wind Speed Effects on “Green Leg” Condemnations in Broilers

Timothy S. Cummings, Scott L. Branton, Philip A. Stayer, and Danny L. Magee

Mississippi State University
College of Veterinary Medicine
P.O. Box 6100
Mississippi State, MS 39762

“Green Leg” condemnations in commercial broilers has been a problem for certain complexes. This condition is caused by ruptured and/or damaged gastroc tendons, and has typically been associated with reovirus infections or the use of certain feed additives. It has been suggested that tendon strain caused by prolonged sitting by the broilers, especially during seasons of heat stress, may be predisposing the tendons to ischemia and subsequent necrosis resulting in the condition described. Thus, the purpose of this project is to assess the effect of wind speed and warm temperature on the incidence of green-leg in commercial broilers.

8:45—9:00 AM

Experiences with Peritonitis in a Commercial Table Egg Layer Complex.


Hugo A. Medina

Sparboe Companies
2625 Zanzibar Lane, Plymouth, MN 55447

Peritonitis is a serious problem for the egg industry. It appears that this problem has been kept out of the research scope, and people who experience this problem only talk about it to close associates. People are reluctant to share their experiences with the challenge, so there is little information about the onset and process of the disease in the field and the successes or failures of treatments implemented to reduce mortality or prevent its presence. Also, it has been discussed to be a problem in breeders (broiler and commercial egg layers).

Characteristic Peritonitis lesions are an inflammatory response. Serous and edematous exudates tend to accumulate in the coelomic cavity. Exudates undergo cessation to form a firm, dry, yellow, irregular, cheese-like mass. Time determines the extent and size of the exudates. The most common sign or symptom is the presence of significant mortality on hens in production.

Peritonitis can be initiated in and affect other organs. Organs that can be affected are ovaries (oophoritis), oviduct (Salpingitis) air sac (air-sacculitis), intestinal tract (enteritis) and systemic (septicemia) infection. The most common bacteria isolated from peritonitis lesions is *Escherichia coli* (*E. coli*) and in a lesser frequency other bacteria types, including *Enterococcus*, *Pasteurella*, *Salmonella*, *Staphylococcus* and *Streptococcus*.



Session A, Monday, July 17, 2006
9:00—9:15 AM

Soft Egg Shell Problem and Mortality: absence of Vit D3 in vitamin mineral premix

A. Singh Dhillon and Curt Nelson
Avian Health and Food Safety Laboratory
Washington State University
Avian Health and Food Safety Laboratory
7613 Pioneer Way E, Puyallup, WA 98371

Nine houses containing 800,000 egg layers were affected by severe softshell egg problems with an increase in mortality. Signs of a respiratory disease were not reported from any of the flocks. Chickens submitted for necropsy from two houses had lesions of soft fragile and crooked breastbone. The ribs were soft and caved in. Rupture of ovarian follicles and associated egg yolk peritonitis was present in all dead birds necropsied. Choanal swab samples tested from two affected flocks were negative for AI and NDV by RRT-PCR. Results of virus isolations were negative. Analysis of vitamin and mineral supplement received was found to be missing Vit D3.

9:15—9:30 AM

Calcium tetany and early lay mortality in broiler breeders

Michael Martin, Harold J. Barnes, and Mike J. Wineland
North Carolina State University
College of Veterinary Medicine
Dept. of Population Health & Pathobiology
4700 Hillsborough Street
Raleigh NC 27606

Early-lay mortality in broiler breeders is a significant cause of economic loss. Causes of early-lay mortality are poorly understood.

“Calcium tetany”, theoretically caused by low blood calcium prior to peak lay, has only been recently described and is not well documented. Affected hens show signs of panting, lethargy, immobility, and death. Mortality can rise until peak egg production. Presently, hens approaching peak production with neural and mobility deficits without obvious traumatic lesions often are classified as having “calcium tetany” without further diagnostics.

We have identified hens with clinical signs consistent with “calcium tetany” without low blood calcium. This unique clinical presentation of early-lay mortality in broiler breeders will be presented.

9:30 – 10:00 AM

BREAK



Session A, Monday, July 17, 2006
10:00—10:15 AM

Moderator: Suzanne Young-Stamey

Multiple Vitamin Deficiencies in Commercial Turkeys

Rocio Crespo, H. L. Shivaprasad, Richard P. Chin, Portia L. Cortes, and Robert Poppenga
CAFHS – Fresno, University of California Davis
2789 South Orange Ave.
Fresno, CA 93725

This paper describes the pathology of lipid soluble vitamins deficiency in commercial turkeys. Birds were between 3 and 8 weeks old. The flock had increased mortality. The birds were depressed, uneven, and had watery contents in the small intestine. Histologically, there was evidence of squamous cell metaplasia, rickets, and/or encephalomalacia. Vitamins A and E were below normal in the liver, serum, and feed. No reliable assay exists for vitamin D analysis. As soon as the diagnosis was made, birds were supplemented with vitamins and feed corrected. Most of the birds recovered; however, flock performance at slaughter was below expected.

10:15—10:30 AM

Can Poults Survive and Thrive without Hatchery Antibiotics?

David Hermes
Perdue Farms, Inc.
P.O. Box 539
Washington IN 47501

Three different hatchery day-one injection protocols were used in a Midwest turkey hatchery for 3 months. Ceftiofur, Gentamicin, and “antibiotic-free” protocols were evaluated for their effect on early livability. Overall field performance and livability was also measured. This paper will review the results.



Session A, Monday, July 17, 2006
10:30—10:45 AM

Effect of electrolytes in feed and water on health, growth and biochemical properties of chickens fed an all vegetable diet

Daniel Venne, Younes Chourfi, Amer N. Silim, and Sylvain Gingras
Couvoir Scott Ltée, Université de Montréal, ITA La Pocatière
1798 route Kennedy, Scott, Québec, Canada, G0S 3G0

This paper will present the clinical problems associated with feeding an all-vegetable diet to broilers and present results of a trial aimed at gathering biochemical blood data from birds given 3 water treatments to accentuate and alleviate hyperkalemia.

Subjects discussed will include the effect of price of soya on potassium levels in feed
Effect of potassium, sodium and chloride in water in addition to the levels in the feed.

Effect of excess potassium on hematocrit, Na, K, Cl, Ca, P, Alp, CK.

Incidence of pasty vents

Effect of potassium in feed and water on early chick mortality

10:45—11:00 AM

The Role of Various Variables on Broiler Performance Parameters When Comparing Two Production Complexes

Don Waldrip
Wayne Farms
Gainesville, GA

Various methods are utilized by broiler production companies to evaluate live operating efficiency, including subscription to anonymous data comparisons from wide access accounting firms such as Agristats Inc. When answers to poorer performances are not forthcoming from these as well as other analytical tools a more in-depth study of potential influencers should be utilized. This paper will discuss such a comparative between two separate production complexes with no commonly identified factor(s) that would explain the differences.



Session A, Monday, July 17, 2006
11:00—11:15 AM

Temperature Programs for Rearing Heavy Broilers

Robert L. Owen and Karen Christensen
University of Pennsylvania, School of Veterinary Medicine
382 West Street Road
Kennett Square, PA 19348

A study was designed to test the effect of reducing the environmental temperature set point during growout of yield-type broilers after 35 days of age from the conventional 70°F to 65°F in an effort to cool the birds and improve air quality. Results showed a 0.18 lb improvement in body weight, a 5 point improvement in feed conversion, and a 1.75% improvement in mortality in the group kept cooler during late growout. Average body temperature for the group reared under conventional conditions was 107.4°F versus 106.8°F for birds reared under cooler conditions.

11:15—11:30 AM


Identification and Correction of Wing Breakage in Broiler Production

Robert M. Williams, DVM, ACPV
Embrex, Inc., Research Triangle Park, NC

Wing breakage has become an integral part of animal welfare concerns. The following is an assessment of catching, unloading and shackling broiler chickens. Interestingly catching is a much smaller issue than is the unloading-shackling process. Data consistently showed wing breakage prior to unloading to be less than 1.00% in two complexes and less than 1.5% in all three complexes studied. Dependent upon changing unloader cage size, conveyance layout and belt drops, breakage between unloading and the kill machine ranges from <1% to greater than 3%. Developing solutions revolve around having a large unloading cage, dark surroundings, straight as possible conveyances and as few drops as possible.

AAAP AWARDS LUNCHEON
SHERATION WAIKIKI
LANA'I BALLROOM

11:30 – 2:00 PM



Session A, Monday, July 17, 2006
2:00—2:15 PM
Grogan

Moderator: Karens Burns

Hurricane Katrina vs. the Mississippi Poultry Industry: Part I – The Storm

Danny L. Magee, Sue A. Hubbard, Philip A. Stayer, and Marshall R. Putnam
Mississippi State University College of Veterinary Medicine
Poultry Research & Diagnostic Laboratory
P. O. Box 97813
Pearl, MS 39288

The standard for measuring and comparing hurricanes in recent times has always been hurricane Camille which struck the Mississippi Gulf Coast in 1969. But in August 2005, hurricane Katrina changed all the rules and standards. The size, power and track of this storm struck a serious blow to Mississippi's economy including its largest agricultural industry – poultry.

2:15—2:30 PM

Hurricane Katrina vs. the Mississippi Poultry Industry: Part II – Short Term Response

Philip A. Stayer, Marshall R. Putnam, Sue A. Hubbard, and Danny L. Magee
Sanderson Farms
Poultry Research & Diagnostic Laboratory
P. O. Box 97813
Pearl, MS 39288

Following the initial impact of hurricane Katrina, the Mississippi Poultry industry realized that it was not prepared for a storm of this magnitude. The first priority was to insure the safety and well being of personnel. Emergency management plans were implemented but efforts were limited in many instances due to lack of communications, electricity and fuel. The industry faced problems everywhere including hatcheries, farms, feed mills and processing plants.



Session A, Monday, July 17, 2006
2:30—2:45 PM

Hurricane Katrina vs. the Mississippi Poultry Industry: Part III – Long Term Response

Marshall R. Putnam, Philip A. Stayer, Danny L. Magee, and Sue A. Hubbard
Wayne Farms
Poultry Research & Diagnostic Laboratory
P. O. Box 97813
Pearl, MS 39288

In the days and weeks following hurricane Katrina, the Mississippi poultry industry had to deal with a multiplicity of unexpected problems. These included such things as dead bird disposal, hatching egg disposal, decreased hatchability, thousands of live birds running loose on farms, increasing live bird size and age, difficulty in making feed deliveries, problems with live haul, decreased processing capabilities and labor problems. These were generally handled by the individual complexes as the problems arose.

2:45—3:00 PM

Hurricane Katrina vs. the Mississippi Poultry Industry: Part IV – Lessons Learned

Sue Ann Hubbard, Danny L. Magee, Marshall R. Putnam, and Philip A. Stayer
Mississippi State University College of Veterinary Medicine
Poultry Research & Diagnostic Laboratory
P. O. Box 97813
Pearl, MS 39288

The Mississippi poultry industry learned a multitude of lessons following hurricane Katrina. Companies in the state have reviewed how they would handle similar situations in the future. The areas of preparedness, response and recovery were all flawed in one way or another, and hurricane Katrina made the industry realize the reality of these flaws. If you can find one good thing that came out of hurricane Katrina it may be that the Mississippi poultry industry is now more prepared to handle any future catastrophe it may encounter.

3:00 PM

ADJOURN



Session B Monday, July 17, 2006
7:30—7:45 AM

Moderator: Fred Hoerr

Influence of Embryonic Incubation Temperature on the Immune Response and Performance of Broiler Chickens

Audrey P. McElroy, David J. Caldwell, Michael Hulet, Richard Kerr, and Robert M. Gogal
Virginia Tech

Department of Animal and Poultry Sciences
3140 Litton Reaves Hall
Blacksburg, VA 24061-0306

The current situation in yield-type poultry incubation has resulted in increased metabolic heat production by embryos that is difficult to dispose of due to poor airflow, and as a result, hatch of fertiles has decreased and problems with embryonic development and subsequent bird performance have become challenges. Experiments investigated a link between incubation temperature and resulting immunocompetence of broilers, which could impact in ovo or day of hatch vaccination efficacy, disease resistance, and performance. Examination of body and organ weight, tissue density, T-cell subset analysis, and lymphocyte transformation assays indicated a significant impact of elevated temperature on early immune response and performance parameters.

7:45—8:00 AM

Bacteriophage: An Alternative to Antibiotics in Avian Medicine and Poultry Production

William E. Huff, Geraldine R. Huff, Narayan C. Rath, and Annie M. Donoghue

USDA/ARS Poultry Production and Product Safety Research Unit
Poultry Science Center
University of Arkansas
Fayetteville, AR 72701

Bacteriophage are viruses that infect and kill bacteria. Bacteriophage do not infect either animal or plant cells making them a natural and potentially safe alternative to antibiotics. We have shown that an aerosol spray of bacteriophage can be used to prevent colibacillosis in poultry, and that bacteriophage can be used to effectively treat colibacillosis in poultry. Our research has demonstrated that bacteriophage have the potential to provide an effective alternative to antibiotics in animal production.



Session B Monday, July 17, 2006

8:00—8:15 AM

In Vitro* Sensitivity Testing of Field Isolates of *Mycoplasma gallisepticum

Sharon Levisohn, Irina Gerchman, and Shimon Perk

Division of Avian and Aquatic Diseases

Kimron Veterinary Institute, POB 12

Bet Dagan, Israel 50250

In vitro sensitivity testing (MIC) of field isolates of *Mycoplasma gallisepticum* (MG) is not routinely performed due to the necessity to isolate the organism in pure culture and the lack of standardized methods and guidelines for interpretation of MIC values. We determined MIC values in reference strains for currently used antibiotics by a standardized microdilution method and the commercial E-test, with excellent correlation between the methods and with literature values. MICs were determined for MG isolates from outbreaks in breeder flocks and from meat-type turkeys with a history of flock and farm treatment with antibiotics. Noteworthy is the presence of increased resistance to enrofloxacin in MG strains in the meat-type turkeys. Pros and cons of the E-test will be discussed.

8:15—8:30 AM

***Mycoplasma gallisepticum* in Commercial Layers**

**Sherrill Davison, Stanley Kleven, Eric Gingerich, Perry Habecker, Maricarmen Garcia,
Susan Casavant, and Robert J. Eckroade**

Laboratory of Avian Medicine and Pathology,


University of Pennsylvania, Kennett Square, PA 19348

Isolation and characterization of the causative strain(s) of MG is often difficult due to overgrowth of non-pathogenic mycoplasmas during culture attempts. To eliminate this problem, Dr. Stanley Kleven suggested the novel approach of a bioassay using sentinel turkeys. Turkeys, which are highly susceptible to pathogenic MGs, act as “filters” for the MG when commingled with MG infected layers in a laboratory setting. The turkeys are cultured for MG rather than the chickens.

Fifteen trials were conducted using the new isolation approach and ten MG isolates were isolated and characterized by random amplified polymorphic DNA (RAPD) and gene-targeted sequencing (GTS). These included four “wild” type MGs, one “ts-11-like” isolate, one “ts-11-derived” isolate, and one “F strain-derived” isolate. Two isolates are pending GTS and one additional isolate was able to be typed as MG and failed to grow in further cultures.

Last year, we reported on pathogenicity trials in layers of one “ts-11-like” MG and two “wild” type MGs. The pathogenicity of the isolates was evaluated by clinical signs, air sac score, and tracheal mucosal thickness. The “ts-11-like” strain demonstrated minimal pathogenicity relative to the negative (no challenge) and positive (R strain) control groups. The two “wild” types demonstrated a greater degree of pathogenicity over the “ts-11-like” strain in every measured parameter including clinical signs of infection, airsacculitis, and tracheitis.

The new information concerning this project involves a vaccine protection study. The “ts-11-like” MG and two “wild” type MGs were used to evaluate MG vaccine efficacy in layer pullets. Pullets were vaccinated with one of the commercially available vaccines and challenged with one of the isolates. Vaccine protection was evaluated by serology, clinical signs, air sac score, and tracheal mucosal thickness. Initial results indicate that both mild, live MG vaccines protected against the “ts-11-like” strain but showed reduced protection against the “wild” type MGs. Ongoing trials are being conducted to evaluate the other commercially available vaccines.



**Session B Monday, July 17, 2006
8:30—8:45 AM**

**Effect of Overlaying F Strain *Mycoplasma gallisepticum* onto Commercial Layer Hens
Previously Vaccinated with 6/85 Strain *Mycoplasma gallisepticum***

**Scott L. Branton, Jeff D. Evans, Spencer A. Leigh, Stephanie D. Collier, William A. Dozier,
William R. Roush, and Joseph R. Olenrewaju**
USDA-ARS, South Central Poultry Research Laboratory
P. O. Box 5367;
Mississippi State, MS 39762

Two trials were conducted wherein a total of 320 commercial layer chickens were placed in the following four treatments: 1) Control (mycoplasma clean), 2) 6/85 *Mycoplasma gallisepticum* (MG) at 10 wk of age, 3) 6/85 MG at 10 wk with overlay of F strain MG at 22 wk, or 4) 6/85 MG at 10 wk with overlay of F strain MG at 45 wk. Egg production, various egg and eggshell quality parameters, and egg size distribution were followed over 58 weeks of age. Significant differences ($P \leq 0.05$) were observed for hen-day egg production, Haugh unit score, and eggshell strength. No significant differences were noted for egg weight, blood or meat spot incidence, or pimpling. No significant differences in egg size distribution were noted among the various treatments.

8:45—9:00 AM

**Role of *Mycoplasma synoviae* with or without *Escherichia coli* Infection in Egg Production
and Peritonitis in Commercial Layers**

**Ziv Raviv, Naola M. Ferguson-Noel, Victoria A. Leiting, Ruth S. Wooten,
and Stanley H. Kleven**

The University of Georgia; Poultry Diagnostic and Research Center;
953 College Station Road; Athens, GA 30602-4875

We studied the possible role of *Mycoplasma synoviae* (MS) infection in laying hens *E. coli* peritonitis (EP), and in production losses. We used four groups of commercial pullets under simulated commercial conditions that were treated as follows: negative control, intratracheal (IT) *E. coli* (E), MS aerosol, and MS aerosol follows by IT E. A typical EP mortality was reproduced by both IT E challenged groups, with significantly higher mortality relative to the negative controls in the MS/E group. The MS/E group demonstrated severe tracheal lesions, and body cavity lesions; suggestive of the pathogenesis contribution of MS in EP.



Session B Monday, July 17, 2006
9:15—9:30 AM

A Comparison of the Efficacy of *Mycoplasma gallisepticum* Vaccines

**Naola Ferguson-Noel, Kalen Cookson, Ziv Raviv, Victoria A. Leiting, Ruth S. Wooten,
and Stanley H. Kleven**

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

In most countries control of *Mycoplasma gallisepticum* (MG) is based on maintaining stock free of infection, but in areas where this is not feasible inactivated and live vaccines have been used. In this study the efficacy of three commercial vaccines were compared - an MG bacterin, a recombinant Pox-vectored MG vaccine and a live F-strain vaccine. The vaccines were evaluated with respect to protection from infection and pathology in the respiratory system and oviduct of layer-type chickens.

9:30—9:45 AM

Delayed Serological Response to *Mycoplasma synoviae*


S. H. Kleven, Naola Ferguson-Noel, Ziv Raviv, and Victoria A. Leiting

University of Georgia
Poultry Diagnostic and Research Center
952 College Station Road
Athens, GA 30602-4875

Chickens were challenged with ten-fold dilutions of one of two *M. synoviae* strains with doses as low as 76 and 24 ccu/bird. Low challenge doses readily infected the birds, and infection spread rapidly to contact controls. Surprisingly, unchallenged controls maintained in the same house remained uninfected for the duration of the study. Serological responses in all challenged groups were timely and typical – there was no delayed serological response, although the response in the lowest challenge titer groups was delayed for about a week. Interestingly, HI titers using homologous HA antigens were higher than with heterologous antigens, suggesting antigenic variability.

9:30 - 10:00 AM

BREAK



**Session B Monday, July 17, 2006
10:00—10:15 AM**

Moderator: Hector Cervantes

REED RUMSEY AWARD

Pathogenesis of Infectious Bronchitis Virus in Chickens expressing Different MHC *B* Complex Genotypes

**Kellye S. Joiner, Frederic J. Hoerr, Sandra J. Ewald, Vicky L. van Santen,
James C. Wright, and Haroldo Toro**

Auburn University
166 Greene Hall
Department of Pathobiology
Auburn University, AL 36849

Major histocompatibility complex (MHC) differences in susceptibility to infectious bronchitis virus (IBV) were evaluated in Leghorn chickens expressing B2/B15 and B2/B21 MHC *B* complex genotypes. Ark DPI IBV vaccine was administered at 18 days of age. Birds were challenged with IBV Ark-type field isolate AL/4614/98 at 14 days post vaccination (DPV), and examined daily for clinical signs. IBV antibody in tears was measured on 5 and 10 DPV, and 3, 6, and 9 days post inoculation (DPI). The study was ended on 10 DPI for histopathology and IBV detection and quantification. Results will be compared among MHC *B* complex genotypes.

10:15—10:30 AM


A New Aid in the Prevention of Avian Colibacillosis

Jerry D. Maiers, K. Cookson, J. Tian, and M. Kumar

Fort Dodge Animal Health
12 Elmwood Lane
Asheville, NC 28803

Avian colibacillosis in domestic poultry is caused by the bacteria *Escherichia coli* (*E. coli*). Infection commonly occurs via the respiratory tract, often after primary bacterial or viral infection or as a result of poor husbandry practices. The most severe manifestation of avian colibacillosis is septicemia that is characterized by pericarditis, perihepatitis, peritonitis, and airsacculitis. Chronic forms of colibacillosis in birds raised for meat production often result in increased condemnations at processing.

This new live attenuated *E. coli* vaccine is a genetic mutant of a highly pathogenic strain of *E. coli* isolated from chickens. The advantage of this technology is that the cell surface structures as well as the fimbriae and flagella remain intact to be expressed in the host. The major benefits of this vaccine is its safety, ease of mass administration, and its ability to stimulate a strong cellular and humoral immune response.



Session B Monday, July 17, 2006
10:30—10:45 AM

Possible Transmission of Campylobacter Between Hog and Turkey Production Systems

Donna K. Carver, Sophia Kathariou, Sandra Wright, and Robin Siletzky

North Carolina State University
Campus Box 7608
North Carolina State University
Raleigh, NC 27695

Turkey flocks are often colonized with *Campylobacter* species. While some flocks are colonized with both *C. jejuni* and *C. coli*, others are colonized with *C. jejuni* exclusively. Historically, *C. coli* have been associated with swine. Objectives are to determine to what extent turkeys grown in the absence of swine are colonized with *C. coli* and to determine if *Campylobacter* species in turkeys and swine are epidemiologically linked. Preliminary data suggest that turkeys grown in close proximity to swine have a higher prevalence of *C. coli* compared to those grown in swine-free areas. Swine do not shed *C. jejuni* and are apparently not colonized with *C. jejuni*. Molecular work to further delineate the epidemiologic link between *C. coli* isolates from turkeys and swine suggest that isolates are not shared between swine and turkeys.

10:45—11:00 AM

Crop Immune Response Post-*Salmonella* Enteritidis Challenge in Eight Commercial Layer Breed-Strains and Specific-Pathogen-Free (SPF) White Leghorns

Lara E. Vaughn, Peter S. Holt, Randle W. Moore, and Richard K. Gast

USDA-ARS, Egg Safety & Quality Research Unit (ESQRU)
1351 White Oak Drive
Athens, Georgia 30606

The pre- and post-SE-challenge mucosal immune responses within the crops of eight commercial layer-breeds (5 white-egg & 3 brown-egg layer strains) and SPF-White Leghorn chickens were evaluated. The hen groups were orally challenged with $\sim 10^8$ cfu/ml *Salmonella* Enteritidis PT13a. Fecal and crop samples were cultured weekly to monitor progression of SE infection. Crop SE-LPS IgA responses were determined via ELISA at weekly intervals from day 0-day 25 post-infection (pi). H&E stained tissue slides from day 34 pi crop sections were assessed for lymphoid tissue via light microscopy & assigned a semi-quantitative numerical score based upon prevalence of lymphoid aggregates & degree of cellularity. Results revealed: crop & fecal SE positive samples at day 6 pi with a decline in percentage positive at day 25 pi; a marked increase in crop SE-LPS specific IgA between day 0 (pre-challenge) to day 11 pi; and well-defined score 3-5 lymphoid tissue aggregates in day 34 pi crop sections.



Session B Monday, July 17, 2006
11:00—11:15 AM

Tracking Salmonella thru the Poultry Production Pyramid

John J. Maurer, Dana Cole, Charles Hofacre, and Michael P. Doyle
Poultry Diagnostic and Research Center,
The University of Georgia
953 College Station Rd.
Athens, GA 30602

The federal government has invested significant resources in diagnosis and prevention of foodborne disease; food inspection and compliance with procedures intended to reduce pathogen levels in food. However these surveillance networks do not address preharvest food safety and therefore a major approach to *Salmonella* control is neglected. By collaborating with the poultry industry, we propose to develop an epidemiological database from prospective and retrospective studies of primary breeder and broiler-breeder poultry flocks in order to *quantitate the risk factors that contribute to the transmission of Salmonella to meat birds*. PFGE was used to determine *Salmonella* strain prevalence and origin.

11:15—11:30 AM

Why has *Salmonella* Serovar Kentucky become Widespread in Poultry?

Dana Cole, Katherine Zamperini, Charles Hofacre, and John J. Maurer
Georgia Division of Public Health; Notifiable Disease Section;
2 Peachtree St. NW, Rm. 14-225; Atlanta, GA 30303

In the United States, we have observed significant decreases in number of foodborne illnesses associated with *Campylobacter* and *E. coli* O157:H7, while only modest decline for *Salmonella*. Simultaneously, there has been a reported increase in *Salmonella* prevalence on poultry carcasses and most notably, *S. serovar Kentucky*. Can this rise in the prevalence of *S. ser Kentucky* be attributed to the dissemination of a *Salmonella* clone uniquely adapted to poultry? By PFGE, we identified four *S. ser Kentucky* strains. However only one was the prevalent strain and this strain was unrelated to *S. ser Kentucky* isolated from reported human cases.

**AAAP AWARDS LUNCHEON
SHERATION WAIKIKI
LANA'I BALLROOM**

11:30 – 2:00 PM



**Session B Monday, July 17, 2006
2:00—2:15 PM**

Moderator: Eric Jensen

Salmonella Bio-Mapping for Salmonella through the Poultry Processing Plant

Bruce Stewart-Brown, Will Morris, and Clay Silas

Perdue Farms, Inc.
PO Box 1537
Salisbury, MD 21802

Poultry are generally presented to the processing plant with some level of Salmonella. The processing plant process has as many as 12 distinct areas where Salmonella can go up, stay the same, or go down. Most of the practices and process management guidelines associated with these distinct areas have been historically developed around controlling temperature (preventing growth of microorganisms) and/or organoleptic properties and meat/carcass quality. In order to define best practices for Salmonella reduction, mapping (level of Salmonella before and after each area) has become necessary and useful. To define best practices for each area, we measured Salmonella thru the area, defined any areas performing in a superior fashion, determined the practices that allow and encourage the most reduction in Salmonella. With these practices in hand, we intend to move from plant to plant implementing best practices for those areas that can be better controlled.

2:15—2:30 PM

Development and evaluation of a recombinant strain based on the SEF14 fimbrial operon against a *Salmonella* Enteritidis challenge in chickens

V. C. Lopes, B. T. Velayudhan, D. N. Foster, D. A. Halvorson, and K. V. Nagaraja.

University of Minnesota
3217 Dakota Ave. South
Saint Louis Park, MN 55416

Several attempts have been made to control *S. enteritidis* infection in poultry. A significant difficulty is the partial protection elicited, which may allow for shedding of the pathogen to horizontal and vertical transmission. In this study, we cloned *sef*ABCD operon of *S. enteritidis* under an anaerobically-inducible promoter. The lethal balance system was exploited to generate a strain that stably express the SEF14 fimbriae to overcome plasmid instability. Our objectives were to develop the *sef*ABCD recombinant strain, to evaluate immune responses and to assess protection in treated and non-treated chickens against a challenge with *S. enteritidis*. Results will be presented.



Session B Monday, July 17, 2006
2:30—2:45 PM

Developing a predictable gangrenous dermatitis model for broilers: lessons learned

**Stephen R. Collett, Young-Jae Cho, John R. Glisson, Charles L. Hofacre,
and Margie D. Lee**

Poultry Diagnostic and Research Center
Department of Population Health; University of Georgia
953 College Station Rd.; Athens, Georgia 30602-4875

In a 2004 survey of US poultry veterinarians, 70% of respondents ranked gangrenous dermatitis (GD) as one of the top three “most serious current disease entities”. Difficulty in consistently reproducing GD under experimental conditions has hindered research on this disease.

Initial screening studies and 6 consecutive experiments with 150 birds in each indicated that the response varied according to; the challenge strain (field isolates vs. reference strains of *Clostridium perfringens* type A or *Clostridium septicum*); culture technique (media and time); form or growth stage (log vs. stationary phase); dose ($0.5 \text{ ml } 1 \times 10^8$ - 1×10^6 cfu/ml); method of administration (subcutaneous injection vs. full-thickness skin scratch contamination) and the use of washed cells alone, toxin alone or cells and toxin combined (broth).

The challenge method of choice is the administration of 0.5 ml of an overnight (16-18 hours) brain heart infusion broth culture of ATCC strain 13124 *Clostridium perfringens* type A containing $\sim 1 \times 10^8$ vegetative cells in the logarithmic growth phase per millilitre, by subcutaneous injection in the breast.

This challenge model consistently induces gangrenous dermatitis lesions in 100% of the challenged birds.

2:45—3:00 PM

The Association of Enteric Infections With *Clostridium perfringens* In Broiler Field Cases Of Gangrenous Dermatitis and the Successful Reproduction of Gangrenous Dermatitis Using Oral or Intravenous Inoculation With *Clostridium septicum*.

Stephen W. Davis

Colorado Quality Research, Inc.
400 E. County Road 72; Wellington, CO 80549

Culture results from fresh mortality broilers during outbreaks of gangrenous dermatitis consistently found heavy growth of *Clostridium perfringens* from enteric cultures and heavy growth of *Clostridium septicum* from the subcutaneous dermatitis lesions. Studies were conducted to attempt to develop a gangrenous dermatitis challenge model. Comparisons of many inoculation methods including scratches, bruises, subcutaneous and/or intravenous injection, and oral dosing was conducted with mixed results. It was found that intravenous inoculation of *Clostridium septicum* consistently created classic gangrenous dermatitis lesions and mortality. A specific mixture of enteric pathogens and an oral challenge with *Clostridium septicum* was necessary to reproduce gangrenous dermatitis lesions with mortality. Clinical signs, gross lesions and culture results from inoculated birds and adjustments necessary to successfully reproduce typical gangrenous dermatitis lesions and mortality will be presented.



**Session A, Tuesday, July 18, 2006
7:00—7:15 AM**

Moderator: Stewart Ritchie

**Findings from Data-Mining Broiler Companies and
Correlating Unique Parameters – Disease Indices, Vaccine Programs,
Production Practices and Timeline Trends**

Lloyd Keck, John F. Tierce, Ph.D., and Greg Rennier

Avian Performance Standards, Inc
Texas Office
6417 Lago Vista Drive
Fort Worth, Texas 76132

Significant trends within the broiler industry will be presented, based on five years of live production data from 3.3 billion broilers. Areas emphasized will include interactions between broiler performance and health variables and their correlation with variables such as vaccines and vaccination programs for both parent stock and broilers. A series of disease indices will be incorporated to assist in describing variation in broiler health variables including, lameness, enteric disorders, dermatitis and early mortality due to bacterial infections. Relationships between feed additives, farm management and hatchery practices will be examined from an economic basis. Quality assurance control charts will summarize the effectiveness of vaccination programs in terms of the level and uniformity of parental and offspring immunity.

7:15—7:30 AM

Prevalence and Antimicrobial Resistance in Organic and Conventional Broilers

Kyle M. Hapner and Teresa Y. Morishita

Department of Veterinary Preventive Medicine, Avian Disease Investigation Laboratory
The Ohio State University
8625 Libra Road
Columbus, Ohio 43016

Escherichia coli was isolated from organically-raised and conventionally-raised broilers. One-hundred and fifty isolates from each production system were then tested for antimicrobial resistance.

Results indicated that E. coli antimicrobial resistance patterns were significantly different in organic and conventional broilers. For example, 62.67% of the E. coli isolates in conventional broilers were resistant to tetracycline and oxytetracycline, while only 24.67% of the E. coli isolates in organic broilers were resistant to both antibiotics. This study indicated that antibiotic usage may play a role in antimicrobial resistance developing on broiler farms.



**Session A, Tuesday, July 18, 2006
7:30—7:45 AM**

Identification of Heat stress Risk Factors in Ontario Broiler Industry

Babak Sanei, Harry Huffman, and Lloyd Weber

Ontario Ministry of Agriculture, Food and Rural Affairs
OMAFRA, Rm # 2154 Department of Pathobiology, Ontario Veterinary College,
Univ. of Guelph, ON, N1G2W1 Canada

The heat stress survey project was conducted in the summer of 2003. During the data collection phase of the study, information was gathered from 63 broiler production farms. Most of these selected farms were visited and pre-designed questionnaires were filled in through face to face interviews with broiler producers. Necessary measurements were also made to calculate certain ventilation parameters (e.g. size of inlet openings, barn dimensions, number and size of fans, and ...). The objectives of this project were: 1- To investigate the extent of losses due to heat stress in Ontario broiler operations 2- To identify the main risk factors that are more important under current housing and management practices. 3- To provide some recommendations, based on the results of the study, in order to reduce the negative impacts of the heat stress on broiler flocks.

The age pattern of majority of flocks that had experienced heat stress problem (HS+ flocks) suggests that most of these flocks were close to their market age and were in the weight categories of 2.2 Kilogram and higher. The majority of Heat stress flocks (75 %) , experienced an average of less than 6.44 percent increased mortality due to heat stress comparing to other flocks raised on the same farms with no problem (other quota periods) or versus other HS- flocks.

The average Feed Conversion in flocks with heat stress problem was higher than those without the heat stress problem (2.01 versus 1.91)

The results of this study clearly demonstrated that farms with the history of heat stress had poor ventilation efficiency and in some cases very high bird density comparing to those with no heat stress history.

7:45—8:00 AM

Temperature and Oxygen Conditions during the last four days of Incubation in Bone Development of Chickens and Turkeys

**Edgar O. Oviedo, Michael J. Wineland, Vern L. Christensen, Debbie T. Ort,
Michael K. Mann, and Sarah L. Funderburk**

North Carolina State University
Department of Poultry Science, Scott Hall O-239
Raleigh, NC. 27695-7608

Skeletal disorders are one of the most prevalent and expensive diseases in broiler and turkey production. Known etiologies are diverse, but it is still considered a metabolic problem. Incubation conditions have an important impact on the hormonal axis that controls bone development. This study evaluated the effects of temperature (36, 37, 38 or 39 °C) and oxygen concentration (17, 19, 21 or 23% oxygen) during the last four days of incubation on bone development in chickens and turkeys. Four experiments were conducted using similar procedures. Samples were taken at pipping and hatch for analyses of hormonal status and bone development.



Session A, Tuesday, July 18, 2006
8:00—8:15 AM

Multistage and Single Stage Incubation Comparison: Field Performance Results with Commercial Yield Breed Broilers in a Paired House Trial

Donna L. Hill , HatchTech
Karen Christensen, OK Foods

Paired eggs from commercial yield breeder flocks will be set in two commercial incubation systems, one multistage and one single stage. The chicks will be placed in two paired commercial broiler houses. Feed and water consumption will be monitored throughout the growout period. At the end of the growout period, the following parameters will be compared: Hatch of fertile, residue breakout results, one week livability, growout livability, mortality patterns, feed conversion, water consumption, and breast meat yield.

