

AAAP/AVMA Symposium & Scientific Programs



Seattle, WA July 12-15, 2009

2009 Annual Meeting Sponsors

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Fort Dodge Animal Health Merial

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AAAP Vision

The American Association of Avian Pathologists strives to prevent and eliminate the suffering and loss of poultry due to disease, to assure a safe poultryassociated food supply and to develop and use cutting edge, science-based techniques to meet these goals through the education of current and future scientists.

Mission

The American Association of Avian Pathologists (AAAP) assures prevention and elimination of suffering and loss of poultry due to disease and the provision of a safe poultry-associated food supply through providing an open exchange of scientific and practical information. Veterinary and non-veterinary researchers, public, private and industry veterinarians, technicians, poultry company owners, industries supporting poultry production representatives, and flock managers are provided cutting edge science concerning the diagnosis, treatment and prevention of poultry disease as well as food safety and animal welfare through national and regional scientific meetings, text books, manuals, and pamphlets developed and authored by the AAAP membership. It is through this effort that the AAAP has received and continues to seek global recognition for this expertise.

Founded in 1957, The American Association of Avian Pathologist (AAAP) is a not-for-profit 501c (6) organization that is allied with the American Veterinary Medical Association. DUNS# 830848508

AAAP, Inc. 12627 San Jose Blvd., Suite 202 Jacksonville, Florida 32223-8638 www.aaap.info



American Association of Avian Pathologists, Inc. 2009-2010 Board of Directors

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Dr. Mark Bland, Director Western region Poultry Veterinary Consultant

Dr. Daniel Bautista, Director Northeast Region Director/Poultry Diagnostician, University of Delaware-Lasher Laboratory

*New candidates will be elected at July 14, 2009 business meeting

Management Support

Bob Bevans-Kerr, Executive Director BK Association Management, LLC

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AAAP Foundation, Inc.

200 -20 Board of Directors

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- **Dr. Hector Cervantes**
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- Dr. Jagdev Sharma
- **Dr. Bruce Stewart-Brown**

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The AAAP Foundation is a 501c (3) public foundation to support the philanthropic goals of AAAP, Inc.

AAAP Foundation Mission

The mission of the AAAP Foundation Board is to administer charitable donations to the AAAP and to advise the AAAP Board regarding qualified charitable expenditures.

AAAP Foundation Objectives

To secure the AAAP Board's approval for the actions of the Foundation Board.

To conduct the annual meeting of the AAAP Charitable Foundation, Inc.

To communicate the actions of the AAAP Foundation to the AAAP Board and to the AAAP membership.

To facilitate efforts to encourage charitable giving to qualified AAAP programs such as scholarships, preceptorships, award endowments, scientific publications and scientific meeting support.

To administer fund raising for special projects that qualify as charitable functions such as hosting international scientific meetings or aiding in funding of archival efforts.

To evaluate proposed new charitable functions and advise the AAAP Board of the results of said evaluation.

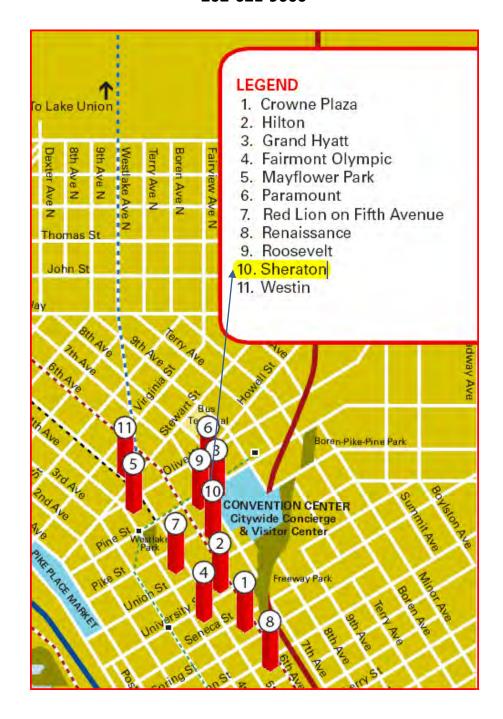
AAAP Foundation, Inc. 12627 San Jose Blvd., Suite 202 Jacksonville, Florida 32223-8638

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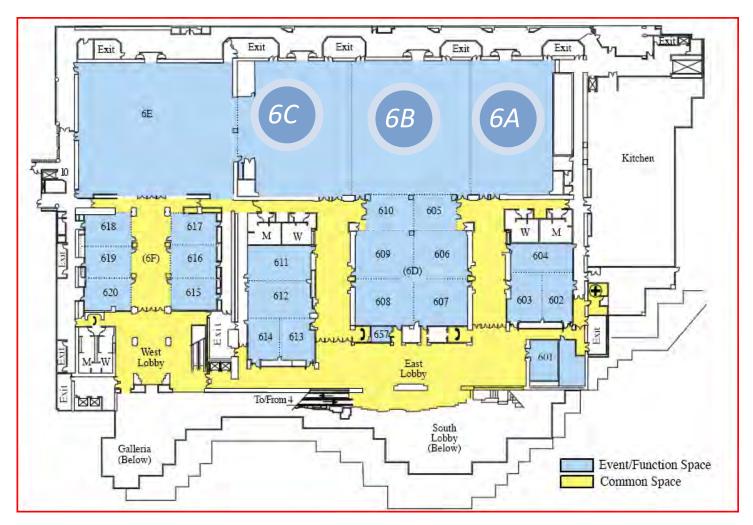
AAAP Awards Luncheon Monday July 12, 2009 12:15-2:45PM Sheraton Seattle Hotel Sheraton Grand Ballroom (2nd Floor)

1400 6th Ave 202-621-9000



Washington State Convention Center

800 Convention Place



See Floor Layout Above

Location of the AAAP Symposium: Washington State Convention & Trade Center, Room 6B

Location of the AAAP Scientific Program: Washington State Convention & Trade Center, Rooms 6B and 6C

Location of the Poultry Posters Presentation: Washington State Convention & Trade Center, Room 6A

Dates/Times of the Poultry Posters Presentation:

- Sunday, July 12 8:00 am 5:50 pm
- Monday, July 13 8:00 am 5:50 pm
- Tuesday, July 14 8:00 am 5:30 pm



Special Presentations

Monday, July 13, 2009 8:00AM- 8:30AM Keynote Speaker, Dr. Richard K. Gast

Strategies for Preharvest Control of Food-born Salmonella in Poultry Washington State Convention Center, Room 6B

Monday, July 13, 2009 3:00PM

Richard B. Rimler Memorial Paper: Jennifer M. Pfeiffer

Effects of Specific Amino Acid Changes on the Antigenicity of Hemagglutinin Molecules of Avian Influenza Isolates from Mexico Washington State Convention Center, Room 6B

Monday, July 13, 2009 3:00PM

AAAP Awards Luncheon

Sheraton Seattle Hotel, Grand Ball Room C

Tuesday, July 14, 2009 8:00AM

Reed Rumsey Award Winner: Taylor Barbosa

Expression of Viral Immunogenic Proteins in Stable Cell Culture Using a Retroviral-Based Gene Delivery System Washington State Convention Center, Room 6C

Tuesday, July 14, 2009 10:00AM- 10:30AM **Lasher History Lecture, Dr. Ray Williams** *A History of Coccidiosis to 1950* Washington State Convention Center, Room 6B

Tuesday, July 14, 2009 10:30AM- 12:00PM **AAAP Business Meeting**

Washington State Convention Center, Room 6B

Wednesday, July 15, 2009 8:00PM **Richard B. Rimler Memorial Paper: Enid T. McKinley** *Evidence of multiple recombination events in an Avian Coronavirus Infectious Bronchitis virus* Westin Hotel, Room Cascade II

Wednesday, July 15, 2009 10:30AM

Reed Rumsey Award Winner: Deirdre I. Johnson

Evaluation of Recombinant Vector-Vaccines against Infectious Laryngotracheitis (ILT) under Experimental Conditions Washington State Convention Center, Room 6C Westin Hotel, Room Cascade II



AVMA Annual Convention July 11-15, 2009

AAAP Schedule of Events The Westin Seattle Hotel

Time	Meeting Name	Contact Person	Room
	Friday, July 10, 2009		
7:00am – 5:00pm	AAAP Board of Directors Meeting	Janece Bevans-Kerr	Elliott Bay
	Saturday, July 11, 2009		1 ?
7:00am – 10:30pm	AAAP Foundation Board Meeting	Janece Bevans-Kerr	Elliott Bay
10:00pm – 5:00pm	AAAP Board of Directors Meeting	Janece Bevans-Kerr	Elliott Bay
7:00am – 8:00 pm	ACPV Exam	Janece Bevans-Kerr	Cascade Ballroom 1
7:00am – 8:00 pm	ACPV Exam # 2	Janece Bevans-Kerr	St. Helens
7:00am – 10:00 pm	ACPV Exam # 3 – (Exam Grading Room)	Janece Bevans-Kerr	Adams
8:00am – 4:00pm	Association of Veterinarians in Broiler Production	Scott Westall	Vashon II
7:00am- 9:00am	AVBP Breakfast c/o Merial Select	Anita Strange	Whidby Room
12:00pm-1:00pm	AVBP Lunch c/o Fort Dodge	0	Whidby Room
	Association of Veterinarians in Turkey Production Lunch Sponsored by		
12:00pm – 5:00pm	Alpharma	Steven Clark	Olympic
2:00pm – 5:00pm	AAAP Histopathology/ Case Report Interest Group	H.L. Shivaprasad	Cascade Ballroom 2
4:00pm – 5:00pm	AAAP Biotechnology Committee	Shane Burgess	Baker
	<u>Sunday, July 12, 2009</u>		
6:00am - 8:00am	AAAP Animal Welfare Committee	Kristi Scott	Elliot Bay
6:00am – 7:30am	Georgia MAM Alumni Breakfast	Karen Grogan	Cascade Ballroom 1
6:30am – 7:30am	AAAP Drugs & Therapeutic Committee	Bret Rings	Adams
6:30am – 7:30am	AAAP Awards Committee	Darrell Kapczynski	Baker
11:30am – 1:30pm	Association of Primary Poultry Breeder Veterinarians	Kate Barger	Adams
12:00pm – 1:30pm	California Poultry Med Alumni Luncheon	Rich Chin	Olympic
2:00pm – 6:00pm	ACPV Board Meeting	Janece Bevans-Kerr	St. Helens
4:00pm – 5:00pm	AAAP Biologics Committee	Charles Broussard	Adams
4:00pm – 5:00pm	AAAP AVMA Liaison Committee	Y.M. Saif	Baker
4:00pm – 5:00pm	AAAP Food Safety Committee	Marty Ewing	Stuart
4:00pm-6:00pm	AAAP Respiratory Diseases Committee	Maricarmen Garcia	Cascade Ballroom 1B
4:00pm – 6:00pm	AAAP Diseases of Poultry Committee	David Swayne	Orcas
4:00pm-5:30pm	AAAP Tumor Virus Committee	Guillermo Zavala	Olympic
4:30pm- 6:00	Education Committee	Isabel Gimeno	Whidby
6:00pm-7:00pm	Univ. of Minnesota Center For Animal Health and Food Safety	Brendan Lee	Cascade Ballroom 1A
	Monday, July 13, 2009		
6:30am – 7:30am	AAAP Enteric Diseases of Poultry Committee	Eric Jensen	Elliott Bay
7:00am – 8:00am	AAAP Avian Diseases Editorial Board Meeting	Jagdev Sharma	Cascade Ballroom 2
6:30am – 7:30am	AAAP History Committee	John Dunn	Baker
	AAAP Toxic, Infectious, Miscellaneous & Emerging Diseases (TIME)		
6:30am – 7:30am	Committee	Scott Fitzgerald	Glacier Peak
7:00am -8:00am	AAAP Epidemiology Committee	Suzanne Young	Adams
7:00am – 9:00am	Association of Veterinarians in Egg Production	Eric Gingerich	Vashon
12:15pm- 2:45 pm	AAAP Awards Luncheon	Janece Bevan-Kerr	Sheraton Grand Ballroom D
7:30pm-11:00 pm	NC State University Poultry Health Management	David Ley	Cascade Ballroom 1C
	Tuesday, July 14, 2009	··	
7:00am – 9:00am	ACPV Reception / Annual Meeting	Janece Bevans-Kerr	Cascade Ballroom 1 & 2
10:30 am – Noon	AAAP Business Meeting (Convention Center)	Janece Bevans-Kerr	Convention Center
4:00pm-5:00pm	AAAP Isolation and Identification Committee	Louise Dufour-Zavala	Glacier Peak
Wednesday, July 15, 2009			
7:00am – Noon	AAAP Board of Directors Meeting	Janece Bevans-Kerr	Elliott Bay



2009 AAAP Symposium North American Broiler Welfare Symposium Sunday, July 12, 2009

Suliday, July 12, 2009				
Washington State Convention CenterBall Room 6B				
Title	Speaker			
Welcome and Overview				
Welcome/ Housekeeping	Dr. Gail Golab Dr. Suzanne Millman			
B:00-8:45 AM A Scientific Approach to Broiler Welfare				
Moderator: James Barton	-			
Genetics & Hatchery				
Broiler Breeder Welfare; Genetics and Management	Dr. Michael Martin			
	Dr. Donna Hill			
Moderator: Linnea Newman				
Broiler Rearing and Harvesting				
Broiler Welfare at the Farm	Dr. Tim Cummings			
Market Age Broiler Harvesting	Dr. Robin Gilbert			
11:45- 1:00 PM Lunch				
Moderator: Robert Williams				
Slaughter				
Poultry Welfare: Farm to Plant	Dr. Ken Opengart			
Technical and Financial Considerations Relating to the Stunning of Broilers in the U.S.	Dr. Simon M. Shane			
Processing Plant Factors Related to Broiler Welfare	Dr. Sarge Bilgili			
Break				
Moderator: Mark Bland				
Auditing				
Welfare Auditing in a Conventional Broiler Farming System	Dr. James T. Barton			
Summary				
Summary and Future	Dr. Suzanne Millman			
	Title Welcome and Overview Welcome/ Housekeeping A Scientific Approach to Broiler Welfare Moderator: James Barton Genetics & Hatchery Broiler Breeder Welfare; Genetics and Management Animal Welfare from a Hatchery Perspective Break Moderator: Linnea Newman Broiler Rearing and Harvesting Broiler Welfare at the Farm Market Age Broiler Harvesting Lunch Moderator: Robert Williams Slaughter Poultry Welfare: Farm to Plant Technical and Financial Considerations Relating to the Stunning of Broilers in the U.S. Processing Plant Factors Related to Broiler Welfare Break Moderator: Mark Bland Auditing Welfare Auditing in a Conventional Broiler Farming System System Summary			



AAAP 2009 Scientific Program

American Association of Avian Pathologists

July 12-15, 2009 Washington State Convention Center Seattle, Washington

	Monday, July 13, 2009	
	Moderator: Gregorio Rosales	
8:00 AM	Keynote Speaker: Dr. Richard K. Gast Room: 6B	
		f Food-borne Salmonella in Poultry
	Session I (Room 6B)	Session II (Room 6C)
	Moderator: Danny Magee	Moderator: Erika Spackman
8:30 AM	Evaluation of Paw Lesions through Broiler Grow	Astrovirus associated with Turkey Viral
	Out: A Field Study	Hepatitis
	Suzanne D. Young, Anel Atencio, and Ken N.Opengart	Shivaprasad H. L. and Mary J. Pantin-Jackwood
8:45 AM	Spinal Abscesses Due to Enterococcus	Inclusion Body hepatits as a vertically
	Cecorum in Broiler Chickens - An Emerging	transmitted disease in Broiler chickens
	Disease?	
	Eric N. Gingerich, John H. Barnes, Robert L. Owen,	Susantha Gomis, Samantha Ekanayake, Davor
	Shelley C. Rankin	Ojkic, Suresh Tikoo, Bob Goodhope, Philip Willson
9:00 AM	Case Report: A Control Strategy for	Development of a recombinant vaccine against
	Histomoniasis on a Replacement Broiler	runting and stunting syndrome in chicken
	Breeder Farm	6 6 7
	Mark A. Burleson	Egbert Mundt, Holly Sellers, Guillermo Zavala
9:15 AM	Histomoniasis in Commercial Tom Turkeys	Changes in pituitary gland, primary and
	Exacerbated by a Faulty Nipple Drinker System	secondary immune organs associated with
		runting and stunting syndrome
	David V. Rives	Mohamed El-Gazzar, Holly Sellers, Egbert Mundt,
		Stephen Collett.
9:30 AM	Epidemiology of an Unique Gangrenous	Enteric disease in broiler chickens following
	Dermatitis Outbreak	experimental infection with chicken parvovirus
	Philip A Stayer, Jackson L McReynolds, Mark A	Laszlo Zsak, Michael J. Day, Keith O. Strother,
	Burleson	
	Break 9:4	
	Moderator: John Smith	Moderator: Mary Pantin-Jackwood
10:00 AM	Review of Virulent Newcastle Disease in	Development of an ELISA assay to detect
	Cormorants A Case Report	chicken parvovirus antibodies
	Sherrill Davison, Eric N. Gingerich and Lynn E.	Keith O Strother, Laszlo Zsak
	Stephens	
10:15 AM	Performance Problems in Free-Range Layers	Assessment of Reovirus Vaccination on
10:15 AM	Performance Problems in Free-Range Layers due to the IBV-QX Strain: Diagnosis and	Assessment of Reovirus Vaccination on Blackleg Trims in Commercial Broilers
10:15 AM	Performance Problems in Free-Range Layers due to the IBV-QX Strain: Diagnosis and Prevention	Blackleg Trims in Commercial Broilers
	Performance Problems in Free-Range Layers due to the IBV-QX Strain: Diagnosis and Prevention Franz Sommer, Verena Seger	Blackleg Trims in Commercial Broilers Timothy S. Cummings, Phil Stayer, Jason Cater
10:15 AM	Performance Problems in Free-Range Layers due to the IBV-QX Strain: Diagnosis and Prevention Franz Sommer, Verena Seger Pathology and economic impact of a severe	Blackleg Trims in Commercial Broilers Timothy S. Cummings, Phil Stayer, Jason Cater Genetic characterization of avian pox viruses
	Performance Problems in Free-Range Layers due to the IBV-QX Strain: Diagnosis and Prevention Franz Sommer, Verena Seger Pathology and economic impact of a severe outbreak of infectious coryza in non-vaccinated	Blackleg Trims in Commercial Broilers Timothy S. Cummings, Phil Stayer, Jason Cater Genetic characterization of avian pox viruses using DNA isolated from formalin fixed tissue
	Performance Problems in Free-Range Layers due to the IBV-QX Strain: Diagnosis and Prevention Franz Sommer, Verena Seger Pathology and economic impact of a severe outbreak of infectious coryza in non-vaccinated broiler breeders.	Blackleg Trims in Commercial Broilers Timothy S. Cummings, Phil Stayer, Jason Cater Genetic characterization of avian pox viruses using DNA isolated from formalin fixed tissue sections
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10:30 AM	Performance Problems in Free-Range Layers due to the IBV-QX Strain: Diagnosis and Prevention Franz Sommer, Verena Seger Pathology and economic impact of a severe outbreak of infectious coryza in non-vaccinated broiler breeders. Guillermo Zavala, Louise Dufour-Zavala, Martin Smeltzer, Mark W. Jackwood and Sunny Cheng Case Report: Septicemic Erysipelas in Chukar	Blackleg Trims in Commercial Broilers Timothy S. Cummings, Phil Stayer, Jason Cater Genetic characterization of avian pox viruses using DNA isolated from formalin fixed tissue sections Deoki N. Tripathy and Bahaa A. Fadl-Alla Determination of the Prevalence of Pigeon

	Session I (Room 6B)	Session II (Room 6C)
11:00 AM	An unusual case of hepatitis in ring-necked	Testing of a New Disinfectant Process for
11.00 AM	pheasants (case report)	Poultry Viruses
	Eva Wallner-Pendleton, Patricia Dunn, Greg Ning,	Lauren Appleby Gay and Egbert Mundt
	Huaguang Lu, Benjamin Lucio, Joan Smyth,	Lauren Appleby Gay and Egbert Mundt
11:15 AM	Elizabeth Buckles	On the offectivity of a veccination achodula
11.15 AM	Oropharnyngitis with Intralesional	On the effectivity of a vaccination schedule
	Dermatophilus-like Bacteria in a Breeder	against aMPV in Brazilian commercial layers as
	Chicken.	measured by RT-PCR, DNA sequencing, ELISA
		and clinical symptoms
	Frederic J. Hoerr, Haroldo Toro, Heather Busby,	Laura YB Villarreal, Delair A Bolis and Paulo E
	Sandra Ewald	Brando
11:30 AM	Water management of Pseudomonas cepacia in	•
	brown-egg layer pullets	Disease Virus (NDV) Maternal Antibodies(Mabs)
		Vaccinated with the Recombinant
		HVF/F(NDV),INNOVAX-ND-SB
	D.A.Anderson and D.Pennock	Rudolf G. Hein , Gwen F. Slacum and Phyllis A.
		Lynch
11:45 AM	Cyanotic Broiler Breeder Males: Why Are Those	Transmission of virulent Newcastle disease
	Heads Purple?	virus (NDV) between unvaccinated, sub-
	•	optimally vaccinated, and well-vaccinated SPF
		chickens
	Danny L. Magee, Jason Cater, Erica Baravik, Sua	Patti J. Miller, Claudio L. Afonso
	Ann Hubbard	
12:15-		de l'anne de seus
2:45PM	AAAP Award	ds Luncheon
	Lunch serve	d at 12:30PM
	Moderator: Dave Suarez	Moderator: Lisa Nolan
	Richard B. Rimler Memorial Paper*:	E. coli survey in a broiler complex using
3:00 PM	Effects of Specific Amino Acid Changes on the	serotyping and pathotyping techniques
	Antigenicity of Hemagglutinin Molecules of	
	Avian Influenza Isolates from Mexico	
	J. Pfeiffer*, C. W. Lee, S. Jadhao, and D. L. Suarez	Kalen C. Cookson, Lisa K. Nolan and Chobi
		Raien C. Cookson, Lisa R. Noian and Chobi
2.15 014		
3:15 PM	Recombinant RCA-free Adenovirus-Vectored	DebRoy
3. 15 PIVI		DebRoy E. coli serogroup O18: Emerging isolates that
3.13 PM	Vaccine for Immunization of Chickens and Pigs	DebRoy
3. 13 MM	Vaccine for Immunization of Chickens and Pigs against Avian Influenza	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry
3.15 PM	Vaccine for Immunization of Chickens and Pigs against Avian Influenza Haroldo Toro, De-chu C. Tang, Bettina Schemera,	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry Kelly A. Tivendale, Yvonne Wannemuehler, Kathy
3. 15 PM	Vaccine for Immunization of Chickens and Pigs against Avian Influenza	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry Kelly A. Tivendale, Yvonne Wannemuehler, Kathy T. Mou, Catherine M. Logue, Ganwu Li,
3. 13 PM	Vaccine for Immunization of Chickens and Pigs against Avian Influenza Haroldo Toro, De-chu C. Tang, Bettina Schemera,	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry Kelly A. Tivendale, Yvonne Wannemuehler, Kathy T. Mou, Catherine M. Logue, Ganwu Li, Subhashinie Kariyawasam, Ashraf Hussein, and
	Vaccine for Immunization of Chickens and Pigs against Avian Influenza Haroldo Toro, De-chu C. Tang, Bettina Schemera, Soren Rodning, Joseph C. Newton	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry Kelly A. Tivendale, Yvonne Wannemuehler, Kathy T. Mou, Catherine M. Logue, Ganwu Li, Subhashinie Kariyawasam, Ashraf Hussein, and Lisa K. Nolan
3:15 PM 3:30 PM	Vaccine for Immunization of Chickens and Pigs against Avian Influenza Haroldo Toro, De-chu C. Tang, Bettina Schemera, Soren Rodning, Joseph C. Newton Pathogenicity of reverse genetics based	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry Kelly A. Tivendale, Yvonne Wannemuehler, Kathy T. Mou, Catherine M. Logue, Ganwu Li, Subhashinie Kariyawasam, Ashraf Hussein, and Lisa K. Nolan Avian Pathogenic E coli (APEC) reduction in
	Vaccine for Immunization of Chickens and Pigs against Avian Influenza Haroldo Toro, De-chu C. Tang, Bettina Schemera, Soren Rodning, Joseph C. Newton Pathogenicity of reverse genetics based reassortant H5N1 avian influenza viruses with	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry Kelly A. Tivendale, Yvonne Wannemuehler, Kathy T. Mou, Catherine M. Logue, Ganwu Li, Subhashinie Kariyawasam, Ashraf Hussein, and Lisa K. Nolan
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	Vaccine for Immunization of Chickens and Pigs against Avian Influenza Haroldo Toro, De-chu C. Tang, Bettina Schemera, Soren Rodning, Joseph C. Newton Pathogenicity of reverse genetics based reassortant H5N1 avian influenza viruses with truncated NS1 gene in chickens Samadhan J. Jadhao, David Swayne, Chang-Won	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry Kelly A. Tivendale, Yvonne Wannemuehler, Kathy T. Mou, Catherine M. Logue, Ganwu Li, Subhashinie Kariyawasam, Ashraf Hussein, and Lisa K. Nolan Avian Pathogenic E coli (APEC) reduction in Commercial Poultry Daniel Karunakaran, Tom Rehberger, Greg
3:30 PM	Vaccine for Immunization of Chickens and Pigs against Avian Influenza Haroldo Toro, De-chu C. Tang, Bettina Schemera, Soren Rodning, Joseph C. Newton Pathogenicity of reverse genetics based reassortant H5N1 avian influenza viruses with truncated NS1 gene in chickens Samadhan J. Jadhao, David Swayne, Chang-Won Lee and David Suarez	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry Kelly A. Tivendale, Yvonne Wannemuehler, Kathy T. Mou, Catherine M. Logue, Ganwu Li, Subhashinie Kariyawasam, Ashraf Hussein, and Lisa K. Nolan Avian Pathogenic E coli (APEC) reduction in Commercial Poultry Daniel Karunakaran, Tom Rehberger, Greg Siragusa, Chris Kromm
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3:30 PM 3:45 PM	Vaccine for Immunization of Chickens and Pigs against Avian Influenza Haroldo Toro, De-chu C. Tang, Bettina Schemera, Soren Rodning, Joseph C. Newton Pathogenicity of reverse genetics based reassortant H5N1 avian influenza viruses with truncated NS1 gene in chickens Samadhan J. Jadhao, David Swayne, Chang-Won Lee and David Suarez Differences between Pekin and Muscovy ducks in response to vaccination against HPAI H5N1 virus Mary J. Pantin-Jackwood, David E. Swayne, and David L. Suarez Analysis of H7 Avian Influenza Viruses by Antigenic Cartography and Correlation to	DebRoy E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry Kelly A. Tivendale, Yvonne Wannemuehler, Kathy T. Mou, Catherine M. Logue, Ganwu Li, Subhashinie Kariyawasam, Ashraf Hussein, and Lisa K. Nolan Avian Pathogenic E coli (APEC) reduction in Commercial Poultry Daniel Karunakaran, Tom Rehberger, Greg Siragusa, Chris Kromm A Novel Autotransporter Contributing to Adherence Is Necessary for the Full Virulence of Avian Pathogenic E. coli Ganwu Li, Subhashinie Kariyawasam, Yvonne Wannemuehler,Kelly Tivendale and Lisa K. Nolan

	Session I (Room 6B)	Session II (Room 6C)
4:15 PM	Passage of low pathogenic avian influenza virus	Real-time PCR in Avian Mycoplasma
	in chickens results in mutations and better	Diagnostics
	transmissibility between birds	
	Daniel Dlugolenski, Ralph A. Tripp, Mark. S.	Ziv Raviv and Stanley H Kleven
	Tompkins, Egbert Mundt	
4:30 PM	Development of DIVA Vaccines for the Control	The Displacement of Mycoplasma gallisepticum
	of Triple Reassortant H3N2 Influenza in Turkeys	Strains by Live Vaccines
	Leyi Wang, Hadi M. Yassine, Smitha Pillai, Yehia	Naola Ferguson-Noel, Victoria Laibinis, Ziv Raviv,
	M. Saif, Chang-Won Lee	Stanley H. Kleven
4:45 PM	Summary of type A influenza virus	Serology results for TS-11 vaccinated and non
	investigations for waterfowl in Ohio and on	vaccinated breeder flocks in Georgia
	Delmarva: 2001-2008	
	R.D. Slemons, D.D. Swayne, C. Driscoll, L.	Len Chappell and Dorene Seabolt
	Hindman, V. Stotts, L. Alexander, M. Shieldcastle,	
	D. Sherman, D. Senne	
5:00 PM	Large death loss in captive/release Mallard	Comparative Testing of Samples from a Broiler
	ducks due to Pasteurella multocida	Breeder Flock for Mycoplasma synoviae
	Richard M. Fulton	David H. Ley, Ziv Raviv, Naola Ferguson-Noel
5:15 PM	Survey of backyard/hobby/small production	Role of Mycoplasma anatis and Interactions
	flocks within a high density poultry area	with other Infectious Agents in the
		Tenosynovitis and Arthritis syndromes in
		commercial ducks.
	Benjamin C. Johnson, Louise Dufour-Zavala, and	Jaime Ruiz, Naola Ferguson-Noel, Benjamin Lucio-
	Martin A. Smeltzer	Martinez, Victoria Laibinis
5:30 PM	Adjo	ourn

