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Research Education and Economics

Agricultural Research Service

Office National Programs

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National Program 103

ANIMAL HEALTH

ACCOMPLISHMENT REPORT 2016-2020

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Executive Summary

Animal health plays a critical role in ensuring a safe and adequate food supply to the United States population and the world. In spite of years of research, challenges remain in animal health with both emerging diseases and diseases that have long been problematic. The mission of the ARS Animal Health National Program is to deliver scientific information and tools to detect, control, and eradicate animal diseases of high national priority. The ultimate goal of the program is to protect and ensure the safety of the Nation's agriculture and food supply through improved disease prevention, protection, mitigation, response, and recovery.

The anticipated products of the animal health research program were captured in an Action Plan at the start of the 5-year national program cycle. Some of the anticipated products in the Action Plan include finding solutions to prevent economic losses from domestic and foreign animal diseases, providing scientific information to maximize on-farm biosecurity practices for naturally or intentionally introduced pathogens, establishing methods to detect, analyze, and respond to new and emerging pathogens, and developing disease prevention tools such as alternatives to antibiotics, vaccines and biotherapeutics.

Research accomplishments in this report are organized under the seven Research Components of the Action Plan. For each component, the report provides the rationale for the research, the anticipated products and impact, followed by examples of accomplishments and contributions to those national research priorities.

Component 1: Biodefense Research
The ARS biodefense research activities under Component 1 include research conducted on foreign animal diseases and emerging diseases that pose the greatest threats to the United States. Foreign animal diseases include Foot-and-Mouth Disease (FMD), which is the only agricultural agent classified as a Tier 1 select agent by both the Centers for Disease Control and Prevention (CDC) and the Animal and Plant Health Inspection Service (APHIS). Since 2015, the ARS Biodefense Research program has made significant contributions towards the defense of the country against disease incursions as well as food security initiatives worldwide. Notable contributions include new vaccine technologies transferred to industry for further research and development for Foot-and-Mouth Disease and African Swine Fever.

Component 2: Antimicrobial Resistance
The research in this component focused on two strategic areas: 1) the ecology of antimicrobial resistance and 2) the research and development of antibiotic alternatives. ARS scientists have made significant progress in several areas, including increasing the understanding of the effects of antibiotic use on swine respiratory pathogens and the animal's natural microbial flora; identifying differences in antibiotic resistant bacteria in human and animal species; and developing and transferring vaccine candidates that can be used to prevent animal diseases that commonly require the use of antibiotics.

Component 3: Zoonotic Diseases
Zoonotic diseases represent one of the leading causes of illness and death in people. The ARS zoonotic disease research program focuses on brucellosis, leptospirosis, tuberculosis (TB), and Q Fever with the strategic goal of developing countermeasures to prevent disease transmission in domestic livestock and wildlife reservoir hosts. While eradication of TB and Brucellosis remain the goal, outbreaks in domestic animals due to the presence of the organisms in wildlife remains a serious concern. As a result, much of the direction of the research conducted in this area is developing strategies to diagnose and control the diseases in wildlife, including bison and white-tail deer. Notable accomplishments include validating laboratory inactivation methods for *Brucella* in response to a request from the Federal Experts Security Advisory Panel (FESAP), furthering the understanding of why some animals become persistently infected with the RB51 vaccine, and development of new culture methods to enhance the understanding of leptospirosis bacterial protein expression during infection.

Component 4: Respiratory Diseases
Despite the wide-ranging use of vaccines and antibiotics in animal agriculture, endemic respiratory diseases remain a primary health threat to livestock and poultry. Most respiratory diseases present themselves as disease complexes involving multiple primary and secondary viral and bacterial pathogens, complicating control and prevention strategies. Importantly, livestock and poultry that develop respiratory diseases have notable decreases in growth performance. ARS scientists have made tremendous strides in understanding host-pathogen interactions, mechanisms of transmission, and the discovery of highly effective diagnostics, and vaccines to control respiratory diseases of livestock and poultry. Since 2015, ARS scientists have made significant contributions to control respiratory pathogens, including the development of vaccines against *Streptococcus suis* in swine; identifying *Mycoplasma ovipneumoniae* in a wide-array of wildlife species; and development of automated vaccine delivery systems for poultry.

Component 5: Production Diseases
Although many production diseases can be prevented through sound biosecurity measures and good management practices, significant scientific gaps remain in our understanding of disease dynamics and host-pathogen interactions. Johne's caused by *Mycobacterium avium* subspecies *paratuberculosis* remains a serious disease for dairy and sheep farmers. This disease causes significant economic loss and to date accurate diagnostic and control strategies remain elusive. There are also significant gaps in our understanding of enteric disease complexes of poultry, and the ecological and host interactions that lead to disease and production losses. Since 2015, ARS scientists have provided key scientific information that has increased our understanding of Johne's disease through the identification of proteins that serve as better diagnostic targets. Another accomplishment includes enhancing the understanding of how selective breeding may make animals more susceptible to mastitis.

Component 6: Parasitic Diseases
Parasites represent one of the most diverse groups of organisms that are responsible for hundreds of insidious diseases ranging from enteric diseases to vector-borne hemoparasitic infections. The livestock and poultry industries are severely affected by significant losses in animal production due to lower weight gain, anemia, diarrhea, and death. Of great concern is the increase in

anthelmintic resistance by parasites over the years. As loss of these important tools for controlling parasitic infections becomes more common, alternatives must be found to maintain healthy animals and a safe source of food. In addition, controlling hemoparasites is important to the economies of both the beef and dairy cattle industries, but also the U.S. sheep, goat and equine industry. Since 2015, ARS scientists have made significant contributions as evidenced by the development of improved diagnostic assays for detecting anaplasmosis in cattle and piroplasmosis in horses; developing treatment techniques to clear piroplasma organisms from infected horses; and the discovery of potential vaccine candidates for control of intestinal parasites in cattle as well as identifying some potential genes that may help to naturally reduce the parasite load.

Component 7: Transmissible Spongiform Encephalopathies
Scrapie of sheep and goats, bovine spongiform encephalopathy (BSE) of cattle, chronic wasting disease (CWD) of deer and elk, and variant Creutzfeldt-Jacob disease (vCJD) of humans are all fatal neurodegenerative disorders classified as transmissible spongiform encephalopathies (TSEs). There are no effective treatments or cure, and the origin of TSEs have yet to be determined. Since 2015, ARS scientists have made significant contributions to our understanding of the pathobiology of prion strains, interspecies transmission, successfully developed new improved diagnostic methods, and the identification of resistant genotypes to breed resistant sheep and goats in support the United States Scrapie eradication program.

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United States Department of Agriculture
Research, Education, and Economics
AGRICULTURAL RESEARCH SERVICE

National Program 103 Animal Health

ACCOMPLISHMENT REPORT 2016-2020

Introduction

Agricultural Research Service
The Agricultural Research Service (ARS) is the principal in-house research agency of the United States Department of Agriculture (USDA). ARS is one of four agencies in the Research, Education, and Economics (REE) mission and is charged with extending the Nation's scientific knowledge with research projects in agriculture, human nutrition, food safety, natural resources, and the environment. ARS supports more than 2,000 scientists and post docs organized into approximately 660 permanent research projects at over 90 locations across the country and five overseas laboratories.

Role
ARS conducts innovative research to find solutions to problems of high National priority that impact the American people daily. ARS often undertakes high-risk research endeavors to make significant breakthroughs in important problem areas. ARS research programs also complement the work of State Colleges and Universities, State Agricultural Experiment Stations, other Federal agencies, and the private sector. Mechanisms for addressing state and local issues often are already in place; therefore, activities within ARS focus on issues having a regional or national scope and where there is a clear federal role. ARS also provides research support to USDA action and regulatory agencies and to several other Federal regulatory agencies, including the Departments of State and Defense, the Food and Drug Administration, and the Environmental Protection Agency.

Mission
The mission of the program is to deliver scientific information and tools to detect, control, and eradicate animal diseases of high national priority.

Goal
The goal of National Program 103 (NP 103), Animal Health, is to protect and ensure the safety of the Nation’s agriculture and food supply through improved disease detection, prevention, control, and treatment. Basic and applied research approaches are used to solve animal health problems of high national priority. Emphasis is given to methods and procedures to control animal diseases through the discovery and development of:

- Diagnostics
- Vaccines
- Biotherapeutics
- Animal genomics applications
- Disease management systems
- Animal disease models
- Farm Biosecurity measures

The vision for the program is to be recognized worldwide as a leader in animal health research that delivers effective solutions to prevent and control animal diseases that impact agriculture and public health.

The Animal Health National Program has ten strategic objectives:

1. Establish ARS laboratories into a fluid, highly effective research network, to maximize use of core competencies and resources.
2. Continue to develop specialized high containment facilities to study zoonotic and emerging diseases.
3. Strengthen the integration of our animal and microbial genomics research programs.
4. Strengthen our core competencies in comparative immunology.
5. Launch a research program to combat antimicrobial resistance bacteria with emphasis on the ecology of antimicrobial resistance and the discovery and development of alternatives to antibiotics.
6. Sustain a technology-driven vaccine and diagnostic discovery research program.
7. Develop core competencies in field epidemiology and predictive biology.
8. Develop internationally recognized expert research laboratories recognized by the World Organization for Animal Health (OIE) and the Food and Agriculture Organization of the United Nations (FAO).
9. Sustain best-in-class training centers for our nation's veterinarians and scientists.
10. Sustain an effective technology transfer program to achieve the full impact of our research discoveries.

Overall Program Impact

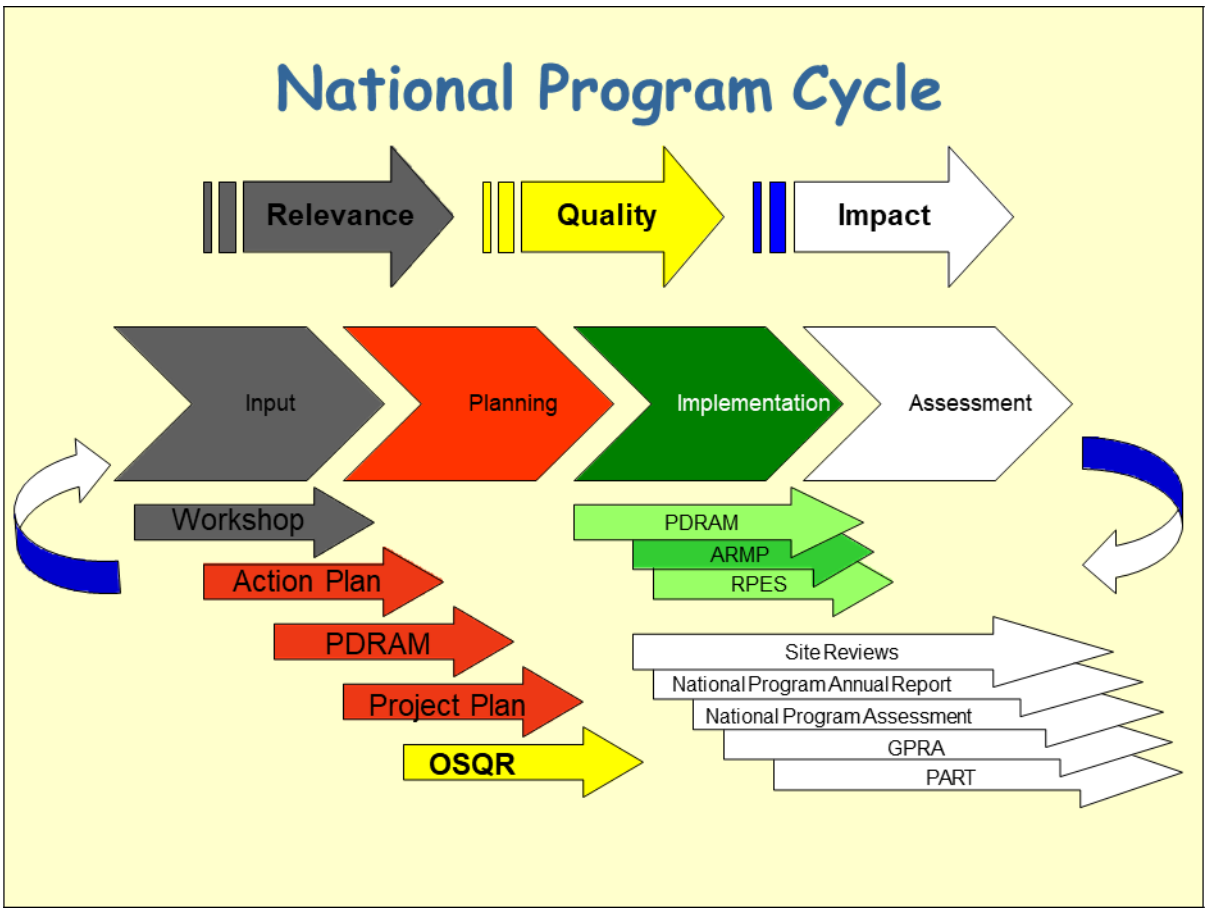
During the 5-year period covered by this report, ARS scientists have provided scientific information that has significantly enhanced our knowledge of endemic and foreign animal diseases, including new and emerging diseases such as Porcine Epidemic Diarrhea Virus and Senecavirus A, as well as the ongoing evolution of influenza A viruses in animals. Scientific information for zoonotic pathogens has been used widely to establish the safety of our food products and support the export of agricultural products. Importantly, several tools for detecting and preventing animal disease outbreaks have been discovered and transferred to action and regulatory agencies and the private sector for full development. Examples include rapid nucleic acid-based diagnostic assays for priority infectious diseases (e.g., highly pathogenic influenza virus) transferred to Animal and Plant Health Inspection Service (APHIS) and the National Animal Health Laboratory Network to support surveillance programs throughout the United States. Although details of Cooperative Research and Development Agreements with the private

sector are considered confidential, new vaccine technologies for priority diseases like Foot-and-Mouth Disease (FMD) and Classical Swine Fever (CSF) have been transferred and are currently being developed by pharmaceutical companies with the support of ARS scientists. A recent example is the first successfully genetically engineered molecular FMD vaccine, which was specifically designed for the U.S. National Veterinary Stockpile.

PLANNING AND COORDINATION FOR NP 103'S 5-YEAR CYCLE

National Program Cycle

The management and execution of all ARS research programs is organized around the five-year National Program Cycle, consisting of four sequential phases designed to ensure the relevance, quality, and impact of every National Program: 1) Input; 2) Planning; 3) Implementation; and 4) Assessment.



National Program Assessment

A National Program Assessment is conducted through two avenues every five-years, one route is the collection of a survey of stakeholders and partners. This survey allows ARS to periodically update the vision and rationale of the NP 103 program and assess the relevancy, effectiveness, and responsiveness of ARS research to stakeholder's needs. The Office of National Programs staff organizes the survey to facilitate the review and simultaneously provide an opportunity for customers, stakeholders, and partners to assess the progress made through the National Program

and provide input for future modifications to the National Program or the National Program's research agenda. A second assessment method is to ask a panel of animal health experts to review the action plan of the National Program and provide input on quality and impact of research over the past five years.

Animal Health National Program (NP 103)
The mission of the Animal Health Program is to conduct basic and applied research on selected diseases of economic importance to U.S. livestock and poultry industries. The goals of the research mission are to produce knowledge and technology to reduce economic and animal losses from infectious, genetic, and metabolic diseases of livestock and poultry. Dr. Cyril G. Gay, Senior National Program Leader, and Dr. Roxann Motroni, National Program Leader, lead this ARS animal health research program. Dr. Gay is lead on projects involving viruses and prions. Dr. Motroni leads the projects on bacterial and parasitic pathogens. The Animal Health National Program currently includes approximately 40 core research projects supported by 86 scientists located at nine research sites throughout the country. The ARS research budget for the Animal Health Program was approximately $80.9 million in FY19. For reference since the last accomplishment report in 2015, the budget has increased $12.9M, and there are 15 fewer scientists and 2 fewer research locations. Approximately, $6M of the $12.9M budget increase is for the NBAF science program.

Background
With a growing global population, increased demands for animal proteins, and the economic dependence on livestock production of over one billion people worldwide, investments in animal health research are critical to achieve global food security and the sustainable growth and resilience of a safe food supply for a growing world population. Enhancing the health of animals in agricultural production systems directly impacts food quality and ensures a sufficient supply of macro and micro-nutrients to meet people's basic needs worldwide. When combined with other investments in agricultural development, research-based innovations address some of the fundamental constraints that give rise to food insecurity by reducing production risks associated with pests and diseases.

Achieving results in animal health research in the 21st century requires a "systems biology" approach in which knowledge obtained from animal studies in functional genomics, immunology, pathology, physiology, nutrition, and epidemiology are integrated in the discovery and development of countermeasures for preventing and controlling disease outbreaks.

Accordingly, this national program fosters the alignment of research expertise and the establishment of strategic partnerships to maximize productivity and impact. For this purpose, NP 103 projects are aligned under seven research components. Each component includes problem statements that define the scope of the action plan. Research components draw upon relevant expertise within NP 103, but also espouse challenges by seeking overarching contributions from scientists working in NP 101 (Animal Production), NP 104 (Veterinary, Medical and Urban Entomology), NP 106 (Aquaculture), NP 107 (Human Nutrition), NP 108 (Food Safety), NP 215 (Grass, Forage, and Rangeland Agroecosystems), and NP 303 (Plant Diseases); thus coordinating and integrating that expertise to develop a specific useful application of the knowledge. Projects within the research components have also attracted additional federal, university, and industry partners at both the national and international level.

The aim of these partnerships is to support component projects. Their inclusion has enabled and enhanced the anticipated products of the component projects. Because a significant number of projects in the animal health research portfolio focus on the discovery of novel technologies, technology transfer strategies were identified to maximize the impact of the research, and where appropriate, help foster investments by the private sector in the development of these technologies.

The anticipated products of the animal health program targeted the following:

- Enhance "Global Food Security" by finding solutions to problems incurred by domestic and transboundary animal diseases of livestock and poultry.
- Support "One Health" initiatives by implementing research programs that will benefit the animal health, public health, and the biomedical research communities.
- Advance the productivity frontier by supporting "Feed the Future" initiatives.
- Develop methods to help producers adjust to changing farming practices that will allow consumer driven issues to be accommodated without compromising financial viability.
- Establish methods to detect, analyze, and effectively respond to new and emerging pathogens that threaten agriculture and public health.
- Find solutions to create and maintain a barrier to pathogens at the domestic-wildlife animal interfaces.
- Build integrated research programs to discover genetic variations associated with disease susceptibility to increase our farmers' productivity and competitiveness.
- Develop experimental animal disease models that will serve the animal and human health research communities to significantly shorten the timelines for developing breakthrough medicines and disease prevention tools and validate the use of countermeasures in disease control programs.
- Conduct and coordinate Animal Health research to support ARS Grand Challenge Synergies and ARS X.

STRUCTURE OF NP 103

Research Component Overview:
The seven research components of the program are:

Component 1: Biodefense Research
Component 2: Antimicrobial Resistance
Component 3: Zoonotic Diseases
Component 4: Respiratory Diseases
Component 5: Priority Production Diseases
Component 6: Parasitic Diseases
Component 7: Transmissible Spongiform Encephalopathies

NP 103 RESEARCH CONTRIBUTES TO THE OVERALL ARS ANIMAL HEALTH RESEARCH EFFORT

Relationship of this National Program to the USDA Strategic Plan:
This Action Plan is relevant to Strategic Goals 3 and 4 of the USDA Strategic Plan for 2014-2018 (http://www.usda.gov/documents/usda-strategic-plan-fy-2014-2018.pdf).

The aim of Strategic Goal 3 is to help America promote agricultural production and biotechnology exports as America works to increase food security. The Action Plan is linked to Objective 3.1: Ensure U.S. Agricultural Resources Contribute to Enhanced Global Security.

The aim of Strategic Goal 4 is to ensure that all of America's children have access to a safe, nutritious and balanced meals. The Action Plan is linked to Objective 4.4: Protect Agricultural Health by Minimizing Major Diseases and Pests to Ensure Access to Safe, Plentiful, and Nutritious Food.

The implementation of the research in the Action Plan will provide critical scientific information and tools to help control and eradicate diseases that threaten animal production and thereby contributes to global food security. This action plan also supports sustainable agriculture production in food-insecure nations by enabling the control of transboundary animal diseases, which the Food and Agriculture Organization of the United Nations (FAO) and the World Organization for Animal Health (OIE) have declared significantly impact the lives of millions of people in developing countries that dependent on livestock and poultry production for their livelihood and well-being.

Relationship of this National Program to the USDA REE Action Plan:
This Action Plan fits under the general guidance of the USDA REE Action Plan's (https://www.usda.gov/sites/default/files/documents/usda-ree-science-action-plan.pdf) Goal 1: Sustainable Intensification of Agricultural Production, Subgoal 1B: Crop and Animal Health, and Subgoal 1C: Crop and Animal Genetics, Genomics, Genetic Resources, and Biotechnology. Specific NP 103 alignment includes the research and development of technologies to mitigate animal diseases to enable sustainable animal production systems.

Relationship of this National Program to the ARS Strategic Plan:
This Action Plan supports the 2012-2017 ARS Strategic Plan, Goal Area 4: Animal Production and Protection:
(http://www.ars.usda.gov/SP2UserFiles/Place/00000000/NPS/OAA/ARS%20Strat%20Plan%202012%20-%202017%20Final.pdf).

Within that Strategic Goal Area, Goal 4.2 (Prevent and Control Pests and Animal Diseases that Pose a Threat to Agriculture, Public Health, and the Well-being of American Citizens) includes eight of the ten strategic objectives in NP 103. Specific emphasis is placed on delivering scientific information and tools to control and eradicate domestic and exotic diseases. The following performance measure sets the targets for NP 103 research within the USDA ARS Strategic Plan:

Performance Measure for Goal 4.4.2: Provide scientific information to protect animals, humans, and property from the negative effects of pests and infectious diseases. Develop and transfer tools to the agricultural community, commercial partners, and government agencies to control or eradicate domestic and exotic diseases and pests that affect animal and human health.

HOW THIS REPORT WAS CONSTRUCTED AND WHAT IT REFLECTS

In this report, NP 103 achievements and their impacts are organized according to the seven Action Plan components. The report first outlines the rationale for the research, followed by the research needs, anticipated products and impact for each of the components. Then, selected accomplishments are listed as examples of contributions toward the high priority needs identified by customer/stakeholders and described in the NP 103 Action Plan.

For the most part, the content of this report is derived from the 2016-2020 annual reports from NP 103 research projects. This report does not include all accomplishments achieved by this national program, rather, only selected accomplishments that illustrate and exemplify the total progress and achievements at the national level.

Research projects in NP 103 are listed in Appendix I; publications in peer-reviewed journals authored by NP 103 scientists are compiled in Appendix II; patents and technology transfer are listed in Appendix III; research collaborations are listed in Appendix IV.

This report was prepared for an external (to ARS) retrospective review of NP 103 research that was conducted from October 2015 to February 2020 to assess how well this national program attained its goals, as outlined in the 2017-2022 Action Plan.

Accordingly, the purpose of the retrospective review is not to review the performance of individual research projects, but rather to gauge the overall impact of NP 103. Consequently, the report does not attempt to catalogue all the accomplishments of the constituent research projects of NP 103. Individual scientists or projects are not identified by name in the narrative text; their achievements are described in the context of contributions to the national program's commitments to U.S. agriculture and progress in animal health research.

This report provides extensive lists of animal health research priorities and anticipated products from the NP 103 Action Plan. Although these research priorities and anticipated products serve to help measure the national program's progress during the last five years, their primary purpose was to provide overarching targets for scientists working in the NP 103 National Program. Importantly, these extensive lists provided direction for acquiring additional resources (extramural funding and research collaborations) to build and expand research programs where needed during the national program cycle. Based on the available funds and resources at the start of the national program cycle, ARS scientists were given specific research objectives to prepare their project plans, which were provided in Program Direction and Resource Allocation Memoranda.

Component 1: Biodefense Research

Rationale for the research:

The health of animals is continuously threatened by diseases naturally, accidently, or deliberately introduced into a naïve healthy population of animals. These diseases vary in the degree of economic loss they cause, their potential to spread, ease of control and our ability to eradicate them. Many of these diseases are caused by high consequence animal pathogens that are not hindered by international borders and are thus labeled transboundary diseases by the Food and Agriculture Organization (FAO) of the United Nations. Since most of these diseases do not exist in the United States they are also referred to as foreign animal diseases. Of particular concern are emerging animal diseases that challenge our disease surveillance systems and our ability to prepare and respond to disease outbreaks. Introduction of these diseases in the United States could have devastating social and economic effects not only in the country's agricultural systems but also in a wide range of economic activities, such as the export and trade of agricultural products.

ARS biodefense research activities under Component 1 include research conducted on Select Agents identified under the Agricultural Bioterrorism Protection Act of 2002. Select Agents pose a severe threat to animal health or animal products. Research on Select Agents is regulated by the USDA Animal and Plant Health Inspection Service (APHIS) and/or the Centers for Disease Control and Prevention (CDC) and requires high containment laboratories and animal facilities. The NP 103 program operates 3 high-containment laboratories that work with a variety of animal species. They are: Plum Island Animal Disease Center (PIADC), U.S. National Poultry Research Center (USNPRC) and the National Animal Disease Center (NADC). The National Bio and Agro-Defense Facility (NBAF) will begin operations in 2022 and eventually provide expanded BSL-4 and biologics development capabilities to replace the aging PIADC. Outputs under Component 1 are used by Federal and State regulatory agencies for surveillance and to mitigate accidental or potential intentional acts of agroterrorism.

The program direction provided to scientists assigned to biodefense research purposely targeted basic research aimed at understanding the mechanisms of disease agent survival outside of the host, movement between susceptible hosts, animal infection, and pathogen escape and shedding. To improve our response to disease incursions, the program also allocated significant resources towards the discovery of veterinary countermeasures for the U.S. National Veterinary Stockpile. The research program has successfully established strategic international research collaborations with scientists in countries where foreign animal diseases (many of which are select agents) are endemic. One notable accomplishment was the building and continuous expansion of the Global African Swine Fever Research Alliance (GARA - http://www.ars.usda.gov/gara). Partnerships with researchers in other countries have contributed significantly to the program by providing access to critical samples, scientific information, resources, and the ability to test countermeasures in endemic settings.

Stakeholders representing the livestock and poultry industries that responded to the 2015 ARS animal health national survey identified biodefense research as a national priority. Foreign animal diseases as well as emerging and/or re-emerging diseases represent a major threat to U.S.

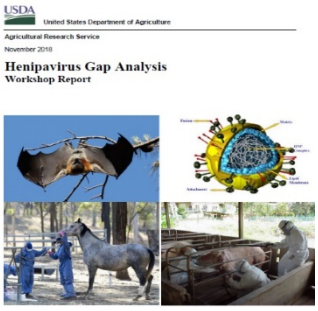
agriculture. Introduction of these agents, either accidental or deliberate, has the potential to result in devastating social and economic effects, not only to the country's agricultural systems but also on a wide range of economic activities.

Research needs:
In order to control foreign animal diseases, a wide variety of agent detection platforms need to be developed and validated. Information for designing these platforms requires knowledge of pathogen genomics and proteomics and understanding the evolution and genetic variability of disease agents. There is a dearth of knowledge for many priority foreign animal diseases, such as understanding the host range of pathogens; their primary site of replication; tissue tropism; carrier state; duration and routes of shedding; transmission mechanisms (e.g. vectors, fomites, aerosols); and ecology and epidemiology (e.g., wildlife reservoirs). If a disease outbreak should occur in the United States, effective veterinary countermeasures are needed to support suitable control strategies compatible with a short time to recover to limit the economic impact. There is a need for developing vaccines and biotherapeutics suitable for the U.S. National Veterinary Stockpile. Importantly, integrated approaches for responding to foreign animal disease outbreaks are needed to enhance our capability to regain country disease-free status and retain economic sustainability.

Since no one can predict with certainty the cause of the next pandemic, there is a need to continuously isolate, identify, and characterize pathogen(s) associated with new disease complexes of unknown etiologies. The capabilities to rapidly detect, identify, and characterize new and emerging animal pathogens are paramount and a primary goal of the biodefense research program. Scientists need to conduct animal studies in the relevant host to fulfill Koch's postulates and determine the pathogenesis of monovalent and multivalent infections. Once a new agent is isolated there is a need to sequence partial or complete microbial genomes to identify unique sequences for diagnostic discovery and molecular epidemiology research. Research is needed to understand mechanisms of disease, disease transmission, and host range specificity to determine the prevalence and emerging potential of new diseases. Ultimately research is needed to identify predictors of disease emergence and disease outbreaks. Establishing strategic international research collaborations is critical to address many of these research needs.

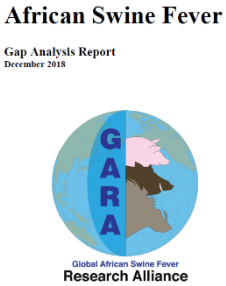
Many of the specific research priorities and anticipated products for Component 1 were derived from gap analyses conducted by worldwide experts in the last five years. These gaps analyses were organized and led by ARS at the request of stakeholders and partners, such as the APHIS National Veterinary Stockpile (NVS) Steering Committee (http://www.aphis.usda.gov), the Global Strategic Alliances for the Coordination of Research on the Major Infectious Diseases of Animals and Zoonoses (http://www.star-idaz.net/), and the United States Animal Health Association (http://www.usaha.org). Some of the results from these gap analyses are confidential due to recommendations made in the reports for stockpiling countermeasures. However, several reports have been amended for public distribution and are available on the following ARS website:
http://www.ars.usda.gov/research/programs/programs.htm?np_code=103&docid=17547. Some of these reports have been instrumental in shaping research priorities worldwide. The following four examples illustrate some of the impacts resulting from gap analysis workshops conducted for priority foreign animal disease threats.

The first example is the Henipavirus Gap Analysis Workshop organized at the Canadian Food Inspection Agency (CFIA) Winnipeg Laboratory in Canada, November 2017. The report from this workshop (http://go.usa.gov/xnHgR) provides a comprehensive assessment of the current scientific knowledge and the available countermeasures to effectively control and mitigate the impact of a Nipah virus outbreak in North America. One of the key outcomes from this workshop was the establishment of a strategic collaboration between the ARS Foreign Animal Diseases Research Unit at the Plum Island Animal Disease Center and the CFIA.

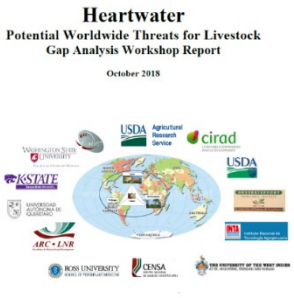


The second example is the Foot-and-Mouth Disease (FMD) Gap Analysis Workshop organized in Buenos Ares, Argentina, June 2018. The report from this workshop (http://go.usa.gov/kCqF) is instrumental in setting the research agenda and activities of the Global FMD Research Alliance (http://www.ars.usda.gov/GFRA). Importantly, the report provides concrete information on the gaps in the scientific information and tools available for controlling FMD, and a list of research priorities for addressing those gaps. This information is also critical in guiding funding agencies as well as enabling strategic research collaborations within the GFRA. In addition, the FMD gap analysis workshop report served as the basis for disseminating critical information in the scientific literature to enable the development of the most promising countermeasures; e.g., Robinson, L., Knight-Jones, T.J., Charleston, B., Rodriguez, L.L., Gay, C.G., Sumption, K.J., Vosloo, W. Global Foot-and-Mouth Disease Research Update and Gap Analysis: 3 – Vaccines (2016) Transboundary and emerging diseases, 63, pp. 30-41.

The third example is the African Swine Fever (ASF) Gap Analysis Workshop organized in Sardinia, Italy, April 2018. The report from this workshop (https://go.usa.gov/xPfWr) was instrumental in setting the research agenda and activities of the Global ASF Research Alliance (http://www.ars.usda.gov/GARA). Importantly, the report also provides information on the gaps in the scientific information and tools available for controlling ASF, and a list of research priorities for addressing those gaps. This information is critical in guiding funding agencies as well as the establishment of strategic research collaborations within the GARA.



The fourth example is the Heartwater Gap Analysis Workshop organized in Guadeloupe, October 2018. The report from this workshop (https://go.usa.gov/xVszZ) provides a comprehensive assessment of the current scientific knowledge and the available countermeasures to effectively control and mitigate the impact of a heartwater outbreak in new geographical areas, as well as support global control and eradication initiatives in heartwater-endemic countries. One of the key outcomes from this workshop was the establishment of a strategic collaboration between the ARS Animal Diseases Research Unit in Pullman, Washington, and CIRAD and CaribVET in the Caribbean.



The reports from gap analyses conducted by USDA-ARS provided research priorities and anticipated products that are expected from the research and now serve to help measure the national program's progress during the last 5 years in meeting the needs of animal producers, researchers, and action and regulatory agencies. The following list of anticipated products from the Action Plan is followed by the expected impact of the research and a sampling of relevant accomplishments.

Anticipated Products from the Action Plan:

- New solutions to prevent economic losses from foreign animal diseases in agricultural and wildlife species.
- Effective countermeasures to prevent and eliminate the threat of zoonotic diseases in agricultural and wildlife species.
- Scientific information to establish on-farm practices that will maximize "biosecurity" to protect farms from naturally or intentionally introduced pathogens that threaten food security, farm productivity, and the trade and export of agricultural products.
- Experimental animal disease models that will serve the veterinary and public health research communities to significantly shorten the timelines for developing breakthrough medicines and disease prevention tools.
- Integrated predictive modeling capability for emerging and/or intentionally released infectious diseases of animals and the collection of data to support these models.
- Novel detection systems and broad spectrum vaccines and biotherapeutics to counter the threat of emerging diseases or engineered biological weapons.
- Novel countermeasures against the natural or intentional introduction of agricultural threats, including new methods for detection, prevention, and characterization of high-consequence agents.
- New pathogens associated with emerging diseases identified.
- Predictors of emerging livestock diseases.
- Methods to rapidly detect and characterize the etiology of new and emerging diseases.
- Tools and expertise to control emerging diseases and rapidly implement intervention strategies to respond to new disease outbreaks.

Impact:

The biodefense research program yields scientific information on disease transmission, pathogenesis, and the discovery of veterinary medical countermeasures to enable the detection,

control and eradication of high consequence foreign animal diseases and/or new emerging diseases of animals and humans.

COMPONENT 1: SELECTED ACCOMPLISHMENTS

Problem Statement 1A: *Foreign Animal Diseases*

USDA Speeds Development of Bird Flu Vaccine

During December 2014-June 2015, the United States experienced a high pathogenicity avian influenza (HPAI) outbreak with devastating consequences for the poultry industry resulting in the death of over 45 million chickens and turkeys. ARS developed, in record time, an effective vaccine against the HPAI virus strains that caused the outbreak. Although these HPAI viruses were not the cause of any human deaths, concerns remain that these viruses could mutate and become more harmful. Vaccines constitute a critical veterinary medical countermeasure to respond to biological threats such as avian influenza viruses. Currently, no vaccines for HPAI are licensed or permitted in the United States. The use of HPAI vaccines is dependent on our ability to rapidly develop vaccines with good efficacy against the virus strains that are the cause of the disease outbreak. In response to the first detections of new HPAI viruses (H5N8 and H5N2) in wild waterfowl and captive raptors in the United States in December 2014, ARS refocused its entire team of scientists working on avian influenza research to the most imminent research needs to address the U.S. outbreak, including the rapid development of a vaccine for emergency use. Within weeks, scientists at the U.S. National Poultry Research Center, Athens, Georgia, rapidly engineered a vaccine (rg-H5 vaccine) using reverse genetics technology that matched the H5N2 and H5N8 HPAI viruses that were the cause of the disease outbreak. The power of the reverse genetics technology is twofold. First, it allowed the manipulation of the genes to change the high pathogenic hemagglutinin (HA) gene from its typical amino acids to having a sequence similar to Low Pathogenic Avian Influenza (LPAI) viruses. This allows the change of a HPAI virus into a LPAI virus for safe production without affecting the efficacy of the vaccine virus. The second factor is that it allowed the creation of a unique vaccine virus by swapping the HA gene with the one that matches the HA genes causing the disease outbreak. ARS with the support of the ARS Office of Technology Transfer and APHIS developed and implemented in record time a technology transfer plan that enabled the transfer of the rg-H5 vaccine to a commercial partner for development and production.

Scientific Publications:

Bertran K, Balzli C, Lee DH, Suarez DL, Kapczynski DR, Swayne DE. 2017. Protection of White Leghorn chickens by U.S. emergency H5 vaccination against clade 2.3.4.4 H5N2 high pathogenicity avian influenza virus. Vaccine. 35(46):6336-6344. doi: 10.1016/j.vaccine.2017.05.051. https://doi.org/10.1016/j.vaccine.2017.05.051

Kapczynski DR, Pantin-Jackwood MJ, Spackman E, Chrzastek K, Suarez DL, Swayne DE. 2017. Homologous and heterologous antigenic matched vaccines containing different H5 hemagglutinins provide variable protection of chickens from the 2014 U.S. H5N8 and H5N2 clade 2.3.4.4 highly pathogenic avian influenza viruses. Vaccine. 35(46):6345-6353. doi: 10.1016/j.vaccine.2017.04.042. https://doi.org/10.1016/j.vaccine.2017.04.042

Discovering Why Some Avian Influenza Vaccines Fail

Vaccination is critical in protecting birds, animals, and people in developing countries like Indonesia where the H5N1 highly pathogenic avian influenza virus has become endemic. Indonesia has implemented a vaccination program, but some of its commercial vaccines have failed to protect poultry. An international collaboration of government officials, regulators, and scientists—including a team from the U.S. National Poultry Research Center, Athens, Georgia, investigated outbreaks in Indonesian flocks that were vaccinated. The team evaluated 14 Indonesian licensed vaccines to identify the seed strain—type of virus—included in the vaccines and variant field viruses to find out why these vaccines were failing. The results of this study showed that 11 of the 14 vaccines contained the manufacturer's listed vaccine seed strains, but 3 vaccines contained different seed strains than the ones listed on the label. Scientists immunized chickens with each of the 14 vaccines and found that protection varied greatly. Tests showed some vaccines contained a lot of antigens and some had only a little. The antigen, a protein from the virus, allows birds to produce antibodies to build up immunity. Vaccinated birds were challenged with three different field viruses. All vaccines protected against one of the viruses, some protected against the second virus, and none protected against the third. This research demonstrated the need to evaluate vaccines often and replace vaccine seed strains with more effective ones as new field viruses emerge that are resistant to older vaccines. Improving vaccines and ensuring they work not only helps control avian influenza outbreaks, but also adds new vaccines to the U.S. emergency stockpile.

Scientific Publication:

Swayne DE, Suarez DL, Spackman E, Jadhao S, Dauphin G, Kim-Torchetti M, McGrane J, Weaver J, Daniels P, Wong F, Selleck P, Wiyono A, Indriani R, Yupiana Y, Sawitri Siregar E, Prajitno T, Smith D, Fouchier R. 2015. Antibody titer has positive predictive value for vaccine protection against challenge with natural antigenic-drift variants of H5N1 high-pathogenicity avian influenza viruses from Indonesia. J Virol. 89(7):3746-62.
https://doi.org/10.1128/JVI.00025-15

H9N2 Low Pathogenic Avian Influenza Viruses with Zoonotic Potential in Pakistan

Significant economic losses from bird mortality and decreased egg production have resulted from H9N2 low pathogenic avian influenza virus (LPAIV) infections in poultry across North Africa, the Middle East and Asia. This group of viruses has also caused sporadic infections in mammalian species, including human beings, and has been associated with some specific genetic changes that suggests increasing pandemic potential. The H9N2 LPAIVs have been endemic in Pakistani poultry since 1996, but no new viruses have been reported since 2010. Because novel genotypes of Pakistani H9N2 contain mammalian host-specific markers, recent surveillance is essential to better understand any continuing public health risk. ARS scientists at the U.S. National Poultry Research Center, Athens, Georgia, in collaboration with Pakistani scientists reported this year the characterization of four new H9N2 LPAIVs, three from 2015 and one from 2012. All of the viruses tested in this study belonged to viruses originating in the Middle East. Importantly, these viruses all contained mammalian host-specific markers, suggesting that Pakistan avian H9N2 viruses have the capacity to infect mammals. Continued active surveillance in poultry and mammals is needed to monitor the spread and understand the potential for zoonotic infections by these H9N2 LPAIVs.

Scientific Publication:
Lee DH, Swayne DE, Sharma P, Rehmani SF, Wajid A, Suarez DL, Afonso C. 2016. H9N2 low pathogenic avian influenza in Pakistan (2012-2015). Vet Rec Open.;3(1):e000171. doi: https://doi.org/10.1136/vetreco-2016-000171

Efficacy of Two Licensed H5 Vaccines Against Challenge with a 2015 United States H5N2 clade 2.3.4.4 Highly Pathogenic Avian Influenza Virus in Domestic Ducks
Highly pathogenic avian influenza (HPAI) clade 2.3.4.4 viruses caused a major outbreak in poultry in the United States in 2015. Although the outbreak was controlled, vaccines were considered as an alternative control method and new vaccines were approved and purchased by the U.S. National Veterinary Stockpile for emergency use. ARS researchers at the U.S. National Poultry Research Center, Athens, Georgia, evaluated the efficacy of two of these vaccines in protecting Pekin ducks against challenge with a H5N2 HPAI poultry isolate. A recombinant alphavirus-based vaccine and an inactivated adjuvanted reverse genetics vaccine (originally developed by ARS and transferred to a commercial partner for manufacturing and stockpiling) were used to immunize the ducks. Both vaccines, regardless of the vaccination strategy used, reduced or prevented disease and virus shedding after challenge. This information is important to prepare and control avian influenza in domestic ducks should another outbreak with H5N2 clade 2.3.4.4 occur in the United States.

Scientific Publication:
Pantin-Jackwood MJ, DeJesus E, Costa-Hurtado M, Smith D, Chrzastek K, Kapczynski DR, Suarez DL. 2019. Efficacy of Two Licensed Avian Influenza H5 Vaccines Against Challenge with a 2015 U.S. H5N2 clade 2.3.4.4 Highly Pathogenic Avian Influenza Virus in Domestic Ducks. Avian Dis. 63(1):90-96. doi: 10.1637/11895-050918-Reg.1. https://doi.org/10.1637/11895-050918-Reg.1

Newcastle Disease Virus that was the Cause of the 2018 California Outbreak is Similar to the Virus that Caused the Outbreak in 2002
Virulent Newcastle disease is a foreign animal disease of poultry and three outbreaks (1971-1974, 2002-2003, and 2018-2020) have occurred in the United States in the last century with the last outbreak currently ongoing in California. These outbreaks have cost several hundreds of millions of dollars to control. To better determine the risks presented by the current California virulent Newcastle disease virus, ARS researchers at the U.S. National Poultry Research Center, Athens, Georgia, conducted studies to compare the virulence and transmission dynamics of the current outbreak strain with the previous 2002 virus. It was concluded that the new Newcastle disease virus presents characteristics that are consistent with a highly virulent velogenic virus that is highly similar to the California 2002 virus. This study is important to researchers and response agencies as it provides key insights into the risk of the present outbreak and demonstrates that previous studies conducted by ARS researchers on the California 2002 virus is still valid.

Scientific Publication:
Dimitrov KM, Ferreira HL, Pantin-Jackwood MJ, Taylor TL, Goraichuk IV, Crossley BM, Killian ML, Bergeson NH, Torchetti MK, Afonso CL, Suarez DL. 2019. Pathogenicity and transmission of virulent Newcastle disease virus from the 2018-2019 California outbreak and related viruses in young and adult chickens. Virology. 2019 May;531:203-218. doi: 10.1016/j.virol.2019.03.010. https://doi.org/10.1016/j.virol.2019.03.010

African Swine Fever Virus Experimental Vaccine Confers Protection Against a Virulent Virus Challenge
African swine fever virus (ASFV) is the etiological agent of a contagious and often lethal disease of domestic pigs that has significant economic consequences for the swine industry. The control of African Swine Fever (ASF) has been hampered by the unavailability of vaccines. ASFV is one of the largest viruses known, and the function of the large majority of the viral genes are unknown. Experimental vaccines have been developed using genetically modified-live attenuated ASFV where viral genes were removed from the genome. However, to date, none of these viruses have proven to be fully attenuated or effective. ARS scientists at the Plum Island Animal Disease Center, Orient Point, New York, have engineered a recombinant virus by specifically deleting six genes thought to be associated with virulence. Studies conducted in pigs showed that when this recombinant virus was inoculated in pigs, the virus was completely attenuated and did not cause disease. Importantly, when these animals were subsequently exposed to highly virulent ASFV strain, no signs of the disease were observed. This is the first report demonstrating the role of these genes acting as independent determinants of ASFV virulence. Additionally, this is the first experimental vaccine reported to induce protection when challenged against this very virulent strain.