8:15—8:30 AM

A Comprehensive Worm Study in Several Broiler Breeder Pullet Flocks

Bret Rings and Tom Yazwinski
Tyson Foods, Inc.
3701 Johnson Rd Springdale AR 72762

Fourteen broiler breeder pullet flocks were evaluated over a 7 month period to determine the worm populations during different ages throughout the pullet growing phase. Entire intestinal tracts from 4 birds/flock were submitted to the parasitology laboratory at the University of Arkansas Animal Science Department every 4 weeks of the flock's age starting at 6 weeks of age. Birds would also be evaluated for worm burdens prior to movement to the hen house. The genus of each worm type would be identified and the numbers of each genus would be recorded. The worms identified included ascarids, *Heterakis gallinarum*, *Ranilletina sp.*, and *Capillaria sp.* It is thought that such a study may indicate specific periods in the growth of replacement breeders when there may be identifiable worm challenges so that an effective treatment plan may be initiated.



Session A, Tuesday, July 18, 2006
8:30—8:45 AM

Evaluation of Vertebral Lesions in Broilers with Gait Abnormalities: A Field Study

Suzanne Young Stamey, John Barnes, Ken Powell
Aviagen, Inc.
5015 Bradford Drive
Huntsville, AL 35805

Lameness and gait abnormalities in broiler chickens > 35 days can be a significant cause of economic loss and are a welfare concern. Often, these gait abnormalities cannot be explained by lesions in the extremities; therefore causes are not well documented. The focus of this study was to determine the frequency of different types of vertebral lesions resulting in gait abnormalities in broilers 5wk through processing age. Multiple lesions were observed and preliminary results will be discussed during this presentation.

8:45—9:00 AM

Cycling Patterns of *Eimeria spp.* and its Correlation with Microscopic Lesions, Shedding of Fecal Oocysts, and Johnson and Reid Scores

Miguel Ruano and Douglas Marvil
Perdue Farms Incorporated
P. O. Box 1537
Salisbury, MD 21802-1537

Cycling patterns of *E. acervulina* and *E. maxima* and its impact on gut health condition is being evaluated by daily histological changes, shedding of fecal oocysts, and development of gross lesions according to the Johnson and Reid Scoring Method. Differences from low and high oocyst challenge dose in non-vaccinated commercial chicks will be discussed as well the interference between the two species after simultaneous challenge.



Session A, Tuesday, July 18, 2006
9:00—9:15 AM

Field Problems with *Eimeria maxima*: Effects of *Eimeria acervulina* on Concurrent *E. maxima* Infections

Greg F. Mathis, Southern Poultry Research, Inc.

The three most commonly occurring species of *Eimeria* infecting chickens are *Eimeria acervulina*, *E. tenella*, and *E. maxima*. Even though *E. maxima* is very immunogenic, lesions are often observed in the field late in a growout. A survey of 50 coccidial field isolates showed that 36 were predominately *E. acervulina*, 4 *E. maxima*, and 10 *E. tenella*. All of the *E. maxima* isolates came from farms where the broilers were over 28 days old. Most of the *E. acervulina* isolates were from broilers that were 18 to 28 days old. The daily oocyst shedding pattern for a commercial coccidial vaccine was examined in floorpen birds. Birds vaccinated for coccidiosis at the hatchery were placed into pens on new pine shaving. The shedding of *E. acervulina* type oocysts peaked around 18 days. A small peak of *E. maxima* was observed around 28 days. A battery cage study was conducted to examine whether *E. acervulina* could be interfering with *E. maxima* development. Birds were challenged at 14 days of age with *E. acervulina* and/ or *E. maxima*. The oocyst per bird challenge levels were none (Trt. 1), *E. acervulina* 100,000 (Trt. 2), *E. acervulina* 100,000 plus *E. maxima* 5,000 (Trt. 3), *E. acervulina* 50,000 plus *E. maxima* 5,000 (Trt. 4), *E. acervulina* 25,000 plus *E. maxima* 5,000 (Trt. 5), and *E. maxima* 5,000 (Trt. 6). Each treatment consisted of 3 replications in a complete randomized block design. *E. maxima* alone caused 21 % weight reduction and 2.75 lesion score. The 100,000 and 50,000 *E. acervulina* oocyst level reduced *E. maxima* lesions to 1.33. The 25,000 *E. acervulina* oocyst level only slightly reduced *E. maxima* lesions to 2.25. The *E. maxima* did not interfere with any of the *E. acervulina* infections. This study suggests that *E. acervulina* interferes with colonization or development of *E. maxima*. As birds become more immune to *E. acervulina* then *E. maxima* has more of an opportunity to develop.

9:15—9:30 AM

Uniformity of infection and growout performance following *in ovo* vaccination with the coccidiosis vaccine Inovocox™

V.W. Doelling, R.P. Poston and A. Martin
Embrex, Inc.; 1040 Swabia Court
P.O. Box 13989; Research Triangle Park, NC 27709-3989

Inovocox™, a live coccidiosis vaccine consisting of four strains of coccidia including *Eimeria acervulina*, *E. tenella*, and two strains of *E. maxima*, is under development for the prevention of coccidiosis in broilers. A series of experiments were conducted to ensure consistent and uniform administration of the vaccine through the Inovoject® system. Additionally, uniformity of infection among individual chicks was monitored to ensure consistent vaccination. Floor pen trials were then conducted to assess the performance of birds on both clean and built up litter when compared to salinomycin. Inovocox was found to be consistently delivered, uniformly infective and birds vaccinated with Inovocox demonstrated equivalent performance to salinomycin.



**Session A, Tuesday, July 18, 2006
10:00—10:30 PM**

Moderator: Jagdev Sharma

LASHER HISTORY LECTURE

**Avian Diseases: The Creation and Evolution of P. Philip Levines's
Enduring Gift**

**Bruce Calnek
Cornell University
Unit of Avian Medicine, Dept. of Micro and Immunology
College of Veterinary Medicine
Ithaca, NY 14853**

10:30 – 12:00

**AAAP BUSINESS MEETING
ROOM 316A**

12:00 – 1:00 PM

LUNCH



Session A, Tuesday, July 18, 2006
1:00—1:15 PM

Moderator: Greg Mathis

A Field Report on Salinomycin Toxicity in Broiler Breeder Chickens

Charles Corsiglia, Bruce Charlton, Portia Cortez, and H. L. Shivaprasad
Foster Farms
14519 Collier Road
Delhi, CA 95315

This field report will describe a case involving a 34 week old Ross x Ross broiler breeder flock that experienced an acute episode of recumbency and mortality after a single feed delivery in one of nine houses on a breeder farm. A description of the field investigation, diagnostics, pathology, and reproduction of the disease using contaminated feed will be presented.

1:15—1:30 PM

Construction and evaluation of recombinant *Salmonella* vaccines expressing *Eimeria acervulina* sporozoite and merozoite antigens

Vjollca Konjufca*, Soo-Young Wanda and Roy Curtiss III.
Biodesign Institute, Arizona State University

Coccidiosis is a ubiquitous disease in poultry and is caused by several distinct species of protozoan parasite *Eimeria* sp. Typical symptoms of this disease include intestinal lesions, poor growth and feed utilization, morbidity and mortality. Efforts to produce an effective vaccine against coccidiosis have been with limited success and the need for an effective vaccine is still evident. *Eimeria* is an intestinal parasite, thus a vaccine capable of inducing both mucosal and systemic immune responses would be most effective in protecting against this parasite. Our approach utilizes bacterial Type Three Secretion System (TTSS) to deliver an antigen directly into the cell cytoplasm of the immunized host and into the MHC I antigen processing pathway for induction of CMI, and antigen-specific CTL responses in particular. To accomplish this goal, *Eimeria* genes encoding antigens EASZ240 and EAMZ250 were fused to *Salmonella* effector protein gene *sptP* in the parental pYA3653 vector, yielding pYA3657. *SptP* effector protein is secreted by TTSS of *Salmonella* and translocated into the cytosol of immunized host cells. Host-strain chromosomal copy of the *sptP* gene was deleted and replaced by a reporter gene *xylE*. Newly constructed vector pYA3657 was introduced into host strain χ 8879 (Δ *phoP233* Δ *sptP1033::xylE* Δ *asdA16*). This strain is an attenuated derivative of highly virulent UK-1 strain. In vitro experiments show that model antigen EASZ240 is secreted into the culture medium by TTSS and it is delivered into the cytoplasm of Int-407 cells by TTSS. Oral immunization of one-week-old chickens with recombinant *Salmonella* expressing a merozoite antigen resulted in significantly higher body weight gain and lower intestinal lesion scores.



**Session A, Tuesday, July 18, 2006
1:30—1:45 PM**

Application of polymerase chain reaction (PCR) and drug-sensitivity testing to compare species composition and anti-coccidial drug resistance in *Eimeria* isolated from vaccine- and coccidiostat-utilizing poultry operations.

M.C. Jenkins, S. Klopp, G. Wilkins, K. Miska
Animal Parasitic Diseases Laboratory, ARS, USDA;
APDL, ARS, USDA, BARC-EAST, Building 1040
Beltsville, MD 20705; Townsends Poultry, Inc.

The purpose of this study was to compare drug resistance and species composition of *Eimeria* isolated from two types of poultry operations that differed in the means of controlling avian coccidiosis. The number of species present in vaccine-control facilities was greater than in drug-control operations, with all 6 species present in the former, and 3-4 species present in the latter. *Eimeria* propagated from drug-utilizing operations displayed greater anti-coccidial resistance compared to *Eimeria* recovered from farms using a live oocyst vaccine. These results suggest that conventional drug treatment leads to lower *Eimeria* species diversity coincident with slightly greater drug resistance.

1:45—2:00 PM

Characterization of Viral Agents associated with runting and stunting syndrome in young broilers

**Holly S. Sellers, Erich G. Linneman, Steven Bell, Susan M. Williams,
and Guillermo Zavala**

The University of Georgia, Poultry Diagnostic Research Center
953 College Station Road

Athens, GA 30602

Novel avian reoviruses and astroviruses have been isolated from young broilers exhibiting a runting and stunting syndrome (RSS). *In vivo* challenge studies in day of age broilers with several reovirus isolates resulted in significant body weight depression, however did not reproduce the disease. Molecular analyses of the sigma C protein reveal a low similarity to current US vaccine isolates indicating that they may be antigenically different. Chicken astroviruses were detected by RT-PCR in the intestines of affected birds. Subsequent sequence analysis of the capsid protein reveals a high degree of sequence similarity to previously described avian nephritis virus. *In vivo* studies of both viruses continue to determine their role in what appears to be a multifactorial disease.



**Session A, Tuesday, July 18, 2006
2:00—2:15 PM**

**The Pathogenesis of Agents Associated with Runting-Stunting Syndrome of Broilers in SPF
Turkeys**

Erica Spackman, Mary J. Pantin-Jackwood, and J. Michael Day
Southeast Poultry Research Lab, USDA, ARS
934 College Station Rd.
Athens GA, 30605

Runting-stunting syndrome (RSS) of broilers is clinically similar to poult enteritis complex. The causes of these disease conditions are poorly understood. In order to determine whether RSS agents could cause disease in turkey poults, specific pathogen free poults were inoculated with untreated or chloroform treated intestinal contents collected from broilers with RSS. Initial characterization of the inoculum revealed that it contained both an avian astrovirus and an avian rotavirus. Both viruses replicated in the poults and clinical signs, including decreased weight gain and gross lesions consistent with RSS, were produced.

2:15—2:30 PM

Molecular characterization of enteric viruses circulating in the United States

Mary J. Pantin-Jackwood, Erica Spackman, and James J. Day
Southeast Poultry Research Laboratory. USDA/ARS
934 College Station Road. Athens, Georgia 30605

This study investigated the genetic diversity of enteric viruses circulating in poultry. Intestinal samples collected from numerous commercial turkey and broiler flocks from different regions of the United States during 2005, were examined for the presence of astrovirus, rotavirus, reovirus and coronavirus by RT-PCR, and for adenoviruses by PCR. Viruses, predominantly astrovirus and rotavirus, were found in samples collected from flocks with enteritis and stunting as well as from healthy flocks. Comparison of these viruses was performed by phylogenetic analysis. Four clearly different astroviruses are circulating in poultry: Turkey Astrovirus 1 (TAsTV-1), Turkey Astrovirus 2 (TAsTV-2), Avian Nephritis Virus (ANV) and a novel Chicken Astrovirus (CAstV). Rotaviruses were widely distributed in chickens and turkeys.



**Session A, Tuesday, July 18, 2006
2:30—2:45 PM**

Comparison of Enteric Virus RT-PCR Results with Production Data from Commercial Turkey Hens

David V. Rives and Mary Pantin-Jackwood
Prestage Farms, Inc.
PO Box 438
Clinton, NC 28328

A survey of eight commercial turkey hen flocks was conducted from March through June of 2005 to determine the presence and persistence of enteric viruses. Flocks were selected based on farm location and the farms' previous performance. Intestinal samples were collected from poults prior to placement and every two weeks thereafter. Samples were examined by RT-PCR for the presence of astrovirus, coronavirus, reovirus, and rotavirus. Results of these tests will be compared to production data from the survey flocks including five-week body weights, processing weights, feed conversion, and daily gain. Clinical observations and parasitology results will also be presented.

2:45—3:00 PM

Experimental Reproduction of Transmissible Viral Proventriculitis by Inoculation Of Chickens with a Novel Adenovirus-like Virus (Isolate R11/3)

James S. Guy, John Barnes, Lynda Smith, and Maria Evans
North Carolina State University, College of Veterinary Medicine,
4700 Hillsborough Street, Raleigh, NC 27606

Transmissible viral proventriculitis (TVP) was experimentally reproduced in two-week-old SPF chickens and commercial broiler chickens by intraocular inoculation of a novel adenovirus-like virus (AdLV), isolate R11/3. AdLV (R11/3) previously was isolated from proventriculi of TVP-affected broiler chickens. No clinical signs and no weight gain depression were observed in inoculated chickens; however, gross and microscopic lesions consistent with TVP were identified. Microscopic lesions in proventriculi consisted of degeneration and necrosis of glandular epithelium, ductal epithelial hyperplasia, and interstitial lymphocytic inflammation. AdLV (R11/3) was recovered by virus isolation from proventriculi of inoculated chickens. No virus or lesions were detected in sham-inoculated chickens.

3:00 PM

ADJOURN



Session B Tuesday, July 18, 2006
7:00—7:15 AM

Moderator: Guillermo Zavala

Evaluation of Roxarsone and/or Bacitracin Methylene Disalicylate on Broiler Performance Using Necrotic Enteritis and Salmonella Challenge Models

Sharon Heins Miller, Mark W. LaVorgna, Stephen W. Davis, and Steven Clark
Alpharma, Inc. – Allied Industry
3595 Hidden Lake Drive
Cumming, GA 30041

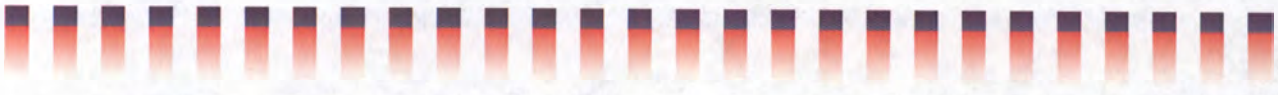
A study was conducted to evaluate the effects of roxarsone and/or bacitracin methylene disalicylate (BMD) when fed *ad libitum* on growth, feed conversion, mortality, necrotic enteritis lesion scores and presence and enumeration of salmonella in cecal content in broilers artificially challenged with *Clostridium perfringens* and a mixture of *Salmonella* serotypes. This is the second study to evaluate these parameters and feed additives to follow up on positive results achieved during the first study.

7:15—7:30 AM

Passive immunity to Clostridium Perfringens Type A: Practical Efficacy Against Necrotic Enteritis Under US and Canadian Broiler Management Systems

Linnea J. Newman, DVM, Charles Broussard, and Richard Phillips
Schering-Plough Animal Health
17 Pine Street
North Creek, NY 12853

An inactivated *Clostridium perfringens*, Type A vaccine was developed for application to broiler breeder replacements to induce high levels of passive immunity against *C. perfringens* in the broiler progeny. Large-scale field trials were implemented at US and Canadian broiler integrators to evaluate the efficacy of passive immunity to *C. perfringens* in lieu of growth promotant antibiotics under real-world conditions. Efficacy of the passive immunity was measured by necrotic enteritis-induced mortality and flock performance of test progeny compared to progeny from breeders that did not receive the vaccine. Field studies included flocks using conventional ionophore anticoccidials, coccidiosis vaccines, corn-soy rations and wheat rations.



Session B Tuesday, July 18, 2006
7:30—7:45 AM

**Evaluation of Elanco Tylan Premix for the Prevention of Necrotic Enteritis
in Broiler Chickens**

Marina L. Brash, Randy N. Bagg, Paul C. Dick, Gordon H. Vessie, and Jeff B. Wilson
Elanco Animal Health
150 Research Lane, Suite 120
Guelph, Ontario, Canada N1G 4T2

Tylan premix in feed was evaluated for its effectiveness in the prevention of necrotic enteritis. Groups A) non-challenged, non-medicated, B) challenged, non-medicated, C) challenged, 11 ppm Tylan, D) challenged, 22 ppm Tylan, E) challenged, 33 ppm Tylan were studied.

NE mortality in Treatment B was significantly ($P < 0.01$) higher (30.4 %) than treatment groups C, D and E (6.2%, 1.0%, and 0.6% respectively). Treatments D and E significantly ($P < 0.01$) improved control of NE mortality over Treatment C.


Tylan premix administered in the feed at the dose of 22 ppm is effective in the prevention of clinical outbreaks NE in broiler chickens.

7:45—8:00 AM

**Organic Acids Effective to Ameliorate the Negative Impact on Broiler Performance
due to Necrotic Enteritis**

**Marco A. Quiroz, D.V.M., Charles L. Hofacre, Grez F. Mathis, Julia Dibner,
and Chris Knight**
Novus International, Inc.
530 Maryville Centre Dr.
St. Louis, MO 633141

This presentation will summarize two experiments conducted to evaluate the effects of organic acids on *Clostridium perfringens* colonization and intestinal microbial populations using a necrotic enteritis model in broiler chickens. It will show that supplementing the drinking water with a blend of organic acids (Activate® US WD Max) was as effective as feeding bacitracin methylene disalicylate (50g/Ton) on controlling necrotic enteritis while maintaining feed conversion.



**Session B Tuesday, July 18, 2006
8:00—8:15 AM**

Molecular basis for Antimicrobial Growth Promoter Effects in Poultry

Margie D. Lee, Jingrang Lu, and Charles Hofacre
The University of Georgia; Dept. of Population Health
Poultry Diagnostic and Research Center
953 College Station Rd., Athens GA 30602

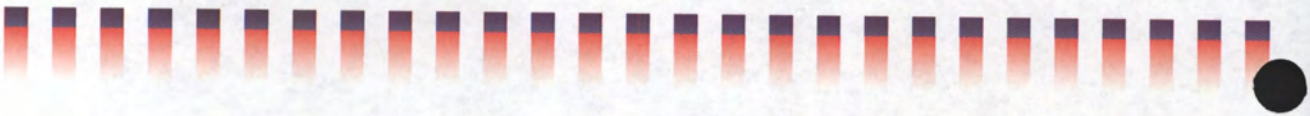
For more than 50 years, the performance-enhancing characteristics of antimicrobial feed additives, also known as antimicrobial growth promoters, (AGPs) have been utilized by the animal production industry. How these antimicrobials enhance performance is unknown, but they may change the population balance of the intestinal microbiota. This microbiota contributes to the maintenance of intestinal health and has a profound effect on the economy of animal production. Studies have shown that the cultivable species represent only about 10% of the microbial diversity in the intestine. Recently, molecular techniques have been developed to characterize complex microbial ecosystems. We used these techniques to reveal the bacterial community structure and the distribution of bacterial species that occurred in response to AGPs and different diet formulations fed to broiler chickens. A better understanding of the effects of these bacterial communities on food animal development and feed conversion will enable replacement of AGPs with novel approaches for managing the ecosystem of the intestine.

8:15—8:30 AM

Evaluation of Oral Administration with Live CU Strain *Pasteurella multocida* Vaccine in Broiler Breeder Pullets

Marilynn N. Finklin, Bradley J. Turner, Charles L. Hofacre, and J. R. Glisson
Department of Population Health, University of Georgia, Athens, GA

Broiler breeder pullet vaccination for fowl cholera via wing web route is costly due to labor and can result in stress and injuries to vaccinated birds. Pullets vaccinated with live vaccine by wing web route also risk acquiring the disease contributing to joint problems, morbidity, and death. Therefore in 2004, in an effort to investigate alternative routes of vaccination, Bruzual et. al, evaluated the use of a commercial *Pasteurella multocida* PM-1 vaccine administered by the drinking water in broiler breeder pullets versus administration by wing web. It was concluded that there was no protection by administration of a 6X dose of PM-1 orally in pullets whereas those given the vaccine by the w.w. route were protected. In this investigation, oral, eye drop, and wing web vaccination will be evaluated using the less attenuated CU (Clemson University) strain of *Pasteurella multocida*. Both post-vaccination mortality and post-challenge mortality after inoculation with a highly virulent serotype 1 *Pasteurella multocida* will be compared between treatment groups vaccinated orally, by eye drop, and via wing web with the CU strain and a group vaccinated with a commercial PM-1 strain via wing web application.



Session B Tuesday, July 18, 2006
8:30—8:45 AM

Performance Differences in Sister Flocks: Is Chicken Anemia Virus (CAV) the Culprit?

Franz Sommer, Carol J. Cardona, and Bruce W. Charlton
CAHFS – UC Davis, Turlock Branch Laboratory
2500 Mira Flores Drive
Turlock, CA, 95380

Sister flocks placed in several houses on a commercial broiler farm were found with differences in mortality and disease rates.

We performed regular necropsies on 10 flocks in two consecutive runs (ages 36 and 44 days, 2 affected, 2 controls; and 19, 28 and 52 days, 3 affected, 3 controls) and did additional testing for the presence of CAV with PCR.

To compare possible differences in the CAV patterns in these flocks, selected strains were sequenced and aligned. Additionally, these strains were compared to strains found in previously tested flocks from other ranches within the area.

The results of these examinations will be presented.


8:45—9:00 AM

The Pivotal Role of Feathers in Chicken Infectious Anemia (CIAV) Horizontal Spread

Irit Davidson, Irena Shkoda, Emmanuel Loebb, and Karel A. Schat
Division of Avian Diseases, Kimron Veterinary Institute;
P.O. Box 12; Bet Dagan, Israel 50250

The increased concern in CIAV derives from the virus-induced immunosuppression, which affects poultry productivity. CIAV spreads vertically and horizontally. Although previously only the oral-faecal route was considered to be important for horizontal transmission, we now suggest feathers as an alternate infection source.

We examined the CIAV presence in feather tips from inoculated and contact chicks, before or after hatch, by histology, immunohistochemistry, and PCR. These studies indicated that feathers are an important source for CIAV transmission. We show for the first time substantial impact of feathers, as a means for CIAV monitoring without sacrificing the birds.



Session B Tuesday, July 18, 2006
9:00—9:15 AM

Chicken Anemia Virus (CAV) Serology Profile of Breeders and Their Progeny

Beatriz Cardoso and Lindolfo Rocha
Lohmann Animal Health International; R. Simpatia 288 ap 21;
Sao Paulo – SP – 05436-020; Brazil

In order to control chicken anemia virus in young birds, is common that breeder producers use serology profile to check the status of the breeders. Some uses this tool also to decide whereas it is appropriate to vaccinate or not those breeders that show low or high titer prior the age of vaccination. The objective here was to check the serological profile of breeders and it's progeny at day of age and 3 days of age. We found that the variation exists among the type of CAV control used on the breeders more than their age.

9:15—9:30 AM

Vaccination of Broilers Using a Live Chicken Infectious Anemia Virus Vaccine

Brett A. Hopkins
Biomune Company
8906 Rosehill Road
Lenexa, KS 66215

Safety, efficacy, serologic, and performance data will be presented from commercial and isolator reared broilers that were vaccinated by the drinking water against chicken infectious anemia virus using a commercially available and USDA licensed live chicken anemia virus vaccine.

9:30 – 10:00 AM

BREAK



**Session B Tuesday, July 18, 2006
10:00—10:30 AM**

LASHER HISTORY LECTURE

ROOM 316A

**Avian Diseases: The Creation and Evolution of P. Philip Levine's
Enduring Gift**

Bruce W. Calnek
Unit of Avian Medicine
Dept. of Microbiology and Immunology
Cornell University
Ithaca NY 14853

10:30—12:00 PM

AAAP BUSINESS MEETING

ROOM 316A

12:00—1:00 PM

LUNCH



**Session B Tuesday, July 18, 2006
1:00—1:15 PM**

Moderator: Pedro Villegas

RICHARD RIMLER PAPER

Detection, Subtyping And Characterization Of Avian Influenza Via cDNA Microarray

Michele N. Maughan, Travis W. Bliss, David L. Suarez, and Calvin L. Keeler, Jr.

University of Delaware
044 Townsend Hall
University of Delaware
Newark, DE 19717

cDNA derived from the hemagglutinin, neuraminidase, and matrix genes from various AI isolates were has been used to construct a glass slide microarray. A panel of unknown AIV isolates was used to evaluate the array. The array was able to detect AI using both vRNA and tracheal swabs as starting material. The AI array can identify type A influenza, differentiate between H1, H2, H3, H5, H7, and H9 hemagglutinin subtypes, N1, N2, and N3 neuraminidase subtypes and, in the case of H5 and H7 isolates, determine the phylogenetic clade from which the strain of AI was isolated.

1:15—1:30 PM

Evidence of immune system dysfunction in poults infected with turkey-origin reoviruses

J. Michael Day, Mary J. Pantin-Jackwood, and Erica Spackman

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

Recently, several avian orthoreoviruses have been isolated from commercial turkeys affected by Poult Enteritis Complex (PEC), a disease syndrome with uncertain etiology. Molecular characterization has shown that these turkey-origin reoviruses (TRVs) are unique among avian orthoreoviruses. The effect of a particularly pathogenic TRV strain on poult cell-mediated and humoral immunity was evaluated using a phytohemagglutinin toe-web inflammation assay and by measuring antibody response to Newcastle Disease Virus vaccine, respectively. The results of these assays suggest that TRV infection can lead to poult immune-dysfunction and may predispose poults to subsequent infections associated with the PEC disease state.



**Session B Tuesday, July 18, 2006
1:30—1:45 PM**

Investigation on the pathogenicity of avian reoviruses

Hafez M. Hafez, Olivia Gooß, Christine Prusas, and Doerte Lueschow
Institute of Poultry Diseases, Free University of Berlin
Königsweg 63, Germany

Reoviruses have been isolated from chickens showing a wide variety of clinical signs and are incriminated as a possible cause of malabsorption syndrome (MAS). Since the isolation of so-called enteric reovirus strains (ERS) from broilers in Poland, no information's are available on the pathogenicity of such strains from other countries. The present investigation describes the pathogenicity of isolates from broiler in Germany in one day old SPF-Broiler. All chicks of the group, which had been infected with the ERS-prototype (polish strain) died within 6 days p.i.. No mortality could be observed in other groups. However, one of tested isolate caused reduction in body weight.

1:45—2:00 PM

Evaluation of field isolates of Infectious Bursal Disease and Chicken Anemia Viruses from broilers with or without Gangrenous Dermatitis in Georgia

Linda B. Purvis, Pedro Villegas, Ivan Alvarado, and Francisco Perozo
The University of Georgia, Dept. of Population Health,
Athens, GA 30602-4875

The objective of this study was to evaluate IBDV and CAV viruses possibly influencing the increase in Gangrenous dermatitis (GD) cases in broiler farms in Georgia over the last several years. Antibody levels against IBDV and CAV were monitored at day of age and 28-30 days of age on farms with and without GD problems. Antibodies at day of age were similar between infected and non-infected farms, however, at 28-30 days of age, differences in antibody levels were found between farms. From farms experiencing GD, IBDV and CAV isolates were obtained and compared with isolates from farms not experiencing GD. Field isolates showing significant molecular differences compared to previous isolates or isolates from farms free of GD will be further evaluated in controlled bird studies to study their effect on the humoral immune system.



**Session B Tuesday, July 18, 2006
2:00—2:15 PM**

Field safety and efficacy Study of Bursal Disease- Marek's Disease vaccine, Serotype 3, Live Marek's Disease Vector administrated to one-day- old Commercial Birds or 18 – day – old Embryonated Commercial Eggs.

Rafael J. Fernandez, Clovis Oliveira, and Jeovane Pereira
Merial Avian Global Enterprise
1112 Airport Parkway.

The objective of the present study was to determine the safety and efficacy of the Bursal Disease- Marek's Disease vaccine, Serotype 3, Live Marek's Disease Vector (Vaxxitek-Merial Brazil) in commercial broilers vaccinated In Ovo or SQ under field conditions. The commercial broilers were challenged with a vvIBD. Results will be presented.

2:15—2:30 PM

Influence of maternal antibodies and differences in genetic background on infectious bursal disease pathogenesis

Silke Rautenschlein, Jung Arne, Rebeski Dierk, and Scharr Heike
Clinic for Poultry, University of Veterinary Medicine Hannover, Foundation
Bünteweg 17
30559 Hannover, Germany

Infectious bursal disease (IBD) pathogenesis depends not only on the virulence of the IBD virus (IBDV) strain but also on the genetic background of the infected birds. Furthermore, maternal antibodies may influence the outcome of an IBDV infection as well. Most of the studies regarding IBD have been conducted in specific pathogen free (SPF) birds. In our studies we compared the pathogenesis of an intermediate IBDV strain in SPF birds and Ross-type broilers. Furthermore, the influence of different levels of maternal antibodies on IBD pathogenesis, and on the stability of the IBDV genome were investigated.



**Session B Tuesday, July 18, 2006
2:30—2:45 PM**

Recombinant Avian Adeno-Associated Virus Expressing the VP2 gene of Infectious Bursal Disease Virus: Development of Immune Response in Chickens.

Iván Alvarado, Pedro Villegas, Carlos Estévez and Linda Purvis
University of Georgia, Department of Infectious Diseases

An avian adeno-associated virus (AAAV) coding for the immunogenic VP2 peptide (rAAAV-VP2) of the Edgar strain of IBDV was constructed by simultaneous transfection of the 293K cells with plasmids containing the *rep* and *cap* genes of AAAV, the entire VP2 gene, and a pHelper plasmid. Appropriate assembly of the rAAAV-VP2 virions and positive expression and conformation of the VP2 protein after transfection of 293K cells were detected by electron microscopy and immunohistochemistry, respectively. The recombinant virus was used to vaccinate specific pathogen free chickens (SPF) by *in ovo* and IM inoculation. Circulating antibodies, measured by a commercial ELISA test were detected from 14 to 42 days of age. Antibody titers were more consistently detected in birds inoculated at day of age when compared with chicken embryos inoculated *in ovo* at 18 days of age.

2:45—3:00 PM

**Is there a Perfect Timing for Vaccination against Infectious Bursal Disease?
Experiences from Field Studies in Broilers**

Hermann Block, Karen Meyer-Block, Dierk Rebeski, and Heike Scharr
Clinic for Poultry and Group Practice Meyer-Block
Am Rott 12
49843 Uelsen
Germany

The right strategy for infectious bursal disease (IBD) control and its success rate depends a lot on the IBD-field pressure, maternal IBD antibody levels, and the IBD vaccine strain. The employment of the 'Deventer formula' may help to estimate the best time point for vaccination based on the level of maternal antibodies and the IBD-vaccine strain to be used. We conducted two field studies applying an IBD-vaccine before, at the best time determined by the 'Deventer formula', and after the estimated time. The active humoral immune response, induction of IBD-lesions, and detection of IBD-virus in bursal tissue were determined and correlated.

3:00 PM

ADJOURN



Session A Wednesday, July 19, 2006

7:00—7:15 AM

Moderator: Mark Jackwood

Case Report: LT in Broiler Breeder Flock Vaccinated with LT Vector Vaccine

Benjamin C. Johnson

Gold Kist, Inc.; PO Box 318; Commerce GA 30529

LT was diagnosed by histopathology in a 48-week-old broiler breeder flock vaccinated at 10 weeks with an LT vector vaccine. Vaccination evaluation indicated an average of 96% of the pullets were vaccinated properly. The average farm mortality during the LT break adjusted for normal mortality 1.5%. The egg production declined during the LT break, but returned to normal in 2-3 weeks. The LT vector vaccine when given at a 96% accuracy rate will adequately protect broiler breeders from LT challenge at 48 weeks of age.

7:15—7:30 AM

New Studies using GIS for Understanding Vaccinal Laryngotracheitis Epidemiology

Louise Dufour-Zavala, Allyson Jason, and Chris Semerjian

Georgia Poultry Laboratory; P.O. Box 20; Oakwood, GA 30566

Occasional outbreaks of vaccinal laryngotracheitis in Georgia have been better understood and controlled using GIS technology. Attempts have been made to use spatial analysis techniques to understand the geographic pattern of cases. The results of these studies will be presented.



Session A Wednesday, July 19, 2006

7:30—7:45 AM

**Evaluation of Heating and Down Time Effects on Poultry Environment
Contaminated with VLTV**

Kelli Holloway Jones, Maricarmen Garcia, Marta Jaramillo, and Susan M. Williams
Aviagen, Inc.
5015 Bradford Dr.
Huntsville, AL

Evaluation of effects of different heating and downtime practices on poultry environment contaminated with vaccinal infectious laryngotracheitis virus. Two consecutive trials conducted, one in a research setting, and one in the field. Three to nine week old sentinel SPF birds placed at PDRC in VLTV contaminated floor pens, and on six northeast Georgia VLTV positive farms. Various heating and downtime protocols were evaluated. Sentinel birds were sacrificed seven to ten days post exposure and tracheas and eyelids were collected. Virus isolation, direct fluorescent antibody testing, histopathological examination, and PCR were performed in attempts to isolate VLTV from the sentinel birds.

7:45—8:00 AM

**The Incidence and Clinical Significance of Hemorrhagic Enteritis Virus (HEV)
Infections in Broilers**

John K. Rosenberger, Sandra Cloud, Nannette Olmeda-Miro, and Conrad Pope
AviServe LLC; Delaware Technology Park;
1 Innovation Way, Suite 100; Newark, DE 19711

Because of the increased incidence of gangrenous dermatitis and other unusual and frequently associated diseases such as inclusion body hepatitis and cystic enteritis (runting-stunting syndrome), we have assessed the role of known and putative immunosuppressive agents as potential predisposing or contributing factors. As part of this assessment, the incidence of HEV infections and clinical significance were evaluated. Sever HEV isolates were obtained by direct isolation from breeder pullets and broilers or via sentinels. The isolates have been evaluated for pathogenic potential in chickens and turkeys and their relative importance as immunosuppressive agents determined in chickens.



Session A Wednesday, July 19, 2006

8:00—8:15 AM

Characterization of Inclusion Body Hepatitis Adenovirus Isolates

Sandra S. Cloud, John Rosenberger, Nannette Olmeda-Miro, and Conrad Pope

AviServe LLC
Delaware Technology Park
1 Innovation Way, Suite 100
Newark, DE 19711 USA

In 2005 a pronounced increase in the incidence of inclusion body hepatitis was diagnosed in broilers produced on the Delmarva peninsula. Type I adenoviruses were readily isolated from livers of affected chickens exhibiting pathognomic inclusion bodies. Isolates were characterized using serological, molecular and in vivo techniques. Virus neutralization demonstrated that the majority of the isolates belong to serotype 11 (US10) or 12 (US12). Molecular characterization results using hexon sequence analysis were variable. Progeny challenge studies have shown a high degree of susceptibility of chickens to both serotypes 11 and 12 and documented horizontal transmission to susceptible contacts.

8:15—8:30 AM

Inclusion Body Hepatitis as a Primary Disease in Broilers in Saskatchewan, Canada

Susantha Gomis¹, Robert Goodhope¹, Davor Ojkic², Philip Willson³

¹Dept. of Veterinary Pathology, Western College of Veterinary Medicine, University of Saskatchewan, Saskatoon, SK, Canada S7N 5B4.

²Laboratory Services Division, University of Guelph, P.O.Box 3612, Guelph ON. Canada N1H 6R8.

³Vaccine and Infectious Disease Organization, 120 Veterinary Road, Saskatoon, SK. Canada S7N 5E3.

In recent years, Inclusion Body Hepatitis (IBH) was an emerging disease in Western Canada. Historically, Infectious Bursal Disease (IBD) and Chicken Infectious Anemia (CIA) have been known to suppress the immune system of broilers which leads susceptible broilers to have a secondary disease such as IBH. Recently, it has been reported that virulent IBH viruses are able to cause IBH as a primary disease in broilers without any immunosuppressive agents.