	Tuesday, July 14, 2009	
	Session I (Room 6B)	Session II (Room 6C)
	Moderator: H. L. Shivaprasad	Moderator: Alejandro Bando
8:00 AM	Detection of Salmonella enterica serovar	Reed Rumsey Award Winner*:
	Enteritidis (SE) Antibodies in Serum Using A	Expression of Viral Immunogenic Proteins in
	Polystyrene Bead/SE Flagella Agglutination	Stable Cell Culture Using a Retroviral-Based
	Assay	Gene Delivery System
	Peter S. Holt, Kyle D. Swayne, Lara E. Vaughn, Richard K. Gast	Taylor Barbosa*, Guillermo Zavala and Sunny Cheng
8:15 AM	Use of CFSE dye and fluorescent imaging to	Detection of chicken birnavirus R11/3 in
	visualize lymphoid tissues in chickens	proventriculi and feces of experimentally
	challenged with Salmonella Enteritidis (SE)	infected chickens using a reverse transcriptase-
		polymerase chain reaction procedure
	Lara E. Vaughn, Peter S. Holt, Richard K. Gast	James S. Guy, Frederick J. Fuller, H. John Barnes, Melissa West
8:30 AM	European Union (EU) Salmonella Enteritidis Control with the Use of an Inactivated Salmonella Vaccine.	Antigenic Characterization and VP2 analysis of Delmarva IBD Field Viruses
	Rik Koopman, Rick v Oort, Ruud Hein	J. Gelb, Jr., D. J. Jackwood, E. Mundt, J. M. Harris,
		B. S. Ladman, D. Bautista, M. Ruano, M. M.
		Troeber, and C. R. Pope
8:45 AM	A Long-Term Survey of Salmonella sp. and	Isolation and Characterization of a new variant
	Campylobacter sp. in a Meat-type Poultry	Infectious Bursal Disease Virus identified by a
	Processing Plant	diagnostic approach using reverse genetics
	Spangler Klopp, Daniel Austria Bautista, Brenda R. Sample, Kathy J. Philips, and Billie Jean Wright	Vijay Durairaj, Holly S. Sellers, Egbert Mundt
9:00 AM	Turkey Slaughter Establishments Operating Under the Salmonella Initiative Program	Efficacy of single dose recombinant HVT-IBDV vaccination (Vaxxitek®) against classical and variant IBDV strains
	Becky J. Tilley, Alice L. Johnson	Francisco Perozo, Pedro Villegas, Linda Purvis, Rafael Fernandez & Julio Cruz
9:15 AM	The Utility of a Commercial Real-Time PCR Assay for Screening Salmonella sp. in Poultry Carcass Rinses and Environmental Samples	Spectrum of protection provided by passive and active immunity to IBD viruses
	Daniel Austria Bautista, Spangler Klopp, Brenda R. Sample, Kathy J. Philips, and Billie Jean Wright	Vilmos Palya, Tamas Mato, Timea Tatar-Kiss, Yannick Gardin
	Break 9:3	0-10:00AM
		Richard Chin
10:00-	Lasher History Lecture: Dr. Ray	Williams Room: 6B ccidiosis to 1950
10:30-		
12:00 PM	AAAP Busin	ness Meeting
12:00-		nah
1:00PM	Lu	nch

	Session I (Room 6B)	Session II (Room 6C)
	Moderator: Phil Stayer	Moderator: Isabel Gimeno
1:00 PM	Correlation between use of various coccidial	Comparison of water based foam depopulation
	control programs and incidence of Gangrenous	with and without CO2 gas to CO2 and Ar-CO2
	Dermatitis in an endemic broiler complex: Part	gassing
	1: Field Observations	g
	G. Donald Ritter	Eric R Benson, Mary Rankin, Keith J. Johnson.
		Robert L. Alphin
1:15 PM	Correlation between use of varioius coccidial	Effects of increased light intensity on tendon
	control programs and incidence of Gangrenous	ruptures in eight pound broilers
	Dermatitis in an endemic broiler complex: Part	
	2: Gastrointestinal microbiota	
	G. Donald Ritter, J. A. Benson, S. M. Dunham, A. P.	Annika L. Hoffman, Philip A. Stayer
	Neumann, T. G. Rehberger and G. R. Siragusa	Annika L. Honnian, T hilip A. Otayer
1:30 PM	Comparison of turkeys from flocks raised on	The beta-tubulin sequence of H. meleagridis
	farms either endemic or non-endemic for	suggests susceptibility against Benzimidazoles
	gangrenous dermatitis: Part 1 Microbiologic	and confirms the phylogenetic position of H.
	differences	meleagridis
	Michelle M. Andersen, Jodi A. Benson, Susan M.	Ruediger Hauck, Hafez M. Hafez
	Dunham , Anthony P. Neumann , Thomas G.	
1:45 PM	Rehberger and Gregory R. Siragusa	Chronological study of the nother series of
1.45 PW	Comparison of turkeys from flocks raised on farms either endemic or non-endemic for	Chronological study of the pathogenesis of
		Marek's disease virus in the eye and brain
	gangrenous dermatitis: Part 2 Gastrointestinal microbiota	
	Michelle M. Andersen, Jodi A. Benson, Susan M.	Arun K.R. Pandiri, Aneg . Cortes, Isabel M. Gimeno
	Dunham, Anthony P. Neumann , Thomas G.	Arun K.K. Fandin, Aneg . Cortes, isaber M. Gimeno
	Rehberger and Gregory R. Siragusa	
2:00 PM	Quantitative detection of C. septicum in turkey	Dimerization partners of Marek's disease virus
	flocks experiencing reoccurring episodes of	encoded Meq protein play an important role in T
	gangrenous cellulitis by real-time PCR	cell transformation
	3 . 3	
	Anthony P. Neumann, Sue M. Dunham, Jodi A.	Paulette F. Suchodolski, Blanca Lupiani, Lucy F.
	Benson, Michelle M. Andersen, Thomas G.	Lee and Sanjay M. Reddy
	Rehberger and Greg R. Siragusa	
2:15 PM	Immune reactive components of Clostridium	Protection properties of mutant rMd5 viruses
	perfringens causing Cellulitis in turkeys	expressing the Meq protein of CVI988 vaccine
		strain
	Anil J. Thachil, David A. Halvorson, and Kakambi V.	Blanca Lupiani, Sanjay M. Reddy
	Nagaraja.	
2:30 PM	Control of Turkey cellulitis using an inactivated	Factors affecting the efficacy of recombinant
	Clostridial toxoid vaccine	Marek's disease vaccine protection
	K.V. Nagaraja, Thachil A.J and D.A. Halvorson	Lucy F. Lee
2:45 PM	Prevalence of netB positive and negative	Characterization of the immune responses
	isolates of C. perfringens from healthy and	elicited by double vaccination against Marek's
	diseased animals and their ability to produce	disease
	necrotic enteritis	
	Thomas G. Martin & Joan A. Smyth	Aneg Lucia Cortes, Richard L. Witter, Isabel M.
	P 1 4 4	Gimeno
	Break 3:0	0-3:30 PM

	Session I (Room 6B)	Session II (Room 6C)
	Moderator: Mike Martin	Moderator: Pat Waknell
3:30 PM	Importance of mixed Clostridium perfringens	Effect of route of vaccination with serotype 1
	isolates in the experimental reproduction of	Marek's disease vaccines on the recruitment of
	necrotic enteritis in broiler chickens, a new	lymphocytes and macrophages in the lungs
	infection model	
	Martine Boulianne, Mathiew Belanger, Philippe	Ricardo A. Cortes, Aneg Lucia Cortes, Oscar
	Fravalo, Robert J. Moore, and Ann Letellier	Fletcher, Isabel M.Gimeno
3:45 PM	Molecular toxinotyping of Clostridium	Virus competition for shedding and tumor
	perfringens isolates from organic farms	formation over time in Marek's disease virus
		dual-infected chickens
	Jessica Brady, Omar Abu-Dahab, Carlyle Bennett,	John R. Dunn, Richard L. Witter, Robert F. Silva,
	Bill Guenter, Juan C. Rodriguez-Lecompte, James	Lucy F. Lee, Scott D. Fitzgerald, Richard M. Fulton,
	D. House	Steve R. Bolin, John B. Kaneene
4:00 PM	Ulcerative enteritis in broiler breeder chickens	Role of Pulmonary Immune Response on the
	and captive capercaillie (Tetrao urogallus) in	Efficacy of Serotype 1 Marek's Disease Vaccines
	Scotland in 2008	
	Alisdair M. Wood, Luca Bano, Ilenia Drigo	Isabel M. Gimeno, Aneg L. Cortes, Richard L.
	, alouali ini irooda, Luca Barlo, norma Brigo	Witter
4:15 PM	A decrease in FOCAL DUODENAL NECROSIS	Detection of VECTORMUNE HVT® Vaccines in
	(FDN) lesion number and severity with use of	Feather Tips by Real-Time PCR
	FDN specific Direct Fed Microbial (DFM).	
	Tammy A. Baltzley, Sue M. Dunham, Firmin Lago,	Motoyuki Esaki, Takanori Sato, Lauren Jensen,
	Jodi A. Benson, Greg R. Siragusa	Shuji Saitoh, Sakiko Saeki, Ayumi Fujisawa and
		Kristi Moore Dorsey
4:30 PM	Turkey Cellulitis Incidence in a Dexamethasone	Immunological Basis for Resistant and
	Immunosuppression Model	Susceptibility to Marek's Disease
	Geraldine R. Huff, William E. Huff, Narayan C. Rath	
4:45 PM	The Environmental Impact, Welfare and	The importance of pp38 splice variants for the
	Production Characteristics of First-Lay and	pathogenesis of Marek's disease
	Molted Turkey Breeder Hens	
	Eric C Gonder	Karel A. Schat, Priscilla H. O'Connell, Micheal S.
		Piepenbrink, Keith W. Jarosinski
5:00 PM	A survey to investigate the adoption of	Insertion of a Reticuloendotheliosis Virus LTR
	biosecurity measures among commercial broiler	into the Marek's Disease Virus Genome
	growers in Georgia	
	Fernanda C. Dorea, Dana J. Cole, Charles Hofacre	Robert F. Silva, Taejoong Kim, Jody Mays and Aly
		M. Fadly
5:15 PM	Biosecurity in a newly constructed egg	Pathogenicity of a molecular clone of Marek's
	production facility: results after 12 years	disease virus with an insert of Long Terminal
	. ,	Repeat (LTR) of reticuloendotheliosis virus
		(REV).
	David A Halvorson	Aly M Fadly, Taejoong Kim, Jody Mays and Robert
		F. Silva
5:30 PM	libA	ourn
0.00 F IVI	Auj	Juin

	Wednesday,	July 15, 2009
	Westin	n Hotel
	Session I (Cascade I)	Session II (Cascade II)
	Moderator: Charlie Broussard	Moderator: Haroldo Toro
8:00 AM	Pathogenesis of vertebral osteorthritis ("Spinal	Richard B. Rimler Memorial Paper*:
	Abscesses") in male broiler breeders casued by	Evidence of multiple recombination events in an
	enterococcus	Avian Coronavirus Infectious Bronchitis virus
	Leslie T. Martin, Michael P. Martin, and H. John	Enid T. McKinley*, Deboah A. Hilt, Mark W.
	Barnes	Jackwood
8:15 AM	Deciphering the role of Polyphosphate kinases	Genomic and Phylogenetic Comparisons of
	in Campylobacter jejuni colonization/	Emerging Coronaviruses
	pathogenesis	
	D. Gangaiah, B. Adhikari, B. Jeon, Q. Zhang , Z.	S. W. Thor, D.A. Hilt, E.T. McKinley, and M.W.
	Liu, Y. Sanad, M. Drozd, and G. Rajashekara	Jackwood
8:30 AM	Reduced fitness associated with macrolide-	Selection of Minor Viral Subpopulations within
	resistant Campylobacter in chickens	Ark-type Infectious Bronchitis Vaccine Effect
		on Tracheal Damage and Mucosal Immune
		Responses
	Oiiing Zhang, Taradan Luangtangkum, Virginia	•
	Qijing Zhang, Taradon Luangtongkum, Virginia	Vicky L. van Santen, Eunice N. Ndegwa, Kellye S.
0.45 AM	Seng, and Orhan Sahin	Joiner, Frederick W. van Ginkel, Haroldo Toro
8:45 AM	Isolation and Characterization of	Assessing Intraspatial Variation of Infectious
	Streptococcus suis from Broiler Chickens	Bronchitis Virus in the Host
	Subhashinie Kariyawasam, Patricia A. Dunn, Eva A.	Rodrigo Gallardo, Vicky van Santen, Haroldo Toro
	Wallner-Pendleton, and Suzanne Myers	
9:00 AM	Consumers: Their Perceptions and How They	Genomic Analysis of Pathogenic and Attenuated
9:00 AM		Genomic Analysis of Pathogenic and Attenuated Strains of Infectious Bronchitis Virus
9:00 AM	Consumers: Their Perceptions and How They	
9:00 AM	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael	
9:00 AM	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman	Strains of Infectious Bronchitis Virus
9:00 AM 9:15 AM	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor,
	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood
	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman The effect of metam sodium on infectivity of	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE
	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman The effect of metam sodium on infectivity of Eimeria oocysts	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE Georgia
	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman The effect of metam sodium on infectivity of Eimeria oocysts Raymond Fetterer, Mark Jenkins, Kate Miska,	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE Georgia Arun B. Kulkarni, Elena Behnke and Reynaldo
	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman The effect of metam sodium on infectivity of Eimeria oocysts Raymond Fetterer, Mark Jenkins, Kate Miska, Ronald Richardson, George Cain	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE Georgia Arun B. Kulkarni, Elena Behnke and Reynaldo Resurreccion
	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman The effect of metam sodium on infectivity of Eimeria oocysts Raymond Fetterer, Mark Jenkins, Kate Miska, Ronald Richardson, George Cain Break 9:3	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE Georgia Arun B. Kulkarni, Elena Behnke and Reynaldo Resurreccion D-10:00AM
9:15 AM	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman The effect of metam sodium on infectivity of Eimeria oocysts Raymond Fetterer, Mark Jenkins, Kate Miska, Ronald Richardson, George Cain Break 9:30 Moderator: Hector Cervantes	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE Georgia Arun B. Kulkarni, Elena Behnke and Reynaldo Resurreccion 0-10:00AM Moderator: Pedro Villegas
9:15 AM	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman The effect of metam sodium on infectivity of Eimeria oocysts Raymond Fetterer, Mark Jenkins, Kate Miska, Ronald Richardson, George Cain Break 9:30 Moderator: Hector Cervantes Molecular Characterization of Eimeria species	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE Georgia Arun B. Kulkarni, Elena Behnke and Reynaldo Resurreccion 0-10:00AM Moderator: Pedro Villegas All-Natural Compound Tested for Activity
9:15 AM	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman The effect of metam sodium on infectivity of Eimeria oocysts Raymond Fetterer, Mark Jenkins, Kate Miska, Ronald Richardson, George Cain Break 9:30 Moderator: Hector Cervantes Molecular Characterization of Eimeria species infecting turkeys	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE Georgia Arun B. Kulkarni, Elena Behnke and Reynaldo Resurreccion 0-10:00AM Moderator: Pedro Villegas All-Natural Compound Tested for Activity Against IBV in Chickens
9:15 AM	Consumers: Their Perceptions and How They Change Robert L Owen, Jeffrey Mellinger, Michael Opperman The effect of metam sodium on infectivity of Eimeria oocysts Raymond Fetterer, Mark Jenkins, Kate Miska, Ronald Richardson, George Cain Break 9:30 Moderator: Hector Cervantes Molecular Characterization of Eimeria species infecting turkeys Katarzyna B. Miska, Thilak Rathinam, Ryan S.	Strains of Infectious Bronchitis Virus J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE Georgia Arun B. Kulkarni, Elena Behnke and Reynaldo Resurreccion 0-10:00AM Moderator: Pedro Villegas All-Natural Compound Tested for Activity Against IBV in Chickens Mark W. Jackwood, Richard Rosenbloom, Michael
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	Session I (Cascade I)	Session II (Cascade II)
10:45 AM	How Eimeria praecox infection affects broiler	Analysis of Climatic Factors during Outbreaks
	intestinal immunity	of Vaccinal Laryngotracheitis
	Lindsay H. Stuard, Kate B. Miska, Mark C. Jenkins,	Louise Dufour-Zavala
	Ray H. Fetterer, Sungwon Kim, Rami A. Dalloul	
11:00 AM	Comparison of broiler performance and vaccine	Benefits of the New Intra-Muscular Wing Killed
	infectivity after Inovocox" vaccination at E18 or	Vaccination on Commercial Egg Pullets
	E19.	••
	Lauren K. Griffin, Larry Charniga, Mohamed	Hugo Medina
	Hamoud, Vivian Doelling, Michelle Miller, Rebecca	
	Poston	
11:15 AM	Improved Coccidiosis Control with Anticoccidial	Nonsuppurative myocarditis in turkeys in
	Drugs Following Vaccination with Coccivac-B	California: a retrospective study of forty five
		cases.
	Greg F. Mathis, and Matilde Alfonso and Charles	Monique S. Franca, Rocio M. Crespo, Richard P.
	Broussard	Chin, Peter Woolcock, Hulimangala L. Shivaprasad
11:30 AM	Paired House Trials for Comparing Body	Association of Gross, Microscopic, and
	Weights at 28 and 42 Days of Age in Broilers	Histopathologic Examination of Enteric Lesions
	Receiving Either Inovocox or Salinomycin	in Young Turkeys
	Jonathan L. Schaeffer, Larry M. Charniga, John	Steven R Clark, David Rives, Steven Matthis, Oscar
	Dickson, David G. Kelly and Rebecca M. Poston	Fletcher
11:45 PM	Is coccidiosis the problem?	Pathology and mortality associated with graded
		levels of melamine and cyanuric acid fed to
		young broiler chickens
	Steve Fitz-Coy	Alex Bermudez, George Rottinghaus, David
		Ledoux, Lindsay Brand, Rita Dourado, Rafael
		Murarolli, and Mengshi Lin
12:00 PM	Adjo	ourn

Avian Influenza

1 Anti-viral activity and toxicity of the Alder tree extract against avian influenza virus subtype H9N2

II Hwan Kim, Hyuk Joon Kwon, Sun Hee Cho, Young Jin Ahn, Sun Joong Kim, Seong Ryul Cho, Jeong Chan Ra, Jae Hong Kim

2 Biological Characterization of Avian Influenza Viruses Isolated from Wild Birds

Seong-Hwan Byun, Min-Jeong Kim, Hwan-Hee Kim, Chang-Hee Lee, Jeong-Hwa Shin, In-Pil Mo

3 Determination of Survivability of Avian Influenza (AI) Viruses in Poultry Litter and Experimentally Infected Chickens

Aline R. Reis, Casey W. Ritz, David E. Stallknecht, and Maricarmen Garcia

4 Establishment of neuraminidase (NA) subtype 2 specific ELISA for Influenza A virus from avian and swine origin

Maricarmen Garcia, Alice Mundt, and Aline Reis

5 Expression of an influenza virus hemagglutinin gene in a fowl adenovirus vector

Eva Nagy, Dan-Hui Yang and Peter J. Krell

- 6 In Vitro Comparison of the Cytokine Response to Avian Influenza Virus from Peripheral Blood Lymphocytes Isolated From Chickens Differing in Mx631 Gene Polymorphism. Darrell R. Kapczynski, Sandra Ewald, Emily Livant, Ludmilla Kaltenboeck, and David L. Suarez
- 7 Increased pathogenicity and altered host responses after adaptation of a mallard H5N2 LPAIV in IBDV-pre-exposed chickens.

Gloria C. Ramirez-Nieto, Chul-Hong Kim, Hyun S. Lillehoj, Haichen Song, H. L. Shivaprasad, Ivan G. Gomez-Osorio, Daniel R. Perez.

8 Interspecies Transmission of Triple Reassortant H3N2 Influenza Viruses Between Swine and Turkeys: Molecular Studies.

Mahesh Khatri, Chang-Won Lee, and Y.M. Saif

9 AIV and NDV surveillance in Peru: An update For The 2006-2007-2008 Migratory Season and H7N3-AIV strain Isolation

Rosa Gonzalez, Eliana Icochea, Armando Gonzalez, Bruno Ghersi, David Blazes, and Joel Montgomery

10 Live-attenuated H7N2 and H9N2 avian influenza viruses for potential use as in ovo vaccines

Cai Y, Song H, Ye J, Araya Y, Padmanabhan R, Perez

- 11 Pathogenicity of J5N1 A/Duck/Vietnam/201/05 reassortants in ducks
- 12 Resurgence of H5N2 Avian Influenza in Live Bird Markets in the Northeast U.S. David L. Suarez, L. Mia Kim, Janice C. Pedersen, and Dennis A. Senne
- 13 Studies of phylogeny and sequence analysis of H9N2 avian influenza virusesYanyan Huang, Beixia Hu, Sanjie Cao, Xintian Wen, Dong Xu, Mazhar Khan, Xiumei Zhang
- 14 **Susceptibility of chicken T cells to low pathogenic H5 influenza viruses.** Mahesh Khatri, Hadi M. Yassine, Yehia M. Saif and Chang-Won Lee
- 15 Viral Adaptation to Host Species-Effect of LP-AIV Passage within a Host Brian S. Ladman, Jack Gelb, Jr., Conrad Pope, Erica Spackman and Richard Slemons

Bacteria, Miscellaneous

16 Amyloid arthropathy associated with various bacteria in Brown Leghorn Chickens

Shivaprasad H. L., M. Franca, R. P. Chin and R. Crespo

17 Application of Bacillus subtilis PB6 in Turkey Field Trials

Andrew G Yersin, and Sally Moore

18 Comparative Genome Analysis of Gallibacterium anatis Causing Peritonitis in Laying Hens

Timothy J. Johnson, Claudia Fernandez, Adam Stell, Anders M. Bojesen, and Lisa K. Nolan

19 Detection of Campylobacter jejuni DNA in Broiler Cecal Dropping by Polymerase Chain Reaction

Sam P. Christenberry, Rick J. Meinersman, John R. Glisson, Kristi M. Moore, Charles M. Corsiglia, Thomas J. Dempsey

- 20 Gallibacterium anatis in broiler breeder flocks in the Republic of Panama: Clinical diagnosis and control measures Erick N. Calderon, Karina Thomas
- 21 Retrospective Study of Pasteurella multocida Isolates in Mississippi

Sue A. Hubbard

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Chicken Anemia Virus

- 23 A Retrospective Study of the Correlation between Chicken Anemia Virus and Avian Leukosis Virus Subgroup J Infection in Broiler and Broiler breeder Chickens Lanqing Li, Michael R. Luther, Alicia Wise, Frederic J. Hoerr
- 24 An 8-year longitudinal survey for the presence of antibodies to chicken infectious anemia virus in two specific-pathogen-free strains of chickens Karel A Schat, Ynte H Schukken
- 25 Serologic Survey of Chicken Infectious Anemia in Backyard Poultry in Argentina

Celina Buscaglia

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Stephan G. Thayer, Charles L. Hofacre, and Charles Broussard

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A. Khatiwara, S. L. Layton, G. Tellez, L. R. Berghman, B. M. Hargis and Y. M. Kwon.

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28 Mechanisms of intestinal Barrier Failure in subclinical Enteritis

Marco A. Quiroz, Julia D. Dibner, and Chris Knight

29 Nicarbazin Anticoccidial Dose Response

Greg F. Mathis

30 The transcriptome of the coccidian parasite Eimeria maxima the merozoite life stage

Ryan S. Schwarz, Raymond H. Fetterer, Katarzyna B. Miska

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Greg F. Mathis, and Jenny A. Fricke, and Charles L. Hofacre

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Catherine M. Logue, Yvonne M. Wannemuehler and Lisa K. Nolan

35 Predicting the virulence of Avian Pathogenic E. coli using serogroup, phylogenetic group and MLST

Kathy T. Mou, Kelly A. Tivendale, Yvonne Wannwmuehler and Lisa K. Nolan

36 Use of GFP- and RFP-Labeled APEC to Evaluate Adherence

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37 R Plasmids Found among Emergent APEC Strains

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Immunology, Immunity, and Vaccines

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39 The turkey genome sequence: implications to immunity, disease and overall health

Rami A. Dalloul, Oswald Crasta, Kent Reed, Audrey McElroy, Otto Folkerts, Ed Smith, Dave Burt, Rick Jensen, Roger Coulombe, Jerry Dodgson

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Evaluation of Paw Lesions through Broiler Grow-Out: A Field Study

Suzanne D. Young, Anel Atencio, and Ken N.Opengart

Keystone Foods Huntsville, Al

Paw lesions have recently become a area of interest in the broiler industry due to both economic and animal welfare implications. A field study was conducted in 3 complexes evaluating broiler paw lesions. Paw lesions, gait scores and litter conditions were monitored for 5 consecutive flocks on choosen farms to determine if litter management and/or farm conditions influenced scores in the field. Litter risk factors including pH, moisture, ammonia and nitrogen were measured and correlated with paw lesion scores through out grow-out. Flocks were followed into the processing plant to follow paw scores. The preliminary results will be presented.

7572

Spinal Abscesses Due to Enterococcus Cecorum in Broiler Chickens - An Emerging Disease?

Eric N. Gingerich, John H. Barnes, Robert L. Owen, Shelley C. Rankin University Of Pennsylvania

An emerging disease of broilers has been seen in Pennsylvania broiler flocks since early 2008 associated with lameness, high culling rate associated with paralysis from spinal cord pressure from abscesses found in the thoracic vertebrae. Enterococcus cecorum has been isolated from the lesion in pure culture. Other outbreaks are being described in other stateas as well. This report will describe the disease clinical signs, lesions, microbiologic findings, and efforts in controlling the disease.

7573

Case Report: A Control Strategy for Histomoniasis on a Replacement Broiler Breeder Farm

Mark A. Burleson

Sanderson Farms, Inc.

For six consecutive flocks, a replacement broiler breeder farm had experienced high mortality due to Histomoniasis and subsequent Staphylococcus infections. A control strategy was implemented and successful in eliminating the disease from this farm. Darkling beetle control, litter management, and a strict deworming program were among the critical aspects of the intervention strategy. Each detail of the program will discussed.

7571

Histomoniasis in Commercial Tom Turkeys Exacerbated by a Faulty Nipple Drinker System

David V. Rives

Prestage Farms, Inc. Po Box 438, Clinton, Nc 28329

An increase in mortality was noted in a house of 8,000 fourteen-week-old tom turkeys. A diagnosis of histomoniasis was made based on gross lesions and confirmed on histopathology. The disease eventually affected three out of four houses on the farm Prior to the increase in mortality, water consumption had begun decreasing in all four houses. Feed consumption had also decreased. Close examination of the nipple drinkers revealed erosion of a plastic plunger in each mechanism which resulted in restricted water flow. Reduced feed and water consumption rendered treatment less effective and negatively affected performance, even in the uninfected house.

7575

Epidemiology of an Unique Gangrenous Dermatitis Outbreak

Philip A Stayer, Jackson L McReynolds, Mark A Burleson Sanderson Farms, Inc.

Gangrenous dermatitis (GD) was unknown in one particular broiler grow-out region for almost ten years. Starting in November of 2007, GD was diagnosed first at processing and then in broiler flocks ranging from 13 to 48 days of age. Two sub-populations by flock age were determined in the outbreak: one group substantially younger at age of GD onset than the other. The younger sub-group appeared primarily in the progeny of one breeder flock. Upon further diagnostic tests, the breeder flock in question was shown to have a large portion of the flock sero-negative to Infectious Chicken Anemia Virus (CAV).

7576

Review of Virulent Newcastle Disease in Cormorants A Case Report

Sherrill Davison, Eric N. Gingerich and Lynn E. Stephens

University Of Pennsylvania, 382 West Street Road, Kennett Square, Pa 19348

Four cormorants suspected as having virulent Newcastle disease (vND) were presented to a local wild bird rehabilitation center. Postmortem examination, virus isolation and PCR were conducted at the state diagnostic laboratory. On virus isolation, one bird had evidence of a paramyxovirus. The PCR was matrix positive for paramyxovirus but negative for NDV. NVSL further characterized the isolate as a vNDV similar to an isolate found in domestic turkeys in 2002. There were several unique aspects of this case that will be reviewed. These include the epidemiology, postmortem examination, PCR testing at the state level, and regulatory issues at the federal and state levels.

7574

7577 Performance Problems in Free-Range Layers due to the IBV-QX Strain: Diagnosis and Prevention

Franz Sommer, Verena Seger

Cutler Associates International, Moorpark, Ca

In October of 2006, several cases of sudden onset of egg shell discoloration and massive drop in egg production, up to 50%, were reported from an Austrian area with high poultry density. Necropsy of hens showed either cystic oviducts with watery contents or partially atrophic oviducts with large cystic dilatations. Later on, IBV-QX virus was confirmed by PCR. In 2008, the same area was affected again. Due to changed flock management and an adapted vaccination regimen, the production drops were generally not as severe, and the recovery period was significantly shortened.A comparison of the course of the infection on several farms will be presented.