Scientific Publication:
O'Donnell V, Holinka LG, Krug PW, Gladue DP, Carlson J, Sanford B, Alfano M, Kramer E, Lu Z, Arzt J, Reese B, Carrillo C, Risatti GR, Borca MV. 2015. African Swine Fever Virus Georgia 2007 with a Deletion of Virulence-Associated Gene 9GL (B119L), when Administered at Low Doses, Leads to Virus Attenuation in Swine and Induces an Effective Protection against Homologous Challenge. J Virol. 89(16):8556-66. doi: 10.1128/JVI.00969-15. Epub 2015 Jun 10. https://jvi.asm.org/content/89/16/8556.short

Using Gene-Editing as a Tool to Engineer an African Swine Fever Vaccine
African swine fever (ASF) is a deadly disease causing near 100% mortality in swine, trade restrictions and significant economic losses globally. The current threat for an introduction of ASF into the United States has never been higher. Since the introduction of ASF into the Republic of Georgia in 2007, 16 countries have reported new ASF outbreaks, including Belgium and China in 2018. Currently there is no commercially available vaccine to prevent this devastating disease. African swine fever is a large and complex double stranded DNA virus. After more than 50 years of research, there are no efficient molecular tools available to help develop a safe and effective live recombinant ASF vaccine. ARS scientists at the Plum Island Animal Disease Center in Orient Point, New York, investigated the use of the "CRISPR-Cas9" gene-editing system as a potentially more robust and efficient system to produce live recombinant ASF viruses. Gene editing is a new type of genetic engineering in which DNA can

be directly inserted, deleted, modified or replaced in the genome of a living organism. Unlike early genetic engineering techniques that randomly inserts genetic material into a host genome, genome editing directs the modification to site specific location. Using the CRISPR-Cas9 system, a recombinant ASF virus was successfully developed in record time compared to the use of traditional genetic engineering techniques. These results demonstrate the potential advantage of using CRISPR/Cas9 over traditional methods and should significantly improve our ability to develop a first generation modified live ASF vaccine.

Scientific Publication:
Borca MV, Holinka-Patterson LG, Berggren K, Gladue, DP. 2018. CRISPR-Cas9, a tool to efficiently increase the development of recombinant African swine fever viruses. Scientific Reports. 8:3154 DOI:10.1038/s41598-018-21575-8. https://doi.org/10.1038/s41598-018-21575-8

New Vaccine to Fight Deadly African Disease in Pigs
African Swine Fever Virus (ASFV) is a deadly disease affecting swine, causing near 100% mortality, trade restrictions and significant economic losses globally. As its name indicates, the disease occurs in Africa. However, in 2007 the disease appeared in the Caucasus (Republic of Georgia), and has subsequently spread into Russia and the Ukraine. ASFV continues to spread and is now present in multiple countries of Eastern Europe, including Poland and most recently the Czech Republic, posing an imminent threat to the European and global swine industries. Currently there are no vaccines to protect swine against ASFV. Therefore, ARS scientists at the Plum Island Animal Disease Center, Orient, New York, have developed an improved live vaccine that is safe and can protect swine as early as 2 weeks post vaccination. This is the first experimental vaccine shown to induce early protection against ASFV in swine. This vaccine could be used globally to protect swine from this deadly disease and also safeguard the U.S. pork industry against the increasing incursions of this devastating disease.

Scientific Publication:
O'Donnell V, Risatti GR, Holinka-Patterson LG, Krug PW, Carlson J, Velazquez-Salinas L, Azzinaro P, Gladue DP, Borca MV. 2017. Simultaneous deletion of the 9GL and UK genes from the African swine fever virus Georgia 2007 isolate results in virus attenuation and may be a potential virus vaccine strain. Journal of Virology. 91(1):e01760-16.
https://doi.org/10.1128/JVI.01760-16

African Swine Fever (ASF) Candidate Vaccines Transferred to Industry for Research and Development
African swine fever was considered a disease of sub-Saharan Africa. However, in 2007, a very virulent viral strain of ASF was introduced into the Republic of Georgia. Subsequently, the virus (Georgia 2007) started to spread to the Russian Federation, reached the European Union in 2014, and in 2018, the disease reached the world's largest pig producer, China. Thus, the disease has conquered three continents over the last decade and ASF now has an unprecedented geographical scope and poses a major threat to the U.S. swine industry. There is no vaccine available for ASF and the control of the disease is strictly dependent on animal quarantine, biosecurity measures, and slaughter. This presents a major gap in the availability of veterinary medical countermeasures to effectively prevent, control, and eradicate an ASF outbreak. A notable accomplishment in 2019 was the successful development and transfer of ASF candidate vaccines

by ARS scientists to five pharmaceutical companies. Important milestones included the submission of patents to the U.S. Patent and Trademark Office, five U.S. Government Patent and Biological Material Licenses, and one Cooperative Research and Development Agreement (CRADA). These important milestones set the stage to begin the process of developing the first safe and effective commercial ASF vaccine.

Scientific Publications:
O'Donnell V, Holinka LG, Gladue DP, Sanford B, Krug PW, Lu X, Arzt J, Reese B, Carrillo C, Risatti GR, Borca MV. 2015. African swine fever virus Georgia isolate harboring deletions of MGF360 and MGF505 genes is attenuated in swine and confers protection against challenge with virulent parental virus. J Virol. 89:6048 –6056. doi:10.1128/JVI.00554-15. https://jvi.asm.org/content/89/11/6048.short

O'Donnell V, Holinka LG, Krug PW, Gladue DP, Carlson J, Sanford B, Alfano M, Kramer E, Lu Z, Arzt J, Reese B, Carrillo C, Risatti GR, Borca MV. 2015. African swine fever virus Georgia 2007 with a deletion of virulence-associated gene *9GL* (B119L), when administered at low doses, leads to virus attenuation in swine and induces an effective protection against homologous challenge. J Virol. 89:8556 –8566. doi:10.1128/JVI.00969-15. https://jvi.asm.org/content/89/16/8556.short

O'Donnell V, Risatti GR, Holinka-Patterson LG, Krug PW, Carlson J, Velazquez-Salinas L, Azzinaro P, Gladue DP, Borca MV. 2017. Simultaneous deletion of the 9GL and UK genes from the African swine fever virus Georgia 2007 isolate offers increased safety and protection against homologous challenge. Journal of Virol. 91(1):e01760-16. https://doi.org/10.1128/JVI.01760-16

News Announcements:
"USDA to grant license for an experimental African swine fever vaccine"
https://www.nationalhogfarmer.com/agenda/usda-grant-license-experimental-african-swine-fever-vaccine

"USDA to grant exclusive license to develop ASF vaccine"
https://www.meatpoultry.com/articles/20321-usda-to-grant-exclusive-license-for-asf-vaccine

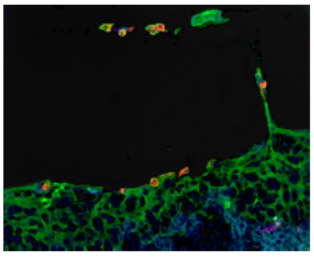
Recoding Classical Swine Fever Virus (CSFV) Structural Glycoprotein E2 and its Potential as a Vaccine Candidate
A DNA codon is a series of three nucleotides that codes for specific amino acids, which are the building blocks that comprise proteins. Some amino acids have more than one codon, resulting in what is called synonymous codon usage. Relative synonymous codon usage (RSCU) refers to the phenomenon during which some synonymous codons are used more often than others. ARS scientists at the Plum Island Animal Disease Center, Orient Point, New York, determined that the E2 glycoprotein, a determinant of virulence of the classical swine fever virus (CSFV), had a similar codon usage as pigs (*Sus scrofa*), its natural host. They explored the effect of just switching native codons in E2 for the less frequently used codons. Their studies showed that changing to synonymous codons in E2 resulted in the full attenuation of CSFV in pigs and demonstrating that the genetically-altered CSFV no longer caused disease. This phenomenon is thought to occur because changing the codon impacts the ability of the gene to be expressed.

Interestingly, when ARS scientists explored the potential use of this phenomenon to generate a vaccine strain, they found that the altered virus was able to protect animals against the disease. By using synonymous codons and not changing a single amino acid, this potential vaccine leaves all natural antigenic epitopes intact. The benefits of producing vaccines using this technology is that the antigenic profile of the virus remains intact, which is important for inducing a protective immune response. Additionally, by changing the nucleotide composition, genetic markers are now available that could be used to differentiate between vaccinated and infected animals.

Scientific Publication:
Velazquez-Salinas L, Risatti GR, Holinka LG, O'Donnell V, Carlson J, Alfano M, Rodriguez LL, Carrillo C, Gladue DP, Borca MV. 2016. Recoding structural glycoprotein E2 in classical swine fever virus (CSFV) produces complete virus attenuation in swine and protects infected animals against disease. Virology 494: 178-189. http://dx.doi.org/10.1016/j.virol.2016.04.007

Understanding the Mechanisms that Drive Persistent Infections in Foot-and-Mouth Disease Infected Cattle

Foot-and-Mouth Disease Virus (FMDV) is one of the most infectious disease of animals and impacts food security in much of the world. A key challenge in the control of FMDV is that 50 percent of vaccinated cattle become persistently infected. Persistent FMDV infection in cattle is largely responsible for the massive depopulation of animals because of the fear that these animals, although asymptomatic with no clinical signs, may spread FMDV and infect other animals. There is currently very little knowledge about the mechanisms that lead to this carrier stage. Tissues obtained post-mortem from cattle persistently infected with FMDV were analyzed by ARS scientists at the Plum Island Animal Disease Center, Orient Point, New York, to characterize the tissue-specific localization of FMDV and assess the expression of genes associated with the host immune response. Analysis of 28 distinct anatomic sites from 21 steers infected with FMDV had the highest prevalence of overall viral detection in the dorsal nasopharynx and dorsal soft palate. FMDV was less frequently detected in laryngeal mucosal tissues, oropharyngeal mucosal sites, and lymph nodes draining the pharynx. Within persistently infected mucosal tissues, FMDV antigens were rarely detectable within few epithelial cells in regions of mucosa-associated lymphoid tissue. Assessment of the genes associated with the host immune response of persistently infected pharyngeal tissues, indicated a general trend of decreased gene expression for 14 genes compared to uninfected control animals. Overall, this study demonstrated that during the FMDV carrier state in cattle, viral persistence is associated with epithelial cells of the nasopharynx in the upper respiratory tract and decreased levels of expression of several genes associated with the immune response in the infected tissues.



Shedding of FMDV from the naso-pharyngeal epithelium of asymptom-atic carrier cattle.

In another study, ARS scientists reported in the Journal of Virology new aspects of viral persistence and host mechanisms associated with chronic asymptomatic FMDV infection. One key finding was that the divergence between carrier animals and FMDV-free animals occurs as early as 10 days post infection. These findings provide new insights into paths that may be taken to develop vaccines that could prevent FMDV carrier animals.

Studying viral persistence of FMDV, ARS scientists investigated whether oral fluids from persistently infected cattle could transmit FMDV to cattle and pigs. Results from these studies showed that naïve cattle inoculated orally with fluids harvested from carrier animals developed clinical FMDV. In contrast, pigs exposed by inoculation of the same fluid samples harvested from the same persistently infected carrier cattle did not develop FMDV. These findings indicate that there is demonstrable contagion for cattle associated with FMDV carrier cattle. The results from this investigation provide new information that should improve response plans for FMDV control and eradication.

In a follow up study, ARS scientists investigated the potential for transmission of FMDV from persistently infected cattle to naive cattle under typical husbandry conditions. Results from a research collaboration with FMD scientists in Vietnam showed that no transmission occurred during six months of contact between FMD carrier and naïve cattle. These results will inform FMD response policy in the event of a large-scale outbreak. The average duration of the carrier state in this study was 27.7 months. The results of this study suggest that the duration of persistent infection in cattle may be longer than previously recognized, but the risk of transmission is low. Additionally, ARS evaluated one carrier animal for 12 months and fully sequenced the genome of seven viruses recovered from that animal during the study period. The analysis of the genome sequence of these seven viruses showed that a number of mutations occurred during the carrier stage. This was the first report of complete sequences of FMDV isolated from one persistently infected animal under natural conditions.

Scientific Publications:

Stenfeldt C, Eschbaumer, M, Rekant, SI, Pacheco, JM, Smoliga, GR, Hartwig, EJ, Rodriguez LL, Arzt J. 2016. The Foot-and-Mouth Disease Carrier State Divergence in Cattle. J Virol. 90(14):6344-64. https://doi.org/10.1128/JVI.00388-16

Pauszek SJ, Eschbaumer M, Brito B, Ferreira De Carvalho HC, Vu LT, Phuong NT, Hoang BH, Tho ND, Tung N, Thuy NT, Long NT, Dung DH, Rodriguez LL, Arzt J. 2016. Site-specific substitution (Q172R) in the VP1 protein of subclinical FMDV isolates collected from asymptomatic carrier ruminants in Vietnam. Virology Reports. 6:90-96. https://doi.org/10.1016/j.virep.2016.10.001

Stenfeldt C, Eschbaumer, M, Smoliga, GR, Rodriguez, LL, Zhu, J, Arzt J. 2017. Clearance of a persistent picornavirus infection is associated with enhanced pro-apoptotic and cellular immune responses. Sci Rep7(1):17800. DOI:10.1038/s41598-017-18112-4. https://doi.org/10.1038/s41598-017-18112-4

Brito B, Pauszek SJ, Eschbaumer M, Stenfeldt C, De Carvalho Ferreira H, Vu LT, Phuong NT, Hoang BH, Tho ND, Dong PV, Minh PQ, Long NT, Dung DH, Rodriguez LL, Arzt J. 2017. Phylodynamics of foot-and-mouth disease virus O/PanAsia in Vietnam 2010-2014. Veterinary Research. 48:24. https://doi.org/10.1186/s13567-017-0424-7

Arzt J, Belsham GJ, Lohse L, Botner A, Stenfeldt C. 2018. Transmission of foot-and-mouth disease from persistently infected carrier cattle to naive cattle via transfer of oropharyngeal fluid.

American Society for Microbiology mSphere 3:e00365-18.
http://doi.org/10.1128/mSphere.00365-18

Protecting livestock producers from the threat of foot-and-mouth disease
Foot-and-mouth disease (FMD), a highly contagious disease affecting cloven-hoofed animals such as cattle, pigs and small ruminants is considered a major global threat to animal agriculture. Although FMD was eradicated from the United States in 1929, it has been estimated that a reintroduction would result in $200 billion in lost revenue over 10 years. While FMD diagnostics and vaccines have been used effectively in controlling the disease, significant gaps remain in the availability of effective veterinary medical countermeasures suited for use in the United States. Therefore, ARS scientists working at the Plum Island Animal Disease Center (PIADC), Orient Point, New York, have dedicated resources and made significant breakthroughs in developing new improved veterinary countermeasures to detect, prevent, and control FMD should an incursion ever occur in the United States. The first is an attenuated vaccine platform called the "leaderless" FMDLL3B3D vaccine. This vaccine mimics the immune response of commercially available inactivated FMD vaccines, which are made with virulent wild type virus strains. But unlike commercial FMD vaccines, the FMDLL3B3D vaccine virus strains are fully attenuated; thus enabling their safe production in the United States. The importance of the vaccine is that its beneficial immune capability is created without the risk of potentially causing a devastating FMD outbreak should the vaccine virus escape from a manufacturing facility. Furthermore, the FMDLL3B3D vaccine virus platform was genetically-engineered with two negative markers to allow the differentiation of infected from vaccinated animals (DIVA). Such differentiation is paramount in the recovery phase of a disease outbreak to establish disease free status and obtain approval to resume the export of agricultural products. This FMD vaccine is now in the advanced development phase with a commercial partner, with approval from regulatory authorities for manufacturing and distribution expected within two years. The second breakthrough is a novel DIVA companion diagnostic test for the FMDLL3B3D vaccine that was developed through a consortium of academic, industry, and federal agencies comprised of APHIS, ARS, and U.S. Department of Homeland Security scientists working at the PIADC. This is the first licensed FMD diagnostic kit approved for manufacturing on the U.S. mainland. Together, the FMDLL3B3D vaccine and companion diagnostic test kit increase national security by providing animal health first responders with important new tools to mitigate the potentially catastrophic economic impact of an FMD outbreak.

Scientific Publication:
Chung CJ, Clavijo IL, Bounpheng MA, Uddowla S, Sayed A, Dancho B, Olesen IC, Pacheco J, Kamicker BJ, Brake DA, Bandaranayaka-Mudiyanselage CL, Lee SS, Rai DK, Rieder E. 2018. An improved, rapid competitive ELISA using a novel conserved 3B epitope for the detection of serum antibodies to foot-and-mouth disease virus. Journal of Veterinary Diagnostic Investigation. 30(5):699-707. https://doi.org/10.1177/1040638718779641

Study Reveals Pigs Can Transmit Foot-and-Mouth Disease Virus Prior to Signs of Sickness
Foot-and-mouth disease (FMD) virus spreads much more aggressively in pigs than previous research suggested, according to ARS scientists at the Plum Island Animal Disease Center, Orient Point, New York. The new study showed that pigs were able to infect other pigs just 24 hours after being themselves infected with the FMD virus, long before showing any clinical signs. Prior to this research, it was believed that FMD transmission in pigs did not occur before

visible signs of illness. Therefore, previous disease-dynamics models to predict disease impacts and estimate outbreak resource requirements did not account for the impact of preclinical transmission. Working with scientists at the Center for Epidemiology and Animal Health, USDA APHIS, ARS scientists used a mathematical modeling approach to estimate the impact of FMD preclinical transmission amongst pigs. With this new information, the models showed FMD outbreaks in the U.S. pig production sector would result in a 40-percent increase in the number of farms affected over previous estimates. That translates into 166 additional farms and more than 664,000 pigs euthanized compared to the previous models. Failure to account for information like this could make the difference between a limited, well-controlled FMD outbreak in the United States with a cost of $3 million over two months as opposed to a catastrophic nationwide epidemic with a cost of $20 billion over one year. Infectious disease modeling is a critical part of preparedness and protection of U.S. livestock. Research such as this provides critical information to help build better models to protect livestock industries from FMD.

Scientific Publications:
Stenfeldt C, Pacheco J, Brito B, Moreno-Torres K, Branan M, Delgado A, Rodriguez L, Arzt J. 2016. Transmission of foot-and-mouth disease virus during the incubation period in pigs. Frontiers in Veterinary Science 3(105). https://doi.org/10.3389/fvets.2016.00105

Arzt J, Branan M, Delgado A, Moreno-Torres K, Yadav, S, Stenfeldt C. 2019. Quantitative impacts of incubation phase transmission of foot-and-mouth disease virus. Scientific Reports. 9:2707. https://doi.org/10.1038/s41598-019-39029-0

Susceptibility of White-Tailed Deer to Rift Valley Fever Virus
Rift Valley fever virus (RVFV), a zoonotic arbovirus, poses major health threats to livestock and humans if introduced into the United States. Although domestic cattle, sheep, and goats are susceptible to RVFV and function as amplification hosts during epidemics, the potential role of wildlife species such as white-tailed deer is unknown. Since white-tailed deer are abundant throughout the country, there is concern they could also serve as an amplifying host and become a reservoir and source of infection for livestock and humans. ARS scientists at the Arthropod-borne Animal Diseases Research Unit (ABADRU), Manhattan, Kansas, in collaboration with Kansas State University scientists, investigated the susceptibility of deer to RVFV and confirmed their susceptibility to this virus. Importantly, infected deer developed hemorrhagic enteritis and bloody diarrhea, resulting in horizontal transmission to control animals. The results of this investigation provide evidence for a potentially major epidemiologic role for white-tailed deer if an outbreak of RVFV ever occurred in the United States.

Scientific Publication:
Wilson WC, Kim I, Trujillo J, Sunwoo S, Noronha LE, Urbaniak K, Mcvey DS, Drolet BS, Morozov I, Faburay B, Schirtzinger EE, Koopman T, Indran S, Balaraman V, Richt J. 2018. Susceptibility of White-Tailed Deer to Rift Valley Fever Virus. Emerging Infectious Diseases. 24(9):1705-1707. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6106403/

ARS Workforce Development: Creating the Next Generation of Biodefense Researchers
In fiscal year 2019, ARS created Workforce Development training agreements with the Auburn University, the University of Connecticut and the University of Minnesota. All three Universities

are new National Bio and Agro-defense Facility (NBAF) training partners with ARS. These agreements provided support for four new trainees in immunology/vaccinology, epidemiology and disease pathogenesis. The goal of this program is to provide a cadre of qualified scientists who will be competitive for NBAF positions when they become available. There are seven trainees currently in place at Mississippi State University and Kansas State University. ARS scientists are collaboratively involved in all of these research training projects. ARS held a research symposium for the workforce development trainees, and their USDA and University mentors in Manhattan, Kansas, on August 27, 2019, which provided trainees the opportunity to visit Manhattan, tour the NBAF and provide updates on their scientific projects as well as meet USDA leadership.

Reference:
National Bio and Agro-Defense Facility (NBAF) Workforce Development
https://www.ars.usda.gov/research/project/?accnNo=433706

Problem Statement 1B: *Emerging Diseases*

Global Migration of Influenza A Viruses in Swine

The emergence of the 2009 A/H1N1 pandemic virus underscores the importance of understanding how influenza A viruses evolve in swine on a global scale. To reveal the frequency, patterns and drivers of the spread of swine influenza virus globally, ARS scientists at the National Animal Disease Center, Ames, Iowa, conducted the largest genetic analysis of swine influenza viruses undertaken to date, integrating demographic and swine trade data. Using genetic and modeling approaches, ARS scientists demonstrated the importance of the asymmetrical global live swine trade on the evolution of influenza virus diversity. The size of a country's swine population was not found to be an important independent factor, as exemplified by China, which hosts the world's largest swine population but has relatively little outgoing swine trade and does not appear to be a major source of viral diversity in neighboring Asian countries or globally. Rather, Japan, Thailand, Vietnam, and South Korea independently import influenza viruses from Europe and North America via long-distance live swine trade. As an extension of these observed patterns, ARS scientists built a population simulation model for the global spread of swine influenza viruses that incorporated trade data and could estimate the likelihood of emergence of the H1N1 pandemic virus in swine in the years leading up to 2009. ARS scientists found that the evolution of swine influenza viruses is most likely to occur in East and South-East Asia. Knowledge of the global linkages between swine influenza virus populations has important implications for designing efficient surveillance strategies in resource-limited settings and predicting future disease threats.

Scientific Publications:
Lewis NS, Russell CA, Langat P, Anderson TK, Berger K, Bielejec F, Burke DF, Dudas G, Fonville JM, Fouchier RA, Kellam P, Koel BF, Lemey P, Nguyen T, Nuansrichy B, Peiris JM, Saito T, Simon G, Skepner E, Takemae N; ESNIP3 consortium, Webby RJ, Van Reeth K, Brookes SM, Larsen L, Watson SJ, Brown IH, Vincent AL. 2016. The global antigenic diversity of swine influenza A viruses. Elife. 5:e12217. doi:10.7554/eLife.12217.
https://doi.org/10.7554/eLife.12217.002

Walia RR, Anderson TK, Vincent AL. 2019. Regional patterns of genetic diversity in swine influenza A viruses in the United States from 2010 to 2016. Influenza Other Respir Viruses. 13(3):262-273. doi: 10.1111/irv.12559. https://doi.org/10.1111/irv.12559

Chang J, Anderson TK, Zeller MA, Gauger PC, Vincent AL. 2019. octoFLU: Automated Classification for the Evolutionary Origin of Influenza A Virus Gene Sequences Detected in U.S. Swine. Microbiol Resour Announc. 8(32). pii: e00673-19. doi: 10.1128/MRA.00673-19. https://doi.org/10.1128/MRA.00673-19

A New United States Swine Pathogen Database

In recent years, several deadly viral diseases of pigs have emerged in the United States causing hundreds of millions of dollars in economic damages. In order to effectively respond to these diseases or detect new disease incursions or viral variants, it is critical to have a database of currently circulating viral genetic sequences and associated tools to analyze the sequences. ARS scientists at the National Animal Disease Center, Ames, Iowa, created such a database (https://swinepathogendb.org) for porcine reproductive and respiratory syndrome virus, Senecavirus A, and porcine epidemic diarrhea virus using nucleotide sequences and related metadata found in GenBank, part of the United States National Center for Biotechnology Information, and from clinical cases detected by key veterinary laboratories. Presently, the South Dakota Animal Disease Research and Diagnostic Laboratory, the Iowa State Veterinary Diagnostic Laboratory and the Kansas State University Veterinary Diagnostic Laboratory have submitted over 2000 sequences. A suite of web-based tools allows stakeholders, researchers and veterinarians to quickly search for genetic sequence information, identify similar viruses, and browse virus genomes to inform research and control efforts. Databases such as these will greatly increase researchers' understanding of endemic circulating viruses as well as speed response efforts by quickly identifying new viral variants.

News Announcement:
"United States Swine Pathogen Database." Pork Checkoff Website
https://www.pork.org/research/united-states-swine-pathogen-database/

Presence of Vaccine-Derived Newcastle Disease Viruses in Wild Birds

Of all the factors influencing disease emergence, likely the most substantial is the loss of ecological species barriers, permitting opportunistic pathogens access to wildlife. ARS scientists at the U.S. National Poultry Research Center, Athens, Georgia, in collaboration with University of Georgia scientists demonstrated the repeated isolation of vaccine-derived Newcastle disease viruses from different species of wild birds across four continents from 1997 through 2014. The data indicate that at least 17 species from 10 avian orders occupying different habitats excrete vaccine-derived Newcastle disease viruses. Examining the extent of vaccine derived Newcastle disease virus from live vaccines, including recently developed recombinant vaccines, transferred from poultry into wild birds is crucial because the downstream epidemiological consequences of such spillovers are still unknown. Circulating live vaccine viruses present additional risks such as reversion of virulence and recombination with wild-type strains. In addition, the immune response of wild birds induced by infection with vaccine strains may provide selective pressures resulting in viral antigenic drift or increased virulence. The finding of live attenuated Newcastle disease virus vaccines in other avian species provides important evidence that the use of these

vaccines should be monitored to assess their potential impact on the environment and the emergence of new viral strains.

Scientific Publication:
Ferreira HL, Taylor TL, Dimitrov KM, Sabra M, Afonso CL, Suarez DL. 2019. Virulent Newcastle disease viruses from chicken origin are more pathogenic and transmissible to chickens than viruses normally maintained in wild birds. Vet Microbiol. 235:25-34. doi:10.1016/j.vetmic.2019.06.004. https://doi.org/10.1016/j.vetmic.2019.06.004

Publication of a Unified Classification and Naming System for New Emerging Newcastle Disease Viruses
The Newcastle disease virus genome is highly variable due to genetic mutations that results in viruses that are often unique to a country or region of the world. Several classification systems have previously been published that have established unique genotypes to describe these differences, but because of a lack of strict rules governing the establishment of new genotypes, a single system has not been widely adopted. ARS researchers at the U.S. National Poultry Research Center, Athens, Georgia, led an effort to establish a consortium of 23 reference and research laboratories to develop new and consistent naming rules to establish a consistent naming system. Of importance to the scientific community, this new nomenclature system is likely to become the *de facto* standard for genotype naming of new emerging Newcastle disease viruses, which will improve our ability to predict and understand the cause of future disease outbreaks.

Scientific Publication:
Dimitrov KM, Abolnik C, Afonso CL, Albina E, Bahl J, Berg M, Briand FX, Brown IH, Choi KS, Chvala I, Diel DG, Durr PA, Ferreira HL, Fusaro A, Gil P, Goujgoulova GV, Grund C, Hicks JT, Joannis TM, Torchetti MK, Kolosov S, Lambrecht B, Lewis NS, Liu H, Liu H, McCullough S, Miller PJ, Monne I, Muller CP, Munir M, Reischak D, Sabra M, Samal SK, Servan de Almeida R, Shittu I, Snoeck CJ, Suarez DL, Van Borm S, Wang Z, Wong FYK. 2019. Updated unified phylogenetic classification system and revised nomenclature for Newcastle disease virus. Infect Genet Evol. 74:103917. doi: 10.1016/j.meegid.2019.103917. https://doi.org/10.1016/j.meegid.2019.103917

Senecavirus A is One of the Causative Agents of Swine Vesicular Disease
Idiopathic vesicular disease in swine is a diagnosis made when none of the pathogens known to cause swine vesicular disease [vesicular exanthema virus, swine vesicular disease virus, vesicular stomatitis virus, and foot-and-mouth disease virus (FMDV)] have been detected in a clinical case. Occasionally, an emerging virus called Senecavirus A (SVA) has been detected in cases of idiopathic vesicular disease, raising the possibility that SVA infection could cause vesicular disease in swine. This assumption was strengthened by the recent emergence of idiopathic vesicular disease in Brazil and the United States from which there was frequent detection of SVA. For the first time, ARS scientists at the National Animal Disease Center, Ames, Iowa, in collaboration with scientists at Iowa State University have been able to experimentally induce clinical signs and gross lesions in nursery age pigs inoculated with SVA, demonstrating a causative relationship between SVA infection and vesicular disease in susceptible pigs. This is noteworthy because SVA disease is clinically indistinguishable from

other vesicular diseases of swine, especially FMDV, a highly transmissible livestock disease that can cause devastating economic losses to farmers. Developing animal disease models is key to animal health research to understand pathogenicity and disease transmission and test experimental vaccines that could be used to prevent the disease.

Scientific Publication:
Montiel N, Buckley A, Guo B, Kulshreshtha V, VanGeelen A, Hoang H, Rademacher C, Yoon KJ, Lager K. 2016. Vesicular Disease in 9-Week-Old Pigs Experimentally Infected with Senecavirus A. Emerg Infect Dis. 22(7):1246-1248. doi: 10.3201/eid2207.151863. https://dx.doi.org/10.3201/eid2207.151863

Early Warning Strategies for Vector-borne Animal Disease Outbreaks
Vesicular stomatitis virus (VSV) is a vector-borne animal pathogen that causes lesions and other symptoms that are clinically indistinguishable from Foot-and-Mouth Disease and is one of the most common vesicular diseases affecting livestock (domestic horses, cattle, pigs) throughout the Americas. VSV is also zoonotic and can cause mild flu symptoms in people. Despite numerous epidemiological studies, there is currently limited understanding of the factors responsible for the VSV outbreaks that have occurred in the United States every decade since 1916. VSV is an excellent research model for understanding and potentially predicting vector-borne animal disease outbreaks because it is complex and there is a lack of understanding of ecological patterns, or the role of environmental factors that may contribute to disease outbreaks through time at local and regional scales. Therefore, scientists with diverse scientific expertise from five locations across the United States (Plum Island Animal Disease Center, Orient Point, New York; Arthropod-Animal Disease Center, Manhattan, Kansas; Jornada Experimental Range Unit, Las Cruces, New Mexico; Rangeland Resources and Systems Research Unit, Cheyenne, Wyoming; Center for Epidemiology and Animal Health, Fort Collins, Colorado), collaborated on developing early warning strategies for VSV. Coupling big data-model integration with human and machine learning, ARS scientists evaluated the relative importance of a large and diverse suite of environmental, insect and livestock variables to patterns in VSV disease outbreaks. Given information on latitude, elevation, and long-term precipitation, veterinarians and livestock owners should now be able to monitor their local conditions to determine the likelihood that VSV will occur in each month of the year. These early VSV warning strategies could also inform public health preparedness and response to other emerging zoonotic vector-borne diseases.

Scientific Publication:
Peters DPC, McVey DS, Elias EH, Pelzel-McCluskey AM, Derner JD, Burruss ND, Shrader TS, Yao J, Pauszek SJ, Lombard J, Rodriguez LL. 2017. Coupling big data-model integration with human and machine learning to develop early warning strategies for vector-borne diseases. Submitted to the journal Science.

Identification of Avian Influenza Epidemiological Risk Factors of Wild Birds in Mexico
The risk of an introduction of avian influenza in North America by infected wild birds is significant. To identify epidemiological risk factors between poultry and synanthropic birds (wild birds living close to humans and farms), ARS researchers at the U.S. National Poultry Research Center, Athens, Georgia, conducted field studies in collaboration with a team of scientists from Mexico. These studies evaluated the risk that synanthropic birds bring to poultry

farms in a highly densely productive area of Mexico. The Altos de Jalisco region in west central Mexico is the location of the largest concentration of poultry farms. Recently, this region witnessed the emergence of low pathogenic H5N2 and the highly pathogenic H7N3 influenza viruses. As a result of these field studies in Mexico, 82 species of wild birds were identified, with some of these species linked to poultry farms. The highest ranked species corresponded to the Mexican Great-tailed Grackle and the Barn Swallow; making those potential hosts for disease transmission of pathogens in the wild bird-poultry interface in the region of Jalisco. The ability to demonstrate epidemiological connections between wildlife and poultry is important to understand the risk to the poultry industry from wild birds. Because of the proximity with the United States, these Mexican regions present a high risk of introduction of avian influenza through trade, wild birds and illegal transport of birds, and therefore pose a significant threat to the U.S. poultry industry.

Scientific Publication:
Valdez-Gomez H, Navarro-Lopez R, Vazques-Mendoza L, Zalapa-Hernandez M, Guerrero-Hernandex I, Fonseca-Delgado V, Marquez-Ruiz M, Afonso CL. 2017. Risk factors for the transmission of infectious diseases agents at the wild birds-commercial birds interface. A pilot study in the region of the Altos de Jalisco, Mexico. BULLETIN DE L'ACADEMIE VETERINAIRE DE FRANCE.170(2):143-150. https://doi.org/10.4267/2042/62332

Epizootic Hemorrhagic Disease Virus (EHDV) Infection Affects Sensory and Neural Tissues in Culicoides Midges

Epizootic Hemorrhagic disease is an emerging disease caused by a virus transmitted by infected insects commonly referred to as midges that transmit the virus when biting animals resulting in significant economic losses in the captive deer industry. This virus has also infected cattle during disease outbreaks resulting in additional economic losses to the cattle industry. With collaborators at Clemson University and University of South Carolina, ARS scientists at the Arthropod-borne Animal Diseases Research Unit, Manhattan, Kansas, identified key changes in female midge gene expression profiles occurring during early infection with EHDV. Genes that had decreased expression included those for sensory functions (especially vision), behavior, learning, and memory. Genes that had increased expression included those for immune processes, odor, and light detection. These results suggest that EHDV infection may have a significant effect on sensory and neural tissues of midges and suggest a change needed in the use of lamps that use light wavelength to optimally attract and trap infected insects during outbreaks, which is important to researchers as well as industries interested in trapping the insects as a means of disease control.

Scientific Publication:
Nayduch D, Shankar V, Mills MK, Robl T, Drolet BS, Ruder MG, Scully ED, Saski CA. 2019. Transcriptome Response of Female *Culicoides sonorensis* Biting Midges (Diptera: Ceratopogonidae) to Early Infection with Epizootic Hemorrhagic Disease Virus (EHDV-2). Viruses. 11(5). pii: E473. doi: 10.3390/v11050473. https://doi.org/10.3390/v11050473

Component 2: Antimicrobial Resistance

Rationale for the research:
Antibiotics are one of the most important medical discoveries of the 20th century and will remain an essential tool for treating animal and human diseases in the 21st century. However, antimicrobial resistance among bacterial pathogens and concerns over the prudent use of antibiotics in animals has garnered global attention. Importantly, the availability of effective medical interventions to prevent and control animal diseases on the farm is likely to impact global food security. Accordingly, more attention needs to be given to understanding the drivers of antimicrobial resistance in farm animals and the discovery of novel technologies that can provide alternatives to antibiotics.

USDA is an active participant in the President's Task Force for Combating Antibiotic Resistant Bacteria (CARB) and fully supports the new initiatives to address antimicrobial resistance announced on September 18, 2014. On November 25, 2014, the USDA released its Antimicrobial Resistance Action Plan (http://www.usda.gov/documents/usda-antimicrobial-resistance-action-plan.pdf). This action plan describes how USDA proposes to obtain and disseminate science-based, actionable, quantitative information about antibiotic drug use and the development of resistance in food-producing animals and their relationship to livestock management practices. USDA is currently working with the interagency to develop the next 2020-2025 version of the CARB National Action Plan. USDA further proposes to address knowledge gaps and develop effective, practical mitigation strategies that will help prolong the effectiveness of existing antibiotics, including the development of alternatives to antibiotics to prevent and treat animal diseases. Additionally, during this reporting period ARS worked with OIE to plan and develop the 3rd International Symposium on Alternatives to Antibiotics, https://www.ars.usda.gov/alternativestoantibiotics/events.html, which brought together researchers and industry from all over the world to discuss on-going research and challenges associated with the development of ATAs.

Stakeholders representing the livestock and poultry industries that responded to the 2015 ARS animal health national survey identified research on antimicrobial resistance bacteria as a national priority.

Research Needs:
There is a need to investigate microbial ecology to decipher the mechanisms that promote antimicrobial resistance in animal production. New integrated strategies are needed to reduce and mitigate antimicrobial resistance resulting from the use of antibiotics to treat respiratory and enteric diseases. One of the priority research areas includes the causes and conditions that induce the increased development of antimicrobial resistance in production animals.

Anticipated Products from the Action Plan:
- Scientific information to decipher the role of the respiratory microbiome in respiratory diseases, including pathogens that are refractory to antimicrobials.
- Scientific information to determine the role of the gut microbiome in enteric diseases,

including the effect of external stressors such as management, feeding practices, environment, transport, and the administration of antimicrobials.
- Scientific information to decipher the microbial composition of the gastrointestinal tract and the mechanisms by which commensal microbial species enhance health and mitigate diseases of livestock and poultry.
- Scientific information to determine the correlation between subclinical infections caused by certain enteric pathogens and feed efficiency.
- Highly effective vaccines that could reduce the use of antibiotics in animal agriculture.
- New biotherapeutic platforms based on protective host proteins to induce and supplement an animal's innate immune response.
- Alternatives to antibiotics with defined mechanisms of action to provide new opportunities for the selection of multiple products that can work synergistically.
- Alternatives to antibiotics that affect the gut microbiome, such as phytochemicals and immune enhancers to provide new opportunities for integrating nutrition, health, and disease research.
- Probiotics that enhance immune development and resistance to enteric pathogens at mucosal surfaces.
- Feed additives and micronutrients that can improve feed efficiency, disease resistance, and health by promoting or inhibiting the growth of specific enteric microorganisms.
- Validated preventive health management programs derived from the re-engineering of gut microbiomes using specially designed feed rations and diets.

Impact:
Understanding the ecology of antimicrobial resistance is critical for developing strategies to minimize the level of antimicrobial resistance in production animals and informs the development of alternatives to antibiotics to prevent or treat respiratory and enteric diseases of livestock and poultry. Developing highly efficacious antibiotic alternatives such as vaccines, phytochemicals, microbial-derived products, immune-related products, and innovative drugs, chemicals, and enzymes to prevent and treat infectious diseases could reduce the need for medically important antibiotics in animal agriculture and strengthen our ability to develop health management strategies that are less reliant on antimicrobial compounds.

COMPONENT 2: SELECTED ACCOMPLISHMENTS

Problem Statement 2A: *Ecology of Antimicrobial Resistance*

Antimicrobial Resistance Distribution Differs Among Methicillin-resistant Staphylococcus aureus (MRSA) Sequence Type (ST) 5 Isolates from Health Care and Agricultural Sources
Antimicrobial resistance is an expanding public health concern and MRSA is a notable example. Since the discovery of livestock associated MRSA (LA-MRSA), public health concerns have arisen surrounding the potential of LA-MRSA isolates to serve as a reservoir for antimicrobial resistance determinants in people. ARS researchers at the National Animal Disease Center, Ames, Iowa, compared swine associated LA-MRSA ST5 and human clinical MRSA ST5 isolates for antimicrobial susceptibilities and genes associated with antimicrobial resistance. Swine associated LA-MRSA ST5 isolates exhibited resistance to fewer antibiotics than clinical MRSA ST5 isolates from humans with no swine contact. Distinct genomic antimicrobial resistance

elements were harbored by each subgroup, with little overlap in shared antimicrobial resistance genes between swine associated LA-MRSA ST5 and clinical MRSA ST5 isolates. Our results demonstrate that antimicrobial susceptibilities and genes that encode antimicrobial resistance among swine associated LA-MRSA ST5 and clinical MRSA ST5 isolates are separate and distinct suggesting that swine do not play a major role in maintaining a MRSA ST5 reservoir for humans.

Scientific Publications:
Hau SJ, Haan JS, Davies PR, Frana T, Nicholson TL. 2018. Antimicrobial resistance distribution differs among methicillin resistant Staphylococcus aureus sequence type (ST) 5 isolates from health care and agricultural sources. Frontiers in Microbiology. 9:2102. https://doi.org/10.3389/fmicb.2018.02102

Hau SJ, Allué-Guardia A, Rusconi B, Haan JS, Davies PR, Frana TS, Eppinger M, Nicholson TL. 2018. Single nucleotide polymorphism analysis indicates genetic distinction and reduced diversity of swine-associated methicillin resistant Staphylococcus aureus (MRSA) ST5 isolates compared to clinical MRSA ST5 isolates. Frontiers in Microbiology. 9:2078. https://doi.org/10.3389/fmicb.2018.02078

Effect of Zinc in the Selection of Methicillin-resistant Staphylococcus aureus (MRSA)
Feeding zinc to young pigs to prevent diarrhea is a common practice in the swine industry. But public health concerns over this practice have increased because using zinc supplements have been partly attributed to the occurrence of livestock-associated methicillin-resistant *Staphylococcus aureus* (LA-MRSA) strains in Europe. Therefore, ARS scientists at the National Animal Disease Center, Ames, Iowa, along with collaborators at Iowa State University and University of Minnesota, compared the prevalence of zinc resistance genes in U.S. swine LA-MRSA isolates with their prevalence in MRSA isolates from humans with no swine contact. None of the swine MRSA isolates were resistant to zinc. In contrast, 21.9 percent of isolates from humans with no swine contact carried the zinc resistance gene, and 24.7 percent were resistant to zinc. These results suggest that the application of zinc in feed, has not led to an increase in zinc resistant MRSA isolates in swine populations, and that these LA MRSA are not the source of zinc resistant MRSA in humans.

Scientific Publication:
Hau SJ, Frana TS, Sun J, Davies PR, Nicholson TL. 2017. Zinc resistance within swine associated methicillin resistant Staphylococcus aureus (MRSA) isolates in the USA is associated with Multilocus Sequence Type Lineage. Applied and Environmental Microbiology. pii: AEM.00756-17. doi: 10.1128/AEM.00756-17. https://doi.org/10.1128/aem.00756-17

Identification of Co-infecting Bacterial Agents in Birds Infected with Newcastle Disease Viruses
Newcastle disease viruses are the most prevalent cause of respiratory disease on poultry farms worldwide, including the United States. To understand causes of Newcastle disease vaccine failure in the field, ARS researchers at the U.S. National Poultry Research Center, Athens, Georgia, conducted random sequencing of nucleic acids obtained from poultry samples isolated in endemic countries. The outcome has been the identification of previously unrecognized

infections with bacteria from the genus *Ochrobactrum*. The bacteria were isolated and determined to contain antibiotic resistance genes. The complete draft genome of a total of eight *Ochrobactrum* species bacteria has been published in two articles. The results of these studies demonstrated that Newcastle disease infection is often accompanied by bacterial infections, and in these cases, multidrug-resistant novel *Ochrobactrum* species strains were isolated from a pigeon, a duck, and chickens. This work is important because it demonstrates that additional factors may be associated with Newcastle disease in the field. Importantly, the discovery of previously unrecognized co-infections with novel multidrug-resistant bacteria highlights the importance of effective Newcastle disease vaccination programs to potentially prevent secondary bacterial infections that may lead to the use of antibiotics and the selection of the antimicrobial resistant bacteria.

Scientific Publication:
Sharma P, Killmaster, LF, Volkering JD, Cardenas-Garcia S, Wajid A, Rehmani SF, Basharatd A, Miller PJ, Afonso CL. 2018. Draft genome sequence of three novel Ochrobactrum spp. isolated from different avian hosts in Pakistan. Genome Announcements. 6:e00269-18. https://doi.org/10.1128/genomeA.00269-18

Antibiotics Alter the Normal Swine Respiratory Microbial Community
Antibiotic stewardship is of the utmost importance to improve animal health outcomes and prevent selection of antimicrobial resistance. There is increasing evidence of the important role the normal respiratory microbial community (microbiota) plays in shaping immune and respiratory health. However, there is little knowledge on the effects of antibiotics on the swine respiratory microbiota. Oxytetracycline is a broad-spectrum antibiotic that is used for treatment of bacterial respiratory disease in swine. ARS researchers at the National Animal Disease Center, Ames, Iowa, showed the respiratory microbiota diversity decreased in response to oxytetracycline administration. In addition, giving the antibiotic in the feed had a greater and longer lasting impact on the nasal microbiota than the injectable route. There were also increased abundances of some pathogenic bacteria and decreased abundances of normal respiratory resident bacteria after antibiotic treatment. These results highlight the need to further assess how these changes can ultimately affect the animal's respiratory health and risk to disease.