The objective of this study was to identify the association of IBH with major immunosuppressive diseases of broilers like IBD and CIA. Serum samples from seventeen broiler breeder flocks, and their progeny at hatch and at the time of slaughter were collected for IBD and CIA serology. Necropsy of daily mortalities was conducted three times during broiler grow-out phase when birds were two, three and four weeks old. The results of this study demonstrated that there is no significant association of IBH with any of the immunosuppressive diseases that were examined. Immunity against IBD and CIA was good in all broiler flocks and their IBH outbreaks were not correlated with IBD and CIA titers. Four serotypes of IBH were isolated in birds infected with IBH (FAdV-7, FAdV-11, FAdV-8a, and FAdV-8b). Serotypes FAd-7 and FAd-11 were isolated in an outbreak of IBH in a broiler breeder flock during the study period. Daily mortality and total mortality were high in broiler flocks with IBH outbreaks. The data support the observation that IBH in broilers in Western Canada is a primary disease with no known association with immunosuppression.



Session A Wednesday, July 19, 2006

8:30—8:45 AM

Protection of Broiler Breeder against Inclusion Body Hepatitis by Vaccination of Grand Parent Flocks with a Bivalent Inactivated Fowl Adenovirus Vaccine

Pedro Villegas, Ivan Alvarado, Eric Jensen, and Gregorio Rosales

University of Georgia, Department of Population Health

953 College Station Road

Athens, GA 30602

An avian adenovirus, characterized as serotype 9 (European classification), was isolated from broiler breeder progenies showing clinical signs and lesions compatible with inclusion body hepatitis. When inoculated in SPF broilers at one and 7 days of age, this isolate induced 100% and 40% mortality, respectively. The level of maternal protection against this isolate in broiler breeders chicks from grand parents vaccinated with an inactivated vaccine was evaluated by challenge at 1 and 7 days of age with serotypes 8, 9 and 11. Significant protection was observed after challenge, suggesting the efficacy of the inactivated bivalent adenovirus vaccine to provide protection against serotype 9.

8:45—9:00AM

Pathogenicity and cross protection studies of IBV field isolates of Ontario, Canada

Helena Grgic, Bruce D. Hunter, Peter Hunton, and Eva Nagy

Department of Pathobiology, Ontario Veterinary College

University of Guelph

Guelph, Ontario, N1G 2W1, Canada

Avian infectious bronchitis (IB) caused by IB virus (IBV) is an economically important viral disease of poultry resulting in reduced performance, reduced egg quality and quantity, significantly increased susceptibility to infection with other agents and costly vaccination programs. To assess the IB situation and incidence of IBV in layers in Ontario (Canada), on an industry wide basis sentinel birds were employed as a biological vehicle. IB viruses isolated from the sentinel birds were not vaccine related as shown by RFLP and sequence analysis of the S1 region. The antigenic characteristics of the field isolates were established, pathogenicity of some of these isolates was determined and protection provided by a commonly applied vaccine against these field isolates was evaluated.



Session A Wednesday, July 19, 2006

9:00—9:15 AM

Experimental TCV and REV Co-infection in Turkeys

Bradley J. Turner

Co authors: G. Zavala, T. Barbosa, S. Cheng, M. Jackwood, D. Hilt
Poultry Diagnostic and Research Center,

The University of Georgia, 953 College Station Road, Athens, GA 30602-4875

The objective of this experiment was to co- infect SPF turkeys with turkey coronavirus and reticuloendotheliosis virus obtained from field cases to determine if there is a potential interaction between the two. Body weights of the experimental groups were compared to controls to determine weight reduction after infection. The digestive tract and immune system tissues were examined microscopically for lesions. RT- PCR was performed on intestinal contents of individual turkeys to detect the presence of TCV RNA and to assess virus persistence in the gut. Virus isolation was performed from plasma to demonstrate viremia induced by REV.

9:15—9:30 AM

Effects of Experimental Infection with CAV and/or IBDV on IBV Infection and Immune Response

Vicky L. van Santen, Haroldo Toro, Kellye S. Joiner, and Frederic J. Hoerr

Auburn University

Department of Pathobiology

264 Greene Hall

Auburn University, AL 36849-5519

CAV and IBDV are endemic in commercial chickens. Concurrent infections with these immunosuppressive viruses and IBV occur frequently in the field. To investigate the effects of these associations, SPF chicks were inoculated with CAV and/or IBDV and inoculated eight days later with IBV. Signs, histological changes, local and systemic antibodies, and levels of IBV genomes in tears and trachea were monitored and compared among groups. Development of IBV-specific IgA in tears was delayed in all immunodeficient groups compared to chickens infected with IBV alone. Peak IBV-specific IgA levels in chickens infected with CAV ultimately exceeded those of all other groups.

9 :30 – 10 :00 AM

BREAK



Session A Wednesday, July 19, 2006

10:00 – 10:15 AM

Moderator: Kenton Kreager

In vivo and in vitro characterization of an oncogenic reticuloendotheliosis virus

Taylor Barbosa, Guillermo Zavala, Sunny Cheng, and Pedro Villegas

Poultry Diagnostic Research Center

The University of Georgia

953 College Station Road

Athens, GA. 30605

The transmissibility and oncogenicity of reticuloendotheliosis virus (REV APC-566) isolated from Attwater Prairie Chickens (APC) was studied in Japanese quail. Lymphomas were induced at an early age and were characterized by immunocytochemistry. Molecular characterization of REV APC-566 was accomplished by genome sequencing and phylogenetic analysis, which included additional field isolates of oncogenic REV. Japanese quail proved to be a useful model for REV pathogenesis studies.

10:15 – 10:30 AM

Current Epidemiology of RNA Tumor Viruses

Guillermo Zavala and Sunny Cheng

The University of Georgia; Dept. of Population Health;

953 College Station Road; Athens, GA 30602-4875

The recent epidemiology of RNA tumor viruses in the U.S. is described. Several hundred field isolates of avian leukosis virus (ALV) were obtained from commercial and backyard poultry from the 1990s until the fall of 2005. ALV subgroups A, C and J as well as avian myeloblastosis-associated virus type 1 (MAV-1) were isolated from multiple flocks of commercial meat- and egg-type chickens, backyard Bantam chickens, and contaminated Marek's disease vaccines. The characteristics of some of these isolates are described. Multiple isolates of oncogenic reticuloendotheliosis virus (REV) were isolated from Attwater's prairie chickens and characterized in the laboratory. A recent multi-stage serological survey for antibodies against REV in poultry and wild birds is also presented.



Session A Wednesday, July 19, 2006

10:30 – 10:45 AM

**Efficacy of MDV and the duration of immunity against NDV and LT of the recombinants
rHVT/F and rHVT/LT**

Rudolf G. Hein, Gwen F. Slacum, and Lilian S. Melson

Intervet Inc
29160 Intervet Lane
PO Box 318

In this paper the Marek's disease virus protection carried out in an one day old sheddertrial of the recombinant HVT with the F protein gene of NDV insert and the HVT with the two glycoproteins gene inserts in combination with SB1 and or CVI988 will be presented. All results will include a comparison with the conventional HVT.

In addition, results will be presented of the duration of immunity in chickens vaccinated at one day of or in ovo for NDV and ILT of both these recombinants.

10:45 – 11:00 AM

Feather PCR Diagnostic Testing to Optimize Marek's Disease Vaccination

Cheryl R. Gustafson, R. Currie, H. LeGalludec, and S. Baigent

Serotype 1 Marek's disease virus is known to replicate in the epithelium of the feather follicles, which is the site where MDV proteins are expressed. By measuring MDV replication, it is a convenient way to measure the level of vaccine multiplication and determine the actual 'take' of a vaccine. Knowing that various factors, such as breed of bird, can affect the effectiveness of vaccination, this diagnostic approach using PCR tests will aid in the determination of the optimum MD vaccination program best suited for an individual production system. (brief video of feather sampling technique included)



Session A Wednesday, July 19, 2006

11:00 – 11:15 AM

The Role of the Marek's Disease Virus UL13 Gene in Generating Cell-free Virus

Robert F. Silva and Isabel Gimeno

USDA/ARS; Avian Disease and Oncology Laboratory;
3606 E. Mount Hope Road; E. Lansing, MI 48823

Varicella-Zoster virus (VZV) and Marek's disease virus (MDV) share many biological characteristics. Both alpha-herpes are initially taken up by macrophages or dendritic cells in the lungs, and quickly spread to CD4⁺ T-lymphocytes. They are both strongly cell-associated. VZV only produces cell-free virus in the hair follicles, while MDV generates cell-free virus only in the feather follicle epithelium (FFE). Cell-free virus production in VZV requires ORF66 gene expression. The MDV UL13 gene is a homolog of the ORF66 gene. We have deleted the MDV UL13 gene and will present data indicating how the UL13 regulates cell-free MDV expression in the FFE,

11:15 – 11:30 AM

Propagation and Molecular Characterization of a Slow Growing Subgroup A Avian Leukosis Virus that was Originally Isolated from Commercial Marek's Disease Vaccines

Aly M. Fadly, Robert F. Silva, and Scott P. Taylor

USDA-ARS; Avian Disease and Oncology Laboratory;
3606 E. Mount Hope Road; E. Lansing, MI 48823

Attempts were made to propagate and obtain high titer of a slow growing subgroup A avian leukosis virus (ALV-A) that was originally isolated from commercial Marek's disease vaccines. Chicken-embryo fibroblasts (CEFs) from two ADOL chicken lines were used; also various concentrations of chicken and calf sera were included in the culture media. Passing this slow growing ALV-A in CEFs obtained from a new ADOL line named RFS resulted in high titer virus stock, as indicated by high ALV p27 ELISA readings (0.8 to 0.9), but only in cultures treated for 24 hours with 10% chicken serum. DNA sequence of this slow growing ALV-A will be presented.



Session A Wednesday, July 19, 2006

11:30 – 11:45 AM

Emergence of Subgroup J avian leukosis virus neutralizing antibody escape variants in meat-type chickens infected with virus at hatch

Arun K.R. Pandiri, Willie M. Reed, Robert F. Silva, and Aly M. Fadly

Department of Pathobiology and Diagnostic Investigation

4125 Beaumont, Ste. 208, Lansing, MI 48910

Infection of meat-type chickens at hatch with field isolates of Subgroup J avian leukosis virus (ALV J) results in a high incidence of chickens with persistent viremia even in the presence of neutralizing antibodies (NAb) against the inoculated parental virus (V+A+). The purpose of this study was to elucidate if the high incidence of V+A+ profile is due to the emergence of NAb escape mutants. Meat-type chickens were infected at hatch with an ALV J molecular clone (ADOL pR5-4). The emergence of NAb escape variants was evaluated by sequential autologous and heterologous virus neutralization (VN). Our results demonstrated that 88% of the chickens had V+A+ infection profile. All V+A+ chickens failed to neutralize autologous viruses demonstrating the emergence of NAb escape variants.

11:45 – 12:00 AM

Methods to distinguish selected serotype 1 MDV in dually infected chickens

John R. Dunn, Shari B. Gross, Richard L. Witter, Robert F. Silva, and Lucy F. Lee

USDA-ARS, Avian Disease and Oncology Laboratory

3606 E. Mount Hope Road, East Lansing, MI 48823

Marek's disease virus (MDV) can spread between vaccinated birds within a flock, leading to potential superinfection by the same or multiple strains of MDV. This study was designed to test methods to distinguish two nearly identical recombinant strains of serotype 1 MDV (rMd5 and rMd5//38CVI) in the same tissue sample. Methods used to differentiate the viruses included immunohistochemistry (IHC) using monoclonal antibodies specific for each strain, as well as pyrosequencing. 112 chickens were infected with one or both viruses. IHC and pyrosequencing were specific, as predicted, for birds singly infected. Using IHC, both virus strains were able to be detected in birds dually infected, however, the relative intensity of staining for each virus was variable between birds. Pyrosequencing was also able to detect both viruses in dually infected birds and results correlated well with IHC. IHC and pyrosequencing were successful for detecting both viruses in dually infected birds and will be used in further studies to understand the effects of serotype 1 MDV superinfection.

12:00 PM

ADJOURN



Session B Wednesday, July 19, 2006


7:00 – 7:15 AM

Moderator: David Swayne

Biological and Molecular Characterization of Recent Asian H5N1 Strains

David L. Suarez, Erica Spackman, Mary Pantin-Jackwood, and David E. Swayne
Southeast Poultry Research Laboratory
934 College Station Rd
Athens, GA 30605

Highly pathogenic H5N1 avian influenza has continued to spread in Asia and recently into Eastern Europe. Although all these viruses have a common ancestor for the hemagglutinin gene, two distinct genetic lineages have been recently characterized. A virus from a wild bird from Mongolia appear to be a good representative of one of the lineages and is closely related to viruses isolated from Russia and Eastern Europe. The other lineage will also be examined by analyzing viruses from Vietnam. Sequence analysis, biological characterization, and antigenic analysis will be performed to evaluate the diversity of viruses in the region. An evaluation of how these differences may affect control by vaccination will be discussed.



Session B Wednesday, July 19, 2006
7:15—7:30 AM

Overview of the USDA H5/H7 Low Pathogenicity Avian Influenza Program in the Live Bird Marketing System

Fidelis N. Hegngi, DVM, MS, Andrew Rhorer, Patrice Klein, Karen Grogan, Bruce Carter, and Thomas J. Myers

USDA, APHIS, VS National Center for Animal Health Programs
Aquaculture, Swine, Equine and Poultry Health Programs
4700 River Road, Unit 46; Riverdale, MD 20737-1231

Proposed by the United State Department of Agriculture, Animal and Plant Health Inspection Service, Veterinary Service, National Center for Animal Health Programs, Aquaculture, Swine, Equine and Poultry Health Programs (Poultry Team), December 1, 2005.

Background

In the past ten years, low pathogenicity avian influenza (LPAI) has become an increasing poultry health concern in the United States and internationally. This concern is based on the persistence of an H7N2 LPAI subtype virus in the Northeastern U.S. live bird marketing system; the ability of H5 and H7 LPAI viruses to mutate to highly pathogenic (HPAI) viruses; the recent examples of transmission of HPAI viruses to humans in certain Asian countries ; and the extensive trade sanctions against U.S. poultry exports that have followed LPAI or HPAI infections in commercial poultry, however limited.

USDA Domestic AI Program

Consequently, the USDA has developed an H5/H7 LPAI monitoring and control program. This program was developed with the encouragement and support of the States and the poultry industries. The program will consist of two components: commercial poultry and the live bird marketing system.

The commercial poultry segment of this program is being developed through the National Poultry Improvement Plan (NPIP). At the NPIP biennial meeting in July 2004, the plan participants adopted a new LPAI program that would provide for H5 and H7 AI monitoring of participating broiler, table egg, and turkey production flocks. The adopted program is currently proceeding through the regulatory process that will fully establish this voluntary program as part of the NPIP.

The live bird market segment of the program is being addressed through the development of a Uniform Standards document. This document defines uniform guidelines for live bird markets, as well as the producers and distributors who supply those markets, in the areas of licensing, AI testing, recordkeeping, sanitation, biosecurity, surveillance, inspections, and response to positive facilities. Participation in this program is voluntary for the States, with the expectation that participating States will enact regulations necessary for compliance of their live bird markets, producers and distributors. The Uniform Standards for this program was published in October 2004.

This program has focused primarily on the Northeast United States. In FY 2004, 10 States participated in the LBMS surveillance (NY, NJ, CT, MA, ME, PA, VT, CA, FL, and TX). In FY 2005, we have initiated cooperative agreements with 21 States (CA, DE, FL, GA, IL, IN, KY, MA, ME, MD, MN MO, NC, NJ, NY, OH, PA, SC, TX, VA, VT). As a result of recent efforts by VS and the States, the incidence of LPAI in LBMs in the Northeastern United States has decreased in FY 2005. For example, the incidence of LPAI in live bird markets in New York decreased from 13 percent of markets positive to 10.4 percent, and in New Jersey there was a decrease in positive markets from 43 percent to 1 percent.

This program will benefit the poultry industries through increased response and control of LPAI infections when they occur, particularly in the live bird marketing system. We also expect that increased monitoring in the commercial industry will lead to a decrease in the severity of LPAI-related trade sanctions as our trading partner's confidence in our monitoring system grows. Finally, early detection and response to LPAI infections is critical to preventing the development of HPAI infections, for the benefit of both poultry and human health.

Considering the global impact and cost of avian influenza, there is a great need for us to take all the steps necessary to educate everyone about this new program.

Our presentation would focus only on the Live Bird Marketing System component of this National Prevention and Control Program.



Session B Wednesday, July 19, 2006
7:30—7:45 AM

Implementation of the NPIP Low Path H5/H7 Monitoring Program in Commercial Poultry

Karen Burns Grogan, Andrew R. Rhorer, and Thomas J. Myers
National Poultry Improvement Plan
1498 Klondike Road, Suite 101
Conyers, GA 30094

The addition of commercial poultry to a voluntary, cooperative monitoring program demands cooperation and understanding between all stakeholders – the industry, the states and the government agencies. To develop and implement this additional program to a long-standing industry driven disease control program will continue to be a struggle to all involved parties. This presentation will highlight the regulatory implications of this program and the challenges that have been faced in implementation and adaptation. Furthermore, the achievements of the program to-date and any significant findings will be discussed.

7:45—8:00 AM

A field report on H3N2 Swine Influenza in Minnesota turkeys: prevalence, effect on breeders in production, serology comparisons, and vaccination strategy

Ron Lippert and Dale C. Lauer
Willmar Poultry Company
PO Box 753
Willmar, Mn 56201

H3N2 Swine Influenza has become more prevalent in the Minnesota hog population in recent years. It has recently “spilled over” into Minnesota’s turkey flocks. Most infected commercial meat flocks seroconvert with no clinical signs, diagnosis being made via the routine processing plant monitoring program. Other than serving as another reservoir of infection, the industry’s primary concern with H3N2 in meat birds has been the potential political import export ramifications.

Like the commercial flocks, turkey breeder flocks don’t get demonstrably sick. Unfortunately, a breeder flock infected during production will lay eggs like a commercial flock as well. (H3N2 infection essentially stops egg production)

Two separate introductions of H3N2 Swine Flu into turkey breeders at different stages in their production cycle will be presented, including serology and virus isolation results. State AIV surveillance results and serology comparisons between AGID, ELISA, and the University of Minnesota’s SIV H3 HI test will also be reviewed. Finally, the use of a commercial H3N4 vaccine will be discussed.



Session B Wednesday, July 19, 2006
8:00—8:15 AM

Can live attenuated avian influenza viruses be prepared for use in poultry?

Haichen Song and Daniel R. Perez

Department of Veterinary Medicine, University of Maryland, College Park
8075 Greenmead Dr.
College Park, MD. 20742

Reverse genetics provide a new avenue for the fast preparation of vaccines against influenza viruses. However, major limitations exist in terms of implementation of vaccination schemes, which basically required the administration the vaccine inoculum to each bird individually. In light of the recent outbreaks of HPAI H5N1 virus in Asia, there is an increasing need for the development of vaccines that allow mass vaccination in a shorter period of time. The cold-adapted (ca) and temperature sensitive (ts) phenotype of the cold-adapted human influenza virus has been mapped to amino acid mutations in two of the polymerase subunits and one in the NP protein. In this study, we introduced the same ca/ts mutations in PB1 and PB2 genes to the avian influenza viruses. The mutant viruses exhibit ca/ts phenotype on both the MDCK and primary chicken kidney cells. In addition, alternative mutations were introduced in the genome of two avian influenza virus, which provided additional attenuation of the viruses in vivo. Our studies suggest that live attenuated avian influenza vaccines can be prepared that are safe and will not interfere with diagnostic tools.


8:15—8:30 AM

Protective efficacy of inactivated influenza vaccines against highly pathogenic Asian H5N1 avian influenza viruses in Chickens

Samadhan J. Jadhao, Chang-Won Lee and David L. Suarez

Southeast Poultry Research Laboratory, USDA-ARS, Athens, GA 30605

We evaluated the efficacy of vaccines prepared from Asian and American avian influenza viruses (AIVs) to protect chickens from lethal challenge with Highly pathogenic (HP) Asian H5N1 AIVs. Vaccination studies were conducted in two week old chickens using inactivated vaccines prepared from reverse genetics rescued viruses or American wild type viruses, A/Turkey/Wisconsin/68 (H5N9) or A/Chicken/Mexico/232/94 (H5N2). The reverse genetics viruses used the hemagglutinin gene of A/Chicken/Indonesia/7/2003, a HP virus that was modified to be low pathogenic with different neuraminidase and internal genes. Oro-nasal challenge of vaccinated birds with a lethal dose of either A/Chicken/Indonesia/07/03 or A/Chicken/Supuranburi/2/04 virus showed all three vaccines provided good protection for chickens. Additionally, some of the vaccines could be used to differentiate infected from vaccinated animals because of different neuraminidase antibody responses.



**Session B Wednesday, July 19, 2006
8:30—8:45 AM**

ELISA test for the differentiation of infected and vaccinated animals (DIVA) using a natural truncated NS1 protein of avian influenza virus

Gloria E. Avellaneda, Chang-Won Lee, and David L. Suarez
USDA-ARS-SEPRL

Southeast Poultry Research Laboratory, Agricultural Research Service,
U.S. Department of Agriculture,
934 College Station Road, Athens, Georgia 30605

The control of Avian Influenza Virus has relied mainly on quarantine and elimination of infected flocks. This strategy allows countries to quickly eliminate disease and return to normal trade. Vaccination has not been more commonly used for control because current killed vaccines interfere with routine serologic surveillance and therefore, with the ability to export poultry. A DIVA (Differentiate Infected from Vaccinated Animals) approach with a natural variant of the NS1 protein has previously been reported, but because of how poultry vaccines are made, this NS1 approach had limitations. An alternative DIVA strategy using a virus with a naturally truncated NS1 gene has been developed. In this strategy, the deleted fragment of the truncated NS1 protein is used in a diagnostic test to provide clearer differentiation of vaccinated and infected birds. Results of conventional NS1 testing with the truncated NS1 fragment will be discussed.

8:45—9:00 AM

Impact of Respiratory Virus Vaccination on Detection of Avian Influenza Virus Infection in Broiler Chickens

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

Dept. of Animal and Food Sciences
531 South College Ave.
O44 Townsend Hall; Dept. of Animal and Food Sciences
University of Delaware; Newark DE 19716-2150

Rapid detection and diagnosis of avian influenza virus (AIV) infections in poultry is vital to eradicating the disease in its earliest stages and preventing a widespread outbreak. Depending on the timing, co-infections with common respiratory viruses may prevent the detection of LP AIV infections in poultry. Two-week-old broiler chickens were inoculated via eyedrop with a commercial modified live infectious bronchitis virus and Newcastle disease virus combination vaccine and were then similarly inoculated with LP H7N2 AIV simultaneously or on days 3 or 7 post IBV and NDV vaccination. Interference was directly assessed by performing real time AI RT-PCR as well as indirectly by determining antibody responses using HI, ELISA, and AGID tests for specific AIV.



**Session B Wednesday, July 19, 2006
9:00—9:15 AM**

***In ovo* Vaccination with an Adenovirus Replicative-defective Recombinant Vaccine
Protects Chickens against Avian Influenza Virus Challenge**

H. Toro, De-Chu Tang, David Suarez, and Kent van Kampen

College of Veterinary Medicine
Department of Pathobiology
264 Greene Hall Auburn University
Auburn, AL 36849-5519

We have produced a recombinant vaccine using a replicative-defective adenovirus vector containing the HA gene of avian influenza virus. The inoculation of this construct by the *in ovo* route has resulted in high antibody titers against the HA of avian influenza virus in the hatched chickens. We will also report on protection conferred by this candidate vaccine against challenge with highly pathogenic avian influenza virus.

9:15—9:30 AM

Swine Flu (H3N2) in Turkey Breeders

Becky J. Tilley, Eric C. Gonder, Chad E. Smith, and Sharon J. Jackson
Goldsboro Milling Co.
P.O. Box 10009
Goldsboro, NC 27532

An outbreak of swine influenza (H3N2) in turkey breeders resulted in a significant drop in production with no other clinical signs. Several commercial turkey flocks in the area were also serologically positive for H3N2, but showed no clinical signs and performed normally. This presentation will discuss the association of index cases with large swine finishing facilities and transmission among turkey breeder farms. Diagnostic challenges will be discussed. Results of a field vaccine trial using various autogenous vaccines containing H3N2 will also be presented.

9:30 – 10:00 AM

BREAK



**Session B Wednesday, July 19, 2006
10:00—10:15 AM**

Moderator: Gregorio Rosales

**Duration of Immunity to the 2002-03 California Virulent Newcastle Disease Virus (vNDV)
Following a Single Newcastle Disease Vaccination of SPF Chickens**

Darrell R. Kapczynski and Daniel J. King

USDA/ARS/SEPRL

Exotic and Emerging Avian Viral Diseases Research Unit

934 College Station Rd

Athens, Ga 30605

Newcastle disease (ND) vaccination is widely practiced in the USA with the majority of commercial chickens receiving multiple vaccinations during their lifetime. The objectives of the present study were to extend the knowledge of onset and duration of immunity of ND vaccines in specific-pathogen-free (SPF) chickens to a lethal challenge with the California 2002-03 virulent NDV (vNDV). Birds were vaccinated at 2 weeks-of-age with either a commercial live or inactivated NDV B1 vaccine. Groups of birds were challenged weekly from 3 to 8 weeks-of-age with vNDV. Vaccination with live or inactivated NDV B1 provided nearly complete protection of all challenged birds, whereas sham-vaccinated chickens were completely susceptible to challenge. Cloacal and oropharyngeal swabs were collected and tested by virus isolation. The results indicate, in the absence of maternal antibodies, single vaccination with either live or inactivated NDV B1 vaccines prevented most all morbidity and mortality following lethal vNDV challenge.

10:15—10:30 AM

Killed Newcastle disease vaccination resulting in significant mortality reduction in the presence of endemic newcastle disease virus

Terry R. Olson, DVM and Mark Cox

Moroni Feed Company

Veterinary Department

15 East 1900 South Feed Mill Road

Endemic newcastle disease virus (NDV) continues to be of worldwide importance. A lentogenic NDV was recently identified (National Veterinary Services Laboratory) as the cause of significant mortality in Utah Turkey flocks. With the introduction of a water-oil-water killed NDV vaccine, a vaccination protocol was implemented consisting of day-of-age injection of killed NDV followed by a live NDV booster (spray). This report will present a historical picture of the original disease situation prior to and following the implementation of day-of-age vaccination. This will include mortality data and important titer information that will be of significant interest to commercial turkey producers.



Session B Wednesday, July 19, 2006
10:30—10:45 AM

Incorporation of Realtime Reverse Transcriptase Polymerase Chain Reaction (rtRT-PCR) in Development of a Mucosal Challenge Model for a Lentogenic Strain of Newcastle Disease Virus (NDV)

Melissa Inman Ph.D., Martin Ficken, and Timothy Miller
Benchmark Biolabs, Inc.
521 West Industrial Lake Dr.
Lincoln, Nebraska 58628

Current models to evaluate NDV protection involves challenging intramuscularly with the Texas GB strain, bypassing innate barriers or mucosal immunological protection. Since this is an unnatural route of exposure and the challenge may not truly reflect pathogenesis of disease, a respiratory challenge model was established using a lentogenic strain of NDV by respiratory route, which causes a very mild disease in chickens. The challenge protocol was performed by inoculating various concentrations of LaSota strain of NDV by ocular/oral route. Tracheal swabs were taken at several times post inoculation for virus quantitation using the conventional ELD₅₀ titration in eggs to detect live virus and rtRT-PCR assay to detect viral RNA. Detection and quantification of viral RNA correlated to ELD₅₀ titers and other parameters of infection. The benefits of using this type of challenge and detection system can be used as an alternate challenge system for vaccine development and immune response evaluation.

10:45—11:00 AM

Chicken mucosal immunity against VG/GA, LaSota and B1 strains of Newcastle Disease Virus

Francisco Perozo, Pedro Villegas, Ivan Alvarado, and Linda Purvis
University of Georgia
Department of Population Health
953 College Station Road
Athens, GA 30602

Newcastle disease virus (NDV) mucosal antiviral immunity depends on locally produced antibodies released across the epithelium onto the mucosal surfaces. Tracheal and intestinal washes and bile samples from SPF chickens vaccinated with the VG/GA, LaSota or B1 strain of NDV will be evaluated for IgA levels. Plasma IgG will also be determined. Trachea, Harderian gland and cecal tonsil tissues will be evaluated histopathologically and screened for the presence of active plasma cells. Virus isolation and reverse transcriptase polymerase chain reaction (RT-PCR) will be used to assess the viruses' replication pattern and persistence.



Session B Wednesday, July 19, 2006

11:00—11:15 AM

Role of the Attachment Glycoprotein in Avian Metapneumovirus Virulence and Pathogenesis

Dhansekaran Govindarajan and Siba K. Samal

VA-MD Regional College of Veterinary Medicine, University of Maryland, College Park
8075 Greenmead Drive; College Park, MD-20742

Avian metapneumovirus (AMPV) causes an acute respiratory disease in turkeys and is also associated with “swollen head syndrome” in chickens. Infections due to AMPV and concomitant secondary bacteria result in serious economic losses to the turkey farmers in the United States. With a long term goal of producing a safer and more effective vaccine, we have recently developed a reverse genetics system for AMPV-C strain Colorado (AMPV/CO). This system allows the genetic manipulation of the viral genome at the DNA level, enabling the engineering of potential vaccine candidates. We have utilized this newly-developed system to recover a recombinant AMPV/CO lacking the attachment glycoprotein gene which encodes the major immunogenic protein. The recombinant mutant virus is currently being characterized for its relative virulence and immunoprotection in turkeys. We believe that these studies will help in better understanding of AMPV pathogenesis and would advance research towards the development of a safe and effective vaccine.

11:15—11:30 AM


Development of a vaccine - challenge model for avian metapneumovirus infection in turkeys: Evaluation of turkey turbinate virus preparations

David A. Halvorson, Binu T. Velayudhan, Anil J. Thachil, Sally L. Noll, Daniel P. Shaw, Sagar M. Goyal, and Kakambi V. Nagaraja

University of Minnesota; Dept of Veterinary Biomedical Sciences
1971 Commonwealth Ave; Saint Paul, MN 55108

In an attempt to develop a challenge model for avian metapneumovirus (aMPV) vaccine, different preparations of aMPV were evaluated for their virulence in turkeys. The overall goal of this study was to arrive at an appropriate preparation and dosage sufficient to produce clinical illness in unvaccinated birds. A recent isolate from 2003 (aMPV/Minnesota/Turkey/19/2003) was selected for this study. This isolate was propagated on chicken embryo fibroblasts and further propagated in turkey poults to generate nasal turbinate homogenate. Three concentrations (20, 2 and 0.2%) of the turbinate homogenate were used in addition to the original preparation. Turkey poults were divided into five groups. Turkeys in four groups were inoculated with one of the preparations. Turkeys in the fifth group were inoculated with virus-free cell culture fluid and kept as non-infected controls. Birds in all the groups were monitored post-inoculation for clinical signs, presence of viral RNA, presence of viral antigen and histopathological changes in tissues.

Nasal turbinate homogenate produced more clinical signs than the original preparation in CEF. Histopathological lesions in nasal turbinate and trachea were more pronounced in birds inoculated with turbinate preparations of MN 19 than the CEF preparation. A significantly higher percentage of birds showed the presence of viral RNA and antigen in nasal turbinate and trachea when inoculated with nasal turbinate homogenate compared to the CEF preparation. The data clearly demonstrated that the nasal turbinate homogenate prepared from birds inoculated with MN 19-CEF produced severe clinical signs and histopathological lesions in infected turkeys.



**Session B Wednesday, July 19, 2006
11:30—11:45 AM**

**Elimination of APV from Commercial Turkey Farm while concurrently stopping
Vaccination**

Brian McComb and Marion Garcia
Jennie-O Turkey Store; 110 W Maple Ave – Q; Barron, WI 54812

This report describes the elimination of Avian Pneumovirus from a turkey farm in conjunction with stopping the use of vaccination. In mid-May 2005, we decided to discontinue our APV vaccination program.

Both older flocks on this 3-age/2-stage farm had been vaccinated, presented with clinical disease, and were diagnosed APV positive. The case flock was not vaccinated for APV and remained negative clinically and serologically throughout its life.

The company veterinarian collected blood samples at irregular intervals and did clinical assessments of the flock throughout its life. It was also sampled serologically at processing. All samples were tested at the University of Minnesota.

This case study demonstrates that APV can be eliminated from a three-age farm without use of vaccination.

11:45—12:00 PM

**Comparative evaluation of avian metapneumovirus spray and eyedrop vaccination
protocols for their efficacy to protect against challenge**

**Binu Velayudhan, Anil J. Thachil, Igor Radovic, Sally Noll, David A. Halvorson,
Daniel Shaw, Sagar M. Goyal, and Kakambi V. Nagaraja**
University of Minnesota; 205 Vet Sci Bldg, 1971 Commonwealth Ave
Saint Paul, MN 55108

A live attenuated avian metapneumovirus vaccine is presently used in turkeys in Minnesota to prevent pneumovirus infection. The objective of this study was to compare two commonly used vaccination protocols (spray and eyedrop vaccinations) for their comparative efficacy. We evaluated immune response, reduction in clinical disease and virus shedding post challenge. Turkeys were challenged with aMPV; three weeks post first and second vaccinations. Following the first vaccination challenge, birds in the spray vaccinated group showed clinical signs almost similar to the non-vaccinated control group whereas birds in the eyedrop vaccinated group showed only minimum clinical signs. However birds in both the spray and eyedrop vaccinated challenged groups showed only minimal signs post second vaccine challenge. Birds in the vaccinated and non-vaccinated groups sero-converted by 14 days post challenge following first vaccination. There was a significant increase in the GMT scores of birds in the vaccinated groups upon challenge post second vaccination. Birds in the non-vaccinated and spray vaccinated challenged groups showed significant virus shedding post first vaccine challenge whereas there was a significant reduction in the virus shedding in the eyedrop vaccinated group. There was a significant reduction in the virus shedding in both the eyedrop and spray vaccinated challenged birds following second vaccination.



POSTERS

Session 1 - Sunday, July 16 and Monday, July 17 – 7:00 AM – 3:00 PM

AVIAN INFLUENZA

1.
Isolation and Characterization of Type A Avian Influenza Viruses (H9N2) from Poultry Flocks in Jordan

Mohammad Q. Al-Natour¹, Nadim M. Amarin², Hisham M. Al-Maaitah² and Ilaria Capua³

¹Department of Pathology and Animal Health, Faculty of Veterinary Medicine, Jordan University of Science and Technology, P.O. Box 3030, Irbid 22110, Jordan.

²Central Veterinary Laboratories, Ministry of Agriculture, Amman, Jordan.

³* Laboratorio Virologia, Istituto Zooprofilattico Sperimentale delle Venezie, Viale dell' Università 10, 35020. Legnaro, Padova, Italy

Forty one samples from poultry farms experiencing signs and lesions typical of airsacculitis were processed for attempted avian influenza virus isolation and identification. All serological and virological investigations were carried out in accordance to EU Directive 92/40/EEC. Characterization of isolates was performed by haemagglutination inhibition test and by RT-PCR. H9N2 viruses were identified in 22 (54%) of the examined flocks.

According to the amino acid sequence of the HA cleavage site of these isolates, it appears that low pathogenic avian influenza viruses of the H9N2 subtype are circulating in the Jordanian poultry industry. This the first report from Jordan.

2.
Transmissibility of H9N2-Avian Influenza Infection Among poultry Farms, Swine, and Farmers in Lebanon

**Elie K. Barbour, Vatche Sagherian, Houssam Shaib, Samar Dankar,
and Mohammed Farran**

Department of Animal Science, American University of Beirut
PO Box: 11-0236; Beirut Lebanon

The purpose of this work is to study the transmissibility of a newly emerging H9N2-Avian influenza virus among poultry farms, swine, and farmers of Lebanon. The presumptive diagnosis at the initiation of the outbreak in the north side of the country followed by diagnostic confirmation using indirect ELISA, Hemagglutination test, PCR, isolation of the virus, and typing proved the presence of the outbreak. The spread of the virus to chicken flocks at the central province of the country took two months, and reaching the south province, by the Israeli border, took another one month. Swine fed the carcasses of poultry infected with H9N2 seroconverted to positives, while the farmers serving the infected poultry flocks showed around 30% seroconversion to H9N2 antigens. The significance of this finding in future research targeting the control of this type of Avian Influenza virus will be discussed.