7578

Pathology and economic impact of a severe outbreak of infectious coryza in nonvaccinated broiler breeders.

Guillermo Zavala, Louise Dufour-Zavala, Martin Smeltzer, Mark W. Jackwood and Sunny Cheng

University Of Georgia

Abstract. An outbreak of infectious coryza in unprotected 47-week-old broiler breeders induced an egg production drop from 62% to 11% within 15 days. Daily mortality peaked at 0.77% by 8 days after the onset of clinical signs. Extreme depression, severe respiratory signs and head swelling were observed. Avibacterium paragallinarum (APG) was cultured from the infraorbital sinuses. Infectious bronchitis virus DEO72 was also detected. Clinical signs were reproduced in SPF chickens inoculated with APG and the microorganism was re-isolated from the experimentally inoculated chickens. Other diseases of concern were ruled out using a variety of diagnostic methods in the laboratory.

Case Report: Septicemic Erysipelas in Chukar Patridge

Scott D. Fitzgerald, DVM, PhD, DACVP, DACPV

Diagnostic Center for Population & Animal Health, Michigan State University

A game-bird producer experienced sudden mortality of 22-week-old chukar partridges (*Alectoris chukar*) in one pen of 250-300 birds. Mortality from the outbreak reached 20% of the pen-mates. Gross lesions were minimal. Histologically, prominent colonies of gram positive bacilli were present in the hearts, lungs, livers, kidneys and spleens. Necrotizing splenitis with RE cell hyperplasia was present; in other tissues bacterial colonization of the endothelium and histiocytes filled with bacteria were the principal histologic findings. Treatment of the pen with a 5 day course of penicillin in the water resulted in rapid clinical response and cessation of mortality.

7580

An unusual case of hepatitis in ring-necked pheasants (case report)

Eva Wallner-Pendleton, Patricia Dunn, Greg Ning, Huaguang Lu, Benjamin Lucio, Joan Smyth, Elizabeth Buckles

Animal Diagnostic Lab, Pennsylvania State University, State College, Pa 16802

An investigation into an unusal cause of mortality in 20 to 28 day ringneck pheasant chicks is described. In most cases, mortality between 5-50% would occur. On necropsy, affected birds would show hepatomegaly, sometimes with diffuse petechial hemorrhages. Duodenums were distended with bloody fluid. A focal ulcer was often found at the gizzard/duodenal junction. Microscopic examination of the livers showed striking necrosis, hemorrhages and bile duct hyperplasia. Results of feed trials, toxicology, disease transmission study and transmission electron microscopy will be discussed.

7581

Oropharnyngitis with Intralesional Dermatophilus-like Bacteria in a Breeder Chicken.

Frederic J. Hoerr, Haroldo Toro, Heather Busby, Sandra Ewald

Thompson-bishop-sparks State Diagnostic Laboratory, 890 Simms Rd., Auburn, Al 36831-2209

A mature pedigreed breeder hen developed open-mouth breathing. The bird was housed in a single-bird cage, raised above the floor in a poultry research building. The flock had a history of acariasis (Ornythonyssus sylviarum). At necropsy, the oropharyngeal mucosa was roughened and discolored yellow. Histologic examination revealed necrosis of superficial epithelial cells with infiltration and laminar separation by Gram-positive filamentous bacteria that were striated with cross tie zoospores. Scrapings of the oral mucosa revealed organisms morphologically consistent with Dermatophilus sp.; attempts to culture agent were not successful. Complete results of the investigation and the comparative pathology of oral lesions in chickens will be presented.

7582

Water management of Pseudomonas cepacia in brown-egg layer pullets

D. A. Anderson and D.Pennock

Georgia Poultry Laboratory Network

Inattention to water sanitation resulted in an acute onset of upper respiratory disease and mortality of a flock of organic brown-egg laying pullets. Diagnostic evaluation ruled out the most serious and more common poultry respiratory diseases. Continued workup revealed pure growths of Pseudomonas cepacia, and the source was the water system. An ORP based flow system was designed and implemented which removed the water line scale deposits and provided treated water even at the most distant watering station and has provided 100% control of the organism

7583

Cyanotic Broiler Breeder Males: Why Are Those Heads Purple?

Danny L. Magee, Jason Cater, Erica Baravik, Robert W. Wills, Sue Ann Hubbard, Lanny W. Pace

Poultry Research & Diagnostic Laboratory, PO Box 97813, Pearl, MS 39288

In commercial broiler breeder operations it is not uncommon to see breeder males with dark combs and wattles. Service technicians often refer to this condition as purple heads. The degree of this discoloration can vary from flock to flock and within a flock. Some have observed a male's coloration change from purple to red during the day. There are multiple suggested causes for the existence of this condition. This presentation will attempt to explain the cause of this clinical presentation.

Effects of Specific Amino Acid Changes on the Antigenicity of Hemagglutinin Molecules of Avian Influenza Isolates from Mexico

J. Pfeiffer, C. W. Lee, S. Jadhao, and D. L. Suarez

Southeast Poultry Research Laboratory, 934 College Station Rd., Athens, Ga 30605.

Amino acid (aa) changes between the hemagglutinin (HA) proteins of a vaccine avian influenza virus and more recent field isolates were detected following prolonged vaccination of Mexican poultry. Using site-directed mutagenesis and reverse genetics (rg), viruses containing identical backbones but different HAs were generated. The cross-hemagglutination inhibition (HI) and virus neutralization tests were used to compare antigenic differences. A vaccine-challenge study on chickens was performed to compare differences in viral shedding between groups, as determined through real-time RT-PCR on oropharyngeal swab sample RNA. The goal was to gain knowledge for selecting vaccine seed strains.

Recombinant RCA-free Adenovirus-Vectored Vaccine for Immunization of Chickens and Pigs against Avian Influenza

Haroldo Toro¹, De-chu C. Tang², Bettina Schemera³, Soren Rodning⁴,

Joseph C. Newton¹

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Protective immunity to avian influenza (AI) virus can be elicited in chickens by *in ovo* or intramuscular (IM) vaccination with RCA-free human adenovirus vector (Ad) encoding AI virus H5 (AdTW68.H5) or H7 (AdChNY94.H7). We evaluated immune responses of 1-day-old chickens vaccinated via spray with increasing doses of these recombinant vaccines. Chickens showed serum and mucosal antibody responses 20 days post vaccination. We evaluated simultaneous *in ovo* vaccination with AdTW68.H5 and AdChNY94.H7. Vaccinated chickens developed robust antibody levels to both H5 and H7 AI strains. We also explored the use of RCA-free-Ad in pigs. All pigs vaccinated IM with a single dose of AdTW68.H5 [10⁹vp] developed strong antibody responses [~6

7584

log₂] 3-w post-vaccination. Few pigs vaccinated intranasally (2/6) developed similar antibody levels. Supported by USDA AI CAP Award# 2007-35203-18070

7586

Pathogenicity of reverse genetics based reassortant H5N1 avian influenza viruses with truncated NS1 gene in chickens

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

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

The NS1 protein of influenza A virus plays an important role in blocking the induction of type I interferon and other regulatory functions in infected cells. However, differences in length of the NS1 protein has been observed in highly pathogenic H5N1, H5N2, and H7N1 subtype avian influenza viruses. To evaluate the influence of truncated NS1 gene on H5N1 virus pathogenicity in chickens, using reverse genetics, we generated polybasic H5 HA containing A/turkey/Ireland/83 and A/chicken/Indonesia/7/03 viruses with full length and truncated NS1 genes. The viruses were compared by intravenous pathotyping and mucosal challenge in 4 week old chickens to determine how NS1 gene truncation affects the virulence.

DIFFERENCES BETWEEN PEKIN AND MUSCOVY DUCKS IN RESPONSE TO VACCINATION AGAINST H5N1 HPAI

Mary J. Pantin-Jackwood, David E. Swayne and David L. Suarez

Southeast Poultry Research Laboratory, USDA-ARS

Ducks and other wild aquatic birds are the natural reservoir of avian influenza viruses which usually are nonpathogenic for these birds. However, since late 2002, H5N1 highly pathogenic avian influenza (HPAI) outbreaks have resulted in mortality among ducks and other wild birds. Vaccination has been used in Vietnam since 2005 to control H5N1 HPAI in domestic ducks with mixed results. One of the observations from the field is that Muscovy ducks (*Cairina moschata*) respond differently to vaccination than other common domestic ducks (*Anas sp.*). The objective of this study was to compare the response to HPAI vaccination between Muscovy and Pekin ducks (*Anas platyrhynchos*). The Re-1 Chinese inactivated recombinant vaccine was used to vaccinate the ducks at 2 and 14 days, at 14 days only, and at 7 and 21 days of age. Ducks were challenged at 30 days of age with a lethal dose of A/duck/Vietnam/88/07. Vaccination provided good protection against mortality; with the best protection obtained when vaccinating ducks at 7 and 21 days of age. However, Muscovy ducks had lower pre-challenge antibody titers, presented higher morbidity and mortality, and shedded virus for a longer period of time than the Pekin ducks. In conclusion, clear differences in response to vaccination were

found between these two different domestic duck species. This information should be taken into account when using vaccination for control of HPAI in ducks.

7589 Analysis of H7 Avian Influenza Viruses by Antigenic Cartography and Correlation to Protection by Vaccination

Erica Spackman, Ron Fouchier, David Swayne, Ahmed Abbas and Luciana Sarmento

Southeast Poultry Research Lab, USDA, Ars, 934 College Station Rd., Athens, Ga 30605

The H7 hemagglutinin subtype one of the most common subtypes of avian influenza virus (AIV) in poultry world wide and since it has the potential to become highly pathogenic, it is among the priority subtypes for vaccination. Selection of the optimal vaccine seed strains may now be aided by antigenic cartography, a relatively new method that can be used to analyze the antigenic relationship among influenza viruses based on hemagglutination inhibition data. Here we evaluate the antigenic relationship among H7 AIVs from poultry and attempt to correlate antigenic relationships with protection by vaccination to challenge with highly pathogenic AIV.

7590

Passage of low pathogenic avian influenza virus isolates from water fowl in chickens results in adaptive mutations in the glycoprotein genes

Daniel Dlugolenski, Ralph A. Tripp, Mark. S. Tompkins, Egbert Mundt University Of Georgia, Poultry Diagnostic Research Center. Department of Infectious

Disease

To gain a better understanding about the mechanisms of IpAIV transmission, adaptation, and disease pathogenesis which might be linked to changes in the genetic composition of HA and NA, six serial passages in chickens were performed. The results show that during passage in chicken with an isolate from a chicken and an isolate from a wild bird several mutations in the HA and NA occurred in both isolates. The observed mutations occurred during early passages of the isolates and remained present throughout the study. This indicates that mutations occur very early during the adaptation of wild bird isolates to chicken.

Development of DIVA Vaccines for the Control of Triple Reassortant H3N2 Influenza in Turkeys

Leyi Wang, Hadi M. Yassine, Smitha Pillai, Yehia M. Saif, Chang-Won Lee

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

To better control triple reassortant (TR) H3N2 influenza in turkeys, a DIVA vaccine with an accompanying serological test can help to screen and stamp out the influenza infected flocks or individual birds. In this study, two different strategies, heterologous NA or truncated NS1 protein-based strategies, were used to generate DIVA vaccines by traditional reassortant method. Efficacy of DIVA vaccines were assessed in young turkeys and older breeder turkeys, followed by serological test for all serum collected from each group. Our results showed that DIVA vaccines significantly reduced challenge virus shedding in oviduct of laying turkeys as well as in trachea and cloaca of young and old turkeys compared to non vaccinated control group. Once serological tests are done for NA or NS1 protein, every aspect of both DIVA vaccines will be compared side by side to select the best DIVA vaccine to control TR H3N2 infection in turkeys.

7592

Summary of type A influenza virus investigations for waterfowl in Ohio and on Delmarva: 2001-2008

R.D. Slemons, D.D. Swayne, C. Driscoll, L. Hindman, V. Stotts, L. Alexander, M. Shieldcastle, D. Sherman, D. Senne Ohio State University

Beginning in 2001, concurrent influenza A virus investigations in waterfowl were conducted at selected sites in Ohio and Maryland. As of 2007, a total of 170 and 391 low pathogenic waterfowl-origin avian influenza virus (WFOAIV) isolates were recovered in Ohio and Maryland, respectively. These WFOAIV isolates represent approximately 50 HA-NA combinations, were recovered yearly and possessed the H5, H7, N1, N2, and/or N3 surface glycoproteins 5, 4, 5, 6 and 6 of the 7 years, respectively. No WFOAIV outbreaks occurred in commercial poultry raised in the study areas.

7593 Survey of backyard/hobby/small production flocks within a high density poultry area

Benjamin C. Johnson¹, Louise Dufour-Zavala¹, and Martin A. Smeltzer² ¹Georgia Poultry Laboratory Network, PO Box 20, Oakwood, GA 30566 ²USDA/APHIS/VS, Dept of Population Health, UGA, Athens, GA, 30602

This study will categorize and produce a statistically accurate census of non-commercial poultry in the city of Gainesville, GA. Eight (1 kilometer radius) base units will be randomly selected within Gainesville. The study will utilize on ground observation surveys to collect information on each base unit, census data, and GIS technologies to project the results to larger areas. This will involve development of an observation survey form, data collection and analysis. The data will be compiled to make statistically valid projections of populations in similar geographic or population density areas.

7594

Astrovirus associated with Turkey Viral Hepatitis

Shivaprasad H. L. and Mary J. Pantin-Jackwood Cahfs Fresno Branch

Turkey Viral Hepatitis (TVH) is a highly contagious viral disease of poults characterized by increased mortality associated with hepatitis and occasionally pancreatitis. Even though the disease has been known since the 1950's, the exact viral cause of the disease is still not known. Recently we investigated three outbreaks of TVH in 25 to 30 day-old poults. History included increased mortality in the flock and many poults had hepatitis and a few birds had pancreatitis. RT-PCR performed on biles and livers revealed eight of ten and two of eight positive for astrovirus respectively. Preliminary sequencing results suggest that this could be a unique astrovirus.

Inclusion boby hepatitis as a vertically transmittedprimary disease in broiler chickens

Susantha Gomis, Samantha Ekanayake, Davor Ojkic, Suresh Tikoo, Bob Goodhope, Philip Willson

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In the last few years, inclusion body hepatitis (IBH) has emerged as an economically important disease in broilers in Canada. In the past, concomitant infections with infectious bursal disease virus (IBDV) and/or chicken anemia virus (CAV) have been known to suppress the immune system of broilers and make them more susceptible to a secondary disease such as IBH. Recently, we reported that virulent adenoviruses are able to cause IBH as a primary disease in broilers without involvement of IBDV and CAV. The objective of this study was to experimentally reproduce IBH in commercial broiler chickens and to demonstrate vertical transmission of fowl adenoviruses (FAdV) in broiler breeders to their progeny. Broiler chickens were intramuscularly inoculated with 1x104- 1x107 pfu of FAdV-8a, FAdV-8a/8b, FAdV-7 or FAdV-11 at day-fourteen of age. After four days following FAdV inoculation, 5% - 15% mortality was observed with hemorrhagic necrotizing hepatitis. To demonstrate vertical transmission of the FAdV, 35-week-old broiler breeders were inoculated with 1x105 pfu of FAdV, eggs were collected and hatched seven days post-injection of FAdV. The broiler progeny derived from broiler breeders inoculated with FAdV-8a/8b had 30% mortality at day-7 posthatch. The results of this study supported the hypothesis that IBH in broilers in Canada is a vertically transmitted primary disease with no apparent immunosuppressive involvement. To the best of knowledge, this is the first report to demonstrate vertical transmission of FAdV by virus isolation in commercial broiler chickens following experimental inoculation of FAdV in broiler breeder parents.

7596

Development of a recombinant vaccine against Runting and stunting syndrome in chicken

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Runting and stunting syndrome (RSS) has been recognized since the late 1970s in broiler chicken and several viruses have been associated with this syndrome. Most of these viruses do not propagate in vitro and in embryonated eggs. To overcome this problem a recombinant baculovirus was generated which encoded for a virus capsid protein. The subsequently purified protein was used as an oil-emulsion vaccine in commercial broiler breeder along with a group of non-vaccinated broiler breeder as control. In challenge experiments the offspring of the vaccinated breeders was protected whereas the off spring of the non vaccinated controls was not protected.

7597 Development of histopathological changes associated with runting and stunting syndrome

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4875

Runting-Stunting Syndrome (RSS) is an old disease affecting broilers leading to poor performances. New wave of this syndrome has emerged in the USA. Reovirus was thought to be the cause of RSS, but Adenovirus, Enterovirus, Rotavirus, Parvovirus and others are involved. The aim of this research is studying other organs besides the intestine, including the primary immune organs, secondary immune organs, and the pituitary glands, using histopathology, and Insituhyperdization targeting Astrovirus in a trial to reach better understanding of the syndrome.

7598

Enteric disease in broiler chickens following experimental infection with chicken parvovirus

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Day-old broiler chickens were inoculated orally with the chicken parvovirus strain, ChPV-P1. In four independent experiments, characteristic clinical signs of enteric disease including watery, mustard color diarrhea and growth retardation were observed following infection. The virus was shedding in feces between 4 and 21 days post infection (DPI) and viremia was detectable from 7 DPI to 21/28 DPI. Anti-parvovirus IgM antibodies appeared as early as 14 DPI and nearly all infected birds seroconverted by 28 DPI. Our data indicate that maternal antibodies have a major role in the pathogenesis of chicken parvovirus in the host.

Development of an ELISA assay to detect chicken parvovirus antibodies

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We describe the development of an ELISA assay to detect parvovirus antibodies in poultry. The major structural VP2 protein of the chicken parvovirus (ChPV) was expressed in a baculovirus expression system. Cell extracts following infection with the baculovirus ChPV-VP2 recombinant were assayed by SDS-PAGE in which a unique band of ~65 kDa was observed in Coomassie stained gels. In Western Blot analysis, the baculovirus ChPV-VP2 protein was specifically labeled using antisera from parvovirus infected chickens. The baculovirus ChPV-VP2 antigen was then used to develop an antigen-down ELISA assay which successfully detected antibodies in chickens following parvovirus infection.

7600

Assessment of Reovirus Vaccination on Blackleg Trims in Commercial Broilers

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"Blackleg" is a common term used in the field to decribe acute tendon damage in broilers. It can result in lameness in the field as well as exessive trim in processing plants, hence minimizing this problem can improve profitability of poultry integrators. Although there can be several factors impacting this condition, the effects of in-ovo reovirus vaccination were assessed in this field study. Broilers which were vaccinated against reovirus were compared with unvaccinated broilers via gross incidence of blackleg lesions on the processing line and trim data. A definite difference was noted and will be addressed in the presentation.

Genetic characterization of avianpox viruses using DNA isolated from formalin fixed tissue sections

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In the absence of virus isolation, diagnosis of avianpox virus infection often rests with histopathological examination of the lesion without genetic characterization of the strain. In this study, we isolated viral DNA from formalin fixed tissue sections. by two different methods. In the first method, the sections were deparaffinized and the DNA was isolated by using the Quiagen Kit. In the second method Wax free DNA kit was used. The isolated DNA was used to amplify three different size fragments of A-type body protein gene. For comparison with the ATI gene sequences, the inclusion amplified products were sequenced. This method will allow rapid genetic characterization of avianpox viruses in the absence of virus isolation.

7602

DETERMINATION OF THE PREVALENCE OF PIGEON CIRCOVIRUS IN **CALIFORNIA SQUAB**

Simone T. Stoute, DVM, Carol J. Cardona, Cathryn R. Bauer, Daphne A. Cooper, and Bruce R. Charlton

University Of California, Davis, California Animal Health And Food Safety Laboratory, Turlock Branch

Pigeon circovirus (PiCV) is a member of the *Circoviridae* family and there is substantial evidence that infection results in immunosuppression. Objectives of this investigation are to determine the prevalence of PiCV in commercial market age N. California squab by PCR, and determine the correlation between PiCV positive birds and birds exhibiting bursal atrophy and intracytoplasmic inclusions within bursas. Electron microscopy on 4 selected bursas was performed to correlate histological detection of botryoid bodies and PCR PiCV detection with the presence of viral particles. DNA sequencing of selected amplified product was done to ensure product was compatible with circovirus and assess the degree of homogeneity among sequenced isolates. All 378 bursas examined, were positive for PiCV by PCR. Interesting, light microscopy detection of PiCV intracytoplasmic botryoid inclusions resulted in detection of only 6.6% (25/378) of PiCV positive bursas. The accuracy of detection of PiCV inclusions by light microscopy also correlated with the detection of inclusions by electron microscopy. 85% (21/25) of the bursas with inclusion bodies also exhibited some degree of bursal lymphoid depletion. Sequencing of the amplified DNA product from 6 samples and the positive control used in the investigation was homologous with other PiCV isolates in the National Center for Biotechnology Information (NCBI) DNA bank.

Testing of a New Disinfectant Process for Poultry Viruses

Lauren Appleby Gay and Egbert Mundt

University Of Georgia

The application of a new chemical compound as a disinfectant against enveloped as well as non-enveloped viruses (AIV, IBDV) was studied. The used biocide forms a gas in this process, thus it has a strong penetrating capability in sealed poultry houses. The results show that both viruses in contaminated chicken litter were inactivated in less than one hour. These results indicate that routine treatment of facilities and poultry litter would greatly control the risk of transmission of virus between flocks which are raised on used litter. Use of this disinfectant after a disease outbreak to disinfect litter seems feasible.

7604

On the effectivity of a vaccination schedule against aMPV in Brazilian commercial layers as measured by RT-PCR, DNA sequencing, ELISA and clinical sig

Laura YB Villarreal, Delair A Bolis and Paulo E Brando

Intervet Schering Plough Animal Health

The aim of this investigation was to evaluate the effectivity of a vaccine schedule against a divergent aMPV lineage. Three flocks of Brazilian layers received live and killed aMPV vaccines, while two flocks received no aMPV vaccination. Vaccinated flocks were negative for aMPV-PCR, with ELISA titres lower than control flocks. Vaccinated flocks showed no signs of aMPV infection. Phylogenetic tree showed that the aMPV lineages involved grouped in an exclusive cluster, apart from other aMPV strains detected in Brazil. These results allow one to conclude that this vaccination schedule was effective in preventing the disease despite the divergent aMPV involved.

7605

Onset of Immunity in Birds with Newcastle Disease Virus (NDV) Maternal Antibodies(Mabs) Vaccinated with the Recombinant HVF/F(NDV), INNOVAX-ND-SB

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Intervet/schering Plough Animal Health P.o.box 318 Millsboro De 19966-0318

Broilers with low and high HI/Elisa NDV Mabs were vaccinated in ovo and/or subcutaneously at one day of age with a recombinant HVT/F(NDV) vaccine. From around one week post-vaccination onwards, birds, including SPF controls, were challenged by mucosal route with velogenic NDVs. Results showed that the NDV Mabs did not interfere with efficacy of the recombinant vaccine.

Transmission of virulent Newcastle disease virus (NDV) between unvaccinated, sub-optimally vaccinated, and well-vaccinated SPF chickens

Patti J. Miller, Claudio L. Afonso

Southeast Poultry Research Laboratory, 934 College Station Road, Athens, Ga 30605

The purpose of this study was to determine the transmissibility of virulent NDV in vaccinated chickens. Chickens were vaccinated with live LaSota and challenged 21 days later with CA02. Two days after challenge, the vaccinated and infected chickens were moved into clean isolators with naÃ⁻ ve or vaccinated contact birds. None of these naÃ⁻ ve contact birds underwent seroconversion or shed virus, even though the infected birds were shedding virus. However, if vaccinated birds where challenged with CA02 prior to 21 days (i.e. sub-optimally 3,5, and 10 days post vaccination), they were able to transmit virus to naÃ⁻ ve and vaccinated contact birds. In experimental conditions, vaccination with live LaSota prevents transmission if a sufficient time is given for an optimal immune response.

7607

E. coli survey in a broiler complex using serotyping and pathotyping techniques

Kalen C. Cookson, Lisa K. Nolan and Chobi DebRoy Fort Dodge Animal Health

Progeny of 11 broiler breeder flocks were submitted for routine bacteriology (yolk sacs) shortly after hatch. A total of 7 broiler farms receiving chicks from these broiler flocks were then tested for E. coli in both litter samples (3-4 weeks) and cellulitis lesions at processing. Finally, attempts were made to culture E. coli from the original 11 breeder flocks. All E. coli positive samples were submitted for both O-serotyping and pathotyping (PCR assay of 9 virulence genes). The results of this survey will be discussed.

E. coli serogroup O18: Emerging isolates that are highly pathogenic in poultry

Kelly A. Tivendale, Yvonne Wannemuehler, Kathy T. Mou, Catherine M. Logue, Ganwu Li, Subhashinie Kariyawasam, Ashraf Hussein, and Lisa K. Nolan Department Of Veterinary Microbiology And Preventive Medicine, Vmri #2, College Of Veterinary Medici

Escherichia coli that can survive and establish an infection outside of the intestine are known as Extraintestinal Pathogenic E. coli (ExPEC). ExPEC are the causative agents of colibacillosis in poultry and urinary tract infections, neonatal meningitis, and sepsis in human hosts. Currently, we are characterizing a group of ExPEC of both human and avian origin that are members of the O18 serogroup, carry large plasmids and are highly pathogenic in poultry. Despite their isolation from different host species, these isolates could not be differentiated by MLST, PFGE, or genotyping, and they were lethal to rat pups, chick embryos, and chickens. Such results suggest the emergence of a group of highly pathogenic E. coli.

7609

Avian Pathogenic E coli (APEC) reduction in Commercial Poultry

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Avian pathogenic E cloi has been implicated in respiratory diseases in broilers and turkeys. As a primary or secondary pathogen APEC results in diseased flocks with poor livability and performance causing the poultry industry severe losses. In the recent years efforts have been made to study the involvement of APEC in diseased poultry and tools to reduce and control its adverse effects. Distribution of five pathogenic genes (iss, iucC,tsh, cvaC and irp2) in US commercial poultry will be reported. Live production performance improvemets resulting from APEC control will be presented and discussed.

A Novel Autotransporter Contributing to Adherence Is Necessary for the Full Virulence of Avian Pathogenic E. coli

Ganwu Li, Subhashinie Kariyawasam, Yvonne Wannemuehler, Kelly Tivendale and Lisa K. Nolan

Department Of Veterinary Microbiology And Preventive Medicine, College Of Veterinary Medicine, 1802

An ORF (ORF APEC O1_O1ColBM96) encoding a novel autotransporter (AT) was located within the pathogenicity island of avian pathogenic Escherichia coli (APEC) O1's virulence plasmid, pAPEC-O1-ColBM. This 3.5-kb gene (aesA) is predicted to encode a 123.7 kDa protein with a 25-amino-acid-long signal peptide, an 857-amino acid-long passenger domain and a 284 amino acid-long Â² domain. Molecular analysis of AesA revealed that it is translocated to the cell surface, is not expressed in vitro, and it elicits antibody production in infected chickens. Gene prevalence analysis suggested that aesA gene is strongly associated with E. coli from avian sources but not with E. coli isolated from human hosts. Also, AesA was shown to enhance adhesion of APEC to chicken fibroblast cells and to be necessary for the full virulence of APEC O1. Therefore, aesA is a novel AT adhesin from E. coli that mediates adherence to chicken fibroblast cells and contributes to APEC virulence.

7611

Egg Yolk Inhibits the Acute Phase Inflammatory Response to E. coli in Egg Yolk Peritonitis

Ingrid Cornax Edwards, Kirk C. Klasing

Uc Davis- School Of Veterinary Medicine, Graduate Group In Immunology

Egg volk peritonitis (EYP) has been found to cause about 50% of all loss in laving flocks. The most commonly isolated pathogen from birds that have died of EYP is E. coli, but E. coli alone is not sufficient to cause disease. Egg yolk appears to play a critical role in the disease pathogenesis of EYP. The goals of this research are to confirm that both egg yolk and E. coli are necessary to cause EYP and to determine how egg yolk increases the pathogenicity of E. coli. Broiler chicks were injected IP with egg yolk and/or E. coli. Only birds injected with both egg yolk and E. coli showed signs consistent with egg yolk peritonitis. E. coli was incubated in vitro with egg yolk and/or macrophages and then streaked on agar plates. Egg yolk significantly decreased E. coli colony number (p=0.0397) and did not affect macrophage antimicrobial function (p=0.3233). In a second injection experiment, egg yolk significantly reduced the E. coliinduced inflammatory reaction, by decreasing blood heterophil: lymphocyte ratios (p=0.0453) and inhibiting IL-1 and IL-6 expression in the spleen (p=0.0361 and p=0.0105, respectively). Egg yolk also tended to increase plasma iron (p=0.0611) and significantly decreased plasma copper (p=0.0014) making conditions more conducive for bacterial growth. Egg yolk did not prevent the induction of acute phase proteins despite lowering II-1 and IL-6 expression.