Scientific Publication:
Mou KT, Allen HK, Alt DP, Trachsel J, Hau SJ, Coetzee JF, Holman DB, Kellner S, Loving CL, Brockmeier SL. 2019. Shifts in the nasal microbiota of swine in response to different dosing regimens of oxytetracycline administration. Vet Microbiol, 237, 108386 Oct 2019: PMID: 31526488 DOI: 10.1016/j.vetmic.2019.108386. https://doi.org/10.1016/j.vetmic.2019.108386

Subinhibitory Concentrations of Antibiotics Commonly Used to Treat Swine Increase *Streptococcus suis* (*S. suis*) biofilm formation
S. suis is the leading bacterial swine pathogen worldwide causing a wide variety of clinical presentations in pigs ranging from asymptomatic carriage to lethal systemic disease. *S. suis* is also a zoonotic pathogen capable of causing invasive disease in humans. A barrier towards the development of improved vaccines or interventions for *S. suis* infections is a gap in understanding the factors contributing to persistence in the host, in which colonized pigs continue to shed and transmit *S. suis*. Biofilms are adherent communities of bacteria that are

protected from clearance mechanisms and are considered a key factor contributing to chronic or persistent bacterial infections. Routine management practices involve treating all pigs in the same pen or barn with appropriate antibiotics upon observing any pig exhibiting clinical signs associated with a bacterial infection. ARS researchers at the National Animal Disease Center, Ames, Iowa, studied the effects of sub-inhibitory concentrations of antibiotics commonly used by the swine industry on the development of *S. suis* biofilms and found amoxicillin, lincomycin, and oxytetracycline increase biofilm formation while bacitracin, carbadox, chlortetracycline, enrofloxacin, gentamicin, neomycin, sulfadimethoxine, tiamulin, and tylosin did not. Collectively, our data demonstrate that exposure to some commonly used antibiotics contributes to increased biofilm formation of *S. suis*, thereby potentially increasing survival and persistence within the respiratory tract of swine and increasing transmission dynamics among animals. This data is critical for proper selection of antibiotics for successful treatment of swine bacterial diseases while minimizing potential collateral consequences.

Scientific Publication:
Waack U, Nicholson TL. 2018. Subinhibitory concentrations of amoxicillin, lincomycin, and oxytetracycline commonly used to treat swine increase Streptococcus suis biofilm formation. Frontiers in Microbiology. 9:2707. https://doi.org/10.3389/fmicb.2018.02707

Problem Statement 2B: *Alternatives to Antibiotics*

Using Genomics to Identify Novel Antimicrobials
There is a critical need in animal agriculture to develop novel antimicrobials and alternative strategies that will help to reduce the use of antibiotics and address the challenges of antimicrobial resistance. High-throughput gene expression analysis is providing new tools that are enabling the discovery of host-derived antimicrobial peptides. One example are the NK-lysins that were first described in mammals and are also found in avian species, which have been shown to have antimicrobial activities that could potentially be used to control important poultry pathogens. ARS scientists at the Animal Biosciences and Biotechnology Laboratory, Beltsville, Maryland, demonstrated alterations following chicken NK-lysin binding to coccidia and *Escherichia coli* membranes, indicating damage and disruption of cell membranes, suggesting that NK-lysin kills pathogenic protozoans and bacteria by direct interaction. DNA analysis revealed that chicken NK-lysin peptides derived from certain genes were more effective at killing pathogens than those derived from other genes, which could potentially result in certain genetic lines of poultry being more resistant to diseases. Although these host-derived antimicrobial peptides may not, by themselves, be able to replace the antibiotics currently used in animal production, their use as specific treatments based on their known mechanisms of action is showing promising results.

Scientific Publication:
Kim WH, Lillehoj HS, Gay CG. 2016. Using genomics to identify novel antimicrobials. Rev Sci Tech. 35(1):95-103. doi: 10.20506/rst.35.1.2420. Review.
https://europepmc.org/article/med/27217171

Direct-fed Microbials as an Antibiotic Alternative

With an increase in concerns regarding the development of antibiotic resistance and efforts to promote the judicious use of antibiotics in food-producing animals, there is a timely need for the development of viable alternatives to ensure and maintain optimal animal health and performance. Direct-fed microbials (DFMs), often referred to as probiotics, are a potential non-antibiotic replacement that has been studied extensively and used in commercial applications. DFMs are beneficial bacteria and often used as a feed supplement to promote gut health. Therefore, to better understand the health benefits of probiotics in enhancing gut health in poultry, and the mechanisms used by the non-pathogenic probiotic bacteria *Bacillus subtilis*, ARS scientists at the Animal Biosciences and Biotechnology Laboratory in Beltsville, Maryland, carried out extensive animal studies to show that certain Bacillus strains stimulate host innate immune responses, decrease harmful inflammatory responses, and promote gut integrity when used as a feed additive in young chickens. These results provide scientific evidence for the beneficial effects of probiotic bacteria and the potential use of some strains of *Bacillus subtilis* as a feed additive to promote gut health in commercial poultry production and reduce the use of medically important antibiotics.

Scientific Publications:

Lee K, Lillehoj HS. 2016. An update on direct-fed microbials in broiler chickens in post-antibiotic era. Animal Production Science. doi.org/10.1071/AN15666.
https://doi.org/10.1071/AN15666

Gadde U, Kim W, Oh S, Lillehoj HS. 2017. Alternatives-to-antibiotics for maximizing the efficiency of growth performance and feed efficiency in poultry: a review. Animal Health Research Reviews. 9:1-20. doi: 10.1017/S1466252316000207.
https://doi.org/10.1017/S1466252316000207

Enhanced Understanding of Mechanisms of Action of Antibiotic Growth Promoters

Although antibiotic growth promoters (AGPs) have been widely used globally to make a significant contribution to the expansion of modern animal agriculture, underlying modes of action of AGPs still remain unknown. In 2017, the FDA released the Veterinary Feed Directive that reduced the ability of producers to use growth performance-promoting antibiotics making it more imperative to understand how AGPs work so rationally designed antibiotic alternatives can be developed and produced to prevent and control animal diseases. ARS scientists at the Animal Biosciences and Biotechnology Laboratory, Beltsville, Maryland, used “omics” technologies to identify biochemical pathways that are associated with the effects of AGPs. The changes in the levels of intestinal small molecular weight metabolites provided distinctive biochemical signatures unique to each antibiotic-supplemented group. These biochemical signatures were characterized by increases in the levels of metabolites of amino acids, fatty acids, nucleosides and vitamins. These results enhance our understanding of the mode of AGP action and provide the framework for future studies to identify natural chemical compounds as antibiotic alternatives to improve poultry growth performance without the use of in-feed antibiotics.

Scientific Publication:
Grant A, Gay CG, Lillehoj HS. 2018. Bacillus spp. as direct-fed microbial antibiotic alternatives to enhance growth, immunity, and gut health in poultry. Avian Pathol.;47(4):339-351. doi: 10.1080/03079457.2018.1464117. https://doi.org/10.1080/03079457.2018.1464117

The Use of Immunomodulators as an Alternative to Antibiotic Use in Swine
The use of immunomodulators is a promising alternative to the use of antibiotics to prevent and combat infectious disease. Previously ARS researchers at the National Animal Disease Center, Ames, Iowa, demonstrated that giving an immunomodulatory protein to pigs elicited a sustained increase in circulating neutrophils, a type of white blood cell that is beneficial in preventing bacterial diseases. In new studies, pigs given the protein had an improved outcome when infected with *Streptococcus suis*, the leading cause of meningitis in weaned pigs. Thus, the use of this immunomodulatory protein in pigs to induce an increase in circulating neutrophil numbers may be a useful alternative to antibiotics for prevention of Streptococcal and other bacterial diseases, especially during times of stress and pathogen exposure such as post-weaning

Scientific Publication:
Brockmeier S, Loving CL, Eberle KC, Hau SJ, Mou KT, Kehrli Jr. M.E. 2019. Administration of granulocyte-colony stimulating factor (G-CSF) to pigs results in a longer mean survival time after exposure to Streptococcus suis. Veterinary Microbiology. 231:116-119. https://doi.org/10.1016/j.vetmic.2019.03.010

Component 3: Zoonotic Diseases

Rationale for the research:

Zoonotic diseases are infectious diseases transmitted from animals to man and represent one of the leading causes of illness and death in people world-wide. Seventy-five percent of emerging diseases are zoonotic. In developing countries, zoonotic diseases stand out as the most prevalent and important threat to public health. Zoonoses also have a negative impact on commerce, travel, and economies worldwide. In industrialized nations, zoonotic diseases are of particular concern to the agricultural sector. Priority diseases include those that are especially difficult to diagnose and cause substantial morbidity and mortality, resulting in significant economic costs to producers when they persist or reemerge. Over the years, USDA has invested significant resources in attempts to eradicate endemic zoonoses from livestock populations in the United States (e.g., brucellosis and tuberculosis). However, their persistence in wildlife reservoirs continues to pose challenges. Moreover, some zoonotic agents have been identified as having the potential to be used for bioterrorism. Because many determinants of zoonotic diseases lie outside the purview of the health sector, agriculture and the animal health community must play an important role in preventing these diseases from propagating in domestic animals, starting with proper surveillance systems. Effective countermeasures are needed to eliminate zoonotic agents at their animal source and protect our Nation from these important public health threats.

The ARS zoonotic bacterial diseases research program focuses on brucellosis, leptospirosis, Q-fever, and tuberculosis with the strategic goal of developing countermeasures to prevent disease transmission in domestic livestock and wildlife reservoir hosts. Zoonotic viral diseases that pose a significant threat to the Nation (e.g., avian influenza, Rift Valley fever) and are exotic to the United States are addressed under Component 1: Biodefense Research. Additional zoonotic diseases are addressed under Component 6, Problem Statement 6B: (Hemoparasitic Diseases) and Component 7: (Transmissible Spongiform Encephalopathies).

Stakeholders representing the livestock industries that responded to the 2015 ARS animal health national survey identified research to prevent and mitigate the impact of zoonotic bacteria a national priority.

Research needs:

Brucella

Research is needed to create new vaccines and diagnostic assays to detect and control infection and transmission with specific emphasis on diagnostic assays that are specifically designed for the control and eradication of brucellosis in wildlife. Genomic analyses of *Brucella* species are needed to identify unique sequences to further the development of serological diagnostic tests that can differentiate *Brucella* species without culturing. New vaccine strategies will include developing oral vaccines that would be economical and reliable for use in wildlife reservoirs.

Leptospira

There is a need to identify and characterize emerging spirochete strains associated with field outbreaks, to determine how these bacteria interact and evade host responses during infection,

and to determine mechanisms of protective immunity. Comparative genomics studies of *Leptospira borgpetersenii* serovar *hardjo* (the most prevalent serovar of cattle) and other *Leptospira* species are needed to identify unique sequences to support diagnostic and vaccine discovery research programs. In order to design vaccines that are effective in reservoir hosts, there is a need to determine *Leptospira* gene expression changes in different hosts. There is also a need to assess mutant strains with defined phenotypic characteristics and analyze how these mutants interact with the host and alter global patterns of gene expression. Diagnostic tools are needed to support molecular epidemiology studies to understand the ecology of *Leptospira* species and the emergence of new serovars.

Mycobacterium bovis

There is a need for improved vaccines and diagnostics for *Mycobacterium bovis*, the causative agent of bovine tuberculosis. Research is needed to identify microbial immunogens critical for the induction of protective immunity and there is a need to sequence the genome of common environmental *Mycobacteria* to develop assays that can differentiate them from pathogenic *Mycobacteria* to eliminate false positive test results. There is a need to characterize *Mycobacterium bovis* infections, pathogenesis, and immune responses in domestic livestock and relevant wildlife reservoir hosts.

Coxiella

There is a need to develop new technologies for diagnosing and mitigating the risk of *Coxiella burnetii* transmission from ruminant livestock that are effective, economically feasible and ecologically responsible.

Anticipated Products from the Action Plan:

- Scientific information to differentiate *Brucella* species by determining the unique sequences associated with phenotypic variations in virulence, host range, and persistent infections that will support diagnostic and vaccine discovery research initiatives.
- Determining the pathogenesis of *Brucella* species to identify mechanisms of persistent infections, host tolerance, and protective immunity.
- Diagnostic and intervention strategies for brucellosis in bison, elk, and feral swine that will enable control and potentially eradication in the wildlife reservoir.
- Diagnostic and intervention strategies for *Brucella melitensis* that will ensure a safe food supply and goat and sheep health.
- New vaccine platforms, with an emphasis on distance and oral delivery, designed to control and eradicate brucellosis in bison, elk, and feral swine.
- In vitro disease models consisting of host cell cultures that will lead to the molecular characterization of host-bacterial interactions, variations in gene expression, and associated pathogenic mechanisms.
- Scientific information on the protective immune responses to spirochete antigens in large and small animal disease models.
- Large-scale sequence analyses to characterize the genome of selected spirochetes and identify strain-specific regions in various *Leptospira* strains.
- Scientific information on the genetic variability of key genes using whole genome sequencing techniques.
- Determine the transcriptome of pathogenic *Leptospira* species to identify differentially

expressed genes to characterize virulence traits and select vaccine candidates.
- Genetically altered bacteria for *in vitro* and *in vivo* studies to establish key links between specific genes and phenotype.
- Efficacious molecular vaccines to prevent the spread of Leptospirosis in domestic animals and wildlife.
- Microbial immunogens critical for development of protective immunity.
- Scientific information to increase our understanding of the molecular pathogenesis of *Mycobacterium bovis* infections.
- Comparative analyses to understand variations of *Mycobacterium bovis* host immune responses to natural infections versus vaccination as well as neonatal versus adult cattle responses.
- Improved sensitive and specific diagnostic platforms amenable to the rapid screening of large cattle herds.
- Diagnostic platforms to differentiate infected versus vaccinated animals.
- Effective vaccine platforms to prevent and control *Mycobacterium bovis* in cattle and relevant wildlife reservoir hosts.
- Host genetic tools to prevent or reduce ruminant shedding of *Coxiella burnetii*.
- Discovery of a vaccine platform that can prevent or reduce ruminant shedding of *Coxiella burnetii*.

Impact:
The discovery of new countermeasures specifically designed to prevent and control brucellosis in wildlife eliminates new sources of infection and increase our ability to eradicate brucellosis in our domestic livestock. The functional genomics analysis of *Leptospira* strains provides the identification of virulence determinants that can lead to vaccine discovery research and new diagnostic platforms for classification of field strains. Developing new generation vaccines for leptospirosis may improve the control of maintenance and accidental host infections in our domestic animals thereby lowering the incidence of disease and protecting farm workers from spirochete-associated zoonoses. New improved countermeasures to control *Mycobacterium bovis* in wildlife and domestic livestock are needed to help prevent new incidences of bovine tuberculosis and support its eradication from the United States. Research on *Coxiella burnetii* is needed to protect cattle, sheep, and goats on U.S. farms and their products in global distribution channels. Importantly, controlling zoonotic agents at the animal source will safeguard people with potential exposure to infected livestock and protect the United States and trading partners from the agricultural, ecological and economic threat posed by zoonotic agents.

COMPONENT 3: SELECTED ACCOMPLISHMENTS

Problem Statement 3A: *Brucellosis*

Long Duration Antibiotic Not Effective Against Brucellosis in Goats
Whole herd treatment of infected goats with a long-lasting antibiotic could be an effective treatment for reducing infection, clinical effects and disease transmission to humans. Therefore, ARS scientists at the National Animal Health Disease Center, Ames, Iowa, treated goats with a commercial antibiotic after experimental infection with *Brucella melitensis* and found that infection and abortion rates were not decreased. Although this antibiotic is expected to remain

efficacious for 21 days after treatment, this study indicates that it would not provide economic or epidemiologic benefits under field conditions. This work eliminates a possible therapeutic approach for managing brucellosis in areas of high disease prevalence and clearly shows that the use of antibiotics as an intervention strategy for brucellosis in goats is not effective.

Scientific Publication:
Boggiatto PM, Olsen SC. 2019. Tulathromycin treatment does not affect bacterial dissemination or clearance of *Brucella melitensis* 16M following experimental infection of goats. PLoS One. 14(12):e0226242. doi: 10.1371/journal.pone.0226242. eCollection 2019. https://dx.doi.org/10.1371%2Fjournal.pone.0226242

Validation of Inactivation Methods for Brucella
Select agents are pathogens that the Department of Health and Human Services (HHS) and/or the U.S. Department of Agriculture (USDA) have deemed to pose a severe threat to public health and safety and are regulated through the Federal Select Agent Program (FSAP). The Federal Experts Security Advisory Panel (FESAP)which provides recommendations related to the security of select agents has emphasized the need to validate methods for inactivation of bacterial and viral agents, because of publicized failures involving select agent pathogens and the danger posed by improper disposal of these agents. ARS scientists at Ames, Iowa, tested several inactivation methods (heat, methanol, acetone, filtration, and formalin) under various conditions. These experiments demonstrated that 95C heating for 1 hour, 67 percent methanol for 5 days, or formalin treatment for 30 minutes were sufficient to prevent recovery of *Brucella* from spiked samples. Filtration of sera through a 0.22 um filter removed all viable *Brucella* from spiked samples. This published data is of great interest to regulatory and biosafety personnel, and the hundreds of laboratories that work with select agents. It provides validated procedures that are effective for inactivating *Brucella* from sera and tissue samples which can mitigate the risk of inadvertent release and ensure laboratory personnel safety.

Scientific Publication:
Olsen SC, Boggiatto PM, Vrentas CE. 2017. Inactivation of virulent *Brucella* species in culture and animal samples. Applied Biosafety. 22(4):145-151. https://doi.org/10.1177/1535676017734202

Olsen SC, Boggiatto PM, White DM, McNunn TB. 2018. Biosafety concerns related to *Brucella* and its potential use as a bioweapon. Applied Biosafety. 23(2)77-90. https://doi.org/10.1177/1535676018771983

Hydrogel Biobullets Maintain Brucella Vaccine Strain Viability
Vaccination of wildlife is important to the control of brucellosis. This is challenging since wildlife vaccines need to be delivered remotely and wild animals often can only be vaccinated once. In order to address some of these challenges, ARS scientists at the National Animal Disease Center, Ames, Iowa, and collaborators evaluated the stability of the *Brucella abortus* strain RB51 vaccine in degradable hydrogel biobullets. The formulation was shown to maintain excellent viability of the vaccine strain during storage and the hydrogels were shown to readily rehydrate within 5 hr in serum. This work establishes a possible remote vaccination option for delivering brucellosis vaccines to wildlife.

Scientific Publication:
Falconer J, Christie RJ, Kaiser EJ, Olsen SC, Grainger DW. 2016. Live RB51 vaccine lyophilized hydrogel formulations with increased shelf life for practical ballistic delivery. International Journal of Pharmaceutics. 498(1-2):187-94. doi:10.1016/j.iipharm.2015.12.040. https://doi.org/10.1016/j.ijpharm.2015.12.040

Efficacy of Brucella Vaccination in Elk
The prevalence of brucellosis in free-ranging elk in the Greater Yellowstone Area (area surrounding Yellowstone National Park) has been epidemiologically linked to infections in cattle herds. In an effort to identify an efficacious vaccine in elk, scientists at the National Animal Disease Center, Ames, Iowa, collaborated with Colorado State University and USDA APHIS to assess the efficacy of a new mucosal brucellosis vaccine. Pregnant elk cows were experimentally challenged with virulent *Brucella abortus* in the third trimester. There was no difference in abortion rates between control and vaccination treatments. Data indicates that the mucosal vaccine did not protect elk against brucellosis. As an efficacious brucellosis vaccine has not yet been identified in elk, intervention strategies to address brucellosis in free ranging elk are lacking. This data will be of interest to regulatory personnel addressing brucellosis in free-ranging elk and scientists working to develop brucellosis vaccines.

Scientific Publication:
Nol P, Olsen SC, Rhyan JC, Sriranganathan N, McCollum MP, Hennager SG, Garner A, Sprino PJ, Berrier RJ, Elzer P, Boyle SM, Salman MD. 2016. Vaccination of elk (Cervus canadensis) with *Brucella abortus* strain RB51 overexpressing superoxide dismutase and glycosyltransferase genes does not induce adequate protection against experimental brucella abortus challenge. Frontiers in Cellular and Infection Microbiology. 6:10. doi: 10.3389/fcimb.2016.00010. https://doi.org/10.3389/fcimb.2016.00010

Problem Statement 3B: *Leptospirosis*

Isolation and Characterization of Pathogenic Leptospires
Leptospirosis is a bacterial disease transmitted from animals to humans worldwide. As leptospires infect the kidney of natural hosts and are excreted in the urine, contact with infected urine or contaminated water can result in disease. Bovine leptospirosis is endemic in the United States and infection in cattle causes reproductive losses. Current commercial vaccines are based on strains isolated from cattle over 25 years ago. ARS scientists at the National Animal Disease Center, Ames, Iowa, characterized leptospires currently circulating in cattle. The study found that 7.2 percent of sampled cattle were actively excreting leptospires and all isolates were found to be serovar Hardjo, historically the predominant serovar infecting cattle worldwide.

Scientific Publication:
Nally JE, Hornsby, RL, Alt DP, Bayles DO, Wilson-Welder JH, Bauer NE. 2018. Isolation and characterization of pathogenic leptospires associated with cattle. Veterinary Microbiology. 218:25-30. https://doi.org/10.1016/j.vetmic.2018.03.023

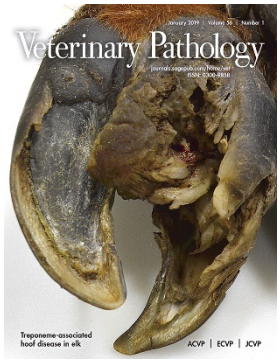
Development of Culture Method to Study Leptospirosis in Animal Hosts
Leptospirosis is an important human and animal disease worldwide. The number of serious human cases of leptospirosis seen annually throughout the world is estimated at over one million, with a case fatality rate above 10 percent. The disease is maintained in soil, water and wildlife reservoirs and because there are many serotypes of this bacterium it is difficult to fully vaccinate against it. In cattle, leptospirosis results in abortion, stillbirth, premature birth and reproductive failure. The composition of cattle vaccines is based on studies that were performed 25 years ago and may not reflect the strains that are currently circulating. An additional challenge to developing improved vaccines against leptospires is that these bacteria are very difficult to grow under laboratory conditions, and do not express the same proteins normally found during natural infection which makes fully characterizing them challenging. ARS researchers at the National Animal Disease Center, Ames, Iowa, developed a novel method for culturing leptospires that more closely mimics natural infection. This unique culture method resulted in expression of *Leptospira* proteins normally found during infection that are usually not expressed under laboratory conditions. These data provide novel insights on colonization and persistence of infection in natural hosts and could lead to new detection and control strategies for this important human and animal pathogen.

Scientific Publication:
Publication pending: ARS is currently pursuing a patent on the culture media.

An Inbred Rat Model to Study Renal Infection
Pathogenic species of *Leptospira* cause leptospirosis, a zoonotic disease with a global distribution. In natural hosts, leptospires localize in the kidney and are excreted in urine. Reservoir hosts of leptospirosis are typically asymptomatic and excrete leptospires for months and years. Surprisingly, little is known about host cellular immune response that facilitate persistent renal infection. ARS scientists at the National Animal Disease Center, Ames, Iowa, developed a laboratory animal model of persistent renal Leptospira colonization. Data demonstrated the inbred rats developed persistent renal infections with excretion of leptospires in urine. Results implicated the local renal lymph node as an important site for immunologic responses to *Leptospira* rather than systemic immune responses. These data allow understanding of immunologic responses that contribute to persistent renal infection and provides critical information on disease pathogenesis.

Scientific Publication:
Nally JE, Wilson-Welder JH, Hornsby RL, Palmer MV, Alt DP. 2018. Inbred rats as a model to study persistent renal colonization and associated cellular immune responsiveness. Frontiers in Cellular and Infection Microbiology. https://doi.org/10.3389/fcimb.2018.00066

Characterization of Treponeme Associated Hoof Disease in Free-ranging Elk



Hoof disease in elk is associated with treponeme, a spirochete bacterium that is pathogenic in humans and warm-blooded animals. ARS scientists at the National Animal Disease Center, Ames, Iowa, collaborated with state officials and university scientists in characterization of hoof disease in free-ranging elk (*Cervus elaphus*) in southwestern Washington State, which continues to spread throughout the region with some herds showing 20-90% of animals affected. In adult elk, lesions of hoof overgrowth, sole ulcers, and sloughed hoof capsules are observed in addition to higher mortality. A field study was conducted to characterize the pathogenesis. Lesions were most severe in elk at nine months of age and mimicked lesions in adult elk, while elk examined at three months of age had the mildest lesions and evidence of treponemes. Data suggests that Treponeme-associated hoof disease in free-ranging elk is a debilitating and progressive condition that is similar to digital dermatitis in cattle and sheep. The work provides critical insight into the pathogenesis of this disease that may lead to intervention strategies that reduce its prevalence in the United States and is of importance to the wildlife community and state-health officials.

Scientific Publication:

Nally JE, Hornsby RL, Alt DP, Bayles DO, Wilson-Welder JH, Bauer NE. 2018. Isolation and characterization of pathogenic leptospires associated with cattle. Veterinary Microbiology. 218:25-30. https://doi.org/10.1016/j.vetmic.2018.03.023

Problem Statement 3C: *Tuberculosis*

Biomarkers for Improved Diagnostic Tests

As bovine tuberculosis is a zoonotic disease, an eradication program was initiated over 100 years ago to protect public health. Complete eradication of bovine tuberculosis in the United States will require development of rapid and accurate diagnostic tests. A blood-based test measuring gamma interferon production has been used for years for diagnosis of bovine tuberculosis. Although it has reasonable accuracy (sensitivity and specificity), to maximize diagnostic sensitivity the blood-based test must be combined with other diagnostic tests (i.e. skin test) that are labor intensive and expensive. ARS scientists at the National Animal Disease Center, Ames, Iowa, used whole blood from experimentally infected cattle to evaluate inflammatory mediators as alternatives to gamma interferon for use in tuberculosis diagnostic tests for cattle. Data suggested that several of the targets may be useful for diagnosis of tuberculosis using whole blood. Development of more accurate diagnostic tests will expedite eradication of bovine tuberculosis and decrease financial losses caused by reduced production and regulatory costs to protect public health.

Scientific Publication:

Palmer MV, Thacker TC, Rabideau MM, Jones GJ, Kanipe C, Vordermeier HM, Ray Waters W. 2020. Biomarkers of cell-mediated immunity to bovine tuberculosis. Vet Immunol Immunopathol. 220:109988. doi: 10.1016/j.vetimm.2019.109988. https://doi.org/10.1016/j.vetimm.2019.109988

Increase in Sensitivity and Specificity of Bovine Tuberculosis Diagnostic Testing
A new serological diagnostic test for tuberculosis in cattle was licensed by the Center for Veterinary Biologics in 2013 and approved for use in the bovine tuberculosis eradication program by USDA APHIS. Research during the fiscal year 2014 showed that the novel serological test was not accurate enough to entirely replace the current testing method (tuberculin skin test). However, it was noted that tuberculin skin testing resulted in a "boosting effect" and an increase in the antibodies detected by the new serological test. The boost in antibody production resulted in an enhanced ability of the new serological test to detect *Mycobacterium bovis* infected cattle (increased accuracy). The two tests used together in this fashion, were better at identifying infected cattle than either test alone. A new paradigm was proposed to administer the tuberculin skin test and then draw blood for the new serological test during the boosting period. However, it was not clear when the boosting period began or ended. In the fiscal year 2015, ARS researchers at the National Animal Disease Center, Ames, Iowa, determined both the beginning (7-10 days) and the ending (60-70 days) of the boosting period. This work allows USDA APHIS Tuberculosis program staff to direct field veterinarians to schedule testing at times that optimize the accuracy of testing, making the diagnosis of bovine tuberculosis more efficient and cost effective for both producers and USDA. This work provides a scientific basis for a change in USDA APHIS regulations directing field veterinarians to obtain blood for the novel test 7 to 70 days after skin testing.

Scientific Publication:
Waters WR, Palmer MV, Stafne MR, Bass KE, Maggioli MF, Thacker TC, Linscott R, Lawrence JC, Nelson JT, Esfandiari J, Greenwald R, Lyashchenko KP. 2015. Effects of serial skin testing with purified protein derivative on the level and quality of antibodies to complex and defined antigens in *Mycobacterium bovis*-infected cattle. Clin Vaccine Immunol. 22(6):641–649. http://dx.doi.org/10.1128/CVI.00119-15

Problem Statement 3D: *Q-Fever*

Domestic Sheep Associated with Human Q Fever
Coxiella burnetii is a globally distributed zoonotic bacterial pathogen that causes abortions in ruminant livestock. In humans, an influenza-like illness results with the potential for hospitalization, chronic infection, abortion, and fatal endocarditis. Ruminant livestock, particularly small ruminants, are thought to be the primary transmission source to humans. A recent Netherlands outbreak from 2007-2010 traced to dairy goats resulted in over 4,100 human cases with estimated costs of more than 300 million euros. Smaller human Q fever outbreaks of small ruminant origin have occurred in the United States, and characterizing shedding is important to understand the risk of future outbreaks. Scientists at the Animal Disease Research Unit, Pullman, Washington, conducted a study to assess bacterial shedding and seroprevalence in 100 sheep from an Idaho location associated with a 1984 human Q fever outbreak. The results from this study indicated that 5 percent of the sheep were seropositive, which was not significantly different from the national average of 2.7 percent for the United states. Importantly, the bacterial pathogen was not detected from placentas, vaginal swabs, or fecal samples, suggesting that none of the sheep were shedding *C. burnetii*. While the presence of seropositive individuals demonstrates some historical *C. burnetii* exposure, the negative placental samples suggest the shedding events were rare or absent. The location where the flock

was maintained had little or no depopulation in 1984, and no *C. burnetii* vaccination during or since 1984. It is not clear how a zero-shedding rate was achieved in these sheep beyond natural immunity; thus more work is required to discover and assess possible factors that may contribute towards achieving zero-shedding status. This study provided the first U.S. sheep placental *C. burnetii* shedding update in over 60 years and demonstrated potential for *C. burnetii* shedding to reach undetectable levels after an outbreak event, even in the absence of targeted interventions, such as vaccination

Scientific Publication:
Oliveira RD, Mousel MR, Pabilonia KL, Highland MA, Taylor JB, Knowles DP, White SN. 2017. Domestic sheep show average *Coxiella burnetii* seropositivity generations after a sheep-associated human Q fever outbreak and lack detectable shedding by placental, vaginal, and fecal routes. PLoS One. 12(11):e0188054. Published 2017 Nov 15. doi:10.1371/journal.pone.0188054. https://dx.doi.org/10.1371%2Fjournal.pone.0188054

Component 4: Respiratory Diseases

Rationale for the research:

In spite of decades of control measures using antibiotics and vaccines, endemic respiratory diseases remain primary health threats to livestock and poultry. The cost of respiratory disease is significant and disease outbreaks often determine the difference between profit and loss for a producer. Most respiratory diseases present themselves as disease complexes involving several primary and secondary viral and bacterial pathogens, complicating control and prevention strategies. The most challenging aspect of dealing with respiratory disease is recognizing that clinical or overt disease is only the tip of the iceberg. The cost goes far beyond the treatment of sick animals and the cost of dead animals. The vast majority of the economic impact is actually due to the hidden cost of sub-clinical disease where animals are infected but show no apparent disease symptoms. Livestock and poultry that develop respiratory diseases have notable decreases in growth performance. Even with livestock and poultry being vaccinated against many of the most common pathogens today, respiratory lesions are still prevalent at slaughter and their impact on weight gain and carcass quality is significant. Important scientific gaps remain in our understanding of respiratory pathogen complexes and the ecological and host interactions that lead to disease and production losses independent of host species. With the current emphasis on reduced usage of antibiotics in livestock and poultry operations, new research approaches are needed to design effective prevention and control programs that will facilitate proper planning, careful attention to livestock and poultry health management, and the discovery of effective countermeasures.

Stakeholders representing the livestock and poultry industries that responded to the 2015 ARS animal health national survey identified research on respiratory diseases as a national priority. Because of the sheer number of pathogens involved in respiratory diseases and the ability of many pathogens to cross the species' barrier, ARS used available resources to focus strategically on priority respiratory pathogens associated with the bovine, porcine, poultry, and sheep respiratory disease complexes. Emphasis was given to the design of experimental animal disease models to test newly discovered technologies and countermeasures, with the eventual goal of validating them under field conditions through strategic partnership with industry.

Research needs:

Research needs identified included identifying and understanding the mechanisms of disease transmission of respiratory pathogens in beef production systems as well as the host responses to respiratory pathogens, including mechanisms of immune evasion and protective immunity. Also recognized as a critical need were epidemiological studies to identify reservoirs of priority respiratory pathogens.

The transmission of malignant catarrhal fever to bison in western grazing lands is a concern to the sheep industry. Two of the most important gaps in the understanding of the transmission between the two species include the pathogenesis and immune response in sheep and bison associated with infection. Research in this area will help develop intervention strategies to minimize the risk of the sheep spreading the devastating disease to bison.

Respiratory disease in swine is one of the most serious and costly concerns to the industry. Research is needed to elucidate the pathogenesis of monovalent and polymicrobial infections of swine respiratory pathogens. Pathogens included in the swine respiratory research included porcine reproductive and respiratory syndrome virus (PRRSV), swine influenza virus (SIV), *Bordetella bronchiseptica*, and *Glaesserella parasuis*. The mechanisms by which swine respiratory pathogens caused disease due to changes in gene expression of both the porcine host and the bacterial respiratory pathogens during the infectious process are important in developing effective intervention strategies for polymicrobial infections. In addition, research to understand the host responses to respiratory pathogens, including mechanisms of immune evasion and protective immunity are needed.

Research gaps were recognized in the elucidation of the mechanisms of disease transmission of respiratory pathogens in relevant poultry production systems including the interaction and pathogenesis of polymicrobial interactions. Studies are needed to investigate the host response to respiratory pathogens, including mechanisms of immune evasion and protective immunity. In addition, novel vaccine candidates need to be developed and evaluated to a number of pathogens. Improved diagnostic capabilities to enable rapid differential diagnosis of respiratory pathogens on poultry farms are also lacking for many of the common poultry respiratory diseases.

Anticipated Products in Action Plan:

- Discovery of determinants of virulence and characterization of mechanisms of infection.
- Scientific information on pathogen interactions that lead to polymicrobial infections and respiratory disease complexes.
- Scientific information on mechanisms of immune evasion and protective immunity.
- Drug and vaccine delivery systems that target the respiratory tract of cattle, sheep and goats.
- Diagnostic platforms that can be used to develop rapid on-site tests.
- Scientific information on the pathogen interactions that lead to polymicrobial infections and respiratory disease complexes in swine.
- Scientific information on changes in gene expression underlying porcine immune responses to infection with respiratory pathogens.
- Scientific information on global changes in gene expression of porcine bacterial pathogens in response to respiratory infection.
- Scientific information on microbial genetic variations associated with differences in virulence and disease transmission.
- Efficacious vaccines that induce targeted immune responses to prevent colonization of the respiratory tract and prevent shedding and disease transmission.
- Scientific information on the characteristics of aerosol spread of priority respiratory pathogens in relevant poultry production systems.
- Drug and vaccine delivery systems that target the avian respiratory tract.
- Differential diagnostics platforms that can be used to develop flock-side tests.
- Scientific information to identify pathogens associated with respiratory disease in domestic and wild sheep.
- Scientific information to determine the differential immune responses between domestic and wild sheep that contribute to population limiting respiratory disease.

- Scientific information to identify the genetic determinants of respiratory disease susceptibility.
- Scientific information to determine management practices that trigger or contribute to population-based respiratory disease.

Impact:
The overall impact of the research resulted from improved diagnosis, control, and prevention of endemic respiratory diseases that benefitted the U.S livestock and poultry industries. The research delivered the identification of disease pathogen reservoirs, understanding pathogen transmission, and the discovery and technology transfer of highly effective diagnostics, vaccines, and biotherapeutics designed to control and eradicate respiratory diseases. Incremental development of these tools provided more predictable costs and better potential returns to enable the U.S. livestock and poultry industries to remain competitive and profitable.

COMPONENT 4: SELECTED ACCOMPLISHMENTS

Problem Statement 4A: *Bovine*

A Genome-Wide Association Study for the Incidence of Persistent Bovine Viral Diarrhea Virus Infection in Cattle
Bovine viral diarrhea viruses (BVDV) comprise a diverse group of viruses that cause disease in cattle. BVDV may establish both transient and persistent infections depending on the developmental stage of the animal at exposure. ARS scientists at the National Animal Disease Center, Ames, Iowa, conducted a study to determine whether genomic regions harboring genetic mutations called single nucleotide polymorphisms (SNP) could be associated with the presence or absence of persistent BVDV infection. A cattle genome-wide association approach based on 777,000 SNP markers was used to test samples from 2400 animals identified as positive or negative for the presence of BVDV in the skin. One SNP marker chromosome 14 was found to be significantly associated with BVDV persistent infection. Fifteen SNP markers, residing on chromosomes 1, 2, 6, 8, 10, 15 and 18, were moderately associated with persistent BVDV infection. The function of the genes harboring these mutations provides leads in understanding the mechanisms involved in BVDV persistent infections.

Scientific Publications:
Casas E, Hessman BE, Keele JW, Ridpath JF. 2015. A genome-wide association study for the incidence of persistent bovine viral diarrhea virus infection in cattle. Anim Genet. 46(1):8-15. doi: 10.1111/age.12239. https://doi.org/10.1111/age.12239

Falkenberg SM, Dassanayake RP, Walz P, Casas E, Neill JD, Ridpath JF. 2019. Frequency of bovine viral diarrhea virus detected in subpopulations of peripheral blood mononuclear cells in persistently infected animals and health outcome. Vet Immunol Immunopathol. 207:46-52. doi: 10.1016/j.vetimm.2018.11.015. https://doi.org/10.1016/j.vetimm.2018.11.015

Taxis TM, Bauermann FV, Ridpath JF, Casas E. 2019. Analysis of tRNA halves (tsRNAs) in serum from cattle challenged with bovine viral diarrhea virus. Genet Mol Biol. 42(2):374-379. doi: 10.1590/1678-4685-GMB-2018-0019. https://doi.org/10.1590/1678-4685-gmb-2018-0019

Problem Statement 4B: *Porcine*

A Computational Tool to Characterize the Antigenic Diversity of Swine Influenza Viruses
Infection with swine influenza A viruses (IAV) is one of the most important respiratory diseases of swine and is the second most common viral diagnosis of respiratory disease in swine in the United States. Furthermore, the global diversity of swine IAV creates substantial risks for both human and swine populations and could be a major contributor to future outbreaks and potential pandemics in humans. There is a keen interest in improving the control of IAV in swine through vaccination. A significant barrier to improve the efficacy of vaccines is lacking the computational expertise to analyze and characterize the hemagglutinin (HA) gene of IAV to properly match vaccines to field strains. The HA protein is a major component of vaccines and target to induce a protective immune response. ARS scientists working at the National Animal Disease Center, Ames, Iowa, in collaboration with virus experts from the OIE/FAO Network of Expertise on Animal Influenza (OFFLU), have developed a computational tool that automatically classifies HA gene sequences from swine IAV isolates. This open-access tool is now widely available and will aid swine producers, veterinarians, vaccine manufacturers, and IAV vaccine researchers in selecting vaccine strains to match the strains that are currently circulating on swine farms.

Scientific Publication:
Zhang Y, Aevermann B, Anderson TK, Burke DF, Dauphin G, Gu Z, He S, Kumar S, Larsen CN, Lee AJ, Li X, Macken C, Mahaffey C, Pickett BE, Reardon B, Smith T, Stewart L, Suloway C, Sun G, Tong L, Vincent AL, Walters B, Zaremba S, Zhao H, Zhou L, Zmasek C, Klem EB, Scheuermann RH. 2017. Influenza research database: an integrated bioinformatics resource for influenza virus research. Nucleic Acids Research. 45(D1):D466-D474. doi:10.1093/nar/gkw857. https://doi.org/10.1093/nar/gkw857

Antiviral Treatment for Porcine Reproductive and Respiratory Syndrome Virus (PRRSV) Infection in Pigs
Interferon is produced in response to virus infections to help the host inhibit viral replication and develop a stronger immune response to the pathogen. Porcine reproductive and respiratory syndrome virus is one of the most devastating and costly viruses to the swine industry world-wide and has been shown to induce a meager interferon response in pigs. ARS researchers at the National Animal Disease Center, Ames, Iowa, explored the use of interferon as an adjuvant given with live-attenuated PRRSV vaccine to determine if it would result in an enhanced immune response to the vaccine. Giving interferon totally abolished replication of the vaccine virus and as a result there was no detectible adaptive immune response after vaccination. Although interferon did not have the desired adjuvant effect, the results are promising for the use of interferon as a treatment for PRRSV infection.

Scientific Publication:
Brockmeier SL, Loving CL, Eberle KC, Hau SJ, Buckley A, Van Geelen A, Montiel NA, Nicholson T, Lager KM. 2017. Interferon alpha inhibits replication of a live-attenuated porcine reproductive and respiratory syndrome virus vaccine preventing development of an adaptive

immune response in swine. Veterinary Microbiology. 212:48-51. https://doi.org/10.1016/j.vetmic.2017.11.004

A Subunit Vaccine against Streptococcus suis in Swine

Streptococcus suis is a bacterium that is an important and common cause of disease in pigs and costs the swine industry millions in losses annually. It is also listed by the World Organization for Animal Health (OIE) as a high priority disease for which improved vaccines would significantly reduce the need for antibiotic administration. ARS researchers at the National Animal Disease Center, Ames, Iowa, with collaborators from the University of Cambridge identified five candidate proteins of *S. suis* which were formulated into a vaccine with different adjuvants to help stimulate an immune response. The vaccine was found to be effective at preventing disease caused by *S. suis*. In addition, antiserum from the vaccinated pigs was reactive against whole *S. suis* bacteria of differing serotypes indicating a potential for cross-protection. These proteins are now being developed into a vaccine by a commercial company that can be used by swine producers to protect against this devastating and costly swine disease. This technology may markedly improve the health and welfare of pigs, reduce the costs of pork production and reduce the use of antibiotics in pigs by decreasing the occurrence of diseases caused by an important bacterial pathogen.

Scientific Publication:

Brockmeier SL, Loving CL, Nicholson TL, Wang J, Peters SE, Weinert L, Chaudhuri R, Seilly DJ, Langford PR, Rycroft A, Wren BW, Maskell DJ, Tucker AW. 2018. Use of proteins identified through a functional genomic screen to develop a protein subunit vaccine that provides significant protection against virulent Streptococcus suis in pigs. Infection and Immunity 86(3):e00559-17. https://doi.org/10.1128/IAI.00559-17

Problem Statement 4C: *Poultry*

Novel Vaccines Effective Against Poultry Diseases

Vaccination is one method used to help prevent the spread of infectious poultry diseases, but current vaccines could be safer and more effective. ARS scientists at the U.S. National Poultry Research Center, Athens, Georgia, have developed a vaccine to help reduce virulent and virus shed excretion of virus by the host and disease transmission from infected birds to healthy ones. This novel vaccine was shown to protect chickens against infectious laryngotracheitis virus (ILTV) and Newcastle disease virus (NDV), two of the most economically important infectious diseases of poultry. Both viruses cause sickness and death in domestic and commercial poultry as well as in some wild birds throughout the world. While current ILTV live-attenuated vaccines are effective, some of the viruses used to make them can regain virulence causing chickens to become chronically ill. Other types of vaccines can protect birds from the disease's clinical signs, but barely reduce the virus shedding in their respiratory secretions after infection. In that sense, those vaccines are not that effective, because they do not reduce the risk of virulent ILTV transmission to uninfected birds. With the support of the ARS Office of Technology Transfer, ARS is currently looking for a commercial partner to develop this vaccine.