3.

Laboratory evaluation of the survivability of H7N2 Avian Influenza and Newcastle Disease Virus to commercial disinfectants

**Eric R. Benson, Robert L. Alphin, Brian S. Ladman, George W. Malone,
Michael D. Dawson, and Megan E. Lombardi**

University of Delaware
242 Townsend Hall
531 South College Avenue
Newark, DE 19716

Current methods of avian influenza (AI) control in poultry require euthanasia of flocks known or suspected to be infected. Environmental concerns have made disposal of AIV infected carcasses a major issue in many poultry-producing areas. Having a rapid, practical, cost-effective and biosecure means of inactivating AIV within the facility, litter, carcasses and on support equipment, is essential in containing disease outbreaks. The decontamination of poultry houses and equipment will be evaluated in a four-phase process.

The objective of phase one of the study (the portion presented herein) was to test the efficacy of five commercially available disinfectants on low pathology avian influenza (LPAI) and Newcastle disease virus (NDV). The viruses were tested on four materials typically found in poultry houses (glass, plastic, wood, and galvanized steel). The samples were separated and inoculated individually with A/chicken/MD/MINH MA/03(H7N2) and LaSota strain of NDV virus and allowed to dry at room temperature. Separate samples were exposed to liquid applied Virkon S, foam applied Virkon S, foam applied DF200, liquid applied BioSentry 904 and fog applied Virocid. Solution containing the decontaminated virus was then collected and inoculated into 10-11 day-old SPF embryos that were incubated and candled daily for five days. The embryos were refrigerated and fluid was tested for hemagglutination activity (HA). HA and positive control activity were used to indicate successful disinfection.

Most disinfectants had similar neutralization activity with the viral agents tested on both glass and plastic. Differences in neutralization activity were observed on both wood and metal surfaces, both of which are typically found in poultry houses. The results will be used to select the agents for the second phase, which will include disinfecting colony houses.



4. Sensitivity of Dry Swabs Utilized in an Avian Influenza Surveillance Program

Bruce R Charlton, David H. Willoughby, Beate M. Crossley, and Sharon K. Hietala
California Animal Health and Food Safety Laboratory System; UC Davis
PO Box 1552; Turlock, CA 95382

The California Department of Food and Agriculture (CDFA) has developed a plan for rapidly conducting surveillance of avian influenza (AI) in a defined geographic area within the Central Valley of California. This plan utilizes pre-positioned surveillance kits stored on individual poultry premises. Kits contain a pre-filled out laboratory submission form, pre-labeled tubes, swabs and instructions for sample collection and submission. Upon notification, farm personnel will collect cloacal swabs and pharyngeal swabs from 5 birds per house and place them in the corresponding tube (dry). Tubes and submission forms will be submitted to the California Animal Health and Food Safety Laboratory System (CAHFS) –Turlock Branch laboratory within 2 to 6 hours of collection. Personnel at the laboratory will receive samples in a biosecure fashion, log and inventory samples, add viral transport media to the tubes, remove swabs from the tubes and forward tubes to CAHFS – Davis Branch for AI RRT-PCR testing.

Ideally, collected swabs should be placed in viral transport medium which had been stored appropriately and utilized prior to any expiration date. Under these ideal circumstances, swab kits would need to be redistributed on a continual basis. Appropriate storage conditions of the viral transport media on a poultry premises maybe a difficulty or an uncertain factor. The submission of dry swabs would alleviate disadvantages of media expiration dates and storage conditions but the loss of sensitivity is unknown. A trial was conducted to determine the loss of sensitivity of the RRT-PCR procedure for detecting AI. Results will be presented.


5. PCR Screening during the First Outbreak of Avian Influenza in Romania

Handan Coste, Mihaela Zaulet, Monica Vanghele, and Mihai Turcitu
Institute for Diagnosis and Animal Health, 63, Dr. Staicovici street, sector 5,
Code 050557, Bucharest, Romania

The first case of AIV in Romania was reported in the Institute for Diagnosis and Animal Health (IDAH), Bucharest, Romania, the virus being isolated at the beginning of October 2005 in some domestic birds from the Danube Delta area. The highly pathogenic serotype H4N1 of AIV was confirmed in a couple of days by the OIE Reference International Laboratory from Weybridge, United Kingdom.

PCR was introduced as a rapid and certain test for the genome presence in the analyzed samples. A combination of Real Time RT-PCR on Light Cycler system (Roche, Germany) for the presence of Matrix protein, followed by the conventional RT-PCR (iCycler, Bio-Rad, US) for identifying the presence of H5 gene was used for screening all the samples tested in IDAH.

During 20 October – 28 December 2005, over 3800 specimens (tracheal and cloacae swabs, homogenate of brain, intestine and spleen) were tested, with 7600 tests on Light Cycler; because of the inhibitor effect of high RNA concentration in the specimen both undiluted samples and 1:10 diluted samples were tested. All the positive PCR results were confirmed by virus isolation and both Real Time PCR and conventional PCR were validated. Till the end of December 2005, a number of 6 counties in the southern part of the country were infected; the molecular exams together with virology ones are on-going.



6. **Analysis and Utilization of CpG-Motif Oligonucleotides as Potential Immunostimulatory Agents Against Avian Influenza Challenge**

John El-Attrache, Adam Jester, Ping Cui, and Blanca Lupiani
Texas A&M University; College of Veterinary Medicine
Department of Pathobiology; 4467 TAMU
College Station, Texas 77843-4467

Several CpG ODN motifs with alternating backbones were evaluated for their immunomodulatory effects in chickens. Initial dose response assays determined the optimal administration of CpGs. A preliminary humoral and mucosal dose response curve was established for CpG ODNs administered via the ocular/nasal and oral routes with and without the presence of virus. For both routes of administration, the 50ug dose was the most consistent in eliciting a specific immune response. qRT-PCR was performed to estimate viral load as well as Type I and II IFN messages elicited in response to AI challenge with and without the addition of CpGs.

7. **Evaluation on Safety and Efficacy of the Killed Vaccine against Low-Pathogenic Avian Influenza in Commercial Layer**

Bong-Do Ha, In-Pil Mo, Hyun-hee So, Ho-gun Won
Chungbuk National University, College of Veterinary Medicine, Avian Disease Laboratory
48 Gaeshin-dong Heungduk-gu Cheongju, Chungbuk 361-763 Korea

In Korea, several outbreaks of low pathogenic AI, H9N2, have been reported from the commercial farms with severe decrease of egg production and mortality resulted in severe economic loss since 1996. Therefore, it has been requested to develop AI vaccines to prevent clinical signs and economic losses from the field infection of AIV. We developed the killed vaccines using H9N2 AI virus isolated in Korea and tested the safety and efficacy of the vaccine in the SPF chickens and 7 different commercial layer farms.



8.

Development of a Multiplex RT-PCR for type A influenza virus and avian H5, H7, and H9 hemagglutinin subtypes

Mazhar I. Khan, Zhixun Xie, Yao-shan Pang, and Jianhua Sun

University of Connecticut

Department of Pathobiology & Veterinary Science

61 North Eagleville Road

Storrs, Ct 06268-3089

A multiplex reverse transcriptase-polymerase chain reaction (mRT-PCR) was developed and optimized for the detection of type A influenza virus; the assay simultaneously differentiates avian H5, H7 and H9 hemagglutinin subtypes. The mRT-PCR DNA products were visualized by gel electrophoresis and consisted of fragments of 860 bp for H5, 634 bp for H7, 488 bp for H9 hemagglutinin subtypes, and 244 bp for type A influenza virus. The common set primers for type A influenza virus were able to amplify a 244 bp DNA band for any of the other subtypes of AIV. The mRT-PCR assay developed in this study was found to be sensitive and specific. Detection limit for PCR-amplified DNA products was 100 pg for the subtypes H5, H7, and H9 and 10 pg for type A influenza virus in all subtypes.

9.

Production of Nucleoprotein (NP) of Avian Influenza Virus (AIV) from Codon-optimized Synthetic NP Gene

Hyuk-Joon Kwon, Tae-Eun Kim, Sun-Hee Cho, and Sun-Joong Kim

Laboratory of Avian Diseases, College of Veterinary Medicine, Seoul National University
(151-744) San 56-1 Shillim-9-dong Kwanag-gu, Seoul, Republic of Korea

Based on the amino acid sequence of A/Pheasant/HK/FY155/01(H5N1) of which NP gene was closely related to Korean H9N2 viruses we synthesized codon-optimized artificial gene by PTDS (PCR-based Two-step DNA Synthesis) for efficient expression in yeast. We successfully cloned a productive clone comparing with NP protein expressed in insect cell. For development of economic rapid diagnosis kit in the field we tested applicability of crude recombinant NP for latex bead agglutination test.



10.

Immunopathogenesis of H9N2 low pathogenic avian influenza virus infection in Immunosuppressed SPF chickens by cyclosporine A (CsA) treatment

Ji-sun Kwon, Huyn-jeong Lee, Yong-kuk Kwon, Youn-Jeong Lee, and Chang-seon Song
Konkuk University
College of Veterinary Medicine
1 Hwayang-dong, Gwangjin-gu,
Seoul, Republic of Korea, 143-701

We previously assessed the effects of T-cell and B-cell suppression on the pathogenesis of H9N2 low pathogenic avian influenza (LPAI) virus by cyclosporine A (CsA) and cyclophosphamide (CY) treatment. The pathogenicity of H9N2 LPAI virus in SPF chickens, determined by pathogenic index (PI) value, mortality and virus shedding, was enhanced by CsA treatment. In this study, we evaluated the immunopathogenesis of H9N2 LPAI virus infection in CsA treated SPF chickens. To clarify the immune mechanism, immunological indicators such as T-cell activity and chicken cytokines were examined. The immunopathogenic roles of H9N2 LPAI virus infection in immunosuppressed SPF chickens will be discussed.

11.

Evaluation of New Rapid Methods for the Surveillance of Avian Influenza

Chinta M. Lamichhane
Synbiotics Corporation
8075 Greenmead Drive
College Park, MD 20742

New diagnostic methods are available for the rapid and efficient large-scale screening of Avian Influenza (AI) virus and associated antibodies with improved sensitivity and specificity. The rapid tests require no special equipment and can be performed in the field. This study examines the efficacy of the new rapid methods for large scale AI surveillance of clinically normal flocks or birds.



12.

Are there better ways to determine the potential pathogenicity of avian influenza virus?

Chang-Won Lee
The Ohio State University
Food Animal Health Research Program
1680 Madison Avenue
Wooster, OH 44691

The current definition of highly pathogenic avian influenza virus (HPAIV) includes potentially pathogenic viruses regardless of their pathogenicities in chickens. However, discordant results between the molecular classification, derived by sequencing the hemagglutinin cleavage site, and virulence in experimentally infected chickens have been observed with several H5 and H7 subtype AIVs. Because the declaration of HPAIV results in severe effects on trade for the entire country, the gap between the genetic and phenotypic markers is an important issue and it requires us to reexamine what should be considered a HPAIV by the OIE standards. To better understand and assess the potential pathogenicity of the virus, the potential pathogenicities of several AIV isolates have been assessed by examining the plaquing efficiency of the virus in chicken embryo fibroblast cells and other avian cell lines, conducting a 14-day-old embryo passage and selection system, and applying in-vitro mutagenesis coupled with reverse genetics. The potential value of these complimentary methods in assessing potential pathogenicity of the AIV will be discussed.

13.

Development and evaluation of H5 subtype-specific monoclonal antibodies for avian influenza diagnostic tests

Huanguang Lu, Lin Lin, and Bill Scheuchenzuber
Wiley Lab, Wiley Lane, Orchard Road
The Pennsylvania State University
University Park, PA 16802

Subtype-specific monoclonal antibodies (MAbs) to H5 hemagglutinin of avian influenza virus (AIV) has been developed to enhance the rapid diagnosis of AIV. The H5 MAbs have been evaluated for detecting AIV by Dot-ELISA, IFA, IHC and Western Bolt assays. Results showed that the H5 MAbs specifically detected H5N2, H5N3, H5N9, and H5N1 subtypes of AIV, but they did not react to H1, H2, H3, H4, H6, H7, and H9 subtypes tested. Our preliminary results indicate that the H5 MAbs can be used for rapid detection and identification of any H5 subtypes including the H5N1 of AIV.



14.

Evaluation of the immune response of chickens to individual avian influenza proteins

Blanca Lupiani, Vinayak Brahmakshatriva, Chinta Laminchane, and Sanjay M. Reddy

Department of Veterinary Pathobiology
College of Veterinary Medicine and Biological Sciences
Texas A&M University, MS 4467
College Station, TX, 77843

Rapid detection of avian influenza virus (AIV) and antibodies in individual birds and flocks are essential for early detection and regulatory surveillance. However, the immunoassays currently available are either time consuming, have low sensitivity, or produce significant numbers of false positives. To overcome these problems, we have expressed 3 avian influenza proteins, NP, M1 and NS1, in a baculovirus expression system. The immunoreactivity of these proteins was determined with serum of chickens infected with AIV. The immune response of individual chickens to avian influenza proteins will be discussed.

15.

Comparison of sensitivity for the detection of H5N1 using two different serology techniques

Ricardo Munoz AND Miao DeYoung

IDEXX Laboratories
One IDEXX Drive
Westbrook, ME 04092

H5N1 has become one of the most important avian influenza viruses in Asia. A study was conducted to evaluate the IDEXX FlockChek* Avian Influenza Virus Antibody test kit's (AI-ELISA) analytical sensitivity for H5N1 antibody detection and correlation to HI titer. A set of SPF chickens was exposed to H5N1 inactivated vaccine. Samples were collected and a dilution series of each sample was tested on the IDEXX AI-ELISA and HI (using homologous antigen - H5N1). Unexposed controls were also tested. The IDEXX AI-ELISA limit of detection was equivalent to the homologous HI. The IDEXX AI-ELISA is an important screening tool for monitoring flocks for H5N1 exposure



16.

Avian Influenza in Romania – Apparition, Diagnostic and Epidemiology

Stefan Nicolae, Eugen Olaru, Juliana Onita, and Handan Coste
Institute for Diagnosis and Animal Health, 63, Dr. Staicovici street, sector 5,
Code 050557, Bucharest, Romania

After the evolution of the avian influenza in ten countries from South – Eastern Asia, the alert of the Romanian veterinary services has led to a backed programme for surveillance of the sensitive birds from our country.

From the beginning of 2005, 7.332 virusological samples have been examined till now and all tests have been negative.

During 4 – 12 October, 2005 the examined samples allowed the first avian influenza diagnostic in Romania and the first isolation of H5N1 virus in Europe from hens and ducks.

In this paper are presented laboratory data and work methodology which has been used (serological and virusological tests, including PCR), as well as daily epidemiological situation concerning the evolution and the control of avian influenza in Romania.

17.

**Molecular Epidemiology and Surveillance of Avian Influenza Virus
in Wild and Domestic Birds**

Annabelle M. Pascua, Nathaniel L. Tablante, and Daniel R. Perez
9116 Saint Andrews Place
College Park, MD 20740

Continued surveillance of the presence of subtypes of Avian Influenza virus in birds is critical in its epidemiology and zoonoses. A total of 591 samples were tested in the laboratory. These samples were collected in the months of December 2004 and May to August 2005 from wild birds, domestic poultry and environment. Out of 591 samples, seven samples were tested positive for H11N3 as detected by Hemagglutination Assay, Real Time RT-PCR and RT-PCR. Full length genome sequencing of H11N3 is presently on its completion for comparative studies with other H11 gene segment sequences and phylogenetic analysis.



18.

Validation of H5 and Matrix Real-Time Reverse Transcriptase Polymerase Chain Reaction (rRT-PCR) Bead Reagents for the Detection of H5 Avian Influenza Virus

**Janice C. Pedersen¹, David L. Suarez², Amaresh Das², Dennis A. Senne¹
and B. Panigrahy¹**

National Veterinary Services Laboratories, Veterinary Services, Animal and Plant Health Inspection Service, U.S. Department of Agriculture, P.O. Box 844, Ames, IA 50010¹, Southeast Poultry Research Laboratory, Agricultural Research Service, U.S. Department of Agriculture, 934 College Station Road, Athens, GA 30605²

Matrix and H5 rRT-PCR primer and probe bead assays were compared to the conventional wet reagent assays currently described in the official rRT-PCR protocol used for the surveillance of AI by the National Animal Health Laboratory Network. The matrix bead includes assay specific primers and probes and an internal control for the detection of false negative results. The H5 bead has multiple primers designed to detect both the Eurasian and North American lineages of H5 AIV. Clinical specimens, including cloacal swabs, were tested by virus isolation and rRT-PCR for validation of the internal control. The H5 bead was compared to NVSL's assay for specificity and sensitivity. The bead assays were comparable in sensitivity to the conventional assay and have been included in the official protocol for distribution to NAHLN laboratories for AI surveillance.

19.

Prevention and Control of Avian Influenza: Advances and Perspectives

Daniel R. Perez

Avian Influenza Coordinated Agricultural Project (PI: Daniel R. Perez, Co-PI: Richard Slemons)
Department of Veterinary Medicine, University of Maryland, College Park
8075 Greenmead Dr., College Park, MD. 20742

In February 2005, the CSREES-USDA funded the single largest project to study, prevent and control a poultry disease: Avian Influenza. The Program "Prevention and Control of Avian Influenza in the US" was created with the participation of 17 US institutions (details can be found at www.agnr.umd.edu/aicap). Projects were concentrated in five major areas including studies interspecies transmission and pathogenesis, surveillance of wild and domestic birds, education and outreach programs, and development of novel and alternative methods of diagnosis and vaccines. Certainly, a project of this magnitude is always in a constant quest for improvement and new challenges, particularly when it comes to coordinate the participation of a large number of institutions. For the past year, important advances have been made in all project areas, with important highlights in the education aspects of the program, in which poultry extension veterinarians have been particularly active. Important advances have been made also in the area of interspecies transmission, pathogenesis and diagnosis. In a time in which, poultry producers and the public are increasingly aware of the implications of avian influenza for birds and humans, our project is building the foundation to confront this disease through an extensive research network. Although there is a lot to be done, we are confident that this project is aimed in the right direction.



20.

Use of the Hemagglutination Inhibition Test To Assess Antigenic and Genetic Relatedness of Avian Influenza H5 Proteins

Jennifer Pfeiffer and David L. Suarez
Southeast Poultry Research Laboratory
934 College Station Rd
Athens, GA 30605

Following an outbreak of avian influenza in Mexico, vaccination was used for control, but the virus persisted in the poultry population in part because of antigenic drift. By using sera from DNA-vaccinated chickens, we used the hemagglutination inhibition test to determine the specific molecular markers related to antigenic drift for the Mexican lineage of viruses. We also want to test these and sera from chickens vaccinated with other types of vaccines using a variety of antigens of various H5 lineages. We want to correlate the genetic and antigenic relationship between different viruses to allow better selection of vaccines during outbreaks.

21.

Pathogenesis of Avian influenza virus with mutation in the NS1 gene

Sanjay M. Reddy, Vinayak Brahmakshatriva, and Blanca Lupiani
Texas A&M University
Veterinary Pathobiology, College of Veterinary Medicine,
TAMU (MS 4467)
College Station, TX 77843

Avian influenza is a respiratory disease of poultry and has worldwide importance. The causative agent is an *Orthomyxovirus* and codes for eight segments of negative polarity. We have generated a reverse-genetic system for the rescue of a moderately pathogenic strain of AI virus. Using this system we have introduced a stop codon in the NS1 gene to generate a truncated NS1 protein (1-142 amino acid residues). This mutant virus grows to high titers in 7-day old embryos but not in older embryos suggesting that it is involved in interferon regulation. The role of NS1 protein as a virulence factor in chickens will be discussed.



22.

Efficacy of a vectored Fowl Pox-Avian Influenza vaccine administered subcutaneously to One-Day-Old Broiler chickens and challenged with Fowl Pox virus at *Five Weeks Post-Vaccination*

Francisco J. Rojo Barrañón, Rafael Fernandez, Enrique Montiel, Héctor Garcia

Merial Avian Global Enterprise
Av. De las Fuentes No. 66
Parque Ind. FINSA
El Marques, Qro.
México 76246

Commercial broilers were vaccinated against Fowl Pox using a Fowl Pox - Avian Influenza recombinant vaccine (Trovac[®]AIV5, Merial-Select laboratories, Gainesville GA), a Fowl Pox Frozen vaccine (FP), and a Pigeon Pox vaccine (PP) to compare protection against challenge. The birds were vaccinated at the hatchery, kept in commercial conditions, and challenged at 5 weeks of age with Fowl Pox virus strain, at a dose of $10^{6.0}$ EID₅₀ per bird, by wing web infection (WW) route.

23.

Efficacy of a vectored Fowl Pox-Avian Influenza vaccine administered subcutaneously to One-Day-Old Broiler chickens and challenged with Fowl Pox virus *Local Strain* at *Five Weeks Post-Vaccination*

Francisco J. Rojo Barrañón, Rafael Fernandez, and Montiel Enrique

Merial Avian Global Enterprise
Av. De las Fuentes No. 66
Parque Ind. FINSA
El Marques, Qro.
México 76246

Commercial broilers were vaccinated against Fowl Pox using a Fowl Pox - Avian Influenza recombinant vaccine (Trovac[®]AIV5, Merial-Select laboratories, Gainesville GA) combined with Marek's vaccine. The birds were vaccinated at the hatchery, kept in commercial conditions, and challenged at 5 weeks of age with a *Local* (Mexican) Fowl Pox virus strain, at a dose of $10^{3.5}$ EID₅₀ per bird, by wing web infection (WW) route. The effect of vaccines dosage was compared as many vaccines are commonly used in partial doses in broilers. Protection was assessed comparing groups with a control group vaccinated with a "traditional vaccination schedule from the region".



24.

Avian Influenza Virus Surveillance in Wild Birds in Lower 48 during 2005

**Richard D. Slemons, Carol J. Cardona, David A. Halvorson, Joseph J. Giambrone,
John El-Attrache, and Daniel R. Perez**

Members of USDA CSREES AIV Surveillance in Wild Birds Network
Department of Veterinary Preventive Medicine, The Ohio State University
1920 Coffey Rd.
Dept. Vet. Prev. Med.
Columbus, OH 4210

In February 2005 the USDA CSREES Competative Grants program announced the funding of a project entitled, "Prevention and Control of Avian Influenza in the United States." The funded research proposal had been in preparation for two years and includes a consortium of 24 researchers/research groups located in 14 states. Objective 4 of this grant is to establishment a systemantic, continent wide avian influenza virus sureviellance program in wild birds and a GIS database for examining temporal-spatial-host-virus relationships associations with avian influenza viruses and the occurance of AI in poultry. Progress reports indicate first year deliverables will be reached. An updated progress report will be presented.

25.

Characterization of H9N2 Avian Influenza Virus Isolated from Korea

**Haan-Woo Sung, Youn-Jeong Lee, Jun-Gu Choi, Eun-Kyoung Lee, Jae-Hong Kim, and
Hyuk-Moo Choi**

Department of Veterinary Medicine, Kangwon National University,
192-1 Hyoja 2 dong, Chunchon,
Kangwon-do, 200-701, Republic of Korea

Thirty four H9N2 avian influenza viruses isolated in Korea were characterized. No isolates caused mortality in inoculated SPF chickens of 6 week-old. Sequence analysis showed that no isolates had multiple basic amino acid motif required for highly pathogenic virus at the hemmagglutinin cleavage site. However, the amino acid motifs varied with isolates.



26.

H5N1 HPAI Virus in Wild Birds of Mongolia

David E. Swayne and David L. Suarez

USDA/ARS/SEPRL; 934 College Station Road; Athens, GA 30605

Outbreaks of H5N1 high pathogenicity avian influenza (HPAI) have occurred in poultry over the past 9 years in Asia beginning with domestic geese (*Anser anser*) in China (1996), followed by outbreaks involving multiple poultry species in Hong Kong (1997, 2001, 2002 and 2003), and widespread geographic outbreaks in 11 Asian countries (2003-2005), including recent outbreaks in Kazakhstan and Russia. The mode of virus spread to individual geographic locations and between countries is not always clear. Spread could be the result of sanctioned or unsanctioned trade in poultry and their products such as commercially exported duck meat or movement of infected birds in the live poultry marketing system; smuggled fighting cocks and other birds; shared use of contaminated equipment; and natural movement of wild birds (1). Recently, an H5N1 HPAI viruses, closely related to poultry viruses of southern China, were detected in wild birds on Qinghai Lake, China. Although wild birds have been suggested as the source of these viruses, other routes of introduction cannot be excluded given there is poultry rearing in the general vicinity of the lake. This report describes wild birds as the source of an outbreak of H5N1 HPAI virus in Mongolia.

27.

Vaccination with Alphavirus-derived Neuraminidase Partially Protects Chickens against Heterologous Avian Influenza Challenge

Sylte, M.J^{1*}, Hubby, B² and Suarez, D.L¹

¹*Southeast Poultry Research Laboratory, Agricultural Research Service, U.S. Department of Agriculture, 934 College Station Road, Athens, GA 30605.*

²*AlphaVax, Inc., P.O. Box 110307, 2 Triangle Drive, Research Triangle Park, North Carolina 27709.*

Antibodies produced against the influenza hemagglutinin protein protect against avian influenza (AI) infection, whereas the role of anti-neuraminidase (NA) antibodies is less clear. We hypothesized that primary response to NA vaccination was important in protection. Using chickens, we compared three different types of vaccines; either NA 2 (N2) replication deficient viral-like particles (VLP), baculovirus-derived protein or DNA vaccines, alone or in combination. Chickens vaccinated with VLP were partially protected against AI challenge, whereas other primary vaccination types failed to protect (e.g., DNA and baculovirus-derived). Results of NA-inhibition titers and AI shedding will be discussed. These results suggest that the VLP are more effective than DNA or baculovirus-derived NA in priming protective immunity against AI challenge.



28.

Responding to an Avian Influenza Outbreak: A Quick Reference Guide

Nathaniel L. Tablante

University of Maryland College Park
VA-MD Regional College of Veterinary Medicine
8075 Greenmead Drive
College Park, MD 20742

Avian Influenza has recently been the focus of media attention due to the sporadic outbreaks of H5N1 “bird flu” in Asia that has caused severe disease and mortality in both poultry and humans. Fearing the possibility of a global Avian Influenza pandemic, many countries are preparing comprehensive programs to prevent bird flu. However, in some cases, outbreaks of AI still occur and preventing the spread of the disease becomes a top priority. For poultry operations, this “Quick Reference Guide” may serve as a checklist for emergency personnel so they can respond promptly to any outbreak of Avian Influenza or any catastrophic disease. The essentials include trained personnel, equipment, supplies, and methods of mass depopulation (euthanasia) and carcass disposal. It is easy to say “be proactive” but do you really know what you need and will they be available at the time of an outbreak?

29.

Properties of an avian influenza virus (H4N8) isolated from adult tom turkeys

Peter R Woolcock, Jinling Li, Nichole Anchell, George Cooper, and Carol J. Cardona
CAHFS, UC Davis
2789 S Orange Ave
Fresno, CA 93725-1919

Tissue samples were submitted for virus isolation as a result of positive serology. AIV was isolated from cecal tonsil on first passage in eggs, HA titer was very low, but Directigen was positive. On serial passage the virus could not be detected. Only after 4 or 5 passages was the HA titer increased and the virus observed by negative stain EM. Virus growth in chicken eggs and MDCK cells has been investigated using real time RT-PCR to evaluate viral load and comparing it with HA activity.



30.

Quail carry sialic acid receptors compatible to bind avian and human influenza viruses

Hongquan wan and Daniel R. Perez

Department of Veterinary Medicine, University of Maryland, College Park
8075 Greenmead Drive, College Park, MD 20742

To better understand the role of quail as an intermediate host for the zoonotic transmission of influenza viruses, we examined the types and distribution of sialic acid receptors in quail. We found that besides the presence of SA α 2,3-gal receptors, there are abundant SA α 2,6-gal receptors in quail trachea and intestine. In trachea, the SA α 2,3-gal are present primarily in nonciliated cells, while SA α 2,6-gal are localized predominantly on ciliated cells. In intestine, both types of receptors were found on epithelial cells as well as in crypts. In agreement with these observations, in a solid-phase overlay binding assay, both avian and human influenza viruses bound to plasma membranes prepared from epithelial cells of quail trachea and intestine, strongly suggesting that these receptors are functional for binding of influenza viruses from different species. These results are consistent with the notion that quail could provide an environment for the spread of reassortants between avian and human influenza viruses.

31.

Antigenic and Genetic Studies on H3N2 Influenza A Viruses Isolated from Swine and Turkeys

Hadi Yassine

Food Animal Health Research Program/ OARDC/OSU
1680 Madison Avenue; Wooster, Oh 44691

Influenza A viruses are highly infectious pathogens that affect various species of animals including mammals and birds. They are classified into different subtypes based on two major surface proteins hemagglutinin (HA) and neuraminidase (NA). Triple reassortants of H3N2 viruses with genes derived from Human, swine and turkeys are becoming endemic in swine and turkey populations in the United States. Turkeys are susceptible to several subtypes of influenza viruses including swine viruses, of which vaccines have been developed and used to protect turkey breeders against H3N2 subtypes. We isolated and characterized two H3N2 viruses from breeder turkey hens in Ohio and Illinois. Infections with these viruses were associated with complete cessation of egg production. The Illinois flock was vaccinated twice with an inactivated virus isolated from swine. This prompted us to initiate a study on the antigenic and genetic relatedness of the turkeys and swine isolates. In this study, four viruses, three from turkeys and one from swine, were tested for their antigenic relatedness using Hemagglutinin Inhibition test and Virus Neutralization test in cell culture. The formula of Archetti and Horsfall was employed to express the antigenic relatedness of the different isolates. Results showed that turkey isolates are highly related, however the swine isolate was very distantly related. In addition, the genetic studies revealed a high degree of similarity between the turkey viruses isolates which were distantly similar to the swine isolate.

In conclusion, the antigenic and genetic studies point to the high degree of relatedness between turkey isolates and a distant similarity between the swine and turkey isolates.

32.

**Differentiation of Highly Pathogenic Strain H5N1 of AIV
by Real Time PCR in the First Outbreak in Romania**

Mihaela Zaulet, Handan Coste, Mihai Turcitu, and Monica Vanghele
Institute for Diagnosis and Animal Health, 63, Dr. Staicovici street, sector 5,
Code 050557, Bucharest, Romania

Fast diagnosis of Avian Influenza is a prerequisite for confining outbreaks. Diagnosis implies the differentiation of virulent and non-virulent Avian Influenza virus. After starting with PCR screening for matrix protein followed by identifying the presence of H5 gene, the diagnosis methodology within Institute for Diagnosis and Animal Health has moved to rapid molecular tests for detecting the virulent and non-virulent strains.

The Real Time RT-PCR test performed on Light Cycler system (Roche, Germany) provides fast, easy and accurate results in identifying and quantifying the virus. Specific primers and probes for detecting influenza virus A Matrix Protein, Influenza virus A H5, Influenza virus A N1 and Influenza A H5N1 were used. PCR results with specific fragments of 250 bp, 189 bp, 198 bp and respectively 161 bp, are obtained within one hour.

RNA extraction was done with MagNA Pure Compact system (Roche, Germany) and two Light Cycler Real Time systems were used simultaneously; the PCR tests were further validated by using over 3400 specimens of tracheal and cloacae swabs, homogenate of brain, intestine and spleen from 16 affected poultry backyards.

BACTERIA, MISCELLANEOUS

33.

**Campylobacter Susceptibility to Ciprofloxacin and Corresponding Fluoroquinolone
Concentrations Within the Gastrointestinal Tracts of Chickens**

P. J. Blore, I. Reyes-Herrera, K. Cole and D. J. Donoghue
Poultry Science Department, Rm O-315
University of Arkansas
1260 W. Maple St
Fayetteville, AR 72701

This study was designed to evaluate the relationship between gut fluoroquinolone concentration and changes in susceptibility of *Campylobacter* to ciprofloxacin. Chickens were challenged With *C. jejuni* at two weeks post hatch and treated at 26 d with 0, 25 or 50 ppm of fluoroquinolone in the drinking water for 3 d or 7 d, respectively. The crop, upper ileum, ceca and colon were collected during dosing and 14 d withdrawal period. *Campylobacter* susceptibility and the corresponding antibiotic concentrations within the gut locations were determined on each collection day. The ciprofloxacin MIC for *Campylobacter* isolated from both enrofloxacin treatments increased within the first day of dosing compared with controls. Gut location did not affect fluoroquinolone concentrations or *Campylobacter* resistance within either treatment group.



34.

Chronic Fowl Cholera with Osteomyelitis of the Skull in Broiler Breeders

^AJose J. Bruzual, ^BMatilde Alfonso, ^AGuillermo Zavala and ^CJohn Smith

^AUniversity of Georgia, 953 College Station Road, Athens, GA 30602-4875

^BNorth Carolina State University, 4700 Hillsborough St., Raleigh, NC 27606 27560

^CFieldale Farms Corp. PO Box 558, Baldwin, GA 30511

During the winter of 2005, two broiler breeder flocks (36-week-old Hubbard UY/Cobb, and 51-week-old Hubbard UY/Hubbard) were reported to have a slight increase in mortality and show torticollis in both males and females. Egg production was slightly below the standard. Necropsy of the affected birds demonstrated no egg production, osteomyelitis of the skull and middle ear, and small discolored focal areas on the liver surface. Histological examination revealed an acute multifocal necrotizing hepatitis with intralesional rod bacteria that were later identified by culture as *Pasteurella multocida* serotype 1.

35.

The Efficacy of NETVAX[®] Necrotic Enteritis Vaccine In Protecting Cobb 500 Broiler Progeny From Vaccinated Breeders When Artificially Challenged With *Clostridium perfringens*

Stephen W. Davis, Charles Broussard, and Richard A. Phillips

Colorado Quality Research, Inc.

400 E. County Road 72

Wellington, CO 80549

A study was conducted to evaluate the efficacy of a new necrotic enteritis vaccine for breeders (NETVAX[®]) using two sources of Cobb 500 chicks (one source from breeders that had received the NETVAX[®] vaccine and the other source from breeders that had not received the vaccine). The study design also compared the performance of the NETVAX[®] vaccinated broilers in Salinomycin coccidiostat or Coccivac-B vaccination programs with or without the inclusion of a growth promoting antibiotic. The treatments were artificially challenged with a *Clostridium perfringens* challenge model and performance results including growth, feed conversion, mortality and necrotic enteritis lesion scores will be presented.



36.

Serotyping and antibiotic susceptibility of *Riemerella anatipestifer* isolates obtained from commercial Pekin duck flocks in Northeastern United States

Rakijah Galloway-Haskins, Alejandro Banda, and Tirath Sandhu
Cornell University Duck Research Laboratory
P. O. Box 217, 192 Old Country Road
Eastport, NY. 11941

Riemerella anatipestifer was isolated in samples from ninety-two commercial Pekin duck flocks submitted for routine diagnostic purposes from November 2003 to November 2005. *R. anatipestifer* was isolated in 43.5% of the samples (299 out of 687). The positive samples were serotyped by plate agglutination using polyclonal sera. Serotype five was most frequently isolated (78%); other serotypes detected were serotype one (3.7%), two (9.4%), seven (0.3%), and eleven (3.7%). In general, the isolates were susceptible to penicillin, enrofloxacin, novobiocin and tetracycline. Antibiotic resistance was observed with neomycin, and kanamycin. Variable results were observed with erythromycin, gentamycin, and with the combination of sulfamethoxazole and trimethoprim.

37.

Hemagglutinin antibody levels and protection in chickens given infectious coryza vaccines

A. García^{1*}, F. Romo², A.M. Ortiz³ and P.J. Blackall⁴

¹Laboratorios Avilab, Porcicultores 80 Tepatitlan, Jalisco, México C.P. 47600, México,
²LIPEPSA Av. Avicultores S/N. Tepatitlan, Jal. 47600, México, ³Facultad de Estudios Superiores Cuautitlán, UNAM. Apdo. Postal 25, Cuatitlán Izcalli, Edo. de México. CP 54740. México and
⁴Queensland Department of Primary Industries and Fisheries, Animal Research Institute, Yeerongpilly 4105, Australia

We have investigated the relationship between hemagglutination-inhibition (HI) antibodies in infectious coryza vaccinated chickens and the protection against challenge. In two experiments, chickens given a single vaccine dose lacked a detectable HI response but showed good levels of protection (from 60 to 100% protection) at 11 weeks post-vaccination. In contrast, in chickens given two doses of a vaccine and challenged three weeks after the second vaccine dose, there was a strong serological response (36/40 birds having a HI titer of $\geq 1/20$) and good protection. Our results suggest that HI titers cannot be regarded as a definitive predictor of vaccine efficacy.