Real-time PCR in Avian Mycoplasma Diagnostics

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Four avian mycoplasmas are commonly recognized as poultry pathogens: *Mycoplasma* gallisepticum (MG), Mycoplasma synoviae (MS), Mycoplasma meleagridis (MM) and Mycoplasma iowae (MI). The avian mycoplasmas are associated with respiratory disease, synovitis and arthritis, poor performance, skeletal deformities or embryo mortality. There are three main approaches to the diagnosis of avian mycoplasmosis: isolation and identification, detection of antibodies, and molecular detection of the organism's nucleic acid by polymerase chain reaction (PCR). In recent years real-time PCR technology has revolutionized the way clinical microbiology laboratories diagnose infectious diseases but so far only a limited number of diagnostic real-time PCRs were proposed for avian mycoplasma diagnostics. We developed a complete set of reliable diagnostic real-time TagMan PCR assays for the 4 pathogenic avian mycoplasmas. The selected genomic targets of the developed assays were species specific and intraspecifically conserved and included the 16S-23S intergenic spacer region of MS and MM, the upstream region to the 16S rDNA of MI and highly conserved foci of the mgc2 gene of MG. The 4 assays were demonstrated highly specific and sensitive to their target avian mycoplasma, with detection limits of 1 copy per reaction mix for the MG assay and 10 copies per reaction mix for the MS, MM and MI assays. In addition the developed assays harbor the potential to be conducted in a multiplex set up. We believe that the incorporation of the developed assays in avian mycoplasma diagnostics will contribute to the accuracy, efficiency and feasibility of these pathogens diagnosis.

7613

The Displacement of Mycoplasma gallisepticum Strains by Live Vaccines

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Mycoplasma gallisepticum (MG) vaccines may be a useful part of eradicating endemic wild-type MG strains from poultry operations. The potential of live vaccines to displace virulent strains was evaluated in this study. We compared the commercial MG vaccines ts-11, 6/85, and F strain; as well as a new vaccine strain - K strain. Vaccinated layer-type chickens were commingled with seeder chickens infected with R strain. Quantitative strain differentiating real-time PCR was conducted to investigate the ability of the vaccines to prevent subsequent infection with R strain.

Serology results for TS-11 vaccinated and non vaccinated breeder flocks in Georgia

Len Chappell and Dorene Seabolt

Georgia Poultry Laboratory Network

During 2007 and 2008, live MG vaccine (ts-11) was used in a large number of meat type breeder flocks in Georgia to control the spread and negative performance impact of field MG infection. ELISA, serum plate agglutination and hemagglutination inhibition test results from vaccinated and non vaccinated breeder flocks at different ages were compared and analyzed and will be presented.

7615

Comparative Testing of Samples from a Broiler Breeder Flock for *Mycoplasma Synovia*e

David H. Ley, Ziv Raviv and Naola Ferguson-Noel

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Serum and tracheal swabs from an out-of-state broiler breeder flock were submitted to NCSU for confirmation of positive *Mycoplasma synoviae* (MS) results obtained locally. Initial MS PCR results using two primers (MSL and *vlh*A) gave very different results: 50% and 0% positive, respectively. Positive MS serum plate agglutination (55%) results, confirmed by hemagglutination inhibition provided sufficient evidence for a diagnosis of MS. However, the disparity of initial MS PCR results stimulated us to examine more closely the outcomes of MS PCR (conventional and real time) and culture at two laboratories. While there was general agreement between laboratories and among MS tests, we also found differences that reinforce the importance of understanding test performance parameters and confirmatory testing.

Role of Mycoplasma anatis and Interactions with other Infectious Agents in the Tenosynovitis and Arthritis syndromes in commercial ducks.

Ruiz, Jaime, Ferguson-Noel, Naola, Lucio-Martinez Benjamin, Laibinis Victoria Cornell University Duck Laboratory

Leg health is one of the most prevalent causes of economic losses in both commercial and breeder ducks. As birds gain weight, tenosynovitis, arthritis and other inflammatory processes are frequent conditions associated with stilted gait and poor flock performance. A Mycoplasma anatis strain has been isolated from affected legs of commercial ducks experiencing severe tenosynovitis in Long Island, NY. In vivo studies, interactions with other potential infectious agents and pathogenicity studies of the role of Mycoplasma anatis in duck leg health problems will be presented.

7617

Detection of Salmonella enterica serovar Enteritidis (SE) Antibodies in Serum Using A Polystyrene Bead/SE Flagella Agglutination Assay

Peter S. Holt, Kyle D. Swayne, Lara E. Vaughn, Richard K. Gast

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Serologic screening of flocks can be an important method to detect Salmonella enteritidis (SE) infections but can be labor intensive or lack specificity. Our goal was to develop a rapid agglutination assay using SE flagella adsorbed to polystyrene beads as a simple, relatively specific test to detect serum SE antibodies. Birds were vaccinated with either a commercial SE bacterin or a bacterin composed of SE cells heated in a boiling water bath which destroyed the flagella but retained LPS O antigens. The serum was tested by mixing 10 ul serum with 20 ul beads. Sera from the birds receiving the commercial bacterin strongly agglutinated the beads while no bead agglutination was observed from the sera of birds receiving the boiled cell bacterin. This latter sera did exhibit strong agglutination of stained whole SE cells, indicating a potent response to the SE LPS antigens. These results indicate that SE flagella coupled to polystyrene can be used as a simple and specific means of serologically screening flocks for SE infection.

Use of CFSE dye and fluorescent imaging to visualize lymphoid tissues in chickens challenged with Salmonella Enteritidis (SE)

Lara E. Vaughn, Peter S. Holt, Richard K. Gast

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Carboxyfluorescein diacetate succinimidyl ester (CFSE) vital dye has been used in leukocyte studies involving mice, rats, sheep, heifers, nonhuman primates, teleost fish and avian embryos. Mice and sheep appear to be the only animals that have received intravenous (IV) CFSE administration, and the purpose was for collection of CFSE positive leukocytes from peripheral blood and subsequent cell transfer for leukocyte migration studies. We have found no literature for IV use of CFSE in mature egg-layer chickens to identify lymphoid tissues in situ. Presently, we explore IV administration of CFSE and the application of a fluorescent imaging system for in situ visualization of lymphoid tissue in egg-layer hens challenged with Salmonella enterica serovar CFSE positive fluorescence was observed within the spleen, Enteritidis (SE). proventriculus, Peyer's patches and cecal tonsils. This novel use of intravenous CFSE in chickens may hold promise for researchers trying to identify often grossly obscure mucosal lymphoid tissue sites for histopathology evaluation or cell harvesting. There appears to be potential for CFSE positive lymphoid tissues to be identified in situ, viable cells collected, and then perhaps CFSE lymphocyte suspensions used for flow cytometric cellular phenotyping, cell culture establishment for cytokine expression research, or cell migration/trafficking and cell division studies. CFSE and fluorescent imaging could be useful tools to advance our knowledge about the avian immune system and the immune response against disease agents such as SE.

7619 European Union (EU) Salmonella Enteritidis Control with the Use of an Inactivated Salmonella Vaccine.

Rik Koopman, Rick v Oort, Ruud Hein

Rik Koopman Dvm, Global Technical Manager Poultry, Intervet/schering-plough Animal Health

Food born Salmonella infection in humans can be related to salmonella infections in poultry. The poultry industry in the EU is obliged by EU law (directives) to actively monitor, report and control the salmonella infections in poultry. Since the late eighties the poultry industry started to implement measurements to reduce salmonella infections in poultry. First mainly by applying increased biosecurity (closed houses, clothes and boots per farm or house) and management tools such single age, rodent control, water& feed control. Some improvement was made in the reduction of salmonella infection in poultry but the addition of vaccination to the salmonella control program brought real reduction. The use of an inactivated vaccine (Nobilis Salenvac) in the control program showed significant reduction of the Salmonella infections in poultry and human cases of Salmonella enteritidis.

7620

A Long-Term Survey of Salmonella sp. and Campylobacter sp. in a Meat-type Poultry Processing Plant

Spangler Klopp, Daniel Austria Bautista, Brenda R. Sample, Kathy J. Philips, and Billie Jean Wright

Klopp Consulting Services

There is a renewed government initiative to reevaluate the USDA Salmonella Performance Standard and potentially establish one for other food safety organisms like Campylobacter sp. A qualitative and quantitative survey of Salmonella sp., Campylobacter sp. was conducted in 2 broiler processing plants for more than 52 weeks. We will present data on Salmonella sp. and Campylobacter sp. prevalence, serotype prevalence, and enumeration in selected sites in the plant. These new collated data will be discussed with live-production management strategies in mind, and will be compared to similar data in published USDA FSIS and scientific literature.

7621 Turkey Slaughter Establishments Operating Under the Salmonella Initiative Program

Becky J. Tilley, Alice L. Johnson

Goldsboro Milling Co./Butterball, LLC

The Salmonella Initiative Program (SIP) is designed to produce voluntary improvements in Salmonella control in broiler and turkey slaughter establishments while allowing a waiver to existing regulations, permitting an increase in the volume of birds slaughtered or allowing the chilling of carcasses in a different manner than is allowed by existing time/temperature requirements. To participate, the establishment must be in Category 1 (percent positive Salmonella samples at 50% or less of the performance standard or guideline in the two most recent completed sample sets). This presentation will provide data from turkey slaughter establishments operating under SIP.

7622

The Utility of a Commercial Real-Time PCR Assay for Screening Salmonella sp. in Poultry Carcass Rinses and Environmental Samples

Daniel Austria Bautista, Spangler Klopp, Brenda R. Sample, Kathy J. Philips, and Billie Jean Wright

University Of Delaware-lasher Laboratory

The performance of a commercially-available the Warnex TM real-time PCR assay in screening for Salmonella in processed broiler carcass rinses, litter drag swabs, and some hatchery samples will be presented. The data is collated from 3 years' worth of Salmonella testing using established culture methods. The sensitivity and specificity of the RT-PCR assay will be discussed.

A History of Coccidiosis to 1950

R.B. Williams

Coxitec Consulting, UK

The early recorded history of coccidiosis occupies about 275 years up to 1950. It will be considered under the following broad topics, which will be addressed and expanded.

- 1) Introduction: known species of homoxenous coccidia of birds, particularly domesticated and game species.
- 2) Archaeological evidence for the domestication and world-wide transport of chickens, turkeys and pheasants during last 4,000 years, enabling the contemporaneous spread of their associated parasites.
- 3) The 17th and 18th centuries. The earliest record of a coccidium was an oocyst in the bile of a rabbit in 1674, by the Dutchman van Leeuwenhoek, a lens grinder and microscope maker. Gregarines were later discovered in 1787 by the Italian Cavolini in crab stomachs.
- 4) The 19th century. In 1839, the Englishman Hake observed oocysts, mistaking them for pus globules associated with liver carcinoma. The rapid development of taxonomy included the identification of new species in many hosts, accompanied by an increased understanding of structure and life cycles, and clarification of nomenclature (*Psorospermium, Coccidium, Eimeria*) particularly in Germany, France and Britain.
- 5) The 20th century. In the first 20 years, the significance of coccidiosis as a disease of domesticated animals began to be realized, and American and British workers became more involved. In Britain, the first eimerian life cycle in a bird was described; the higher taxonomy was being worked out; and coccidiosis was becoming a problem on farms in Britain and South Africa. The supposed connection between coccidiosis and cancer during the 19th century may have stimulated Tyzzer (a cancer specialist at Harvard) to begin his work on coccidiosis. Having discovered *Cryptosporidium* in mice, he became the world leader in avian *Eimeria* research, while others initiated research in the west coast states.
- 6) The 1920s and 1930s saw significant developments in the USA of laboratory methods, and an increased understanding of epidemiology and its impact on farm animals. The confusion between coccidiosis and "white diarrhoea" and "blackhead" was also unravelled. Early attempts at chemotherapy culminated in success with sulphonamides in 1939.
- 7) Improved epidemiological knowledge contributed to disease control by hygienic measures, and the application of immunization was followed up in the 1940s, with attempts to combine chemotherapy and immunization. This era ended in 1949 with the first specialist coccidiosis conference in New York, a series still continuing today.

Correlation between use of various coccidial control programs and incidence of Gangrenous Dermatitis in an endemic broiler complex: Part 1: Field Observations

G. Donald Ritter

Mountaire Farms Inc., Po Box 1320 Millsboro, De 19966

The incidence of Gangrenous Dermatitis (GD) was documented over time in a large commercial broiler complex endemic for this disease condition. A seasonal pattern of GD incidence was identified. Next the impact of the use of different coccidial control programs, including ionophores, chemicals and biologics, on the incidence of GD diagnosed in the complex was investigated. Clear correlations between the use of various coccidial control programs and the incidence of GD were observed.

7625

Correlation between use of varioius coccidial control programs and incidence of Gangrenous Dermatitis in an endemic broiler complex: Part 2: Gastrointestinal microbiota

G. Donald Ritter, J. A. Benson, S. M. Dunham, A. P. Neumann, T. G. Rehberger and G. R. Siragusa

Mountaire Farms Inc., Po Box 1320 Millsboro, De 19966

Samples from broilers on farms identified as endemic GD farms were collected for gastrointestinal microbiota determination while on different coccidial control programs. Terminal restriction fragment length polymorphism (TRFLP) was used to examine the bacterial community profile of gastrointestinal tracts collected from broilers. The community profiles of broilers grown on endemic farms receiving different coccidial control GD farms were then compared and related to the expression of clinical GD during that specific flock cycle.

Comparison of turkeys from flocks raised on farms either endemic or nonendemic for gangrenous dermatitis: Part 1 Microbiologic differences

Michelle M. Andersen, Jodi A. Benson, Susan M. Dunham, Anthony P. Neumann, Thomas G. Rehberger and Gregory R. Siragusa Jennie-o Turkey Store, Inc.

Within a single turkey production system, there can be distinct differences between farms with regards to prevalence of gangrenous dermatitis. The purpose of this study was to determine if differences exist in the prevalence and genotype of key bacterial species, i.e. Clostridium perfringens, C. septicum and avian pathogenic Escherichia coli, in turkeys grown on endemic dermatitis farms versus those grown on non-endemic farms. Flocks matched based on age and feed source were sampled biweekly starting at approximately five weeks of age. Sampling continued until a flock was treated for dermatitis. Gastrointestinal tracts, livers, spleens and litter were collected for analysis.

7627

Comparison of turkeys from flocks raised on farms either endemic or nonendemic for gangrenous dermatitis: Part 2 Gastrointestinal microbiota

Michelle M. Andersen, Jodi A. Benson, Susan M. Dunham, Anthony P. Neumann, Thomas G. Rehberger and Gregory R. Siragusa Jennie-o Turkey Store, Inc.

Previous work has identified differences in the bacterial communities of the gastrointestinal tract in birds with dermatitis when compared to the bacterial community profile found in healthy flockmates. The exact role these changes play in the development of disease remains unclear. For this study, terminal restriction fragment length polymorphism (TRFLP) was used to evaluate the bacterial communities of gastrointestinal tracts collected from turkeys in the weeks leading up to a dermatitis break. The community profiles of turkeys grown on endemic farms were then compared to those of age-matched turkeys raised on non-endemic farms.

Quantitative detection of *C. septicum* in turkey flocks experiencing reoccurring episodes of gangrenous cellulitis by real-time PCR

Anthony P. Neumann, Susan M. Dunham, Jodi A. Benson, Michelle M. Andersen, Thomas G. Rehberger and Greg R. Siragusa

Clostridium septicum is a motile, spore-forming anaerobe that has been implicated as a causative agent of spontaneously occurring myonecrosis, also known as gangrenous cellulitis or dermatitis, in poultry. In recent years these diseases have consistently ranked as top concerns for both turkey and broiler producers. Although C. septicum has been commonly recovered from diseased tissues, its prevalence and impact under normal rearing conditions remains not well understood due to a lack of sensitive and specific detection methods. For this study a real-time PCR assay that specifically detects the C. septicum alpha-toxin gene (csa) was utilized to quantitatively assess the prevalence of C. septicum bacteria in turkey flocks experiencing reoccurring episodes of infection. Template DNA was isolated from gastrointestinal tract sections (duodenum, jejunum and ileum) as well as organs (liver and spleen) and necrotic muscle, in cases of disease, in order to trace the route of infection. Of the tissues tested, gastrointestinal tracts were the most likely to be positive for csa, although at an average level roughly 2.4 logs lower than colonized organs. Necrotic muscle was found to contain the target at average levels 5.5 logs greater, or 300,000 times more than the average positive gut and 3.1 logs, or about 1200 times more than the average colonized organ. These results support the hypothesis that C. septicum is common in low levels in the GI-tract, capable of translocation to organs and the musculature where substantial proliferation with accompanying toxigenesis leads to severe necrosis and systemic disease.

Immune reactive components of Clostridium perfringens causing Cellulitis in turkeys

Anil J. Thachil, David A. Halvorson, and Kakambi V. Nagaraja University Of Minnesota

Clostridium perfringens is widely considered as one of the major organism responsible for cellulitis in turkeys, a major cause of economic loss to turkey producers over the last few years. Though Clostridium perfringens toxoid was found to protect turkeys against cellulitis the role of immunity is not studied in detail. The objective of our study was to identify the secretory components of Clostridium perfringens in eliciting the protective immune response in turkeys. C. perfringens isolated from cases of cellulitis was grown and allowed to produce toxins in suitable media. The culture supernatant was subjected to SDS-PAGE to separate the toxin components. A western blot was performed using convalescent sera from the birds exposed and non-exposed to C. perfringens toxins. Our results suggested involvement of different toxins in eliciting protection against cellulitis in turkeys. The results of this experiment will be presented.

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Control of Turkey cellulitis using an inactivated Clostridial toxoid vaccine

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We have consistently isolated C.perfringenes and C. septicum from cases of Cellulitis in turkeys. We screened several Isolates of Clostridium perfringens and Clostridium septicum from cases of cellulitis for Production of maximum spore & Toxin under laboratory conditions We examined the role of these toxin preparations in cellulitis induction in turkeys. We prepared a Formalin inactivated anaculture toxoid preparations from isolates of C. perfringens and C. septicum that produced more toxins. We tested for its safety and efficacy in 5-week-old turkeys at two dose levels and booster vaccinations at our research facilities. The vaccinated birds were also challenged. A field trial was also conducted. A reduction in mortality and antibiotic usage were attained in outbreaks of cellulitis in the field following the use of experimental toxoid.

Prevalence of *netB* positive and negative isolates of *C. perfringens* from healthy and diseased animals, and their ability to produce necrotic enteritis

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The newly discovered NetB toxin, which is produced by some strains of *Clostridium* perfringens, has been reported to be critical to the development of necrotic enteritis (NE) in poultry. We investigated the prevalence of the *netB* gene among local US C. perfringens isolates. We found the gene in some, but not all, strains recovered from cases of NE. netB was also detected in isolates from normal birds and a cow. The pathogenicity of *netB* positive and negative strains from cases of NE, and *netB* positive strains recovered from normal birds and a cow were investigated.

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Importance of mixed Clostridium perfringens isolates in the experimental reproduction of necrotic enteritis in broiler chickens, a new infection model

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The effect of various concentrations of wheat in the diet and pre-challenge doses of Eimeria spp. as risk factors in a necrotic enteritis infection model were compared. Field isolates of Clostridium perfringens (Cp) obtained from clinical cases were characterized. A mixed inoculum was used and naive chickens were placed with the experimentally infected birds. After challenge, clinical signs were recorded, macroscopic lesions scoring and intestinal bacterial counts were done. A pulse-field gel electrophoresis was performed on pre- and post-infection Cp isolates. Clinical signs consistent with necrotic enteritis and mortality were observed in two groups. Cp horizontal transmission will be described.

7633 **Molecular Toxinotyping of Clostridium perfringens Isolates from Organic Farms**

Jessica C. Brady, Omar Abu-Dahab, Carlyle Bennett, Bill Guenter, James D. House, Juan C. Rodriguez-Lecompte

Department Of Animal Science, University Of Manitoba

Organic agriculture has been growing in popularity due to concerns raised by antibiotic bans. Organic poultry farms are unable to use growth promoting antibiotics to control Clostridium perfringens. Our goal was to characterize the genetic toxinotype of Clostridium perfringens isolates. Ten different C. perfringens strains were isolated from animals showing signs of necrotic enteritis. Capillary DNA sequencing and diverse PCR assays were done to create a phylogenetic analysis and toxin type and quorum sense profile, respectively. Sensitivity tests were run on C. perfringens and associated bacteria. This preliminary in vitro assay showed that the bacterial profile is similar to that of conventional systems and seems to be associated with a farm factor.

Ulcerative enteritis in broiler breeder chickens and captive capercaillie (*Tetrao urogallus*) in Scotland in 2008.

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Ulcerative enteritis (UE) in broiler breeder chickens in the early part of lay, and in two captive capercaillies was investigated. Lesions were consistent with UE. Spore forming bacilli resembling *Clostridium colinum* were seen in direct smears of liver lesions, but difficulty was experienced in isolating this organism. Others have recently reported difficulty with isolation of *C. colinum* (1.) The cases in Scotland are described and anaerobic culture versus alternative ways of confirming involvement of *C. colinum* discussed. *C. colinum* was demonstrated by PCR in the capercaillies.

A decrease in FOCAL DUODENAL NECROSIS (FDN) lesion number and severity with use of FDN specific Direct Fed Microbial (DFM).

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Agtech Products, Waukesha, Wi

Focal Duodenal Necrosis (FDN) is a disease affecting layer facilities. The recognition of this disease is increasing within the layer industry. As FDN is better understood it is clear that a prevention/treatment other than antibiotics would benefit the industry. A DFM was developed to reduce the levels of Clostridium colinum, an organism correlated to the FDN process. This DFM was fed over the course of 53 weeks. Monthly samples were taken, duodenal samples were analyzed, a lesion score given based on lesion severity. A decrease in FDN lesion number and severity was seen in the DFM treated verses control houses.

Turkey Cellulitis Incidence in a Dexamethasone Immunosuppression Model

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USDA/ARS Poultry Production and Product Safety Research Unit

Stress-induced immunosupression results in lesions of turkey osteomyelitis complex (TOC) in a Dexamethasone (Dex) model using air sac inoculation of 50-100 cfu of Escherichia coli or Staphylococcus aureus. In this model, cellulitis lesions identical to those seen in field cases of Clostridial Dermatitis are consistent and are associated with late mortality. We hypothesize that production stress alone can undermine resistance to opportunistic pathogens in fast-growing male turkeys by disruption of the skin's antimicrobial barrier resulting in the cutaneous and subcutaneous lesions referred to as cellulitis.

Some of the common characteristics between TOC and Clostridial Dermatitis are that they are both caused by opportunistic bacterial species that are part of the normal flora and are prevalent in the environment, they are both most common in male birds, and in both diseases the affected birds are usually large, healthy, and from the best performing flocks. Stress is known to result in diuresis, and wet litter is associated with Clostridial Dermatitis and is a hallmark of the Dex model.

The objective of this study was to determine the incidence of cellulitis lesions in 5 separate studies using the Dex model that were conducted from 1998 – 2001 and to determine the relative impact of Dex treatment and bacterial challenge.

In three studies using an *E. coli* challenge Dex increased the incidence of cellulitis with a main effect mean (MEM) P value of < 0.0001. In one of these studies there was no

effect of *E. coli* challenge on the incidence of cellulitis (P = 0.85), and in the other two studies *E. coli* challenge significantly decreased cellulitis incidence compared to Dextreated and non-challenged birds (P = 0.002 and 0.05). In two studies using a *S. aureus* challenge, Dex increased the incidence of cellulitis with MEM *P* values of <0.0001 and 0.002, and the retrospective *S. aureus* challenge had no effect on cellulitis incidence (*P* values = 0.6 and 0.2).

These data clearly show that bacterial challenge had no effect in the etiology of cellulitis lesions in these studies, suggesting that the effect of Dex treatment on the host immune response was the major factor in the disease and that causative bacteria were acquired from the environment or from the birds' normal flora. While Clostridia species have not been studied using this model, their natural presence in the turkey gut and production environment suggests that they may have been concomitant pathogens.

The high incidence of cellulitis lesions in the Dex model and the prevalence of both TOC and Clostridial Dermatitis in fast-growing male turkeys suggest that stress may be the most important element in the etiology of both production problems and that a stress model may be useful for development of preventative treatment for Clostridial Dermatitis.

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The Environmental Impact, Welfare and Production Characteristics of First-Lay and Molted Turkey Breeder Hens

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Some information is available on the environmental impact, welfare, and production characteristics of first-lay and molted table-egg chickens. Very little such information is available for turkey breeder hens - the only other commercial poultry species that is molted with some frequency. Absent such information on turkey breeders, regulatory and welfare policy for both species is frequently determined by using the information available on caged layers, despite the marked differences in housing, production and genetics. This presentation should aid in providing information specific to turkey breeders so that such interspecies extrapolations would be unnecessary.

A survey to investigate the adoption of biosecurity measures among commercial broiler growers in Georgia

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Biosecurity is a major part of poultry disease prevention and containment. To investigate the compliance in Georgia, an anonymous survey was distributed among commercial broiler growers in 6 companies. Questions assessed biosecurity practices required of personnel and visitors, visitor access to birds, methods of cleaning and disposal, and the sharing of equipment. The results show that the biosecurity required of visitors is higher than that adopted by personnel, but overall compliance is still low. Visitors with no direct role in poultry services may be allowed inside the poultry houses. Equipment is usually not disinfected, but sharing of equipment is rare.

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Biosecurity in a newly constructed egg production facility: results after 12 years

David A Halvorson

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Egg production in the United States (USA) predominantly takes place in large multipleage egg production complexes. The reason for this concentration of hens is the efficiency gained by conveying the eggs directly from the hen house to an attached processing facility. Because eggs are processed on site, scores of employees are needed and there may be frequent indirect contacts with other farms through equipment, crews, egg movement as well as the many necessary visitors that come to the location. These complexes are known for their tendency to harbor infections with Mycoplasma gallisepticum (MG), infectious laryngotracheitis (ILT) virus, some live vaccines as well as infestations with ectoparasites such as Ornithonyssus sylviarum (the northern fowl mite). A new farm was planned, designed, built and operated with the goal of having as few unnessary diseases as possible. After 12 years of operation this farm remains free of MG, ILT and northern fowl mites.

Expression of Viral Immunogenic Proteins in Stable Cell Culture Using a Retroviral-Based Gene Delivery System

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Inactivated vaccines are largely used to increase levels of circulating antibodies. Several avian viruses are difficult to propagate in laboratorial condition and others have high biosecurity regulations. We have developed a system for subunit vaccine production using a replication-defective retroviral vector derived from non-oncogenic natural recombinant viruses. Avian fibroblastoid DF-1 cells have been successfully transduced to express GFP and IBDV VP234 polyprotein. DF-1 transduced cells are clone selected for G-418 resistance. The system is now been used for expression of several other avian viral immunogenic proteins, which included Chicken Anemia Virus, Infectious Laryngotracheitis virus and Avian Influenza Virus.

7641

Detection of R11/3 virus in experimental and naturally-occurring cases of transmissible viral proventriculitis using a reverse transcriptase-polymerase chain reaction procedure

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R11/3 virus previously was determined to be the etiology of transmissible viral proventriculitis (TVP); physiochemical and genome sequence analyses identified the virus as a novel chicken birnavirus. A reverse transcriptase-polymerase chain reaction (RT-PCR) procedure was developed based on nucleic acid sequences within the R11/3 virus VP1 (RNA-dependent RNA polymerase) gene region. Preliminary studies have indicated that this RT-PCR procedure is a highly sensitive and specific detection method. Further evaluation of the RT-PCR procedure for detection of R11/3 virus in proventriculi of chickens with experimentally-induced and naturally-occurring TVP will be based on comparison with virus isolation and immunohistochemical detection methods.

Antigenic Characterization and VP2 analysis of Delmarva IBD Field Viruses

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Infectious bursal disease virus (IBDV) isolates were obtained from three- or four- weekold commercial broiler chickens raised on the Delmarva peninsula region of the United States in 2007. The VP2 genes of sixteen isolates recovered from broilers were sequenced to determine their relatedness to reference strains. Isolates representing five different phylogenetic clades were further analyzed by virus-neutralization and monoclonal antibody typing to determine their relatedness to known IBDV strains. The isolates produced gross and microscopic lesions consistent with IBDV infection in the commercial broilers and experimentally in specific-pathogen-free leghorn chickens.

7643

Isolation and Characterization of a new variant Infectious Bursal Disease Virus identified by a diagnostic approach using reverse genetics

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The identification of the emergence of antigenic variant IBDV is of great importance due to their ability to break through existing vaccination programs. We describe the identification of a new variant IBDV using reverse genetics which showed an unusual reaction panel using monoclonal antibodies. In animal experiments using SPF birds the virus was able to induce complete destruction of the Bursa Fabricii (BF) showing a high portion of necrosis. In commercial broiler with high levels of maternal derived antibodies this isolate was able to induce complete destruction of the bursal follicles indicating a new antigenic subtype of IBDV.

Efficacy of Single dose recombinant HVT-IBDV vaccination (Vaxxitek ®) against classical and variant IBDV strains

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To assess the protection conferred by day-one vaccination using VAXXITEK®, (a recombinant HVT expressing the VP2 from a classical IBDV), one-day old SPF and/or commercial broilers were vaccinated and then challenged with classical or variant E IBDV strains at 18 or 28 days. Clinical signs, bursa/bodyweight ratio and bursal histopathology were evaluated. No clinical signs or IBDV challenge related lesions were observed in the VAXXITEK® vaccinated birds at both early and late challenge. Unvaccinated challenged birds showed significantly lower bursal indexes. Cross-protection was demonstrated for the variant E challenge. Results indicate that single dose recombinant HVT-IBDV vaccination protects against classical and variant IBDV strains.