Scientific Publications:
Yu Q, Spatz S, Li Y, Yang J, Zhao W, Zhang Z, Wen G, Garcia M, Zsak L. 2017. Newcastle disease virus vectored infectious laryngotracheitis vaccines protect commercial broiler chickens in the presence of maternally derived antibodies. Vaccine. 35(5):789-795. doi: 10.1016/j.vaccine.2016.12.038. https://doi.org/10.1016/j.vaccine.2016.12.038

Yu Q, Li Y, Dimitrov K, Afonso CL, Spatz S, Zsak L. 2019. Genetic stability of a Newcastle disease virus vectored infectious laryngotracheitis virus vaccine after serial passages in chicken embryos. Vaccine. 38(4):925-932. doi: 10.1016/j.vaccine.2019.10.074. https://doi.org/10.1016/j.vaccine.2019.10.074

Development of an improved vaccine evaluation protocol to compare the efficacy of Newcastle disease vaccines
ARS scientists at the U.S. National Poultry Research Center, Athens, Georgia, developed an improved animal challenge model to evaluate the efficacy of Newcastle disease vaccines. This challenge model is more stringent than currently used models in that it allows the detection of statistically significant differences in mortality between Newcastle disease vaccines. This is important because different areas of the world are affected by different strains of Newcastle disease virus and this system may improve the ability of veterinarians to choose a more appropriate vaccine for their region. This animal model also addresses the importance of evaluating vaccines for suppression of viral shedding. Current vaccine evaluation protocols do not evaluate the capacity of vaccines to reduce viral shedding. This deficiency is a problem in countries that do not practice culling (endemic poor countries) as the virus continues to be maintained in poorly vaccinated poultry farms.

Scientific Publication:
Cardenas-Garcia, S, Giel, DG, Susta, L, Lucio-Decanini, E, Yu, Q, Brown, CC, Miller, PJ, Afonso, CL. 2015. Development of an improved vaccine evaluation protocol to compare the efficacy of Newcastle disease vaccines. Biologicals. 43(2):136-145. https://doi.org/10.1016/j.biologicals.2014.11.003

Automated Vaccine Delivery System to Improve Biosecurity in Poultry Houses
Traditional methods of vaccinating poultry often involve an operator entering the poultry house to spray vaccine using a backpack-mounted device, which increases the likelihood of pathogens being inadvertently brought into the barn with equipment and increasing biosecurity risks. Although vaccines are often added to poultry water supplies, there can also be problems with contamination using this method of vaccination. To solve these problems, ARS scientists at the Poultry Research Unit, Mississippi State, Mississippi, developed a new, fully automated system for spraying the vaccines inside the barns. The idea behind the machine is to trigger birds to drink from the water lines using changes in lighting. After the birds go to the water lines, vaccine spray nozzles are lowered from the ceiling and spray the birds. The nozzles are strategically placed above the water lines, to ensure maximum coverage over the birds. The target is the important eye area, where scientists know that vaccines are easily absorbed by the birds, but the vaccine will also be picked up by the birds through preening and contact with other birds. The scientists compared the performance of the automated system against a three-man vaccination crew with backpack sprayers using a combination infectious bronchitis and Newcastle disease

vaccine. The results of the blood samples taken from automatically vaccinated flocks showed improved immune protection against the viruses compared with the backpack method, as well as reducing biosecurity risks and the number of personnel required to vaccinate a flock.

News Announcement:
"How could automated vaccine delivery help poultry producers"
http://www.thepoultrysite.com/poultrynews/36455/ippe-how-automated-vaccine-delivery-could-help-poultry-producers/

Scientific Publication:
Purswell JL, Branton, SL, Evans JD. 2019. Performance of an automated whole-house spray vaccination system. Journal of Applied Poultry Research. 28(1):215-220.

Problem Statement 4D: *Sheep*

Mycoplasma ovipneumoniae Identified in a Widening Array of Animals
Mycoplasma ovipneumoniae, currently believed to be the primary agent of pneumonia in bighorn sheep, has been described as having a host range restricted to *Caprinae* species (sheep, goats, musk ox) with domestic sheep and goats being implicated as the historic and continued source for introduction of this bacterium in bighorn sheep. This assumption has dictated grazing practices and policy implementation by other federal and state agencies. ARS researchers at the Animal Disease Research Unit, Pullman, Washington, performed epidemiological studies and identified *M. ovipneumoniae* in the following numbers of tested animals: 21 of 421 (5 percent) of caribou, 9 of 362 (2.5 percent) of moose, 23 of 184 (12.5 percent) of Dall's (thinhorn) sheep, and 5 of 43 (12.5 percent) mountain goats. ARS scientists in Pullman, Washington, in collaboration with several veterinarians and a state agriculture department identified this bacterium in captive white-tailed and mule deer exhibiting respiratory illness, and in a bison (no history reported). Determining the true host range of this bacterium is of utmost importance to livestock, federal and state stakeholders, as current policies to restrict grazing and thereby interactions between domestic small ruminants and bighorn sheep will not fully address possible transmission routes and remain ineffective until all vectors are considered.

Scientific Publications:
Highland MA, Herndon DR, Bender S, Hansen L, Gerlach RF, Beckman KB. 2018. Mycoplasma ovipneumoniae in wildlife species beyond the subfamily caprinae. Emerging Infectious Diseases. 24(12):2384-2386. https://doi.org/10.3201/eid2412.180632.

Rovani ER, Beckman KB, Highland MA. 2019. Mycoplasma ovipneumoniae associated with polymicrobial pneumonia in a free-ranging yearling barren ground caribou (Rangifer tarandus granti) from Alaska, USA. Journal of Wildlife Diseases. 55(3):733-736.
https://doi.org/10.7589/2018-08-188

Component 5: Priority Production Diseases

Rationale for the research:

Production diseases affect agricultural animals universally and are the cause of significant production losses, mortality, and waste. Many production diseases are difficult to prevent with on-farm biosecurity measures alone. Significant scientific gaps remain in our understanding of infection disease complexes, disease transmission, and the ecological and host interactions that lead to disease and production losses. With the continued concern over animal welfare, the use of antibiotics in animal agriculture, and impact on the environment, there is a need to find safe and practical countermeasures to prevent and control production diseases. Research is needed to identify the pathogens responsible for production diseases, molecular tools for epidemiological studies, and the discovery of improved diagnostics and vaccines that can be integrated in the design of effective prevention and control programs.

Stakeholders representing the livestock and poultry industries that responded to the 2015 ARS animal health national survey identified research on the following endemic diseases as a national priority: 1) Johne's disease; 2) Oncogenic Viruses of Poultry; 3) Enteric Diseases of Poultry; and 4) Bovine Mastitis

Research Needs:

Johne's disease (paratuberculosis) is a chronic, progressive enteric disease of domestic and wild ruminants caused by infection with the intracellular pathogen, *Mycobacterium avium* subsp. *paratuberculosis*. It is estimated that 20-30 percent of U.S. dairy and cattle herds are infected with this pathogen. There is a need to complete the sequencing of the *M. paratuberculosis* genome to provide new research tools to identify *M. paratuberculosis*-specific genes and proteins that may be useful as diagnostic tools or vaccine candidates.

Oncogenic viruses of poultry are endemic in the United States and cause periodic outbreaks with severe economic loss. Depending on the virus, control measures consist of either blanket vaccination of all commercial birds or diagnostic testing procedures to ensure breeder flocks remain virus free. However, the continued circulation of these viruses in commercial flocks lead to shifts in viral virulence or the emergence of new subgroups through mutation and/or recombination, rendering the latest control measures ineffective. There is a need to implement genomics-based research programs to identify and decipher genetic and biological determinants of virulence, immune evasion mechanisms, and the emergence of new tumor viral strains.

Enteric diseases continue to pose a significant threat to the poultry industry and countermeasures to prevent and control them are needed. There are several pathogens known but production losses associated with unclassified enteric disease in both turkeys and chickens are continual problems for the poultry industry. There is a need to understand and characterize the immune response during enteric infections and apply immunological and genomic approaches to identify host and pathogen genes involved in resistance to enteric infections. Tools are needed to study the epidemiology and genetic relationships of enteric infectious organisms and the processes that regulate host response to enteric infection to enable the development of effective strategies to

prevent enteric diseases. The use of comparative studies designed to better understand the gut microbiome in healthy and underperforming poultry flocks are needed to allow researchers to identify disease-associated pathogens to develop targeted interventions.

Mastitis continues to be the single most costly dairy disease. Antibiotics are often used to treat and prevent mastitis, but their use in food producing animals remains a major concern due to antimicrobial resistance. There is a need to develop alternatives to antibiotic and immunologic-based strategies to prevent and control bovine mastitis. Bovine functional genomics studies are needed to understand variations in gene expression in response to bacterial challenges of the mammary gland.

Anticipated Products from the Action Plan:

- Scientific information on the *Mycobacterium paratuberculosis* proteome to identify unique bacterial proteins that will enable the development of highly sensitive and specific diagnostic tests for the detection of Johne's disease in cattle and sheep.
- Scientific information on the host immune response to understand the mechanisms of control in early stages of disease and the switch in immunity that results in progression from subclinical to clinical disease.
- Highly effective vaccine platform that prevents subclinical disease, shedding of *M. paratuberculosis*, and progression to clinical disease.
- Scientific information on how genetic variations influence the immune response to Marek's disease virus (MDV) infection.
- Scientific information on how the interplay between specific host and MDV genes, and the variation within these genes, leads to disease susceptibility or resistance.
- Simple molecular tests to pathotype field strains of MDV.
- Viral genes responsible for pathogenesis and identification of predictors of virulence shifts.
- Scientific information on the biological pathways that lead to the development of Marek's disease.
- Safe and effective vaccines with mass vaccination capability that convey protection against emerging MDV strains in defined host animal genotypes.
- Molecular-based techniques to rapidly speciate and quantify *Eimeria* oocysts in litter samples.
- Rapid tests to identify drug resistance markers in *Eimeria* field isolates.
- Recombinant vaccines that are safe and effective against heterologous field challenges with mass vaccination capability to prevent outbreaks of coccidiosis in poultry farms.
- Recombinant vaccines targeting specific enteric viruses early during the poultry production period.
- Discovery of quantifiable factors associated with disease risk.
- Discovery of modulators of stress in production systems that affect enteric disease development and severity.
- Cytokines and expression profiles associated with processes involved in host defense during enteric infection.
- Discovery and characterization of pathogens responsible for poultry enteric disease complexes.

- Pathogen-specific markers useful for molecular or immunological detection.
- Molecular tools to study the epidemiology, ecology, and evolution of enteric pathogens.
- Intervention strategies that enhance the clearance of enteric pathogens.
- Immuno-intervention strategies that prevent the development of enteric infections.
- New biotherapeutic platforms/immune stimulators based on protective host proteins to induce and supplement the cow's innate immune response.
- Therapeutics to reduce cell damage and enhance repair during mastitis.
- New management and nutritional schemes to prevent metabolic stresses contributing to immunosuppressive states in the dairy cow.
- New non-antibiotic dry cow therapies are to be developed and tested for efficacy.
- Scientific information to define bacterial pathogenesis with the outcome of new targets for intervention of persistent or chronic infections.

Impact:
The impact of the ARS Animal Health Research Program on production diseases consisted of providing information on key host-pathogen responses during the infection process, leading to the development and application of genomic-based countermeasures. Importantly, this research program provided critical scientific information to support disease management programs.

COMPONENT 5: SELECTED ACCOMPLISHMENTS

Problem Statement 5A: *Johne's Disease*

New Diagnostics for Johne's Disease
Improved diagnostic tests are needed to aid producers in controlling this disease. Using a protein array, scientists at the National Animal Disease Center, Ames, Iowa, identified patterns of serological reactivity in cows with Johne's disease. Some antigens induced antibody responses throughout disease stages, where responses to other antigens were detected late in the course of disease, or early in infection with a decline in responses at later stages of disease. The most promising proteins identified from the protein array were tested by ELISA and a multiplex bead assay and distinguished infected from non-infected healthy animals with sensitivities ranging from 65 percent to 93 percent. Some of the proteins had better sensitivity on clinical samples when compared to a commercially available Johne's disease diagnostic tests. These data will be of interest to livestock producers, veterinarians, and diagnostic laboratories who want more accurate tests identifying animals infected with Johne's disease.

Scientific Publication:
Capsel RT, Thoen CO, Reinhardt TA, Lippolis JD, Olsen R, Stabel JR, Bannantine JP. 2016. Composition and potency characterization of *Mycobacterium avium* subsp. *paratuberculosis* purified protein derivatives. Veterinary Microbiology. 11(5):e0154685. doi: 10.1371/journal.pone.0154685. https://dx.doi.org/10.1371%2Fjournal.pone.0154685

Problem Statement 5B: *Oncogenic Viruses of Poultry*

Differences in T-Lymphocytes and Cecal Microbiome Community During Marek's Disease Virus Infection Are Associated with Genetic Resistance to Marek's Disease
Marek's disease (MD) is an important oncogenic disease of chickens caused by Marek's disease virus (MDV). In previous studies, ARS scientists demonstrated that certain genetic lines are resistant to MD. Recent interest in the role of the microbiome in preventing diseases led ARS scientists at the Avian Diseases and Oncology Laboratory, East Lansing, Michigan, in collaboration with Michigan State University scientists, to investigate the potential role of the microbiome in MD resistance. The results of this research showed differences in two chicken genetic lines, one resistant and the other susceptible to MD. Splenic helper CD4+ T cells were similar; however, cytotoxic CD8 sub-populations were different with a greater number of lymphocytes with cell surface markers associated with innate immunity in the MD resistant chicken line. Interestingly, the microbiome composition was different between resistant and susceptible birds. With MDV challenge, both chicken lines showed lower numbers of beneficial *Faecalibacterium* species and increased number of *Lactobacillus* species. Metabolic profiles between chicken types were similar but with MDV challenge, there were differences in metabolism in both chicken lines, with amino acid metabolism impacted in resistant birds and lipid metabolism in susceptible birds. These results provide insights into differences in the immune response of MD-resistant chickens and potential interplay with the microbiome during infection with an oncogenic virus. Information on the role of the microbiome in preventing diseases may be used in the future to reduce the loss from Marek's disease.

Scientific Publication:
Perumbakkam S, Hunt HD, Cheng HH. 2016. Differences in CD8αα and cecal microbiome community during proliferation and late cytolytic phases of Marek's disease virus infection are associated with genetic resistance to Marek's disease. FEMS Microbiol Ecol. 92(12):fiw188. https://doi.org/10.1093/femsec/fiw188

Problem Statement 5C: *Enteric Diseases of Poultry*

Comparative Analysis of the Intestinal Viral Communities from Birds on Chicken Farms
There is a great deal of interest in characterizing the complex microbial communities in the poultry gut, and in understanding the effects of these dynamic communities on poultry performance, disease status, animal welfare, and microbes with human health significance. Field investigations undertaken by ARS scientists at the U.S. National Poultry Research Center, Athens, Georgia, to characterize viruses in the gut of poultry identified several novel poultry viruses, but the roles these viruses play in disease and performance problems have yet to be fully understood. The complex bacterial community present in the poultry gut influences gut development, immune status, and animal health, each of which can be an indicator of overall performance. Analysis of the intestinal viruses from birds placed on poultry farms revealed colonization by members of the *Picornaviridae*, *Picobirnaviridae*, *Reoviridae*, and *Astroviridae*. Analysis of the birds gut bacterial community revealed an altered community, notably by members of the *Lachnospiracea*/*Clostridium* and *Lactobacillus* families and genera. Members of the avian enteric *Reoviridae* and *Astroviridae* have been well-characterized and have historically

been implicated in poultry enteric disease; members of the *Picobirnaviridae* and *Picornaviridae* have only relatively recently been described in the poultry and avian gut, and their roles in the recognized disease syndromes and in poultry performance in general have not been determined. This study has provided insight into the colonization of the poultry gut by enteric microbes circulating in commercial broiler flocks and has identified enteric viruses and virus communities that warrant further study in order to understand their role(s) in avian gut health and disease.

Scientific Publications:
Day JM, Zsak L. 2015. Investigating turkey enteric picornavirus and its association with enteric disease in poults. Avian Dis. 59(1):138-42. https://doi.org/10.1637/10940-092414-RegR

Day JM, Zsak L. 2016. Molecular characterization of enteric picornaviruses in archived turkey and chicken samples from the United States. Avian Dis. 60(2):500-505. https://doi.org/10.1637/11289-092415-ResNote

Host-pathogen Responses to Necrotic Enteritis in Two Inbreed Chicken Lines
Necrotic enteritis (NE) is an important intestinal infectious disease of commercial poultry flocks caused by *Clostridium perfringens*. Using an experimental model of NE, ARS scientists at the Animal Biosciences and Biotechnology Laboratory, Beltsville, Maryland, and Avian Diseases and Oncology Laboratory East Lansing, Michigan, collaborated to identify the genetic mechanisms that might regulate the host response to this disease. Genomic tools were used to measure gene expression, resulting in the identification of 1,049 genes whose levels of expression were altered in intestinal lymphocytes from infected Ross chickens compared with uninfected controls. Five biological functions, all related to host immunity and inflammation, and 11 pathways were identified from this dataset. To further elucidate the role of host genetics in NE susceptibility, two inbred chicken lines, ADOL line 6 and line 7, which have identical major histocompatibility genes (enables immune cells to recognize pathogens) but differ in their susceptibility to virus infection, were compared for clinical symptoms and the expression levels of a panel of immune-related genes during experimental NE. Line 6 chickens were more susceptible to development of experimental NE compared with line 7. Of 21 immune-related genes examined, 15 were increased in infected line 6 versus line 7 chickens. These results suggest that immune pathways are activated in response to experimental NE infection and that genetic determinants outside of the chicken major histocompatibility genes influence resistance to this disease.

Scientific Publications:
Truong AD, Hong YH, Lillehoj HS. 2015. High-throughput sequencing reveals differing immune responses in the intestinal mucosa of two inbred lines afflicted with necrotic enteritis. Vet Immunol Immunopathol. 166(3-4):116-124. doi: 10.1016/j.vetimm.2015.06.008. https://doi.org/10.1016/j.vetimm.2015.06.008

Oh ST, Lillehoj HS. 2016. The role of host genetic factors and host immunity in necrotic enteritis. Avian Pathol. 45(3):313-316. doi: 10.1080/03079457.2016.1154503. https://doi.org/10.1080/03079457.2016.1154503

Truong AD, Rengaraj D, Hong Y, Hoang CT, Hong YH, Lillehoj HS. 2017. Analysis of JAK-STAT signaling pathway genes and their microRNAs in the intestinal mucosa of genetically

disparate chicken lines induced with necrotic enteritis. Vet Immunol Immunopathol.187:1-9. doi: 10.1016/j.vetimm.2017.03.001. https://doi.org/10.1016/j.vetimm.2017.03.001

Problem Statement 5D: *Bovine Mastitis*

Genetic and Gene Expression Differences Between Escherichia coli Strains that Cause Persistent Versus Transient Mastitis in Dairy Cows

Infection of the mammary gland of cows with *Escherichia coli* can result in a self-resolving infection that last a few days or a persistent infection that can last months. It is important to determine the mechanisms that allows certain *E. coli* strains to persistently infect cows to allow for rapid detection and treatment of those cows likely to have a persistent infection. ARS researchers at the National Animal Disease Center, Ames, Iowa, have published work on the genomic and transcriptome differences between *E. coli* strains that can cause a persistent infection and those strains that cause transient mammary gland infections in dairy cows. Research has identified genes that were only present in the strains that cause persistent disease, these genes help bacteria to generate a protective capsid that may protect them from the immune system. The loss of these genes in bacteria that are correlated with persistent disease will be tested for a corresponding loss of persistent mastitis in cows.

Scientific Publications:

Lippolis JD, Holman DB, Brunelle, BW, Thacker TC, Bearson BL, Reinhardt TA, Sacco RE, Casey T. 2017. Genomic and transcriptomic analysis of *Escherichia coli* strains associated with persistent and transient bovine mastitis and the role of colanic acid. Infection and Immunity. 86(1):e00566-17. https://doi.org/10.1128/IAI.00566-17

Thacker TC, Lippolis JD, Brunelle BW, Casey T, Reinhardt TA, Sacco RE, Holman DB. 2017. Genome sequences of *Escherichia coli* strains that cause persistent and transient mastitis. Genome Announcements. 5(34):e00775-17. https://doi.org/10.1128/genomeA.00775-17

Component 6: Parasitic Diseases

Rationale for the research:

Parasites represent one of the most diverse groups of organisms that live on a host (ectoparasites) or within a host (endoparasites) and are responsible for hundreds of insidious diseases ranging from enteric diseases to vector-borne hemoparasitic infections. The livestock industries are severely affected by these parasitic diseases, which cause significant losses in animal production due to lower weight gain, anemia, diarrhea, and death from parasites. Moreover, many parasites are invasive and exotic to the United States and impact international trade. Most importantly, the emergence of drug resistant parasites against many commonly used pharmaceutical drugs has huge economic implications. To further complicate control, the populations of parasites may change with the climate changes anticipated with global warming.

Stakeholders representing the cattle, sheep, goat, and equine industries that responded to the 2015 ARS animal health national survey identified research on parasitic diseases of livestock as a national priority.

Research Needs:

Currently, drug resistance has emerged as the single most important problem confronting the control of gastrointestinal parasites of livestock worldwide. Although the use of drugs continues to be the primary treatment against parasites, the intensive use of these products has resulted in some degree of resistance to the majority of the drugs currently available. There is a need to define the mechanisms of anti-helminthic resistance to drugs such as Ivermectin and Fenbendazole used to treat nematodes of small ruminants and cattle. There is a need to elucidate the genetics of the immune response to parasites at both the host and parasite level to enable the development of novel intervention strategies to reduce resistance to drugs by parasites.

Hemoparasitic diseases result in significant export and production problems for the U.S. cattle and equine industries and continue to be a national priority for these industries. Priority hemoparasitic diseases include Anaplasmosis, Babesiosis, and Equine Piroplasmosis. Research is needed to discover new improved detection, control, and elimination strategies, including vector-related contributions to reduce disease risks from priority hemoparasites in areas within the United States characterized as endemic.

Anticipated Products from the Action Plan:

- Scientific information on cases of drug resistance related to parasite species; e.g., *Haemonchus contortus, H. placei, Cooperia punctata, C. oncophora, Ostertagia ostergii, Nematodirus helvetianus*, and *Trichostrongylus*.
- Scientific information on the effect of genotypic and phenotypic differences of the host and parasite of drug resistance in sheep, cattle and goats.
- Molecular probes to better define parasite species in the field to enable tracking if their range changes due to climate change.
- Molecular markers of drug resistance based on mode of action and measure the allele frequency of parasite genes involved in drug resistance.
- Scientific information on patterns of gene flow in nematode populations to manage drug

resistance in different production systems to reduce the impact of drug resistance on productivity.
- Novel control strategies such as vaccines and natural anti-parasiticides to control parasites.
- Scientific information on the transmission competence of vectors within the United States and trading partners (Canada and Mexico).
- Vaccines that prevent production losses from clinical disease and transmission (transfection technology is the center of our vaccine strategy for babesiosis).
- Scientific information on the effectiveness of current chemotherapeutics for *A. marginale, Babesia caballi, B. equi* and variant piroplasma species in clearing persistent infections.

Impact:
The ARS Animal Health Research Program on gastrointestinal parasites provided a greater understanding of the extent and type of drug resistance of nematodes of U.S. cattle and sheep, improved molecular probes for speciating nematodes in the farm environment, and identified markers of drug resistance. The research program on ectoparasites resulted in new research tools and the discovery of novel diagnostics and vaccines to prevent clinical disease and block vector borne- disease transmission.

COMPONENT 6: SELECTED ACCOMPLISHMENTS

Problem Statement 6A: *Gastrointestinal (GI) Parasitic Diseases*

Climatological Variation and Ecological Perturbation Drive Geographic and Host Colonization Events for Parasites and Pathogens
ARS researchers in collaboration with academic scientists, characterized how episodic shifts in climate and environmental conditions combine with host switching and other ecological mechanisms to hasten parasite diversification. This view runs counter to more than a century of co-evolutionary thinking about the nature of complex host-parasite assemblages. The Animal Parasitic Diseases Laboratory, Beltsville, Maryland, introduced the concepts of hard tipping points and shifting balances (related to temperature thresholds for parasite development) to explain how the range expansion and establishment likely occurs. These new insights provide a framework to understand and predict how ongoing and accelerating climate change will influence the distribution of parasites and the emergence of disease in wild and domestic animals, with consequences for food sustainability, availability, and animal and human health.

Scientific Publication:
Hoberg EP, Cook JA, Agosta SJ, Boeger W, Galbreath KE, Laaksonen S, Kutz SJ, Brooks DR. 2017. Arctic systems in the Quaternary: ecological collision, faunal mosaics and the consequences of a wobbling climate. J Helminthol. 91(4):409-421. doi: 10.1017/S0022149X17000347. https://doi.org/10.1017/S0022149X17000347

Novel Screening System Developed to Identify Anti-parasite Molecules
A microfluidic device ("chip") was developed to record the rhythmic contraction of the pharynx of parasitic worms that control worm feeding. These electropharyngeograms (EPGs) can now be

used to record responses from multiple worms per chip and can be used to evaluate novel drugs that target worm feeding to damage worm development. Therefore, ARS scientists at the Animal Parasitic Diseases Laboratory, Beltsville, Maryland, and colleagues at the University of California - San Francisco, George Washington University, and the University of Oregon validated this microfluidic technology using larval stages of parasitic human hookworms and the pig large round worm *Ascaris suum* against known anti-parasite drugs. Novel drugs are now being tested with this device. This showed that the microfluidic EPG platform provides a new tool for screening candidate drugs that can be further tested to eliminate parasitic worms from animals and humans.

Scientific Publication:
Weeks JC, Roberts WM, Robinson KJ, Keaney MI, Vermeire, JJ, Urban Jr JF, Lockery SR, Hawdon JM. 2016. Microfluidic platform for electrophysiological recordings from host-stage hookworm Ascaris suum larvae: a new tool for anthelmenthic research. International Journal for Parasitology: Drugs and Drug Resistance. 6(3):314-328. doi: 10.1016/j.ijpddr.2016.08.001. https://doi.org/10.1016/j.ijpddr.2016.08.001

Nanoparticles Improve Vaccines against Coccidiosis
Coccidiosis, a gut disease of poultry, costs U.S. producers $350 million annually due to poor weight gain in affected animals and the costs of treatment. It is also listed by the World Organization for Animal Health (OIE) as a high priority disease for which improved vaccines would significantly reduce the need for antibiotic administration. Current vaccines are comprised of low doses of highly infectious organisms and better vaccines are needed. ARS scientists at the Animal Parasitic Diseases Laboratory, Beltsville, Maryland, discovered that attaching a protective vaccine antigen to nanoparticles significantly improved efficacy. Chickens given the vaccine by oral administration at hatch showed improved weight gain and feed conversion efficiency, as compared to chickens vaccinated with the same antigen but without nanoparticles. This technology may markedly improve the health and welfare of poultry flocks, reduce the costs of poultry production and reduce the use of antibiotics in poultry by decreasing the occurrence of concomitant bacterial infections.

Scientific Publication:
Jenkins MC, Stevens L, O'Brien CN, Parker CC, Miska KB, Konjufca V. 2018. Incorporation of a recombinant Eimeria maxima IMP1 antigen into nanoparticles confers protective immunity against E. maxima challenge infection. Vaccine. 36(8):1126-1131. https://doi.org/10.1016/j.vaccine.2017.11.014

Development of an In-vitro Drug-sensitivity Test for Eimeria
Medication of poultry feed with ionophore drugs or synthetic chemicals represents a major way to control avian coccidiosis in the poultry industry. However, this approach has become less efficacious because of drug-resistance in Eimeria parasites. While there are several different anti-coccidial drugs available, it is impossible to know ahead of time which drug to use on a particular farm because there are no rapid tests for estimating drug-sensitivity in the resident Eimeria population. ARS researchers at the Animal Parasitic Diseases Laboratory, Beltsville, Maryland, developed an in vitro cell culture assay that utilizes chicken cells inoculated with the parasite in the presence or absence of ionophore drugs. The effect of these drugs on parasite

invasion and development was measured by using microscopy or molecular methods which indicate that in vitro cell culture constitutes a viable, rapid, and less costly alternative to evaluating drug-sensitivity in a broiler house. Poultry companies and poultry farmers will benefit from this technology by knowing which anti-coccidial drugs to use to prevent avian coccidiosis outbreaks.

Scientific Publication:
Jenkins MC, O'Brien CN, Fuller L, Mathis GF, Fetterer R. 2015. A rapid method for determining salinomycin and monensin sensitivity in Eimeria tenella. Vet Parasitol. 206(3-4):153-158. doi: 10.1016/j.vetpar.2014.09.017. https://doi.org/10.1016/j.vetpar.2014.09.017

A New Cure for Hookworm
Hookworms are intestinal nematode parasites that infect nearly half a billion people globally. They cause iron-deficiency, anemia and productivity losses of up to $139 billion annually. In some places, available drugs have less than a 40% cure rate, increasing the threat posed by emerging drug resistance. Fortunately, a pore-forming protein (Cry5B) produced by the soil bacterium *Bacillus thuringiensis* (Bt) has demonstrated good efficacy against *Ancylostoma ceylanicum* hookworm infections in hamsters. ARS scientists at the Beltsville Agricultural Research Center, Beltsville, Maryland, broadened the application of Cry5B to dogs infected with *Ancylostoma caninum*, and hamsters as a model for human hookworm infection with *Necator americanus.* This protein, Cry5B, was shown to be highly effective against all hookworm parasites tested and can be improved by neutralizing the effects of stomach acids. Efficacy did not depend on the host immune system and did not decline with repeated dosing. A pan-hookworm therapy with excellent properties for use in humans and other animals has thereby been discovered.

Scientific Publication:
Hu Y, Nguyen T, Lee AC, Urban Jr JF, Miller MM, Zhan B, Koch DJ, Noon J, Abraham A, Fujiwara T. 2018. Bacillus thuringiensis Cry5B protein as a new pan-hookworm cure. International Journal for Parasitology: Drug and Drug Resistance. 8:287-294. https://doi.org/10.1016/j.ijpddr.2018.05.001

Comparative Genomics of the Major Parasitic Worm
Parasitic nematodes (roundworms) and platyhelminths (flatworms) cause debilitating chronic infections of humans and animals, decimate animal production and are a major impediment to socioeconomic development. ARS scientists at the Animal Parasitic Diseases Laboratory, Beltsville, Maryland, and University collaborators undertook a major survey of genes that modulate host immune responses, enable parasite migration through host tissues and allow parasites to feed. They identified extensive lineage-specific differences in core metabolism and protein families that have historically been targeted for drug development. From wide-ranging analyses involving computer modeling and simulations, they identified and prioritized new potential drug targets and compounds for testing. This study is the broadest and most comprehensive comparative study to date utilizing the genomes of parasitic and non-parasitic worms, providing a transformative new resource for the research community to understand and combat the diseases that parasitic worms cause.

Scientific Publication:
International Helminth Genomes Consortium. Coghlan, A, Tyagi, R, Cotton, J.A. *et al.* 2019. Comparative genomics of the major parasitic worms. Nat Genet. 51**:**163–174. https://doi.org/10.1038/s41588-018-0262-1

Problem Statement 6B: *Hemoparasitic Diseases*

A New Research Tool for Tick-transmitted Diseases that Eliminates the Need for Animals
Laboratory methods to enhance large-scale preparations of purified pathogens from infected tick vectors are paramount to advance research of tick-transmitted diseases such as bovine anaplasmosis, bovine babesiosis, Lyme disease, and Heartwater. But these methods require the use of live animals to propagate these pathogens. Therefore, scientists at the Animal Diseases Research Unit, Pullman, Washington, designed and developed a novel continuous flow laboratory tick feeding system that facilitates isolating pure, free, and infectious tick-borne pathogens, thus eliminating the traditional method of using animals for isolating pure cultures of pathogens. Ticks are fed on a silicone membrane covering blood circulated at a constant temperature, mimicking living animals. The laboratory tick feeding system will be a useful method to improve live vaccine development for tick-borne diseases, to study pathogen-tick vector interaction, and the tick-mammalian host interface. This new laboratory method not only improves pathogen isolation but is responsive to the goal of improving animal welfare by replacement, reduction and refinement of animal use in the study of tick transmitted diseases.

Scientific Publication:
Vimonish R, Johnson WC, Mousel R, Brayton KA, Scoles GA, Noh SM, Ueti MW. 2020. Quantitative analysis of *Anaplasma marginale* acquisition and transmission by *Dermacentor andersoni* fed *in vitro.* Scientific Reports. 10:470. https://doi.org/10.1038/s41598-019-57390-y

Improving the Accuracy of Diagnostic Tests for Bovine Babesiosis
Bovine babesiosis (also known as cattle tick fever) is a tick-transmitted disease caused by the protozoan parasites *Babesia bovis* and *B. bigemina. Babesia* parasites can be transmitted by tick vectors to cattle of any age, which can result in 90% mortality in naive adults. Babesiosis was a significant problem in the southern United States until the eradication of the tick vector in the 1940's. The United States imports a million head of cattle yearly from Mexico, where Babesiosis and cattle fever ticks are present. The control measure used to prevent babesiosis from coming to the United States is treating all cattle arriving from Mexico with acaricides to eliminate cattle fever ticks. But, the recent discovery of acaricide resistant tick populations capable of transmitting *babesia species,* and the re-emergence and spread of cattle fever ticks by wildlife on the Texas - Mexico border is increasing the risk of the return of bovine babesiosis, which is a major concern for the U.S. livestock industry. Therefore, scientists at the Animal Diseases Research Unit, Pullman, Washington, have developed a modified diagnostic test with greater accuracy than the current standard tests to determine the infection prevalence of bovine babesiosis on the United States-Mexico border. This improved diagnostic test was fully developed in collaboration with a commercial partner and is now available for use to determine whether bovine babesiosis infections of cattle and wildlife are moving beyond the United States-Mexico border.

Scientific Publication:
Chung JC, Suarez CE, Bandaranayake-Mudiya NL, Bandaranayake-Mudiya N, Rzepka, J, Heiniger TJ, Chung G, Lee SS, Adams E, Yun G. Waldron SJ. 2017. A novel modified-indirect ELISA based on spherical body protein 4 for detecting antibody during acute and long-term infections with diverse babesia bovis strains. Parasites & Vectors. 10(1):77. https://doi.org/10.1186/s13071-017-2016-9

Identifying Conserved Epitopes for use in a Broadly Protective Vaccine Against Bovine Anaplasmosis

Anaplasma marginale, the causative agent of bovine anaplasmosis, is a tick-borne bacterial pathogen of cattle that causes economic losses to cattle producers throughout the world. This pathogen is widespread in the United States, with greater than 80 percent of cattle in some regions of Texas being infected. One of the constraints of vaccine development is the variability of *A. marginale* strains such that one vaccine is not protective against all strains. In collaboration with colleagues at Washington State University in Pullman, Washington, and the University of Ghana in Legon, Ghana, ARS researchers at the Animal Diseases Research Unit, Pullman, Washington, have identified a vaccine candidate that is broadly conserved among geographically diverse strains, including those from North America, Mexico, Ghana and Australia. Importantly, this vaccine candidate is recognized by T cells, is surface exposed and antigenic. This finding is important for the development of a vaccine that is broadly protective against diverse *A. marginale* strains.

Scientific Publication:
Deringer JR, Forero-Becerra EG, Ueti MW, Turse JE, Futse JE, Noh SM, Palmer GH, Brown WC. 2016. Identification of a T-cell epitope that is globally conserved among outer membrane proteins (OMPs) OMP7, OMP8, and OMP9 of Anaplasma marginale strains and with OMP7 from the A. marginale subsp. centrale vaccine strain. Clinical and Vaccine Immunology. 24(1):e00406-16. doi: 10.1128/CVI.00406-16. https://doi.org/10.1128/CVI.00406-16

Transgenic Babesia bovis Lacking 6-Cys Sexual-stage Genes as the Foundation for a Non-transmissible Live Vaccine Against Bovine Babesiosis

Babesia bovis, a tick-borne parasite responsible for bovine babesiosis has a complex life cycle including sexual development in its *Rhipicephalus microplus* tick vector. Understanding the molecular mechanisms involved in sexual development in ticks may provide a new approach for developing future-generation transmission blocking vaccines. Recently discovered sexual markers 6-CysA and 6-CysB genes of *B. bovis* were found to be expressed during the tick-stage of *B. bovis*. ARS scientists at the Animal Diseases Research Unit, Pullman, Washington, hypothesized that disruption of both 6-CysA and 6-CysB in *B. bovis* would result in unaltered ability of the parasite to invade and grow in red blood cells but lose the ability to transmit to ticks. Parasites deficient in genes required for sexual reproduction could be the foundation for genetically-defined, non-transmissible live vaccines against bovine babesiosis that could help reduce the burden of disease globally.

Scientific Publication:
Alzan HF, Lau A, Knowles DP, Herndon DR, Ueti MW, Scoles GA, Kappmeyer LS, Suarez CE. 2016. Expression of 6-Cys gene superfamily defines *Babesia bovis* sexual stage development

within *Rhipicephalus microplus*. PLoS One. doi:10.1371/journal.pone.0163791. https://journals.plos.org/plosone/article/file?type=printable&id=10.1371/journal.pone.0163791

Alzan A, Silva M, Davis W, Herndon DR, Schneider DA, Suarez CE. 2017. Geno- and phenotypic characteristics of a transfected *Babesia bovis* 6-Cys-E knockout clonal line. Parasites & Vectors. 10(1):214. https://doi.org/10.1186/s13071-017-2143-3

Component 7: Transmissible Spongiform Encephalopathies

Rationale for the research:

Transmissible spongiform encephalopathies (TSEs) include several fatal diseases of people and animals involving degeneration of the nervous system and brain function. Important TSEs include Creutzfeldt-Jakob disease (CJD), the primary human prion disease; Scrapie of sheep and goats; Chronic Wasting Disease (CWD) of deer, elk, and moose; and Bovine Spongiform Encephalopathy (BSE), also called "mad cow," which is the cause of variant CJD (vCJD) in people and the only TSE known to have crossed the species barrier from animals to people.

Although significant scientific advances have been made, the research conducted to date has yet to deliver many of the concrete solutions needed to safeguard people and animals from devastating prion diseases. A critical concern is the potential for environmental, genetic, or iatrogenic events to lead to new variant TSEs that are infectious and zoonotic.

Stakeholders representing the cattle, sheep, goat, captive cervids, and wildlife industries that responded to the 2015 ARS animal health national survey identified research to control and eradicate TSEs a national priority.

Research Needs:

Important gaps remain in our basic understanding of the pathobiology of animal prion diseases. It is widely assumed that the oral route of infection is important in the pathogenesis of naturally occurring TSEs of livestock and cervids; however, basic research is needed to understand the mechanisms of transmission of TSE agents from the initial site of entry to the central nervous system. A notable feature of prion diseases is a lack of detectable immune responses and inflammation during the course of a prion infection, even though immune system cells may carry prions to target tissues. To date, research in animals suggests that prion accumulation may be largely influenced by the host species affected rather than the TSE involved. An investment in comparative pathology, which has not received much experimental attention, is needed to advance research programs in epidemiology and diagnostic discovery.

Prion diseases have stimulated intense scientific scrutiny since it was first proposed that the infectious agent was devoid of nucleic acid. Despite this finding, host genetics has played a key role in understanding the pathobiology and clinical aspects of prion diseases through the effects of a series of polymorphisms and mutations in the prion protein gene. Nevertheless, the functional genomics of disease resistance are not completely understood, and recent research suggests genetic variations may lead to different clinical outcomes. There is a need to implement research aimed at utilizing powerful computational biology and bioinformatic approaches, along with traditional animal breeding experiments, to steadily improve our understanding of mechanisms of genetic disease resistance.

Important gaps remain in our arsenal of diagnostic tools for early detection and countermeasures to prevent disease outbreaks, transmission, and spread. Current diagnostic tests were validated for use only on post-mortem samples; simple, sensitive, cost-effective ante mortem tests have yet

to be developed. There is a critical need to improve diagnostics methods for surveillance, including the discovery of an ante mortem test for early detection and implementation of intervention strategies.

Anticipated Products from the Action Plan:

- Scientific information on the mechanisms responsible for the development of multiple TSE strains within a host species.
- Scientific information on the manner in which prions enter the nervous system from peripheral sites of exposure such as a host's gastrointestinal tract, nasal mucosa, skin, and eyes.
- Scientific information on the mechanisms by which prion spread within the nervous system.
- Scientific information on the mechanisms that control prion disease incubation times.
- Scientific information on prion neuropathogenesis.
- Scientific information on prion distribution in goats infected with Scrapie.
- Scientific information on prion distribution in sheep infected with atypical Scrapie.
- Identification of genetic variations associated with disease susceptibility.
- Scientific information on the correlation between host genotypes and the phenotypes of prion agents.
- Identification of genetic factors controlling susceptibility of goats to sheep Scrapie.
- Scientific information to evaluate the effectiveness of disease resistance breeding programs in sheep.
- Scientific information to evaluate sheep ARR/ARR genotype for resistance to different TSE strains.
- Scientific information on the influence of genetics on BSE incubation time and the frequency of animals carrying the E211K allele.
- TSE diagnostic test capable of detecting low levels of abnormal prions (i.e., key step to enable the development of an ante mortem test that can identify disease during the early stages of incubation).
- Validation of existing biopsy-based TSE tests in goats, deer, and elk.
- Rapid biochemical methods for strain typing.
- Validated murine models for strain typing.
- Improved diagnostics for TSEs in bodily fluids, including blood in host species where this might be possible.
- Technologies to distinguish infectious prions from normal cellular prion proteins.
- Determination of the prevalence of proteinase K sensitive prion in the various TSEs and potential of this form to cause disparate results between IHC, WB, and ELISA tests.

Impact:

The impact of the research included scientific information to enable regulatory and action agencies to promulgate science-based control programs. The development of diagnostics and countermeasures has enhanced current federal and state control and eradication programs for scrapie and CWD.

COMPONENT 7: SELECTED ACCOMPLISHMENTS

Problem Statement 7A: *Pathobiology of Prion Strains*

Swine are Potential Hosts for the Scrapie Agent

A naturally occurring prion disease has not been recognized in swine, but the agent of bovine spongiform encephalopathy does transmit to swine by experimental routes. Swine are thought to have a robust species barrier when exposed to the naturally occurring prion diseases of other species, but the susceptibility of swine to the agent of sheep scrapie has not been thoroughly tested. Therefore, ARS scientists at the National Animal Disease Center, Ames, Iowa, conducted this experiment to test the susceptibility of swine to U.S. scrapie isolates by intracranial and oral inoculation. Necropsies were done on a subset of animals at approximately 6 months post inoculation (PI): the time the pigs were expected to reach market weight. Remaining pigs were maintained and monitored for clinical signs of transmissible spongiform encephalopathies (TSE) until study termination at 80 months PI or when removed due to disease. Brain samples were examined by multiple diagnostic approaches, and for a subset of pigs in each inoculation group, bioassay in mice expressing porcine prion protein. At 6 months PI, no evidence of scrapie infection was noted by any diagnostic method. However, at 51 months of incubation or greater, five animals were positive by one or more diagnostic methods. Furthermore, positive bioassay results were obtained from all inoculated groups (oral and intracranial; market weight and end of study) suggesting that swine are potential hosts for the agent of scrapie.

Scientific Publication:

Greenlee JJ, Kunkle RA, Smith JD, West Greenlee, MH. 2016. Scrapie in swine: a diagnostic challenge. Food Safety. 4(4):110-114. https://doi.org/10.14252/foodsafetyfscj.2016019

Problem Statement 7B: *Genetics of Prion Diseases Susceptibility*

The Potential Role of Prion Genetics in Cattle with Classical Versus Atypical Bovine Spongiform Encephalopathy

In 2006, a case of atypical bovine spongiform encephalopathy (BSE-H) was diagnosed in a cow that was associated with a heritable genetic mutation in the bovine prion protein gene (PRNP). Unlike classical BSE, which is caused by feeding cattle with contaminated BSE material, it is thought that atypical BSE cases may occur spontaneously in cattle due to genetic mutations in the PRNP. ARS scientists at the National Animal Disease Center, Ames, Iowa, conducted a series of pathogenicity studies and showed that the survival time of the cattle with the genetic mutation inoculated with BSE-H was shorter (10 months) than cattle without the mutation (18 months). This genetic effect was not observed when cattle, with or without the genetic mutation, were inoculated with classical BSE. Their survival time was 26 months, regardless of whether the cattle had the genetic mutation or not. The results of these studies demonstrate that the genetic mutation associated with atypical BSE exhibits a number of features that differ from classical BSE. Understanding the association between this genetic mutation and BSE provides important information on the potential public health risk of atypical BSE.