38.

Development of competitive ELISA to determine antibody titer in birds vaccinated with *C. perfringens* type A alpha toxoid (NetVax™)

Huchappa Jayappa, Suzan Dimmick, Lindy Dierks, Joan Schrader, and Terri Wasmoen
Schering Plough Animal Health
21401 West Center Road

The role of *Clostridium perfringens* Type A in the pathogenesis of necrotic enteritis has been firmly established. The vaccine containing *C. perfringens* Type A alpha toxoid has been shown to provide passive protection in progeny chicks. A competitive ELISA has been developed to determine antibody response to alpha toxin in egg yolk, sera of hens vaccinated with *C. perfringens* type A alpha toxoid (NetVax™). The assay was shown to be sensitive and specific. The assay can be used to monitor the flocks for the presence of antibody titer to alpha toxin.

39.

Molecular characterization of *L. monocytogenes* isolated from poultry meat

Nitin V Kurkure, Dewanand R. Kalorey, Pramod K. Jallewar, Prashant S. Gunjal, Arun G. Bhandarkar, and Sukhadev B. Barfuddhe
Maharashtra Animal & Fishery Sciences University,
Nagpur Veterinary College, High land drive, Seminary Hills
NAGPUR 440 006 INDIA

The 92 poultry meat samples were collected from the local poultry meat market. Isolation of the pathogen was attempted from the samples by selective enrichment in University of Vermont Medium and plating onto Dominguez-Rodriguez isolation agar. Confirmation of the isolates was based on biochemical tests followed by in vitro pathogenicity testing. Pathogenicity of the isolates was tested by Christie, Atkins, Munch Petersen (CAMP) test, phosphatidylinositol-specific phospholipase C (PI-PLC). The isolates were subjected to PCR assay for the five virulence associated genes namely, *plcA*, *hlyA*, *actA* and *iapA*. A total of six *Listeria* spp. were isolated and out of them three were confirmed as *L. monocytogenes*, two were confirmed as *L. innocua* and one isolate was confirmed as *L. sellegiri*. Out of three cultures of *L. monocytogenes* two culture harbored all pathogenic genes *plcA*, *hlyA*, *actA* and *iapA* gene. While remaining one culture harbored only *hlyA* and *iapA* genes. The multiplex PCR reaction was also developed for the detection of *plcA*, *hlyA*, *actA* and *iapA* and the results were consistent.



40.

A serotypic survey of poultry *Pasteurella multocida* isolates from 1996 to 2005

Christine M. Lobsinger, Charles L. Hofacre, and Stephan G. Thayer

University of Georgia, Poultry Diagnostic Research Center
953 College Station Rd.
Athens, GA 30605

A survey of *P. multocida* serotypes from cultures submitted to PDRC and cultures isolated at PDRC between the years of 1996 to 2005. The isolates were submitted from varying states and mostly from broiler breeders. Other species were rabbit, broilers, layers, turkeys, ducks, quails and pheasants. Each isolate was tested against 16 known antisera using AGID method. Results will be presented.

41.

Virulence and Resistance Genotyping of *Campylobacter* from Production Turkeys in the Midwest

Catherine M. Logue, Mohamed K. Fakhr, Shana R. Petermann, Ellen Lutgen Johnson, and Julie S. Sherwood

Department of Veterinary and Microbiological Sciences,
130a Van Es Hall
North Dakota State University, Fargo, ND 58105, USA

Genotyping was used to characterize virulence and resistance of *Campylobacter* recovered from production turkeys in the Midwest in order to develop a pathotype to define pathogenic *Campylobacter* as it relates to host species. *Campylobacter* isolates recovered from production turkeys were subjected to PCR and probed to determine the presence of virulence and resistance genes associated with type IV secretion systems, invasion, toxin production, host resistance and drug resistance. Results from the study found that some genes associated with the resistance and persistence were common and located either on the chromosome or on plasmids, suggesting a role for plasmids as a potential factor in the pathogenesis of *Campylobacter*. There appear to be key genes which may be useful in defining a pathotype.



42.

**Prevalence of Bacterial Pathogens and their Antimicrobial Resistance
in Backyard Poultry Flocks**

Kimberly L. Moherman, Teresa Y. Morishita, and J. David Latshaw
OSU, Department of Veterinary Preventive Medicine
1920 Coffey Road
Columbus, OH 43210

Although there is much information on disease outbreaks in the commercial poultry industry, there is a lack of information on diseases and health programs for the small (or backyard) owner. It is important to gather such information since these small flocks can serve as reservoirs for diseases to commercial poultry flocks. Because there is a lack of information on the health and management status of this particular poultry population, it is important to collect such data. Hence, the objective is to determine the exposure status of small (or backyard) poultry flocks to common disease agents through culture and serological tests. Results will indicate the prevalence of bacterial pathogens such as *Salmonella* and their antimicrobial resistance in these backyard flocks based on 4 main focus groups: 1) exhibition poultry; 2) youth groups (i.e. FFA and 4-H); 3) organic producers; and 4) backyard "pet chicken" flock owners.

43.

A Bacteriologic Survey of Wing Web-Applied Vaccine Reservoirs

Andy McRee, DVM, dipl. ACPV
Intervet, Inc.
1203 Linton Court
Cary NC 27511

A bacteriologic survey of reservoirs used during live fowl cholera vaccination of breeder pullets was conducted subsequent to observations of severe post-vaccination wing web tissue reaction. Accumulated dust, debris and feathers were evident upon visual inspection of vaccine reservoirs used for wing web application, especially those used during the entire vaccination handling process (a period of ~5-7 hours). Reservoirs were swabbed for bacterial culture utilizing blood, MacConkey, Pea and Baird-Parker agars. Culture yielded extensive growth of a variety of bacterial contaminants including potential pathogens such as *E. coli* and *S. aureus*.



44.

Bacterial Orchitis and Epididymo-orchitis in Broiler Breeders

Rafael Monleon and Harold John Barnes

North Carolina State University – College of Veterinary Medicine
4700 Hillsborough St.; Raleigh, NC 27606

Two cases of orchitis identified during investigations of broiler breeder male health are described. The first case involved only one testicle and was due to *Staphylococcus aureus*; the second was a case of bilateral epididymo-orchitis caused by *Escherichia coli*. The affected testicle in Case 1 was enlarged, irregularly shaped, edematous, swollen, yellow color, had fibrin on the surface, and displayed prominent vessels. The contralateral testicle was normal. In Case 2, the affected testicles were enlarged, irregularly shaped, had raised spotted and more normal appearing depressed areas, and prominent vessels. Multiple petechiae were present on the *tunica albuginea*. On cut section, they exhibited extensive multifocal areas containing pale, granular foci. Microscopically, heterophilic interstitial-intratubular orchitis with intralesional bacterial colonies was observed in both cases. Epididymitis also was observed in Case 2. These findings are indicative of an ascending infection.

45.

Virulence and Immunogenicity of a *Riemerella anatipestifer* serotype 3 strain

Tirath Sandhu

Cornell University Duck Research Laboratory
P. O. Box 217; Eastport, NY 11941

Riemerella anatipestifer causes a serious economically important disease in ducks, turkeys and other birds. Although serotype 1, 2 and 5 cause majority of outbreaks in ducks in the United States, other serotypes such as 3, 6, 7 and 11 have also been reported to produce high mortality in ducks at some duck farms.

Virulence of serotype 3 isolates was tested to look for avirulent or low-virulent strains as potential vaccine candidates. One isolate (DRL-31009) did not cause any mortality or disease in 10-day-old ducklings exposed by intranasal route to 5×10^7 CFU (Colony Forming Unit). This isolate was further tested for safety and immunogenicity. Two-week-old ducklings exposed to 10^8 CFU by aerosol showed protection against a challenge with virulent type 3 strain that produced 65% mortality in susceptible control ducklings. Isolate DRL-31009 was tested alone and in combination with a trivalent (serotypes 1, 2 and 5) live vaccine against challenge with serotypes 1, 2, 3 and 5 virulent strains. Three groups of 80, one-day-old ducklings in each group were vaccinated with isolate DRL-31009, trivalent vaccine, and isolate DRL-31009 in combination with trivalent vaccine by aerosol spray in separate units. Eighty susceptible hatch mates were held separately as an unvaccinated control group. At 4 weeks of age, 20 ducklings from each group including unvaccinated control were challenged with virulent strains of serotype 1, 2, 3 and 5. No mortality or unfavorable reaction due to vaccination was observed in ducklings in any group. Isolate DRL-31009 and the trivalent vaccine in combination with isolate DRL-31009 produced good protection when challenged with type 3 virulent strain. Isolate DRL-31009 did not provide protection against challenge with serotype 1, 2 and 5 virulent strains and vice versa. A combination of isolate DRL-31009 and trivalent vaccine provided significant protection against challenge with virulent serotypes 1, 2, 3 and 5 strains.



46.

M 13 and ERIC 1R PCR fingerprinting of *Ornithobacterium rhinotracheale*

**Anil. J. Thachil, Binu T. Velayudhan, Vanessa Lopes,
and Kakambi V. Nagaraja**

University of Minnesota
205 Vet Sci Bldg
1971 Commonwealth Ave
Saint Paul, MN 55108

Ornithobacterium rhinotracheale (ORT) is an infectious respiratory pathogen of chickens, turkeys and wild birds. There are eighteen serotypes of ORT so far reported worldwide. Our studies revealed that DNA fingerprints obtained by M13 fingerprinting were more discriminative in differentiating ORT isolates than with ERIC1R fingerprinting. ORT M13 fingerprinting revealed distinct fingerprint patterns for all the reference serotypes of ORT viz: A,C,D,E,F,I,J, and K tested. However when large number of ORT field isolates was analyzed there appeared to be no correlation between serotype specificity and fingerprint patterns. But within ORT serotypes A, C and I we could find distinct and different fingerprint patterns. More than 18 distinct fingerprint patterns were obtained in this study and this procedure is very valuable in the epidemiological investigation of ORT infection.

47.

**Histopathologic Lesions of Experimentally Induced Gangrenous Dermatitis
in Commercial Broilers**

Susan M. Williams and Stephen Collett

University of Georgia
PDRC, 953 College Station Road
Athens, GA 30605

A gangrenous dermatitis challenge model based on subcutaneous injection of *Clostridium perfringens* type A broth culture in the breast of 4 week old commercial broilers consistently induces gangrenous dermatitis lesions in all challenged birds. Unlike field cases, these experimentally induced lesions resolve very rapidly despite the persistence of viable *C. perfringens* type A organisms in the sub-cutis up to 7 days after challenge.

To learn more about the pathogenesis and progression of this re-emerging disease, skin lesion samples were collected every 24 hours for 7 days after challenge. Samples were fixed in 10% formalin and routinely processed for microscopic examination. The sequence of events following challenge will be described histologically.



48.

Macrolide-resistance in *Campylobacter*: emerging frequency and resistance mechanisms

Qijing Zhang, Meiguan Yan, Sonia Pereira, and Orhan Sahin
Department of Veterinary Microbiology, Iowa State University,
1116 Vet. Med., Ames, IA 50011

Erythromycin, a macrolide antibiotic, is one of the key antibiotics used for treating human foodborne illnesses caused by *Campylobacter*. Since macrolides are also used for poultry production, there is a concern that use of macrolide antibiotics may promote the occurrence of macrolide-resistant *Campylobacter* in the food chain. To address this concern, we conducted both *in vitro* and *in vivo* studies to determine the emergence of macrolide-resistant *Campylobacter* mutants in response to treatment with macrolide antibiotics. Our results indicate that that in contrast to fluoroquinolones, *Campylobacter* has extremely low mutation rates to macrolide antibiotics and the treatment of poultry with tylosin does not promote the occurrence of macrolide-resistant *Campylobacter*. These findings reveal the complexity of macrolide resistance in *Campylobacter* and suggest that the emergence of macrolide-resistant *Campylobacter* on poultry farms can not be simply explained by use of this class of antibiotics.

E. COLI

49.

**Role of iron acquisition systems in virulence of an avian pathogenic
Escherichia coli strain**

Melissa Caza, Roy Curtiss III, and Charles M. Dozois
University of Quebec, INRS-Institut Armand-Frappier
531 boul. des Prairies
Laval, Quebec H7V 1B7 Canada

Avian pathogenic *Escherichia coli* strain chi7122 (O78:K80:H9) causes aerosacculitis/colibacillosis in chickens. Chi7122 encodes several iron acquisition systems including the plasmid-encoded siderophores aerobactin and salmochelin, the ferrous iron transporter Sit, and the chromosome-encoded siderophore enterobactin. These systems were investigated for their roles in the establishment of a systemic infection in 3 week-old chickens. Bacteria were recovered from blood, lungs, liver and spleen and lesions on liver, airsacs and pericardium were scored. Single mutations of these iron acquisition systems caused some attenuation in virulence. Moreover, double-mutants lacking aerobactin / salmochelins and enterobactin / aerobactin were avirulent in chickens.



50.

**Bacterial adhesins and colonization of respiratory tissues by avian pathogenic
Escherichia coli (APEC)**

**Charles M. Dozois, Maria H. Lymberopoulos, Maryvonne Moulin-Schouler,
and Roy Curtiss III**

University of Quebec, INRS-Institut Armand-Frappier
531 boul. des Prairies
Laval, Quebec H7V1B7, CANADA

APEC cause respiratory and systemic disease in poultry. APEC strains produce a number of different adhesins including Type 1 fimbriae, curli, and the temperature sensitive hemagglutinin (Tsh). These adhesins and a newly identified Iron-regulated fimbriae, Irf, were investigated for their role in colonization of respiratory tissues of experimentally infected chickens. The individual or combined loss of Type 1 fimbriae, curli, and Tsh had no significant effect on the capacity of APEC strain chi7122 (O78:K80:H9) to colonize respiratory tissues. However, a mutant lacking Irf was less able to colonize the air sacs, but demonstrated improved colonization of the trachea.

51.

**Effects of a dietary yeast extract on the response to transport stress of turkey
poults previously challenged with *Escherichia coli***

**Geraldine Huff, William E. Huff, Narayan C. Rath, Morgan B. Farnell, Fausto Solis de los
Santos, and Anne M. Donoghue**

USDA/ARS
POSC O-303, University of Arkansas, Fayetteville, AR 72701

A yeast extract antibiotic alternative, Alphamune™, was used to supplement turkey poult diets at two levels, 1lb/ton or 2 lb/ton. Male Hybrid Converter poults were challenged by airsac injection with 60 cfu of *E. coli* at 1 wk of age. At 3 wk of age challenged birds were subjected to transport stress. Birds were bled and necropsied 12 hours after transport stress. Stress decreased body weights and gain which were protected by both levels of supplementation. The heterophil/lymphocyte ratio was increased by stress and this increase was prevented by both levels of supplementation.



52.

Distribution of Plasmids Among Avian Pathogenic *Escherichia coli*

Sara J. Johnson, Timothy J. Johnson, and Lisa K. Nolan
Department of Veterinary Microbiology and Preventive Medicine,
Iowa State University
1802 Elwood Drive, VMRI #2, Ames, IA, 50011

Virulence plasmids are a defining characteristic of the avian pathogenic *Escherichia coli* (APEC) pathotype. Additionally, R plasmids encoding multidrug resistance often co-transfer with these virulence plasmids. In an effort to further characterize the plasmids that occur among APEC, 541 isolates were screened for 16 different plasmid replicon types using multiplex PCR. Results, presented in this study, indicate that the RepFIB plasmid replicon, which typifies APEC virulence plasmids, was very common among the APEC examined. Additionally, isolates possessing the RepN, RepP, or RepI1 replicons, characteristic of R plasmids, were frequently found in these isolates.

53.

Exploring the Evolution of APEC Plasmids through Comparative Genomics

Timothy J. Johnson* and Lisa K. Nolan

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

Plasmid-associated virulence genes are a defining characteristic of the avian pathogenic *Escherichia coli* (APEC) pathotype. In APEC, such genes are often linked to ColV plasmids. We recently completed the first sequence of a ColV plasmid, and it was found to contain a cluster of virulence genes associated with the ColV operon. However, gene prevalence studies revealed that this cluster was not always associated with the ColV operon. Further studies have shown that this cluster also occurs on ColBM plasmids. Consequently, we have generated the first complete sequence of a ColBM plasmid. In this study, these plasmids are compared to provide insight into the evolution of APEC virulence.



54.

A Novel Virulence Marker 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

We have recently described a pathogenicity island, PAI I_{APEC-01}, in an avian pathogenic *Escherichia coli* (APEC) isolate that contains the *tia* gene among other genes. We hypothesized that *tia* is a virulence marker of APEC that is present predominantly in APEC rather than in fecal commensal *E. coli* of health poultry (AFEC). 500 APEC and 175 AFEC were screened for the presence of *tia* by PCR. 39% of APEC were positive for *tia* while only 7% of AFEC isolates carried the gene. This study indicates that *tia* can be used in a PCR-based diagnostic assay to augment detection of APEC from clinical specimens.

55.

Molecular characterization of avian pathogenic *E. coli* (APEC) in Korea

Sun-Joong Kim, Tae-Eun Kim, Sun-Hee Cho, and Hyuk-Joon Kwon

Laboratory of Avian Diseases, College of Veterinary Medicine, Seoul National University (151-744) San 56-1 Shillim-9-dong Kwanag-gu, Seoul, Republic of Korea

We characterized virulence genes (*fimC*, *fyuA*, *irp2*, *tsh*, *iucD*, *iss*, LT and ST) and class 1 integron in 121 isolates of APEC from diseased chickens in Korea between 1985 and 2005. The most frequent virulence gene was *fimC* (111/121, 91.7%) followed by *iss* (82/107, 76.6%), *iucD* (86/121, 71.1%), *tsh* (76/121, 62.8%), *fyuA* (57/121, 47.1%) and *irp2* (56/121, 46.3%). All virulence-gene-positive isolates were dominant (30/107, 28.0%) followed by *tsh-iucD-fimC-iss* positive (22/107, 20.6%) and *fimC-iss* positive isolates (14/107, 13.1%). The first class 1 integron was detected in 1985 and the size of gene cassettes was about 1.0kb. Various sizes of gene cassettes (about 1.0 to 3.0kb) were detected in 44.6% (54/121) of APEC.



56.

Detection of Iss Protein on the Outer Membrane of an Avian Pathogenic *E. coli* Isolate

Lynne, A.M., Logue, C.M, and Nolan, L.K.

Iowa State University, Dept. of Veterinary Microbiology & Preventive Medicine, 1802 Elwood Drive, VMRI #2, Ames, IA 50011

iss has a strong association with avian pathogenic *Escherichia coli* (APEC) virulence. *iss*, thought to be derived from lambda *bor*, encodes Iss, an 11-kDa protein. Here, *iss* and *bor* knockout mutants were created to assess the specificity of anti-Iss monoclonal antibodies (Mabs). These Mabs detected Iss and Bor in outer membrane protein (OMP) preparations of wild-type *E. coli* but not in the mutants. Use of these Mabs in immunofluorescent microscopy also distinguished between wild-type and mutant strains. This report demonstrates that Iss and Bor are located in the outer membrane and shows that our anti-Iss Mabs cross-react with Bor protein.

57.

Analysis of the Avian Pathogenic *Escherichia coli* O78:K80:H9 Virulon

Suman Mukhopadhyay and Chris D. Herren

University of Maryland, 8075 Greenmead Drive, College Park, MD 20742

Avian pathogenic *Escherichia coli* (APEC) cost considerable damage to the poultry industry. The regimen of APEC virulent, has yet to be identified and the APEC genome has not been sequenced. A complex regulatory network exists in APEC that senses the surrounding environment and activates genes that are required for infection of poultry. The BarA-UvrY two component system controls expression of some virulence factors. Preliminary data in our lab shows that mutations in *barA*, *uvrY*, and *barA-uvrY* affect critical aspects of APEC O78:K80:H9 infection including adherence, colonization, and mortality.



58.

Emergence of Virulent *Escherichia coli* Isolates in Poultry Production

Lisa K. Nolan*, Timothy J. Johnson, and Yvonne Wannemuehler

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

Large virulence plasmids appear to typify the extant APEC pathotype. Curiously, this does not appear to be true of the APEC isolated from previous decades. APEC isolated in the 1970s have relatively few large plasmids and plasmid-associated virulence genes, as compared to recent isolates. Moreover, APEC of the past are classified into the “commensal” phylotypes, while more recent isolates are classified into the “pathogenic” phylotypes. These results suggest that a more virulent type of APEC is emerging among poultry, which may mean that management strategies that have controlled colibacillosis in the past may not prove as effective in the future.

59.

**Relationship between Host Age and Virulence Capacity of Avian Pathogenic
Escherichia coli (APEC)**

**Jennifer A. Scaccianoce*, Yvonne M. Wannemuehler, Timothy J. Johnson,
and Lisa K. Nolan**

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

It is commonly assumed that the APEC of older birds may be more virulent than the ones of young hosts. To test this assumption, 133 APEC isolates from young and old host birds with colibacillosis have been subjected to phylotyping and genotyping. Phylotyping classifies *E. coli* isolates into one of four groups, two of which are thought to contain pathogens and the other opportunists. Genotyping studies involve about 100 different genes thought to contribute to APEC’s disease causing ability. Results of phylotyping and genotyping of the APEC from young and old hosts will be presented.



60.

**Characterization of *E. coli* Isolates from Peritonitis Lesions
in Commercial Laying Hens**

Darrell W. Trampel, Lisa K. Nolan, and Yvonne Wannemuehler
Iowa State University
1802 Elwood Drive
VMRI – Building 1
Ames, Iowa 50011-1240

A study was conducted in order to isolate, identify, and characterize *E. coli* strains from the cloaca, oviduct, and peritoneum of healthy laying hens and those with peritonitis. Five clinically normal chickens and ten dead chickens with lesions of peritonitis were examined from each of three commercial egg-laying operations. Culture swabs were taken by aseptic procedures from each anatomical location. *E. coli* isolates were characterized by O serogrouping, virulence genotyping, and phylogenetic grouping. *E. coli* isolates from the cloaca, oviduct, and peritoneum were identical in dead hens within the same flock and different from those found in healthy chickens.

61.

**Distribution of Plasmid-Mediated Resistance and Resistance Genes Among Avian
*Escherichia coli***

Yvonne M. Wannemuehler, Timothy J. Johnson, and Lisa K. Nolan
Department of Veterinary Microbiology and Preventive Medicine, Iowa State University
1802 Elwood Drive, VMRI #2, Ames, IA, 50011

Recently, we described the first sequence of an avian pathogenic *Escherichia coli* resistance (R) plasmid, pAPEC-O2-R. Such plasmids encode resistance to multiple antibiotics, disinfectants, and heavy metals. Here, we compare 451 APEC and 105 commensal *E. coli* of poultry for their possession of the resistance genes found on pAPEC-O2-R. Additionally, we assessed these isolates for plasmid-associated resistances. Our results indicate that APEC possess significantly more plasmid-associated resistances and resistance genes than found in the commensals. Though the prevalence of the resistance genes are lower in commensals, a number of the commensal strains are resistant, suggesting that they may be important reservoirs of resistance genes for pathogenic strains.

62.

Histopathological Effects of Intravenous Injections of *E. coli* on the Bone Marrow in Broilers Including Quantitative Histomorphometric Changes

Floyd D. Wilson, Timothy S. Cummings, and Mark A. Burluson

MVRDL & Poultry Laboratory,
CVM, Mississippi State University
2531 N. West Street
Jackson, MS 39216

Bone marrow changes on post-injection days 1, 3 and 6 in response to intravenous injections of a pathogenic *E. coli* were evaluated using routine histological and quantitative histomorphometric techniques. Development of foci of bone marrow hyperplasia comprised of blastic or primitive heterophilic progenitors were present on day 1. Over extended time the hyperplastic foci increased in size and maturity resulting in hyperplastic marrows consisting mainly of mature heterophils. Thus, the marrows by six days were similar to those previously observed for broilers manifesting with naturally occurring systemic bacterial infections. Possible applications of the studies to slaughter disposition will be discussed.

GENERAL DISEASES

63.

Performance of broilers chickens raise in reuse litter vs new litter

**Mónica Alba, Eliana Icochea Pablo Reyna, Rosa Gonzalez, Branko Alva
and Pilar Vejarano**

School of Veterinary Medicine, University of San Marcos Lima - Las Esmeraldas 1009 Coop.
Huancayo, Lima 10 – Perú

This study was to evaluate the performance and sanitary status of broilers raised in new litter and reuse litter. 250 male broilers from the Ross 308 line were raised as follows: 125 in new and 125 in reused litter during 5 flocks. Performance data was collected weekly, as well as ammonia levels and oocysts count per gram of litter. The levels of antibodies against Infectious Bronchitis (IB) Newcastle Disease (ND) Infectious Bursal Disease (IBD) Chicken Anemia virus (CAV) and Avian Reovirus were evaluated by ELISA test at the beginning and ending of the flock (day 49). The broilers raised in new litter obtained a feed conversion of 2.07 and an average body weight of 3.2 Kg, against 2.09 feed conversion and 3.87 body weight found in the second group. At the end of the study there was no statistic difference ($P < 0.05$) between the performance parameters for both groups. The ammonia levels and oocyst count were higher during the first weeks in the reused litter group; nevertheless, there was no statistic difference. The serology results did not show any seroconversion to the virus tested. Upon this, it will be expected that the litter coming from poultry farms without a history of infectious problems and with an adequate handling of environmental conditions of the farm can be safely reused for up to flocks.

64.

Utility of Molecular Diagnostic Tests in Timely and Accurate Avian Disease Diagnosis

Daniel A. Bautista, DVM, MS
Maryland Department of Agriculture
Salisbury Animal Health Diagnostic Laboratory
PO Box 2599, Salisbury MD 21802

We present cases that underline the role of molecular diagnostic tests in disease diagnosis when faced with inadequate samples, uncommon or mixed clinical presentations and lesions, and the need for fast turn-around time. The first is a Newcastle Disease RRT-PCR diagnosis in layers from eggs with a high incidence of internal calcium carbonate deposits. The second is ILT, IBV, MG/MS, and blackhead-related mortality in auction market-purchased poultry. RRT-PCR ruled out AI and NDV, and ruled in IBV, ILT after 3 hours, and MG/MS shortly afterwards. The third is a WNV outbreak in Impeyan pheasants/Himalayan monals diagnosed by RRT-PCR.

65.

Laboratory evaluation of the survivability of H7N2 Avian Influenza and Newcastle Disease Virus to commercial disinfectants

Eric R. Benson, Robert L. Alphin, Brian S. Ladman, George W. Malone, Michael D. Dawson, and Mega E. Lombardi

University of Delaware, 242 Townsend Hall
531 South College Avenue, Newark, DE 19716

Current methods of avian influenza (AI) control in poultry require euthanasia of flocks known or suspected to be infected. Environmental concerns have made disposal of AIV infected carcasses a major issue in many poultry-producing areas. Having a rapid, practical, cost-effective and biosecure means of inactivating AIV within the facility, litter, carcasses and on support equipment, is essential in containing disease outbreaks. The decontamination of poultry houses and equipment will be evaluated in a four-phase process.

The objective of phase one of the study (the portion presented herein) was to test the efficacy of five commercially available disinfectants on low pathology avian influenza (LPAI) and Newcastle disease virus (NDV). The viruses were tested on four materials typically found in poultry houses (glass, plastic, wood, and galvanized steel). The samples were separated and inoculated individually with A/chicken/MD/MINH MA/03(H7N2) and LaSota strain of NDV virus and allowed to dry at room temperature. Separate samples were exposed to liquid applied Virkon S, foam applied Virkon S, foam applied DF200, liquid applied BioSentry 904 and fog applied Virocid. Solution containing the decontaminated virus was then collected and inoculated into 10-11 day-old SPF embryos that were incubated and candled daily for five days. The embryos were refrigerated and fluid was tested for hemagglutination activity (HA). HA and positive control activity were used to indicate successful disinfection.

Most disinfectants had similar neutralization activity with the viral agents tested on both glass and plastic. Differences in neutralization activity were observed on both wood and metal surfaces, both of which are typically found in poultry houses. The results will be used to select the agents for the second phase, which will include disinfecting colony houses.



66.

Hurricane Katrina's Effect on the Poultry Industry in Mississippi

Mark A. Burleson, Danny Magee, Sue Ann Hubbard, and Phil Stayer
Mississippi State University
302 E Gillespie St
Starkville, MS 39759

This poster attempts to summarize the effect of Hurricane Katrina on the poultry industry in Mississippi. While mentioning the short-term and long-term responses of the industry, emphasis will be placed on the lessons learned, from the approach of the storm to the days following the storm. Different hardships encountered by the industry will also be discussed. This poster will serve as an adjunct to the oral presentations that will be given on this topic, while highlighting topics that were not discussed.

67.

A Serological Study of One Company's Vaccination Program

Mark A. Burleson, Phil Stayer, and Tim Cummings
Mississippi State University/Sanderson Farms, Inc.
302 E Gillespie St
Starkville, MS 39759

Three years of data was gathered to study the history of a company's serological titers in different geographical locations and to compare the data with conventional vaccination programs. The program in study involves a one-time killed IBDV and Reovirus vaccination at 15 weeks of age, as well as a complete live IBV and NDV vaccination program. Geographical and seasonal changes were noted. IBDV and Reovirus titers were comparable with comparison titers, while IBV and NDV were lower. The study shows the serological titers of this company's vaccination program and the decisions that will be made based on this data.



68.

Incidence of Subclinical Diseases and Pathological Conditions in Clinically Normal Broilers from 3 Production Complexes by Sex and Age

Hector M. Cervantes
Phibro Animal Health Corp.
1031 Westchester Court
Watkinsville GA 30677

Necropsy records from a total of 47 routine broiler chicken necropsy sessions, involving a total of 2,805 broiler chickens, from 3 broiler complexes in the southeastern United States were analyzed to determine incidence of subclinical diseases and pathological conditions in regards to the sex and age of the birds. The number of necropsy sessions per broiler complex ranged from 12 to 19, the data was analyzed by bird sex within each age group. The age groups were as follows: 1-21 days; 22-35 days; 36-42 days and >42 days. This presentation will summarize the findings and will discuss possible explanations for any differences found between sexes and age groups.

69.

Mixed Tumor In The Right Oviduct Of A White Leghorn

Portia L. Cortes, Rocio M. Crespo, H. J. Barnes, and H. L. Shivaprasad
California Animal Health and Food Safety Laboratory,
Fresno Branch, University of California, Davis
2789 South Orange Avenue
Fresno, CA93725

This case report intends to highlight the histological lesions observed in this unusual presentation of carcinoma in the right oviduct of a 2-year-old White Leghorn raised as a 4-H project. The bird was submitted due to a large tumorous growth in the neck area. On post-mortem examination, a second mass, measuring 5-6 cm in diameter was observed in the right oviduct. On histopathology, a neoplasia of well-differentiated squamous epithelial cells and glandular cells were observed pallisading in a concentric manner with very few inflammatory cells. Immunohistochemistry and other characteristics of the neoplasia will be described.

70.

Histologic Lesions in the Proventriculus of Broilers and Broiler Breeders

Oscar J. Fletcher, James S. Guy, and H. John Barnes

Poultry Health Group, Department of Population Health and Pathobiology, College of Veterinary Medicine, NC State University, Raleigh, NC 27606

Lesions of transmissible viral proventriculitis (TVP) as described by Goodwin *et al* (1996) were reproduced by inoculation of an adenovirus-like virus (Guy, *et al*. 2005). Samples from these experimental studies serve as comparisons with lesions found in the proventriculus from broilers and broiler breeders submitted to diagnostic laboratories in NC and GA in June 2005 through October 2005.

Proventriculus was available for microscopic examination in 58 of 243 cases involving broilers or broiler breeders and lesions were found in 24 of the 58 cases. Histopathology confirmed TVP in 9 cases. The relationships between clinical and microscopic evaluations are presented in Table 1.

Table 1: Distribution of cases based on clinical diagnosis with microscopic evaluation for confirmation of TVP.

Clinical Dx	Microscopic			Total Cases
	TVP Confirmed	Other Lesions	No Dx Lesions	
TVP Yes	3	4	3	10
TVP No	6	11	31	48
Total	9	15	34	58

These observations support the conclusion that TVP can not be diagnosed by clinical signs and gross necropsy evaluation.

Proventricular lesions considered diagnostic for TVP are necrosis of glandular epithelium, lymphocytic inflammation, hyperplasia of ductal epithelium, and replacement of lost glandular epithelium by the hyperplastic ductal epithelium.

Other lesions, not diagnostic for TVP, consisted of focal necrosis of the mucosa, submucosal edema, dilation of glands, mild to moderate ductal hyperplasia, variable degrees of sub-ductal fibrosis, and serosal inflammation. Many sections contained some lymphoid foci in the glandular area and this finding is considered normal.



71.

Teaching Poultry Disease and the Importance of Poultry in Afghanistan

Richard M. Fulton and Robert M. Smith

Diagnostic Center for Population and Animal Health, Michigan State University,
PO Box 30076, Lansing, MI 48909-7576

In the reconstruction of Afghanistan, the United States Department of Agriculture invited me to evaluate the pathology instructional program and facilities at the veterinary school of Kabul University in Afghanistan. While there, veterinary students and veterinarians and poultry service workers that are part of the United Nations' Food and Agriculture Organization Village Chicken Program were taught diseases of poultry. The village chicken program trains illiterate people in small villages to raise chickens and collect eggs which were sold in Kabul. Through this program, the FAO has been able to provide food and a source of income for poor people in isolated villages as well as spark a desire to learn to read and write.

72.

Use of Composting for Disposing Dead Poultry in Venezuela in Normal and Emergency Situations

Luis B. Gomez and Sonia Puche

Agropecuaria Ls2000 S.C.
Calle Vargas #17 El Limon
Maracay, Aragua 2101, Venezuela

There are only two approved methods of disposing dead poultry in Venezuela. Under current conditions of increased production and productivity plus the increase challenge of producing disease-free poultry and other consumer concerns, the use of techniques that will handle dead birds in an efficient way without harming the user, the environment and that the final product will be economically beneficial is highly desirable. Composting is a viable alternative for all parties involved and the use of the final product as a fertilizer and possible use as an additive for ruminants has been proposed and are explained in this study.

73.

Efficiency of an Aluminosilicato in the Ammonia Control in Litter of Broiler Chickens

**Eliana Icochea¹, Pablo Reina¹, Jhon Guzmán¹, Mónica Alba¹, Rosa González¹,
Anthony Berríos²**

1. School of Veterinary Medicine, San Marcos University, Lima-Perú

2. SUD - CHEMIE Peru' S.A. Laboratories

The objective of this study was to evaluate the effectivity of the litter treatment with an aluminosilicate from peruvian resources. The study was carried out using the physical facilities of the Vet College at San Marcos University, Lima - Peru, from June to July, 2005. One day-old 400 broilers ROSS 308 (200 males and 200 females) were divided in two groups of 200 animals as hatched each one. Group A, raised on litter pen chemically treated twice. In the first treatment was applied 0,55 kg per m² three days before the reception of the birds, and in the scond treatment was used 0,45 kg. of the product per m². at 35 days of age. Group B was raised on litter pen without treatment (Control)

Were registered Ammonia levels and pH of litter pen, Humidity and litter pen caking, Pectoral and plantar lesions. Shank pigmentation and Productive parameters. The quantitative variables as body weight, European Efficiency Index (EEL) and Feed Conversion (FC) were analyzed at 44 days of age by Student Test,

The Pectoral and plantar lesions by Kruskall wallis test.

74.

The Effect of Two Broiler Catching Techniques on Wing and Leg Damage

**Kelli H. Jones^A, Stephen R. Collett^A, Suzanne D. Young^A, Marilynn Finklin^A, and
Benjamin C. Johnson^B**

^ADepartment of Avian Medicine, College of Veterinary Medicine, The University of Georgia, Athens, GA 30602-4875

^BGold Kist Inc., 244 Perimeter Center Pkwy., N.E., Atlanta, GA 30346-2397

Carcass downgrades, as a consequence of wing and leg damage have increased in parallel with slaughter weights resulting in meat loss and concern for bird welfare during the catching process. Traditional, one-leg catching method was compared with a two-leg catching method in both small (4 lb) and large (7 lb) birds in commercial poultry operations. While two-leg catching reduced the prevalence of wing and leg injuries in 4lb birds, it increased the prevalence of these injuries in 7lb birds. In addition, two-leg catching took 2-3 times longer than one-leg catching regardless of bird size.