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Spectrum of protection provided by passive and active immunity to IBD viruses

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The authors investigated the value of passive (maternally derived antibodies) and active immunity (elicited by vaccination with live IBDV vaccine) in protection against homologue and heterologue antigenic type IBDV infection. For this purpose chicks carrying MAB to classical antigenic type IBDV and chickens vaccinated with classical antigenic type live vaccines were challenged with either homologue or heterologue antigenic type IBD viruses. The protection against the clinical disease as well as the protection against infection was evaluated by gross, histopathological and molecular (RT-PCR) methods. The results that will be presented indicate an antigenic type-independent protection for passive immunity and a broad, antigenic type-independent protection for active immunity.

Comparison of water based foam depopulation with and without CO2 gas to CO2 and Ar-CO2 gassing

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Current control strategies for Avian Influenza (AI) and other highly contagious poultry diseases include surveillance, guarantine, depopulation, disposal, and disinfection. United States Department of Agriculture (USDA) conditionally approved the use of water based foam in 2006. In this experiment, carbon dioxide gassing (CO₂), Argon -CO2 gassing, water based foam with CO₂ gas, and water based foam with ambient air were compared. A total of 68 broilers were individually instrumented and depopulated with one treatment. Foam was more consistent than the two gassing methods tested; however, there were no statistically significant differences between foam methods. The addition of CO₂ to the foam appears to have no practical advantage, minimal impact on bird welfare, and does not materially change cessation time.

7648

Effects of increased light intensity on tendon ruptures in eight pound broilers

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Four trials were conducted to investigate if effects of light intensity would decrease tendon ruptures in 8 pound broiler chickens. Each trial was conducted on a different contracted poultry farm, where 3 houses were used; 1 test and 2 controls. All birds were raised by company standards until the last two weeks before processing. Control houses continued company standards; continuous lighting of 0.03 Foot-candles. The test house had intermittent lighting of 0.3 Foot-candles for 30 minutes, and 0.03 Footcandles for 1 hour, 30 minutes. Samples were taken at processing to assess chronicity of lesions and efficacy of the light trial.

The beta-tubulin sequence of H. meleagridis suggests susceptibility against Benzimidazoles and confirms the phylogenetic position of H. meleagridis

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Benzimidazoles are antiparasitic drugs that act on beta-tubulin. The sequence of betatubulin allows predictions about their efficacy. We sequenced and analyzed the betatubulin gene of five H. meleagridis strains and of the close relative Dientamoeba fragilis. All histomonal amino acid sequences predicted a susceptibility to Benzimidazoles. In spite of this five Benzimidazoles showed no efficacy against H. meleagridis in vitro, even in higher concentrations than concentrations found to be effective against other flagellates. A phylogenetic tree of trichomonal beta-tubulin genes placed the histomonal sequences on a branch with D. fragilis, close to Monocercomonas sp. and Tritrichomonas foetus.

7650

Dimerization partners of Marek's disease virus encoded Meq protein play an important role in T-cell transformation

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Marek's disease virus (MDV), the etiologic agent of Marek's disease, is a potent oncogenic herpesvirus. MDV encodes a Meq protein that shares resemblance with the Jun/Fos family of transcription factors and we have earlier shown that Meq is essential for transformation of T-cells. Similar to Jun, the leucine zipper region of Meq allows the formation of homo and heterodimers. Earlier studies indicate that Meq homo and heterodimers may differentially mediate transcription regulation of viral and cellular genes. In order to study the role of Meq dimerization in the pathogenicity of MDV, mutant viruses that allow for the exclusive homo- or heterodimerization of Meq were generated. In vivo characterization of these mutant viruses indicates that Meq heterodimerization is required for T-cell transformation.

Protection properties of mutant rMd5 viruses expressing the Meq protein of CVI988 vaccine strain

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Meq is a proven oncoprotein of Marek's disease virus (MDV). Though the vaccine strain CVI988 is non-oncogenic, it encodes for two forms of Meq, CVI988 Meq and CVI988 long Meq. We have previously shown that the Meq proteins from both pathogenic (Md5) and vaccine (CVI988) strains are capable of transforming fibroblasts in vitro. To study the role of CVI988 Meq proteins in pathogenesis, we have generated mutant rMd5 viruses in which the meq gene was replaced with those from CVI988 vaccine strain. In vivo characterization of these viruses showed that these viruses were attenuated suggesting that the meq gene from CVI988 is involved in pathogenesis. In maternal antibody MDV positive chickens these mutant viruses did not cause any MD. In this report we will discuss the protection properties of these mutant viruses when challenged with a very virulent plus MDV.

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Factors affecting the efficacy of recombinant Marek's disease vaccine protection

Lucy E. Lee

Many factors have the potential to influence the efficacy of Marek's disease (MD) vaccination. Some of these factors include maternal antibody, vaccine dose, age of birds at vaccination or challenge, challenge virus strain and genetic background of chickens. The objective of this study was to evaluate the effect of challenge virus dose and age of chickens at exposure on the efficacy of recombinant vaccine protection. The vaccine used in this study was a gene deletion mutant of rMd5 (rMd5 Δ Meg) and the challenge virus was 686, very virulent and very virulent plus strains of MDV, respectively. In the first experiment, six groups of 17 birds each were vaccinated at day of age with 2000 PFU of rMd5∆Meg and challenged on different days ranging from zero to 5 days post vaccination. In the second experiment, six groups of 17 birds each were vaccinated at day of age with rMd5 Δ Meg and challenged with 686MDV, ranging from 500 to 100,000 PFU per group five days later. Data from the first experiment shows a higher protective index at day zero than day 1. A protection index of 100% was obtained only when birds were challenged 5 days after vaccination. The data from the second experiment indicates that the vaccine dose of 2000 PFU could protect 100% of chickens challenged at a dose from 500 to 32,000 PFU/bird. However, at a dose of 100,000 PFU/bird, the protective index was only 85%. These data taken together indicate that dose and time post vaccination plays an importance role in the efficacy of vaccine protection.

Characterization of the immune responses elicited by double vaccination against Marek's disease

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Population Health And Pathobiology Colege Of Veterinary Medicine North Carolina State University

We have recently demonstrated that in ovo vaccination (HVT) against Marek's disease followed by a second vaccine (HVT + SB-1) at day of age confer higher protection than either vaccine alone. Double vaccination reduced not only the mortality and percentage of MD but also decreased the load of challenge MDV DNA in the feather pulp. In this work, we have studied the immune response elicited by the double vaccination protocol. Activation and expansion of T cells as well dynamics of MD vaccine infection have been evaluated after administering single vaccination or after double vaccination.

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Effect of route of vaccination with serotype 1 Marek's disease vaccines on the recruitment of lymphocytes and macrophages in the lungs

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In previous studies, we have demonstrated that strong replication in the lung is a common feature to highly protective serotype 1 Marek's disease vaccines and also that the route of inoculation greatly affects the pulmonary tropism. The objective of this study was to further evaluate the inflammatory infiltration in the lungs following vaccination in ovo (18 days of embryonation) and subcutaneously (hatch) with vaccine strains R2, R2/23, and CVI988. Samples of lungs were collected at 3, 5, and 10 days post vaccination and evaluated for the presence of CD4, CD8, macrophages, B cells, MHC-II, and viral antigens pp38 and gB.

Virus competition for shedding and tumor formation over time in Marek's disease virus dual-infected chickens

John R. Dunn, Richard L. Witter, Robert F. Silva, Lucy F. Lee, Scott D. Fitzgerald, John B. Kaneene, Richard M. Fulton

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This study was designed to determine what effect multiple virulent Marek's disease viruses have on each other over time during dual-infection. Serotype 1 viruses able to be differentiated were administered either simultaneously or with a short (24 hours) or long (13 days) interval. Virus frequency and quantity was measured in tumors at termination and from feathers at three time points from 226 birds with tumors. The same virus tended to dominant at all time points, but the particular virus that dominated was more affected by time interval between challenges than by virulence. Even after the long interval, the second virus can still be important.

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Role of Pulmonary Immune Response on the Efficacy of Serotype 1 Marek's **Disease Vaccines**

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We have recently demonstrated that highly protective (HP) serotype 1 Marek's disease vaccines replicate more in the lung than lower protective (LP) serotype 1 vaccines. In this study we have evaluated differences in the pattern of cytokine mRNAs in the lung and in the spleen induced by HP and LP vaccines within the first week after vaccination. Expression of cytokines in the lung had a distinct pattern from that observed in the spleen regarding onset of expression and level of expression of certain cytokines. HP vaccinated chickens had similar or higher levels of cytokine expression at 3 and 5 dpv than LP vaccinated chickens but much lower levels at 10 dpv with the exception of IL6 and IFNÂ³.

Detection of VECTORMUNE® HVT Vaccines in Feather Tips by Real-Time PCR

Motoyuki Esaki, Takanori Sato, Lauren Jensen, Shuji Saitoh, Sakiko Saeki, Ayumi Fujisawa and Kristi Moore Dorsey Ceva-Biomune

VECTORMUNE® HVT vaccines contain a turkey herpesvirus backbone expressing protective antigens from various pathogens, such as infectious bursal disease virus, Newcastle disease virus and laryngotracheitis virus. The objective of this study was to establish a method to confirm successful vaccination with these vaccines and to evaluate replication of vaccine viruses in chickens. We developed real-time PCR assays that specifically detect and quantify each of VECTORMUNE® HVT vaccines in feather tips of chickens vaccinated with VECTORMUNE® HVT vaccines.

Immunological Basis for Resistant and Susceptibility to Marek's Disease

Mohammad Heidari

Marek's disease (MD) is a contagious lymphoproliferative disease of domestic chickens caused by a highly cell-associated α -herpesvirus, Marek's disease virus (MDV). MDV replicates in chicken lymphocytes and establishes a latent infection within CD4⁺ T cells. Mechanisms of viral pathogenesis, resistance to MDV infection/oncogenesis, and the protection process of attenuated vaccine strains are poorly understood. Clearly, a complex set of interactions between viral genes and host's immune responses are involved. To better understand the molecular mechanisms overriding these events, the expression pattern of cytokines, chemokines, and other immune related genes were investigated between MDV-infected chickens of lines 6₃ (MD-resistant) and 7₂ (MDsusceptible). Real-Time PCR analysis revealed high expression levels of IL-1_β, IL-6, IL-8, iNOS, IFN β , IFN γ , and IFN λ in the spleen tissues of MDV-infected susceptible chickens when compared to un-infected control birds. In contrast, only a limited number of cytokines were up-regulated in the MDV-inoculated resistant chickens (IL-6, IL12p35, IFN_{γ}, and IFN_{λ}). In addition, immunohistochemical analysis showed an extensive infection and inflammation in the bursa and spleen tissues of MDV-infected birds from line 7₂. No infection was observed in the tissues of MDV-inoculated chickens from line 6₃. This study reveals significant differences in immunological responses to MD between the resistant and susceptible lines.

The importance of pp38 splice variants for the pathogenesis of Marek's disease

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Previously we have shown that Marek's disease virus pp38 produces 2 splice variants. To determine the importance of the splice variants we generated recombinant RB-1B virus strains using the rBB-1B BAC clone, in which pp38 is mutated to only express splice variant A (rRB-1B/A), splice variant B(rRB-1B/B), or with the two potential splice acceptor sites mutated (rRB-1B/M). In vitro growth curves indicate that rRB-1B/M, rRB-1B/A and rRB-1B/B yield 3, 146, and 70 times less virus than the wild-type RB-1B. The results of in vivo experiments examining the effects on the pathogenesis of infection and tumor development will be reported.

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Insertion of a Reticuloendotheliosis Virus LTR into the Marek's Disease Virus Genome

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USDA, Agricultural Research Service

Marek's disease virus (MDV) had previously been co-cultivated in culture with Reticuloendotheliosis virus (REV). During co-cultivation, a long terminal repeat (LTR) from REV was inserted into the MDV genome. The resulting MDV, designated RM1, was attenuated but still induced severe thymic and bursal atrophy in inoculated chickens. To determine whether the LTR insertion was responsible for the altered phenotype, we isolated and cloned an REV LTR into a bacterial artificial chromosome clone of MDV. We will report on the cloning procedure and preliminary characterization of the chimeric MDV.

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Pathogenicity of a molecular clone of Marek's disease virus with an insert of Long Terminal Repeat of (LTR) of reticuloendotheliosis virus (REV).

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Recently, we have inserted reticuloendotheliosis virus (REV) long terminal repeat (LTR) sequences into strain Md5 of Marek's disease (MD) virus (MDV) using rMd5 bacterial artificial chromosome (BAC). The rMd5 BAC with REV LTR inserts was passed in duck-embryo fibroblast for 40 passages. Chickens of ADOL line 15 X 7 with and without MD maternal antibodies were used to study the pathogenicity of rMd5 BAC with REV LTR insert, and to compare with that of wild type strain Md5 of MDV and rMd5 BAC without REV LTR insert. The influence of REV LTR inserts on the pathogenicity of rMd5 will be discussed.

Pathogenesis of vetebral osteoarthritis ("Spinal Abscesses") in male broiler breeders caused by enterococcus cecorum

Leslie T. Martin, Michael P. Martin, and H. John Barnes

University Of North Carolina - Chapel Hill

Vertebral osteoarthritis is an emerging disease of young male broiler breeders first identified in 2006. Cases continue to occur in male breeders and broilers in the US and Canada. The disease is characterized clinically by lameness, paresis, and/or paralysis. Abscesses are found in the free thoracic vertebra and cause damage to the spinal cord. Enterococcus cecorum is isolated from almost all the abscesses found in field cases. Mortality has ranged from 5-20%. A pathogenesis study in male broiler breeders was conducted. Challenge routes included intravenous, intra-air sac, and oral gavage. Data including gross necropsy, histopathology, and bacteriology will be presented.

7663

Deciphering the role of Polyphosphate kinases in Campylobacter jejuni colonization/ pathogenesis

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Campylobacter jejuni is a major cause of bacterial food poisoning worldwide accounting for up to 6 billion losses in the United States alone. Despite the availability of complete genome sequence, the pathogenesis/colonization mechanisms of this bacterium are poorly understood in contrast to other major food borne pathogens. Polyphosphate kinase (PPK) is a critical enzyme in the synthesis polyphosphate. In many bacteria, inorganic polyphosphate has been demonstrated to be important for stress response. survival and infection of the host. Campylobacter stress response, in the absence of conventional stress genes seen in other enteric pathogens, is important for this fastidious pathogen to survive outside the animal host. In the present study, we have investigated the importance of homologs of ppk genes in C. jejuni physiological functions and colonization. Our study showed that ppk mutants were defective in motility, biofilm formation and In vivo colonization in chicken. Electron microscopy examination showed reduced poly P accumulation in the cytoplasm, increased sensitivity to various antimicrobials and decreased ability to take DNA from the environment. Further, the contribution of PPK to the ability of Campylobacter to resist various environmental stresses would be discussed. The present work will have implications for developing strategies to control Campylobacter as well as emergence of drug resistant Campylobacter.

7664 **Reduced fitness associated with macrolide-resistant Campylobacter in chickens**

Taradon Luangtongkum, Virginia Seng, Orhan Sahin, and Qijing Zhang

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Campylobacter, a major foodborne human pathogen, is prevalent in poultry and is increasingly resistant to macrolide antibiotics. In this study we determined if macrolide-resistant Campylobacter was able to persist in chickens in the absence of antibiotic selection. Multiple chicken experiments consistently showed that macrolide-resistant Campylobacter was outcompeted by macrolide-susceptible strains in chickens, indicating that acquisition of macrolide resistance incurs a fitness burden in Campylobacter. This fitness cost was linked to the resistance-conferring mutations in the 23S rRNA gene. Our results suggest that appropriate management practices can be implemented to minimize the development of macrolide resistance in Campylobacter.

Isolation and Identification of *Enterococcus cecorum* from Commercial Broiler Farms in Central Pennsylvania

S. Kariyawasam, P. A. Dunn, E. A. Wallner-Pendleton, S. Myers, and Valerie Lintner

Animal Diagnostic Laboratory, The Pennsylvania State University, University Park, PA 16802

Multiple outbreaks of osteomyelitis and septicemia caused by *Enterococcus cecorum* were identified recently from commercial broiler farms in central Pennsylvania. Affected chickens were between 16 to 70 days of age. Clinical characteristics of the process included lameness, increased culling rate and increased mortality. Histologically, dyschondroplasia and necrotizing osteomyelitis of the proximal tibiotarsus, femur and/or vertebrae were noted. Gram-positive, non-hemolytic or weakly alpha-hemolytic cocci were isolated from bones, joints, livers, pericardial fluid, and/or yolk sacs of the affected chickens. A Trek Diagnostic Systems Sensititre, gram-positive identification (GPID) plate misidentified the organism as *Streptococcus suis*. Further identification by sequencing the 16S ribosomal RNA (16S rRNA) gene identified the organism as *E. cecorum*. Due to misidentification, we suggest that *E. cecorum* may be an under-reported cause of bacterial infections in poultry. This presentation describes growth characteristics, and biochemical and molecular identification of *E. cecorum* from poultry.

Consumers: Their Perceptions and How They Change

Robert L Owen, Jeffrey Mellinger, Michael Opperman

Alpharma Animal Health 2228 Oxford Rd, New Oxford, Pa 17350

In order to better understand consumer preference as it relates to antibiotic use in food producing animals, a customer survey was conducted to investigate consumer perception before and after education about antibiotic use. Results of the survey showed that Americans when uneducated about the importance of antibiotics tended to prefer meat and poultry from animals raised without antibiotics. When educated, consumers are comfortable eating meat and poultry from animals treated with antibiotics. This work shows the importance of educating consumers about the realities of animal agriculture.

7668

Molecular Characterization of Eimeria species infecting turkeys

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Pure isolates of two species of Eimeria infectious to turkeys, Eimeria adenoeides and Eimeria meleagridis, were obtained. Genomic DNA was extracted and three genomic regions: internal transcribed spacer regions 1 and 2 (ITS-1 and 2), the cytochrome oxidase subunit I (cox-1), and small subunit ribosomal RNA (SSU), were PCR amplified, cloned, and sequenced. This work represents the first molecular characterization of Eimeria from turkeys. The data suggests that turkey Eimeria form a specific clade distinct from Eimeria sequences that infect other galliforme birds. Data comparing genetic variability of Eimeria in turkeys to that of chicken Eimeria will be presented.

7669

Using molecular techniques to differentiate vaccine and field strains of Eimeria spp. oocysts in coccidiostat- and vaccine-utilizing poultry farms

Mark Jenkins, Katarzyna Miska, Raymond Fetterer, Spangler Klopp Animal Parasitic Diseases Laboratory, Ars, USDA, Beltsville, Md 20705

The technique of rapidly isolating Eimeria oocysts and identifying the species composition of coccidia in poultry litter was improved further by using DNA probes followed by automated DNA sequencing that can differentiate vaccine-originating strains from the resident Eimeria population. The value of this improved technique is that as new field variants arise in poultry facilities, controls using either anti-coccidial drugs or live oocyst vaccines can be modified to minimize coccidiosis outbreaks. Research is on-going to identify DNA markers for drug-resistance in Eimeria, and an application of molecular typing to track drug-resistant Eimeria acervulina is being pursued.

7670 Histopathological changes in the small intestine associated with Eimeria praecox infection.

Jenny A. Fricke, Greg. F. Mathis, Charles L. Hofacre, Blair E. Telg, Andres F. Montoya and Susan M. Williams

Department Of Population Health, Poultry Diagnostic And Research Center, The University Of Georgia

Eimeria praecox is generally regarded as a lesser pathogenic to non-pathogenic coccidial parasite of chickens. This parasite develops in the duodenum and jejunum, often with no obvious gross lesions, and has a short pre-patent period. Because of these characteristics, E. praecox infection is often overlooked. However, previous and current work has documented loss of pigmentation, decreased weight gain and poor feed conversion in association with E. praecox infection. The histopathological changes associated with E. praecox infection of the small intestine, which play a role in these pathologic effects, are presented.

7671

How Eimeria praecox Infection Affects Broiler Intestinal Immunity

Lindsay H. Stuard, Kate B. Miska, Mark C. Jenkins, Ray H. Fetterer, Sungwon Kim, Rami A. Dalloul

Virginia Tech

Many parasitic species of the *Eimeria* genus act as the causative agent of avian coccidiosis, a costly enteric disease to the broiler industry. The lesser pathogenic *Eimeria praecox* is usually prevalent on poultry farms but rarely included in live oocyst vaccines. While *E. praecox* effect on concurrent *E. maxima* infection was a positive impact on weight gain and feed efficiency, little is known about its influence on gut immunity of broilers. This study assessed the immunological effects that an invasion with *E. praecox* may have on the expression of innate immune parameters within the gut of infected broilers. The results of this study indicate that the expression of avian specific Toll-like receptor (TLR)-15 is downregulated (P < 0.05) in response to *E. praecox* intestinal colonization. This observation suggests that the parasite may be evading the innate immune system by affecting TLRs in a unique way. Toll-like receptors (TLRs) are transmembrane proteins that modulate non-specific immunity through cytokine and chemokine signaling and participation in the interferon pathway. TLR expression is upregulated in response to *E. praecox* infections of avian species has not been well characterized.

Comparison of broiler performance and vaccine infectivity after Inovocox" vaccination at E18 or E19.

Lauren K. Griffin, Larry Charniga, Mohamed Hamoud, Vivian Doelling, Michelle Miller, Rebecca Poston

Pfizer Animal Health, Poultry Health Division

This study examined vaccine infectivity at E18 or E19 and compared broiler performance with hatchmates vaccinated post-hatch with Coccivac-B® or non-vaccinated birds fed salinomycin-medicated rations. Rate of vaccine infectivity in individual birds was 80% and 90% for E18 and E19 Inovocox administration. Both Inovocox groups were not significantly different in BW compared to either Coccivac-B or salinomycin at all timepoints. Both Inovocox groups demonstrated superior FC at 35, 42 and 48 days when compared to the salinomycin-fed birds and on days 35 and 42 when compared to Coccivac-B. Inovocox at E19 demonstrated superior feed conversion on D48 as compared to Coccivac-B.

7673

Improved Coccidiosis Control with Anticoccidial Drugs Following Vaccination with Coccivac-B

Greg F. Mathis, and Matilde Alfonso and Charles Broussard

Southern Poultry Research, Inc., 2011 Brock Rd., Athens, Ga 30607

All Eimeria that comprise Coccivac-B are highly sensitive to anticoccidial drugs. This study examined the degree of change in anticoccidial sensitivity to the anticoccidial Salinomycin 66 ppm after repeated usage of the vaccine. Coccidia were isolated from five houses where only drugs were used or five house that had birds vaccinated with Coccivac-B. The results clearly showed that the coccidia isolated from the vaccine houses had a much higher degree of sensitivity to Salinomycin than the drug only isolates. This demonstrates that vaccination with Coccivac-B has the potential during routine commercial usage to replace resistant coccidia with sensitive coccidia.

Paired House Trials for Comparing Body Weights at 28 and 42 Days of Age in Broilers Receiving Either Inovocox or Salinomycin

Jonathan L. Schaeffer, Larry M. Charniga, John Dickson, David G. Kelly and Rebecca M. Poston

Pfizer Poultry Health Division, 1040 Swabia Court, Durham, NC 27703

Day 28 and 42 body weights were collected in paired commercial houses to compare broilers vaccinated with Inovocox to broilers fed salinomycin. Three different bird slaughter age programs were involved. No significant differences were seen at Day 28; however, on Day 42, overall (across programs and sexes) and for each sex overall, Inovocox birds weighed significantly more than salinomycin birds. Also, Inovocox bird weights showed significantly lower variability than salinomycin on Day 42. These data demonstrate that Inovocox does not adversely impact body weight during early growout and does not adversely impact the weight of broilers processed at 42 days.

7675

Is coccidiosis the problem?

Steve Fitz-Coy

Intervet/Schering-Plough Ah

Houses with high prevalence of pododermatitis also had watery and or mucoid droppings. The level of coccidia was mild. Feed samples from the chickens (A and B) and turkey (A) a basic poultry diet was control (C). Composite samples prepared from fresh feces (F) and gastrointestinal (G) content collected from the turkey farms. Young broiler chickens consumed one of the three diets for two hrs, then each bird gavaged with a 1ml dye. The feed passage rate for A was 15% faster than C and 49% faster than B. Young birds were fed one of several concoctions of the F or GI mixtures for 12 days. Chickens or poults fed C had no clinical signs, but birds fed C, spiked with F or GI exhibited signs of gastro-intestinal abnormalities. Birds fed (A) spiked with F or GI had signs of gastro- intestinal abnormalities. The signs and lesions were similar to those seen in the commercial birds. The findings indicated diet might influence the feed passage rate and moisture levels in the feces. The level of coccidia did not appear to be the main contributor in the syndrome.

Evidence of multiple recombination events in an Avian Coronavirus Infectious Bronchitis virus field isolate

Enid T. McKinley, D eboah A. Hil, t Mark W. Jackwood

Poultry Diagnostic And Research Center, University Of Georgia, Athens, Georgia 30602

RNA viruses such as avian infectious bronchitis are able to evolve rapidly by generating genotypic variation through mutations acquired from the error prone RNA-dependent RNA polymerase and also from recombination events. Evolution of these viruses has lead to the emergence of new serotypes that have crossed species barriers. In our study we sequenced and analyzed the entire genome of IBV viruses isolated over several decades to investigate mutation and evolutionary rates of these rapidly changing viruses. Our data revealed that a Mass field virus isolated in 2006 resulted from multiple recombination events between a Mass vaccine virus and an unknown number of field or vaccine viruses. Determined by the spike gene sequence, the 2006 isolate was designated as a Mass virus, however, sequence analysis of the entire genome indicated higher similarity with Cal and Conn viruses. Phylogenetic analysis of all the genes revealed incongruent trees providing additional evidence of recombination events in viruses isolated from the field. This data adds to the growing evidence that recombination plays a role in coronavirus evolution.

Genomic and Phylogenetic Comparisons of Emerging Coronaviruses

S. W. Thor, D.A. Hilt, E.T. McKinley, and M.W. Jackwood The University Of Georgia

Viruses in the Family Coronaviridae are of global economic importance. These viruses are widespread both with respect to geography and host tropism. Because coronaviruses have high rates of both mutation and recombination, it is not surprising that they have proven to be a major source of emerging zoonotic diseases. Although the spike region for many of the coronaviruses has been sequenced, there are very few full genomic sequences available. This is especially true for the coronaviruses that infect animals that are in close contact with humans; including dogs, cats, rats, mice, chickens and turkeys. Therefore, we were interested in examining the complete genomic sequence of selected animal coronaviruses from group I, II, and III. Relevance of the viruses to both animal and human health will be shown using genomic and phylogenetic analysis.

7641

Detection of chicken birnavirus R11/3 in proventriculi and feces of experimentally infected chickens using a reverse transcriptase-polymerase chain re

James S. Guy, Frederick J. Fuller, H. John Barnes, Melissa West

North Carolina State University, College Of Veterinary Medicine, 4700 Hillsborough St., Raleigh, NC

R11/3 virus previously was determined to be the etiology of transmissible viral proventriculitis (TVP); physiochemical and genome sequence analyses identified the virus as a novel chicken birnavirus. A reverse transcriptase-polymerase chain reaction (RT-PCR) procedure was developed based on nucleic acid sequences within the R11/3 virus VP1 (RNA-dependent RNA polymerase) gene region. Preliminary studies have indicated that this RT-PCR procedure is a highly sensitive and specific detection method. Further evaluation of the RT-PCR procedure for detection of R11/3 virus in proventriculi and feces of chickens with experimentally-induced TVP will be based on comparison with virus isolation and immunohistochemical detection methods.

7573

Case Report: A Control Strategy for Histomoniasis on a Replacement Broiler Breeder Farm

Mark A. Burleson

Sanderson Farms, Inc.

For six consecutive flocks, a replacement broiler breeder farm had experienced high mortality due to Histomoniasis and subsequent Staphylococcus infections. A control strategy was implemented to eliminate the disease from this farm. Darkling beetle control, litter management, and a strict deworming program were among the critical aspects of the intervention strategy. Each detail of the program will discussed.

7678 SELECTION OF MINOR VIRAL SUBPOPULATIONS WITHIN ARK-TYPE INFECTIOUS BRONCHITIS VACCINE - EFFECT ON TRACHEAL DAMAGE AND MUCOSAL IMMUNE RESPONSES

Vicky L. van Santen, Eunice N. Ndegwa, Kellye S. Joiner, Frederick W. van Ginkel, Haroldo Toro

Department of Pathobiology, College of Veterinary Medicine, Auburn University

We previously reported that commercial Ark IBV vaccines contain different proportions of a minor subpopulation that apparently replicates more efficiently in the upper respiratory tract of SPF chickens than the "major" vaccine population and is "selected" in chickens after ocular vaccination. Furthermore, SPF chickens vaccinated with vaccines containing higher proportions of the selected subpopulation had significantly higher viral loads 5 and 8 days post-vaccination (DPV). We expected the higher viral loads might result in greater damage to the trachea, but might also result in greater immune responses. Therefore, in the present report we compared tracheal damage, assessed by histopathological examination and morphometric analysis, among SPF chickens vaccinated with four different Ark IBV vaccines. At 7 DPV, chickens vaccinated with the vaccine having the highest proportion of "selected" subpopulation had more severe epithelial necrosis and deciliation in the trachea than chickens vaccinated with one of the vaccines with the lowest proportion of "selected" subpopulation, and more lymphocytic infiltration of the trachea than chickens vaccinated with the other three vaccines. In addition we compared mucosal immune responses by analysis of IgA and interferon gamma gene expression in the Harderian glands. We found great individual variation among chickens and few statistically significant differences among vaccinated groups.