Scientific Publication:
Moore SJ, West Greenlee MH, Smith JD, Vrentas CE, Nicholson EM, Greenlee JJ. 2016. A Comparison of Classical and H-Type Bovine Spongiform Encephalopathy Associated with E211K Prion Protein Polymorphism in Wild-Type and EK211 Cattle Following Intracranial Inoculation. Front Vet Sci. 3:78. https://doi.org/10.3389/fvets.2016.00078

Identifying and Breeding Goats Resistant to Scrapie
Scrapie is a fatal brain disease of goats and sheep for which there is no treatment. Scrapie is caused by the progressive accumulation of an abnormal form of the prion protein and loss of brain cells that often leads to abnormal behavior, lack of coordination, gait abnormalities and reduced mobility and body condition, but which always leads to death. Historically, a single diagnosis of scrapie results in permanent quarantine or euthanasia of all goats and sheep on the farm. The sheep industry has had an important tool in the struggle to eradicate scrapie in the form of genetic resistance. The strong resistance to scrapie in sheep comes from the R171 gene allele, which changes the amino acid at position 171 of the prion protein to arginine (written simply as "R"). Conversely, genetic resistance has not been available to the goat industry. This recently changed with the discovery of two naturally occurring prion gene alleles in goats that have shown exceptional promise for conferring resistance. The first is S146, which denotes a serine (S) amino acid at prion protein position 146. The second is K222, which denotes a lysine (K) amino acid at position 222. Through an experiment that has lasted close to 10 years, ARS scientists at the Animal Diseases Research Unit, Pullman, Washington, along with other laboratories around the world have shown that even one copy of either S146 or K222 confers strong resistance to classical scrapie in goats, much like R171 in sheep. Although the USDA National Scrapie Eradication Program has not yet formally recognized these alleles, USDA is planning pilot genetic based cleanup plans for goats similar to what has been done for sheep. Scrapie resistance should enhance goat breeding programs and goat health in major ways. Importantly, breeding scrapie resistant goats will benefit all small ruminant producers by reducing scrapie in the United States and help work towards import/export status for the United States as a scrapie-free country.

Scientific Publication:
Cinar MU, Schneider DA, Waldron DF, O'Rourke KI, White SN. 2018. Goats singly heterozygous for PRNP S146 or K222 orally inoculated with classical scrapie at birth show no disease at ages well beyond six years. Vet J. 233:19-24. doi: 10.1016/j.tvjl.2017.12.019. https://doi.org/10.1016/j.tvjl.2017.12.019

Problem Statement 7C: *Diagnostics, Detection, and Surveillance*

Improved Diagnosis of Transmissible Spongiform Encephalopathies (TSEs) of Sheep, Goats, and Elk
Gold standard diagnostic testing for the TSEs of small and wild ruminants in the United States is performed by immunohistochemistry analysis of formalin fixed tissues using an automated, monoclonal antibody-based system with reagents and procedures developed by the Animal Disease Research Unit, Pullman, Washington. ARS researchers in Pullman, Washington, and their partners at Washington State University demonstrated that infectious prions can be detected from much smaller blood sample volumes, even during preclinical infection. This study supports

further development of a safe and highly efficient blood-based diagnostic test for preclinical scrapie infection in sheep.

Scientific Publication:
Dassanayake RP, Truscott TC, Zhuang D, Schneider DA, Madsen-Bouterse SA, Young AJ, Stanton JB, Davis WC, O'Rourke KI. 2015. Classical natural ovine scrapie prions detected in practical volumes of blood by lamb and transgenic mouse bioassays. J Vet Sci. 16(2):179-86. https://doi.org/10.4142/jvs.2015.16.2.179

Sensitive and Specific Detection of Classical Scrapie Prions in the Brains of Goats
Scrapie is a transmissible spongiform encephalopathy that causes fatal neurodegenerative disorders in goats and sheep. "Real-time quaking-induced conversion" is a rapid, specific and highly sensitive detection assay used to detect low levels of abnormal scrapie prion proteins (PrPSc). Although this sensitive assay has been successfully used to detect abnormal prion proteins in various tissues from humans and animals, including sheep, tissues from goats infected with scrapie have not yet been tested. ARS scientists at the Animal Diseases Research Unit, Pullman, Washington, in collaboration with scientists at Washington State University, evaluated whether abnormal prion proteins could be detected in the brain tissues of goats with scrapie using this sensitive assay, optimized reaction conditions to improve scrapie detection in goats, and compared the performance of this sensitive assay for the detection of scrapie with the more commonly used prion detection methods. ARS scientists further optimized assay conditions for sensitive and specific detection of goat scrapie in clinical animals. The results from these studies provided good discrimination between scrapie-infected and normal goat brain samples. Importantly, these studies indicated that this highly sensitive assay was at least 10,000-fold more sensitive than the commonly used prion detection methods for the detection of scrapie activity in goat brain samples.

Scientific Publication:
Dassanayake RP, Orrú CD, Hughson AG, Caughey B, Graça T, Zhuang D, Madsen-Bouterse SA, Knowles DP, Schneider DA. 2016. Sensitive and specific detection of classical scrapie prions in the brains of goats by real-time quaking-induced conversion. J Gen Virol. 97(3):803-812. doi: 10.1099/jgv.0.000367. https://dx.doi.org/10.1099%2Fjgv.0.000367

A Method for Long Term Storage of Prion Protein Used in Diagnostic Tests
Prion diseases include Scrapie, Chronic Wasting Disease and mad cow disease. Recent advances in prion disease diagnostic methods include the amplification of the amount abnormal prion protein is in a sample. One such technique, known as RT-QuIC, requires a steady supply of freshly purified prion protein, which necessitates constant production that is not sustainable in a diagnostic laboratory setting. ARS researchers at the National Animal Disease Center, Ames, Iowa, developed a method to dry and preserve the prion protein for long term storage (months or years). This allows for production of the prion protein in larger quantities, and it can be shipped to diagnostic laboratories facilitating widespread use of RT-QuIC as a diagnostic method.

Scientific Publication:
Hwang S, Tatum T, Lebepe-Mazur S, Nicholson EM. 2018. Preparation of lyophilized recombinant prion protein for TSE diagnosis by RT-QuIC. BMC Res Notes. 11(1):895. doi: 10.1186/s13104-018-3982-5. https://doi.org/10.1186/s13104-018-3982-5

Research Impact beyond Animal Health: Biomedical Research

The Silver Bullet: Utilizing Vesicular Stomatitis Virus to Treat Cancer
Vesicular stomatitis virus (VSV) is an animal pathogen that causes vesicular disease in horses, cattle and pigs. VSV is a bullet-shaped enveloped virus that grows rapidly producing powerful immune response that can be used as a vaccine to prevent and/or treat infectious disease and cancer in humans and animals. Modified VSV has been shown to replicate selectively in and kill cancer cells and were not pathogenic during clinical trials in humans and dogs. However, there is concern that these VSV vectors could be pathogenic and transmissible to farm animals (e.g. pigs). ARS scientists at the Plum Island Animal Disease Center, Orient, New York, collaborated with scientists at the Mayo Clinic, Rochester, Minnesota, and showed that VSV is safe not only to humans and dogs, but also in pigs. These studies pave the way for further development of this promising cancer therapy.

Scientific Publication:
Velazquez-Salinas L, Niak S, Peng K, Pauszek SJ, Rodriguez LL. 2017. Oncolytic recombinant vesicular stomatitis virus (VSV) is nonpathogenic and non-transmissible in pigs, a natural host of VSV. Human Gene Therapy. 28(2):108-115. https://doi.org/10.1089/humc.2017.015

Appendices

APPENDIX 1 – *Research Projects in National Program 103*
APPENDIX 2 – *Publications by Research Project*
APPENDIX 3 – *ARS Technology Transfer*
APPENDIX 4 – *Research Collaborations*

APPENDIX 1

National Program 103 – Animal Health ACCOMPLISHMENT REPORT 2016 – 2020

List of Research Projects

Albany, California Western Regional Research Center Produce Safety and Microbiology Research Unit	
2030-32000-010-00D	Immunodiagnostics to Detect Prions and Other Important Animal Pathogens; Robert Hnasko (P), Christopher Silva and Vivian Wu
Ames, Iowa National Animal Disease Center Infectious Bacterial Diseases Research Unit	
5030-32000-221-00D	Characterization of Antigens, Virulence Markers, and Host Immunity in the Pathogenesis of Johne's Disease; Judith Stabel (P) and John Bannantine
5030-32000-222-00D	Characterize the Immunopathogenesis and Develop Diagnostic and Mitigation Strategies to Control Tuberculosis in Cattle and Wildlife; Mitchell Palmer (P)
5030-32000-223-00D	Characterization of the Pathogenesis and Antigen Expression in Spirochete Diseases; David Alt (P), Jarlath Nally, Darrell Bayles and Jennifer Wilson-Welder
5030-32000-224-00D	Pathogenesis and Development of Improved Diagnostic and Control Strategies for Brucellosis in Livestock and Wildlife; Steven Olsen (P) and Paola Boggiatto
National Animal Disease Center Ruminant Diseases and Immunology Research Unit	
5030-32000-115-00D	Non-antibiotic Approaches to Control Mastitis; John Lippolis (P) and Timothy Reinhardt
5030-32000-116-00D	Identification of Disease Mechanisms and Control Strategies for Bacterial Respiratory Pathogens in Ruminants; Randy Sacco (P), Fred Tatum, Hao Ma, Rohana Dassanayake, Eduardo Casas, Robert Briggs and Karen Register
5030-32000-117-00D	Identification of Disease Mechanisms and Control Strategies for Viral Respiratory Pathogens of Ruminants; John Neill (P), Hao Ma, Rohana Dassanake, Randy Sacco, Shollie Falkenberg and Eduardo Casas
National Animal Disease Center Virus and Prion Research Unit	
5030-32000-114-00D	Pathobiology, Genetics, and Detection of Transmissible Spongiform Encephalopathies; Eric Nicholson (P), Kelly Lager and Justin Greenlee
5030-32000-118-00D	Intervention Strategies to Control Endemic and New and Emerging Viral Diseases of Swine; Kay Faaberg (P), Alexandra Buckley, Laura Miller and Kelly Lager
5030-32000-119-00D	Non-Antibiotic Strategies to Control Priority Bacterial Infections in Swine; Susan Brockmeier (P), Kelly Lager and Tracy Nicholson
5030-32000-120-00D	Intervention Strategies to Control Influenza A Virus Infection in Swine; Amy Vincent (P), Tavis Anderson and Kelly Lager

Athens, Georgia U.S. National Poultry Research Center Southeast Poultry Research Laboratory Endemic Poultry Viral Diseases Research Unit	
6040-32000-073-00D	Intervention Strategies to Prevent and Control Enteric Diseases of Poultry; Qingzhong Yu (P) and John Dunn
6040-32000-074-00D	Genetic and Biological Determinants of Avian Herpesviruses Pathogenicity, Transmission, and Evolution to Inform the Development of Effective Control Strategies; Stephen Spatz (P), Taejoong Kim, John Dunn and Hans Cheng *****Project managed by Avian Disease and Oncology Research*****
6040-32000-075-00D	Intervention Strategies to Prevent and Control Immunosuppressive Viruses of Poultry Associated with Secondary Pathogen Infections; David Swayne (P) and John Dunn
6040-32000-076-00D	Integrative Strategies to Mitigate Enteric Diseases of Poultry; David Swayne (P)
Southeast Poultry Research Laboratory Exotic and Emerging Avian Viral Diseases Research Unit	
6040-32000-066-00D	Intervention Strategies to Prevent and Control Disease Outbreaks Caused by Emerging Strains of Avian Influenza Viruses; Mary Pantin-Jackwood (P), David Swayne, David Suarez, Erica Spackman and Darrell Kapczynski
6040-32000-072-00D	Intervention Strategies to Predict, Prevent and Control Disease Outbreaks Caused by Emerging Strains of Virulent Newcastle Disease Viruses; Claudio Afonso (P) and David Suarez
Beltsville, Maryland Animal Biosciences & Biotechnology Laboratory	
8042-32000-107-00D	Non-antibiotic Strategies to Control Enteric Diseases of Poultry; Hyun Lillehoj (P) and Charles Li
Animal Parasitic Diseases Laboratory	
8042-31000-107-00D	Molecular Approaches to Control Intestinal Parasites that Affect the Microbiome in Swine and Small Ruminants; Joseph Urban (P), Robert Li and Dante Zarlenga
8042-32000-102-00D	Evaluation of Swine Immunity and Development of Novel Immune and Genomic Intervention Strategies to Prevent and/or Treat Respiratory Diseases of Swine; Joan Lunney (P)
8042-32000-105-00D	Immune, Molecular, and Ecological Approaches for Attenuating GI Nematode Infections of Ruminants; Dante Zarlenga (P) and Wenbin Tuo
8042-32000-111-00D	Development of Control and Intervention Strategies for Avian Coccidiosis; Mark Jenkins (P)
Clay Center, Nebraska U.S. Meat Animal Research Center Animal Health Research	
3040-32000-034-00D	Genomic Intervention Strategies to Prevent and/or Treat Respiratory Diseases of Ruminants; Carol Chitko-McKown (P), Aspen Workman, Gregory Harhay, Michael Clawson and Michael Heaton
Manhattan, Kansas Center for Grain and Animal Health Research Arthropod-Borne Animal Diseases Research Unit	
3020-32000-009-00D	Rift Valley Fever Pathogenesis, Epidemiology, and Control Measures; William Wilson (P), Leela Noronha, Dana Mitzel and Barbara Drolet

3020-32000-010-00D	Orbivirus Pathogenesis, Epidemiology, and Control Measures; Barbara Drolet (P), William Wilson, Dana Mitzel, Dana Nayduch and Leela Noronha
3020-32000-013-00D	Ecology of Vesicular Stomatitis Virus (VSV) in North America; Barbara Drolet (P), Lee Cohnstaedt, Dana Mitzel and William Wilson
3020-32000-014-00D	Japanese Encephalitis Virus Prevention and Mitigation Strategies; Leela Noronha (P), David McVey, Barbara Drolet, Dana Mitzel and William Wilson
Manhattan, Kansas National Bio and Agro-Defense Facility	
3022-32000-012-00D	Training of Biodefense Research Workforce for the National Bio and Agro-defense Facility (NBAF); David McVey (P)
3022-32000-017-00D	National Bio and Agro-Defense Facility Scientists Project; David McVey (P)
Mississippi State, Mississippi Poultry Research Center	
6064-13000-013-00D	Systems Approach to Understanding and Mitigating Avian Escherichia coli Infections and Antimicrobial Resistance in the Poultry Environment; Jeffrey Evans (P), Leigh Spencer and Joseph Purswell
6064-32000-012-00D	Transmission, Pathogenesis, and Control of Avian Mycoplasmosis; Jeffrey Evans (P), Joseph Purswell, Scott Branton and Spencer Leigh
Orient Point, New York Plum Island Animal Disease Center	
8064-32000-059-00D	Ecology of Vesicular Stomatitis Virus (VSV) in North America; Luis Rodriguez (P), James Zhu and Jonathan Arzt
8064-32000-060-00D	Countermeasures to Control and Eradicate Foreign Animal Diseases of Swine; Manuel Borca (P), James Zhu, Luis Rodriguez, Jonathan Arzt and Douglas Gladue
8064-32000-061-00D	Intervention Strategies to Support the Global Control and Eradication of Foot-and-Mouth Disease Virus (FMDV); Luis Rodriguez (P), Manuel Borca, Teresa De Los Santos, Aida Rieder, James Zhu and Jonathan Arzt
Pullman, Washington Animal Disease Research Unit	
2090-32000-035-00D	Genetic Impact and Improved Diagnostics for Sheep and Goat Transmissible Spongiform Encephalopathies; David Schneider (P), Stephen White, Naomi Taus, David Herndon, Lowell Kappmeyer, Cristina Cunha and Michelle Mousel
2090-32000-036-00D	Identification of Host Factors and Immunopathogenesis of Pneumonia in Domestic and Bighorn Sheep; Stephen White (P), David Herndon, Lowell Kappmeyer and Michelle Mousel
2090-32000-037-00D	Diagnostic and Control Strategies for Malignant Catarrhal Fever; Cristina Cunha (P)
2090-32000-038-00D	Identification of Tick Colonization Mechanisms and Vaccine Development for Anaplasmosis; Susan Noh (P), David Herndon, Lowell Kappmeyer and Massaro Ueti
2090-32000-039-00D	Development of Detection and Control Strategies for Bovine Babesiosis and Equine Piroplasmosis; Massaro Ueti (P), Lowell Kappmeyer, Naomi Taus, Carlos Suarez and Lindsay Fry

* For the sake of consistency, projects are listed and organized in Appendix 1 according to the ARS project number used to track projects in the Agency's internal database. A (P) after a scientist's name indicates the project's principal investigator.

APPENDIX 2

National Program 103 – Animal Health
ACCOMPLISHMENT REPORT 2016 – 2020

Publications by Research Project

* For the sake of consistency, projects are listed and organized in Appendix 2 according to the ARS project number used to track projects in the Agency's internal database. A (P) after a scientist's name indicates the project's principal investigator.

Albany, California
Western Regional Research Center
Produce Safety and Microbiology Research Unit

2030-32000-010-00D
Immunodiagnostics to Detect Prions and Other Important Animal Pathogens; Robert Hnasko (P), Christopher Silva and Vivian Wu

Silva, C.J., Erickson-Beltran, M.L., Duque Velásquez, C., Aiken, J.M., McKenzie, D. 2020. A General Mass Spectrometry-Based Method of Quantitating Prion Polymorphisms from Heterozygous Chronic Wasting Disease-Infected Cervids. Analytical Chemistry. 92(1):1276-1284. https://doi.org/10.1021/acs.analchem.9b04449

Hnasko, R.M., Lin, A.V., McGarvey, J.A., Stanker, L.H. 2018. Enhanced detection of infectious prions by direct ELISA from the brains of asymptomatic animals using DRM2-118 monoclonal antibody and Gdn-HCl. Journal of Immunological Methods. 456:38-43. https://doi.org/10.1016/j.jim.2018.02.010.

Hnasko, R.M., Lin, A.V., Stanker, L.H., McGarvey, J.A. 2018. A bioassay for the optimization of macrophage conditioned medium (MCM) as culture supplement used to promote hybridoma cell survival and growth. Monoclonal Antibodies in Immunodiagnosis and Immunotherapy. 37(3):126-133. https://doi.org/10.1089/mab.2018.0008.

Sevillano, A.M., Fernández-Borges, N., Younas, N., Wang, F., Elezgarai, S.R., Bravo, S., Vázquez-Fernández, E., Rosa, I., Eraña, H., Gil, D., Veiga, S., Vidal, E., Erickson-Beltran, M.L., Guitián, E., Silva, C.J., Nonno, R., Ma, J., Castilla, J., Requena, J.R. 2018. Recombinant PrPSc shares structural features with brain-derived PrPSc suggesting that they have a similar architecture: Insights from limited proteolysis. PLoS Pathogens. 14(1):e1006797. https://doi.org/10.1371/journal.ppat.1006797.

Silva, C.J. 2018. Food forensics: using mass spectrometry to detect foodborne protein contaminants as exemplified by Shiga toxin variants and prion strains. Journal of Agricultural and Food Chemistry. 66(32):8435-8450. https://doi.org/10.1021/acs.jafc.8b01517.

Babrak, L.M., McGarvey, J.A., Stanker, L.H., Hnasko, R.M. 2017. Identification and verification of hybridoma-derived monoclonal antibody variable region sequences using recombinant DNA technology and mass spectrometry. Molecular Immunology. 90:287-294. https://doi.org/10.1016/j.molimm.2017.08.014.

Silva, C.J., Erickson-Beltran, M.L., Martín-Burriel, I., Badiola, J., Requena, J.R., Bolea, R. 2017. Determining the relative susceptibility of four prion protein genotypes to atypical scrapie. Analytical Chemistry. 90(2):1255-1262. https://doi.org/10.1021/acs.analchem.7b03985.

Silva, C.J., Erickson-Beltran, M.L., Hui, C., Badiola, J.J., Nicholson, E.M., Requena, J.R., Bolea, R. 2016. Quantitating PrP polymorphisms present in prions from heterozygous scrapie-infected sheep. Analytical Chemistry. 89(1):854-861. https://doi.org/10.1021/acs.analchem.6b03822

Llorens, F., Thüne, K., Schmitz, M., Ansoleaga, B., Frau-Méndez, M.A., Cramm, M., Tahir, W., Gotzmann, N., Berjaoui, S., Carmona, M., Silva, C.J., Fernandez-Vega, I., José Zarranz, J., Zerr, I., Ferrer, I. 2016. Identification of new molecular alterations in fatal familial insomnia. Human Molecular Genetics. 25(12):2417-2436. https://academic.oup.com/hmg/article/25/12/2417/2525733

Silva, C.J., Erickson-Beltran, M.L., Dynin, I.C. 2016. Covalent Surface Modification of Prions: A Mass Spectrometry-Based Means of Detecting Distinctive Structural Features of Prion Strains. Biochemistry. 55(6):894-902. https://pubs.acs.org/doi/10.1021/acs.biochem.5b01068

Ames, Iowa

National Animal Disease Center

Infectious Bacterial Diseases Research Unit

5030-32000-221-00D

Characterization of Antigens, Virulence Markers, and Host Immunity in the Pathogenesis of Johne's Disease; Judith Stabel (P) and John Bannantine

Bannantine, J. P., J. R. Stabel, E. Laws, D. Cardieri MC, and C. D. Souza. 2015. Mycobacterium Avium Subspecies Paratuberculosis Recombinant Proteins Modulate Antimycobacterial Functions of Bovine Macrophages. PLoS One. 10(6):e0128966. http://doi.org/10.1371/journal.pone.0128966.

Bannantine, J. P. and A. M. Talaat. 2015. Controlling Johne's Disease: Vaccination Is the Way Forward. Front Cell Infect Microbiol. 5:2. http://doi.org/10.3389/fcimb.2015.00002.

Everman, J. L., T. M. Eckstein, J. Roussey, P. Coussens, J. P. Bannantine, and L. E. Bermudez. 2015. Characterization of the Inflammatory Phenotype of Mycobacterium Avium Subspecies Paratuberculosis Using a Novel Cell Culture Passage Model. Microbiology. 161(7):1420-34. http://doi.org/10.1099/mic.0.000106.

Godden, S. M., S. Wells, M. Donahue, J. Stabel, J. M. Oakes, S. Sreevatsan, and J. Fetrow. 2015. Effect of Feeding Heat-Treated Colostrum on Risk for Infection with Mycobacterium Avium Ssp. Paratuberculosis, Milk Production, and Longevity in Holstein Dairy Cows. J Dairy Sci. 98(8):5630-41. http://doi.org/10.3168/jds.2015-9443.

Leite, F. L., L. B. Eslabao, B. Pesch, J. P. Bannantine, T. A. Reinhardt, and J. R. Stabel. 2015. Zap-70, Ctla-4 and Proximal T Cell Receptor Signaling in Cows Infected with Mycobacterium Avium Subsp. Paratuberculosis. Vet Immunol Immunopathol. 167(1-2):15-21. https://doi.org/10.1016/j.vetimm.2015.06.017

Leite, F. L., T. A. Reinhardt, J. P. Bannantine, and J. R. Stabel. 2015. Envelope Protein Complexes of Mycobacterium Avium Subsp. Paratuberculosis and Their Antigenicity. Vet Microbiol 175(2-4):275-85. http://doi.org/10.1016/j.vetmic.2014.11.009.

Magombedze, G., S. Eda, and J. Stabel. 2015. Predicting the Role of IL-10 in the Regulation of the Adaptive Immune Responses in Mycobacterium Avium Subsp. Paratuberculosis Infections Using Mathematical Models. PLoS One. 10(11):e0141539. http://doi.org/10.1371/journal.pone.0141539.

Martcheva, M., S. Lenhart, S. Eda, D. Klinkenberg, E. Momotani, and J. Stabel. 2015. An Immuno-Epidemiological Model for Johne's Disease in Cattle. Vet Res. 46:69. http://doi.org/10.1186/s13567-015-0190-3.

Mitchell, R. M., Y. Schukken, A. Koets, M. Weber, D. Bakker, J. Stabel, R. H. Whitlock, and Y. Louzoun. 2015. Differences in Intermittent and Continuous Fecal Shedding Patterns between Natural and Experimental Mycobacterium Avium Subspecies Paratuberculosis Infections in Cattle. Vet Res. 46:66. http://doi.org/10.1186/s13567-015-0188-x.

Bannantine, J. P., C. K. Lingle, P. R. Adam, K. X. Ramyar, W. J. McWhorter, J. R. Stabel, W. D. Picking, and B. V. Geisbrecht. 2016. Nlpc/P60 Domain-Containing Proteins of Mycobacterium Avium Subspecies Paratuberculosis That Differentially Bind and Hydrolyze Peptidoglycan. Protein Sci. 25(4):840-51. http://doi.org/10.1002/pro.2884.

Capsel, R. T., C. O. Thoen, T. A. Reinhardt, J. D. Lippolis, R. Olsen, J. R. Stabel, and J. P. Bannantine. 2016. Composition and Potency Characterization of Mycobacterium Avium Subsp. Paratuberculosis Purified Protein Derivatives. PLoS One. 11(5):e0154685. http://doi.org/10.1371/journal.pone.0154685.

Hempel, R. J., J. P. Bannantine, and J. R. Stabel. 2016. Transcriptional Profiling of Ileocecal Valve of Holstein Dairy Cows Infected with Mycobacterium Avium Subsp. Paratuberculosis. PLoS One. 11(4):e0153932. http://doi.org/10.1371/journal.pone.0153932.

Krueger, L. A., D. C. Beitz, S. B. Humphrey, and J. R. Stabel. 2016. Gamma Delta T Cells Are Early Responders to Mycobacterium Avium Ssp. Paratuberculosis in Colostrum-Replete Holstein Calves. J Dairy Sci. 99(11):9040-50. http://doi.org/10.3168/jds.2016-11144.

Krueger, L. A., T. A. Reinhardt, D. C. Beitz, R. L. Stuart, and J. R. Stabel. 2016. Effects of Fractionated Colostrum Replacer and Vitamins a, D, and E on Haptoglobin and Clinical Health in Neonatal Holstein Calves Challenged with Mycobacterium Avium Ssp. Paratuberculosis. J Dairy Sci. 99(4):2884-95. http://doi.org/10.3168/jds.2015-10395.

Kugadas, A., E. A. Lamont, J. P. Bannantine, F. M. Shoyama, E. Brenner, H. K. Janagama, and S. Sreevatsan. 2016. A Mycobacterium Avium Subsp. Paratuberculosis Predicted Serine Protease Is Associated with Acid Stress and Intraphagosomal Survival. Front Cell Infect Microbiol. 6:85. http://doi.org/10.3389/fcimb.2016.00085.

Park, K. T., M. M. ElNaggar, G. S. Abdellrazeq, J. P. Bannantine, V. Mack, L. M. Fry, and W. C. Davis. 2016. Phenotype and Function of Cd209+ Bovine Blood Dendritic Cells, Monocyte-Derived-Dendritic Cells and Monocyte-Derived Macrophages. PLoS One. 11(10):e0165247. http://doi.org/10.1371/journal.pone.0165247.

Rathnaiah, G., J. P. Bannantine, D. O. Bayles, D. K. Zinniel, J. R. Stabel, Y. T. Grohn, and R. G. Barletta. 2016. Analysis of Mycobacterium Avium Subsp. Paratuberculosis Mutant Libraries Reveals Loci-

Dependent Transposition Biases and Strategies for Novel Mutant Discovery. Microbiology. 162(4):633-41. http://doi.org/10.1099/mic.0.000258.

Bannantine, J. P., J. J. Campo, L. Li, A. Randall, J. Pablo, C. A. Praul, J. A. Raygoza Garay, J. R. Stabel, and V. Kapur. 2017. Identification of Novel Seroreactive Antigens in Johne's Disease Cattle by Using the Mycobacterium Tuberculosis Protein Array. Clin Vaccine Immunol. 24(7):e00081-17. http://doi.org/10.1128/CVI.00081-17.

Bannantine, J. P., G. Etienne, F. Laval, J. R. Stabel, A. Lemassu, M. Daffe, D. O. Bayles, C. Ganneau, F. Bonhomme, M. Branger, T. Cochard, S. Bay, and F. Biet. 2017. Cell Wall Peptidolipids of Mycobacterium Avium: From Genetic Prediction to Exact Structure of a Nonribosomal Peptide. Mol Microbiol. 105(4):525-39. http://doi.org/10.1111/mmi.13717.

Jenvey, C. J. and J. R. Stabel. Autofluorescence and Nonspecific Immunofluorescent Labeling in Frozen Bovine Intestinal Tissue Sections: Solutions for Multicolor Immunofluorescence Experiments. J Histochem Cytochem. 65(9):531-41. http://doi.org/10.1369/0022155417724425.

Albarrak, S., Hostetter, J., Waters, W.R., Stabel, J.R. 2017. WC1+ gamma delta T cells from cattle naturally infected with Mycobacterium avium subsp. paratuberculosis respond differentially to stimulation with PPD-J. Veterinary Immunology and Immunopathology. 190:57-64. https://doi.org/10.1016/j.vetimm.2017.07.007.

Li, L., J. P. Bannantine, J. J. Campo, A. Randall, Y. T. Grohn, R. Katani, M. Schilling, J. Radzio-Basu, and V. Kapur. 2017. Identification of Sero-Reactive Antigens for the Early Diagnosis of Johne's Disease in Cattle. PLoS One. 12(9):e0184373. http://doi.org/10.1371/journal.pone.0184373.

Li, L., B. Wagner, H. Freer, M. Schilling, J. P. Bannantine, J. J. Campo, R. Katani, Y. T. Grohn, J. Radzio-Basu, and V. Kapur. 2017. Early Detection of Mycobacterium Avium Subsp. Paratuberculosis Infection in Cattle with Multiplex-Bead Based Immunoassays. PLoS One 12(12):e0189783. http://doi.org/10.1371/journal.pone.0189783.

Magombedze, G., T. Shiri, S. Eda, and J. R. Stabel. 2017. Inferring Biomarkers for Mycobacterium Avium Subsp. Paratuberculosis Infection and Disease Progression in Cattle Using Experimental Data. Sci Rep. 7:44765. http://doi.org/10.1038/srep44765.

Park, K. T., M. M. Elnaggar, G. S. Abdellrazeq, J. P. Bannantine, V. Mack, L. M. Fry, and W. C. Davis. 2017. Correction: Phenotype and Function of CD209+ Bovine Blood Dendritic Cells, Monocyte-Derived-Dendritic Cells and Monocyte-Derived Macrophages. PLoS One. 12(1):e0171059. http://doi.org/10.1371/journal.pone.0165247

Rathnaiah, G., D. K. Zinniel, J. P. Bannantine, J. R. Stabel, Y. T. Grohn, M. T. Collins, and R. G. Barletta. 2017. Pathogenesis, Molecular Genetics, and Genomics of Mycobacterium Avium Subsp. Paratuberculosis, the Etiologic Agent of Johne's Disease. Front Vet Sci. 4:187. http://doi.org/10.3389/fvets.2017.00187.

Souza, C. D., J. P. Bannantine, W. C. Brown, M. G. Norton, W. C. Davis, J. K. Hwang, P. Ziaei, G. S. Abdellrazeq, M. V. Eren, J. R. Deringer, E. Laws, and M. C. D. Cardieri. 2017. A Nano Particle Vector Comprised of Poly Lactic-Co-Glycolic Acid and Monophosphoryl Lipid a and Recombinant Mycobacterium Avium Subsp Paratuberculosis Peptides Stimulate a Pro-Immune Profile in Bovine Macrophages. J Appl Microbiol. 123(1):54-65. http://doi.org/10.1111/jam.13491.

Abdellrazeq, G. S., M. M. Elnaggar, J. P. Bannantine, K. T. Park, C. D. Souza, B. Backer, V. Hulubei, L. M. Fry, S. A. Khaliel, H. A. Torky, D. A. Schneider, and W. C. Davis. 2018. A Mycobacterium Avium Subsp. Paratuberculosis Rela Deletion Mutant and a 35 Kda Major Membrane Protein Elicit Development of Cytotoxic T Lymphocytes with Ability to Kill Intracellular Bacteria. Vet Res. 49(1):53. http://doi.org/10.1186/s13567-018-0549-3.

Albarrak, S. M., W. R. Waters, J. R. Stabel, and J. M. Hostetter. 2018. Evaluating the Cytokine Profile of the Wc1(+) Gammadelta T Cell Subset in the Ileum of Cattle with the Subclinical and Clinical Forms of MAP Infection. Vet Immunol Immunopathol. 201:26-31. http://doi.org/10.1016/j.vetimm.2018.05.003.

Bannantine, J. P., J. R. Stabel, J. D. Lippolis, and T. A. Reinhardt. 2018. Membrane and Cytoplasmic Proteins of Mycobacterium Avium Subspecies Paratuberculosis That Bind to Novel Monoclonal Antibodies. Microorganisms. 6(4):127. http://doi.org/10.3390/microorganisms6040127.

Barkema, H. W., K. Orsel, S. S. Nielsen, A. P. Koets, Vpmg Rutten, J. P. Bannantine, G. P. Keefe, D. F. Kelton, S. J. Wells, R. J. Whittington, C. G. Mackintosh, E. J. Manning, M. F. Weber, C. Heuer, T. L. Forde, C. Ritter, S. Roche, C. S. Corbett, R. Wolf, P. J. Griebel, J. P. Kastelic, and J. De Buck. 2018. Knowledge Gaps That Hamper Prevention and Control of Mycobacterium Avium Subspecies Paratuberculosis Infection. Transbound Emerg Dis. 65(Suppl 1):125-48. http://doi.org/10.1111/tbed.12723.

Greenstein, R. J., L. Su, P. S. Fam, J. R. Stabel, and S. T. Brown. 2018. Failure to Detect M. Avium Subspecies Paratuberculosis in Johne's Disease Using a Proprietary Fluorescent in Situ Hybridization Assay. BMC Res Notes 11(1):498. http://doi.org/10.1186/s13104-018-3601-5.

Jenvey, C. J., J. M. Hostetter, A. L. Shircliff, and J. R. Stabel. 2018. Relationship between the Pathology of Bovine Intestinal Tissue and Current Diagnostic Tests for Johne's Disease. Vet Immunol Immunopathol. 202:93-101. http://doi.org/10.1016/j.vetimm.2018.06.012.

Samba-Louaka, A., E. Robino, T. Cochard, M. Branger, V. Delafont, W. Aucher, W. Wambeke, J. P. Bannantine, F. Biet, and Y. Hechard. 2018. Environmental Mycobacterium Avium Subsp. Paratuberculosis Hosted by Free-Living Amoebae. Front Cell Infect Microbiol. 8:28. http://doi.org/10.3389/fcimb.2018.00028.

Abdellrazeq, G. S., M. M. Elnaggar, J. P. Bannantine, D. A. Schneider, C. D. Souza, J. Hwang, A. H. A. Mahmoud, V. Hulubei, L. M. Fry, K. T. Park, and W. C. Davis. 2019. A Peptide-Based Vaccine for Mycobacterium Avium Subspecies Paratuberculosis. Vaccine. 37(21):2783-90. http://doi.org/10.1016/j.vaccine.2019.04.040.

Bannantine, J. P., A. Wadhwa, J. R. Stabel, and S. Eda. 2019. Characterization of Ethanol Extracted Cell Wall Components of Mycobacterium Avium Subsp. Paratuberculosis. Vet Sci. 6(4):88. http://doi.org/10.3390/vetsci6040088.

Bannantine, J. P., D. K. Zinniel, and R. G. Barletta. 2019. Transposon Mutagenesis in Mycobacterium Avium Subspecies Paratuberculosis. Methods Mol Biol. 2016:117-25. http://doi.org/10.1007/978-1-4939-9570-7_11.

Franceschi, V., A. H. Mahmoud, G. S. Abdellrazeq, G. Tebaldi, F. Macchi, L. Russo, L. M. Fry, M. M. Elnaggar, J. P. Bannantine, K. T. Park, V. Hulubei, S. Cavirani, W. C. Davis, and G. Donofrio. 2019. Capacity to Elicit Cytotoxic Cd8 T Cell Activity against Mycobacterium Avium Subsp. Paratuberculosis

Is Retained in a Vaccine Candidate 35 Kda Peptide Modified for Expression in Mammalian Cells. Front Immunol. 10:2859. http://doi.org/10.3389/fimmu.2019.02859.

Jenvey, C. J., J. M. Hostetter, A. L. Shircliff, J. P. Bannantine, and J. R. Stabel. 2019. Quantification of Macrophages and Mycobacterium Avium Subsp. Paratuberculosis in Bovine Intestinal Tissue During Different Stages of Johne's Disease. Vet Pathol 56(5):671-80. http://doi.org/10.1177/0300985819844823.

Jenvey, C. J., A. L. Shircliff, J. P. Bannantine, and J. R. Stabel. 2019. Phenotypes of Macrophages Present in the Intestine Are Impacted by Stage of Disease in Cattle Naturally Infected with Mycobacterium Avium Subsp. Paratuberculosis. PLoS One. 14(5):e0217649. http://doi.org/10.1371/journal.pone.0217649.

Li, L., J. P. Bannantine, J. J. Campo, A. Randall, Y. T. Grohn, M. A. Schilling, R. Katani, J. Radzio-Basu, L. Easterling, and V. Kapur. Identification of Sero-Diagnostic Antigens for the Early Diagnosis of Johne's Disease Using Map Protein Microarrays. Sci Rep. 9(1):17573. http://doi.org/10.1038/s41598-019-53973-x.

Stabel, J., L. Krueger, C. Jenvey, T. Wherry, J. Hostetter, and D. Beitz. 2019. Influence of Colostrum and Vitamins a, D3, and E on Early Intestinal Colonization of Neonatal Holstein Calves Infected with Mycobacterium Avium Subsp. Paratuberculosis. *Vet Sci* 6, no. 4 (Nov 20 2019). http://doi.org/10.3390/vetsci6040093.

Stabel, J. R. and J. P. Bannantine. 2019. Divergent Antigen-Specific Cellular Immune Responses During Asymptomatic Subclinical and Clinical States of Disease in Cows Naturally Infected with Mycobacterium Avium Subsp. Paratuberculosis. Infect Immun. 88(1):e00650-19. http://doi.org/10.1128/IAI.00650-19.

Stabel, J. R., T. A. Reinhardt, and R. J. Hempel. 2019. Short Communication: Vitamin D Status and Responses in Dairy Cows Naturally Infected with Mycobacterium Avium Ssp. Paratuberculosis. J Dairy Sci. 102(2):1594-600. http://doi.org/10.3168/jds.2018-15241.

Abdellrazeq, G. S., L. M. Fry, M. M. Elnaggar, J. P. Bannantine, D. A. Schneider, W. M. Chamberlin, A. H. A. Mahmoud, K. T. Park, V. Hulubei, and W. C. Davis. 2020. Simultaneous Cognate Epitope Recognition by Bovine Cd4 and Cd8 T Cells Is Essential for Primary Expansion of Antigen-Specific Cytotoxic T-Cells Following Ex Vivo Stimulation with a Candidate Mycobacterium Avium Subsp. Paratuberculosis Peptide Vaccine. Vaccine. (Jan 2 2020). http://doi.org/10.1016/j.vaccine.2019.12.052.

5030-32000-222-00D
Characterize the Immunopathogenesis and Develop Diagnostic and Mitigation Strategies to Control Tuberculosis in Cattle and Wildlife; Mitchell Palmer (P)

Maggioli, M. F., M. V. Palmer, T. C. Thacker, H. M. Vordermeier, and W. R. Waters. 2015. Characterization of Effector and Memory T Cell Subsets in the Immune Response to Bovine Tuberculosis in Cattle. PLoS One. 10(4): e0122571. http://doi.org/10.1371/journal.pone.0122571.

Maggioli, M. F., M. V. Palmer, H. M. Vordermeier, A. O. Whelan, J. M. Fosse, B. J. Nonnecke, and W. R. Waters. 2015. Application of Long-Term Cultured Interferon-Gamma Enzyme-Linked Immunospot Assay for Assessing Effector and Memory T Cell Responses in Cattle. J Vis Exp. 101:e52833. http://doi.org/10.3791/52833.

Palmer, M. V., T. C. Thacker, and W. R. Waters. 2015. Analysis of Cytokine Gene Expression Using a Novel Chromogenic in-Situ Hybridization Method in Pulmonary Granulomas of Cattle Infected Experimentally by Aerosolized Mycobacterium Bovis. J Comp Pathol. 153(2-3):150-9. http://doi.org/10.1016/j.jcpa.2015.06.004.

Ramdas, K. E., K. P. Lyashchenko, R. Greenwald, S. Robbe-Austerman, C. McManis, and W. R. Waters. 2015. Mycobacterium Bovis Infection in Humans and Cats in Same Household, Texas, USA, 2012. Emerg Infect Dis. 21(3):480-3. http://doi.org/10.3201/eid2103.140715.

Stahl, R. S., C. K. Ellis, P. Nol, W. R. Waters, M. Palmer, and K. C. VerCauteren. 2015. Fecal Volatile Organic Ccompound Profiles from White-Tailed Deer (Odocoileus Virginianus) as Indicators of Mycobacterium Bovis Exposure or Mycobacterium Bovis Bacille Calmette-Guerin (Bcg) Vaccination. PLoS One. 10(6):e0129740. http://doi.org/10.1371/journal.pone.0129740.

Thacker, T. C., M. V. Palmer, S. Robbe-Austerman, T. P. Stuber, and W. R. Waters. 2015. Anatomical Distribution of Mycobacterium Bovis Genotypes in Experimentally Infected White-Tailed Deer. Vet Microbiol. 180(1-2): 75-81. http://doi.org/10.1016/j.vetmic.2015.07.006.

Tuggle, C. K. and W. R. Waters. 2015. Tuberculosis-Resistant Transgenic Cattle. Proc Natl Acad Sci U S A. 112(13):3854-5. http://doi.org/10.1073/pnas.1502972112.

Waters, W. R. and M. V. Palmer. 2015. Mycobacterium Bovis Infection of Cattle and White-Tailed Deer: Translational Research of Relevance to Human Tuberculosis. ILAR J. 56(1):26-43. http://doi.org/10.1093/ilar/ilv001.

Waters, W. R., M. V. Palmer, M. R. Stafne, K. E. Bass, M. F. Maggioli, T. C. Thacker, R. Linscott, J. C. Lawrence, J. T. Nelson, J. Esfandiari, R. Greenwald, and K. P. Lyashchenko. 2015. Effects of Serial Skin Testing with Purified Protein Derivative on the Level and Quality of Antibodies to Complex and Defined Antigens in Mycobacterium Bovis-Infected Cattle. Clin Vaccine Immunol. 22(6):641-9. http://doi.org/10.1128/CVI.00119-15.

Alexander, K. A., C. E. Sanderson, M. H. Larsen, S. Robbe-Austerman, M. C. Williams, and M. V. Palmer. 2016. Erratum for Alexander Et Al., Emerging Tuberculosis Pathogen Hijacks Social Communication Behavior in the Group-Living Banded Mongoose (Mungos Mungo). mBio. 7(3):e00921-16. http://doi.org/10.1128/mBio.00921-16.

Alexander, K. A., C. E. Sanderson, M. H. Larsen, S. Robbe-Austerman, M. C. Williams, and M. V. Palmer. 2016. Emerging Tuberculosis Pathogen Hijacks Social Communication Behavior in the Group-Living Banded Mongoose (Mungos Mungo). mBio. 7(3)e00281-16. http://doi.org/10.1128/mBio.00281-16.

Bigi, M. M., F. C. Blanco, F. R. Araujo, T. C. Thacker, M. J. Zumarraga, A. A. Cataldi, M. A. Soria, and F. Bigi. 2016. Polymorphisms of 20 Regulatory Proteins between Mycobacterium Tuberculosis and Mycobacterium Bovis. Microbiol Immunol. 60(8):552-60. http://doi.org/10.1111/1348-0421.12402.

Maggioli, M. F., M. V. Palmer, T. C. Thacker, H. M. Vordermeier, J. L. McGill, A. O. Whelan, M. H. Larsen, W. R. Jacobs, Jr., and W. R. Waters. 2016. Increased Tnf-Alpha/Ifn-Gamma/Il-2 and Decreased Tnf-Alpha/Ifn-Gamma Production by Central Memory T Cells Are Associated with Protective Responses against Bovine Tuberculosis Following Bcg Vaccination. Front Immunol. 7:421. http://doi.org/10.3389/fimmu.2016.00421.

Palmer, M. V., T. C. Thacker, and W. R. Waters. 2016. Differential Cytokine Gene Expression in Granulomas from Lungs and Lymph Nodes of Cattle Experimentally Infected with Aerosolized Mycobacterium Bovis. PLoS One. 11(11):e0167471. http://doi.org/10.1371/journal.pone.0167471.

Palmer, M. V., T. C. Thacker, and W. R. Waters. 2016. Multinucleated Giant Cell Cytokine Expression in Pulmonary Granulomas of Cattle Experimentally Infected with Mycobacterium Bovis. Vet Immunol Immunopathol. 180:34-39. http://doi.org/10.1016/j.vetimm.2016.08.015.

Waters, W. R., M. F. Maggioli, M. V. Palmer, T. C. Thacker, J. L. McGill, H. M. Vordermeier, L. Berney-Meyer, W. R. Jacobs, Jr., and M. H. Larsen. 2016. Interleukin-17a as a Biomarker for Bovine Tuberculosis. Clin Vaccine Immunol. 23(2):168-80. http://doi.org/10.1128/CVI.00637-15.