75.

Pigeon Disease in Georgia 1996-2006

Donna J. Kelly

Georgia Poultry Laboratory Network
PO Box 6025
Forsyth, GA 31029

Pigeon accessions to the Georgia Poultry Laboratory Network for the past ten years will be summarized. The summary will include the most common diagnoses and disease trends. These diagnoses will be further subdivided into production type: racing, breeder, exhibition, wildlife, research. Common practices that perpetuate and exacerbate these diseases will also be discussed.

76.

Intestinal Intussusception in 11 wk-old Broiler Breeder Pullets

Jose A. Linares, DVM, ACPV

Texas Veterinary Medical Diagnostic Laboratory

Ten 11 wk-old broiler breeder pullets were submitted to the diagnostic laboratory from a flock with increased mortality. All ten chickens had severe intestinal intussusception, seven had intestinal prolapse, two had lesions compatible with necrotic enteritis and two had coccidiosis based on intestinal scrapings. Intestinal intussusception was the primary cause of mortality. Intestinal intussusception is caused by a mix of factors such as feed management, coccidiosis and necrotic enteritis. The fact that previous cases of intussusception, with lower incidence, had been documented at the same premise suggests an increased intestinal challenge.

77.

Acidification of Drinking Water: An Overview

Trisha Marsh Johnson, DVM, MAM

125 Fox Trace
Athens, GA 30606

The usage of water acidification as a preventive or treatment tool for disease management or bird performance is probably one of the most poorly understood areas of clinical poultry medicine. This stems partly from the fact that until recently, no controlled research had been done investigating the preferred pH for poultry water consumption. This paper will summarize the available data on water acidification and its use in poultry management, the different types of water delivered acids and their appropriate use, and explore plausible explanations for the disease prevention and performance responses documented in recent research.



78.

Therapeutic and Prophylactic Anticoccidial Sensitivity of Coccivac

Greg F. Mathis

Southern Poultry Research, Inc.
2011 Brock Road, Athens, GA 30607

Coccivac is a commercial chicken coccidial vaccine comprised of *Eimeria acervulina*, *E. maxima*, *E. mivati*, and *E. tenella*. All strains of *Eimeria* used in this vaccine were isolated prior to the introduction of the major anticoccidials used today. Using traditional anticoccidial sensitivity battery testing procedures, the anticoccidial sensitivity was determined for sulfadimethoxine, amprolium, diclazuril, monensin, salinomycin and nicarbazine. The challenge caused an approximate 45 % weight depression and 2.5 to 3.5 regional lesion score in the NMI birds. All medicated challenged birds had less than 5 % weight depression and less than 1.0 lesion score.

79.

Intestinal Changes Associated with Feed Deprivation and Recovery from Feed Deprivation in Laying Hens

Randle W Moore, Peter S Holt, Deana R Jones

USDA, ARS, Egg Safety & Quality Research Unit, Russell Research Center
950 College Station Road, Athens, GA 30605

The effect of progressive feed deprivation and recovery from feed deprivation on the Leghorn hen intestines was observed. Hens were divided into two groups, full fed (FF) and feed deprived (FD). In trial 1, washed intestinal (proventricular-duodenal junction to vent) samples were obtained from hens on 1, 4, 6, 12 days of feed withdrawal (FW) and 1, 4 days of return of feed (RF). In trial 2, washed intestinal samples and individual measurements of crops, duodena, jejunum, ilea, and ceca were obtained at 2.5, 8 hr, and 1, 2, 3, 7, 10, 14 days FW and at 6 hr, and 1, 2, 5 days RF. In trial 1, FD intestinal lengths and weights were decreased by 24 hr and 4 days FW, respectively, and remained decreased for the duration of FW. Intestinal lengths and weights returned to FF levels by 24 hr RF. In trial 2, weight and length of intestine and intestinal sections were decreased from FF by 8 hr and 48 hr FW, respectively, and remained decreased at almost every time point for the duration of FW. The weights and lengths of intestines and intestinal sections of FD returned to FF levels by 24 hr and 6 hr RF, respectively. The percent dry weight of FD intestinal sections were mostly unaltered during FW, but decreased for at least the initial 48 hr RF. FD proventricular and ileal pH decrease and crop, duodenal, and jejunal pH increased with FW, and most returned to FF levels by 48 hrs RF.



80.

Addressing Poultry Health Needs of Private Veterinary Practitioners

Teresa Y. Morishita and Meredith F. Davis

Department of Veterinary Preventive Medicine, The Ohio State University
1920 Coffey Road; Columbus, Ohio 43221

Poultry medicine is a specialized area in the veterinary profession and often requires more advanced studies and/or field experiences in poultry medicine. Veterinarians in private clinical practices are sometimes presented chickens and other poultry species as patients. This can cause some anxiety as poultry medicine is often not a required course in veterinary school. Veterinary extension units in land grant universities are often requested information on poultry health topics. We present here a study on the informational topics requested by veterinarians in clinical practice. Our results indicate that there is a need to expand the written resources on poultry medicine.

81.

**Interfering Effect Of Multiple Poultry Agents On Same FTA[®] Cards
On The Detection Of Poultry Pathogens**

Hugo Moscoso, Gwen Brown and Charles L. Hofacre.

Poultry Diagnostic and Research Center. Department of Population Health.
The University of Georgia.

In recent years we have adapted molecular techniques for the identification and characterization of avian pathogens in specimens collected on FTA[®] cards. We have shown that viruses and bacteria are rendered non-infectious upon contact with the FTA[®] thus allowing the import of samples from overseas in compliance with federal regulations. The number of tests requested by clients from foreign countries has increased 10 fold in the past 2 years and we anticipate that this trend will continue. In addition, we believe that request for multiple tests i.e. infectious bronchitis (IBV), Newcastle disease virus (NDV), and Mycoplasma from one FTA[®]/specimen i.e. tracheal impressions will also increase in the near future as sampling is facilitated and transportation cost is reduced using the FTA[®] cards.

Artificial mixes of avian viruses and mycoplasma were spotted on FTA[®] cards as 5 ul aliquots and stored at 25 C for 24 h. FTA[®] samples were processed for DNA or RNA to perform PCR or RT-PCR respectively. Detection/reactivity of viral DNA or RNA or bacterial DNA was qualitatively determined by the presence or absence of specific amplicon bands or by visual intensity of the bands on agarose gels.

Infectious laryngotracheitis (ILT), Mycoplasma gallisepticum (MG), and Mycoplasma synoviae (MS) were readily detected on FTA[®] inoculated with mixes containing any combination of 2 or 3 of these organisms. In addition the presence of RNA viruses (IBV or NDV) did not interfere with the detection of ILT, MG or MS. On the other hand, the detection of NDV was totally inhibited by concomitant presence of IBV or ILT and partially inhibited by MG or MS while the detection of IBV was completely suppressed by any of these pathogens spotted in the same FTA[®].

These preliminary results indicate that DNA from multiple organisms can be detected on the same FTA[®] sample by PCR but the detection of viral RNA by RT-PCR may not be possible in the presence of other DNA or RNA organisms spotted on the same FTA[®] card.



82.

Prevalence of Antimicrobial Resistance in Migratory Passerines

Crystal D. Newcomer-Decker, B.S. and Teresa Y. Morishita, DVM, PhD

The Ohio State University, Department of Veterinary Preventive Medicine

2550 Farrel Dr., #219, Columbus, OH 43235

The prevalence of intestinal bacteria and parasites was studied in a population of spring migratory passerines in 2004. Out of 192 birds sampled, 112 had a bacteria recovered from sampling. For the passerines with bacteria, antimicrobial resistance will be tested. It will be determined if these wild birds will have a resistance to certain antibiotics or antibiotic groups. Prevalence of antimicrobial resistance can be a good indicator of the types of antibiotics these birds have been exposure to in nature.

83.

A case of acute intoxication with carbofuran in ducks

Daniela Nica and Elisabetha G. Bianu

Institute for Diagnosis and Animal Health

Str. Dr. Staicovici Nr. 63, Sector 5, Cod 050557

Bucharest, Romania

In a morning the owner of an individual farm found 12 of his ducks dead. He observed that near the hedge inside his yard there were a lot of corn grains coloured in red. To establish the cause of the death he sent seven birds and some crop for toxicological investigations. The pathological exam showed no characteristic post mortem lessions, only congestion and edema of lungs and liver. Using thin layer cromathography and gas cromatograph – mass spectrometer technique we identified carbofuran in gizzard content, so the death of the birds was an acute intoxication with this carbamate.



84.

Biomechanical Factors that Influence Femoral Spiral Fractures of Turkeys

**Edgar O. Oviedo, Peter R. Ferket, H. John Barnes, Diego V. Bohórquez,
and Jesse L. Grimes**

North Carolina State University, Department of Poultry Science,
Scott Hall O-239, Raleigh, NC 27695-7608

The average incidence of skeletal problems in turkeys is about 2-6%, but it may exceed 15% in some flocks. Spiral femoral fractures (SFF) are common in rapidly growing tom flocks around 15 to 20 weeks of age and may account for 5% mortality, costing the US turkey industry over \$150 million per year. We hypothesized that SFF is due to biomechanical stress. Samples of turkeys raised in experimental conditions and obtained from field cases of SFF were evaluated for structural and biomechanical properties. A model formulated to explain SFF based on body conformation, skeletal balance, and bone strength will be presented.

85.

Litter Impaction of the Lower Intestinal Tract of Broiler-Breeders

Kristen Roza, Michael P. Martin, and H. John Barnes

North Carolina State University

4700 Hillsborough St.

Raleigh, NC 27606

Clinical case presentation: Increased mortality occurred in 23-25 week old male broiler breeders. On necropsy, the birds were found to have had a severe impaction of litter in the lower intestinal tract. This case was unique in that lower intestinal tract impaction of adult chickens has not, to the authors' knowledge, been previously reported. The condition was believed to be due to feeder access changes, leading to an inability to find feed and increased feed competition between male birds. The affected birds appear to have starved out and eaten litter.



86.

Dermal Squamous Cell Carcinoma in a Six-month-old New Hampshire Red Chicken

Craig F. Sarver and Teresa Y. Morishita

Ohio Department of Agriculture, Animal Disease Diagnostic Laboratory
8995 E. Main Street, Reynoldsburg, OH 43068

Multiple dermal squamous cell carcinomas have been reported in the carcasses of young broiler hens at the time of slaughter. Squamous cell carcinomas have been reported rarely in older birds. A six month old, New Hampshire Red hen was presented for necropsy examination for "sores on back and neck." Four, raised and thickened nodules were noted on the neck (1), chest (2) and back (1) of the bird. The lesions involved the feather shafts and extended into the subcutaneous tissues. Histopathological examination revealed multiple squamous cell carcinomas. Bacteriological and virological studies did not detect any underlying agents.

87.

Resident Canada Geese: Vectors of Disease

**Jordan Schaul, Lori Martin, Amna El Tayeb, Teresa Morishita, Peter Kobalka,
and Walt Threlfall**

Laboratory for Wildlife & Environmental Health
Department of Veterinary Preventive Medicine
College of Veterinary Medicine, The Ohio State University
1920 Coffey Road, Columbus, OH 43212

Given widespread interest in an avian flu pandemic, there is a need for avian veterinarians to educate allied health professionals on measures for epizootic preparedness. Nuisance Canada Geese populations are on the rise. As natural vectors for zoonotic diseases, it is important to disseminate information regarding Canada Geese biology, the interface between domestic avifauna and wild birds, as well as the pathobiology of the avian influenza virus and associated avian pathogens. The objective of our education initiative is to provide information to reduce the risk of disease transmission between humans and wild birds, particularly Canada geese.



88.

Development of Multiplexed Fluorometric Immunoassay for Poultry Diagnostics

Elena Seletskaja, Joe H. Simmons, Rajeev Dhawan, and William R. Shek
Charles River Laboratories, Diagnostic Laboratory Research and Development
251 Ballardvale St.
Wilmington, MA 01887

Rapid and accurate detection of infectious agents in domestic poultry is of critical diagnostic importance. We have developed a multiplexed serologic assay for nine avian viruses (infectious bronchitis, bursal disease, influenza, Newcastle's, adenovirus, fowl pox, reovirus, reticulendotheliosis, and paramyxovirus 2) using Luminex® xMAP® Technology. The multiplex also includes additional tissue and antibody controls to ensure optimum assay performance. The multiplexed avian infectious disease assay is highly sensitive and specific, and it has been validated by comparison to traditional serologic techniques including ELISA, IFA, and AGP. Ongoing research and development will allow us to include additional infectious agents in this multiplex.

89.

Severe Neuropathy in Broiler Breeder Pullets Associated with High Levels of Dietary Salt

**C. Gabriel Senties-Cué, Danny L. Magee, Floyd D. Wilson, Philip A. Stayer,
and William R. Maslin**
Poultry Research and Diagnostic Laboratory, College of Veterinary Medicine,
Mississippi State University
P.O. Box 97813, Pearl, MS 39288

A sudden increase in mortality occurred in a 17-week-old, broiler breeder pullet flock. At necropsy, the most remarkable lesions were oral and gizzard erosions and darkened kidneys. No obvious gross lesions were observed in the brain. Histopathology revealed extensive mononuclear perivascular cuffing and severe degenerative changes in the brain. High levels of salt were measured in the diet. Histopathological findings and differential diagnosis will be discussed.



90.

The use of oxydative reduction potential (ORP) as a measure of the effect of water sanitisers on Gumboro vaccine

Amer Silim, Daniel Venne, Younes Chorfi, and Dianne Frenette

University of Montreal

Faculte de Medicine Veterinaire

Université de Montreal, 3200 Rue Sicotte, Saint Hyacinthe, Quebec

Canada, J2S 7C6

This study was undertaken to verify the effect of ORP in water used in the dilution of Gumboreo vaccines administered in drinking water. Municipal tap water and three commercial bottled waters were used in the experiment. Each water was used as is and at an adjusted pH of 6 and 8. The waters were then treated with chlorine or were used untreated. The ORP was measured before adding a known quantity of virus, followed by incubation at room temperature for one or two hrs. Each virus treatment was titrated in chicken embryo fibriblast culture and readings were made after five days of incubation

91.

Serologic evidence of five poultry pathogens in free-ranging chickens in Grenada

R. N. Sharma, M. I. Bhaiyat, S. Tawde and C. N. I. Macpherson

Department of Paraclinical studies, School of Veterinary Medicine, St. George's University, Grenada, West Indies

Sera samples from 92 free-ranging chickens in Grenada, were tested for the presence of antibodies against five poultry pathogens (Newcastle disease virus [NDV], infectious bronchitis virus [IBV], Mycoplasma gallisepticum/Mycoplasma synoviae [MG-MS], Pasteurella multocida [PM] and Salmonella enteritidis [SE] using a commercial enzyme linked immunosorbent assay (ELISA) kit. The survey revealed seropositivity of 99% for NDV, 71% for IB, 78% for MG-MS, 35% for PM and 45% for SE. The presence of antibodies for more than one disease agent in the same chicken indicates that several infections circulate within the same premises. Chickens were between 12 and 24 months of age. This is the first report on disease survellance in the chicken population in Grenada.



92.

A Comparison of Two Customer Requested and One Industry Derived Lighting Program

P. A. Stayer, J. Paul Thaxton, Martha L. Ewing, and John Rice

Sanderson Farms, Inc.

P.O. Box 988

225 North 13th Ave.

Laurel, MS 39441

By customer request, two brighter light intensity programs were compared to one company's corporate lighting program for production and welfare effects. The tests involved increased light intensity to 0.3 foot-candles for 8 hours each day after 23 days of age: one continuous program, the other was split. Production parameters showed a growth benefit, with either test program, but feed conversion and cost were adversely affected. Non-production measures, such as gait scores and corticosterone levels, indicated increased stress with brighter lights. Both test lighting programs appeared to adversely affect broiler well-being when compared to the corporate lighting program.

93.

Lactic Acid Fermentation of Poultry Carcasses Prior to Rendering

Nada M. Tamim, Rami A. Dalloul, Timothy A. Shellem, and John A. Doerr

University of Maryland

3404 Elenoir Court

Bowie, MD 20716

Lactic fermentation, one method for preserving poultry mortality on farm prior to rendering could be directly related to the state of carcass putrefaction. A series of experiments were conducted to study the influence of putrefaction on fermentation and biogenic amine accumulation. Carcasses putrefied for more than 18 hours yielded a product that did not always ferment properly and drop to expected pH levels. Furthermore, several biogenic amines were found to accumulate during putrefaction of broiler carcasses especially after 24 hours of putrefaction. These amines continued to accumulate during fermentation especially in putrefied carcasses and at high fermentation temperatures.



94.

The Harderian Gland a Mucosal Effector Site for Viral Infections

Frederik W. van Ginkel, Vicky L. van Santen, and Haroldo E. Toro

Department of Pathobiology, College of Veterinary Medicine,
Auburn University, 217 Scott Ritchey
Auburn, AL 36849

Mucosal immunity plays an important role in the defense against infectious bronchitis virus (IBV). To better understand the events that constitute a mucosal immune response in the Harderian glands, we developed IgA and IgM IBV-specific ELISPOT assays to directly measure the induction of IBV-specific IgM and IgA secreting B cells in Harderian glands and we also performed FACS analyses to measure T and B cell kinetics in this mucosal effector site, when challenged in the presence of immunosuppressive viruses CAV and/or IBDV, which are endemic in the poultry industry. We will present results from both virus-immunocompromized and normal chickens.

95.

Cutaneous aspergillosis in broiler breeder pullets

Francene Sophia Van Sambeek, Fred J. Hoerr, and Susan Lockaby

Alabama Department of Ag and Industry: Hinton Mitchem Poultry
Diagnostic Laboratory
266 COUNTY ROAD 699
CULLMAN, AL 35055

The case report will summarize the gross and histopathologic findings in several flocks of broiler breeder pullets submitted to the diagnostic lab for swellings on the heads. These smooth nodules were found on a percent of the young flock with little other signs of illness.

96.

Some Answers to the Causes of 'Femoral Head Necrosis'

Floyd D. Wilson, DVM, Phil A. Stayer, Lanny W. Pace, and Fred Muhammad

MVRDL & Poultry Laboratory, CVM. MSU

The cause[s] of separation of the femoral head during coxofemoral joint disarticulation at routine necropsy is controversial. We investigated the microscopic anatomy of femoral heads obtained from clinically normal 26-50 day-old broilers, but which either did or did not exhibit such detachment. Using histomorphometric methods we observed a quantifiable decrease in the chondrocyte density and increased pyknotic nuclei along the surface of the hyperplastic zone immediately adjacent to the tear site and an increase area of the vascular canals within the hypertrophic zone for affected femurs. We compare these results to other histopathological findings for poultry with clinical lameness.



POSTERS

Session 2 - Tuesday, July 19 and Wednesday, July 19 – 7:00 AM – 3:00 PM

IMMUNOLOGY, IMMUNITY, AND VACCINES

97.

Development of microsphere assays for the detection of antibodies against avian pathogens

Scott A. Callison, Tye O. Boynton, Deborah A. Hilt, and Mark W. Jackwood

The University of Georgia, College of Veterinary Medicine
PDRC, Department of Population Health
953 College Station Road, Athens, GA 30602-4875

Microsphere immunoassays utilize an antigen bound to a colored microsphere, which can be combined with other coated microspheres to create assays that detect antibodies to several disease agents in the same sample. Herein, we report the development of microsphere assays that can detect antibodies against several different avian pathogens. In addition, data using a triplex microsphere assay that concurrently detects specific antibodies against Newcastle disease virus, infectious bronchitis virus, and reovirus in the same sample will be presented.

98.

Investigating the Avian Macrophage Responses to Different *Eimeria* Species Using cDNA Microarray

Rami A. Dalloul, Hyun S. Lillehoj, Travis W. Bliss, Yeong H. Hong, Dong W. Park, and Calvin L. Keeler, Jr.

Animal Parasitic Diseases Laboratory
Animal & Natural Resources Institute, ARS, USDA
Bldg. 1040 / BARC-East, Beltsville, MD 20705

Coccidiosis is recognized as the major parasitic disease of poultry and is caused by the apicomplexan protozoa *Eimeria*. Increasing evidence shows the magnitude of complexity involved in host immune responses to *Eimeria* where microarray technology presents a powerful tool for the study of such intricate biological processes. Using avian macrophage microarray containing 4,906 unique gene elements obtained from chicken macrophage cDNA library and spotted in triplicates, we identified important host genes whose expression changed following infection of macrophages with sporozoites of *E. tenella*, *E. acervulina*, and *E. maxima*. This approach enabled us to identify common genetic immune-related elements whose transcriptional expression is induced or repressed by exposure to *Eimeria* sporozoites and to identify transcription patterns unique to each individual *Eimeria* species. Fundamental analysis of avian chemokine and cytokine expression patterns offers insight into the unique avian immunological responses to these related but biologically unique pathogens.



99.

Development and Use of an Avian Innate Immunity Microarray (AIIM)

Calvin L. Keeler, Jr., Hyun Lillehoj, Michael Kogut, Susan Lamont, and Travis Bliss
Dept. of Animal and Food Sciences, University of Delaware,
040 Townsend Hall, Newark, DE 19716-2150

Elements of the innate immune system, including macrophages, heterophils, dendritic cells, and natural killer cells are involved in destroying intracellular and extracellular pathogens and in providing instructions for the initialization of the proper acquired immune response. Recognition of evolutionarily conserved pathogen associated molecular patterns (PAMPs) are mediated by specific pattern recognition receptors (PRRs). Our laboratory has developed a cDNA microarray, containing ~5,000 elements, which is being used to characterize changes in the transcriptome of cells of the avian innate immune system when exposed to various avian pathogens. A summary and comparison of results obtained from heterophils and macrophages exposed to a variety of bacterial (*E. coli*, *Salmonella*, *Mycoplasma*), viral (MDV, AIV), and protozoan (coccidia) agents will be presented.

INFECTIOUS BRONCHITIS VIRUS

100.

siRNA inhibition of Infectious Bronchitis Virus Replication

Mark W. Jackwood and Ralph A. Tripp
The University of Georgia
Department of Population Health
953 College Station Road
Athens, GA 30602

Small interfering RNA (siRNA) molecules are short RNA segments that serve as template with the RNA-induced silencing complex (RISC) to specifically silence genes. We designed siRNAs against infectious bronchitis virus and tested them in vitro and in vivo. In vitro viral replication was inhibited by 3 of the constructs, but not by a negative control siRNA. The IBV inhibiting siRNA constructs were mixed together and given to 1 week old chicks and the level of protection was assessed following challenge. The results of the experiments will be presented along with the practicality of this approach to control IBV.



101.

Construction and evaluation of DNA vaccines coding for S1 and N of infectious bronchitis virus

Hyuk Moo Kwon, Ena Kim, Son Il Pak, and Haan Woo Sung
Department of Veterinary Medicine, Kangwon National University
Chunchon, 200-701, Kangwondo,
Republic of Korea

Infectious bronchitis viruses (IBVs) isolated in Korea were used to construct DNA vaccines in this study. S1 glycoprotein (S1) and nucleocapsid (N) genes of IBV were amplified by reverse transcriptase polymerase chain reaction (RT-PCR). To construct plasmid DNA vaccines, each PCR product of S1 and N genes was purified and cloned into eukaryotic expression vector (pcDNA 3.1/V5-His TOPO vector) and transformed into competent *Escherichia coli*. Cells carrying recombinant plasmid were selected on LB plates containing ampicillin. The nucleotide sequence and orientation of each constructed plasmid were verified by DNA sequencing. Verified plasmids were prepared and used to confirm the expression of cloned S1 and N gene. The expression of S1 and N proteins from constructed DNA vaccines were confirmed by an in vitro transcription / translation system. The protective immunity of constructed plasmids was tested in chickens.

INFECTIOUS BURSAL DISEASE

102.

Detection by PCR and nucleotide sequence analysis of a duck circovirus detected in Pekin ducks in the United States

Alejandro Banda, Rakijah Galloway-Haskins, Tirath Sandhu, Karel Schat
Cornell University Duck Research Laboratory
P. O. Box 217, 192 Old Country Road
Eastport, NY. 11941

A duck circovirus (DuCV) was detected by polymerase chain reaction (PCR) in bursal and thymic samples from Pekin ducks from New York. These birds exhibited bursal and thymic atrophy, as well as arthritis caused by *Staphylococcus aureus*. A pair of degenerate primers were designed to amplify a product of 408 bp partially encompassing the *Rep* gene. A second pair of primers were designed to amplify the remaining portion of the genome (1583 bp). These two portions were cloned and sequenced. Phylogenetic analysis of the complete genome was performed by the neighbor-joining method. The circovirus detected in NY is closely related to the virus detected in Germany (93.7% of nucleotide identity). The genomic organization of this new circovirus included the presence of four open reading frames (ORF), one stem-loop structure and four copies of 44 bp repeats. The two major ORFs corresponded to the genes *Rep* and *Cap*, that were located in the viral strand and complementary strand respectively. A stem-loop structure was observed which is characteristic of other circoviruses. The development of a specific DuCV PCR technique for diagnostic purposes is also presented.



103.

Efficacy of a Turkey Herpesvirus (HVT-MDV serotype-3)-Infectious Bursal Disease (IBD) Vaccine, Live HVT Vector, IBD-VP2, Administered *in ovo* and to One-Day-Old SPF Chickens

**Julio S. Cruz-Coy, Clovis Oliveira, Jeovane Pereira, Fernanda Ambrosino, Airtton Gaudenci, Francois-Xavier Le-gros, and Nikki Pritchard
Merial Select, Inc.**

The purpose of the study was to demonstrate the efficacy of a recombinant Marek's Disease serotype 3-Infectious Bursal Disease vaccine (VAXXITEK HVT-IBD TM, Merial Saúde Animal Ltda.), expressing genes of the IBD viral protein 2 (VP2) against the virulent MDV serotype-1 strain GA 22, and the two IBDV pathogenic strains 52-70 and the Brazilian F6023 (molecular group 15). The studies were conducted at the Merial Research Facility in Paulinia, Sao Paulo, Brazil.

Eighteen-day-old SPF embryos and one-day-old SPF chicks were vaccinated with a full dose, 9000 pfu's per bird, of a recombinant HVT-MDV serotype-3/IBD- VP2 vaccine by the *in ovo* or SQ routes of administration, respectively. Additional birds were kept non-vaccinated to serve as challenged and non-challenged controls. Five days after hatching and SQ vaccinations, 35 each *in ovo* or SQ vaccinated chicks, as well as 35 non-vaccinated chicks, were challenged by the IP route with the virulent MDV GA 22 strain, 2000 pfu's per bird. The remainder of the vaccinated chicks, 20 *in ovo*, 20 SQ and 10 non-vaccinated, were challenged at 21 days-of-age with the IBDV challenge strain 52-70 by the IO route, $10^{2.0}$ EID₅₀ per bird. After the challenges the birds were observed for morbidity and mortality for 44 days for the MDV challenge, and 4 days for the IBDV challenge. At the end of the respective observation period the remaining birds were necropsied and MDV or IBDV lesions recorded. Non-vaccinated, non-challenged birds were also necropsied at the end of the respective MDV or IBDV observation periods.

Results demonstrated that the MDV-HVT serotype-3/IBD- VP2 recombinant vaccine is efficacious against the MDV and IBDV challenges by both, *in ovo* and SQ routes of administration. The *in ovo* administration of the recombinant vaccine provided 86% and 95% protection against the MDV and IBDV challenges, respectively, while the SQ route provided 87% and 100% protection against MDV and IBDV, respectively.

The non-vaccinated, challenged control birds showed infections rates of 83% for MDV, and 100% for IBDV.

Further challenge studies against IBDV where the virulent Brazilian strain F6023 ($10^{2.0}$ EID₅₀ per bird) was used, also indicated that a full dose of the MDV-HVT serotype-3/IBD- VP2 recombinant vaccine administered *in ovo* or SQ provided 90% and 100% protection, respectively, against this challenge. The incidence of IBDV in the non-vaccinated, challenged control birds was 90% in this trial.

These results indicate that the licensed MDV-HVT serotype-3/IBD- VP2 recombinant vaccine is fully effective in the control of both MDV and IBDV.



104.

Construction and Evaluation of Turkey Herpesvirus Vectored Newcastle Disease Vaccine

Motoyuki Esaki, Kristi M. Moore, Takanori Sato, Shuji Saitoh, Sakiko Saeki, Ayumi Fujisawa, Atsushi Yasuda and Joan D. Leonard

The *fusion* gene of Newcastle disease virus was inserted into the genome of a turkey herpesvirus vaccine. Insertion and expression of the *fusion* gene was confirmed by several molecular assays. The recombinant vaccine was stable after sequential passages in cell culture and specific pathogen free chickens. Safety was demonstrated in chickens as well as multiple avian species. For efficacy studies, the recombinant vaccine was mixed with the SB1 vaccine strain of Marek's disease virus and administered *in ovo* and subcutaneously to day of age chicks. Protection was shown following challenge with the Texas GB strain of Newcastle disease virus as well as a very virulent strain of Marek's disease virus.

105.

Serological response with the use of three different bivalent killed vaccines against IBD and Reovirus in Broiler Breeders

Mauricio Gonzalez, Miguel M. Melchor, Odette Urquiza, and Mario G. Lechuga

Fort Dodge Animal Health
Sevilla 821, Colonia Portales, CP 03300
Delegación Benito Juárez
Mexico City, Mexico

120 Broiler breeders, divided in three groups (40 each) and identify individually, were vaccinated at 12 weeks of age with a Killed vaccine containing IBD (Standard and variant strain) and Reovirus antigen, Bursa Tissue origin. Serums for each birds were obtained at 12 (befor killed vaccine application) 16 and 20 weeks of age.

The serums were tested by ELISA test (IBD and Reovirus) and Micro Virus Serum Neutralization (IBD and Reovirus) in two different Diagnostic laboratories : UNAM and Biovet Serological results were plotted and compared statically (ANOVA test) in order to look for significant differences between the 3 product used.

106.

Profiling of infectious bursal disease virus (IBDV) in the U.S.A. and some foreign countries based on sequencing genomic material during 2003-2005

Mohamed Hamoud and Pedro Villegas

Poultry Diagnostic Research Center; College of Veterinary Medicine
The University of Georgia; 953 College Station Rd. Athens, GA 30602 U.S.A.

The hypervariable region of the VP2 gene for IBDV was extracted from formalin fixed paraffin embedded tissues. Using real-time RT-PCR and sequencing, the IBDV strains were identified. The ability to identify the actual virus strain causing the lesions observed microscopically in the bursa of Fabricius allows for direct correlation between viral identity and pathology. This will help in designing better vaccination strategies, and identifying new emerging viruses as they appear in the environment.

107.

Efficacy of the vaccination against Infectious Bursal Disease in Broilers using an Immune Complex Vaccine at one day old.

**Eliana Icochea¹, Pablo Reina¹, Mónica Alba¹, Rosa González¹, Jhon Guzmán¹
Karina Vidal¹, Walter Paredes¹ Paola Cruz².**

1. School of Veterinary Medicine, San Marcos University, Lima-Perú

2. Ceva Sante Animale Laboratory .

This study evaluated three vaccination programs against Gumboro Disease in broilers chickens. Were used 240 Ross 308 broilers chickens one day old, distributed in four groups of 60 birds each one. Group A was vaccinated at one day old using a commercial vaccine containing an antigen – antibody complex (2512 strain), applied subcutaneously. Group B was vaccinated twice at 10 and 18 days old using intermediate commercial vaccines containing the Lukert (mildly) and CE (intermediate) strains, respectively. Group C was vaccinated once with a vaccine containing LZD228 strain applied at 21 days old. Group D was a control. The groups were growing in separated environments at Vet School at San Marcos University until challenge at 35 days old. The F52/70 Gumboro strain was used for challenge. In order to evaluate the three vaccination programs were registered mortality, clinic signs and macroscopic lesions. Mortality rates were not observed. The clinical signs were depression and diarrhea. Were observed 15 and 20% depressed birds at 2, 3 and 4 days post challenge in vaccinated groups and between 20 and 40% in unvaccinated group. Furthermore until 60% of unvaccinated group had moderate to severe diarrhea 10 days post challenge. All groups presented bursal edema until 10 days post challenge, but the birds of the unvaccinated control group presented severe edema affecting 87 % of these birds at 10 days post challenge. In order to determine the safety of the vaccines applied in the three vaccination programs, bursal microscopic lesions and bursal Index were evaluated. The microscopic bursal lesions were analyzed by Kruskal-wallis. There were significant differences ($p < 0.05$) among vaccinated groups. The group A obtained the highest bursal microscopic lesions scores (3.8) and group B the lowest (1.8). Similarly Bursal index was evaluated to determine bursal atrophy as a vaccination result. Any vaccinated group showed bursal index values of atrophy. These values were analyzed by the Kruskal-Wallis test and there were not statistical differences ($p > 0.05$) among groups. The lower serologic response at 45 days of age in the groups B, C and control corresponded with the presence of clinical disease signs (bursal edema) in these groups until the end of the experiment. This was not happening with the group A. Regarding productive performance analyzed by Anova, significant statistical differences in body weight were obtained among vaccinated groups A (3426), B (3264) and C (3215) ($p < 0.05$) versus unvaccinated control group (3082), obtaining from 134 to 345 gr more at the slaughter, 10 days post challenge, in these vaccinated groups. Accumulated daily weight gain, Feed conversion rate and performance index was better than the control group nevertheless these differences were not significant.



108.

Phylogenetic analysis of very virulent infectious bursal disease viruses.

Daral J. Jackwood and Susan E. Sommer-Wagner

The Ohio State University/OARDC
Food Animal Health Research Program
1680 Madison Ave., Wooster, Ohio 44691

The very virulent form of infectious bursal disease virus (vvIBDV) causes high mortality and immune suppression in chickens. Genome samples from vvIBDV strains were collected from countries in Asia, Europe, Africa, Middle East, South America and Central America. The hypervariable sequence region of the VP2 gene was determined and used to compare the viruses. A phylogenetic analysis was conducted on the sequences to determine their evolutionary relatedness. In most cases, vvIBDV strains from a continent were more closely related compared to viruses from different continents, indicating the viruses were evolving locally. However, some vvIBDV strains from Europe, Africa, South America and Central America had close evolutionary ties suggesting that vvIBDV has spread from one continent to another. Our data do not support independent mutation events for the emergence of these viruses.

109.

Mechanisms of cell destruction by infectious bursal disease virus.

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 economic loss to poultry industry worldwide. The virus infects and destroys actively dividing IgM-bearing B cells and macrophages in the bursa and at peripheral locations. Mechanisms of IBDV-induced cell death are not well understood. Apoptosis (programmed cell death) has been implicated in the pathogenesis of IBDV, although it is unclear whether proteases of the caspase family are involved. Caspases comprise a family of cysteine-dependent aspartate-directed proteases which catalyze key steps in the death pathway. In the present study, we measured the activation of caspase-3 by immunohistochemistry *in vivo* and *in vitro*. At 4 and 6 days following *in ovo* inoculation, caspase-3 activation was considerably elevated in bursa of IBDV inoculated chickens in comparison to normal controls. Similarly, caspase-3 activation was observed in chicken embryo fibroblast cells infected with IBDV. A caspase inhibitor, Q-VD-OPH, significantly inhibited IBDV induced caspase-3 activation and apoptosis. These results indicated that IBDV induced apoptosis is caspase dependent. A clearer understanding of the pathogenic mechanisms of virus-induced cell death is crucial for the development of effective therapeutic strategies.



110.

The role of 4 different Infectious Bursal Disease vaccines in the control of field Infectious Bursal Disease Virus (IBDV) in a broiler ranch in California

**A. Mazaheri^A, B. R. Charlton^A, M. C. Bland^A, G. L. Cooper^A, A. A. Bickford^A,
F. Sommer^A and D. J. Jackwood^B**

^ACalifornia Animal Health and Food Safety Laboratory System - Turlock Branch, University of California, Davis

^BFood Animal Health Research Program, Department of Veterinary Preventive Medicine, Ohio Agriculture Research and Development Center, The Ohio State University

Recent investigations in a broiler ranch in California with a history of runting and stunting revealed the presence of an IBDV strain comparable to the designated T1 strain isolated first in Georgia (Jackwood et al, 2001). Severe bursa damage detected via histopathology during this study and the detection of T1-like IBDV strain suggested a sub clinical bursal infection in the ranch. Based on this previous investigation, a field trial was initiated to test the protective potential of 4 different commercial IBDV vaccines via histopathological examination.