7679

Assessing Intraspatial Variation of Infectious Bronchitis Virus in the Host

Rodrigo Gallardo, Vicky van Santen, Haroldo Toro

Department of Pathobiology, 264 Greene Hall, Auburn University, Auburn AL 36849

Infectious bronchitis virus (IBV) strains show different proportions of distinct subpopulations (predominant and minor). The differences between these subpopulations are mainly encountered in the Spike (S) protein, responsible for virus attachment. We have shown previously that some of these subpopulations are apparently more fit to replicate in the upper respiratory tract. We hypothesize that different tissues within the host will exert a distinct selective pressure on the virus population which might change the dominant genotypic structure. We inoculated chickens with an Ark-type IBV strain via the ocular route and examined the S gene sequence of IBV obtained from lachrymal fluid, trachea, kidney, oviduct, and cecal tonsils of these chickens at different times post-inoculation.

7680

Genomic Analysis of Pathogenic and Attenuated Strains of Infectious Bronchitis Virus

J.E. Phillips, D.A. Hilt, E.T McKinley, S.W. Thor, and M.W. Jackwood University Of Georgia, Department Of Infectious Diseases, Poultry Diagnostic And Research Center

Infectious Bronchitis Virus (IBV) is a group III Coronavirus that infects chickens causing a highly contagious upper-respiratory disease. In this study the full- length genome of pathogenic parent and attenuated progeny strains of IBV were sequenced and examined. Comparative genomics were conducted on IBV strains: Ark-DPI pathogenic, Ark-DPI attenuated, GA98 pathogenic, GA98 attenuated, Mass41 pathogenic, and Mass41 attenuated. Nucleotide and amino acid comparisons to identify differences associated with pathogenicity will be presented.

7681 Isolation and Characterization of Recent Infectious Bronchitis Virus Isolates from NE Georgia

Arun B. Kulkarni, Elena Behnke and Reynaldo Resurreccion

Georgia Poultry Laboratory Network, Oakwood, Ga 30566

The case report describes the isolation and identification of infectious bronchitis virus variants from laboratory submissions during 2008. Clinical signs ranging from respiratory rattles, nasal discharge to excessive flushing have been reported from these flocks. Gross and microscopic lesions will be described, as well as direct fluorescent antibody results and embryo lesions. Molecular and phylogenetic analysis of the 750 bp PCR product from the HVR region of the S1 gene compared to reference strains showed that all the isolates belong to the same genetic lineage that is distinct from the known groups in the US.

7682

All-Natural Compound Tested for Activity Against IBV in Chickens

Mark W. Jackwood, Richard Rosenbloom, Michael Petteruti, Deborah A. Hilt, Amber W. McCall, and Susan M. Williams

Dept. Of Population Health, Pdrc, 953 College Station Road, University Of Georgia, Athens, Ga 30602

In this study, we examined a mixture of all-natural ingredients designated QR448(a) for its effect against infectious bronchitis virus (IBV) in chickens. The best route of inoculation, minimum effective dose, duration of activity, and effect on transmission of the virus was examined. Delivery or QR448(a) by fine spray was effective against challenge with IBV for up to 4 days post-treatment, and transmission of Arkansas vaccine virus was significantly reduced. The effect of QR448(a) on IBV in vivo appears to be virucidal in nature.

Proliferative and Lymphocytic Pneumonia in Broiler Chickens

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Histopathology done on over 60 clinical cases of respiratory disease showed a proliferative and lymphocytic pneumonia. This response pattern differs from the exudative pneumonias associated with bacterial infections such as E. coli and from granulomatous pneumonias caused by fungi such as Aspergillus. Proliferative, lymphocytic pneumonia commonly was associated with hyperplasia of tracheal epithelium and lymphocytic tracheitis. Lymphocytic nephritis was present in many, but not all, kidneys submitted. Lymphocytic airsacculitis was common, but air sacs were not submitted in all cases. Most of these cases came from flocks located in areas where the GA08 variant of infectious bronchitis virus was present. Many cases came from flocks that tested negative for mycoplasma. Histologic lesions will be illustrated and differential diagnoses considered. Emphasis is on raising awareness that infectious bronchitis virus could be causing proliferative, lymphocytic pneumonia in broilers.

7684

Evaluation of Recombinant Vector-Vaccines against Infectious Laryngotracheitis (ILT) under Experimental Conditions

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Poultry Diagnostic And Research Center, 953 College Station Rd, Athens, GA 30602

Despite efforts to control the disease through vaccination with live-attenuated vaccines, infectious laryngotracheitis (ILT) continues to be a significant problem to the industry in the United States. Two recombinant products are currently available, the Herpes virus of turkey (HVT) vaccine INNOVAX"-ILT, and the Poxvirus vaccine VECTORMUNE FP-LT. The broiler industry is using both recombinant products off-label, via in ovo application, achieving variable results in the field. The objective of this study was to evaluate the efficacy of both recombinant vaccines to protect broilers against the standard USDA challenge strain after in ovo application under controlled-experimental conditions.

7685 Analysis of Climatic Factors during Outbreaks of Vaccinal Laryngotracheitis

Louise Dufour-Zavala

GA Poultry Laboratory Network

Factors affecting the behavior, length and severity of vaccinal laryngotracheitis (VLT) outbreaks in broilers in dense poultry production areas are multiple. Human factors include biosecurity procedures and the use of vaccines. Spatial factors include the distance of farms to major roads. In this presentation, climatic factors such as wind speed and direction, day and night temperatures, and amounts of rain fall are analyzed in relation to cases of VLT in one area over time.

7686

Benefits of the New Intra-Muscular Wing Killed Vaccination on Commercial Egg Pullets

Hugo Medina

Sparboe Companies

We will review and present Intra-Muscular Wing Killed Vaccination on Commercial Egg Pullets in comparison to the historically other body sites used for the application of killed vaccines. This presentation is a practical- field overview of this practices with advantages and disadvantages. This new site as IM killed vaccine was researched using field, practical, immuno responses and safety procedures and results for its implementation.

NONSUPPURATIVE MYOCARDITIS IN TURKEYS IN CALIFORNIA: A RETROSPECTIVE STUDY

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Affiliation: California Animal Health and Food Safety Laboratory System, University of California, Davis

Fifty cases of nonsuppurative myocarditis in turkeys were diagnosed at California Animal Health and Food Safety Laboratory System between 1991 and 2009. The age of the affected birds ranged from 6 days to 10 weeks and 44 cases occurred in turkey poults up to 4 weeks of age. Clinical history included increase in mortality in the flock in 44 cases. Grossly, isolated birds had pale areas in the epicardium and myocardium with or without enlarged heart with ventricular dilatation, ascites, pale areas in the liver and enteritis. Microscopically, there was mild to severe nonsuppurative myocarditis characterized by myocardial degeneration and necrosis accompanied by infiltration of primarily macrophages and lymphocytes and a few plasma cells, heterophils and giant cells. Centrolobular degeneration and necrosis in the liver and lymphoid depletion in the bursa of Fabricius were also commonly seen. Analysis for concentration of vitamin E was performed in liver samples in 5 cases and in 3 follow up submissions and revealed very low concentration of this vitamin. Reovirus was isolated from the heart in 9 cases and in 3 follow up submissions. In one case, this virus was also detected in the heart by Reverse Transcriptase Polymerase Chain Reaction (RT-PCR) and immunohistochemistry, and viral particles resembling reovirus were seen by transmission electron microscopy. The cause of the nonsuppurative myocarditis seen in turkeys is still unknown although these findings suggest reovirus as a possible etiology. Low vitamin E might have contributed to the development of myocarditis.

Association of Gross, Microscopic, and Histopathologic Examination of Enteric Lesions in Young Turkeys

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Enteritis in turkeys is a common complaint of poultry professionals. The aim of this project was to develop a teaching tool utilizing digital photography to associate gross lesions with microscopic and histopathology exam. Both still and video digital images of common enteric lesions were collected with digital images from microscopic evaluations of both wet smears and histopathology. The project demonstrated that not all gross

lesions are associated with microscopic lesions or etiology. This is the first time known to the authors of such a practical application being developed for a training tool.

Pathology and mortality associated with graded levels of melamine and cyanuric acid fed to young broiler chickens

Alex Bermudez, George Rottinghaus, David Ledoux, Lindsay Brand, Rita Dourado, Rafael Murarolli, and Mengshi Lin

Dept. Of Veterinary Pathobiology, College Of Veterinary Medicine; University Of Missouri, Columbia

Recent studies in our laboratory have determined that 2% to 3% inclusion of melamine in the diet of broiler chickens caused significant mortality (16% to 36%) associated with renal pathology. Similar studies with cyanuric acid revealed that 0 to 3% cyanuric acid in a broiler ration did not cause mortality or significant renal pathology. It has been suggested that the ingestion of the combination of melamine (M) and cyanuric acid (CA) results in M-CA crystal formation in the kidneys of cats and dogs leading to acute renal failure and that the two chemicals together are more pathogenic that either one alone. The current study describes the combined effects of graded levels of M (0.5, 1.0 and 1.5%) and CA (0.5, 1, and 1.5%) in the diets of broiler chickens. No treatment related mortality was observed over the 21 day study. Body weight gain was decreased by 1% and 1.5% M alone and by all combinations of M and CA, but was not affected by CA alone. Relative kidney weight was increased by all levels of M alone, and by all combinations of M and CA, but was not affected by CA alone. Histopathology of kidney sections of the high dose combination treatments of M and CA revealed polarizable crystals morphologically similar to those seen in other animals with M-CA renal toxicity. The toxic effects observed in this study were attributed to melamine with the M-CA crystals observed on renal histopathology being the only synergistic M and CA effect.

Anti-viral activity and toxicity of the Alder tree extract against avian influenza virus subtype H9N2

II Hwan Kim, Hyuk Joon Kwon, Sun Hee Cho, Young Jin Ahn, Sun Joong Kim, Seong Ryul Cho, Jeong Chan Ra, Jae Hong Kim

Out of 1,500 plant extract purchased from the Plant Extract Bank in Korea, the crude extract of the alder tree was selected for high anti-viral activity against AIV subtype H9N2. The toxicity was immeasurable in vitro and in vivo. The EC50 of the extract on MDCK cell line ranged from $<50 \text{Å} \cdot /\text{ml}$ to $100 \text{Å} \cdot /\text{ml}$ according to the ways of extraction. The efficacy tests with SPF chickens also revealed that the extract (8mg/chicken/day) reduced virus titre. From the extract several novel single compounds were identified to be effective on AIV in vitro. Therefore, the single compounds may be promising candidates for anti-viral agents.

7692

Biological Characterization of Avian Influenza Viruses Isolated from Wild Birds

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Because of the outbreaks of highly pathogenic avian influenza in Korea, avian influenza (AI) monitoring for the wild birds has been performed since 2006. All 8 segments of total 31 AI viruses isolated from wild birds were sequenced and phylogenetically analyzed. From this study, we found that several AI viruses were genetically clustered with those of domestic poultry. This result may be one of the clues about the interspecies transmission of AI virus. Therefore, biological characterization of these viruses was performed to find the possibility of viral replication and adaptation in the different hosts including SPF chickens.

Determination of Shedding and Survivability of Avian Influenza (AI) Viruses in Experimentally Infected Chickens and Survivability of AI in Feces and Poultry Litter.

Aline R. Reis¹ DVM, Casey W. Ritz² PHD, David E. Stallknecht³ PHD, and Maricarmen García¹ PHD

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Transmission of AI viruses to humans has shown to require direct contact with infected poultry or contaminated poultry environment. To predict the risk of human infections associated with exposure to the poultry environment further knowledge on the survivability of AI viruses in the environment will be necessary. The objective of this study was to determine the shedding of poultry adapted virus A/Ck/CA/431/00(H6N2) and duck isolate A/Mallard/MN/355779/00(H5N2) in chicken feces and to determine the survivability of these viruses in poultry litter and feces. To determine the shedding rate of these viruses broilers and layers were infected by the intravenous and intranasal routes with a dose of $10^{7.2}$ EID₅₀. Feces and cloacal swabs were collected at 3, 7, 11, 12, 13 and 14 days post infection (PI) and virus isolation and titration were performed in 9 to 11 day old chicken embryos. Both type of birds shed H6N2 viruses in the feces and the cloaca up to day 7 PI, with average titers in the feces of $10^{2.8}$ EID₅₀ for birds inoculated intravenously, and $10^{1.5}$ EID₅₀ for intranasaly inoculated birds. At 12 and 14 days PI the

H6N2 virus was recovered only from feces of layers inoculated intravenously at titers of $10^{2.5}$ to $10^{2.7}$ EID₅₀. Birds inoculated intravenously with H5N2 shed virus up to day 7 PI at an average titer of $10^{2.1}$ EID₅₀, while birds inoculated intranasally only shed virus at day 3 PI at an average titer of $10^{1.5}$ EID₅₀. Evaluation of the survivability of H6N2 and H5N2 viruses in feces and in direct contact with broiler litter is been conducted.

Baculovirus expression of neuraminidase (NA) subtype 2 for establishment of specific ELISA for Influenza A virus from avian and swine origin

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The genes encoding neuraminidases from A/GF/MA/14081-11/02(H7N2) and A/TK/Ohio/313053/04(H3N2) were amplified from viral genomic RNA by RT-PCR and cloned. After replacing the natural signal peptide coding sequences with a signal peptide sequence from *Leucania separata* nucleopolyhedrovirus (LsNPV) and addition of an RGS-His-tag, recombinant Baculoviruses were generated using the Bac-to-Bac system (Invitrogen). Expression of recombinant engineered neuraminidase was analyzed by Western blot and IFA using monoclonal anti-6xHis antibodies and sera from infected chickens. Recombinant proteins were purified by immobilized metal affinity chromatography for establishment of N2 specific ELISA.

Expression of an influenza virus hemagglutinin gene in a fowl adenovirus vector

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Using our fowl adenovirus (FAdV-9) based FAdmid we developed a vector for integration of foreign genes into a left end 2.4 kb non-essential region of the genome. Influenza A virus hemagglutinin was cloned under the control of a CMV promoter in leftward and rightward orientations into the 2.4 kb non-essential region of the FAdmid. Expression of HA by both viruses, FAdV-9Â"L-HA-R and FAdV-9Â"L-HA-L, was confirmed by Western immunoblotting using anti HA polyclonal antibodies, hemadsorption and, in cell lysates, hemagglutination. We have thus demonstrated that our FAdmid technology can be used to generate FAdV-9 expressing influenza virus HA.

7696

In Vitro Comparison of the Cytokine Response to Avian Influenza Virus from Peripheral Blood Lymphocytes Isolated From Chickens Differing in Mx631 Gene Polymorphism.

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Following viral infection, the host innate immune response triggers induction of cytokine and interferon genes. Interferon-alpha induction stimulates expression interferonstimulated response elements (ISRE), including the Mx protein. This protein has been shown to confer protection against influenza in mice. In chickens, a single nucleotide polymorphism (SNP) at position 631(Ser-> Asn) has been determined to enhance protection against avian influenza (AI). Here, we evaluated the cytokine response in vitro of PBL's to AI infection. Preliminary analysis indicates increased cytokine and interferon gene induction in Mx-Asn631 lymphocytes compared to Mx-Ser631, which may provide insight into the increased protection observed in chickens with the MX-631Asn allele.

7697

INCREASED PATHOGENICITY AND ALTERED HOST RESPONSES AFTER PASSING A MALLARD H5N2 LPAIV IN IBDV-PRE-EXPOSED CHICKENS .

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After 22 passages of a mallard H5N2 low pathogenic avian influenza virus (LPAIV) in infectious bursal disease virus (IBDV) pre-exposed chickens a LPAIV was obtained that replicated substantially better than the WT mallard virus in both IBDV-pre-exposed and normal chickens. IBDV-pre-exposed chickens showed less than optimal humoral responses to LPAI infection and altered local molecular pathways that eventually lead to an exacerbated disease and death. We suggest that prior IBDV exposure provides a port of entry for avian influenza in an otherwise resistant chicken population.

7698

Interspecies Transmission of Triple Reassortant H3N2 Influenza Viruses Between Swine and Turkeys: Molecular Studies.

Mahesh Khatri, Chang-Won Lee, and Y.M. Saif

Hadi M. Yassine (ph.d. Student)

Triple Reassortant (TR) H3N2 influenza A viruses were first isolated from swine in 1998 in the United States. Similar viruses were then isolated from turkeys in 2003. Previously, we identified viruses (H3N2 TR) that transmit both ways between swine and turkeys, some which transmit one way from swine to turkeys and others that did not transmit either way between the two species. Using reverse genetics we rescued three viruses that represent the above three categories of transmission potential. We performed directed reassortment between the above rescued viruses and tested some of these reassortments for their transmission amongst turkeys. Transmission experiments revealed that the HA gene plays a major role in the transmission of these viruses amongst turkeys. Generated reassortment viruses with replacement in the HA gene will be tested for their transmission from swine to turkeys. Additionally, these viruses will be tested for their in vivo replication in bronchial/tracheal primary cell culture derived from human, swine and turkey origin.

AIV and NDV surveillance in Peru: An update For The 2006-2007-2008 Migratory Season and H7N3-AIV strain Isolation

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Migratory waterfowl are currently considered to be the primary reservoirs for avian influenza viruses. Although intense influenza virus surveillance in wild bird has been occurring in Europe, North America, Asia and Africa, few activities are present to date in South America. Environmental fecal samples were collected from four wetlands along the coast of central Peru from June 2006 to December 2008. Samples were processed for viral isolation in SPF embryonated chicken eggs. Allantoic fluids were evaluated for presence of hemmaglutinatin agents (PMV and AI). A total of 4000 samples were collected during thirty months. Sixteen avian influenza isolates, representing eight different subtypes, and nine paramixovirus isolates were obtain. Between the avian influenza isolates we report the presence of a low pathogenic H7N3 subtype from wild waterfowl (Anas cyanoptera and Anas bahamensis) in Pantanos de Villa.

Live-attenuated H7N2 and H9N2 avian influenza viruses for potential use as *in* ovo vaccines

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In ovo vaccination with live attenuated viruses is widely used in the poultry industry to protect against various infectious diseases. The worldwide outbreaks of low pathogenic and highly pathogenic avian influenza (LPAI and HPAI, respectively) highlight the pressing need for the development of a similar mass vaccination strategy against avian influenza viruses. We have previously shown that a genetically modified avian influenza virus was amenable for in ovo vaccination and provided optimal protection against H5 HPAI viruses. However, in ovo vaccination against other subtypes resulted in poor hatchability and therefore it seemed impractical. In this study, we modified the H7 and H9 hemagglutinin (HA) genes by substituting the cleavage site amino acids with alternative cleavage site sequences. Hatchability was greatly improved even when using large concentrations of the attenuated virus inoculum. In addition, these viruses were not able to transmit to either quail or chickens and provided adequate protection against either H7 or H9 LPAI strains.

Pathogenicity of H5N1 A/Duck/Vietnam/201/05 reassortants in ducks

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In order to understand which viral genes contribute to the high virulence of A/Dk/Vietnam/201/05 H5N1 highly pathogenic avian influenza (HPAI) virus in ducks, we used reverse genetics to generate single-gene reassortant viruses with genes from A/Ck/Indonesia/7/03, a virus that produces mild disease in ducks. Ducks were intranasaly inoculated with the reassortant viruses and mortality, mean death times, viral replication and host gene expression was determined. This study permitted the identification of the genes involved in increased pathogenicity of H5N1 HPAI viruses in ducks.

7702

Resurgence of H5N2 Avian Influenza in Live Bird Markets in the Northeast U.S.

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Live bird markets in the Northeast U.S. have been a source of low pathogenic avian influenza isolates ever since surveillance began in the 80s. The most persitent lineage (H7N2)was successfully eradicated in 2006, but since then a small but increasing number of H5N2 viruses have been isolated from the markets. Whole genome sequence analysis of over 30 H5 avian influenza isolates was conducted to evaluate if these viruses were from a single or multiple introductions. Evidence of multiple introductions was observed, but one lineage appears to be persistent and poses a risk of spread to the integrated poultry industry.

Studies of Phylogeny and Sequence Analysis of H9N2 Avian Influenza Viruses

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It has been shown that H9N2 low pathogenic virus posses a significant threat both to birds and humans. Since its emergence in 1994 in southern China, H9N2 avian influenza subtype virus has rapidly spread throughout the country. In this study, fourteen H9N2 avian influenza viruses (AIV) were isolated from sick chickens in China from 1998 to 2008. The phylogenetic and sequences analyses of the nonstructural (NS) gene of these isolates were determined. The entire ORF sequences of NS1 and NS2 protein were obtained. Homology of these nucleotide sequences and putative amino acid sequences were compared with several classic reference viruses of H9N2. These H9N2 isolates were highly homologous (92.9%-99.9% identity) in their NS gene sequences. These H9N2 isolates belong to A/Chicken/Beijing/1/1994-like group in the Asia birds-swine branch of Allele A of the NS gene on phylogenetic tree. There is a 13 amino acid deletion on the C-terminal of NS1 protein of these isolates, which differentiates the NS gene of H9N2 viruses in mainland China from that of Hong Kong, Korea and Pakistan. According to this study and previous reports of other researchers, NS genes of H9N2 subtype isolates in chickens of China are genetically stable and there is not enough data to support the establishment of other sub lineages in chickens yet.

Susceptibility of chicken T cells to low pathogenic H5 influenza viruses.

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Influenza A virus continues to represent a major cause of morbidity and mortality in human and animal populations. Highly pathogenic avian influenza virus (HPAI) is circulating in wild birds and domestic poultry in Asia, Africa, and Europe, where it has caused more than 240 human deaths since 2003. Low pathogenic avian influenza A strains (LPAI) of the H5 type due to reassortment can transform into HPAI. The pathogenesis of influenza infections has been associated with alteration in the lymphohemopoietic system. In this study we examined the susceptibility of T cells to LPAI H5 influenza viruses. We stimulated peripheral blood mononuclear cells (PBMCs) with phytohemagglutinin (PHA: 5 \hat{A}_{4}^{\prime} g/ml) for 3 days. The T-cell blasts were then infected in vitro with H5N2 and H5N3 viruses. Both viruses induced lysis of T cells and progeny virus was produced in the infected cultures. These data indicate that T cells are target for infection with LPAI H5 viruses.

7705

Viral Adaptation to Host Species-Effect of LP-AIV Passage within a Host

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The objective of the research is to identify and analyze subpopulations of LPAIV that are associated with host adaptation. A U.S. Low Path (LP) avian influenza virus (AIV) H5N1 isolate (A/duck/H5N1/Maryland/2006) was serially passaged 10-times in turkeys and quail to adapt the virus to these poultry species. Infected turkeys displayed clinical signs (conjunctivitis, nasal exudate, and swelling of the sinus) consistent with LPAIV infection more frequently as the passages progressed, while the quail appeared normal throughout the experiment. Moreover, AI antibodies were not detected by AGID in the quail whereas in the turkey, rates of seroconversion ranged from 60-100%.

Amyloid arthropathy associated with various bacteria in Brown Leghorn Chickens

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Amyloidosis is accumulation of homogenous eosinophilic material in various organs causing dysfunction of the organs and the whole body. Brown Leghorn chickens are highly susceptible to amyloidosis of the joints resulting in swollen joints, ataxia and increased mortality. Various bacteria primarily Enterococcus faecalis, and others such as Staphylococcus aureus, Mycoplasma gallisepticum and M. synoviae, etc., have been involved with this condition. Retrospective examination of the data on amyloid arthropathy in chickens in our laboratory revealed numerous similar cases. Information will be presented on the incidence, age, clinical signs, serology, bacteriology and pathology of amyloidosis.

7707

Application of Bacillus subtilis PB6 in Turkey Field Trials

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A Bacillus bacterium was isolated, identified, and registered as Bacillus subtilis PB6. The inhibitory activity of PB6 against isolates of Clostridium perfringens and Clostridium septicum obtained from diagnostic cases was positive using an in vitro method (streak and/or well diffusion assay). Two field trials were also conducted to evaluate the performance characteristics of Large White turkeys after application of PB6 as a direct fed microbial (DFM) in the feed. Livability was improved by 0.5 to 1.0% and feed conversion was improved by 3-6 points. The incidence of cellulitis was reduced, but not eliminated in any of the trials.

Comparative Genome Analysis of *Gallibacterium anatis* Strains Isolated from Peritonitis Lesions and Healthy Birds

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Introduction: Peritonitis is a major problem for the commercial table egg industry. *Gallibacterium* spp., along with *Escherichia coli*, are frequently isolated from the lesions of birds affected with peritonitis. However, *Gallibacterium* are also highly prevalent in the healthy bird's normal flora, and thus its exact mechanisms of disease remain unclear. To better understand the underlying mechanisms of pathogenesis, we utilized a pathogenomics approach involving comparative genome sequencing of *Gallibacterium* isolates.

Methods: Draft genome sequencing was performed on 2 *Gallibacterium* isolates in an effort to identify virulence-associated traits. The prevalence of these traits was examined among a collection of Iowa *Gallibacterium* isolates using multiplex polymerase chain reaction and Southern hybridization. These isolates were also examined for their genetic similarities using pulsed field gel electrophoresis and multilocus sequence typing. Hierarchical clustering was performed to identify any correlations between clonal type and gene profile. A detailed genomic comparison was also performed using comparative genomics tools.

Results: This study demonstrates the potential use of genotyping-based techniques to examine *Gallibacterium* populations from diverse sources. In Iowa commercial layer flocks, certain *Gallibacterium* clonal types appear to dominate, both within and between flocks. Overall, the genomes of *Gallibacterium* strains are highly diverse, and our results suggest that some strains possess virulence factors providing them with an added ability to cause disease.

Detection of Campylobacter jejuni DNA in Broiler Cecal Dropping by Polymerase Chain Reaction

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Campylobacter jejuni is of little clinical concern in the live bird but is a major concern relative to food borne illness in people. Much effort has been expended toward decreasing its prevalence during process while trying to identify the source(s) of infection on the farm. The source of C. jejuni on the farm is a conundrum. Chicks are placed on the farm without culturable C. jejuni only to become infected at 2-3 weeks of age without a readily identifiable source. Previously polymerase chain reaction (PCR) has been used to detect C. jejuni in water. In this study, primers used to amplify a 383 base pair region of the flaA portion of the flagellar gene were validated so investigation into other possible environmental sources of C. jejuni infection could be pursued.

7710 Gallibacterium anatis in broiler breeder flocks in the Republic of Panama: Clinical diagnosis and control measures

Erick N. Calderon, Karina Thomas

Productos Toledano, S.a.

G. anatis (formerly known as Pasteurella haemolytica) is an important infectious agent in poultry that is sometimes misdiagnosed. The clinical presentation may show different forms that are not usually considered in the regular differential diagnosis for reproductive or respiratory outbreaks. The agent has a slow dissemination rate between houses. G. anatis can be isolated in pure culture from the lesions of affected birds, and it can be considered as the primary etiological agent, however, it is often ignored as such because of the reduced information available The agent was isolated and identified by biochemical and enzymatic reactions, and the clinical disease was prevented in other flocks by using an autogenous bacterin that included the identified serovars. A similar product has been used as a commercial bacterin in Mexico.

7711 Large death loss in captive/release Mallard ducks due to Pasteurella multocida

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A flock of approximately 500 captive-reared Mallard ducks that were being raised for release experienced a death loss of over 20 ducks per day. Birds were submitted to the diagnostic laboratory and they had lesions of epicardial and intestinal serosal hemorrhage with prominence of the lyphoid bands. The differential diagnosis included highly pathogenic avian influenza, Newcastle disease, duck viral enteritis and fowl cholera. Histopathology revealed necrosis in multiple tissues with the presence of numerous bacteria. Pasteurella multocida was cultured from liver and spleen.

7712

Retrospective Study of Pasteurella multocida Isolates in Mississippi

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Pasteurella multocida isolates that are isolated from routine diagnostic cases at the Poultry Research and Diagnostic Laboratory are routinely serotyped. This will be a retrospective study of those isolates and their serotypes. Determination of antibacterial susceptibility patterns will be evaluated. The seasonality and geographical characteristics of P. multocida in Mississippi will also be described.