Ellis, C. K., S. Rice, D. Maurer, R. Stahl, W. R. Waters, M. V. Palmer, P. Nol, J. C. Rhyan, K. C. VerCauteren, and J. A. Koziel. 2017. Use of Fecal Volatile Organic Compound Analysis to Discriminate between Non-Vaccinated and Bcg-Vaccinated Cattle Prior to and after Mycobacterium Bovis Challenge. PLoS One. 12(7):e0179914. http://doi.org/10.1371/journal.pone.0179914.

Lyashchenko, K. P., A. Grandison, K. Keskinen, A. Sikar-Gang, P. Lambotte, J. Esfandiari, G. C. Ireton, A. Vallur, S. G. Reed, G. Jones, H. M. Vordermeier, J. R. Stabel, T. C. Thacker, M. V. Palmer, and W. R. Waters. 2017. Identification of Novel Antigens Recognized by Serum Antibodies in Bovine Tuberculosis. Clin Vaccine Immunol. 24(12):e00259-17. http://doi.org/10.1128/CVI.00259-17.

Petruzzi, B., Briggs, R.E., Tatum, F.M., Swords, W.E., De Castro, C., Molinaro, A., Inzana, T.J. 2017. Capsular polysaccharide interferes with biofilm formation by Pasteurella multocida Serogroup A. mBio. 8(6):e01843-17. https://doi.org/10.1128/mBio.01843-17.

Klima, C.L., Zaheer, R., Briggs, R.E., McAllister, T.A. 2017. A multiplex PCR assay for molecular capsular serotyping of Mannheimia haemolytica serotypes 1, 2, and 6. Journal of Microbiological Methods. 139:155-160. doi. 10.1016/j.mimet.2017.05.010. https://doi.org/10.1016/j.mimet.2017.05.010.

Lyashchenko, K. P., R. Greenwald, A. Sikar-Gang, A. A. Sridhara, A. Johnathan, P. Lambotte, J. Esfandiari, M. F. Maggioli, T. C. Thacker, M. V. Palmer, and W. R. Waters. 2017. Early Detection of Circulating Antigen and IgM-Associated Immune Complexes During Experimental Mycobacterium Bovis Infection in Cattle. Clin Vaccine Immunol. 24(6):e00069-17. http://doi.org/10.1128/CVI.00069-17.

Palmer, M. V., R. J. Cox, W. R. Waters, T. C. Thacker, and D. L. Whipple. 2017. Using White-Tailed Deer (Odocoileus Virginianus) in Infectious Disease Research. J Am Assoc Lab Anim Sci. 56(4):350-60. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5517323/

Patane, J. S., J. Martins, A. Beatriz Castelao, C. Nishibe, L. Montera, F. Bigi, M. J. Zumarraga, A. A. Cataldi, A. Fonseca Junior, E. Roxo, A. Luiza, A. R. Osorio, K. S. Jorge Ufms, T. C. Thacker, N. F. Almeida, F. R. Araujo, and J. C. Setubal. 2017. Patterns and Processes of Mycobacterium Bovis Evolution Revealed by Phylogenomic Analyses. Genome Biol Evol. 9(3):521-535. http://doi.org/10.1093/gbe/evx022.

Rusk, R. A., M. V. Palmer, W. R. Waters, and J. L. McGill. 2017. Measuring Bovine Gammadelta T Cell Function at the Site of Mycobacterium Bovis Infection. Vet Immunol Immunopathol. 193-194:38-49. http://doi.org/10.1016/j.vetimm.2017.10.004.

Vu, D. M., R. M. Sakamuri, W. R. Waters, B. I. Swanson, and H. Mukundan. 2017. Detection of Lipomannan in Cattle Infected with Bovine Tuberculosis. Anal Sci. 33(4):457-60. http://doi.org/10.2116/analsci.33.457.

Wanzala, S. I., M. V. Palmer, W. R. Waters, T. C. Thacker, M. Carstensen, D. A. Travis, and S. Sreevatsan. 2017. Evaluation of Pathogen-Specific Biomarkers for the Diagnosis of Tuberculosis in White-Tailed Deer (Odocoileus Virginianus). Am J Vet Res. 78(6):729-34. http://doi.org/10.2460/ajvr.78.6.729.

Waters, W. R., H. M. Vordermeier, S. Rhodes, B. Khatri, M. V. Palmer, M. F. Maggioli, T. C. Thacker, J. T. Nelson, B. V. Thomsen, S. Robbe-Austerman, D. M. Bravo Garcia, M. A. Schoenbaum, M. S. Camacho, J. S. Ray, J. Esfandiari, P. Lambotte, R. Greenwald, A. Grandison, A. Sikar-Gang, and K. P. Lyashchenko. 2017. Potential for Rapid Antibody Detection to Identify Tuberculous Cattle with Non-Reactive Tuberculin Skin Test Results. BMC Vet Res. 13(1):164. http://doi.org/10.1186/s12917-017-1085-5.

Alexander, K. A., P. N. Laver, M. C. Williams, C. E. Sanderson, C. Kanipe, and M. V. Palmer. 2018. Pathology of the Emerging Mycobacterium Tuberculosis Complex Pathogen, Mycobacterium Mungi, in the Banded Mongoose (Mungos Mungo). Vet Pathol. 55(2):303-09. http://doi.org/10.1177/0300985817741730.

Hadi, S. A., W. R. Waters, M. Palmer, K. P. Lyashchenko, and S. Sreevatsan. 2018. Development of a Multidimensional Proteomic Approach to Detect Circulating Immune Complexes in Cattle Experimentally Infected with Mycobacterium Bovis. Front Vet Sci. 5:141. http://doi.org/10.3389/fvets.2018.00141.

Lyashchenko, K. P., C. Gortazar, M. A. Miller, and W. R. Waters. 2018. Spectrum of Antibody Profiles in Tuberculous Elephants, Cervids, and Cattle. Vet Microbiol. 214:89-92. http://doi.org/10.1016/j.vetmic.2017.12.013.

Palmer, M. V. 2018. Emerging Understanding of Tuberculosis and the Granuloma by Comparative Analysis in Humans, Cattle, Zebrafish, and Nonhuman Primates. Vet Pathol. 55(1):8-10. http://doi.org/10.1177/0300985817712795.

Palmer, M. V. and T. C. Thacker. 2018. Use of the Human Vaccine, Mycobacterium Bovis Bacillus Calmette Guerin in Deer. *Front Vet Sci* 5 (2018): 244. http://doi.org/10.3389/fvets.2018.00244.

Cox, R. J., P. Nol, C. K. Ellis, and M. V. Palmer. 2019. Research with Agricultural Animals and Wildlife. ILAR J. ilz006. http://doi.org/10.1093/ilar/ilz006.

Guerra-Maupome, M., M. V. Palmer, W. R. Waters, and J. L. McGill. 2019. Characterization of Gammadelta T Cell Effector/Memory Subsets Based on Cd27 and Cd45r Expression in Response to Mycobacterium Bovis Infection. Immunohorizons. 3(6): 208-18. http://doi.org/10.4049/immunohorizons.1900032.

Maurer, D. L., C. K. Ellis, T. C. Thacker, S. Rice, J. A. Koziel, P. Nol, and K. C. VerCauteren. 2019. Screening of Microbial Volatile Organic Compounds for Detection of Disease in Cattle: Development of Lab-Scale Method. Sci Rep. 9(1):12103. http://doi.org/10.1038/s41598-019-47907-w.

Miller, M. A., P. Buss, E. O. Roos, G. Hausler, A. Dippenaar, E. Mitchell, L. van Schalkwyk, S. Robbe-Austerman, W. R. Waters, A. Sikar-Gang, K. P. Lyashchenko, S. D. C. Parsons, R. Warren, and P. van

Helden. 2019. Fatal Tuberculosis in a Free-Ranging African Elephant and One Health Implications of Human Pathogens in Wildlife. Front Vet Sci. 6:18. http://doi.org/10.3389/fvets.2019.00018.

Palmer, M. V., C. Kanipe, R. Cox, S. Robbe-Austerman, and T. C. Thacker. 2019. Characteristics of Subclinical Mycobacterium Avium Ssp. Paratuberculosis Infection in a Captive White-Tailed Deer Herd. J Vet Diagn Invest. 31(6):844-51. http://doi.org/10.1177/1040638719873028.

Palmer, M. V., T. C. Thacker, M. M. Rabideau, G. J. Jones, C. Kanipe, H. M. Vordermeier, and W. Ray Waters. 2019. Biomarkers of Cell-Mediated Immunity to Bovine Tuberculosis. Vet Immunol Immunopathol. 220:109988. http://doi.org/10.1016/j.vetimm.2019.109988.

Palmer, M. V., J. Wiarda, C. Kanipe, and T. C. Thacker. 2019. Early Pulmonary Lesions in Cattle Infected Via Aerosolized Mycobacterium Bovis. Vet Pathol. 56(4):544-54. http://doi.org/10.1177/0300985819833454.

Wanzala, S. I., J. Nakavuma, D. Travis, P. Kia, S. Ogwang, W. R. Waters, T. Thacker, T. Johnson, S. A. Hadi, and S. Sreevatsan. 2019. Retrospective Analysis of Archived Pyrazinamide Resistant Mycobacterium Tuberculosis Complex Isolates from Uganda-Evidence of Interspecies Transmission. Microorganisms. 7(8):221. http://doi.org/10.3390/microorganisms7080221.

Krajewska-Wedzina, M., A. Didkowska, A. A. Sridhara, R. Elahi, A. Johnathan-Lee, L. Radulski, M. Lipiec, K. Anusz, K. P. Lyashchenko, M. A. Miller, and W. R. Waters. 2020. Transboundary Tuberculosis: Importation of Alpacas Infected with Mycobacterium Bovis from the United Kingdom to Poland and Potential for Serodiagnostic Assays in Detecting Tuberculin Skin Test False-Negative Animals. Transbound Emerg Dis. http://doi.org/10.1111/tbed.13471.

5030-32000-223-00D
Characterization of the Pathogenesis and Antigen Expression in Spirochete Diseases; David Alt (P), Jarlath Nally, Darrell Bayles and Jennifer Wilson-Welder

Almeida, A. M., A. Bassols, E. Bendixen, M. Bhide, F. Ceciliani, S. Cristobal, P. D. Eckersall, K. Hollung, F. Lisacek, G. Mazzucchelli, M. McLaughlin, I. Miller, J. E. Nally, J. Plowman, J. Renaut, P. Rodrigues, P. Roncada, J. Staric, and R. Turk. 2015. Animal Board Invited Review: Advances in Proteomics for Animal and Food Sciences. Animal. 9(1):1-17. http://doi.org/10.1017/S1751731114002602.

Alt, D. P., J. H. Wilson-Welder, D. O. Bayles, C. Cameron, B. Adler, D. M. Bulach, T. Seemann, M. J. Lehane, L. R. Haines, A. C. Darby, N. Hall, A. D. Radford, and R. L. Zuerner. 2015. Complete Genome Sequence of Leptospira Interrogans Serovar Bratislava, Strain Pigk151. Genome Announc. 3(3):e00678-15. http://doi.org/10.1128/genomeA.00678-15.

Goeijenbier, M., M. H. Gasem, J. C. Meijers, R. A. Hartskeerl, A. Ahmed, M. G. Goris, B. Isbandrio, S. S. Schuller, A. D. Osterhaus, B. E. Martina, E. C. van Gorp, J. E. Nally, and J. F. Wagenaar. 2015. Markers of Endothelial Cell Activation and Immune Activation Are Increased in Patients with Severe Leptospirosis and Associated with Disease Severity. J Infect. 71(4):437-46. http://doi.org/10.1016/j.jinf.2015.05.016.

Grassmann, A. A., A. J. McBride, J. E. Nally, and M. J. Caimano. 2015. Generation of Mammalian Host-Adapted Leptospira Interrogans by Cultivation in Peritoneal Dialysis Membrane Chamber Implantation in Rats. Bio Protoc. 5(14)e1536. http://doi.org/10.21769/bioprotoc.1536.

Looft, T., D. O. Bayles, D. P. Alt, and T. B. Stanton. 2015. Complete Genome Sequence of Coriobacteriaceae Strain 68-1-3, a Novel Mucus-Degrading Isolate from the Swine Intestinal Tract. Genome Announc. 3(5)e01143-15. http://doi.org/10.1128/genomeA.01143-15.

Nally, J. E., W. Mullen, J. J. Callanan, H. Mischak, and A. Albalat. 2015. Detection of Urinary Biomarkers in Reservoir Hosts of Leptospirosis by Capillary Electrophoresis-Mass Spectrometry. Proteomics Clin Appl. 9(5-6):543-51. http://doi.org/10.1002/prca.201400205.

Piras, C., A. Soggiu, V. Greco, P. A. Martino, F. Del Chierico, L. Putignani, A. Urbani, J. E. Nally, L. Bonizzi, and P. Roncada. 2015. Mechanisms of Antibiotic Resistance to Enrofloxacin in Uropathogenic Escherichia Coli in Dog. J Proteomics. 127(Pt B):365-76. http://doi.org/10.1016/j.jprot.2015.05.040.

Prager, K. C., D. P. Alt, M. G. Buhnerkempe, D. J. Greig, R. L. Galloway, Q. Wu, F. M. D. Gulland, and J. O. Lloyd-Smith. 2015. Antibiotic Efficacy in Eliminating Leptospiruria in California Sea Lions (Zalophus Californianus) Stranding with Leptospirosis. Aquat Mamm. 41(2):203-12. http://doi.org/10.1578/AM.41.2.2015.203.

Schuller, S., Z. J. Arent, C. Gilmore, and J. Nally. 2015. Prevalence of Antileptospiral Serum Antibodies in Dogs in Ireland. Vet Rec. 177(5):126. http://doi.org/10.1136/vr.102916.

Schuller, S., J. J. Callanan, S. Worrall, T. Francey, A. Schweighauser, B. Kohn, R. Klopfleisch, H. Posthaus, and J. E. Nally. 2015. Immunohistochemical Detection of IgM and IgG in Lung Tissue of Dogs with Leptospiral Pulmonary Haemorrhage Syndrome (Lphs). Comp Immunol Microbiol Infect Dis. 40:47-53. http://doi.org/10.1016/j.cimid.2015.04.002.

Schuller, S., T. Francey, K. Hartmann, M. Hugonnard, B. Kohn, J. E. Nally, and J. Sykes. 2015. European Consensus Statement on Leptospirosis in Dogs and Cats. J Small Anim Pract. 56(3):159-79. http://doi.org/10.1111/jsap.12328.

Schuller, S., K. Sergeant, J. Renaut, J. J. Callanan, C. Scaife, and J. E. Nally. 2015. A 2-D Guinea Pig Lung Proteome Map. Data Brief. 4:140-5. http://doi.org/10.1016/j.dib.2015.04.009.

Schuller, S., K. Sergeant, J. Renaut, J. J. Callanan, C. Scaife, and J. E. Nally. 2015. Comparative Proteomic Analysis of Lung Tissue from Guinea Pigs with Leptospiral Pulmonary Haemorrhage Syndrome (Lphs) Reveals a Decrease in Abundance of Host Proteins Involved in Cytoskeletal and Cellular Organization. J Proteomics. 122:55-72. http://doi.org/10.1016/j.jprot.2015.03.021.

Wilson-Welder, J. H., D. P. Alt, and J. E. Nally. 2015. Digital Dermatitis in Cattle: Current Bacterial and Immunological Findings. Animals (Basel). 5(4):1114-35. https://doi.org/10.3390/ani5040400

Wilson-Welder, J. H., D. P. Alt, and J. E. Nally. 2015. The Etiology of Digital Dermatitis in Ruminants: Recent Perspectives. Vet Med (Auckl). 6:155-64. http://doi.org/10.2147/VMRR.S62072.

Allen, H. K., D. O. Bayles, T. Looft, J. Trachsel, B. E. Bass, D. P. Alt, S. M. Bearson, T. Nicholson, and T. A. Casey. 2016. Pipeline for Amplifying and Analyzing Amplicons of the V1-V3 Region of the 16s rRNA Gene. BMC Res Notes. 9:380. https://doi.org/10.1186/s13104-016-2172-6

Benkirane, A., S. Noury, R. A. Hartskeerl, M. G. Goris, A. Ahmed, and J. E. Nally. 2016. Preliminary Investigations on the Distribution of Leptospira Serovars in Domestic Animals in North-West Morocco. Transbound Emerg Dis. 63(2):e178-84. http://doi.org/10.1111/tbed.12252.

Nally, J. E., Z. Arent, D. O. Bayles, R. L. Hornsby, C. Gilmore, S. Regan, A. D. McDevitt, J. Yearsley, S. Fanning, and B. J. McMahon. 2016. Emerging Infectious Disease Implications of Invasive Mammalian Species: The Greater White-Toothed Shrew (Crocidura Russula) Is Associated with a Novel Serovar of Pathogenic Leptospira in Ireland. PLoS Negl Trop Dis. 10(12):e0005174. http://doi.org/10.1371/journal.pntd.0005174.

Nally, J. E., D. O. Bayles, D. Hurley, S. Fanning, B. J. McMahon, and Z. Arent. 2016. Complete Genome Sequence of Leptospira Alstonii Serovar Room22 Strain Gwts #1. Genome Announc. 4(6):e1230-16. http://doi.org/10.1128/genomeA.01230-16.

Sharma, V. K., D. O. Bayles, D. P. Alt, and T. Looft. 2016. Complete Genome Sequences of Curli-Negative and Curli-Positive Isolates of Foodborne Escherichia Coli O157:H7 Strain 86-24. Genome Announc. 4(6):e01323-16. http://doi.org/10.1128/genomeA.01323-16.

Trachsel, J., D. O. Bayles, T. Looft, U. Y. Levine, and H. K. Allen. 2016. Function and Phylogeny of Bacterial Butyryl Coenzyme A:Acetate Transferases and Their Diversity in the Proximal Colon of Swine. Appl Environ Microbiol. 82(22): 6788-98. http://doi.org/10.1128/AEM.02307-16.

Wilson-Welder, J. H., A. T. Frank, R. L. Hornsby, S. C. Olsen, and D. P. Alt. 2016. Interaction of Bovine Peripheral Blood Polymorphonuclear Cells and Leptospira Species; Innate Responses in the Natural Bovine Reservoir Host. Front Microbiol. 7:1110. http://doi.org/10.3389/fmicb.2016.01110.

Bearson, B. L., S. M. D. Bearson, B. W. Brunelle, D. O. Bayles, I. S. Lee, and J. D. Kich. 2017. Salmonella Diva Vaccine Reduces Disease, Colonization and Shedding Due to Virulent S. Typhimurium Infection in Swine. J Med Microbiol. 66(5): 651-61. http://doi.org/10.1099/jmm.0.000482.

Furuya, Y., G. S. Kirimanjeswara, S. Roberts, R. Racine, J. Wilson-Welder, A. M. Sanfilippo, S. L. Salmon, and D. W. Metzger. 2017. Defective Anti-Polysaccharide Igg Vaccine Responses in Iga Deficient Mice. Vaccine. 35(37):4997-5005. http://doi.org/10.1016/j.vaccine.2017.07.071.

Johnson, T. A., T. Looft, A. J. Severin, D. O. Bayles, D. J. Nasko, K. E. Wommack, A. Howe, and H. K. Allen. 2017. The in-Feed Antibiotic Carbadox Induces Phage Gene Transcription in the Swine Gut Microbiome. mBio 8(4):e00709-17. http://doi.org/10.1128/mBio.00709-17.

Nally, J. E., A. A. Grassmann, S. Planchon, K. Sergeant, J. Renaut, J. Seshu, A. J. McBride, and M. J. Caimano. 2017. Pathogenic Leptospires Modulate Protein Expression and Post-Translational Modifications in Response to Mammalian Host Signals. Front Cell Infect Microbiol. 7:362. http://doi.org/10.3389/fcimb.2017.00362.

Scanlan, E., L. Ardill, M. V. Whelan, C. Shortt, J. E. Nally, B. Bourke, and O. Croinin T. 2017. Relaxation of DNA Supercoiling Leads to Increased Invasion of Epithelial Cells and Protein Secretion by Campylobacter Jejuni. Mol Microbiol. 104(1):92-104. http://doi.org/10.1111/mmi.13614.

Sharma, V. K., D. O. Bayles, D. P. Alt, T. Looft, B. W. Brunelle, and J. A. Stasko. 2017. Disruption of Rcsb by a Duplicated Sequence in a Curli-Producing Escherichia Coli O157:H7 Results in Differential Gene Expression in Relation to Biofilm Formation, Stress Responses and Metabolism. BMC Microbiol. 17(1):56. http://doi.org/10.1186/s12866-017-0966-x.

Bakker, M. G., T. Looft, D. P. Alt, K. Delate, and C. A. Cambardella. 2018. Bulk Soil Bacterial Community Structure and Function Respond to Long-Term Organic and Conventional Agricultural Management. Can J Microbiol. 64(12):901-14. http://doi.org/10.1139/cjm-2018-0134.

Ferrer, M. F., E. Scharrig, N. Charo, A. L. Ripodas, R. Drut, E. A. Carrera Silva, A. Nagel, J. E. Nally, D. P. Montes de Oca, M. Schattner, and R. M. Gomez. 2018. Macrophages and Galectin 3 Control Bacterial Burden in Acute and Subacute Murine Leptospirosis That Determines Chronic Kidney Fibrosis. Front Cell Infect Microbiol. 8:384. http://doi.org/10.3389/fcimb.2018.00384.

Nally, J. E., R. L. Hornsby, D. P. Alt, D. Bayles, J. H. Wilson-Welder, D. E. Palmquist, and N. E. Bauer. 2018. Isolation and Characterization of Pathogenic Leptospires Associated with Cattle. Vet Microbiol. 218:25-30. http://doi.org/10.1016/j.vetmic.2018.03.023.

Nally, J. E. and S. Schuller. 2018. Proteomic Analysis of Lung Tissue by DIGE. Methods Mol Biol. 1664:167-83. http://doi.org/10.1007/978-1-4939-7268-5_14.

Nally, J. E., J. H. Wilson-Welder, R. L. Hornsby, M. V. Palmer, and D. P. Alt. 2018. Inbred Rats as a Model to Study Persistent Renal Leptospirosis and Associated Cellular Immune Responsiveness. Front Cell Infect Microbiol. 8:66. http://doi.org/10.3389/fcimb.2018.00066.

Wilson-Welder, J. H., J. E. Nally, D. P. Alt, S. B. Humphrey, and S. C. Olsen. 2018. Short Communication: Lymphocyte Proliferative Responses in Cattle Naturally Infected with Digital Dermatitis Consist of Cd8+ and Gammadelta-T Cells but Lack Cd4+ T Cells. J Dairy Sci. 101(9):8301-07. http://doi.org/10.3168/jds.2017-13913.

Wilson-Welder, J. H., J. E. Nally, D. P. Alt, M. V. Palmer, J. Coatney, and P. Plummer. 2018. Experimental Transmission of Bovine Digital Dermatitis to Sheep: Development of an Infection Model. Vet Pathol. 55(2):245-57. http://doi.org/10.1177/0300985817736572.

Han, S., K. G. Mansfield, D. S. Bradway, T. E. Besser, D. H. Read, G. J. Haldorson, D. P. Alt, and J. H. Wilson-Welder. 2019. Treponeme-Associated Hoof Disease of Free-Ranging Elk (Cervus Elaphus) in Southwestern Washington State, USA. Vet Pathol. 56(1):118-32. http://doi.org/10.1177/0300985818798108.

Hernandez-Castellano, L. E., J. E. Nally, J. Lindahl, M. Wanapat, I. A. Alhidary, D. Fangueiro, D. Grace, M. Ratto, J. C. Bambou, and A. M. de Almeida. 2019. Dairy Science and Health in the Tropics: Challenges and Opportunities for the Next Decades. Trop Anim Health Prod. 51(5):1009-17. http://doi.org/10.1007/s11250-019-01866-6.

Nally, J. E., R. L. Hornsby, D. P. Alt, and J. P. Whitelegge. 2019. Phenotypic and Proteomic Characterization of Treponemes Associated with Bovine Digital Dermatitis. Vet Microbiol. 235:35-42. http://doi.org/10.1016/j.vetmic.2019.05.023.

O'Neill, L. M., O. M. Keane, P. J. Ross, J. E. Nally, J. Seshu, and B. Markey. 2019. Evaluation of Protective and Immune Responses Following Vaccination with Recombinant Mip and Cpaf from Chlamydia Abortus as Novel Vaccines for Enzootic Abortion of Ewes. Vaccine. 37(36):5428-38. http://doi.org/10.1016/j.vaccine.2019.06.088.

Sharma, V. K., S. Akavaram, R. G. Schaut, and D. O. Bayles. 2019. Comparative Genomics Reveals Structural and Functional Features Specific to the Genome of a Foodborne Escherichia Coli O157:H7. BMC Genomics. 20(1):196. http://doi.org/10.1186/s12864-019-5568-6.

Wafa, E. I., J. H. Wilson-Welder, R. L. Hornsby, J. E. Nally, S. M. Geary, N. B. Bowden, and A. K. Salem. 2020. Poly (Diaminosulfide) Microparticle-Based Vaccine for Delivery of Leptospiral Antigens. Biomacromolecules. (Jan 2 2020). http://doi.org/10.1021/acs.biomac.9b01257.

5030-32000-224-00D
Pathogenesis and Development of Improved Diagnostic and Control Strategies for Brucellosis in Livestock and Wildlife; Steven Olsen (P) and Paola Boggiatto

Olsen, S. C. and S. A. Carlson. 2015. In Vitro Bactericidal Activity of Aminoglycosides, Including the Next-Generation Drug Plazomicin, against Brucella Spp. Int J Antimicrob Agents. 45(1):76-8. http://doi.org/10.1016/j.ijantimicag.2014.09.011.

Olsen, S. C., J. L. McGill, R. E. Sacco, and S. G. Hennager. 2015. Immune Responses of Bison and Efficacy after Booster Vaccination with Brucella Abortus Strain Rb51. Clin Vaccine Immunol. 22(4):440-7. http://doi.org/10.1128/CVI.00746-14.

Falconer, J. L., R. J. Christie, E. J. Pollard, S. C. Olsen, and D. W. Grainger. 2016. Live Rb51 Vaccine Lyophilized Hydrogel Formulations with Increased Shelf Life for Practical Ballistic Delivery. Int J Pharm. 498(1-2):187-94. http://doi.org/10.1016/j.ijpharm.2015.12.040.

Nol, P., S. C. Olsen, J. C. Rhyan, N. Sriranganathan, M. P. McCollum, S. G. Hennager, A. A. Pavuk, P. J. Sprino, S. M. Boyle, R. J. Berrier, and M. D. Salman. 2016. Vaccination of Elk (Cervus Canadensis) with Brucella Abortus Strain Rb51 Overexpressing Superoxide Dismutase and Glycosyltransferase Genes Does Not Induce Adequate Protection against Experimental Brucella Abortus Challenge. Front Cell Infect Microbiol. 6:10. http://doi.org/10.3389/fcimb.2016.00010.

Olsen, S. C. and F. M. Tatum. 2017. Swine Brucellosis: Current Perspectives. Vet Med (Auckl). 8:1-12. http://doi.org/10.2147/VMRR.S91360.

Boggiatto, P. M., D. Fitzsimmons, D. O. Bayles, D. Alt, C. E. Vrentas, and S. C. Olsen. 2018. Coincidence Cloning Recovery of Brucella Melitensis RNA from Goat Tissues: Advancing the in Vivo Analysis of Pathogen Gene Expression in Brucellosis. BMC Mol Biol. 19(1):10. http:/doi.org/10.1186/s12867-018-0111-x.

Vrentas, C. E., P. M. Boggiatto, R. G. Schaut, and S. C. Olsen. 2018. Collection and Processing of Lymph Nodes from Large Animals for RNA Analysis: Preparing for Lymph Node Transcriptomic Studies of Large Animal Species. J Vis Exp. 135:e57195. http://doi.org/10.3791/57195.

Vrentas, C. E., R. G. Schaut, P. M. Boggiatto, S. C. Olsen, F. S. Sutterwala, and M. Moayeri. 2018. Inflammasomes in Livestock and Wildlife: Insights into the Intersection of Pathogens and Natural Host Species. Vet Immunol Immunopathol. 201:49-56. http://doi.org/10.1016/j.vetimm.2018.05.008.

Boggiatto, P. M. and S. C. Olsen. 2019. Tulathromycin Treatment Does Not Affect Bacterial Dissemination or Clearance of Brucella Melitensis 16m Following Experimental Infection of Goats. PLoS One 14(12):e0226242. http://doi.org/10.1371/journal.pone.0226242.

Boggiatto, P. M., R. G. Schaut, C. Kanipe, S. M. Kelly, B. Narasimhan, D. E. Jones, and S. C. Olsen. 2019. Sustained Antigen Release Polyanhydride-Based Vaccine Platform for Immunization against Bovine Brucellosis. Heliyon. 5(8):e02370. http://doi.org/10.1016/j.heliyon.2019.e02370.

Schaut, R. G., P. M. Boggiatto, C. L. Loving, and V. K. Sharma. 2019. Cellular and Mucosal Immune Responses Following Vaccination with Inactivated Mutant of Escherichia Coli O157:H7. Sci Rep. 9(1):6401. http://doi.org/10.1038/s41598-019-42861-z.

Vrentas, C. E., P. M. Boggiatto, S. C. Olsen, S. H. Leppla, and M. Moayeri. 2019. Characterization of the Nlrp1 Inflammasome Response in Bovine Species. Innate Immun. (Nov 11 2019): 1753425919886649. http://doi.org/10.1177/1753425919886649.

Vrentas, C. E. 2019. Teaching Trek: Reflections on Participation in the Indo-U.S. Teaching Professorship Program. J Microbiol Biol Educ. 20(1). http://doi.org/10.1128/jmbe.v20i1.1553.

Woolums, A. R., B. B. Karisch, J. A. Parish, J. Park, K. S. Seo, P. Badial, and S. C. Olsen. 2019. Effect of a DNA-Based Immunostimulant on Growth, Performance, and Expression of Inflammatory and Immune Mediators in Beef Calves Abruptly Weaned and Introduced to a Complete Ration. J Anim Sci. 97(1):111-21. http://doi.org/10.1093/jas/sky392.

Lindahl, J. F., C. E. Vrentas, R. P. Deka, R. A. Hazarika, H. Rahman, R. G. Bambal, J. S. Bedi, C. Bhattacharya, P. Chaduhuri, N. M. Fairoze, R. S. Gandhi, J. P. S. Gill, N. K. Gupta, M. Kumar, S. Londhe, M. Rahi, P. K. Sharma, R. Shome, R. Singh, K. Srinivas, and B. B. Swain. 2020. Brucellosis in India: Results of a Collaborative Workshop to Define One Health Priorities. Trop Anim Health Prod. 52(1): 387-396. http://doi.org/10.1007/s11250-019-02029-3.

National Animal Disease Center

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5030-32000-115-00D

Non-antibiotic Approaches to Control Mastitis; John Lippolis (P) and Timothy Reinhardt

Casas, E., J. D. Lippolis, L. A. Kuehn, and T. A. Reinhardt. 2015. Seasonal Variation in Vitamin D Status of Beef Cattle Reared in the Central United States. Domest Anim Endocrinol. 52:71-4. http://doi.org/10.1016/j.domaniend.2015.03.003.

Merriman, K. E., M. F. Kweh, J. L. Powell, J. D. Lippolis, and C. D. Nelson. 2015. Multiple Beta-Defensin Genes Are Upregulated by the Vitamin D Pathway in Cattle. J Steroid Biochem Mol Biol 154:120-9. http://doi.org/10.1016/j.jsbmb.2015.08.002.

Monteiro Ferreira, A., A. Tomas Marques, M. Bhide, V. Cubric-Curik, K. Hollung, C. H. Knight, K. Raundrup, J. Lippolis, M. Palmer, E. Sales-Baptista, S. S. Araujo, and A. M. de Almeida. 2015. Sequence Analysis of Bitter Taste Receptor Gene Repertoires in Different Ruminant Species. PLoS One. 10(6):e0124933. http://doi.org/10.1371/journal.pone.0124933.

Rossdeutscher, L., J. Li, A. L. Luco, I. Fadhil, B. Ochietti, A. Camirand, D. C. Huang, T. A. Reinhardt, W. Muller, and R. Kremer. 2015. Chemoprevention Activity of 25-Hydroxyvitamin D in the Mmtv-Pymt

Mouse Model of Breast Cancer. Cancer Prev Res (Phila). 8(2):120-8. http://doi.org/10.1158/1940-6207.CAPR-14-0110.

Ryman, V. E., G. M. Pighetti, J. D. Lippolis, J. C. Gandy, C. M. Applegate, and L. M. Sordillo. 2015. Quantification of Bovine Oxylipids During Intramammary Streptococcus Uberis Infection. Prostaglandins Other Lipid Mediat. 121(Pt B):207-17. http://doi.org/10.1016/j.prostaglandins.2015.09.006.

Sun, J., K. Aswath, S. G. Schroeder, J. D. Lippolis, T. A. Reinhardt, and T. S. Sonstegard. 2015. MicroRNA Expression Profiles of Bovine Milk Exosomes in Response to Staphylococcus Aureus Infection. BMC Genomics. 16: 806. http://doi.org/10.1186/s12864-015-2044-9.

Weiss, W. P., E. Azem, W. Steinberg, and T. A. Reinhardt. 2015. Effect of Feeding 25-Hydroxyvitamin D3 with a Negative Cation-Anion Difference Diet on Calcium and Vitamin D Status of Periparturient Cows and Their Calves. J Dairy Sci. 98(8):5588-600. http://doi.org/10.3168/jds.2014-9188.

Hernandez-Castellano, L. E., A. M. Ferreira, P. Nanni, J. Grossmann, A. Arguello, J. Capote, G. Cai, J. Lippolis, N. Castro, and A. M. de Almeida. 2016. The Goat (Capra Hircus) Mammary Gland Secretory Tissue Proteome as Influenced by Weight Loss: A Study Using Label Free Proteomics. J Proteomics. 145:60-69. http://doi.org/10.1016/j.jprot.2016.03.030.

Li, J., A. L. Luco, B. Ochietti, I. Fadhil, A. Camirand, T. A. Reinhardt, R. St-Arnaud, W. Muller, and R. Kremer. 2016. Tumoral Vitamin D Synthesis by Cyp27b1 1-Alpha-Hydroxylase Delays Mammary Tumor Progression in the Pymt-Mmtv Mouse Model and Its Action Involves Nf-Kappab Modulation. Endocrinology 157(6):2204-16. http://doi.org/10.1210/en.2015-1824.

Lippolis, J. D., B. W. Brunelle, T. A. Reinhardt, R. E. Sacco, T. C. Thacker, T. P. Looft, and T. A. Casey. 2016. Differential Gene Expression of Three Mastitis-Causing Escherichia Coli Strains Grown under Planktonic, Swimming, and Swarming Culture Conditions. mSystems. 1(4):e00064-16. http://doi.org/10.1128/mSystems.00064-16.

Nelson, C. D., J. D. Lippolis, T. A. Reinhardt, R. E. Sacco, J. L. Powell, M. E. Drewnoski, M. O'Neil, D. C. Beitz, and W. P. Weiss. 2016. Vitamin D Status of Dairy Cattle: Outcomes of Current Practices in the Dairy Industry. J Dairy Sci. 99(12):10150-60. http://doi.org/10.3168/jds.2016-11727.

Weiss, W. P., E. Azem, W. Steinberg, and T. A. Reinhardt. 2017. Corrigendum to Effect of Feeding 25-Hydroxyvitamin D3 with a Negative Cation-Anion Difference Diet on Calcium and Vitamin D Status of Periparturient Cows and Their Calves (J. Dairy Sci. 98:5588-5600). J Dairy Sci. 99(2):1693. http://doi.org/10.3168/jds.2016-99-2-1693.

Thacker, T. C., J. D. Lippolis, B. W. Brunelle, T. A. Casey, T. A. Reinhardt, R. E. Sacco, and D. B. Holman. 2017. Genome Sequences of Escherichia Coli Strains That Cause Persistent and Transient Mastitis. Genome Announc. 5(34):e00775-17. http://doi.org/10.1128/genomeA.00775-17.

Lippolis, J. D., D. B. Holman, B. W. Brunelle, T. C. Thacker, B. L. Bearson, T. A. Reinhardt, R. E. Sacco, and T. A. Casey. 2018. Genomic and Transcriptomic Analysis of Escherichia Coli Strains Associated with Persistent and Transient Bovine Mastitis and the Role of Colanic Acid. Infect Immun. 86(1):e00566-17. http://doi.org/10.1128/IAI.00566-17.

Looft, T., G. Cai, B. Choudhury, L. X. Lai, J. D. Lippolis, T. A. Reinhardt, M. J. Sylte, and T. A. Casey. 2018. Avian Intestinal Mucus Modulates Campylobacter Jejuni Gene Expression in a Host-Specific Manner. Front Microbiol. 9:3215. http://doi.org/10.3389/fmicb.2018.03215.

Powell, E. J., T. A. Reinhardt, E. Casas, and J. D. Lippolis. 2018. The Effect of Pegylated Granulocyte Colony-Stimulating Factor Treatment Prior to Experimental Mastitis in Lactating Holsteins. J Dairy Sci. 101(9):8182-93. http://doi.org/10.3168/jds.2018-14550.

Dominguez-Perez, D., J. Lippolis, M. Dennis, B. Miller, K. Tiley, V. Vasconcelos, A. M. de Almeida, and A. Campos. 2019. The Queen Conch (Lobatus Gigas) Proteome: A Valuable Tool for Biological Studies in Marine Gastropods. Protein J. 38(6):628-39. http://doi.org/10.1007/s10930-019-09857-0.

Lippolis, J. D., E. J. Powell, T. A. Reinhardt, T. C. Thacker, and E. Casas. 2019. Symposium Review: Omics in Dairy and Animal Science-Promise, Potential, and Pitfalls. J Dairy Sci. 102(5):4741-54. http://doi.org/10.3168/jds.2018-15267.

Putz, E. J., J. M. Eder, T. A. Reinhardt, R. E. Sacco, E. Casas, and J. D. Lippolis. 2019. Differential Phenotype of Immune Cells in Blood and Milk Following Pegylated Granulocyte Colony-Stimulating Factor Therapy During a Chronic Staphylococcus Aureus Infection in Lactating Holsteins. J Dairy Sci. 102(10):9268-84. http://doi.org/10.3168/jds.2019-16448.

Putz, E. J., A. M. Putz, H. Jeon, J. D. Lippolis, H. Ma, T. A. Reinhardt, and E. Casas. 2019. MicroRNA Profiles of Dry Secretions through the First Three Weeks of the Dry Period from Holstein Cows. Sci Rep. 9(1):19658. http://doi.org/10.1038/s41598-019-56193-5.

Snyder, N. A., M. V. Palmer, T. A. Reinhardt, and K. W. Cunningham. 2019. Milk Biosynthesis Requires the Golgi Cation Exchanger Tmem165. J Biol Chem. 294(9):3181-91. http://doi.org/10.1074/jbc.RA118.006270.

5030-32000-116-00D
Identification of Disease Mechanisms and Control Strategies for Bacterial Respiratory Pathogens in Ruminants; Randy Sacco (P), Fred Tatum, Hao Ma, Rohana Dassanayake, Eduardo Casas, Robert Briggs and Karen Register

Abrahante, J. E., S. S. Hunter, S. K. Maheswaran, M. J. Hauglund, F. M. Tatum, and R. E. Briggs. 2015. Draft Genome Sequence of Pasteurella Multocida Isolate P1062, Isolated from Bovine Respiratory Disease. Genome Announc. 3(5):e01254-15. http://doi.org/10.1128/genomeA.01254-15.

Andersson, L., A. L. Archibald, C. D. Bottema, R. Brauning, S. C. Burgess, D. W. Burt, E. Casas, H. H. Cheng, L. Clarke, C. Couldrey, B. P. Dalrymple, C. G. Elsik, S. Foissac, E. Giuffra, M. A. Groenen, B. J. Hayes, L. S. Huang, H. Khatib, J. W. Kijas, H. Kim, J. K. Lunney, F. M. McCarthy, J. C. McEwan, S. Moore, B. Nanduri, C. Notredame, Y. Palti, G. S. Plastow, J. M. Reecy, G. A. Rohrer, E. Sarropoulou, C. J. Schmidt, J. Silverstein, R. L. Tellam, M. Tixier-Boichard, G. Tosser-Klopp, C. K. Tuggle, J. Vilkki, S. N. White, S. Zhao, H. Zhou, and Faang Consortium. 2015. Coordinated International Action to Accelerate Genome-to-Phenome with Faang, the Functional Annotation of Animal Genomes Project. Genome Biol. 16:57. http://doi.org/10.1186/s13059-015-0622-4.

Hauglund, M. J., F. M. Tatum, D. O. Bayles, S. K. Maheswaran, and R. E. Briggs. 2015. Genome Sequences of Mannheimia Haemolytica Serotype A2 Isolates D171 and D35, Recovered from Bovine Pneumonia. Genome Announc. 3(2). http://doi.org/10.1128/genomeA.00093-15.

Hauglund, M. J., F. M. Tatum, D. O. Bayles, S. K. Maheswaran, and R. E. Briggs. 2015. Genome Sequences of Serotype A6 Mannheimia Haemolytica Isolates D174 and D38 Recovered from Bovine Pneumonia. Genome Announc. 3(2). http://doi.org/10.1128/genomeA.00086-15.

Ramirez, O., W. Burgos-Paz, E. Casas, M. Ballester, E. Bianco, I. Olalde, G. Santpere, V. Novella, M. Gut, C. Lalueza-Fox, M. Sana, and M. Perez-Enciso. 2015. Genome Data from a Sixteenth Century Pig Illuminate Modern Breed Relationships. Heredity (Edinb). 114(2):175-84. http://doi.org/10.1038/hdy.2014.81.

Register, K. B., L. Thole, R. F. Rosenbush, and F. C. Minion. 2015. Multilocus Sequence Typing of Mycoplasma Bovis Reveals Host-Specific Genotypes in Cattle Versus Bison. Vet Microbiol. 175(1):92-8. http://doi.org/10.1016/j.vetmic.2014.11.002.

Casas, E., G. Cai, L. A. Kuehn, K. B. Register, T. G. McDaneld, and J. D. Neill. 2016. Association of MicroRNAs with Antibody Response to Mycoplasma Bovis in Beef Cattle. PLoS One. 11(8):e0161651. http://doi.org/10.1371/journal.pone.0161651.

Casas, E. and M. E. Kehrli, Jr. 2016. A Review of Selected Genes with Known Effects on Performance and Health of Cattle. Front Vet Sci. 3:113. http://doi.org/10.3389/fvets.2016.00113.

McGill, J. L., R. A. Rusk, M. Guerra-Maupome, R. E. Briggs, and R. E. Sacco. 2016. Bovine Gamma Delta T Cells Contribute to Exacerbated Il-17 Production in Response to Co-Infection with Bovine RSV and Mannheimia Haemolytica. PLoS One. 11(3):e0151083. http://dx.doi.org/10.1371/journal.pone.0151083.

Tait, R. G., R. A. Cushman, A. K. McNeel, E. Casas, T. P. Smith, H. C. Freetly, and G. L. Bennett. 2016. Estimates of Epistatic and Pleiotropic Effects of () and () Genetic Markers on Beef Heifer Performance Traits Enhanced by Selection. J Anim Sci. 94(3):920-6. http://dx.doi.org/10.2527/jas.2015-9860.

Dassanayake, R. P., S. M. Falkenberg, R. E. Briggs, F. M. Tatum, and R. E. Sacco. 2017. Antimicrobial Activity of Bovine Nk-Lysin-Derived Peptides on Bovine Respiratory Pathogen Histophilus Somni. PLoS One. 12(8):e0183610. http://dx.doi.org/10.1371/journal.pone.0183610.

Dassanayake, R. P., S. Shanthalingam, W. Liu, E. Casas, and S. Srikumaran. 2017. Differential Susceptibility of Bighorn Sheep (Ovis Canadensis) and Domestic Sheep (Ovis Aries) Neutrophils to Mannheimia Haemolytica Leukotoxin Is Not Due to Differential Expression of Cell Surface Cd18. J Wildl Dis. 53(3):625-29. http://doi.org/10.7589/2016-11-244.

Klima, C. L., R. Zaheer, R. E. Briggs, and T. A. McAllister. 2017. A Multiplex PCR Assay for Molecular Capsular Serotyping of Mannheimia Haemolytica Serotypes 1, 2, and 6. J Microbiol Methods. 139:155-60. http://doi.org/10.1016/j.mimet.2017.05.010.

Petruzzi, B., R. E. Briggs, F. M. Tatum, W. E. Swords, C. De Castro, A. Molinaro, and T. J. Inzana. 2017. Capsular Polysaccharide Interferes with Biofilm Formation by Pasteurella Multocida Serogroup A. mBio. 8(6):e01849-17. http://doi.org/10.1128/mBio.01843-17.