The flock under investigation was placed on 3rd run litter and under the assumption that the birds were naturally exposed to this field virus. Four wood and wire paneled pens were constructed within a standard broiler house, resulting in 5 groups of birds. Forty birds were placed in each pen and the remaining house contained about 22000 birds.

Four groups were vaccinated with different IBD vaccines via drinking water, the 5th group remained as unvaccinated control group. Five birds per group were euthanized every 5 days during the first two weeks of the trial and then every 10 days until the end of grow out. Bursa samples were collected, and each bursa was examined by histopathology.

The first severe bursa lesions were found at 25 days of age. In both control and the vaccinated groups, histopathology results suggest ongoing bursa destruction until day 36 and 45. In order to prove the presence of an IBDV field strain, bursa pool samples were sent to Ohio State University for RT-PCR/RFLP. No IBDV field strain was detected in birds younger than 25 days. T1 strain was detected from bursa samples of the 5 different groups at the age of 25 days, which confirmed the histopathological results. In conclusion, the birds were probably challenged by T1 strain around 15 days of life, at the time when the level of maternal antibodies wanes. Histopathological results suggest that none of the commercial vaccines was able to protect the birds against the IBDV field challenge.



111.

Lymphocyte depletion, vaccine virus detection and bursal lesions in commercial broilers vaccinated with commercially available and experimental Infectious bursal disease vaccines

Enrique Montiel N, Nikki Pritchard, Amy Holderfield, Julio Cruz, Robert Smith, and Kari Pack

Merial Avian Global Enterprise
P.O. Drawer 2497; Gainesville GA 30503

Commercial broilers were vaccinated with commercially available and experimental Infectious bursal disease vaccines either *in ovo* or after hatch. An additional group of birds was kept unvaccinated to serve as challenge controls. The birds were housed in isolation units. Bursa, spleen and thymus samples were collected in 10% formalin at 3,7,10,14,17 and 21 days post vaccination for histopathology and fresh tissues for IBD PCR. The degree of bursa damage was assessed by lesion scoring and lymphocyte depletion using a bursa imaging system. The dissemination of the virus in the tissues was assessed by PCR. All PCR products were sequenced to establish the identity of the vaccine virus present.

112.

Infectious Bursal Disease Virus (IBDV) Surveillance

Nannette Olmeda-Miro, Sandra Cloud, and John Rosenberger

AviServe LLC
Delaware Technology Park
1 Innovation Way, Suite 100
Newark, DE 19711 USA

Surveillance of broiler flocks from different geographical areas, complexes, etc. can be a valuable means for monitoring IBDV antigenic and pathotype shifts, to evaluate progeny susceptibility to field challenge and for establishing base lines for assessing the effectiveness of control programs within and among companies.

For IBDV surveillance, bursae of Fabricius are collected from five birds per broiler flock. The farms selected are ranked in the lower one third of producers and the birds assessed are between the ages of 16 to 26 days. Virus isolations and initial pathotyping are done in embryos and IBDV isolates further characterized with specific antibodies. If deemed appropriate the viruses may be sequenced and pathotyped in chickens.

Selected isolates have been used for progeny challenge and preparation of autogenous vaccine (s).



113.

Identification of Very Virulent Infection Bursal Disease (VVIBD) virus in Colombia. A Five Year Study

Maritza Tamayo, Sergio Velez, and Kalen Cookson
Fort Dodge Animal Health; Sevilla 821 Col Portales Sur
Del. Benito Juarez CP 03300; Mexico D.F. MEXICO

Infectious Bursal Disease (IBD) continues to be an economic problem throughout Latin America. Immunodepression resulting from an IBD virus infection could be associated to other different infectious conditions that affect in the region.

In order to have a better understanding of the Disease we support in different diagnostic tools, from the field using a Bursameter continuing with Imaging Processing (IP) and Reverse Transcriptase, Polymerase Chain Reaction Fragment Length Polymorphism. (RT.PCR/RFLP). We are presenting results for the past five years.

114.

Infecting Chickens and Inducing Immune Response by a VP3-deleted Infectious Bursal Disease Virus expressing the GFP Reporter Gene

Ching Ching Wu, Michelle Peters, and Tsang Long Lin
Purdue University, Animal Disease Diagnostic Laboratory,
406 South University Street, West Lafayette, IN 47907-2065

The utilization of viral vectors as vaccine vectors for the delivery of recombinant genes has arisen to take advantage of the inherent capacity of viruses to transducer target cells in a highly efficient and specific manner. The present study was undertaken to determine whether the VP3 deletant of infectious bursal disease virus (IBDV) could be rescued in a complementing cell culture system and trigger immune response in chickens following oral inoculation. A VP3 deletion in IBDV was substituted with the GFP reporter gene (GFP⁺, VP3⁻ IBDV). The virus vector was replication deficient due to the VP3 deletion, but virus particles could be rescued at titers of 10^{8.5} pfu/ml from Vero cells. The infectivity of GFP⁺, VP3⁻, IBDV was verified by GFP expression in infected cells and virus particles with typical birnavirus morphology were demonstrated by electron microscopy. Specific-pathogen-free chickens were infected with 10⁷ EID₅₀ GFP⁺, VP3⁻, IBDV. High levels of viral and reporter gene expression were detected in the bursa, thymus, and spleen. Transduction with GFP⁺, VP3⁻, IBDV induced strong humor immunity with peak serum antibody titers against the viral and reporter proteins observed 8-15 days after inoculation. Virus neutralization antibodies were present between 8-22 days after inoculation. Cell-mediated immunity directed against viral and reporter proteins was evident by up-regulation of specific lymphocyte stimulation indices and marked early up-regulation of IFN- γ and IL-2 transcripts in the bursa and spleen in response to GFP⁺, VP3⁻, IBDV transduction.



115.

The Use of Live IBD Boosts in Breeders

John E. McCarty
Schering-Plough Animal Health
7744 Peters Rd #312
Plantation, FL 33324

A live IBD boost program has been established in several breeder flocks. Sister flocks of these vaccinated flocks are also being followed. A live IBD boost is being used in the IBD boost vaccinated flocks to see if more consistent IBD titers can be maintained in these flocks compared to the sister flocks which are not receiving the live IBD field boost. The poster will present the IBD titer results of the study and discuss the pros and cons of such a program.

LARYNGOTRACHEITIS

116.

An outbreak of infectious laryngotracheitis in meat chickens

**Rocio Crespo, H. L. Shivaprasad, Richard P. Chin, Portia L. Cortes, Peter R. Woolcock,
and Maricarmen Garcia**
CAFHS – Fresno, University of California Davis
2789 South Orange Ave.
Fresno, CA 93725

An outbreak of infectious laryngotracheitis occurred in meat chickens, between 4 and 21 weeks of age, from the California Central Valley. The morbidity was as high as 20%. Syncytial cells with intranuclear inclusion bodies were found in conjunctiva, larynx, trachea, lung, and air sac. Virus isolation was attempted in every case, but isolation of herpesvirus was successful only in a couple of cases. ILTV DNA characteristic of CEO vaccine strain was detected by Real Time PCR-RFLP. This is an unusual finding since no vaccinated flocks are near the outbreak area. Epidemiology and management of the outbreak will be discussed.



117.

Vaccination of Broilers Using a Recombinant Fowl Pox-Infectious Laryngotracheitis Vaccine Inoculated in ovo

Brett A. Hopkins
Biomune Company
8906 Rosehill Road
Lenexa, KS 66215

Commercial broiler eggs were injected with Marek's, Infectious bursal disease and a recombinant fowl pox-infectious laryngotracheitis vaccine using the Inovo-ject[®] system at 19 days of incubation at commercial hatcheries in the Southeast United States. The safety of administering a recombinant fowl pox-laryngotracheitis vaccine in ovo in commercial hatcheries was evaluated in broilers by several methods that included but were not limited to hatchability, gross and histology evaluations of tissues, mortality and growth performance. The efficacy for protection against infectious laryngotracheitis was evaluated by eye-drop, spray, and contact challenge in colony houses, and also by performance in commercial housing environments.

MISCELLANEOUS VIRUS

118.

Cross Virus-Neutralization Studies on Turkey Coronavirus

Tye O. Boynton, Mark W. Jackwood, Scott A. Callison, and Deborah A. Hilt
The University of Georgia
Department of Population Health
953 College Station Road
Athens, GA 30602

Hypervariable regions in the sequence of the spike glycoprotein of turkey coronavirus (TCoV) suggests that different serotypes of the virus exist. In this study, we conducted two-way cross virus-neutralization experiments in embryonating turkey eggs. Specific rabbit antiserum against two different strains of TCoV was reacted with the viruses and virus-neutralization was assessed by quantitative real-time reverse-transcriptase polymerase chain reaction. The results when correlated with ELISA data and sequencing data, indicate that different serotypes of the virus do exist.



119.

**Sequence comparison of the right end of fowl adenovirus genomes
representing each viral species**

Amalia Garceac and Eva Nagy

Department of Pathobiology, Ontario Veterinary College,
University of Guelph, Guelph, Ontario, N1G 2W1, Canada

Fowl adenoviruses (FAdV) are classified in the family *Adenoviridae*, genus *Aviadenovirus*. FAdVs can be involved in important diseases like inclusion body hepatitis and hydropericardium syndrome.

We have sized the entire genome and sequenced the right end of four FAdV serotypes from different species, FAdV-4 and FAdV-10 strain C-2B (*FAdV-C*), FAdV-2 (*FAdV-D*) and FAdV-8 (*FAdV-E*) and compared them to the known FAdV-1 (*FAdV-A*) and FAdV-9 (*FAdV-D*). The sizing was based on digestion with restriction enzymes and separation in agarose gels. The genome length was found to be 46-47 kb for FAdV-4, -10 and -2, and 43-44 kb for FAdV-8. The right end fragments for all viruses were cloned, sequenced and overlapped by PCR. Nucleotide and amino acid sequence analyses demonstrated a relative diversity among the right end ORFs of all viruses, with the exception of GAM-1, lipase and fiber protein, which are highly conserved. Less than half of the identified ORFs had similarity with fowl adenoviruses. Some of the ORFs showed sequence identity with sequences from herpesviruses and poxviruses, suggesting ancestral recombination between them and fowl adenoviruses.

120.

**Genetic Diversity in Turkey Coronavirus Viral RNA following Passage
in Embryonating eggs**

Deborah A. Hilt, Mark W. Jackwood, and Tye O. Boynton

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

Sequence data for coronaviruses is biologically significant because it furthers our understanding of viral genetic mutation, recombination, genome diversity, and evolution. That information is extremely important for predicting the emergence of new coronaviruses like severe acute respiratory syndrome coronavirus (SARS-CoV). In this study we examined the entire 3' end (spike gene to 3' un-translated region) of two different strains of turkey coronavirus (TCoV) at low and high passage in embryonating turkey eggs. Genome diversity, mutation rates, and recombination sites were examined and related to evolutionary trends for the virus. The data provides important parameters for predicting how new coronaviruses become widespread and persistent in the field.



121.

Selecting the Optimal Age to Vaccinate Turkeys Against Hemorrhagic Enteritis Virus

Brett A. Hopkins
Biomune Company
8906 Rosehill Road
Lenexa, KS 66215

The rate of decline and titers of maternal antibodies were determined using ELISA detectable antibodies from multiple commercial turkey flocks originating from three distinct sources. The age for vaccination against HEV was adjusted based on the maternal antibody results and the effect of this adjustment was evaluated by ELISA HEV serology post-vaccination and by flock settlement performance.

122.

Dynamics of the Chicken Anemia Virus at Commercial Broiler Farms

Mario Lechuga, Elena Salazar, Susano Medina, and Maritza Tamayo
Fort Dodge Animal Health
Sevilla 821, Colonia Portales, CP 03300
Delegación Benito Juárez
Mexico City, Mexico

The objective of this paper is to show the observations that we had made from laboratory studies when they were tested for Chicken Anemia Virus. The samples come from seven broiler farms, and the parameters to measure were serology (ELISA), histopathology and PCR to detect viral DNA.

123.

Detection and Sequencing of Avian Astrovirus from Broilers Using RT-PCR

**Lanqing Li, PhD, Emily Handley, MS, Michael R. Luther, BS,
and Frederic J. Hoerr DVM, PhD.**

Alabama Department of Agriculture and Industries, C. S. Roberts Veterinary Diagnostic Laboratory,
Auburn, AL 36831-2209

Broilers with enteric disease-stunting syndrome from eight poultry companies in three states (Alabama, Arkansas, Georgia) were tested for avian astrovirus by RT-PCR. The samples included 65 intestine and six mealworm larvae samples from chicken litter; 35/65 intestine samples (53.8%) and 1/6 mealworm samples were positive. The positive chickens were four- to 23-days-old. Twelve PCR amplicons (about 600 bases, partial of ORF1b gene of astrovirus) were sequenced; all shared 82.4-86.8% similarity with avian nephritis virus 1 at the nucleotide level and 90-94.9% similarity at the amino acid level. Three sequences derived from samples from two different broiler companies shared 100% similarity at the amino acid level.



124.

Spike protein gene-based genetic analysis of turkey coronavirus isolates from different geographic locations of the U.S.

Tsang Long Lin, Chien Chang Loa, Ching Ching Wu, Thomas Bryan, Thomas Hooper, and Donna Schrader

Purdue University; Animal Disease Diagnostic Laboratory
406 South University Street; West Lafayette, IN 47907-2065

The study was conducted to determine the genetic relationship of turkey coronavirus (TCoV) isolates from various geographic locations of the U.S. by sequence analysis and comparison of spike (S) protein gene, the major structural protein gene of TCoV with the most genetic variability. Seventeen isolates of TCoV from turkey farms located in different geographic regions were recovered from clinical cases submitted to Animal Disease Diagnostic Laboratory, Purdue University. The entire S protein genes of TCoV isolates were amplified by reverse-transcription-polymerase chain reaction (RT-PCR) from RNA extracted from turkey embryo intestines infected with TCoV. The PCR amplicons were cloned and sequenced and the similarity of their nucleic acid and deduced amino acid sequences were analyzed. The size of S protein gene of TCoV isolates varied from 3,609 nucleotides (a Texas isolate) to 3,642 nucleotides (a North Carolina isolate). Pairwise comparisons of S genes among different TCoV isolates revealed high level of similarity at 92.2 to 99.7 % for nucleic acid sequences and 90.2 to 99.3 % for amino acid sequences. Most of the sequence variations among the different isolates were observed in the amino terminal half of S protein. Phylogenetic analysis based on the deduced amino acid sequences of S protein demonstrated that all TCoV isolates were clustered within the same genomic lineage while the IBV was grouped into a separate cluster. The results indicated that the TCoV isolates from various geographic locations in the U.S. had high genetic homology among the S protein genes and shared high genetic similarity.

125.

Gene function studies of fowl adenovirus type 9 genome

Éva Nagy and Juan Carlos Corredor

Department of Pathobiology, Ontario Veterinary College
University of Guelph; Guelph, Ontario, N1G 2W1, Canada

The molecular biology of fowl adenoviruses (FAdVs) is poorly understood. Early genes found at the right and left end regions of the FAdV genomes do not have homologies to their counterparts in mastadenoviruses. The mastadenoviral E1 genes, at the left end of the genome, play essential roles in virus replication such as inhibition of apoptosis, cell cycle progression, counteraction of innate immune response, etc. Therefore, we hypothesized that genes at the left end of FAdVs also have similar functions. To test this hypothesis, we used FAdV-9 as a model and modified the left end of the virus. The altered viruses were analyzed *in vitro* and *in vivo*.



126.

Genetic and serologic evidence of reticuloendotheliosis virus (REV) integrated avian poxvirus (APV-REV) infection of wild and exotic birds.

Theodros Tadese, E.A.Potter, and W.M. Reed

Diagnostic Center for Population and Animal Health/
Pathobiology and Diagnostic Investigation, Michigan State University,
East Lansing, MI 48824

Reticuloendotheliosis virus (REV) has been found to be intimately associated with very limited numbers of vaccine strains but almost all field isolates of fowl pox virus (FPV) examined thus far in recent years. In light of that, we attempt to shed light on the ecology of transmission of avian pox virus (APV) among wild birds carrying integrated REV. A sensitive PCR technique; a commercial enzyme linked immuno- sorbent assay (ELISA) and an in-house REV coated ELISA were used to test sera derived from avian poxvirus infected chickens. While most sera indicated presence of anti -REV antibodies; only 4 out of 23 (17 %) APVs were positive for REV-LTR by PCR confirming the presence of integrated REV in APVs of wild birds. These results suggest that APVs from wild birds also carry integrated REV. This study highlights the magnitude and intricate association of REV with APV strains, isolated over an extended period of time, from various species of wild birds. This study will help lead to more surveillance and control programs.

127.

**Rapid Diagnosis and Differentiation of Avianpox viruses
by Amplification of Specific Gene Fragments**

Deoki N. Tripathy and Trina Westerman

University of Illinois
2001 S. Lincoln Avenue, College of Veterinary Medicine
Urbana, Illinois 61802

Poxviruses that infect wild birds can also infect domestic poultry. With a view to rapidly diagnose and differentiate these viruses, by using seven sets of primers for amplification of specific regions in their genome, we have made interesting observations. For example, EGF and A-type inclusion body genes are conserved in all avian poxviruses. Reticuloendotheliosis (REV) related sequences are absent in viruses of wild birds. Differences in the size of 39K gene occur among strains of fowlpox viruses. Field and vaccine strains of fowl poxvirus as well as those, which infect the wild birds, can be differentiated by using these primers.



128.

Production of Recombinant VP1 Protein and Specific Antibodies of Avian Polyomavirus

Hsiang-Jung Tsai, Chih-Ming Hsu, and Ing-Cherng Guo

Graduate Institute of Veterinary Medicine, National Taiwan University

No. 1, Sec. 4, Roosevelt Road, Taipei 106, Taiwan

PCR products of VP1 gene of avian polyomavirus (APV) were cloned into the pET vector. Following transformation into the host cells BL21(DE3), IPTG induction and purification with nickel-NTA His-bind resin column, the yield of highly purified recombinant VP1, recognized specifically by anti-His tag antibodies and anti-APV antibodies, is around 275 mg per liter of *E. coli* culture. The APV-specific antiserum has been produced by recombinant VP1 and monoclonal antibodies are under screening. The high yield and purity of recombinant VP1 and specific antibodies are the good material source for developing diagnostic kit and vaccine of APV infection.

129.

A Reproducible Model for Runting and Stunting Syndrome (RSS)

Guillermo Zavala, Taylor Barbosa, and Holly S. Sellers

The University of Georgia

Department of Population Health

953 College Station Road

Athens, GA 30602

Runting and stunting syndrome (RSS) has been previously reproduced by gavage inoculations with intestinal homogenates from RSS-affected chickens. A reproducible model was designed to study early RSS in broiler chickens. Contaminated litter and affected broilers from a field case were used to seed isolated pens. Multiple growouts of 2-3 weeks with downtime intervals of 24-72 hours successfully and consistently reproduced: a) RSS clinical signs; b) approximately 50% body weight depression compared to control chickens; and c) gross and microscopic lesions. The model was used to study the potential role of breed cross, gender, downtime, thermal treatment of contaminated litter, maternal antibody against CAV, IBDV and reovirus, and antiprotozoal drugs.



MYCOPLASMA

130.

Is F Strain *Mycoplasma gallisepticum* (Mg) Vaccine as Contagious and as Easily Spread as Thought? Experiences From A Back Passage Study Shared

Stephen W. Davis, C. Broussard and R.A. Phillips
Colorado Quality Research, Schering-Plough Animal Health

A study was conducted to evaluate the working seed for a commercially licensed F strain *Mycoplasma gallisepticum* (Mg) vaccine through back passage in susceptible SPF leghorn chickens. The results from this research found that back passing the Mg vaccine working seed was more difficult than expected. The challenge dose and incubation time after bird challenge was significantly increased to successfully back pass the organism. The route of inoculation of the Mg was also necessary to successfully pass the Mg vaccine from one group of challenged birds to the next group of susceptible birds. Clinical signs and culture results from inoculated birds and adjustments necessary to successfully back pass the vaccine working seed will be presented.

131.

***Mycoplasma gallisepticum* Vaccine Strain 6/85 Penetrates into Chick Embryo Fibroblasts in High Numbers**

John E. Dohms and Cynthia M. Boettger
Dept. of Animal and Food Sciences, University of Delaware,
040 Townsend Hall, Newark, DE 19716-2150

M. gallisepticum (MG) 6/85 is a vaccine strain shown to be effective in field trials, but produces little serological response. In a previous report, MG R strain was able to enter HeLa and chick embryo fibroblast cells (CEF) cultures with 4-5% invasion rates. In a series of experiments, MG 685 had an average 452% invasion rate in CEF after 3 hours of incubation. This is conundrum because intracellular invasion is thought to be a virulence factor. We surmise that the low MG 685 antibody responses may be due to MHC class I presentation that might produce cell mediated immune response.



132.

Field Evaluation of TS- 11 and Vectormune FP-MG+AE Vaccines in Controlling MG Outbreak in Commercial Cage Layer Operation in Arkansas and It's Effects on Flock Performance

Hashim M. Ghori, Lisa A. Newberry, and Stanley H. Kleven
Veterinary Diagnostic Laboratory, Arkansas Livestock & Poultry Commission,
#1 Natural Resources Drive, Little Rock, AR 72205,
Department of Avian Medicine, University of Georgia, Athens, Georgia

Hens were submitted to the Laboratory for evaluation following an increase in mortality and a drop in egg production. MG was confirmed in the flock by PCR and serology. Subsequent evaluation of the pullet farms and the hen flocks demonstrated the presence of MG in the hen flocks only. Pullet flocks were located 45 miles away from the hen complex and were initially vaccinated with TS-11 and changed to Vectormune FP-MG+AE vaccine. Data on mortality, feed conversion and egg production are under evaluation. DNA sequencing of select MG isolates and serology titers between vaccinated and unvaccinated flocks will be provided.

133.

***Mycoplasma gallisepticum* Detection and Genotyping in Vaccinated Layer Flocks**

Alecia Godoy, Maricarmen Garcia, Kristi Moore, Motoyuki Esaki, and Joan Leonard
Biomune Company
8906 Rosehill Road, Lenexa, Ks 66215

This study was conducted in commercial layer flocks located in different geographic regions of the USA. Tracheal swabs, taken at different times during production, were analyzed applying the method of polymerase chain reaction (PCR) using primers targeted to the gene *mgc2*. These primers have the characteristic of encoding a cytoadhesin-related surface protein uniquely present in *Mycoplasma gallisepticum* (MG) strains. The identification of MG strains was carried out by purification of the PCR products and sequencing analysis. Results obtained from sequencing portions of *mgc2* genes indicated that the MG field strains detected in the tracheal swabs had sequences that ranged from 97.0% similar to 100% identical to 6/85, F, ts-11 and house finch 51 strains. An interesting observation during this evaluation was the findings of a combined MG infection specifically detected in the same tracheal swab. The PCR analysis of this sample showed two different banding patterns of approximately 237bp and 300bp, which in the sequencing results were identified as MG strain 6/85 (97% similar) and as F strain (100% identical), respectively. The use of this method showed to be useful for epidemiological evaluations of MG infections.



134.

A Case Study of a Mycoplasma Problem Breeder Farm

Tami F. Kelly, Fred Hoerr, and Marshall Putnam

Alabama State Veterinary Diagnostic Lab

501 Usury Ave

Boaz, AL 35957

A breeder farm has broken with MS or MG for the last three years in a row at approximately 35 to 36 weeks of age. The breeder farm has neighbors with a vast array of yard birds: guineas, ducks, chickens, and turkeys. The results will be shown for these 3 years of testing along with the affected broiler flocks and the neighbor's yard birds that we were able to test. DNA fingerprinting was used to compare all the different isolates and these results will also be presented.

135.

Modulation Of Gene Expression In Chicken Macrophages By *Mycoplasma Synoviae*

**Miha Lavrič^{1,2}, Travis W. Bliss², John E. Dohms², Dušan Benčina¹, Mojca Narat¹,
Calvin L. Keeler Jr.²**

¹ Biotechnical Faculty, University of Ljubljana

² Department of Animal and Food Sciences, University of Delaware

Mycoplasma synoviae (MS) is the causative agent of chronic respiratory disease and infectious synovitis in chickens and turkeys. Previous studies showed that specific MS membrane proteins induce secretion of IL-1 β , IL-6 and nitric oxide in chicken macrophages. The transcriptional response of infected chicken macrophage cell line HD11 was evaluated using the AIIM (Avian Innate Immunity Microarray). A variety of chemokine and cytokine genes involved in the avian inflammatory response (e.g. K60, MIP-1 β , IL-1 β , iNOS) were found to respond (induced or repressed up to 100-fold) to the MS infection. These studies will be extended to different target cells (e.g. PBL derived macrophages) as well as using different MS strains, MS derived membrane fractions and specific membrane proteins.

The research project is supported by the Fulbright Grant.



136.

Comparison of culture and PCR for the detection of *M. iowae* in turkey embryos

V.A. Leiting and S.H. Kleven

The University of Georgia
953 College Station Road
Dept. of Population Health
Athens, GA 30602-4875

Dead in-shell turkey embryos were cultured via esophagus, yolk-sac, or other tissue, to isolate *M. iowae*, (*MI*). Esophageal cultures yielded the best results, with less contamination than that of yolk-sac. Polymerase chain reaction (PCR) was used for *MI* specific determination. The PCR method was found to be sensitive in the determination of *MI* infection in the field. Random Amplified Polymorphic DNA analysis (RAPD) was used to determine if a specific type of *MI* was highly transmissible in the field, and was associated with pathogenicity.

137.

Further Western Spread of *Mycoplasma gallisepticum* Infection of House Finches

David H. Ley, Deborah S. Sheaffer, and André A. Dhondt

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

Mycoplasma gallisepticum (MG) emerged in 1994 as the cause of conjunctivitis in house finches in their eastern range of North America. We report further western expansion of MG-conjunctivitis in the native range of house finches based on positive PCR results with samples from birds captured near Portland, Oregon. Furthermore, we found evidence of genomic variability among MG isolates by amplified-fragment length polymorphism. Extension of MG-conjunctivitis in house finches to Oregon and evidence that the strains involved are showing genotypic variability are reminders that commercial poultry producers should maintain biosecurity measures that minimize contact between songbirds, especially house finches, and poultry.

138.

Mycoplasmosis In Free Living Water Fowls

Salah Moussa

Vet. Service, Assiut, EGYPT

A total of 96 samples were collected from 96 free living water-fowls (*Anas crecca* and *Fulica arta*). Samples were subjected to mycoplasma isolation and identification. Two mycoplasma serotypes (*M. anatis* and *M. iners*) were serologically identified. Results of pathogenicity test proved that both serotypes were pathogenic for three-week-old muscovy ducks, but were nonpathogenic for day old chicks.

139.

Natural coinfection of *Avibacterium paragallinarum* and *A. gallinarum* in *Mycoplasma* spp. seropositive game chickens

Edgardo Soriano V., Vladimir E. Morales, Vicente H. Vega, Andrea P. Zepeda, Nydia R. Reyes, Saul A. Ramirez, and Salvador B. Lagunas

Centro de Investigación y Estudios Avanzados en Salud Animal, FMVZ-UAEM.

Instituto Literario No. 100, Col . Centro

Toluca, 50000, México, México

Here is reported the isolation of *Avibacterium paragallinarum* (serogroup A) and *A. gallinarum* in *Mycoplasma* spp.-seropositive game chickens. Furthermore, biochemical identification and PCR-based typing are discussed. Association of these bacteria have been widely assumed or reported in a number of papers. However, the present work appears to be the first report of the natural coinfection of *A. paragallinarum* and *A. gallinarum* in chickens.



140.

Use of *M. cloacale* Bacterin in Control of Infectious Sinusitis in Pheasants and Partridges Negative for *M. gallisepticum*, *M. synoviae*, and *M. melleagridis*

Radivoje Spasojevic
Willmar Poultry Company
4546 15th St. NE
Willmar, MN 56201

Pheasants and partridges on several game farms experienced a Mycoplasma-like disease. Flocks were plate agglutination, ELISA, HI and culture negative for MG, MM, and MS and PCR negative for MG. Numerous non-pathogenic Mycoplasmae were isolated from tracheas. Commercial MG and MS bacterins produced no improvement. Autogenous bacterin consisting of *M. cloacale*, *M. pullorum*, and *M. gallinaceum* produced a 4-6 month delay of clinical signs. A monovalent *M. cloacale* bacterin was as effective as the trivalent bacterin. *M. cloacale* bacterin has been used with great success since 1997 on farms in 5 states. Non-vaccinated flocks continue to develop full blown disease.

NEWCASTLE

141.

Phylogenetic Characterization of Endemic Newcastle Disease Viruses Isolated from Wild Birds during 2000 to 2004 in Delaware, Maryland and New Jersey.

C. L. Afonso, Daniel J. King, David J. Stallknecht, and Richard Slemmons
South East Poultry Research Laboratories, USDA, ARS
934 College Station Rd, Athens, Georgia 30605

Virulent strains of avian paramyxovirus type 1 (APMV-1), the etiologic agent of Newcastle disease, are a significant threat to the United States poultry industry. The common finding of lentogenic APMV-1 viruses in live bird markets in the U.S. is a cause of concern because it suggests epidemiological connections between viruses found in wild birds and poultry and because there is evidence that some low virulence viruses can mutate and become pathogenic for poultry. Here we describe the epidemiological and biological characterization of APMV-1 present in migratory waterfowl and shore birds during 2000 to 2004 in New Jersey, Delaware and Maryland and the relationship of these viruses with other wildlife viruses found worldwide and in bird markets



142.

Exotic Newcastle Disease Virus: A Histopathologic Characterization In Encephalic Regions Of Specific Pathogen Free Chickens

Adriana Barri, Nestor Ledesma, Guillermo Tellez, and John El-Attrache

Virginia Polytechnic Institute and State University
14210 Stroubles Creek Rd.
Blacksburg, VA. 24060

This study identified, localized, and characterized lesions produced in the different anatomical regions of the encephalon of SPF chickens infected with an exotic Newcastle Disease virus strain. Birds were separated into two groups of 54 four weeks old chicks: Group 1) 36 birds inoculated I.M. with 0.2 ml of 10^6 EID₅₀ eNDV. Group 2) 18 birds inoculated I.M. with sterile PBS. Clinical signs and histological lesions were determined. Histopathological changes were more frequent in Telencephalon (Neostriatum and Hiperestriatum), Mesencephalon (optic-quiasm) and Cerebellum. Lesions consisted of: 95% gliosis, 95% necrosis, 81% neuronophagia, 30% dysmyelination, 5% hemorrhages and 5% non suppurative meningitis.

143.

The Interaction between *E. coli* and Newcastle Disease Virus in chickens

Amna B. El Tayeb^{A, B}, and Robert P. Hanson

^ADepartment of Veterinary Preventive Medicine, The Ohio State University,

^BDepartment of Veterinary Science, The University of Wisconsin-Madison

The interaction between Newcastle disease virus (NDV) and *E. coli* was investigated in cell cultures, embryonated chicken eggs, and 8-week-old chickens. The interactions were measured on the basis of bacterial adherence to chick embryo cell culture and NDV hemagglutination titers in both chickens and chicken embryos. Depending on the inoculation order of *E. coli*, a significant ($p < 0.05$) alteration of the growth of NDV was observed in both chicken and chickens embryos. Adherence of *E. coli* to chicken embryo kidney (CEK) cells was significantly increased ($p < 0.05$) when the CEK cells were infected with NDV first and then followed by *E. coli*.

^A Corresponding author



144.

Construction and Evaluation of Turkey Herpesvirus Vectored Newcastle Disease Vaccine

Motoyuki Esaki, Kristi M. Moore, Takanori Sato, Shuji Saitoh, Sakiko Saeki, Ayumi Fujisawa, Atsushi Yasuda and Joan D. Leonard

The *fusion* gene of Newcastle disease virus was inserted into the genome of a turkey herpesvirus vaccine. Insertion and expression of the *fusion* gene was confirmed by several molecular assays. The recombinant vaccine was stable after sequential passages in cell culture and specific pathogen free chickens. Safety was demonstrated in chickens as well as multiple avian species. For efficacy studies, the recombinant vaccine was mixed with the SB1 vaccine strain of Marek's disease virus and administered *in ovo* and subcutaneously to day of age chicks. Protection was shown following challenge with the Texas GB strain of Newcastle disease virus as well as a very virulent strain of Marek's disease virus.

145.

Real time RT-PCR developed for a Phylogenetically divergent Group of Newcastle viruses not detected by Current Tests

L. Mia Kim, Claudio L. Afonso, David L. Suarez, and D. Jack King
USDA-ARS-Southeast Poultry Research Lab
934 College Station Road, Athens, GA 30605

Newcastle disease virus, also known as avian paramyxovirus 1 (APMV-1), is an important poultry pathogen worldwide. Sensitive rapid diagnostic tests exist to detect APMV-1; however, due to the heterogeneous genetic nature of this virus, a unique and phylogenetically separated group of low-virulence viruses may be missed with current tests. These viruses are commonly found in waterfowl and shorebirds, but are also known to infect domestic poultry with at least some being virulent. This study outlines the development of a primer and probe set that detects members of this divergent group. Additionally, this set is compatible with the currently validated matrix real-time RT-PCR as a multiplex test.



146.

Stability of Selected Newcastle Disease Virus (NDV) Strains at Environmental Temperatures that Range from Hot to Cold

Daniel J. King

USDA, ARS, Southeast Poultry Research Laboratory
934 College Station Road
Athens, GA 30605

During the 2002-2003 Newcastle disease outbreak in the Southwestern U.S. there was concern about movement of manure from farms with infected flocks. Virus shed in the feces would be subjected to varying environmental temperatures at different geographic locations and treatment of that infective manure by composting or drying would extend temperatures to those considerably above ambient temperature. Comparative thermostability profiles are being developed for low virulence and virulent NDV strains including LaSota, Ulster, and Chicken/California/212676/2002 as the basis for assessing risk of NDV persistence in manure held at different temperatures. Initial data has been obtained at a typical composting temperature of 55C. Data is being collected at increments of 5C above and below 55C.

147.

Protection against CA02ENDV challenge of chickens vaccinated with inactivated vaccines of Newcastle Disease Virus (NDV) from different genetic lineages

Patti J. Miller, Daniel J. King, and David L. Suarez

Southeast Poultry Research Laboratory
934 College Station Road
Athens GA 30605

A comparison of the serological response and protection induced by vaccines prepared from NDV strains that represent genetically diverse isolates of the avian paramyxovirus type 1 serotype was made. Four-week-old leghorns were vaccinated with one of six different BPL inactivated vaccines; B1, Ulster, CA 2002ENDV, Pigeon 84, Alaska 196 and a normal allantoic fluid control vaccine. Three weeks post vaccination, serum was collected for antibody analysis and the birds were challenged with the velogenic strain, CA2002 ENDV. The birds were examined daily and were monitored for virus shedding at regular intervals. All vaccines except for the allantoic fluid control induced greater than 90% protection to clinical disease. Further analysis among vaccines will be presented.



148.

The Role of Intergenic Sequences in Pathogenesis of Newcastle Disease Virus

Yongqi Yan, Daniel J. King, and Siba K. Samal
VA-MD Reg. College of Vet Med
8075 Greenmead Drive
College Park MD 20742

Newcastle disease is an important infectious disease of poultry worldwide. The virulence and pathogenesis of Newcastle Disease Virus (NDV) are not well understood. We have examined the role of the non-transcribed intergenic sequences (IGS) in NDV transcription and pathogenesis. Several recombinant NDVs with altered intergenic regions, both in length and sequence, were generated by reverse genetic techniques. The level of transcription of mRNAs from these recombinant viruses was compared to that of the parental virus. Pathogenesis and virulence of the recombinant NDVs were evaluated in chickens. Our results showed that the IGS can modulate the pathogenesis of NDV.

PARASITIC DISEASES

149.