Unusual Pathology Manifested with an Outbreak of *Ornithobacterium rhinotracheale* Associated Meningoencephalitis in Commercial Broiler Chickens

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An outbreak of elevated mortality associated with clinical neurological signs including torticollis and incoordination was observed in 32-day-old mixed-sex, Ross-Ross cross commercial broiler chickens in Southern Mississippi. Histopathology of the brain demonstrated the occurrence of variable pleocelluar or mixed heterophilic and histiolymphocytic meningitis that often was associated with mononuclear perivascular inflammatory cell infiltration or "lymphocytic cuffing" of the brain parenchyma, and sometimes multilfocal gilosis and hemorrhage. Half of the birds submitted to PRDL for evaluation also grossly demonstrated either unilateral or bilateral hypopyon and microscopically extensive enopthalmitis [or panopthalmitis] was observed. In addition, the bone marrow examined for several bird exhibited dramatic granulocytic hyperplasia with elevations in primitive and mature heterophils and as documented by morphometric studies. *Ornithobacterium rhinotracheale* was isolated from about half of the brains cultured, and several of the eyes and ears of the affected birds. The eye and bone marrow pathology appears to represent unusual findings and to our knowledge not previously reported (1-3).

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7714

A Retrospective Study of the Correlation between Chicken Anemia Virus and Avian Leukosis Virus Subgroup J Infection in Broiler and Broiler breeder Chickens

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Chicken anemia virus (CAV) causes acute aplastic anemia and lymphoid atrophy with high morbidity and mortality rates in young susceptible chickens. Avian leukosis virus subgroup J (AVL-J) causes myeloid leukosis in chickens as young as 4 weeks of age. The purpose of this retrospective study was to determine whether there is a correlation between CAV and ALV-J infections. A total of 765 cases or sample sets collected from broiler and broiler breeder chickens between years 1997 to 2006 were tested for CAV and ALV-J by PCR. Based on the PCR results, 399/765 (52%) samples were CAV positive. Of CAV positive samples, 175/399 samples (44%) were ALV-J positive. The total CAV PCR negative samples were 366. Of CAV negative samples, 285/366 samples (78%) were ALV-J positive. Compared with CAV negative chickens, ALV-J infection in CAV-positive chickens was 34% lower, a significant decline (QS=92.0981, P<.0001). This data may indicate that chickens infected with CAV as determined by PCR may have some resistance to ALV-J infection. The virus sequencing analysis, age analysis of the birds infected by CAV and ALV-J, and histopathology summary are in progress and will be presented with these findings.

7715

An 8-year longitudinal survey for the presence of antibodies to chicken infectious anemia virus in two specific-pathogen-free strains of chickens

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Cornell University maintains two genetic lines of specific-pathogen-free chickens in a filtered-air, positive-pressure (FAPP) house. All generations are maintained in the FAPP house without clean-out between generations. Both lines are latently infected with chicken anemia virus. Each flock was monitored for seroconversion 2 to 3 times over a 65-week period. Analysis of the data over an eight-year period included hatch cohort follow up, graphical analysis of prevalence and statistical comparison of seroconversion rates between sires and dams. Sires showed significantly higher rates of seroconversion than dams in one line. Two periods of high seroconversion rates were observed separated by approximately 5 years.

7717 Serologic Survey of Chicken Infectious Anemia in Backyard Poultry in Argentina

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Chicken Infectious Anemia (CIA) was diagnosed for the first time in Argentina in 1991 and the virus was isolated in 1993. Buscaglia (2008) reported at the Argentinean Virology Congress the presence of antibodies for CIA virus (CIAV) antibodies in 91% of backyard hens provided to poor families in a social program called Pro Huerta. Since then I examined sera from backyard chickens unrelated to the Pro Huerta program from different locations in the province of Buenos Aires. Interestingly none of these sera were positive. Possible reasons will be discussed and consequences evaluated.

7718

Development of a Clostridium perfringens alpha toxin (Phospholipase C) antibody ELISA assay using a single serum dilution

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An ELISA was developed that uses a single serum dilution to measure antibody titers to Clostridium perfringens alpha-toxin (Phospholipase C). This assay replaced a previous version that used serial doubling dilutions to titrate antibody titers. The conversion to the new format reduced test costs and materials consumption by 10-fold. The new test format increased efficiency by permitting the testing of 90 samples per plate instead of 9 samples using the previous format.

7719

Development of bacteria-vectored vaccines for necrotic enteritis

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Necrotic enteritis (NE) is a common disease of poultry associated with poor performance and mortality. There is an increasing demand for the development of alternative measures to antibiotics to prevent the incidence of NE. The objective of this study was to develop and evaluate bacterial vectored vaccines for NE. We have constructed attenuated Salmonella and probiotic Bacillus subtilis strains that display immunoprotective epitopes of Clostridium perfringens and Eimeria spp on the surface along with immune-enhancing oligopeptide. Research is currently in progress to evaluate their immune responses and protective efficacies using NE-disease model of chicken. Key words: Necrotic enteritis, vaccine, Salmonella

Mechanisms of intestinal Barrier Failure in subclinical Enteritis

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Novus International, Inc.

One commom consequence of enteritis, regardless of its cause, is a breakdown in the barrier function that normally protects the animal against invasion by commensal and pathogenic gut microbiota. The objective of this review is to describe the structure and function of the intertinal barrier that prevents invasion of the host, to summarize the evidence that loss of barrier function accompanies oxidative stress associated with enteritis, and to discuss possible consequences of the resulting bacterial translocation.

7721

Nicarbazin Anticoccidial Dose Response

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A series of anticoccidial sensitivity tests were designed to determine an anticoccidial dose response to Nicarbazin. The treatments were Nonmedicated, noninfected and infected, Nicarbazin 36, 72, 99 and 125 ppm. Three mixed species field isolates (all contained highly pathogenic levels of E. acervulina, E. maxima, and E. tenella) were examined. An increase in control was observed with each increasing level of Nicarbazin. Approximately a 1 point reduction in lesion scores was observed with Nicarbazine 36 ppm, followed by a 2 point reduction with 72 ppm. Very few lesions were observed with either Nicarbazine at 99 or 125 ppm.

7722

The effect of metam sodium on infectivity of Eimeria oocysts

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Metam sodium (MS, sodium N-methyl dithiocarbamate) is a widely used soil fumigant. In the current study the effect of MS on the infectivity of oocysts of Eimeria tenella, E. acervulina and E. maxima was investigated. Isolated oocysts were exposed for 1 hr to aqueous concentrations of MS ranging from 0 to 1000 ug/ml. Treated oocysts were inoculated into chickens and parameters of coccidiosis infection were compared to chickens receiving equal numbers of untreated oocysts. The results indicate that MS alters infectivity of ooycsts in a dose related manner and may serve as an effective sterilant for coccidia oocysts.

7723

The transcriptome of the coccidian parasite Eimeria maxima the merozoite life stage

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To study the genes expressed from one of seven described species of Eimeria parasites from chickens, we report here the analysis of approximately 2,500 ESTs obtained from the merozoite life stage of E. maxima. More than 60% of these ESTs (~1,700) formed clusters (419) and were categorized according to 20 putative functional categories. The most abundantly categorized ESTs were: unknown function (57%), translation (14.8%), cytoskeletal (6.7%), glycolysis (3.1%), metabolism (2.8%), surface antigen (2.5%), protein folding (2.3%), transport (2.3%), and microneme (2.3%). These data are contrasted with previous transcriptome analyses of E. tenella and E. acervulina.

7724

Which Eimeria Species Most Affects the Production of Necrotic Enteritis in Broiler Chickens

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Coccidiosis damage to the intestinal mucosa leads to proliferation of Clostridium perfringens and production of Necrotic Enteritis (NE). The objective of this study was to examine association of various species of Eimeria (E. acervulina, E. maxima, E. tenella, and E. praecox) to production of NE. Birds infected with E. maxima were more significantly affected by NE than birds challenged with the other species. Some NE mortality occurred with the E. acervulina challenge. There was no NE mortality with the E. praecox/CP challenge but there was a reduction of performance. The least affected birds were those inoculated with E .tenella.

7725

Relationship between Active Infection with Cryptosporidium baileyi and anti-C.baileyi Antibody Responses

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It is well known that serology using anti-Cryptosporidium IgG or total immunoglobulins (Igs) can be used to provide information about the prevalence of a prior exposure to the parasite; but is of no value in determining the period of infection and to distinguish a recent infection from an old one. We noticed during a previous study a correlation between the chronic shedding of C.baileyi oocysts in the feces and the IgA response in SPF chickens experimentally co-infected with C.baileyi and HPRS16 strain of MDV (Abbassi et al, Avian Diseases 2000). This current study is carried out to investigate the correlation between the active infection with C.baileyi evaluated by the detection of oocysts in feces and the antibody response using all anti-C.baileyi immunoglobulin isotypes (IgG, IgM, IgA) and total Igs. We used SPF chicks experimentally infected with C.baileyi alone, or in combination with IBDV. The feces were examined every other day and the serology performed by ELISA every week during a period of over 10 weeks.

7726

APEC Virulence Plasmids: Multi-Purpose Contributors to Disease

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Most APEC harbor large virulence plasmids. These plasmids are modular in nature, often containing a pathogenicity island, a region of unknown function, and a transfer region. Some also harbor islands of genes encoding multidrug resistance. Here, we summarize the disease-associated phenotypes encoded by APEC's virulence plasmids.

Among these are multidrug resistance, adherence, invasiveness, ability to grow in human urine, low iron, and avian and human serum, and ability to cause avian colibacillosis and murine urinary tract infection, sepsis, and meningitis. We will also provide evidence that APEC containing such plasmids are emergent and will discuss the implications of this emergence.

Does the Use of Aresenic-containing Compounds Promote the Selection of Multidrug-Resistant APEC

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Arsenic-containing compounds are commonly used in poultry feeds to control intestinal parasites and promote growth of broilers. Plasmid-mediated resistance to arsenate and arsenite has been recognized in both Gram negative and positive organisms. In this study, the prevalence and transmissibility of arsenic resistance was assessed in APEC isolates of poultry. Additional work assessed the genomic location of arsenic-resistance genes and the potential role of arsenic-containing compounds in the selection of multi-drug resistant APEC.

Predicting the virulence of Avian Pathogenic *E. coli* using serogroup, phylogenetic group and MLST

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Highly virulent avian pathogenic *E. coli* (APEC) cause significant economic losses within the poultry industry. Some APEC are more virulent than others, but it is not known why. We have characterized a group of APEC isolates by virulence genotype, phylogenetic group, O serogroup, pulsed field gel electrophoresis profiles and multi locus sequence type. Whilst the majority of these isolates were from the B2 phylogenetic group, they were found to belong to a range of O serogroups. The virulence of a subgroup of these isolates was determined using the embryo lethality assay. The pathogenicity of these isolates varies widely although some correlation with specific types was observed.

Use of GFP- and RFP-Labeled APEC to Evaluate Adherence

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Adherence is an important step in establishment of bacterial infection. Thus, adherence assays are often used to evaluate the potential virulence of bacterial strains. Here, the ability of different avian pathogenic Escherichia coli (APEC) to adhere to cultured cells was evaluated using competition assays. To facilitate identification of 'competing' strains, a novel bacterial labeling procedure was used. APEC of interest were labeled with green or red fluorescent protein by introduction of the gfp or rfp gene into their chromosomes using a simple and efficient, one-step procedure. Differences in adherence between the two competing APEC strains that were differentially labeled were readily apparent in competition assays.

7730

Holistic treatment of immunosuppressed broilers coinfected with Mycoplasma gallisepticum and Infectious Bronchitis Virus

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The purpose of this study is to evaluate the pathology and performance following an essential oil treatment of immunosuppressed broilers, challenged with Mycoplasma gallisepticum (MG) at 5 days of age, live B1 and LaSota BewCastle disease vaccine strains at 10 and 16 d of age respectively, Infectious Bursal Disease Virus (IBDV) at 21 days of age, and with Infectious Brochitis Virus (IBV) at 26 days of age. The pathological (weight gain, feed conversion and immunities) will be assessed, and compared statistically to controls within a complete randomized design

The Draft Turkey Genome Sequence: Implications for Immunity, Disease and Overall Health

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As the Turkey Genome Sequencing Consortium, we have employed next generation sequencing technology to determine the genome of the domesticated turkey, Meleagris gallopavo. Economically, US turkey production and consumption rank fourth behind beef, chicken, and pork nationally. Also, the turkey serves as a model organism for a number of human metabolic and medical diseases. The source DNA for sequencing was isolated from a female turkey (NT-WF06-2002-E0010, referred to as "Nici" (Nicholas inbred)) of an inbred sub-line, originally derived from a commercially significant breeding line. Roche/454 GS-Titanium sequencing at Virginia Tech has already produced more than 5x random and paired-end genome coverage (> 8 billion bases sequenced). The latest sequence assembly contains ~880 million base pairs in > 428,000 large contigs, with average size of ~2 kb. This initial sequencing phase is paving the way for the development of many more genomic resources expected out of the fully sequenced genome. Preliminary analysis of turkey immune related genes indicates relatively high sequence homology to those of the chicken. For example, Tolllike receptor (TLR)-4 and interleukin (IL)-10, showed > 90% sequence homology to their chicken orthologs. Further analyses will undoubtedly uncover species-specific genetic markers that could be related to disease resistance and turkey immune response. Those markers would also be employed in the development of SNP panels for genomebased selection and improvement, as well as comparative genomics in poultry and other avian species. Such discoveries may also help direct our focus to enhance the turkey immune competence and develop new and/or more effective disease prevention strategies.

7732

Characterization of infectious bronchitis virus field strains isolated in 2008 from poultry farms in Mississippi.

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Field strains of infectious bronchitis virus were isolated from tracheas and cecal tonsils collected from poultry farms in Mississippi. Most of the isolates were isolated within the first three passages in specific pathogen free embryonated eggs. Forty eight hours post inoculation, most of the isolates hemoagglutinated chicken red blood cells after treatment with neuraminidase and induced characteristic embryonic lesions as observed six days after the inoculation. After the nucleotide, amino acid sequence and phylogenetic analysis, it was determined that most isolates exhibited genetic characteristics of the Arkansas DPI strain.

Comparison of the Pathogenicity of Two Infectious Bronchitis Strains, Qu16 and Qu_MV, Isolated from Quebec, Canada

Leni Corrand, Mona Morin, Carl Gagon, Amer Silim, Davor Ojkic, and Jean-Pierre Vaillancourt

University Of Montreal

Two viral strains of infectious bronchitis virus- Qu16 and Qu_MV- were isolated from respiratory affected broilers farms in Quebec between 1996 and 1999. No studies had been done about their pathogenicity until now. In order to evaluate and compare pathogenic effects, SPF Leghorns were infected at 14 days of age with the Qu16 or Qu_MV strain. Serological monitoring, clinical signs, macroscopic (airsacs) and microscopic (trachea) lesions were graded. Significant tracheal lesions were detected in infected birds, although no clinical signs, nor airsac lesions were observed. Considering that these strains were considered highly pathogenic in the field, these results suggest that environmental conditions during rearing play a paramount role in the expression of the disease with these strains.

7734

Evaluation of safety and protective efficacy of bivalent live nephropathogenic infectious brochitis virus K2 vaccine combined with nonpathogenic Newcastle virus DSB-HP strain (Live K2-ND bivalent vaccine) by coarse spray iin 1-day-old commercial broilers

Jeong-Yong Park, Hyun-Jeong Lee, Ha-Na Yoon, Seung-Hwan Jung, Tae-Hyun Lim, Dong-Hoon Lee, Youn-Jeong Lee, Won Hur, Joong-Bok Lee, Seung-Yong Park, In-Soo Choi, Chang-Seon Song

Avian Disease Laboratory, College Of Veterinary Medicine, Konkuk University

In previous study, we developed a live attenuated infectious bronchitis virus (IBV) vaccine, named as the K2, derived from Korean nephropathogenic strain and confirmed its safety and efficacy in SPF chickens. In this study, we performed evaluation of safety and protective efficacy of live K2-ND bivalent vaccineTM in commercial broilers using cabinet and potable coarse sprayer for practical application in field condition. For evaluating vaccine safety, tracheal ciliostasis, histological lesion, body weight gain, livability and performance index was examined. For evaluating vaccine efficacy, mortality after vvNDV challenge and virus reisolation in trachea and kidney after QX-like nephropathogenic IBV challenge was evaluated. Further, interference effect between IBV and NDV was examined.

Evaluation of the Effectiveness of Two Infectious Bronchitis Vaccine Programs for Preventing Disease Caused by the IBV Qu_MV Field Isolate from Quebec

Leni Corrand, Mona Morin, Carl Gagon, Davor Ojkic, and Jean-Pierre Vaillancourt University Of Montreal

A new viral strain of infectious bronchitis virus - Qu_MV - has appeared in Quebec broiler farms in conjunction with a high prevalence of airsacculitis at slaughter. Recently, this strain has also been isolated in other provinces. In order to evaluate the efficacy of commercial vaccines against an experimental Qu_MV infection, two vaccine programs were tested. SPF Leghorns were vaccinated at 1 and 14 days of age with Mass/Conn or Ark vaccines, then challenged at 31 days. Serological monitoring, clinical signs, macroscopic (airsacs) and microscopic (trachea) lesions were graded. A significant reduction in tracheal lesions was noted in vaccinated birds compared to infected non vaccinated controls; mainly with the Ark strain.

7736

Investigations into the genotype of the S1 gene and the tissue distribution of a unique IBV strain present in the United States

Peter R Woolcock, Carol J Cardona, Mingke Yu, Richard P Chin, Rocio Crespo H L Shivaprasad, Bruce R Charlton, Hailu Kinde Cahfs, Uc Davis

An infectious bronchitis isolate, designated PA1220/98 was first reported in 1998 from 20-week-old layer pullets in Pennsylvania. It has also been detected in Iowa and Ontario, Canada. In California 12 isolations have been made from both meat and layer chickens, first in 1998, then ten times between 1999 and 2002. It had not been detected again in California until September 2008. PA1220/98 is unique in that it has less than a 50% relationship to any other IBV sequences logged in GenBank. Results of investigations into the genotype of the S1 gene and immunohistochemistry of infected tissues will be reported.

7737 Pathological and molecular diversity of Brazilian strains of IBV: multiple lineages can cause multiple diseases

Laura Y. B. Villarreal, Delair A. Bolis, Thaisa L. Sandri and Paulo E. Brandeo Intervet Schering Plough Animal Health - Brazil

Though variant types of IBV exist in Brazil, vaccination is based solely on the Massachusetts serotype. Samples were collected from grandparents and broilers showing different manifestations. Six IBV strains detected grouped as a Brazilian exclusive genotype, with no disease-specific lineage, with one 4/91 strain. The Brazilian genotype can be associated with all kind of IB manifestations, what could allow a faster and more disseminated transmission of IBV with high evolution rates due to a lower selective pressure. For an efficient control of IBV, it's necessary to include a vaccination schedule based on protectotypes close to the genotype described herein.

7738

Unusual lesions of nephritis associated with Infectious Bronchitis virus in Chickens

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Infectious Bronchitis (IB) caused by coronavirus is a highly contagious disease of chickens characterized by respiratory signs, airsacculitis and increased condemnations in young chickens and decreased egg production and poor egg quality in layers. There are many serotypes of IB virus including Cal 99 which causes respiratory disease in chickens. IBV Cal 99 was isolated from the kidneys of several five to ten week-old game chickens with a history of respiratory signs and increased mortality and lesions of pneumonia and severe nephritis. Immunohistochemistry for IBV revealed antigen not only in the kidneys but also in the respiratory system.

Update on the Molecular Epidemiology of Infectious Bursal Disease Virus in Latin America

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Infectious Bursal Disease virus (IBDV) continues to be present in the poultry industry. In Latin America a very virulent form of IBDV virus is present in South America. Due to high economic losses from this condition, a survey was conducted in Mexico and Central America in order to monitor the presence of the IBDV. Chickens from farms with a history of poor performance and high secondary problems were selected for a bursal health survey. This survey consisted in Imaging Processing, PCR and sequencing in some cases. Results of the genetic diversity of this virus along this geographic area are reported

7740

Identification of infectious bursal disease viruses in broiler chickens at processing plants.

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Maternal immunity can delay infectious bursal disease virus (IBDV) infection until broilers are several weeks of age. We have observed gross and microscopic lesions in the bursa and identified IBDV in broilers 3- 4 weeks of age. Late field virus infections suggest IBDV may still be present when the birds are processed. We observed IBDV in the bursa tissue of broilers at processing plants. The export of U.S. broiler products to some foreign countries has been restricted because of the potential for IBDV contamination. Until now, the presence of IBDV in broilers at the processing plant was unknown.

Recombinant Sub-Unit VP2 vaccine protects against challenge with US isolates of standard and variant Infectious Bursal Disease strains

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Recombinant sub-unit VP2 vaccine was developed in Israel from a very virulent Infectious Bursal Disease Virus (IBDV). The vaccine protected against challenge with three different IBDV strains prevalent in the US: 2512, Delaware E and AL-2. The degree of protection and ELISA antibodies titers was similar to those induced by Bursal Tissue Origin vaccine that is used in the US. Induced antibodies neutralize both Standard and Variant strains of IBDV. The VP2 subunit vaccine prevents IBDV infection of chickens and eliminate the need to use bursal tissue for the preparation of killed IBD vaccines.

7742

Specific humoral immunity elicited by DNA encoding infectious bursal disease virus large segment gene and avian influenza virus hemagglutinin gene

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The present study was carried out to determine whether infectious bursal disease virus (IBDV) large segment gene-based DNA fused with avian influenza virus (AIV) hemagglutinin (HA) gene could trigger immune response to both IBDV and AIV. Hemagglutinin gene of AIV was amplified from cDNA of A/turkey/WI/68 (H5N9) strain by PCR and inserted into IBDV VP243 gene in a pcDNA vector carrying IBDV VP243 gene. One-day-old specific pathogen free (SPF) chickens were intramuscularly injected with vector DNA or plasmid DNA carrying VP243, H5, or VP243-H5 gene weekly for three times, followed by a two-week interval for the fourth injection. The virus neutralization (VN) titers to IBDV were significantly higher (p<0.05) in chickens inoculated with VP243/pcDNA than those with VP243-H5/pcDNA 2 to 6 weeks after the first inoculation. The hemagglutination inhibition (HI) titers to AIV were significantly higher (p<0.05) in chickens inoculated with H5/pcDNA than those with VP243-H5/pcDNA 2 to 6 weeks after the first inoculation. The results indicated that IBDV large segment gene-based DNA fused with AIV HA gene in DNA vaccination can elicit specific humoral immune response to both IBDV and AIV.

7743 The Isoleucine at position 451 is not critical for pep46 activity in Infectious Bursal Disease Viruses.

Daral J. Jackwood and Susan E. Sommer-Wagner

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During replication, the IBDV polyprotein (pVP2-VP4-VP3) is cleaved to yield pVP2 which is further processed to the mature VP2 capsid protein. One peptide that is cleaved off pVP2 during this maturation process is pep46; a 46 amino acid peptide responsible for the formation of pores in cell endosomal membranes. Galloux et al. (2007) showed that substitution of I451 with Alanine blocked IBDV replication. Our results demonstrate that the amino acid Leucine at position 451 does not inhibit replication of the virus. Our results further show that L451 seems to be restricted to only very virulent strains of IBDV.

7744

Characterization of infectious laryngotracheitis virus involving in a severe outbreak in Peruvian commercial layers flocks

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The infectious laryngotracheitis (ILT) is highly contagious respiratory disease of chickens, which leads to significant economic losses. A severe outbreak of respiratory disease occurred amongst commercial egg-laying flocks from the Chincha city, Peru, in October 2008. The clinical signs and lesions observed, including nasal discharge, marked dyspnea, expectoration of blood, stained mucus, and increased mortality, were compatible with ILT. The ILT virus was detected in trachea and trigeminal ganglia samples by PCR. The sequences analysis of two regions of the ICP4 gene and the PCR/RFLP analysis showed that the severe outbreak was caused by a non-vaccine and virulent strain.

7745 Developing a Practical ILT Control Program for Non-Commercial Poultry Flocks

Nathaniel L. Tablante, Jennifer Timmons, Connie Hodgson

Virginia-maryland Regional College Of Veterinary Medicine, University Of Maryland College Park

Infectious Laryngotracheitis (ILT) is an acute viral respiratory disease of chickens that causes severe economic losses in commercial poultry operations due to high mortality and morbidity. Unlike commercial poultry operations, non-commercial (small-scale or backyard) poultry operations usually have poor biosecurity and are often multi-age and multi-species. These flocks may serve as reservoirs for ILT and may pose a threat to commercial poultry operations. ILT outbreaks in non-commercial flocks require a comprehensive program that includes regular testing, isolation and selective culling of infected chickens, vaccination of susceptible birds with tissue culture or recombinant vaccine, increased biosecurity, and education.

7746

Differentiation of vaccine strains and field isolates of infectious laryngotracheitis virus by sequence analysis of ICP4 gene

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Two sets of primers were designed to amplify two fragments of the ICP4 gene of infectious laringotracheitis virus (ILTV). Thirty ILTV field isolates, one TCO vaccine strain and two CEO vaccine strains were analyzed. The primers amplified two fragments of 688 and 631 bp. The aminoacid analysis permitted to differentiate vaccine strains from field isolates when PCR products were submitted to sequencing. It was possible to distinguish between the TCO and CEO vaccine strains. In addition, the nucleotide analysis permitted to discriminate between the two CEO strains included in this study. The sequencing results were confirmed by RFLP of PCR products from four genes.

Heterologous Expression of Glycoproteins B, I and J of Infectious Laryngotracheitis Virus

Alice Mundt, PhD, and Maricarmen Garcia, PhD

Poultry Diagnostic and Research Center, Department of Population Health, University of Georgia

Infectious laryngotracheitis virus (ILTV) is one of the major pathogens affecting commercial poultry production. Contrary to other herpes viruses no genetically engineered vaccine strain is currently in use. As a prerequisite for the engineering of mutants we have expressed glycoproteins B, I and J of a recent US isolate of ILTV transiently in LMH-cells and from a Baculovirus vector in Sf-9 cells. Sera from vaccinated or infected chickens were analyzed by IFA on transfected LMH-cells. Baculovirus-expressed gl and gJ were purified from infected Sf-9 cells by immobilized metal affinity chromatography and used to produce hyperimmune sera in chickens.

7748

A Novel and Practical Method Utilizing the Latin Square as a Basis for Statistical Analysis of Variance Between Two Experimental Treatments in the Hatchery

Mark A. Dekich, Robert W. Keirs, E. David Peebles Scientific Director Avitech LLC

The U.S. Poultry Industry is financially challenged with grain and energy cost. The industry seeks new products, technology, or equipment that potentially improves efficiency and lowers cost. Chick cost has exceeded \$.25/chick and is a major part of live production cost. Increasing hatch by 1% can be significant.

,Evaluating new products, technologies, or equipment to assesse .5-2%consistant improvement is difficult due to biological diversity of the egg pack and hatching system variance.

,A novel approach for statistical experiments utilizing the Latin Square has been practically adapted to the hatchery for simple evaluation of variance.

A novel chlorination technique to improve the safety, efficacy, and handling of chlorine in poultry watering systems.

Douglas A. Anderson and Douglas Pennock.

Georgia Poultry Laboratory Network

To resolve a waterborne bordetellosis problem, a uniquely engineered ORP system was developed to deliver effective levels of active chlorine throughout the water delivery system. It proved to be successful, but had the drawback of excessive storage quantities of liquid chemicals that could present a hazardous situation. To circumvent this problem a novel combination of dry chemicals were tested in the current system and assessed for efficacy, safety, and improved ease of handling. The novel chlorination system proved to be equally efficacious, safe, and was a vast improvement over the handling and cost of the liquid chemicals.

7750

BIOSECURITY RISK ASSESSMENT OF LIVE BIRD MARKETS IN BANGLADESH AND WEST AFRICA

Jarra F. Jagne, DVM, DACPV, Mick Fulton PhD, DVM, DACPV, Linda Spink MSW, David Tardif-Douglin, PhD

DAI- STOP AI Project; 7600 Wisconsin Ave. Suite 200 Bethesda, Md 2081Highly pathogenic avian influenza (HPAI) H5N1 first received widespread recognition following a 1997 outbreak in poultry in Hong Kong SAR with subsequent spread of the virus to humans. During that outbreak, 18 people were infected and a total of 6 persons died. Live bird markets (LBMs) were implicated in the infections. Live bird markets or wet markets as they are also known; sell poultry to the public as live or freshly slaughtered poultry. Most of these markets operate in urban settings in many parts of the world where populations prefer to eat fresh rather than frozen meat. Since the Hong Kong outbreak, surveillance studies in many Asian countries have shown that several avian influenza subtypes including H5N1 exist in LBMs. Live bird markets play a big role in harboring the H5N1 virus and the potential for dissemination back into the community.

Day of Hatch vs Day after Hatch Placement of Turkey Poults: the influence of environment on turkey health and performance

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Rapid post-hatch development of the gastrointestinal tract, influenced by immediate post-hatch feed, is necessary for the overall health and growth of chicks and poults. The effect of day of placement and environment on turkey performance was investigated in poults placed day of (d0), and day after (d1) hatch at both the NCSU Teaching Animal Unit and with a commercial grower (sister flock, SF). Bird weight, disease, and mortalities were monitored throughout the brooding and grow-out periods. D1 birds in both groups weighed less and had higher mortality than d0 birds; these differences were accentuated in the SF and are attributable to increased GIT disease.