Casas, E., G. Cai, L. A. Kuehn, K. B. Register, T. G. McDaneld, and J. D. Neill. 2018. Association of Circulating Transfer RNA Fragments with Antibody Response to Mycoplasma Bovis in Beef Cattle. BMC Vet Res. 14(1):89. http://doi.org/10.1186/s12917-018-1418-z.

Dassanayake, R. P., S. M. Falkenberg, K. B. Register, D. Samorodnitsky, E. M. Nicholson, and T. A. Reinhardt. 2018. Antimicrobial Activity of Bovine Nk-Lysin-Derived Peptides on Mycoplasma Bovis. PLoS One. 13(5):e0197677. http://doi.org/10.1371/journal.pone.0197677.

Elnaggar, M. M., G. S. Abdellrazeq, R. P. Dassanayake, L. M. Fry, V. Hulubei, and W. C. Davis. 2018. Characterization of Alphabeta and Gammadelta T Cell Subsets Expressing Il-17a in Ruminants and Swine. Dev Comp Immunol 85:115-24. http://doi.org/10.1016/j.dci.2018.04.003.

Madsen-Bouterse, S. A., M. A. Highland, R. P. Dassanayake, D. Zhuang, and D. A. Schneider. 2018. Low-Volume Goat Milk Transmission of Classical Scrapie to Lambs and Goat Kids. PLoS One. 13(9):e0204281. http://doi.org/10.1371/journal.pone.0204281.

Petruzzi, B., R. E. Briggs, F. M. Tatum, W. E. Swords, C. De Castro, A. Molinaro, and T. J. Inzana. 2018. Erratum for Petruzzi Et Al., Capsular Polysaccharide Interferes with Biofilm Formation by Pasteurella Multocida Serogroup A. mBio. 9(1). http://doi.org/10.1128/mBio.00176-18.

Register, K. B., W. D. Boatwright, K. M. Gesy, T. C. Thacker, and M. D. Jelinski. 2018. Mistaken Identity of an Open Reading Frame Proposed for Pcr-Based Identification of Mycoplasma Bovis and the Effect of Polymorphisms and Insertions on Assay Performance. J Vet Diagn Invest. 30(4):637-41. http://doi.org/10.1177/1040638718764799.

Register, K. B., S. C. Olsen, R. E. Sacco, J. Ridpath, S. Falkenberg, R. Briggs, C. Kanipe, and R. Madison. 2018. Relative Virulence in Bison and Cattle of Bison-Associated Genotypes of Mycoplasma Bovis. Vet Microbiol. 222:55-63. http://doi.org/10.1016/j.vetmic.2018.06.020.

Tait, R. G., Jr., R. A. Cushman, A. K. McNeel, E. Casas, T. P. L. Smith, H. C. Freetly, and G. L. Bennett. 2018. Mu-Calpain (Capn1), Calpastatin (Cast), and Growth Hormone Receptor (Ghr) Genetic Effects on Angus Beef Heifer Performance Traits and Reproduction. Theriogenology. 113:1-7. http://doi.org/10.1016/j.theriogenology.2018.02.002.

Bennett, G. L., R. G. Tait, Jr., S. D. Shackelford, T. L. Wheeler, D. A. King, E. Casas, and T. P. L. Smith. 2019. Enhanced Estimates of Carcass and Meat Quality Effects for Polymorphisms in Myostatin and Micro-Calpain Genes. J Anim Sci. 97(2):569-77. http://doi.org/10.1093/jas/sky451.

Dassanayake, R. P., S. M. Falkenberg, E. M. Nicholson, R. E. Briggs, F. M. Tatum, V. K. Sharma, and T. A. Reinhardt. 2019. Synthetic Bovine Nk-Lysin-Derived Peptide (Bnk2a) Does Not Require Intra-Chain Disulfide Bonds for Bactericidal Activity. PLoS One. 14(6):e0218507. http://doi.org/10.1371/journal.pone.0218507.

Register, K. B., M. D. Jelinski, M. Waldner, W. D. Boatwright, T. K. Anderson, D. L. Hunter, R. G. Hamilton, P. Burrage, T. Shury, R. Bildfell, P. L. Wolff, D. Miskimins, R. J. Derscheid, and M. R. Woodbury. 2019. Comparison of Multilocus Sequence Types Found among North American Isolates of Mycoplasma Bovis from Cattle, Bison, and Deer, 2007-2017. J Vet Diagn Invest. 31(6):899-904. http://doi.org/10.1177/1040638719874848.

Register, K. B., L. Thole, R. F. Rosenbush, and F. C. Minion. 2019. Corrigendum to 'Multilocus Sequence Typing of Mycoplasma Bovis Reveals Host-Specific Genotypes in Cattle Versus Bison' Vet. Microbiol. 228:264-65. http://doi.org/10.1016/j.vetmic.2018.11.012.

5030-32000-117-00D
Identification of Disease Mechanisms and Control Strategies for Viral Respiratory Pathogens of Ruminants; John Neill (P), Hao Ma, Rohana Dassanake, Randy Sacco, Shollie Falkenberg and Eduardo Casas

Hwang, S., J. J. Greenlee, and E. M. Nicholson. 2020. Role of Donor Genotype in Rt-Quic Seeding Activity of Chronic Wasting Disease Prions Using Human and Bank Vole Substrates. PLoS One. 15(1):e0227487. http://doi.org/10.1371/journal.pone.0227487.

Michael, A. V., J. J. Greenlee, T. A. Harm, S. J. Moore, M. Zhang, M. S. Lind, M. H. W. Greenlee, and J. D. Smith. 2020. In Situ Temporospatial Characterization of the Glial Response to Prion Infection. Vet Pathol. 57(1):90-107. http://doi.org/10.1177/0300985819861708.

Hofstetter, A. R. and R. E. Sacco. 2020. Oxidative Stress Pathway Gene Transcription after Bovine Respiratory Syncytial Virus Infection in Vitro and Ex Vivo. Vet Immunol Immunopathol. 219:109956. http://doi.org/10.1016/j.vetimm.2019.109956.

Falkenberg, S. M., R. P. Dassanayake, P. Walz, E. Casas, J. D. Neill, and J. F. Ridpath. 2019. Frequency of Bovine Viral Diarrhea Virus Detected in Subpopulations of Peripheral Blood Mononuclear Cells in Persistently Infected Animals and Health Outcome. Vet Immunol Immunopathol. 207:46-52. http://doi.org/10.1016/j.vetimm.2018.11.015.

Neill, J. D., B. M. Crossley, A. C. Mosena, J. F. Ridpath, D. O. Bayles, S. K. Hietala, M. L. Killian, and S. M. Falkenberg. 2019. Genomic and Antigenic Characterization of a Cytopathic Bovine Viral Diarrhea Virus 1i Isolated in the United States. Virology. 535:279-82. http://doi.org/10.1016/j.virol.2019.07.020.

Neill, J. D., A. M. Workman, R. Hesse, J. Bai, E. P. Porter, B. Meadors, J. Anderson, D. O. Bayles, and S. M. Falkenberg. 2019. Identification of Bvdv2b and 2c Subgenotypes in the United States: Genetic and Antigenic Characterization. Virology. 528:19-29. http://doi.org/10.1016/j.virol.2018.12.002.

Silveira, S., S. M. Falkenberg, R. P. Dassanayake, P. H. Walz, J. F. Ridpath, C. W. Canal, and J. D. Neill. 2019. In Vitro Method to Evaluate Virus Competition between Bvdv-1 and Bvdv-2 Strains Using the Primeflow RNA Assay. Virology. 536:101-09. http://doi.org/10.1016/j.virol.2019.07.029.

Silveira, S., S. M. Falkenberg, B. S. Kaplan, B. Crossley, J. F. Ridpath, F. B. Bauermann, C. P. Fossler, D. A. Dargatz, R. P. Dassanayake, A. L. Vincent, C. W. Canal, and J. D. Neill. 2019. Serosurvey for Influenza D Virus Exposure in Cattle, United States, 2014-2015. Emerg Infect Dis. 25(11):2074-80. http://doi.org/10.3201/eid2511.190253.

Pecora, A., M. S. Perez Aguirreburualde, J. F. Ridpath, and M. J. Dus Santos. 2019. Molecular Characterization of Pestiviruses in Fetal Bovine Sera Originating from Argentina: Evidence of Circulation of Hobi-Like Viruses. Front Vet Sci. 6:359. http://doi.org/10.3389/fvets.2019.00359.

Guerra-Maupome, M., M. V. Palmer, J. L. McGill, and R. E. Sacco. 2019. Utility of the Neonatal Calf Model for Testing Vaccines and Intervention Strategies for Use against Human RSV Infection. Vaccines (Basel). 7(1):7. http://doi.org/10.3390/vaccines7010007.

McGill, J. L., S. M. Kelly, M. Guerra-Maupome, E. Winkley, J. Henningson, B. Narasimhan, and R. E. Sacco. 2019. Vitamin A Deficiency Impairs the Immune Response to Intranasal Vaccination and RSV Infection in Neonatal Calves. Sci Rep. 9(1):15157. http://doi.org/10.1038/s41598-019-51684-x.

Falkenberg, S. M., R. P. Dassanayake, J. D. Neill, and J. F. Ridpath. 2018. Evaluation of Bovine Viral Diarrhea Virus Transmission Potential to Naive Calves by Direct and Indirect Exposure Routes. Vet Microbiol. 217:144-48. http://doi.org/10.1016/j.vetmic.2018.03.012.

Flores, E. F., J. F. Cargnelutti, F. L. Monteiro, F. V. Bauermann, J. F. Ridpath, and R. Weiblen. 2018. A Genetic Profile of Bovine Pestiviruses Circulating in Brazil (1998-2018). Anim Health Res Rev. 19(2):134-41. http://doi.org/10.1017/S1466252318000130.

Gomez-Romero, N., F. J. Basurto-Alcantara, A. Verdugo-Rodriguez, R. Lagunes-Quintanilla, F. V. Bauermann, and J. F. Ridpath. 2018. Detection of Border Disease Virus in Mexican Cattle. Transbound Emerg Dis. 65(1):267-71. http://doi.org/10.1111/tbed.12641.

Kuca, T., T. Passler, B. W. Newcomer, J. D. Neill, P. K. Galik, K. P. Riddell, Y. Zhang, and P. H. Walz. 2018. Identification of Conserved Amino Acid Substitutions During Serial Infection of Pregnant Cattle and Sheep with Bovine Viral Diarrhea Virus. Front Microbiol. 9:1109. http://doi.org/10.3389/fmicb.2018.01109.

Silveira, S., L. F. Baumbach, M. N. Weber, A. C. S. Mosena, M. S. da Silva, S. P. Cibulski, M. R. Borba, R. D. Maia, V. C. S. Coimbra, G. M. de Moraes, J. F. Ridpath, and C. W. Canal. 2018. Hobi-Like Is the Most Prevalent Ruminant Pestivirus in Northeastern Brazil. Transbound Emerg Dis. 65(1):e113-e20. http://doi.org/10.1111/tbed.12689.

Silveira, S., S. M. Falkenberg, M. J. Elderbrook, K. S. Sondgeroth, R. P. Dassanayake, J. D. Neill, J. F. Ridpath, and C. W. Canal. 2018. Serological Survey for Antibodies against Pestiviruses in Wyoming Domestic Sheep. Vet Microbiol. 219:96-99. http://doi.org/10.1016/j.vetmic.2018.04.019.

Walz, P. H., K. P. Riddell, B. W. Newcomer, J. D. Neill, S. M. Falkenberg, V. S. Cortese, D. W. Scruggs, and T. H. Short. 2018. Comparison of Reproductive Protection against Bovine Viral Diarrhea Virus Provided by Multivalent Viral Vaccines Containing Inactivated Fractions of Bovine Viral Diarrhea Virus 1 and 2. Vaccine. 36(26):3853-60. http://doi.org/10.1016/j.vaccine.2018.04.005.

McGill, J. L., S. M. Kelly, P. Kumar, S. Speckhart, S. L. Haughney, J. Henningson, B. Narasimhan, and R. E. Sacco. 2018. Efficacy of Mucosal Polyanhydride Nanovaccine against Respiratory Syncytial Virus Infection in the Neonatal Calf. Sci Rep. 8(1):3021. http://dx.doi.org/10.1038/s41598-018-21292-2.

Bauermann, F. V., S. M. Falkenberg, and J. F. Ridpath. 2017. Hobi-Like Virus RNA Detected in Foetuses Following Challenge of Pregnant Cows That Had Previously Given Birth to Calves Persistently Infected with Bovine Viral Diarrhoea Virus. Transbound Emerg Dis. 64(5):1624-32. http://doi.org/10.1111/tbed.12556.

Bauermann, F. V., J. F. Ridpath, and D. A. Dargatz. 2017. Bovine Leukemia Virus Seroprevalence among Cattle Presented for Slaughter in the United States. J Vet Diagn Invest. 29(5):704-06. http://doi.org/10.1177/1040638717702183.

Bauermann, F. V., J. F. Ridpath, and D. A. Dargatz. 2017. A Serosurvey for Ruminant Pestivirus Exposure Conducted Using Cattle Sera Collected for Brucellosis Surveillance in the United States. J Vet Diagn Invest. 29(1):76-82. http://doi.org/10.1177/1040638716680251.

Dias, R. K., J. F. Cargnelutti, M. N. Weber, C. W. Canal, F. V. Bauermann, J. F. Ridpath, R. Weiblen, and E. F. Flores. 2017. Antigenic Diversity of Brazilian Isolates of Hobi-Like Pestiviruses. Vet Microbiol. 203:221-28. http://doi.org/10.1016/j.vetmic.2017.03.021.

Falkenberg, S. M., F. V. Bauermann, and J. F. Ridpath. 2017. Characterization of Thymus-Associated Lymphoid Depletion in Bovine Calves Acutely or Persistently Infected with Bovine Viral Diarrhea Virus 1, Bovine Viral Diarrhea Virus 2 or Hobi-Like Pestivirus. Arch Virol. 162:3473-80. http://doi.org/10.1007/s00705-017-3523-x.

Falkenberg, S. M., R. P. Dassanayake, J. D. Neill, and J. F. Ridpath. 2017. Improved Detection of Bovine Viral Diarrhea Virus in Bovine Lymphoid Cell Lines Using Primeflow RNA Assay. Virology. 509:260-65. http://doi.org/10.1016/j.virol.2017.06.032.

Fulton, R. W., A. W. Confer, N. J. Sorensen, J. F. Ridpath, and L. J. Burge. 2017. Bovine Viral Diarrhea Virus 1b Fetal Infection with Extensive Hemorrhage. J Vet Diagn Invest. 29(6) 880-84. http://doi.org/10.1177/1040638717726825.

Fulton, R.W., Neill, J.D., Saliki, J.T., Landis, C., Burge, L.J., Payton, M.E. 2017. Genomic and antigenic characterization of bovine parainfluenze-3 viruses in the United States including modified live virus vaccine (MLV) strains and field strains from cattle. Virus Research. 235:77-81. https://doi.org/10.1016/j.virusres.2017.04.009.

Gomez-Romero, N., F. J. Basurto-Alcantara, A. Verdugo-Rodriguez, F. V. Bauermann, and J. F. Ridpath. 2017. Genetic Diversity of Bovine Viral Diarrhea Virus in Cattle from Mexico. J Vet Diagn Invest. 29(3):362-65. http://doi.org/10.1177/1040638717690187.

Newcomer, B. W., J. D. Neill, P. K. Galik, K. P. Riddell, Y. Zhang, T. Passler, B. T. Velayudhan, and P. H. Walz. 2017. Serologic Survey for Antibodies against Three Genotypes of Bovine Parainfluenza 3 Virus in Unvaccinated Ungulates in Alabama. Am J Vet Res. 78(2):239-43. http://doi.org/10.2460/ajvr.78.2.239.

Ridpath, J. F., J. D. Neill, M. V. Palmer, F. V. Bauermann, S. M. Falkenberg, and P. L. Wolff. 2017. Isolation and Characterization of a Novel Cervid Adenovirus from White-Tailed Deer (Odocoileus Virginianus) Fawns in a Captive Herd. Virus Res. 238:198-203. http://doi.org/10.1016/j.virusres.2017.06.020.

Runyan, C. A., E. D. Downey-Slinker, J. F. Ridpath, T. B. Hairgrove, J. E. Sawyer, and A. D. Herring. 2017. Feed Intake and Weight Changes in Bos Indicus-Bos Taurus Crossbred Steers Following Bovine Viral Diarrhea Virus Type 1b Challenge under Production Conditions. Pathogens. 6(4):66. http://doi.org/10.3390/pathogens6040066.

Silveira, S., M. N. Weber, A. C. Mosena, M. S. da Silva, A. F. Streck, C. A. Pescador, E. F. Flores, R. Weiblen, D. Driemeier, J. F. Ridpath, and C. W. Canal. 2017. Genetic Diversity of Brazilian Bovine Pestiviruses Detected between 1995 and 2014. Transbound Emerg Dis. 64(2):613-23. http://doi.org/10.1111/tbed.12427.

Stanton, J. B., B. Swanson, E. Orozco, J. F. Munoz-Gutierrez, J. F. Evermann, and J. F. Ridpath. 2017. Immortalized Sheep Microglial Cells Are Permissive to a Diverse Range of Ruminant Viruses. Vet Q. 37(1):52-56. http://doi.org/10.1080/01652176.2017.1297550.

Weber, M. N., F. V. Bauermann, C. W. Canal, D. O. Bayles, J. D. Neill, and J. F. Ridpath. 2017. Temporal Dynamics of 'Hobi'-Like Pestivirus Quasispecies in Persistently Infected Calves Generated under Experimental Conditions. Virus Res. 227:23-33. http://doi.org/10.1016/j.virusres.2016.09.018.

Weber, M. N., F. V. Bauermann, N. Gomez-Romero, A. D. Herring, C. W. Canal, J. D. Neill, and J. F. Ridpath. 2017. Variation in Pestivirus Growth in Testicle Primary Cell Culture Is More Dependent on the Individual Cell Donor Than Cattle Breed. Vet Res Commun. 41(1):1-7. http://doi.org/10.1007/s11259-016-9666-5.

Bauermann, F. V., S. M. Falkenberg, and J. F. Ridpath. 2017. Hobi-Like Virus RNA Detected in Foetuses Following Challenge of Pregnant Cows That Had Previously Given Birth to Calves Persistently Infected with Bovine Viral Diarrhoea Virus. Transbound Emerg Dis. 64(5):1624-32. http://doi.org/10.1111/tbed.12556.

Bauermann, F. V., J. F. Ridpath, and D. A. Dargatz. 2017. Bovine Leukemia Virus Seroprevalence among Cattle Presented for Slaughter in the United States. J Vet Diagn Invest. 29(5):704-06. http://doi.org/10.1177/1040638717702183.

Bauermann, F. V., J. F. Ridpath, and D. A. Dargatz. 2017. A Serosurvey for Ruminant Pestivirus Exposure Conducted Using Cattle Sera Collected for Brucellosis Surveillance in the United States. J Vet Diagn Invest. 29(1):76-82. http://doi.org/10.1177/1040638716680251.

Dias, R. K., J. F. Cargnelutti, M. N. Weber, C. W. Canal, F. V. Bauermann, J. F. Ridpath, R. Weiblen, and E. F. Flores. 2017. Antigenic Diversity of Brazilian Isolates of Hobi-Like Pestiviruses. Vet Microbiol. 203:221-28. http://doi.org/10.1016/j.vetmic.2017.03.021.

Falkenberg, S. M., F. V. Bauermann, and J. F. Ridpath. 2017. Characterization of Thymus-Associated Lymphoid Depletion in Bovine Calves Acutely or Persistently Infected with Bovine Viral Diarrhea Virus 1, Bovine Viral Diarrhea Virus 2 or Hobi-Like Pestivirus. Arch Virol. 162(11):3473-80. http://doi.org/10.1007/s00705-017-3523-x.

Falkenberg, S. M., R. P. Dassanayake, J. D. Neill, and J. F. Ridpath. 2017. Improved Detection of Bovine Viral Diarrhea Virus in Bovine Lymphoid Cell Lines Using Primeflow RNA Assay. Virology. 509:260-65. http://doi.org/10.1016/j.virol.2017.06.032.

Fulton, R. W., A. W. Confer, N. J. Sorensen, J. F. Ridpath, and L. J. Burge. 2017. Bovine Viral Diarrhea Virus 1b Fetal Infection with Extensive Hemorrhage. J Vet Diagn Invest. 29(6):880-84. http://doi.org/10.1177/1040638717726825.

Fulton, R.W., Neill, J.D., Saliki, J.T., Landis, C., Burge, L.J., Payton, M.E. 2017. Genomic and antigenic characterization of bovine parainfluenze-3 viruses in the United States including modified live virus vaccine (MLV) strains and field strains from cattle. Virus Research. 235:77-81. https://doi.org/10.1016/j.virusres.2017.04.009.

Gomez-Romero, N., F. J. Basurto-Alcantara, A. Verdugo-Rodriguez, F. V. Bauermann, and J. F. Ridpath. 2017. Genetic Diversity of Bovine Viral Diarrhea Virus in Cattle from Mexico. J Vet Diagn Invest. 29(3):362-65. http://doi.org/10.1177/1040638717690187.

Newcomer, B. W., J. D. Neill, P. K. Galik, K. P. Riddell, Y. Zhang, T. Passler, B. T. Velayudhan, and P. H. Walz. 2017. Serologic Survey for Antibodies against Three Genotypes of Bovine Parainfluenza 3 Virus in Unvaccinated Ungulates in Alabama. Am J Vet Res. 78(2):239-43. http://doi.org/10.2460/ajvr.78.2.239.

Ridpath, J. F., J. D. Neill, M. V. Palmer, F. V. Bauermann, S. M. Falkenberg, and P. L. Wolff. 2017. Isolation and Characterization of a Novel Cervid Adenovirus from White-Tailed Deer (Odocoileus

Virginianus) Fawns in a Captive Herd. Virus Res. 238:198-203. http://doi.org/10.1016/j.virusres.2017.06.020.

Runyan, C. A., E. D. Downey-Slinker, J. F. Ridpath, T. B. Hairgrove, J. E. Sawyer, and A. D. Herring. 2017. Feed Intake and Weight Changes in Bos Indicus-Bos Taurus Crossbred Steers Following Bovine Viral Diarrhea Virus Type 1b Challenge under Production Conditions. Pathogens. 6(4):66. http://doi.org/10.3390/pathogens6040066.

Silveira, S., M. N. Weber, A. C. Mosena, M. S. da Silva, A. F. Streck, C. A. Pescador, E. F. Flores, R. Weiblen, D. Driemeier, J. F. Ridpath, and C. W. Canal. 2017. Genetic Diversity of Brazilian Bovine Pestiviruses Detected between 1995 and 2014. Transbound Emerg Dis. 64(2):613-23. http://doi.org/10.1111/tbed.12427.

Stanton, J. B., B. Swanson, E. Orozco, J. F. Munoz-Gutierrez, J. F. Evermann, and J. F. Ridpath. 2017. Immortalized Sheep Microglial Cells Are Permissive to a Diverse Range of Ruminant Viruses. Vet Q. 37(1):52-56. http://doi.org/10.1080/01652176.2017.1297550.

Weber, M. N., F. V. Bauermann, C. W. Canal, D. O. Bayles, J. D. Neill, and J. F. Ridpath. 2017. Temporal Dynamics of 'Hobi'-Like Pestivirus Quasispecies in Persistently Infected Calves Generated under Experimental Conditions. Virus Res. 227:23-33. http://doi.org/10.1016/j.virusres.2016.09.018.

Weber, M. N., F. V. Bauermann, N. Gomez-Romero, A. D. Herring, C. W. Canal, J. D. Neill, and J. F. Ridpath. 2017. Variation in Pestivirus Growth in Testicle Primary Cell Culture Is More Dependent on the Individual Cell Donor Than Cattle Breed. Vet Res Commun. 41(1):1-7. http://doi.org/10.1007/s11259-016-9666-5.

Flowers, E. M., T. R. Bachvaroff, J. V. Warg, J. D. Neill, M. L. Killian, A. S. Vinagre, S. Brown, A. S. Almeida, and E. J. Schott. 2016. Genome Sequence Analysis of Csrv1: A Pathogenic Reovirus That Infects the Blue Crab Callinectes Sapidus across Its Trans-Hemispheric Range. Front Microbiol. 7:126. http://doi.org/10.3389/fmicb.2016.00126.

Fulton, R. W., J. M. d'Offay, C. Landis, D. G. Miles, R. A. Smith, J. T. Saliki, J. F. Ridpath, A. W. Confer, J. D. Neill, R. Eberle, T. J. Clement, C. C. Chase, L. J. Burge, and M. E. Payton. 2016. Detection and Characterization of Viruses as Field and Vaccine Strains in Feedlot Cattle with Bovine Respiratory Disease. Vaccine. 34(30):3478-92. http://doi.org/10.1016/j.vaccine.2016.04.020.

Ridpath, J. F. and J. D. Neill. 2016. Challenges in Identifying and Determining the Impacts of Infection with Pestiviruses on the Herd Health of Free Ranging Cervid Populations. Front Microbiol. 7:921. http://doi.org/10.3389/fmicb.2016.00921.

Ridpath, J. F. and T. Passler. 2016. Editorial: Control of Pestivirus Infections in the Management of Wildlife Populations. Front Microbiol. 7:1396. http://doi.org/10.3389/fmicb.2016.01396.

Schaut, R. G., J. F. Ridpath, and R. E. Sacco. 2016. Bovine Viral Diarrhea Virus Type 2 Impairs Macrophage Responsiveness to Toll-Like Receptor Ligation with the Exception of Toll-Like Receptor 7. PLoS One. 11(7:e0159491. http://doi.org/10.1371/journal.pone.0159491.

Weber, M. N., F. V. Bauermann, D. O. Bayles, C. W. Canal, J. D. Neill, and J. F. Ridpath. 2016. Comparison of 'Hobi'-Like Viral Populations among Persistent Infected Calves Generated under Experimental Conditions and to Inoculum Virus. Virology. 492:225-31. http://doi.org/10.1016/j.virol.2016.03.001.

Weber, M. N., A. C. Mosena, S. V. Simoes, L. L. Almeida, C. R. Pessoa, R. F. Budaszewski, T. R. Silva, J. F. Ridpath, F. Riet-Correa, D. Driemeier, and C. W. Canal. 2016. Clinical Presentation Resembling Mucosal Disease Associated with 'Hobi'-Like Pestivirus in a Field Outbreak. Transbound Emerg Dis. 63(1):92-100. http://doi.org/10.1111/tbed.12223.

Wolff, P. L., C. Schroeder, C. McAdoo, M. Cox, D. D. Nelson, J. F. Evermann, and J. F. Ridpath. 2016. Evidence of Bovine Viral Diarrhea Virus Infection in Three Species of Sympatric Wild Ungulates in Nevada: Life History Strategies May Maintain Endemic Infections in Wild Populations. Front Microbiol. 7:292. http://doi.org/10.3389/fmicb.2016.00292.

McGill, J. L., R. A. Rusk, M. Guerra-Maupome, R. E. Briggs, and R. E. Sacco. 2016. Bovine Gamma Delta T Cells Contribute to Exacerbated Il-17 Production in Response to Co-Infection with Bovine RSV and *Mannheimia haemolytica*. PLoS One. 11(3): e0151083. http://dx.doi.org/10.1371/journal.pone.0151083.

McGill, J. L. and R. E. Sacco. 2016. Gammadelta T Cells and the Immune Response to Respiratory Syncytial Virus Infection. Vet Immunol Immunopathol. 181:24-29. http://dx.doi.org/10.1016/j.vetimm.2016.02.012.

Bauermann, F. V., S. M. Falkenberg, N. Decaro, E. F. Flores, and J. F. Ridpath. 2015. Experimental Infection of Calves, Sheep, Goats and Pigs with Hobi-Like Viruses by Direct Inoculation or Exposure to Persistently Infected Calves. Vet Microbiol. 181(3-4):289-93. http://doi.org/10.1016/j.vetmic.2015.10.011.

Bauermann, F. V. and J. F. Ridpath. 2015. Hobi-Like Viruses--the Typical 'Atypical Bovine Pestivirus'. Anim Health Res Rev. 16(1):64-9. http://doi.org/10.1017/S146625231500002X.

Casas, E., G. Cai, and J. D. Neill. 2015. Characterization of Circulating Transfer RNA-Derived RNA Fragments in Cattle. Front Genet. 6:271. http://doi.org/10.3389/fgene.2015.00271.

Darweesh, M. F., M. K. Rajput, L. J. Braun, J. F. Ridpath, J. D. Neill, and C. C. Chase. 2015. Characterization of the Cytopathic BVDV Strains Isolated from 13 Mucosal Disease Cases Arising in a Cattle Herd. Virus Res. 195:141-7. http://doi.org/10.1016/j.virusres.2014.09.015.

Eberle, K. C., J. D. Neill, S. K. Venn-Watson, J. L. McGill, and R. E. Sacco. 2015. Novel Atlantic Bottlenose Dolphin Parainfluenza Virus Ttpiv-1 Clusters with Bovine Piv-3 Genotype B Strains. Virus Genes. 51(2):198-208. http://doi.org/10.1007/s11262-015-1224-7.

Fulton, R. W., H. R. Herd, N. J. Sorensen, A. W. Confer, J. W. Ritchey, J. F. Ridpath, and L. J. Burge. 2015. Enteric Disease in Postweaned Beef Calves Associated with Bovine Coronavirus Clade 2. J Vet Diagn Invest. 27(1):97-101. http://doi.org/10.1177/1040638714559026.

Giammarioli, M., J. F. Ridpath, E. Rossi, M. Bazzucchi, C. Casciari, and G. M. De Mia. 2015. Genetic Detection and Characterization of Emerging Hobi-Like Viruses in Archival Foetal Bovine Serum Batches. Biologicals. 43(4):220-4. http://doi.org/10.1016/j.biologicals.2015.05.009.

Kane, S. E., L. D. Holler, L. J. Braun, J. D. Neill, D. B. Young, J. F. Ridpath, and C. C. Chase. 2015. Bovine Viral Diarrhea Virus Outbreak in a Beef Cow Herd in South Dakota. J Am Vet Med Assoc. 246(12):1358-62. http://doi.org/10.2460/javma.246.12.1358.

Kirkland, P. D., M. J. Frost, K. R. King, D. S. Finlaison, C. L. Hornitzky, X. Gu, M. Richter, I. Reimann, M. Dauber, H. Schirrmeier, M. Beer, and J. F. Ridpath. 2015. Genetic and Antigenic Characterization of Bungowannah Virus, a Novel Pestivirus. Vet Microbiol. 178(3-4):252-9. http://doi.org/10.1016/j.vetmic.2015.05.014.

Neill, J. D., E. J. Dubovi, and J. F. Ridpath. 2015. Identification of Amino Acid Changes in the Envelope Glycoproteins of Bovine Viral Diarrhea Viruses Isolated from Alpaca That May Be Involved in Host Adaptation. Vet Microbiol. 179(3-4):299-303. http://doi.org/10.1016/j.vetmic.2015.06.007.

Neill, J. D., J. F. Ridpath, and B. T. Valayudhan. 2015. Identification and Genome Characterization of Genotype B and Genotype C Bovine Parainfluenza Type 3 Viruses Isolated in the United States. BMC Vet Res. 11:112. http://dx.doi.org/10.1186/s12917-015-0431-8.

Ridpath, J. F. 2015. Emerging Pestiviruses Infecting Domestic and Wildlife Hosts. Anim Health Res Rev. 16(1):55-9. http://doi.org/10.1017/S1466252315000067.

Ridpath, J. F., D. O. Bayles, J. D. Neill, S. M. Falkenberg, F. V. Bauermann, L. Holler, L. J. Braun, D. B. Young, S. E. Kane, and C. C. Chase. 2015. Comparison of the Breadth and Complexity of Bovine Viral Diarrhea (Bvdv) Populations Circulating in 34 Persistently Infected Cattle Generated in One Outbreak. Virology. 485:297-304. http://doi.org/10.1016/j.virol.2015.07.022.

Ridpath, J. F. and J. D. Neill. 2015. Pestiviruses: Old Enemies and New Challenges. Anim Health Res Rev. 16(1):1-3. http://doi.org/10.1017/S1466252315000134.

Schaut, R. G., J. L. McGill, J. D. Neill, J. F. Ridpath, and R. E. Sacco. 2015. Bovine Viral Diarrhea Virus Type 2 in Vivo Infection Modulates Tlr4 Responsiveness in Differentiated Myeloid Cells Which Is Associated with Decreased Myd88 Expression. Virus Res. 208:44-55. http://doi.org/10.1016/j.virusres.2015.05.017.

Downey-Slinker, E. D., J. F. Ridpath, J. E. Sawyer, L. C. Skow, and A. D. Herring. 2016. Antibody Titers to Vaccination Are Not Predictive of Level of Protection against a Bvdv Type 1b Challenge in Bos Indicus - Bos Taurus Steers. Vaccine. 34(42):5053-59. http://doi.org/10.1016/j.vaccine.2016.08.087.

Eberle, K. C., J. L. McGill, T. A. Reinhardt, and R. E. Sacco. 2015. Parainfluenza Virus 3 Blocks Antiviral Mediators Downstream of the Interferon Lambda Receptor by Modulating Stat1 Phosphorylation. J Virol. 90(6):2948-58. http://doi.org/10.1128/JVI.02502-15.

Hansen, T. R., N. P. Smirnova, B. T. Webb, H. Bielefeldt-Ohmann, R. E. Sacco, and H. Van Campen. 2015. Innate and Adaptive Immune Responses to in Utero Infection with Bovine Viral Diarrhea Virus. Anim Health Res Rev. 16(1):15-26. http://doi.org/10.1017/S1466252315000122.

Sacco, R. E., R. K. Durbin, and J. E. Durbin. 2015. Animal Models of Respiratory Syncytial Virus Infection and Disease. Curr Opin Virol. 13:117-22. http://doi.org/10.1016/j.coviro.2015.06.003.

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5030-32000-114-00D
Pathobiology, Genetics, and Detection of Transmissible Spongiform Encephalopathies; Eric Nicholson (P), Kelly Lager and Justin Greenlee

Comoy, E. E., J. Mikol, S. Luccantoni-Freire, E. Correia, N. Lescoutra-Etchegaray, V. Durand, C. Dehen, O. Andreoletti, C. Casalone, J. A. Richt, J. J. Greenlee, T. Baron, S. L. Benestad, P. Brown, and J. P. Deslys. 2015. Transmission of Scrapie Prions to Primate after an Extended Silent Incubation Period. Sci Rep. 5:11573. http://doi.org/10.1038/srep11573.

Greenlee, J. J. and M. H. Greenlee. 2015. The Transmissible Spongiform Encephalopathies of Livestock. ILAR J. 56(1):7-25. http://doi.org/10.1093/ilar/ilv008.

Greenlee, M. H., J. D. Smith, E. M. Platt, J. R. Juarez, L. L. Timms, and J. J. Greenlee. 2015. Changes in Retinal Function and Morphology Are Early Clinical Signs of Disease in Cattle with Bovine Spongiform Encephalopathy. PLoS One. 10(3):e0119431. http://doi.org/10.1371/journal.pone.0119431.

Munoz-Gutierrez, J. F., D. A. Schneider, T. V. Baszler, J. J. Greenlee, E. M. Nicholson, and J. B. Stanton. 2015. Htert-Immortalized Ovine Microglia Propagate Natural Scrapie Isolates. Virus Res. 198:35-43. http://doi.org/10.1016/j.virusres.2014.10.028.

Nicholson, E. M. 2015. Detection of the Disease-Associated Form of the Prion Protein in Biological Samples. Bioanalysis. 7(2):253-61. http://doi.org/10.4155/bio.14.301.

Greenlee, J. J., J. D. Smith, and A. N. Hamir. 2016. Oral Inoculation of Neonatal Suffolk Sheep with the Agent of Classical Scrapie Results in Prp(Sc) Accumulation in Sheep with the Prnp Arq/Arq but Not the Arq/Arr Genotype. Res Vet Sci. 105:188-91. http://doi.org/10.1016/j.rvsc.2016.02.016.

Haley, N. J., C. Siepker, J. J. Greenlee, and J. A. Richt. 2016. Limited Amplification of Chronic Wasting Disease Prions in the Peripheral Tissues of Intracerebrally Inoculated Cattle. J Gen Virol. 97(7):1720-24. http://doi.org/10.1099/jgv.0.000438.

Haley, N. J., C. Siepker, W. D. Walter, B. V. Thomsen, J. J. Greenlee, A. D. Lehmkuhl, and J. A. Richt. 2016. Antemortem Detection of Chronic Wasting Disease Prions in Nasal Brush Collections and Rectal Biopsy Specimens from White-Tailed Deer by Real-Time Quaking-Induced Conversion. J Clin Microbiol. 54(4):1108-16. http://doi.org/10.1128/JCM.02699-15.

Moore, S. J., R. Kunkle, M. H. Greenlee, E. Nicholson, J. Richt, A. Hamir, W. R. Waters, and J. Greenlee. 2016. Horizontal Transmission of Chronic Wasting Disease in Reindeer. Emerg Infect Dis. 22(12):2142-45. http://doi.org/10.3201/eid2212.160635.

Moore, S. J., J. D. Smith, M. H. Greenlee, E. M. Nicholson, J. A. Richt, and J. J. Greenlee. 2016. Comparison of Two Us Sheep Scrapie Isolates Supports Identification as Separate Strains. Vet Pathol. 53(6):1187-96. http://doi.org/10.1177/0300985816629712.

Moore, S. J., M. H. West Greenlee, J. D. Smith, C. E. Vrentas, E. M. Nicholson, and J. J. Greenlee. 2016. A Comparison of Classical and H-Type Bovine Spongiform Encephalopathy Associated with E211k Prion Protein Polymorphism in Wild-Type and Ek211 Cattle Following Intracranial Inoculation. Front Vet Sci. 3:78. http://doi.org/10.3389/fvets.2016.00078.

West Greenlee, M. H., M. Lind, R. Kokemuller, N. Mammadova, N. Kondru, S. Manne, J. Smith, A. Kanthasamy, and J. Greenlee. 2016. Temporal Resolution of Misfolded Prion Protein Transport,

Accumulation, Glial Activation, and Neuronal Death in the Retinas of Mice Inoculated with Scrapie. Am J Pathol. 186(9):2302-9. http://doi.org/10.1016/j.ajpath.2016.05.018.

Hwang, S., J. J. Greenlee, and E. M. Nicholson. 2017. Use of Bovine Recombinant Prion Protein and Real-Time Quaking-Induced Conversion to Detect Cattle Transmissible Mink Encephalopathy Prions and Discriminate Classical and Atypical L- and H-Type Bovine Spongiform Encephalopathy. PLoS One. 12(2):e0172391. http://doi.org/10.1371/journal.pone.0172391.

Hwang, S., M. H. West Greenlee, A. Balkema-Buschmann, M. H. Groschup, E. M. Nicholson, and J. J. Greenlee. 2017. Real-Time Quaking-Induced Conversion Detection of Bovine Spongiform Encephalopathy Prions in a Subclinical Steer. Front Vet Sci. 4:242. http://doi.org/10.3389/fvets.2017.00242.

Kondru, N., S. Manne, J. Greenlee, H. West Greenlee, V. Anantharam, P. Halbur, A. Kanthasamy, and A. Kanthasamy. 2017. Integrated Organotypic Slice Cultures and Rt-Quic (Oscar) Assay: Implications for Translational Discovery in Protein Misfolding Diseases. Sci Rep. 7:43155. http://doi.org/10.1038/srep43155.

Mammadova, N., S. Ghaisas, G. Zenitsky, D. S. Sakaguchi, A. G. Kanthasamy, J. J. Greenlee, and M. H. West Greenlee. 2017. Lasting Retinal Injury in a Mouse Model of Blast-Induced Trauma. Am J Pathol. 187(7):1459-72. http://doi.org/10.1016/j.ajpath.2017.03.005.

Moore, S. J., M. H. West Greenlee, N. Kondru, S. Manne, J. D. Smith, R. A. Kunkle, A. Kanthasamy, and J. J. Greenlee. 2017. Experimental Transmission of the Chronic Wasting Disease Agent to Swine after Oral or Intracranial Inoculation. J Virol. 91(1). http://doi.org/10.1128/JVI.00926-17.

Silva, C. J., M. L. Erickson-Beltran, C. Hui, J. J. Badiola, E. M. Nicholson, J. R. Requena, and R. Bolea. 2017. Quantitating Prp Polymorphisms Present in Prions from Heterozygous Scrapie-Infected Sheep. Anal Chem. 89(1): 854-61. http://doi.org/10.1021/acs.analchem.6b03822.

Vrentas, C. E., J. J. Greenlee, G. H. Foster, J. West, M. M. Jahnke, M. T. Schmidt, and E. M. Nicholson. 2017. Effects of a Naturally Occurring Amino Acid Substitution in Bovine Prp: A Model for Inherited Prion Disease in a Natural Host Species. BMC Res Notes. 10(1):759. http://doi.org/10.1186/s13104-017-3085-8.

Hwang, S., J. J. Greenlee, N. M. Vance, and E. M. Nicholson. 2018. Source Genotype Influence on Cross Species Transmission of Transmissible Spongiform Encephalopathies Evaluated by Rt-Quic. PLoS One 13(12):e0209106. http://doi.org/10.1371/journal.pone.0209106.

Hwang, S. and E. M. Nicholson. 2018. Thermodynamic Characterization for the Denatured State of Bovine Prion Protein and the Bse Associated Variant E211k. Prion. 12(5-6):301-309. http://doi.org/10.1080/19336896.2018.1534485.

Hwang, S., T. Tatum, S. Lebepe-Mazur, and E. M. Nicholson. 2018. Preparation of Lyophilized Recombinant Prion Protein for Tse Diagnosis by Rt-Quic. BMC Res Notes. 11(1):895. http://doi.org/10.1186/s13104-018-3982-5.

Moore, S. J., C. E. Vrentas, S. Hwang, M. H. West Greenlee, E. M. Nicholson, and J. J. Greenlee. 2018. Pathologic and Biochemical Characterization of Prp(Sc) from Elk with Prnp Polymorphisms at Codon 132 after Experimental Infection with the Chronic Wasting Disease Agent. BMC Vet Res. 14(1):80. http://doi.org/10.1186/s12917-018-1400-9.

Samorodnitsky, D. and E. M. Nicholson. 2018. Differential Effects of Divalent Cations on Elk Prion Protein Fibril Formation and Stability. Prion. 12(1):63-71. http://doi.org/10.1080/19336896.2017.1423187.

Cassmann, E. D., S. J. Moore, J. D. Smith, and J. J. Greenlee. 2019. Sheep with the Homozygous Lysine-171 Prion Protein Genotype Are Resistant to Classical Scrapie after Experimental Oronasal Inoculation. Vet Pathol. 56(3):409-17. http://doi.org/10.1177/0300985818817066.

Cassmann, E. D., S. J. Moore, J. D. Smith, and J. J. Greenlee. 2019. Sheep Are Susceptible to the Bovine Adapted Transmissible Mink Encephalopathy Agent by Intracranial Inoculation and Have Evidence of Infectivity in Lymphoid Tissues. Front Vet Sci. 6:430. http://doi.org/10.3389/fvets.2019.00430.

Greenlee, J. J. 2019. Review: Update on Classical and Atypical Scrapie in Sheep and Goats. Vet Pathol. 56(1):6-16. http://doi.org/10.1177/0300985818794247.

Hwang, S., R. P. Dassanayake, and E. M. Nicholson. 2019. Pad-Beads Enrichment Enhances Detection of Prp(Sc) Using Real-Time Quaking-Induced Conversion. BMC Res Notes. 12(1):806. http://doi.org/10.1186/s13104-019-4842-7.

Mammadova, N., C. M. Summers, R. D. Kokemuller, Q. He, S. Ding, T. Baron, C. Yu, R. J. Valentine, D. S. Sakaguchi, A. G. Kanthasamy, J. J. Greenlee, and M. Heather West Greenlee. 2019. Accelerated Accumulation of Retinal Alpha-Synuclein (Pser129) and Tau, Neuroinflammation, and Autophagic Dysregulation in a Seeded Mouse Model of Parkinson's Disease. Neurobiol Dis. 121:1-16. http://doi.org/10.1016/j.nbd.2018.09.013.

Moore, S. J., J. D. Smith, J. A. Richt, and J. J. Greenlee. 2019. Raccoons Accumulate Prp(Sc) after Intracranial Inoculation of the Agents of Chronic Wasting Disease or Transmissible Mink Encephalopathy but Not Atypical Scrapie. J Vet Diagn Invest. 31(2):200-209. http://doi.org/10.1177/1040638718825290.