Tiamulin and Semduramicin: Effects of Simultaneous Administration on Performance and Health of Growing Broiler Chickens

Kenneth W. Bafundo, Annette Schuhmacher, and Jurgen Gropp
Phibro Animal Health Corporation
65 Challenger Road
Ridgefield Park, New Jersey 07660

A 35-day study was performed to evaluate the compatibility of the anticoccidial semduramicin (SEM) and tiamulin (TIA), a pleuromutilin antibiotic known to negatively interact with several ionophores. Results indicated that simultaneous administration of TIA and SEM during the third week of the test transiently reduced water and feed intake resulting in a temporary growth depression. By day 35 however, performance, histopathological and hematological parameters were unaffected by treatment. Neither mortality nor long-term effects on performance occurred in broilers. These results differ markedly from those reported for TIA and other ionophores, and indicate that TIA and SEM can be used concurrently with little risk to broiler flocks.



150.

The characterization and localization of the protective antigen SO7 in developmental stages of *Eimeria tenella*

Raymond H Fetterer, Mark C. Jenkins, and Katarzyna B. Miska

Animal Parasitic Diseases Laboratory, Animal and Natural Resources Institute, Henry A. Wallace Beltsville Agricultural Research Center, Bld 1040 Room 206, BARC-East, Beltsville, Maryland 20704

We have undertaken the characterization of the recombinant antigen SO7, which has been previously shown to protect chickens against infection by several *Eimeria* species. Molecular, biochemical and microscopic techniques demonstrate that SO7 is transcribed in the unsporulated oocyst of *E. tenella* but distribution of the protein is restricted to the sporulated oocysts and sporozoites (SZ). SO7 is highly concentrated in the SZ refractile bodies with some limited distribution in the apical complex and is released from the SZ upon intracellular invasion. The results suggest that SO7 is closely associated with the sporozoite refractile body and may be necessary for cell invasion.

151.

Biological Characteristics of the Lesser Species of Chicken *Eimeria*

Steve Fitz-Coy

Schering-Plough Animal Health
5638 Royal Mile Blvd, Salisbury, MD 21801

Round to almost round, *Eimeria mitis* oocysts are distinguishable from *E. acervulina*, *E. mivati*, *E. hagani*, and *E. praecox*. *Eimeria acervulina* and *E. praecox* are ovoid, *E. mivati* broadly ovoid to almost ellipsoidal. *Eimeria hagani* are broadly ovoid. The oocysts of *E. mivati* averaged smallest and *E. praecox* largest. *Eimeria acervulina*, *E. praecox* and *E. hagani* parasitized the anterior third of the small intestine, anterior to the yolk sac diverticulum (ysd) and not found invading the ysd. *Eimeria mivati* and *E. mitis* parasitized the entire small intestine, ysd, large intestine and ceca.



152.

Toxoplasmosis in Aviary Zebra Finches

Scott D. Fitzgerald, Matti Kiupel, and Heather Teater

Diagnostic Center for Population & Animal Health, Michigan State University
DCPAH, 4123 Beaumont Road
Lansing, MI 48910-8104

A midwestern zoological park had a walk-through aviary containing a mixed display of zebra finches (*Taeniopygia guttata*) and budgerigars. During the summer of 2005, three finches died without premonitory signs over a three week period. The entire viscera from each bird was fixed in formalin and submitted for histopathologic examination. All three birds had varying degrees of necrotizing and granulomatous hepatitis and splenitis, associated with the presence of numerous intracellular protozoal cysts. Principal differentials included *Toxoplasma gondii*, *Atoxoplasma* sp., and *Isospora* sp. Based on histomorphology, tissue distribution, ultrastructure, and immunohistochemical staining, a diagnosis of Toxoplasmosis was made.

153.

**Investigation of the Efficacy of Essential Oil Product on *Histomonas meleagridis*
in-vitro and *in-vivo***

Hafez M. Hafez and Ruediger Hauck

Institute of Poultry Diseases, Free University of Berlin
Königsweg 63
D-14163 Berlin, Germany

Herbal compounds and essential oils which are used as feed aroma substances, were shown to have antibacterial and antiprotozoal effects. The aim of this study was to investigate the effect of an herbal aroma product on *Histomonas meleagridis* in turkey poults in vivo and in vitro. The obtained results revealed different product depending results. And some products seem to have some effects in-vivo.



154.

Necrotic Enteritis Association with *Eimeria acervulina* and *E. maxima*

G. F. Mathis and C. L. Hofacre

Southern Poultry Research, Inc.; 2011 Brock Road, Athens, GA 30607
The University of Georgia; Population Health, Athens, GA 30602-4875

The disease Necrotic enteritis (N.E.) is caused by the bacteria *Clostridium perfringens* (C.P.). C.P. is a normal inhabitant of the broiler chickens' intestines; therefore, it is an opportunistic bacteria when it causes disease. The poultry industry has used antibiotics in the feed to prevent the growth of C.P. and its' subsequent exotoxin production for the last 40 years. Today with legislative initiatives and especially consumer concerns for the use of antibiotics in feed, many companies are limiting or discontinuing their use altogether. This has resulted in an increasing incidence of N.E. If the poultry industry is going to be successful in growing broilers without antibiotics, we must determine what factors predispose chickens to allow the ubiquitous organism C.P. to grow and elicit its toxin. It is known that intestinal damage by coccidia can increase the incidence of N.E. We also know that the control for 1 *Eimeria* species may not fully control another *Eimeria* species. There are numerous publications where N.E. has been reproduced with *E. acervulina* or *E. maxima* or both. However, there has been no work that has looked at whether the disease severity of N.E. is affected by the species or the interaction of 2 species of *Eimeria*. In this study, we reproduced N.E. as evidenced by mortality using *E. acervulina* alone (8%), *E. maxima* alone (42%) or both (18%). We were also able to show that the *E. maxima* alone resulted in greater weight reduction than the combination of the 2 coccidia or the *E. acervulina* alone. This research indicates that the primary coccidian to control for prevention of N.E. must be *E. maxima* and that having a concurrent *E. acervulina* infections present in the same intestine may limit the ability of *E. maximia* to damage the intestine resulting in clinical N.E.

155.

Poultry Coccidiosis Control Programs: Live Vaccines

Greg F. Mathis

Southern Poultry Research, Inc.
2011 Brock Road, Athens, GA 30607

Resistance, banning certain anticoccidial, tissue residue issues, and increased interest in drug free birds have simulated the use of live coccidial vaccine usage. When properly utilized, vaccines can perform at parity with anticoccidial programs in birds of similar weight. New methods of administration such as by in-ovo injection, vaccine dilutes, SPF bird usage, and attenuated vaccines are being introduced. There is variation among vaccines in degree of anticoccidial sensitivity. It has been demonstrated that vaccination with a vaccine that contains sensitive strains has the potential to shift a coccidial population toward being more sensitive.



156.

Molecular analysis of the content and diversity of *Eimeria* species present in litter of local poultry facilities using ribosomal DNA (rDNA) sequencing

Katarzyna B. Miska, Mark C. Jenkins, and Spangler Klopp

USDA/ARS

10300 Baltimore Ave Bldg. 1042 BARC-East
Beltsville, MD 20705

Understanding the genetic diversity of *Eimeria* species present in poultry facilities may assist in tracking virulence and drug-resistance patterns of these coccidian parasites. As a first step in this effort, the ITS-1, ITS-2, and 5.8S rDNA regions of DNA derived from *Eimeria* oocysts in litter from 5 poultry facilities were amplified, cloned, and sequenced. Sequences from all but *E. necatrix* were obtained. *Eimeria maxima* sequences displayed the most sequence diversity, but this diversity did not appear to be specific to any of the 5 samples. Also, *Eimeria praecox* sequences were obtained which were significantly different to those previously described.

157.

Chemical Constituents and Preliminary Antiparasitic Activity of *Ficus Platyhylla* (DEL)

By

Rehab Mousa* and Salah Moussa**

*-Pharmacist, Al-Azhar Univ.

** - Dept. Of Poultry dis.

Fac. Vet. Med., Assiut Univ., Egypt

10% methanolic extract of the plant was prepared and tested for its anthelmintic and anticoccidial effect . A marked decrease in the number of *Ascaridea galli* worms was noticed in in treated chickens. A marked decrease in lesion score of *Eimeria tenella* infected chicks was noticed in treated birds. The antiparasitic effect increased markedly with increase of the given dose.

158.

Cytological Diagnosis of Coccidia Infection in Turkeys

Claudia Osorio,^A Steven Clark,^B Michael Martin,^A and H. John Barnes^A

^A Poultry Health Management, College of Veterinary Medicine,
North Carolina State University, 4700 Hillborough Street, Raleigh, NC 27606

^B Alpharma Animal Health, Inc., Fort Lee, NJ 07024

Coccidia infection in turkeys causes significant economic losses because of decreased growth and feed utilization. To detect infection, gross evaluation of the intestinal tract and a microscopic evaluation of a wet smear are used as routine diagnostic method. Cytology is infrequently used as a method for the diagnosis of poultry diseases. The aims of this study were to determine how useful cytology would be for diagnosing coccidia infection in turkeys, and to compare cytology with other methods of diagnosis (wet smear, histopathology). Histopathology was used as the "gold standard" for comparing diagnostic technics. Sixty-one turkeys between 2 and 6 weeks of age from 16 flocks belonging to two integrators in North Carolina were evaluated. Wet smears, mucosal impressions, and a tissue sample were obtained from the upper and middle jejunum, ileum, and cecum of each turkey. Mucosal impression smears were stained with a commercial "dip" stain used for hematology.

Coccidia in various stages were readily identified by cytology, especially when the bird was heavily infected. Cytology, wet smears, and histopathology detected coccidia infection in 23 (37.7%), 17 (27.8%), and 27 (44.3%) birds respectively. Cytology correlated well (83.6 %) with histopathology and wet smears correlated well (80.3 %) with cytology. These results indicate that cytology compares favorably with other diagnostic methods and is a rapid, inexpensive, and accurate method for diagnosing coccidia infections in turkeys.

159.

Histomonas meleagridis: genotyping of isolates using a novel technique: C-profiling

Harold M.J.F. van der Heijden, Wil J.M. Landman, Sophie Greve, and Ron Peek

GD (Animal Health Service)

R&D; P.O.Box 9

NL-7400 AA Deventer; The Netherlands

Identification of *Histomonas meleagridis* subtypes might prove useful for epidemiological studies, e.g. correlation of infections between flocks or species, monitoring of intervention measures or virulence differences between strains. C-profiling is a novel genotyping method for protozoan pathogens, based on PCR and sequencing of AT-rich ITS-1 sequences. Among isolates from histomoniasis outbreaks in six Dutch turkey and chicken flocks three *H. meleagridis* genotypes were identified. Type I and II were associated with clinical disease. In two flocks recovered from an histomoniasis outbreak, a type III strain was found that was also morphologically slightly different from the type I and II isolates.



PNEUMOVIRUS

160.

Comparative analysis of the virulence of early and recent isolates of avian metapneumovirus of turkey origin from Minnesota

Dr. K. V. Nagaraja, Binu T. Velayudhan, Sally Noll, and David A. Halvorson
University of Minnesota
205 Vet Sci Bldg
1971 Commonwealth Ave
Saint Paul, MN 55108

The virulence of early (1997) and recent (2003) isolates of avian metapneumovirus (aMPV) from USA was examined in turkeys. The change in disease causing potential of these viruses was analysed. Briefly, eighty two-week-old turkeys were divided into five groups. Group 1 was kept as non-infected controls. Group 2 was inoculated with Vero cell propagated early isolate. Birds in group 3 were inoculated with Vero cell propagated recent isolate. Groups 4 and 5 were inoculated with allantoic fluid propagated early isolate and chicken embryo fibroblast propagated recent isolate, respectively. We examined clinical signs and histopathological changes in turkeys post-infection. Clinical sign scoring of infected turkeys demonstrated a more pronounced clinical disease in birds inoculated with the recent isolate. Nasal turbinate and trachea showed more severe histological changes in the birds inoculated with recent isolates than those with early isolates. The findings of the present work clearly indicated that the recent isolate produced more severe clinical signs and histopathological changes in the infected turkeys compared to the early US isolate of avian metapneumovirus from turkeys.

161.

Production and Characterization of Monoclonal Antibodies Produced against Avian Metapneumovirus Subtype C which React against the Nucleocapsid Protein

Qingzhong Yu, Carlos Estevez, and Darrell R. Kapczynski
Southeast Poultry Research Laboratory, Agricultural Research Service,
United States Department of Agriculture, 934 College Station Road
Athens, Georgia 30605

Monoclonal antibodies (MAbs) were prepared against avian metapneumovirus (aMPV) subtype C virus (aMPV/Minnesota/turkey/1a/97). Six MAbs were selected based on ELISA activities and characterized by isotyping, neutralization test, Western blot analysis, and immunohistochemistry assay. The results showed all six MAbs reacted with the nucleocapsid (N) protein of aMPV, but did not neutralize aMPV infectivity at a detectable level. Three MAbs (3E, 9D and 12C) belonged to IgG1 subclass, whereas the other three (5D, 8E and 16E) were related to the IgG2a subclass. These MAbs provide new tools and methods for investigating aMPV infection, pathogenesis, and diagnosis of aMPV disease.



REOVIRUS

162.

Characterization of a Novel Reovirus Isolated from Meat Birds in Central Georgia, USA

Sam Christenberry and Holly S. Sellers

Perdue Farms, Inc.

P.O. Box 550

Candor, NC 27229

Reported clinical manifestations of Reovirus infection in the literature include Runting and Stunting Syndrome (RSS) as well as viral arthritis/tenosynovitis. Clinical manifestations are dependent upon the age of the bird at the time of infection, the virus pathotype and the route of exposure.

A novel Reovirus isolated from meat type chickens in Central Georgia, USA was characterized by molecular techniques and challenge studies in SPF chickens. Sequence analysis of the viral genome revealed the virus to be significantly different than the types commonly isolated in the United States. In challenge studies, both footpad and subcutaneous inoculation of the virus produced histopathologic lesions consistent with a viral challenge. Significant differences in bursa weight and body weight were found among both subcutaneous and footpad challenged birds when compared to controls. There were no significant differences between bursa weight and body weight of the subcutaneous and footpad challenge groups when compared to each other.

163.

Reovirus Progeny Challenge Protection in Broiler Flocks with Differing Maternal Immune Status

Kalen Cookson¹ and Joe Giambrone²

Fort Dodge Animal Health¹ and Auburn University²

963 Hawk Creek Trail

Lawrenceville, GA 30043

Reovirus has received growing attention over the past 3-4 years. Early reovirus infections can result in poor weight gains, uniformity issues and leg problems. Reovirus infections might also play a role in cystic enteritis, a condition affecting broilers in the Southeast. To date, we have conducted three progeny challenge studies in 11 broiler flocks to measure protection from malabsorption strain 2408 at either 3 days of age (intratracheal challenge) or 10 days of age (foot pad inoculation). Protection rates were compared to the serological profiles to determine what level of passive immunity is necessary to protect against reovirus-related problems in a high challenge environment.



164.

Case Report: Accidental Vaccination of Baby Chicks with a Virulent Reovirus Vaccine

Louise Dufour-Zavala and Guillermo Zavala

GA Poultry Lab

P.O.Box 20, Oakwood, GA 30566

Broiler Breeder Pullet chicks were accidentally vaccinated with a virulent reovirus vaccine at one day old, resulting in over 40% mortality over the first 5 days. The case was documented through histopathology and virus isolation, and experimental reproduction. The virus caused severe inflammatory lesions in the heart and liver.

165.

Epidemiological Studies of Avian Reovirus Infection in the Broilers

Jong-man Kim, In-Pil Mo, and Jin-Seok Song

Chungbuk National University College of Veterinary Medicine Avian Disease Laboratory
48 Gaeshin-dong Heungduk-gu Cheongju, Chungbuk 361-763 Korea

Avian reovirus (ARV) infection has been associated with enteric disease, characterized by mainly poor feed conversion and stunting in broiler. Although ARV infection has been recognized as one of the most important disease in the Korea poultry industry, detailed epidemiological study has not been conducted. We selected 5 different commercial broiler farms with a history of growth retardation, lameness and poor feathering and investigated the infection of ARV using serological, virological and molecular techniques. We found the increased antibody titers against ARV and also isolated ARVs which will be evaluated for their pathogenicity.



166.

Molecular investigations of avian reoviruses

Doerte Lueschow, Olivia Gooß, Christine Prusas, and Hafez M. Hafez
Institute of Poultry Diseases, Free University of Berlin
Königsweg 63
D-14163 Berlin, Germany

Avian reoviruses are a diverse group of pathogens which are involved in a wide variety of diseases in poultry. In the present investigations a RT-PCR combined with restriction enzyme analysis was established for identification and characterization of avian reoviruses. The designed primers framed a region within the S1 segment and were able to amplify reovirus RNA isolated from chicken and turkey with various disease conditions. Restriction profiles revealed genetic variations among the different isolates, but allowed no definitive classification. In addition, a further characterization of selected isolates was carried out by polyacrylamide gel electrophoresis of viral RNA.

SALMONELLA

167.

Histopathological, bacteriological, and serological investigation of *S. Enteritidis*-challenged broilers fed with diets supplemented with non-immunized egg yolk powder

Agnes C. Agunos, Umaporn Silphaduang, and Yoshinori Mine
Department of Food Science, University of Guelph
Guelph, Ontario, Canada, N1G2W1

Chicken consumption is a risk factor in *Salmonella enterica* serovar Enteritidis (SE) infection in humans. SE is widely distributed in commercial chicken flocks and high levels of cecal carriage and shedding may lead to broiler meat contamination. The present study assessed the efficacy of non-immunized egg yolk (EYP) powder for the reduction of SE in broilers, a follow-up study on the effects of EYP in layers. Feeding EYP during the first two weeks of life reduced SE shedding at 23 days post-infection (dpi) with a concomitant reduction of cecal carriage and clearance of SE in the livers. When fed in the face of an infection, EYP significantly reduced the SE shedding after a 10 day feeding with a restricted SE colonization in the liver, spleen, and cecal wall. Histopathological evaluations and serological responses of the birds to SE are presented.

168.

Use of an inactivated *Salmonella* vaccine in an Australian Poultry operation

Peter J. Groves and Anthony Pavic

Zootechny Pty Ltd
PO Box 49
Austral NSW 2179
AUSTRALIA

An integrated broiler producing company in Australia began using a trivalent inactivated *S. enterica* vaccine in its breeder farms in early 2004. The vaccine contained inactivated cultures of *S. Typhimurium*, *S. Mbandaka* and *S. Orion*, representing sero-groups B, C and E respectively.

Over the subsequent 18 months, a major shift in the *S. enterica* serovars isolated from environmental swabs from the breeder farms was observed, with the prevalence of smooth serogroups being replaced by rough types. The overall outcome has shown promising trends in reduction the prevalence of *S. enterica* subspecies of concern to human health.

169.

**Salmonella Shedding following Necrotic Enteritis Challenge:
Efficacy of 2 Feed Additives in this Model**

Charles L. Hofacre,¹ Greg Mathis,² Sharon Heins Miller,³ and Mark LaVorgna³

¹The University of Georgia, College of Veterinary Medicine,
Department of Population Health, 953 College Station Road, Athens, GA;

²Southern Poultry Research, 2011 Brock Road, Athens, GA;

³Alpharma Inc., One Executive Drive, Fort Lee, NJ

The poultry industry is experiencing consumer and regulatory pressures from 3 directions. There is pressure to reduce the level of salmonella on the final product. There is pressure to reduce or eliminate the use of antibiotic feed additives. There is environmental pressure to eliminate the use of organic arsenical feed additives. All 3 of these issues affect or are affected by the intestinal microbial flora. It has been documented by Lee et. al that use of the antimicrobial feed additive bacitracin methylene disalicylate (BMD) can shift the intestinal microflora from primarily a lactobacilli population to a primarily clostridia population. It has also been shown that *Eimeria tenella* infection can result in higher infection and shed of *Salmonella enteritidis* and *S. typhimurium*. Additionally, it has been theorized that necrotic enteritis caused by a type A *Clostridium perfringens* may increase salmonella shedding.

This study used a *C. perfringens* challenge model using fishmeal in a corn soy diet for the first 14 days of feed, a 25 X dose of a broiler coccidial vaccine and a toxigenic *C. perfringens* to determine if treatment with 3-Nitro-4 hydroxyphenylaronic acid (3-Nitro) or BMD or both would reduce the N.E. lesions, mortality and broiler performance, as well as, reduce the colonization and shedding of *S. heidelberg* in this 30 pen floor pen study to 42 days of age broilers.



170.

Prevalence of *C. jejuni*, *E. coli*, and *Salmonella spp.* at Public Picnic Areas in Metropolitan Parks

**Peter Joseph Kobalka, Stephanie S. Gfeller-Samples, Teresa Y. Morishita,
and Sara Jankovsky**

Department of Veterinary Preventive Medicine, The Ohio State University

Campylobacter jejuni, *Escherichia coli*, and *Salmonella spp.* are human pathogens and can survive in the intestines of waterfowl. Waterfowl are prevalent in park areas and humans can come into contact with their feces or the fecal contaminated environment. For this reason, soil samples from picnic areas of several metropolitan parks were collected and cultured. Prevalence of *E. coli* was 42.5%, but no *Campylobacter* or *Salmonella* was isolated. *Escherichia coli* showed resistance to several antibiotics. The results indicated that *E. coli* can survive in the soil and can serve as a source of transmission and infection for humans.

171.

Critical Review of Egg Quality Improvement Programs in Relation to Suppression of *Salmonella Enteritidis*

Simon M. Shane

North Carolina State University (Adjunct Professor)

205 Landreth Court

Durham NC 27713

The requirements for compliance with State and the national UEP Egg Quality Improvement Programs will be evaluated in relation to their capacity to detect SE infection in flocks. The inadequacy of the current single litter drag swab assay at the end of each production cycle will be demonstrated in relation to surveillance performed at an egg production complex with known SE infection.

Recommendations for enhanced surveillance for SE will be provided based on published literature and assessment of the sensitivity of sampling techniques and sites on US multi-age in-line egg production complexes



172.

A Survey of Salmonella Serotypes from Southeastern United States from 1999-2005

Stephan G. Thayer, Christine M. Lobsinger, and Charles L. Hofacre

University of Georgia
Poultry Diagnostic and Research Center
953 College Station Road
Athens, GA 30605

Salmonella group and serotype data will be obtained from submissions to the Diagnostic Services and Teaching Laboratory PDRC for the period January 1999 through December 2005. The data will be analyzed and presented using tabular and graphical representation to present by year, by serogroup, and serotype.

173.

Systematic review of intervention strategies for *Salmonella* in broiler production and processing

Robert W. Wills, R. Hartford Bailey, and Kristin M. Clements

College of Veterinary Medicine, Mississippi State University
P.O. Box 6100
Mississippi State, MS 39762

Systematic reviews are a method of identifying effective treatments or processes based on the available evidence from a variety of sources. They differ from traditional narrative or critical reviews of literature by using a replicable, scientific methodology to collect all available information on a subject. We conducted a formal systematic review to identify evidence for effectiveness of interventions for *Salmonella* in the production and processing of broilers. This topic was chosen based on its public health significance, its importance to the poultry industry, and the volume of national and international research literature/experiences available for review.


TOXINS

174.

Poisoning of Wild Geese by Organophosphated Pesticides

Elisabeta Bianu and Daniela A. Nica
Institute For Diagnosis And Animal Health
Str. Dr. Staicovici Nr. 63 Sector 5 Cod 050557

At the beginning of November 2004 a lot of dead wild geese were found in an area situated at the north-east border of Romania. The local veterinarian sent seven bodies for laboratory analysis. Post mortem lesions observed were haemorrhagic gastroenteritis, pulmonary oedema and degenerative changes in liver and kidney. The results of bacteriological and virusological exams were negative. Chemical analysis performed on goitrous and gizzard content using TLC and GC-MS showed that the death of geese was due the poisoning with diazinon.

175.

Sodium hypochlorite toxicity in 5-day-old turkey poults

R.P. Chin
California Animal Health & Food Safety Laboratory, University of California, Davis
CAHFS-Fresno
2789 S. Orange Ave
Fresno, CA 93725

A flock of 5-day-old turkeys were accidentally administered a toxic level of sodium hypochlorite. A water sampled tested and found to contain >750ppm of chlorine in the drinking water. 160 gallons of sodium hypochlorite was used during a 3 day period. Normally 3-4 gallons would have been used in the drinking water. There was 5% mortality on day 5. Birds appeared depressed, unable to walk with apparent neurological signs. Histologically, there was bilateral encephalomalacia. Brain sodium levels ranged from 1520 to 2080ppm in 4 birds. It appears that the birds stopped drinking and eating and died of sodium toxicity.



176.

A Field Investigation of the Total Tissue Arsenic Content of Broilers Medicated with and without 3-Nitro® (Roxarsone™)

Steven R. Clark, Mark W. LaVorgna, Kurt N. Dobson, and Sharon Heins Miller
Alpharma Animal Health
599 Chestnut Creek Rd
West Jefferson, NC 28694

The purpose of this trial was to provide field data on the total arsenic content of liver and muscle tissues of commercially grown broiler chickens medicated with or without 3-Nitro® (roxarsone). Tissue samples from 205 birds (liver and breast meat) were collected, August – November 2004, at the processing plants of various companies and complexes; 105 had been medicated with roxarsone at some time during their lives and 100 were never treated with the drug. Arsenic levels of all breast muscle and liver samples were well below the FDA's tolerance of 0.5 and 2 ppm, respectively.

177.

Evaluation of Tissue Arsenic Levels Following the Withdrawal of 3-Nitro® (Roxarsone) in the Diet

Kurt N. Dobson, Mark LaVorgna, Steven Clark, and Sharon Heins Miller
Alpharma, Inc., 2149 Hartford Dr., Fayetteville, AR 72701

The level of arsenic in animal tissue has recently been in the press. A recent study was conducted to look at the tissue level (breast meat, liver) of total arsenic after the withdrawal of Roxarsone from the diet. Broiler chickens were fed 3-Nitro® (Roxarsone) at 45.4 g/t from 30, 35 or 40 days of age. Tissues were collected from the birds at 40, 45, 50 and 55 days of age and assayed for total arsenic. The breast meat from birds fed Roxarsone that received a 5 day withdrawal or more did not differ significantly from controls.



178.

Toxicopathological effects of Ochratoxin A and its interaction with Newcastle disease in Indian layer chicken

S. Gounalan, C. Balachandran, and B. Murali Manohar

Madras Veterinary College, Tamilnadu Veterinary and Animal Sciences University
Department of Veterinary Pathology
Madras Veterinary College
Vepery, Chennai – 600 007, India

Experimental ochratoxicosis was induced in vaccinated layer chicken by feeding diet containing 0.5ppm ochratoxin A (OA) from 0 to 12 weeks of age. There was no mortality in the OA fed birds. Birds were sacrificed at 4th, 8th and 12th week of age. Grossly, slightly enlarged and congested kidneys were observed in the OA group. Microscopically, hepatitis, nephritis, ingluvitis, proventriculitis, ventriculitis and enteritis were observed in ochratoxin fed birds. There was damage to all the lymphoid organs. Decrease in the humoral and cell mediated immunity was observed in OA group. Feeding OA (0.5 ppm) for 36h in 12 weeks old layer chicken resulted in significant induction of apoptosis and necrosis in the spleen and thymus of OA group.

For ND interaction studies, experimental ochratoxicosis was induced by feeding diets containing OA (0.25 ppm) from 0 to 14 weeks of age in layer chicks. Vaccinated birds were challenged with velogenic NDV three weeks after each vaccination. Lowered humoral immunity was observed in vaccinated OA group. Cent percent mortality in the unvaccinated groups and 33-66 percent mortality in the vaccinated ochratoxin fed groups was observed. There was damage to all lymphoid organs. Thus, the ochratoxin fed birds were immunocompromised even if adequately vaccinated and predisposed to ND on challenge. Grossly, haemorrhages around proventricular papillae and petechiae in the caecal tonsils were noticed in the dead birds. Thus even at 0.25ppm level, OA affected the performance and health of layer chicken.

TUMOR VIRUSES

179.

Characterization of Various Isolates of a Naturally Occurring Recombinant Avian Leukosis Virus using Biological Assays and Polymerase Chain Reaction

Banat, Ghida R., Robert F. Silva, Willie M. Reed, and Aly M. Fadly

USDA, ARS, Avian Diseases and Oncology Laboratory,
and Department of Pathobiology and Diagnostic Investigation, Michigan State University,
3606 East Mount Hope Highway, East Lansing, MI 48823

Recently, we have isolated a naturally occurring recombinant avian leukosis virus (ALV) containing the envelope of ALV- β and LTR of ALV-J from commercial layer flocks affected with myeloid leukosis. Seven new isolates of the recombinant ALV, isolated from the same flock, were characterized using biological assays and polymerase chain reaction (PCR). The results suggested that the seven isolates are similar to the original isolate termed ADOL-AF-115-4. DNA sequences of the seven isolates of this recombinant ALV will be compared.



180.

Sensitivity and Specificity of Subgroup-Specific PCR for Detection of Avian Leukosis Virus

Sunny Cheng and Guillermo Zavala
Poultry Diagnostic and Research Center
Department of Population Health, University of Georgia
Vet. School, UGA. Athens, GA. 30602

The specificity and sensitivity of subgroup-specific PCR primers designed by Dr. K. Venugopal (Institute of Animal Health, Compton, U.K.) were examined using as templates an array of ALV proviral DNA samples from reference isolates and clinical samples. The minimum amount of detectable viral DNA was calculated using as template subgroup-specific PCR product cloned into a pCR 2.1 plasmid that was subsequently serially diluted to a PCR endpoint. All PCR primer sets designed specifically for subgroups A-D and J were highly specific and sensitive in general. However, recent unusual field isolates of avian leukosis virus in commercial layers (MAV-1) could not be detected by any of the PCR primers herein described.

181.

Co-infection and Vertical Transmission of Avian Leukosis Virus Subgroup J and Reticuloendotheliosis Virus in Chicken Flocks in China

Zhizhong Cui, Shuhong Sun, and Lucy F. Lee
Shandong Agricultural University,
College of Animal Science and Veterinary Medicine
Shandong Agricultural University
Taian 271018, P. R. China

An epidemiological survey of avian tumor viruses from 1999 to 2005 in China were performed. The results showed 50% of field isolates containing both subgroup J avian leucosis viruses (ALV-J) and reticuloendotheliosis virus from white meat-type birds or local yellow meat-type birds with myeloblastomas in livers and spleens. These viruses were propagated in SPF chicken embryo fibroblasts (CEF) cultures and verified by monoclonal antibodies specific for ALV-J and REV. Vertical transmission of ALV-J or REV in the eggs were detected individually or both. Co-infection of both viruses in chickens had significant synergism in pathogenesis of the diseases and this type of infection was commonly found in chicken flocks in China.



182.

Dissemination and Spread of a Chimeric Marek's Disease Virus Vaccine

Linda R. Gergen, Joan S. Schrader, Stephanie M. Cook, and Terri L. Wasmoen
Schering-Plough Animal Health
21401 West Center Road
Elkhorn, Nebraska 68022

A chimeric Marek's Disease Virus (MDV) vaccine has been developed by Schering-Plough Animal Health. Dissemination of the vaccine virus was evaluated in SPF chickens vaccinated at day of age. Results of virus isolation and PCR analysis will be presented.

183.

Load of MDV DNA in peripheral blood as criterion for early diagnosis of Marek's disease

Isabel M. Gimeno and Robert F. Silva
USDA ARS Avian Disease and Oncology Laboratory
3606 East Mount Hope Road, East Lansing, MI, 48823

Outbreaks of Marek's disease (MD) in vaccinated flocks occur sporadically leading to economical losses. In previous work, we have demonstrated that high load of MDV DNA in tumors is a good diagnostic criterion but it does not permit an early diagnosis of MD. In this study we have evaluated if high load of MDV DNA in peripheral blood could aid in the early diagnosis of MD. A series of experiments combining various MD vaccines and challenge viruses were conducted to simulate field conditions. Samples of blood were taken periodically and gross lesions were evaluated at the end of the experiments. Our results show that chickens that developed MD by the end of the experiments had high load of MDV DNA as early as 3 week post inoculation. Comparison of MDV DNA load in whole blood and buffy coats is also discussed.

184.

Skin leucosis induced by MDV

Mohammad Heidari, Scott D. Fitzgerald, Robert F. Silva, and Huanmin M. Zhang
US Department of Agriculture
3606 East Mount Hope Road
East Lansing, MI 48823

The aim of the present study was to evaluate the impact of feathers on the infection, pathology, and horizontal spread of Marek's Disease Virus (MDV) in featherless chickens. Layer type of featherless chickens were infected with a highly oncogenic strain of MDV and housed with their non-inoculated counterpart contact birds. The birds were kept for 8 weeks post infection and were observed on a daily basis for clinical signs, skin lesions, and mortality. Periodically, various tissues were collected from selected birds for histology, immunohistochemistry, and PCR analysis.

185.

Immunological Evaluation of Marek's Disease Virus Early Infection on the Pathogenesis of Infectious Bronchitis virus infection in broiler chickens

Hyun-Jeong Lee, Ji-Sun Kwon, Youn-Jeong Lee, Yong-Kuk Kwon, Hyung-Kwan Jang, and Chang-Seon Song
302, Laboratory of Avian Disease, College of Veterinary Medicine,
Konkuk University, #1 Hwayang-dong, Kwangjin-gu
Seoul, Korea (143-701)

In previous study, we found that Marek's disease virus (MDV) early infection in broiler farms in Korea seemed to be common features and MDV-induced immunosuppression could increase the susceptibility of respiratory virus infection such as IBV, NDV and APV.

In this study, we evaluated the effectiveness of MDV vaccination in broilers to protect the MDV-induced immunosuppression as well as to reduce the susceptibility to IBV infection. Immune status of chickens was assessed periodically by examine the T-cell activity, cytokine levels and other immunological indicators. The immunological evaluation results from this experimental study will be presented.



186.

The Role of Marek's Disease Virus Gene pp38 in Transactivation of Promoter Activities

Lucy Lee, Zhizhong Cui, and Sanjay Reddy

USDA-ARS-Avian Disease and Oncology Laboratory
3606 East Mount Hope Road, East Lansing, MI 48823

Marek's disease virus contains bi-directional promoters located between pp38 gene and 1.8-kb mRNA in the IRL region of the viral genome. The involvement of pp38 gene in up-regulating the activity of these promoters was analyzed by transient expression of chloramphenicol acetyltransferase (CAT) reporter gene. In order to study the transcriptional activity of pp38, a mutant was constructed in which pp38 gene was deleted. The CAT activities between parental rMd5 and mutant rMd5/ Δ pp38 were analyzed and compared. The results show that pp38 is critical in up-regulating MDV bi-directional promoters and producing protein products necessary for the pathogenesis of MDV.

187.

Immunohistochemical Detection of a B-cell Antigen in Lymphoid Tumors in Chickens

Susan B. Lockaby and Frederic J. Hoerr

State of Alabama Veterinary Diagnostic Laboratory
P.O. Box 2209
Auburn, AL 36831-2209

An immunohistochemical staining procedure was developed using a mouse monoclonal antibody against Bu-1, a chicken B cell antigen. The procedure was applied to lymphoid tumors occurring in formalin-fixed, paraffin-embedded chicken tissues previously submitted to the laboratory for diagnostic evaluation. Results will be presented and discussed.

188.

Polymorphisms in the Genomes of Oncogenic and Attenuated Pathotypes of Marek's Disease Virus Serotype 1

Stephen J. Spatz and Venugopal Nair
Southeast Poultry Research Laboratory (SEPRL)
934 College Station Road
Athens, GA 30605

A comparative genomics study involving Marek's Disease virus serotype 1 was initiated to determine polymorphisms present in the vaccine strain, CVI988. To achieve this, the complete DNA sequences of CVI988 and RB1B were determined and compared to the published sequences of virulent pathotypes. Nine open reading frames in the CVI988 genome differ from homologues found in the virulent pathotypes. These polymorphisms grouped as ORFs containing insertions (meq, 23 kD, RLORF6 and UL36), truncations (ORF49.1, ORF 5.5 and vIL8) and deletions (UL36 and UL49). Only one ORF (ORF 6.2), encoding a previously unidentified protein, was absent in the genome of CVI988.





50th Birthday Party

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

AAAP/AVMA Scientific Program



Honolulu, Hawaii
July 15-19, 2006