7752

Effect of Fresh Dietary Garlic Powder on Some of Serum Biochemical Parameters in Broiler Chicks

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Two hundred and eighty, two-day-old, Ross chicks were randomly divided into groups A, B (52 each), C and D (88 each). The chicks in groups A & B were fed control diet, but those in groups C & D received diet supplemented with 1 and 3% garlic powder, respectively. Also, to evaluate the effect of consumption period of garlic on immune response, half of the chicks in groups C & D were separated after 2nd bleeding as groups E and F, and were fed control diet until the end of the experiment. All groups except A were inoculated against ND, AI and IBD. Fifteen blood samples were taken from each of the groups at 21, 32 and 42 days of age and 5 at 2 d. The sera were assayed for Pre-ALB, ALB, ±-1, ±-2, Â² and Â³-globulins using bi-dimensional electrophoresis. The results showed that the above-mentioned parameters, except Â³globulins, were not affected by the diet (P>0.05). Group B, compared with group A, had higher values in all parameters, but \hat{A}^3 -globulins, at 42 d (p<0.05). Whereas, they were different in Â³-globulins from the age of 32 d onwards. Furthermore, the amount of Â³globulins had a dose-dependent increase in treated groups from 21 d onwards. Also, the removal of garlic from the diet of group E resulted in a significant decrease in Â³globulins, in relation to group C, at 42 d. It is concluded that garlic powder has the potential to increase serum Â³-globulins of broiler chicks. Keywords: garlic, electrophoresis, biochemical parameters, broiler chicken

Evaluation of Biosecurity and Monitoring System in the Breeder Farm Specialized for the Production of Human Influenza Vaccine

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The production of embryonated eggs used for the human influenza vaccine were needed high level of biosecurity. Therefore, most countries including Korea imported the vaccines as end-product form because of economic concern. However, recent outbreaks of HPAI in human gave rise to the necessity of their own production system to prepare the pandemic in the future. Recently, the HS breeder farm specialized for the human vaccine was constructed in Korea. We regularly monitored the biosecurity of the farm using microbiological techniques especially to detect infectious agents. We are going to present detailed data collected from the HS farm at the meeting.

7754

The Identification of Fungi Collected from Commercial Poultry Houses using an Automated rep-PCR System.

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In poultry, fungi are normally ignored unless clinical signs are reported, while the role of fungi in the laying house environment is largely unknown. The goal of the present study was to monitor fungi in a commercial poultry house conditions. Broilers and layers were evaluated for fungi and yeast recovered from cecae at placement, and every two weeks-of-age. Fungal samples were isolated from the cecae and characterized using a rep-PCR methodology to track fungal genera changes during production. The results from the present study will provide a better understanding of the ecology of fungi and yeast in commercial poultry facilities.

Use of the I-Stat Serum Chemistry Analyzer for Evaluation of Poult Flip-Over Syndrome

Michael P. Martin, Rosemary Stoertz, Summer Russell, David Rives, Oscar Fletcher, and H. John Barnes

NCSU, College Of Veterinary Medicine, Poultry Health Management Team

The I-Stat serum analyzer has been used in our previous clinical evaluations of broiler breeders and turkey poults and has been established as a useful diagnostic tool for poultry flocks. Previous studies presented last year showed how the instrumentation may be useful in evaluations of poult quality, health, and comparative management techniques Poult flip-over can be a source of significant mortality in turkeys shortly after placement. A serological investigation following a flock from the hatchery to farm was performed and blood chemistries were performed. Poults without access to feed or water were compared to 'Flip-Over' poults and normal poults.

7756

Detection of ts-11 vaccine in the presence of wild-type Mycoplasma gallisepticum using a unique restriction enzyme site

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Current practices employed to diagnose *Mycoplasma gallisepticum*, (MG) include serological testing, PCR, real-time PCR, and isolation by culture methods. None of these methods differentiate between the ts-11 vaccine strain and field isolates. This study has defined a unique restriction enzyme site (Taq I) within the MGA0319 gene of ts-11. The site allows differentiation of ts-11 from other MG strains by restriction enzyme digest using the Taq I enzyme. Results show specificity, sensitivity, and the ability to detect ts-11 even in the presence of wild-type MG strains.

EVALUATION OF A NEW MULTIPLEX REAL-TIME PCR ASSAY FOR THE DETECTION OF MYCOPLASMA GALLISEPTICUM AND MYCOPLASMA SYNOVIAE IN POULTRY

Scott A. Callison, Ph.D.

GTCAllison, LLC

Two of the most important pathogenic avian mycoplasmas to the commercial poultry industry are *Mycoplasma gallisepticum* (MG) and *Mycoplasma synoviae* (MS). Infections cause poor bird performance leading to economic loss. Since MG and MS can be vertically and horizontally transmitted, the basic control strategy for these infections is to maintain mycoplasma free flocks. To that end, different diagnostic assays are employed to monitor the mycoplasma status of flocks. Herein, a new multiplex real-time PCR assay for the detection of MG and MS in poultry was evaluated with respect to specificity, limit of detection, reproducibility, and correlation with other diagnostic methods.

Serological results with an improved Serum Plate Agglutination antigen for Mycoplasma synoviae

Gloria Avellaneda and Theodore Girshick

Charles River Laboratories Avian Products And Services (SPAFAS)

Serologic testing of *Mycoplasma synoviae* infection in commercial poultry is often accomplished by serum plate agglutination (SPA), hemagglutination inhibition (HI), and/or enzyme-linked immunosorbent assay (ELISA). Immune response to *M. synoviae* can be detected sooner by the SPA test than by HI or ELISA, making it a qualitative test of choice for screening large numbers of samples from chickens and turkeys. However, the specificity of SPA for a rapid diagnosis of *M. synoviae* has shown to be less than adequate due to the high incidence of false positives. The objective of the present work was to determine the sensitivity and specificity of an improved antigen for the SPA test when compared with a commercial *M. synoviae* ELISA kit. After infecting groups of chickens with *M. synoviae*, serum samples were collected periodically and tested for antibodies against *M. synoviae* to determine the onset of detected immunity with both systems. Results obtained will be presented and differences will be discussed.

A Highly Virulent Strain of Newcastle Disease Virus Elicits a Strong Innate Immune Response and Nitric Oxide Production In Vivo

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The avian immune response to Newcastle Disease Virus (NDV) and the contribution of this response to disease are not well understood. In this study, the transcriptional host response of chickens to a virulent NDV outbreak strain (California 2002) was characterized in vivo. Using microarray, a strong host response was observed in spleens at early times post-infection with the induction of numerous innate anti-viral and pro-inflammatory response genes. The robust host response to NDV shown here, in conjunction with severe pathological damage observed, suggest that the host response itself may significantly contribute to the rapid mortality of this outbreak strain.

7760

An experimental egg transmission of Newcastle disease virus

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Because some ND cases in Korea were found in chickens less than 10 days old, vertical transmission of NDV was suspected. Although the presence of NDV in the reproductive organs following infection has been reported, the evidence of egg transmission of NDV is lacking. Because velogenic NDVs usually cause death of chicken embryo within 60 hrs after infection, it was suggested that egg transmission of NDV could not be possible. We thought that very low titers of NDV in the egg, especially in commercial chicken eggs with high titers of maternal antibody to NDV, might not cause embryo death, so egg transmission of NDV could be possible. In this study, the possibility of the egg transmission of NDV was studied through the inoculation of low titer of NDV in commercial chicken embryos. In experiment of egg transmission of NDV, a few chicken embryos inoculated with low titer of NDV can hatch and contain NDV after hatching. These results suggest that NDV can be transmitted through the eggs infected with a low virus titer, which is not enough to induce the death of embryos during incubation.

Characterization of Paramyxovirus Isolated from Migratory Wild Birds

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Avian paramyxovirus type I has been isolated from numerous hosts including chickens and wild birds throughout the world. Three strains, neutralized by Newcastle disease (ND) antibodies, were isolated from wild birds in Korea and biologically characterized to evaluate the possibility as a live vaccine against NDV. These isolates were classified into geno-group I of NDV based on the sequence of fusion protein gene. In preliminary results using SPF chickens, these strains showed significant seroconversion after inoculation, and 96% protection against the challenge with viscerotropic velogenic NDV. The detailed data for safety and efficacy of the live vaccine using these isolates will be discussed at the meeting.

7762

Immunogenecity and safety of thermostable Newcastle disease virus vaccine candidate, K17 strain, originated from wild bird

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We recently isolated novel Newcastle disease virus (NDV), named as the K17, in feces collected from wild bird habitat in Korea. In this study, we analyzed the biological and molecular characteristics of the NDV K17 and examined the immunogenicity and safety in SPF chicks to evaluate the potential vaccine candidate strain for recent epizootic viscerotopic velogenic NDV strain type VII. The NDV K17 was shown to mean-death-time of 144 hours and intracerebral-pathogenecity-index of 0.2 and it was stable at 56 for 1 hour. The molecular study showed that NDV K17 was included in the cluster of NDV genotype I and possessed a motif of lentogenic strain. The efficacy and safety of NDV K17 was compared with NDV V4 vaccine strain. The result of this experiment will be discussed.

Presence of apoptosis, as determined by immunohistochemistry, in lymphoid tissues of chickens infected with strains of Newcastle disease virus of varying virulence

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Virus-induced apoptosis is an anti-viral mechanism of host defense utilized by eukaryotic cells to minimize viral replication and to reduce damage caused by infection while clearing the invading pathogen. Thus, we have analyzed the expression of caspases, key enzymes in the induction of apoptosis. Formalin-fixed, paraffin embedded sections from tissues of chickens infected with 8 NDV strains of different virulence were evaluated via immunohistochemistry (IHC) to detect the active isoforms of various caspases in the main lymphoid organs. The number of positive cells was numerically assessed and the differences in expression were statistically evaluated

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Development of a selective MPN assay for the enumeration of Salmonella in poultry carcass rinses

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Historically the performance of Most-probable Number assessments have been used to enumerate whole carcass rinses for bacterial load. We wished to limit that assessment to Salmonella only by using the MPN format but by making two changes. The first modification was the use of Tetrathionate Brilliant green with lodine as the primary enrichment followed by a secondary enrichment in Rappaport-Vassiliadis broth. Second was the application of a microtechnique using 2ml deep-well microplates instead of the cumbersome tube approach. This greatly facilitated the management of large numbers of carcass rinse samples

Identification of Salmonella in backyard chicks and characterization of the salmonella isolates by serotyping and multiplex PCR

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For estimation of the prevalence of Salmonella, Samples of visceral organs including gastrointestinal tract, liver, and heart were taken from 500 backyard chicks (at early ages) and subjected to culture according to standard methods. The gastrointestinal tract, liver, and heart samples identified to be contaminated with salmonella in 62 (12.4%), 77 (15.4%), and 71 (14.2%) cases respectively. The results indicated that of all 210 isolated samples, the serovars were S. typhimurium (93 samples), S. enteritidis (112 samples), and S. gallinarum (5 samples). By using multiplex PCR, all S. typhimurium serovars were positive for rfbJ, fljB, fliC, and invA genes. For S. enteritidis serovars, they were positive to spv, and sefA genes. With crp-1, crp-2 and IS10as2 primers, a fragment was produced inside the crp gene of S. gallinarum. Keywords: Salmonella, prevalence, serotype, multiplex PCR

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Protection from Salmonella Enteritidis (SE) contamination in eggs derived from hens vaccinated with various SE PROGRAMS

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Eggs were collected from commercial egg laying hens at 25 and 45 weeks of age to compare Salmonella enteritidis (SE) vaccination programs involving live, and/or inactivated SE vaccines, containing inactivated Newcastle disease virus (NDV) and infectious bronchitis virus (IBV). Egg pools were prepared and inoculated with an SE culture to ascertain whether SE growth was suppressed by the antibodies in the egg contents derived from the vaccinated hens. The results regarding the reduction in SE growth will be presented.

Reduction of Salmonella egg contamination by a commercial bivalent bacterin after intravenous challenge of layer chickens at the onset of lay

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A commercial bivalent Salmonella bacterin was evaluated under the semi-field condition. On-farm vaccinated (85-day old) and unvaccinated birds were challenged intravenously with Salmonella serovars Enteritidis and Typhimurium at the onset of egg laying (150-day old). As a result, the vaccine could alleviate the decreased egg production and increased egg contamination. At necropsy, the shrunken ovaries and oviducts were observed only in unvaccinated hens that had stopped egg production after challenge. From these results the vaccine can decrease egg contamination even after challenge at the onset of egg laying, which is associated with a high susceptibility to the infection.

RE-EVALUATION OF THE CONVENTIONAL METHODS FOR ISOLATING SALMONELLA PULLORUM USING STRAINS ISOLATED FROM BACKYARD FLOCKS

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Salmonella pullorum was isolated from separate backyard poultry flocks in 2002 and 2003. Since *pullorum* was isolated from only a few birds we wanted to determine if the conventional isolation methods were sufficient to detect these strains. After experimentally infecting day-old chicks, 9 different isolation methods were tested, including direct plating, non-selective, and selective enrichments. Differences were observed in the isolation of the two *pullorum* field strains and a laboratory control strain for the various methods. However, it was concluded that the isolation method described in the National Poultry Improvement Plan was satisfactory for detecting these *pullorum* strains isolated from backyard flocks.

Salmonella typhimurium Infection in a Finch Aviary

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This is a case report of a commercial aviary producing Lady Gouldian Finches that exhibited acute onset of mortality after trying to expand the genetic pool. Salmonella typhimurium was determined to be the cause. Treatment and a cleaning and disinfection protocol was suggested. After the initial mortality and the diagnosis, there were no problems in the birds. The suggestions were not followed. There was another increase in mortality about 60 days later due to S. typhimurium. The protocol was then followed. The origin of the infection remains uncertain. The case report will discuss the findings in the birds and how the aviary was attempted to be cleared of the infection.

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Transcriptional Analysis of the Avian Innate Immune Response to Salmonella enteritidis

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University Of Delaware

The molecular mechanisms of the avian immune response to Salmonella challenge have not been extensively characterized. The avian innate immunity microarray (AIIM) is a genomics tool designed to study the transcriptional activity of the avian immune response. Day old chicks from two F8 advanced intercross lines were challenged with Salmonella enteritidis. RNA was obtained from spleens and analyzed. Thirty-three microarray elements exhibited significant (p<0.001) >1.5-fold changes in gene expression in both lines. One line was also found to induce the expression of the cytokine K60 gene as well as the beta defensin, gallinacin-1, and complement C3 genes.

Twenty Years of Salmonella Monitoring: Milestones and Setbacks

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The goal of University of Maine's salmonella program was to reduce exposure of laying flocks to Salmonella enteriditis infection, and thereby reduce contamination of eggs for human consumption. Initially, vertical transmission was addressed by testing and eliminating infected breeder flocks, and testing pullets prior to repopulating layer houses. Then, the program was expanded to include monitoring to detect environmental reservoirs and sources of infection. Control measures included sanitizing infected houses, rodent control and vaccination of replacement pullets for salmonella. Indicators for successful control of SE in commercial flocks will be discussed.

UPDATE ON THE PULLORUM-TYPHOID AGGLUTINATION TEST AND WHAT IT IS DETECTING

W. Douglas Waltman PhD

Georgia Poultry Laboratory Network

Over the last 20 years greater than 1.5 million sera have been screened with the pullorum-typhoid agglutination (PT) test resulting in 2673 positive sera. Each of these positive birds was cultured for *Salmonella*. *Salmonella* were isolated from 768 birds. The 4 most frequently isolated *Salmonella* were *heidelberg* (426), *kentucky* (144), *pullorum* (54) and *typhimurium* (38). Nine birds were positive for *enteritidis* (*SE*).

Ninety-three birds were PT tested and cultured that was submitted from 6 SE environmentally positive houses. Forty-three birds were PT and culture negative and 6 were PT and culture positive. There were 37 birds that were culture positive, but PT negative and 7 that were PT positive and culture negative. These results show the relative failure of the PT test to detect SE infected birds.

A comparative study of three antigen-capture ELISA kits for detection of Avian leukosis virus group specific antigen

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Three commercially produced antigen-capture ELISA kits were compared for their ability to detect avian leukosis virus group specific antigen (p27, gs, or gag) in meconium, cloacal swabs, egg albumen and infected cell cultures. The comparative study included ALV subgroups A, B, C, D, and J as well as MAV-1 which were inoculated in susceptible chickens and also in cell cultures. Biological materials obtained from chickens or quail infected with reticuloendotheliosis virus (REV) and/or Marek's disease virus (MDV) were used as negative controls for samples obtained in vivo. Observations on sensitivity, specificity and repeatability are reported.

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Accumulation of Quasi-Species during Serial Passage-Induced Attenuation of Marek's Disease Virus-1 (MDV-1)

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Attenuation of MDV-1 involves the serial passage of virulent isolates in cell culture 100 times. The pathobiology in chickens of a single virulent strain (648A) at defined passage intervals (p10 through p100) was determined along with their nucleotide sequences using pyrosequencing that on average generated a 50 fold base pair coverage. In theory this should allow a correlation between phenotypes (lost of neoplastic lesions, transient paralysis, etc) and genotypes. What was discovered was a collection of polymorphisms within higher passages. Few genetic changes were absolute after the attenuation passage of 80, indicating changes in specific genes are at best suggestive.

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Chronological study of the pathogenesis of Marek's disease virus in the eye and brain

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We have recently demonstrated that there is a temporal relationship between the lesions induced by Marek's disease virus (MDV) in the eye and in the brain. Early lesions in the eye (11 dpi) correlated with brain lesions associated with transient paralysis (8-11 dpi). Late lesions in the eye (later than 18 dpi) correlated with brain lesions associated with persistent neurological disease (later than 21 dpi). In the present work, we have conducted a chronological evaluation to correlate the pattern of cytokine mRNA and MDV gene expression in the eye and in the brain

Marek's Disease Seortype 1 virus detection in the field by Qualitative PCR method

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Marek's disease is a common lymphoproliferative disease of chickens, usually characterized by mononuclear cellular infiltrates in peripheral nerves and other organs and tissues including iris and skin. For diagnostic purposes, we relied on histopathology and clinical signs, being also difficult to differentiate vaccinated from unvaccinated birds. Qualitative PCR test detects the 132 bp repeat region of the MDV genome. This region is specific to serotype 1 and shows differences between Rispens vaccine strain and field strains. This test allows to detect serotype 1 MDV and to differentiate Rispens vaccine strain from field viruses and will give us a better approach for disease control.

Marek's Disease Virus MicroRNA Expression in Feather Tips

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Marek's disease virus (MDV) encodes microRNAs, which are small RNA molecules (~22-23 nucleotides) that regulate gene expression by inhibiting translation of mRNAs into protein or by directing the degradation of mRNAs. They have been identified in MDV tumors, MDV-infected chicken embryo fibroblasts (CEF), and MSB1 lymphoblastoid cells. We have identified MDV microRNAs in feather tips of MDVinfected chickens. MDV-miR-M4 is found at higher abundance in isolated feather tips than in feather tips that are surrounded by skin tissue. This indicates that microRNAs in feathers are most abundant in feather follicle epithelium, as is MDV, and do not accumulate in surrounding tissue. Chicken microRNAs miR 221 and miR 155 are also found in feather tips, with miR 221 in greater abundance. However, unlike MDV-miR-M4, there was no difference in miR 221 and miR 155 accumulation between isolated feather tip and feather tip with surrounding skin. This is consistent with the idea that miR 221 and miR 155 are host microRNAs found in all chicken tissues. Finally, the expression of MDV1 microRNAs was analyzed and compared among MDV1-infected chickens, some of which were vaccinated with one of the following: HVT, HVT+SB1, and CVI988. Understanding microRNA expression in young chickens may have utility in predicting disease outcome.

Tumors and Tumor-like Lesions of the Female Reproductive Tract: A Proposed Classification

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Classification schemes exist for tumors and tumor-like lesions that occur in the female reproductive tracts of women and mammals, but no such classification has been developed for birds. Tumors of the female avian reproductive tract, notably viral tumors (Marek's disease, avian leukosis, reticuloendotheliosis), adenomas, adenocarcinomas, and leiomyomas, are relatively common. Adenocarcinomas of the ovary and leiomyomas serve as animal models of human disease. Other, less common tumors include cystadenomas, sex cord/gonadostromal tumors (granulosa cell tumors, ovarian Sertoli cell tumors), and germ cell tumors (dysgerminomas). Teratomas and other mesenchymal tumors are rare. Characteristics of each type of tumor and a proposed classification scheme will be presented. Additionally, lesions that might be confused with tumors such as cysts and inflammatory diseases will be described.

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An unpredicted subgenomic mRNA produced by turkey coronavirus

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Coronavirus transcribes its genome to a set of 5'- and 3'-coterminal subgenomic mRNA (sqRNA) for structure gene translation. The key element in sqRNA transcription is transcription-regulating sequences (TRS) located at the 3' end of leader and upstream of structure genes (body TRS). The objective of the present study was to determine the TRS of each sgRNA. Both positive (+) and negative (-) strand cDNA were synthesized from total RNA isolated from turkey coronavirus (TCoV) infected turkey small intestines using TCoV-specific primers. PCR was then used to amplify sequences flanking TRS region using synthesized cDNA as the template. Sequence of cloned PCR product revealed a new sgRNA for 3' untranslated region (sg3UTR). The finding was further confirmed by using newly designed primers to amplify sg3UTR from three isolates of TCoV and strain Ark99 of infectious bronchitis virus (IBV). Both (+) and (-) sgRNA were amplified and sequenced, demonstrating UAAAC as body TRS for sgRNA production. An open reading frame (ORF) was identified within sg3UTR which encodes a transmembrane protein of 74 residues with predicted RNA binding activity at the Cterminal. The results indicated that TCoV has six sgRNA with the 6th sgRNA in the 3' UTR.

Biological assessment of recombinant avian metapneumovirus subgroup C (aMPV-C) viruses containing different length of the G gene in cultured cells and SPF turkeys

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Genetic variation in length of the G gene among different aMPV-C isolates has been reported. However, its biological significance in virus replication and pathogenicity is unknown. In this study, we generated two Colorado (CO) strain-based recombinant viruses containing either the full-length G gene derived from a Canadian goose isolate or a truncated G gene of the CO strain. The biological evaluation of these two recombinant G variants in Vero cells and SPF turkeys revealed differences in virus growth dynamics and severity in clinical signs, demonstrating the effect of the G protein truncation on virus replication and pathogenicity.

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Characterization and Complete Genome Sequence of Avian Paramyxovirus Serotype-3

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The avian paramyxoviruses consist of nine antigenic serotype. The Newcastle disease virus belongs to serotype-1 and is well characterized among all avian paramyxoviruses. Very little is known about avian paramyxovirus serotypes 2 through 9. Avian paramyxovirus serotype-3 (APMV-3) has been isolated from chickens and turkeys in different parts of the world. The pathogenicity of this virus varies from conjunctivitis, enteritis to signs of central nervous system disease. The detailed molecular, pathological and epidemiological characteristics of APMV-3 are not known. We have analyzed the growth characteristics of APMV-3 in different cell lines and chicken embryos. The pathogenicity studies of this virus in embryonated chicken eggs showed hemorrhages and mortality. Furthermore, virus infection in chickens and turkeys of different age groups showed virus replications in different organs. Complete genome sequence of APMV-3 strains PKT/Netherland/449/75 and turkey/Wisconsin/68 revealed unusually long trailer region, which is largest among all other avian paramyxovirus till date. Work is underway to develop a reverse genetics system for this virus, which will facilitate development of vaccine against this disease.

Detection of Turkey type 2 astroviruses (TAstV2) in guinea fowl buildings: assessment of the cleaning -disinfection procedures

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Turkey Astrovirus type 2 (TAstV2) has been associated with enteritis in turkey and recently, in guinea fowl. A real-time RT-PCR assay was designed so as to amplify a part of the viral polymerase (ORF1b) of turkey or guinea poults TAstV2. This assay was applied to the detection of astroviruses in the environment of poultry farms, after the application of the cleaning-disinfection procedure. Swabs were applied on the walls, drinkers and litter and dark beetles were also sampled. Some RT-PCR-positive samples were inoculated to day-old guinea poults, so as to assess if the material was actually infectious. Using this approach, the efficacy of different cleaning-disinfection procedures was compared.

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Development of reference antisera to enteric-origin avian viruses

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Recent molecular surveys have revealed geographically distinct lineages of avian reovirus, rotavirus and astrovirus circulating in commercial poultry. To improve our understanding of enteric virus pathogenesis, specific immunological reagents are needed to detect viruses in histological samples. To this end, we prepared an inactivated oil emulsion turkey rotavirus vaccine and used it to produce reference antiserum in chickens. This antiserum was utilized to detect rotavirus antigen in the mature enterocytes of experimentally infected turkey poults via immunohistochemistry (IHC). This reference antiserum also resulted in less background staining than IHC using turkey convalescent serum. Antisera produced using this methodology should prove useful in tracking enteric virus infection in the organs of experimentally infected birds.

Metagenomics analysis of intestinal contents from runting-stunting-syndrome diseased broiler

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Several viruses have been associated with the runting and stunting syndrome (RSS) in broiler chicken. Although non-infectious issues may influence the manifestation of RSS, viral agents play an important role. Most of these viruses do not propagate in in vitro or in ovo. Viruses of different families have been identified using the 454 technology for a global analysis of the viral metagenome of RSS infected chicken in comparison to non-infected chickens. Sequences were categorized based on a viral database created for this purpose. The obtained results will increase the understanding of the role of certain viruses in RSS.

Pathogenicity of recently re-emerged fowl adenovirus (serotype 4, 9 and 11) isolated from broilers vaccinated with infectious bursal disease intermediated plus type vaccine in Korea

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Fowl adenovirus (FAdV) infection associated with inclusion body hepatitis (IBH) and hepatitis-hydropericardium syndrome (HHS) was re-emerged recently in infectious bursal disease (IBD) intermediated plus type vaccinated broilers in Korea. In these cases, we isolated 3 serotypes of FAdV (serotype 4, 9 and 11). In the present study, we compared pathogenicity of these FAdV isolates in specific pathogen free (SPF) chickens with broilers vaccinated with IBD intermediated plus type vaccine to evaluate the potential pathogenesis of FAdV in vaccine induced transient immunocompromised broilers. The pathogenicity of FAdV infection was evaluated by mortality, histological lesion and virus shedding in feces. Immune status of chickens was also assessed by T-cell activity, cytokine levels and other immunological indicators. The results of experimental study will be discussed.

Safety and Efficacy of Fowl Adenovirus Serotype-4 Inactivated Oil Emulsion Vaccine in Chicken

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Hydropericardium syndrome (HPS) occurs usually in 3~6week olds broilers with mortality of up to 80% and characterized by hydropericardium and multifocal hepatic necrosis with intranuclear inclusion body. Because the economic impact was severe due to several outbreaks of HPS in Korea, the development of inactivated vaccine of fowl adenovirus serotype-4 (FAdV-4) was needed. The present study describes safety and efficacy of FAdV-4 inactivated oil emulsion vaccine. At present, the developed FAdV-4 vaccine is effective in the aspect of viral shedding, mortality and pathological lesions. However, we need further study to develope standard challenge system using different FAdV-4 with variable virulence.

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The Comparison of Pathogenicity in the SPF Chickens Challenged with Fowl Adenovirus and/or Avian Reovirus

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Recently, hydropericardium syndrome (HPS) caused by fowl adenovirus (FAdV) have been reported frequently in Korea. However, the clinical severity of HPS was variable depends on the associated factors such as type of transmission, level of sanitation and concurrent diseases. One of the diseases related with HPS was avian reovirus (ARV) which induce various clinical diseases especially malabsorption syndrome. Therefore, this study was designed to evaluate the differences of clinical severity when inoculated with ARV and/or FAdV in the SPF chickens. After inoculation, we observed and compared the weight change, clinical signs, pathological lesions, blood chemistry and virus reisolation among groups.

The design of a SYBR Green real-time PCR assay for the detection of Duck and Goose circoviruses (DuCV and GoCV) and its application to diagnosis.

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Circoviruses are small, non enveloped viruses, with a single stranded DNA genome. Waterfowl flocks affected by GoCV or DuCV may show growth retardation, feathering disorders and secondary infections. We developed a SYBR Green real-time PCR assay for detecting and quantifying both circoviruses. A set of PCR primers was designed on the replication gene, based on the alignment of the sequence of 3 GoCVs and 6 DuCVs. Clinically suspected Mule ducks were sampled, including for each bird cloacal swab, spleen, bursa and blood. After DNA extraction, the SYBR Green real-time PCR assay was performed and the viral load determined. This strategy will be used to investigate the pathobiology of waterfowl circoviruses and to evaluate their putative immunosuppressive role.

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Turkey Stunting and Enteritis Syndrome - Pennsylvania Cases

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An unusual increase in the number of cases of stunting and enteritis of turkey poults up to 8 weeks of age in 2008 stimulated interest into the possible causes, effective treatments, and preventative measures. These will be reported along with clinical signs, gross lesions, histologic lesions, and microbiologic findings.

R Plasmids Found among Emergent APEC Strains

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R plasmids are common among avian pathogenic *Escherichia coli* (APEC). These plasmids often encode resistance to multiple antibiotics and disinfectants and harbor class 1 integrons. Some even harbor pathogenicity islands. Recent study of APEC isolated over a 30 year period has revealed that R plasmid-containing strains of APEC are emergent. In this study, classic and molecular bacteriologic techniques are used to characterize the plasmids of these emerging strains.