Smith, J. D., T. A. Harm, and J. J. Greenlee. 2019. Pathology in Practice. J Am Vet Med Assoc. 254(7):819-21. http://doi.org/10.2460/javma.254.7.819.

5030-32000-118-00D

Intervention Strategies to Control Endemic and New and Emerging Viral Diseases of Swine; Kay Faaberg (P), Alexandra Buckley, Laura Miller and Kelly Lager

Cheung, A. K., T. F. Ng, K. M. Lager, D. P. Alt, E. Delwart, and R. M. Pogranichniy. 2015. Identification of Several Clades of Novel Single-Stranded Circular DNA Viruses with Conserved Stem-Loop Structures in Pig Feces. Arch Virol. 160(1):353-8. http://doi.org/10.1007/s00705-014-2234-9.

Crawford, K., K. Lager, L. Miller, T. Opriessnig, P. Gerber, and R. Hesse. 2015. Evaluation of Porcine Epidemic Diarrhea Virus Transmission and the Immune Response in Growing Pigs. Vet Res. 46:49. http://doi.org/10.1186/s13567-015-0180-5.

Kappes, M. A. and K. S. Faaberg. 2015. Prrsv Structure, Replication and Recombination: Origin of Phenotype and Genotype Diversity. Virology. 479-480:475-86. http://doi.org/10.1016/j.virol.2015.02.012.

Kappes, M. A., C. L. Miller, and K. S. Faaberg. 2015. Porcine Reproductive and Respiratory Syndrome Virus Nonstructural Protein 2 (Nsp2) Topology and Selective Isoform Integration in Artificial Membranes. Virology. 481:51-62. http://doi.org/10.1016/j.virol.2015.01.028.

Loving, C. L., F. A. Osorio, M. P. Murtaugh, and F. A. Zuckermann. 2015. Innate and Adaptive Immunity against Porcine Reproductive and Respiratory Syndrome Virus. Vet Immunol Immunopathol. 167(1-2):1-14. http://doi.org/10.1016/j.vetimm.2015.07.003.

Miller, L. C., D. O. Bayles, E. L. Zanella, and K. M. Lager. 2015. Effects of Pseudorabies Virus Infection on the Tracheobronchial Lymph Node Transcriptome. Bioinform Biol Insights. 9(Suppl 2):25-36. http://doi.org/10.4137/BBI.S30522.

Renukaradhya, G. J., X. J. Meng, J. G. Calvert, M. Roof, and K. M. Lager. 2015. Live Porcine Reproductive and Respiratory Syndrome Virus Vaccines: Current Status and Future Direction. Vaccine. 33(33):4069-80. http://doi.org/10.1016/j.vaccine.2015.06.092.

Renukaradhya, G. J., X. J. Meng, J. G. Calvert, M. Roof, and K. M. Lager. 2015. Inactivated and Subunit Vaccines against Porcine Reproductive and Respiratory Syndrome: Current Status and Future Direction. Vaccine. 33(27):3065-3072. http://doi.org/10.1016/j.vaccine.2015.04.102.

Sang, Y., L. C. Miller, and F. Blecha. 2015. Macrophage Polarization in Virus-Host Interactions. J Clin Cell Immunol. 6(2):311. http://doi.org/10.4172/2155-9899.1000311.

Spear, A. and K. S. Faaberg. 2015. Development of a Genome Copy Specific Rt-Qpcr Assay for Divergent Strains of Type 2 Porcine Reproductive and Respiratory Syndrome Virus. J Virol Methods. 218:1-6. http://doi.org/10.1016/j.jviromet.2015.02.007.

Crawford, K., K. M. Lager, V. Kulshreshtha, L. C. Miller, and K. S. Faaberg. 2016. Status of Vaccines for Porcine Epidemic Diarrhea Virus in the United States and Canada. Virus Res. 226:108-16. http://doi.org/10.1016/j.virusres.2016.08.005.

Fleming, D. S. and L. C. Miller. 2016. Leading Edge Analysis of Transcriptomic Changes During Pseudorabies Virus Infection. Genom Data. 10:104-06. http://doi.org/10.1016/j.gdata.2016.09.014.

Miller, L.C., Crawford, K., Lager, K.M., Kellner, S.G. and Brockmeier, S.M. 2016. Evaluation of a real-time polymerase chain reaction assay for porcine epidemic diarrhea virus surveillance purposes. J. Vet. Diagn. Invest. 28(1):20-29. 2016. https://doi.org/10.1177%2F1040638715621949.

Schlink, S.N., Lager, K.M., Brockmeier, S., Loving, C.L., Miller, L.C., Vorwald, A.C., Yang, H., Kehrli Jr, M.E., Faaberg, K.S. 2016. Enhancement of innate immunity with granulocyte colony-stimulating factor did not mitigate disease in pigs infected with a highly pathogenic Chinese PRRSV strain. Veterinary Immunology and Immunopathology 179:70-76. 2016. https://doi.org/10.1016/j.vetimm.2016.08.007.

Gerber, P. F., C. T. Xiao, K. Lager, K. Crawford, V. Kulshreshtha, D. Cao, X. J. Meng, and T. Opriessnig. 2016. Increased Frequency of Porcine Epidemic Diarrhea Virus Shedding and Lesions in Suckling Pigs Compared to Nursery Pigs and Protective Immunity in Nursery Pigs after Homologous Re-Challenge. Vet Res. 47(1):118. http://doi.org/10.1186/s13567-016-0402-5.

Langel, S. N., F. C. Paim, K. M. Lager, A. N. Vlasova, and L. J. Saif. 2016. Lactogenic Immunity and Vaccines for Porcine Epidemic Diarrhea Virus (Pedv): Historical and Current Concepts. Virus Res. 226:93-107. http://doi.org/10.1016/j.virusres.2016.05.016.

Miller, L. C., K. K. Crawford, K. M. Lager, S. G. Kellner, and S. L. Brockmeier. 2016. Evaluation of Two Real-Time Polymerase Chain Reaction Assays for Porcine Epidemic Diarrhea Virus (Pedv) to Assess Pedv Transmission in Growing Pigs. J Vet Diagn Invest. 28(1):20-9. http://doi.org/10.1177/1040638715621949.

Montiel, N., A. Buckley, B. Guo, V. Kulshreshtha, A. VanGeelen, H. Hoang, C. Rademacher, K. J. Yoon, and K. Lager. 2016. Vesicular Disease in 9-Week-Old Pigs Experimentally Infected with Senecavirus A. Emerg Infect Dis. 22(7):1246-1248. http://doi.org/10.3201/eid2207.151863.

Niederwerder, M. C., J. C. Nietfeld, J. Bai, L. Peddireddi, B. Breazeale, J. Anderson, M. A. Kerrigan, B. An, R. D. Oberst, K. Crawford, K. M. Lager, D. M. Madson, R. R. Rowland, G. A. Anderson, and R. A. Hesse. 2016. Tissue Localization, Shedding, Virus Carriage, Antibody Response, and Aerosol Transmission of Porcine Epidemic Diarrhea Virus Following Inoculation of 4-Week-Old Feeder Pigs. J Vet Diagn Invest. 28(6):671-78. http://doi.org/10.1177/1040638716663251.

Brockmeier, S. L., C. L. Loving, K. C. Eberle, S. J. Hau, A. Buckley, A. Van Geelen, N. A. Montiel, T. Nicholson, and K. M. Lager. 2017. Interferon Alpha Inhibits Replication of a Live-Attenuated Porcine Reproductive and Respiratory Syndrome Virus Vaccine Preventing Development of an Adaptive Immune Response in Swine. Vet Microbiol. 212:48-51. http://doi.org/10.1016/j.vetmic.2017.11.004.

Liu, Q., L. C. Miller, F. Blecha, and Y. Sang. 2017. Reduction of Infection by Inhibiting Mtor Pathway Is Associated with Reversed Repression of Type I Interferon by Porcine Reproductive and Respiratory Syndrome Virus. J Gen Virol. 98(6):1316-28. http://doi.org/10.1099/jgv.0.000802.

Miller, L. C., D. S. Fleming, X. Li, D. O. Bayles, F. Blecha, and Y. Sang. 2017. Comparative Analysis of Signature Genes in Prrsv-Infected Porcine Monocyte-Derived Cells to Different Stimuli. PLoS One 12(7):e0181256. http://doi.org/10.1371/journal.pone.0181256.

Bester, S. M., C. M. Daczkowski, K. S. Faaberg, and S. D. Pegan. 2018. Insights into the Porcine Reproductive and Respiratory Syndrome Virus Viral Ovarian Tumor Domain Protease Specificity for Ubiquitin and Interferon Stimulated Gene Product 15. ACS Infect Dis. 4(9):1316-26. http://doi.org/10.1021/acsinfecdis.8b00068.

Buckley, A., N. Montiel, B. Guo, V. Kulshreshtha, A. van Geelen, H. Hoang, C. Rademacher, K. J. Yoon, and K. Lager. 2018. Dexamethasone Treatment Did Not Exacerbate Seneca Valley Virus Infection in Nursery-Age Pigs. BMC Vet Res. 14(1):352. http://doi.org/10.1186/s12917-018-1693-8.

Fleming, D. S., S. J. Anderson, and L. C. Miller. 2018. Identification of Coding and Non-Coding RNA Classes Expressed in Swine Whole Blood. J Vis Exp. 141:e58689. http://doi.org/10.3791/58689.

Fleming, D. S. and L. C. Miller. 2018. Identification of Small Non-Coding RNA Classes Expressed in Swine Whole Blood During Hp-Prrsv Infection. Virology 517:56-61. http://dx.doi.org/10.1016/j.virol.2018.01.027.

Shi, P., Y. Su, R. Li, L. Zhang, C. Chen, L. Zhang, K. Faaberg, and J. Huang. 2018. Dual Regulation of Host Traip Post-Translation and Nuclear/Plasma Distribution by Porcine Reproductive and Respiratory

Syndrome Virus Non-Structural Protein 1alpha Promotes Viral Proliferation. Front Immunol. 9:3023. http://doi.org/10.3389/fimmu.2018.03023.

Spear, A., F. X. Wang, M. A. Kappes, P. B. Das, and K. S. Faaberg. 2018. Progress toward an Enhanced Vaccine: Eight Marked Attenuated Viruses to Porcine Reproductive and Respiratory Disease Virus. Virology. 516:30-37. http://doi.org/10.1016/j.virol.2017.12.029.

van Geelen, A. G. M., T. K. Anderson, K. M. Lager, P. B. Das, N. J. Otis, N. A. Montiel, L. C. Miller, V. Kulshreshtha, A. C. Buckley, S. L. Brockmeier, J. Zhang, P. C. Gauger, K. M. Harmon, and K. S. Faaberg. 2018. Porcine Reproductive and Respiratory Disease Virus: Evolution and Recombination Yields Distinct Orf5 Rflp 1-7-4 Viruses with Individual Pathogenicity. Virology. 513:168-79. http://doi.org/10.1016/j.virol.2017.10.002.

Buckley, A., V. Kulshreshtha, A. van Geelen, N. Montiel, B. Guo, K. J. Yoon, and K. Lager. 2019. Experimental Seneca Valley Virus Infection in Market-Weight Gilts. Vet Microbiol. 231:7-10. http://doi.org/10.1016/j.vetmic.2019.02.019.

Deng, X., A. van Geelen, A. C. Buckley, A. O'Brien, A. Pillatzki, K. M. Lager, K. S. Faaberg, and S. C. Baker. 2019. Coronavirus Endoribonuclease Activity in Porcine Epidemic Diarrhea Virus Suppresses Type I and Type Iii Interferon Responses. J Virol. 93(8). http://doi.org/10.1128/JVI.02000-18.

Fleming, D. S. and L. C. Miller. 2019. Differentially Expressed MiRNAs and TRNA Genes Affect Host Homeostasis During Highly Pathogenic Porcine Reproductive and Respiratory Syndrome Virus Infections in Young Pigs. Front Genet. 10:691. http://doi.org/10.3389/fgene.2019.00691.

Lager, K. M. and A. C. Buckley. 2019. Porcine Anti-Viral Immunity: How Important Is It? Front Immunol. 10:2258. http://doi.org/10.3389/fimmu.2019.02258.

Langel, S. N., F. C. Paim, M. A. Alhamo, A. Buckley, A. Van Geelen, K. M. Lager, A. N. Vlasova, and L. J. Saif. 2019. Stage of Gestation at Porcine Epidemic Diarrhea Virus Infection of Pregnant Swine Impacts Maternal Immunity and Lactogenic Immune Protection of Neonatal Suckling Piglets. Front Immunol. 10:727. http://doi.org/10.3389/fimmu.2019.00727.

Langel, S. N., F. C. Paim, M. A. Alhamo, K. M. Lager, A. N. Vlasova, and L. J. Saif. 2019. Oral Vitamin A Supplementation of Porcine Epidemic Diarrhea Virus Infected Gilts Enhances Iga and Lactogenic Immune Protection of Nursing Piglets. Vet Res. 50(1):101. http://doi.org/10.1186/s13567-019-0719-y.

Shields, L. E., J. Jennings, Q. Liu, J. Lee, W. Ma, F. Blecha, L. C. Miller, and Y. Sang. 2019. Cross-Species Genome-Wide Analysis Reveals Molecular and Functional Diversity of the Unconventional Interferon-Omega Subtype. Front Immunol. 10:1431. http://doi.org/10.3389/fimmu.2019.01431.

5030-32000-119-00D
Non-Antibiotic Strategies to Control Priority Bacterial Infections in Swine; Susan Brockmeier (P), Kelly Lager and Tracy Nicholson

Hau, S. J., D. O. Bayles, D. P. Alt, S. L. Brockmeier, T. S. Frana, and T. L. Nicholson. 2015. Draft Genome Sequences of Nine Streptococcus Suis Strains Isolated in the United States. Genome Announc. 3(6). http://doi.org/10.1128/genomeA.01301-15.

Hau, S. J., J. Sun, P. R. Davies, T. S. Frana, and T. L. Nicholson. 2015. Comparative Prevalence of Immune Evasion Complex Genes Associated with Beta-Hemolysin Converting Bacteriophages in Mrsa St5 Isolates from Swine, Swine Facilities, Humans with Swine Contact, and Humans with No Swine Contact. PLoS One. 10(11):e0142832. http://doi.org/10.1371/journal.pone.0142832.

Ivanov, Y. V., N. Shariat, K. B. Register, B. Linz, I. Rivera, K. Hu, E. G. Dudley, and E. T. Harvill. 2015. A Newly Discovered Bordetella Species Carries a Transcriptionally Active Crispr-Cas with a Small Cas9 Endonuclease. *BMC* Genomics. 16:863. http://doi.org/10.1186/s12864-015-2028-9.

Register, K. B., Y. V. Ivanov, E. T. Harvill, L. Brinkac, M. Kim, and L. Losada. 2015. Draft Genome Sequences of Six Bordetella Hinzii Isolates Acquired from Avian and Mammalian Hosts. Genome Announc. 3(2). http://doi.org/10.1128/genomeA.00081-15.

Register, K. B., Y. V. Ivanov, E. T. Harvill, N. Davison, and G. Foster. 2015. Novel, Host-Restricted Genotypes of Bordetella Bronchiseptica Associated with Phocine Respiratory Tract Isolates. Microbiology. 161(Pt 3):580-592. http://doi.org/10.1099/mic.0.000035.

Register, K. B., Y. V. Ivanov, N. Jacobs, J. A. Meyer, L. L. Goodfield, S. J. Muse, W. E. Smallridge, L. Brinkac, M. Kim, R. Sanka, E. T. Harvill, and L. Losada. 2015. Draft Genome Sequences of 53 Genetically Distinct Isolates of Bordetella Bronchiseptica Representing 11 Terrestrial and Aquatic Hosts. Genome Announc. 3(2). http://doi.org/10.1128/genomeA.00152-15.

Trainor, E. A., T. L. Nicholson, and T. J. Merkel. 2015. Bordetella Pertussis Transmission. Pathog Dis. 73(8):ftv068. http://doi.org/10.1093/femspd/ftv068.

Allen, H. K., D. O. Bayles, T. Looft, J. Trachsel, B. E. Bass, D. P. Alt, S. M. Bearson, T. Nicholson, and T. A. Casey. 2016. Pipeline for Amplifying and Analyzing Amplicons of the V1-V3 Region of the 16s rRNA Gene. BMC Res Notes. 9:380. http://doi.org/10.1186/s13104-016-2172-6.

Ivanov, Y. V., B. Linz, K. B. Register, J. D. Newman, D. L. Taylor, K. R. Boschert, S. Le Guyon, E. F. Wilson, L. M. Brinkac, R. Sanka, S. C. Greco, P. M. Klender, L. Losada, and E. T. Harvill. 2016. Identification and Taxonomic Characterization of Bordetella Pseudohinzii Sp. Nov. Isolated from Laboratory-Raised Mice. Int J Syst Evol Microbiol. 66(12):5452-59. http://doi.org/10.1099/ijsem.0.001540.

Linz, B., Y. V. Ivanov, A. Preston, L. Brinkac, J. Parkhill, M. Kim, S. R. Harris, L. L. Goodfield, N. K. Fry, A. R. Gorringe, T. L. Nicholson, K. B. Register, L. Losada, and E. T. Harvill. 2016. Acquisition and Loss of Virulence-Associated Factors During Genome Evolution and Speciation in Three Clades of Bordetella Species. BMC Genomics. 17(1):767. http://doi.org/10.1186/s12864-016-3112-5.

McCaig, W. D., C. L. Loving, H. R. Hughes, and S. L. Brockmeier. 2016. Characterization and Vaccine Potential of Outer Membrane Vesicles Produced by Haemophilus Parasuis. PLoS One. 11(3):e0149132. http://doi.org/10.1371/journal.pone.0149132.

Nicholson, T. L., S. M. Shore, K. B. Register, D. O. Bayles, R. A. Kingsley, and B. W. Brunelle. 2016. Comparative Genomic Analysis of the Swine Pathogen Bordetella Bronchiseptica strain Km22. Vet Microbiol. 182:87-94. http://doi.org/10.1016/j.vetmic.2015.10.026.

Register, K. B., T. L. Nicholson, and B. W. Brunelle. 2016. Comparison of Ribotyping and Sequence-Based Typing for Discriminating among Isolates of Bordetella Bronchiseptica. J Microbiol Methods. 129:117-26. http://doi.org/10.1016/j.mimet.2016.08.008.

Schlink, S. N., K. M. Lager, S. L. Brockmeier, C. L. Loving, L. C. Miller, A. C. Vorwald, H. C. Yang, M. E. Kehrli, Jr., and K. S. Faaberg. 2016. Enhancement of Innate Immunity with Granulocyte Colony-Stimulating Factor Did Not Mitigate Disease in Pigs Infected with a Highly Pathogenic Chinese PRRSV Strain. Vet Immunol Immunopathol. 179:70-6. http://doi.org/10.1016/j.vetimm.2016.08.007.

Taha-Abdelaziz, K., L. L. Bassel, M. L. Harness, M. E. Clark, K. B. Register, and J. L. Caswell. 2016. Cilia-Associated Bacteria in Fatal Bordetella Bronchiseptica Pneumonia of Dogs and Cats. J Vet Diagn Invest. 28(4):369-76. http://doi.org/10.1177/1040638716646806.

Brockmeier, S. L., C. L. Loving, K. C. Eberle, S. J. Hau, A. Buckley, A. Van Geelen, N. A. Montiel, T. Nicholson, and K. M. Lager. 2017. Interferon Alpha Inhibits Replication of a Live-Attenuated Porcine Reproductive and Respiratory Syndrome Virus Vaccine Preventing Development of an Adaptive Immune Response in Swine. Vet Microbiol. 212:48-51. http://doi.org/10.1016/j.vetmic.2017.11.004.

Brockmeier, S. L., C. L. Loving, M. V. Palmer, A. Spear, T. L. Nicholson, K. S. Faaberg, and K. M. Lager. 2017. Comparison of Asian Porcine High Fever Disease Isolates of Porcine Reproductive and Respiratory Syndrome Virus to United States Isolates for Their Ability to Cause Disease and Secondary Bacterial Infection in Swine. Vet Microbiol. 203:6-17. http://doi.org/10.1016/j.vetmic.2017.02.003.

Hau, S. J., D. O. Bayles, D. P. Alt, P. R. Davies, J. S. Haan, and T. L. Nicholson. 2017. Draft Genome Sequences of Nine Livestock-Associated Methicillin-Resistant Staphylococcus Aureus Sequence Type 5 Isolates from Humans with Long-Term Swine Contact. Genome Announc. 5(41). http://doi.org/10.1128/genomeA.01079-17.

Hau, S. J., D. O. Bayles, D. P. Alt, T. S. Frana, and T. L. Nicholson. 2017. Draft Genome Sequences of 14 Livestock-Associated Methicillin-Resistant Staphylococcus Aureus Sequence Type 398 Isolates from Swine Farms in the United States. Genome Announc. 5(44). http://doi.org/10.1128/genomeA.01082-17.

Hau, S. J., D. O. Bayles, D. P. Alt, T. S. Frana, and T. L. Nicholson. 2017. Draft Genome Sequences of 63 Swine-Associated Methicillin-Resistant Staphylococcus Aureus Sequence Type 5 Isolates from the United States. Genome Announc 5(44). http://doi.org/10.1128/genomeA.01081-17.

Hau, S. J., D. O. Bayles, D. P. Alt, T. S. Frana, and T. L. Nicholson. 2017. Complete Genome Sequence of a Livestock-Associated Methicillin-Resistant Staphylococcus Aureus Sequence Type 5 Isolate from the United States. Genome Announc. 5(32). http://doi.org/10.1128/genomeA.00791-17.

Hau, S. J., D. O. Bayles, D. P. Alt, T. S. Frana, and T. L. Nicholson. 2017. Complete Genome Sequence of Livestock-Associated Methicillin-Resistant Staphylococcus Aureus Sequence Type 398 Isolated from Swine in the United States. Genome Announc. 5(32). http://doi.org/10.1128/genomeA.00790-17.

Hau, S. J., D. O. Bayles, D. P. Alt, and T. L. Nicholson. 2017. Draft Genome Sequences of 50 Methicillin-Resistant Staphylococcus Aureus Sequence Type 5 Isolates Obtained from a U.S. Hospital. Genome Announc 5(44). http://doi.org/10.1128/genomeA.01083-17.

Hau, S. J., D. O. Bayles, D. P. Alt, and T. L. Nicholson. 2017. Draft Genome Sequences of One Methicillin-Sensitive and Seven Methicillin-Resistant Staphylococcus Aureus Sequence Type 5 Isolates Obtained in California. Genome Announc. 5(42). http://doi.org/10.1128/genomeA.01084-17.

Hau, S. J., D. O. Bayles, D. P. Alt, and T. L. Nicholson. 2017. Complete Genome Sequences of Two Staphylococcus Aureus Sequence Type 5 Isolates from California, USA. Genome Announc. 5(13). http://doi.org/10.1128/genomeA.00099-17.

Hau, S. J., D. O. Bayles, D. P. Alt, and T. L. Nicholson. 2017. Draft Genome Sequences of 14 Staphylococcus Aureus Sequence Type 5 Isolates from California, USA. Genome Announc. 5(13). http://doi.org/10.1128/genomeA.00098-17.

Hau, S. J., T. Frana, J. Sun, P. R. Davies, and T. L. Nicholson. 2017. Zinc Resistance within Swine-Associated Methicillin-Resistant Staphylococcus Aureus Isolates in the United States Is Associated with Multilocus Sequence Type Lineage. Appl Environ Microbiol. 83(15). http://doi.org/10.1128/AEM.00756-17.

Kumar, S., B. E. Bass, M. Bandrick, C. L. Loving, S. L. Brockmeier, T. Looft, J. Trachsel, D. M. Madson, M. Thomas, T. A. Casey, J. W. Frank, T. B. Stanton, and H. K. Allen. 2017. Fermentation Products as Feed Additives Mitigate Some Ill-Effects of Heat Stress in Pigs. J Anim Sci. 95(1):279-90. http://doi.org/10.2527/jas.2016.0662.

Nicholson, T. L., S. L. Brockmeier, N. Sukumar, A. E. Paharik, J. L. Lister, A. R. Horswill, M. E. Kehrli, Jr., C. L. Loving, S. M. Shore, and R. Deora. 2017. The Bordetella Bps Polysaccharide Is Required for Biofilm Formation and Enhances Survival in the Lower Respiratory Tract of Swine. Infect Immun. 85(8). http://doi.org/10.1128/IAI.00261-17.

Brockmeier, S. L., C. L. Loving, T. L. Nicholson, J. Wang, S. E. Peters, L. Weinert, R. Chaudhuri, D. J. Seilly, P. R. Langford, A. Rycroft, B. W. Wren, D. J. Maskell, and A. W. Tucker. 2018. Use of Proteins Identified through a Functional Genomic Screen to Develop a Protein Subunit Vaccine That Provides Significant Protection against Virulent Streptococcus Suis in Pigs. Infect Immun. 86(3). http://doi.org/10.1128/IAI.00559-17.

Gestal, M. C., I. Rivera, L. K. Howard, K. K. Dewan, I. H. Soumana, M. Dedloff, T. L. Nicholson, B. Linz, and E. T. Harvill. 2018. Blood or Serum Exposure Induce Global Transcriptional Changes, Altered Antigenic Profile, and Increased Cytotoxicity by Classical Bordetellae. Front Microbiol 9:1969. http://doi.org/10.3389/fmicb.2018.01969.

Hau, S. J., A. Allue-Guardia, B. Rusconi, J. S. Haan, P. R. Davies, T. S. Frana, M. Eppinger, and T. L. Nicholson. 2018. Single Nucleotide Polymorphism Analysis Indicates Genetic Distinction and Reduced Diversity of Swine-Associated Methicillin Resistant Staphylococcus Aureus (Mrsa) St5 Isolates Compared to Clinical Mrsa St5 Isolates. Front Microbiol 9:2078. http://doi.org/10.3389/fmicb.2018.02078.

Hau, S. J., J. S. Haan, P. R. Davies, T. Frana, and T. L. Nicholson. 2018. Antimicrobial Resistance Distribution Differs among Methicillin Resistant Staphylococcus Aureus Sequence Type (St) 5 Isolates from Health Care and Agricultural Sources. Front Microbiol. 9:2102. http://doi.org/10.3389/fmicb.2018.02102.

Hau, S. J., S. Kellner, K. C. Eberle, U. Waack, S. L. Brockmeier, J. S. Haan, P. R. Davies, T. Frana, and T. L. Nicholson. 2018. Methicillin-Resistant Staphylococcus Aureus Sequence Type (St) 5 Isolates from Health Care and Agricultural Sources Adhere Equivalently to Human Keratinocytes. Appl Environ Microbiol. 84(2). http://doi.org/10.1128/AEM.02073-17.

Hughes, H. R., S. L. Brockmeier, and C. L. Loving. 2018. Bordetella Bronchiseptica Colonization Limits Efficacy, but Not Immunogenicity, of Live-Attenuated Influenza Virus Vaccine and Enhances Pathogenesis after Influenza Challenge. Front Immunol. 9:2255. http://doi.org/10.3389/fimmu.2018.02255.

Nicholson, T. L., B. W. Brunelle, D. O. Bayles, D. P. Alt, and S. M. Shore. 2018. Comparative Genomic and Methylome Analysis of Non-Virulent D74 and Virulent Nagasaki Haemophilus Parasuis Isolates. PLoS One. 13(11):e0205700. http://doi.org/10.1371/journal.pone.0205700.

Waack, U. and T. L. Nicholson. 2018. Subinhibitory Concentrations of Amoxicillin, Lincomycin, and Oxytetracycline Commonly Used to Treat Swine Increase Streptococcus Suis Biofilm Formation. Front Microbiol. 9:2707. http://doi.org/10.3389/fmicb.2018.02707.

Brockmeier, S. L., C. L. Loving, K. C. Eberle, S. J. Hau, K. T. Mou, and M. E. Kehrli, Jr. 2019. Administration of Granulocyte-Colony Stimulating Factor (G-Csf) to Pigs Results in a Longer Mean Survival Time after Exposure to Streptococcus Suis. Vet Microbiol. 231:116-19. http://doi.org/10.1016/j.vetmic.2019.03.010.

Faulds-Pain, A., H. A. Shaw, V. S. Terra, S. Kellner, S. L. Brockmeier, and B. W. Wren. 2019. The Streptococcus Suis Sortases Srtb and Srtf Are Essential for Disease in Pigs. Microbiology. 165(2):163-73. http://doi.org/10.1099/mic.0.000752.

Hau, S. J., K. T. Mou, D. O. Bayles, and S. L. Brockmeier. 2019. Transcriptomic Differences Noted in Glaesserella Parasuis between Growth in Broth and on Agar. PLoS One. 14(8):e0220365. http://doi.org/10.1371/journal.pone.0220365.

Mou, K. T., H. K. Allen, D. P. Alt, J. Trachsel, S. J. Hau, J. F. Coetzee, D. B. Holman, S. Kellner, C. L. Loving, and S. L. Brockmeier. 2019. Shifts in the Nasal Microbiota of Swine in Response to Different Dosing Regimens of Oxytetracycline Administration. Vet Microbiol. 237:108386. http://doi.org/10.1016/j.vetmic.2019.108386.

Stuart, K. L., S. M. Shore, and T. L. Nicholson. 2019. Complete Genome Sequence of Escherichia Coli Antibiotic-Resistant Isolate Ar Bank #0349. Microbiol Resour Announc. 8(48). http://doi.org/10.1128/MRA.01078-19.

5030-32000-120-00D
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Anderson, T. K., B. A. Campbell, M. I. Nelson, N. S. Lewis, A. Janas-Martindale, M. L. Killian, and A. L. Vincent. 2015. Characterization of Co-Circulating Swine Influenza A Viruses in North America and the Identification of a Novel H1 Genetic Clade with Antigenic Significance. Virus Res. 201:24-31. http://doi.org/10.1016/j.virusres.2015.02.009.

Henningson, J. N., D. S. Rajao, P. Kitikoon, A. Lorusso, M. R. Culhane, N. S. Lewis, T. K. Anderson, and A. L. Vincent. 2015. Comparative Virulence of Wild-Type H1N1pdm09 Influenza A Isolates in Swine. Vet Microbiol. 176(1-2):40-9. http://doi.org/10.1016/j.vetmic.2014.12.021.

Hughes, H. R., A. L. Vincent, S. L. Brockmeier, P. C. Gauger, L. Pena, J. Santos, D. R. Braucher, D. R. Perez, and C. L. Loving. 2015. Oral Fluids as a Live-Animal Sample Source for Evaluating Cross-

Reactivity and Cross-Protection Following Intranasal Influenza A Virus Vaccination in Pigs. Clin Vaccine Immunol. 22(10):1109-20. http://doi.org/10.1128/CVI.00358-15.

Nelson, M. I., J. Stratton, M. L. Killian, A. Janas-Martindale, and A. L. Vincent. 2015. Continual Reintroduction of Human Pandemic H1N1 Influenza A Viruses into Swine in the United States, 2009 to 2014. J Virol. 89(12):6218-6226. http://doi.org/10.1128/JVI.00459-15.

Nelson, M. I., C. Viboud, A. L. Vincent, M. R. Culhane, S. E. Detmer, D. E. Wentworth, A. Rambaut, M. A. Suchard, E. C. Holmes, and P. Lemey. 2015. Global Migration of Influenza A Viruses in Swine. Nat Commun. 6:6696. http://doi.org/10.1038/ncomms7696.

Nelson, M. I. and A. L. Vincent. 2015. Reverse Zoonosis of Influenza to Swine: New Perspectives on the Human-Animal Interface. Trends Microbiol. 23(3):142-153. http://doi.org/10.1016/j.tim.2014.12.002.

Obadan, A. O., B. J. Kimble, D. Rajao, K. Lager, J. J. Santos, A. Vincent, and D. R. Perez. 2015. Replication and Transmission of Mammalian-Adapted H9 Subtype Influenza Virus in Pigs and Quail. J Gen Virol. 96(9):2511-2521. http://doi.org/10.1099/vir.0.000190.

Rajao, D. S., P. C. Gauger, T. K. Anderson, N. S. Lewis, E. J. Abente, M. L. Killian, D. R. Perez, T. C. Sutton, J. Zhang, and A. L. Vincent. 2015. Novel Reassortant Human-Like H3N2 and H3n1 Influenza A Viruses Detected in Pigs Are Virulent and Antigenically Distinct from Swine Viruses Endemic to the United States. J Virol. 89(22):11213-22. http://doi.org/10.1128/JVI.01675-15.

Rajao, D. S. and A. L. Vincent. 2015. Swine as a Model for Influenza A Virus Infection and Immunity. ILAR J. 56(1):44-52. http://doi.org/10.1093/ilar/ilv002.

Thebault, S., P. Waters, M. D. Snape, D. Cottrell, N. Darin, T. Hallbook, A. Huutoniemi, M. Partinen, A. J. Pollard, and A. Vincent. 2015. Neuronal Antibodies in Children with or without Narcolepsy Following H1N1-As03 Vaccination. PLoS One. 10(6):e0129555. http://doi.org/10.1371/journal.pone.0129555.

Abente, E. J., T. K. Anderson, D. S. Rajao, S. Swenson, P. C. Gauger, and A. L. Vincent. 2015. The Avian-Origin H3N2 Canine Influenza Virus That Recently Emerged in the United States Has Limited Replication in Swine. Influenza Other Respir Viruses. 10(5):429-32. http://doi.org/10.1111/irv.12395.

Abente, E. J., J. Santos, N. S. Lewis, P. C. Gauger, J. Stratton, E. Skepner, T. K. Anderson, D. S. Rajao, D. R. Perez, and A. L. Vincent. 2015. The Molecular Determinants of Antibody Recognition and Antigenic Drift in the H3 Hemagglutinin of Swine Influenza A Virus. J Virol. 90(18):8266-8280. http://doi.org/10.1128/JVI.01002-16.

Anderson, T. K., C. A. Macken, N. S. Lewis, R. H. Scheuermann, K. Van Reeth, I. H. Brown, S. L. Swenson, G. Simon, T. Saito, Y. Berhane, J. Ciacci-Zanella, A. Pereda, C. T. Davis, R. O. Donis, R. J. Webby, and A. L. Vincent. 2015. A Phylogeny-Based Global Nomenclature System and Automated Annotation Tool for H1 Hemagglutinin Genes from Swine Influenza A Viruses. mSphere. 1(6). http://doi.org/10.1128/mSphere.00275-16.

Balzli, C., K. Lager, A. Vincent, P. Gauger, S. Brockmeier, L. Miller, J. A. Richt, W. Ma, D. Suarez, and D. E. Swayne. 2016. Susceptibility of Swine to H5 and H7 Low Pathogenic Avian Influenza Viruses. Influenza Other Respir Viruses. 10(4):346-52. http://doi.org/10.1111/irv.12386.

Goodell, C. K., J. Zhang, E. Strait, K. Harmon, D. Patnayak, T. Otterson, M. Culhane, J. Christopher-Hennings, T. Clement, P. Leslie-Steen, R. Hesse, J. Anderson, K. Skarbek, A. Vincent, P. Kitikoon, S.

Swenson, M. Jenkins-Moore, J. McGill, R. Rauh, W. Nelson, C. O'Connell, R. Shah, C. Wang, R. Main, and J. J. Zimmerman. 2016. Ring Test Evaluation of the Detection of Influenza A Virus in Swine Oral Fluids by Real-Time Reverse-Transcription Polymerase Chain Reaction and Virus Isolation. Can J Vet Res. 80(1):12-20. https://www.ncbi.nlm.nih.gov/pubmed/26733728.

Gutierrez, A. H., C. Loving, L. Moise, F. E. Terry, S. L. Brockmeier, H. R. Hughes, W. D. Martin, and A. S. De Groot. 2016. In Vivo Validation of Predicted and Conserved T Cell Epitopes in a Swine Influenza Model. PLoS One. 11(7):e0159237. http://doi.org/10.1371/journal.pone.0159237.

Lewis, N. S., C. A. Russell, P. Langat, T. K. Anderson, K. Berger, F. Bielejec, D. F. Burke, G. Dudas, J. M. Fonville, R. A. Fouchier, P. Kellam, B. F. Koel, P. Lemey, T. Nguyen, B. Nuansrichy, J. M. Peiris, T. Saito, G. Simon, E. Skepner, N. Takemae, Esnip consortium, R. J. Webby, K. Van Reeth, S. M. Brookes, L. Larsen, S. J. Watson, I. H. Brown, and A. L. Vincent. 2016. The Global Antigenic Diversity of Swine Influenza A Viruses. Elife. 5:e12217. http://doi.org/10.7554/eLife.12217.

Rajao, D. S., H. Chen, D. R. Perez, M. R. Sandbulte, P. C. Gauger, C. L. Loving, G. D. Shanks, and A. Vincent. 2016. Vaccine-Associated Enhanced Respiratory Disease Is Influenced by Haemagglutinin and Neuraminidase in Whole Inactivated Influenza Virus Vaccines. J Gen Virol. 97(7):1489-1499. http://doi.org/10.1099/jgv.0.000468.

Rajao, D. S., M. R. Sandbulte, P. C. Gauger, P. Kitikoon, R. Platt, J. A. Roth, D. R. Perez, C. L. Loving, and A. L. Vincent. 2016. Heterologous Challenge in the Presence of Maternally-Derived Antibodies Results in Vaccine-Associated Enhanced Respiratory Disease in Weaned Piglets. Virology. 491:79-88. http://doi.org/10.1016/j.virol.2016.01.015.

Sandbulte, M. R., P. C. Gauger, P. Kitikoon, H. Chen, D. R. Perez, J. A. Roth, and A. L. Vincent. 2016. Neuraminidase Inhibiting Antibody Responses in Pigs Differ between Influenza A Virus N2 Lineages and by Vaccine Type. Vaccine. 34(33):3773-9. http://doi.org/10.1016/j.vaccine.2016.06.001.

Shriner, S. A., J. J. Root, M. W. Lutman, J. M. Kloft, K. K. VanDalen, H. J. Sullivan, T. S. White, M. P. Milleson, J. L. Hairston, S. C. Chandler, P. C. Wolf, C. T. Turnage, B. J. McCluskey, A. L. Vincent, M. K. Torchetti, T. Gidlewski, and T. J. DeLiberto. 2016. Surveillance for Highly Pathogenic H5 Avian Influenza Virus in Synanthropic Wildlife Associated with Poultry Farms During an Acute Outbreak. Sci Rep. 6:36237. http://doi.org/10.1038/srep36237.

Souza, C. K., D. S. Rajao, C. L. Loving, P. C. Gauger, D. R. Perez, and A. L. Vincent. 2016. Age at Vaccination and Timing of Infection Do Not Alter Vaccine-Associated Enhanced Respiratory Disease in Influenza A Virus-Infected Pigs. Clin Vaccine Immunol. 23(6):470-82. http://doi.org/10.1128/CVI.00563-15.

Abente, E. J., P. C. Gauger, R. R. Walia, D. S. Rajao, J. Zhang, K. M. Harmon, M. L. Killian, and A. L. Vincent. 2017. Detection and Characterization of an H4N6 Avian-Lineage Influenza A Virus in Pigs in the Midwestern United States. Virology. 511:56-65. http://doi.org/10.1016/j.virol.2017.08.021.

Abente, E. J., P. Kitikoon, K. M. Lager, P. C. Gauger, T. K. Anderson, and A. L. Vincent. 2017. A Highly Pathogenic Avian-Derived Influenza Virus H5N1 with 2009 Pandemic H1N1 Internal Genes Demonstrates Increased Replication and Transmission in Pigs. J Gen Virol. 98(1):18-30. http://doi.org/10.1099/jgv.0.000678.

Bowman, A. S., R. R. Walia, J. M. Nolting, A. L. Vincent, M. L. Killian, M. M. Zentkovich, J. N. Lorbach, S. E. Lauterbach, T. K. Anderson, C. T. Davis, N. Zanders, J. Jones, Y. Jang, B. Lynch, M. R.

Rodriguez, L. Blanton, S. E. Lindstrom, D. E. Wentworth, J. Schiltz, J. J. Averill, and T. Forshey. 2017. Influenza A(H3N2) Virus in Swine at Agricultural Fairs and Transmission to Humans, Michigan and Ohio, USA, 2016. Emerg Infect Dis. 23(9):1551-55. http://doi.org/10.3201/eid2309.170847.

Gao, S., T. K. Anderson, R. R. Walia, K. S. Dorman, A. Janas-Martindale, and A. L. Vincent. 2017. The Genomic Evolution of H1 Influenza A Viruses from Swine Detected in the United States between 2009 and 2016. J Gen Virol. 98(8):2001-10. http://doi.org/10.1099/jgv.0.000885.

Gutierrez, A. H., V. J. Rapp-Gabrielson, F. E. Terry, C. L. Loving, L. Moise, W. D. Martin, and A. S. De Groot. 2017. T-Cell Epitope Content Comparison (Epicc) of Swine H1 Influenza A Virus Hemagglutinin. Influenza Other Respir Viruses. 11(6):531-542. http://doi.org/10.1111/irv.12513.

Kaplan, B. S., M. K. Torchetti, K. M. Lager, R. J. Webby, and A. L. Vincent. 2017. Absence of Clinical Disease and Contact Transmission of HPAI H5Nx Clade 2.3.4.4 from North America in Experimentally Infected Pigs. Influenza Other Respir Viruses. 11(5):464-70. http://doi.org/10.1111/irv.12463.

Martin, B. E., H. Sun, M. Carrel, F. L. Cunningham, J. A. Baroch, K. C. Hanson-Dorr, S. G. Young, B. Schmit, J. M. Nolting, K. J. Yoon, M. W. Lutman, K. Pedersen, K. Lager, A. S. Bowman, R. D. Slemons, D. R. Smith, T. DeLiberto, and X. F. Wan. 2017. Feral Swine in the United States Have Been Exposed to Both Avian and Swine Influenza A Viruses. Appl Environ Microbiol. 83(19). http://doi.org/10.1128/AEM.01346-17.

Olson, Z. F., M. R. Sandbulte, C. K. Souza, D. R. Perez, A. L. Vincent, and C. L. Loving. 2017. Factors Affecting Induction of Peripheral IFN-Gamma Recall Response to Influenza A Virus Vaccination in Pigs. Vet Immunol Immunopathol 185 (Mar 2017): 57-65. http://doi.org/10.1016/j.vetimm.2017.01.009.

Rajao, D. S., C. L. Loving, E. H. Waide, P. C. Gauger, J. C. Dekkers, C. K. Tuggle, and A. L. Vincent. 2017. Pigs with Severe Combined Immunodeficiency Are Impaired in Controlling Influenza A Virus Infection. J Innate Immun. 9(2):193-202. http://doi.org/10.1159/000451007.

Rajao, D. S., R. R. Walia, B. Campbell, P. C. Gauger, A. Janas-Martindale, M. L. Killian, and A. L. Vincent. 2017. Reassortment between Swine H3N2 and 2009 Pandemic H1N1 in the United States Resulted in Influenza A Viruses with Diverse Genetic Constellations with Variable Virulence in Pigs. J Virol. 91(4). http://doi.org/10.1128/JVI.01763-16.

Vincent, A. L., D. R. Perez, D. Rajao, T. K. Anderson, E. J. Abente, R. R. Walia, and N. S. Lewis. 2017. Influenza A Virus Vaccines for Swine. Vet Microbiol. 206: 35-44. http://doi.org/10.1016/j.vetmic.2016.11.026.

Zhang, Y., B. D. Aevermann, T. K. Anderson, D. F. Burke, G. Dauphin, Z. Gu, S. He, S. Kumar, C. N. Larsen, A. J. Lee, X. Li, C. Macken, C. Mahaffey, B. E. Pickett, B. Reardon, T. Smith, L. Stewart, C. Suloway, G. Sun, L. Tong, A. L. Vincent, B. Walters, S. Zaremba, H. Zhao, L. Zhou, C. Zmasek, E. B. Klem, and R. H. Scheuermann. 2017. Influenza Research Database: An Integrated Bioinformatics Resource for Influenza Virus Research. Nucleic Acids Res. 45(D1):D466-D74. http:/doi.org/10.1093/nar/gkw857.

Abente, E. J., D. S. Rajao, J. Santos, B. S. Kaplan, T. L. Nicholson, S. L. Brockmeier, P. C. Gauger, D. R. Perez, and A. L. Vincent. 2018. Comparison of Adjuvanted-Whole Inactivated Virus and Live-Attenuated Virus Vaccines against Challenge with Contemporary, Antigenically Distinct H3N2 Influenza A Viruses. J Virol. 92(22). http://doi.org/10.1128/JVI.01323-18.

Duwell, M. M., D. Blythe, M. W. Radebaugh, E. M. Kough, B. Bachaus, D. A. Crum, K. A. Perkins, Jr., L. Blanton, C. T. Davis, Y. Jang, A. Vincent, J. Chang, D. E. Abney, L. Gudmundson, M. G. Brewster, L. Polsky, D. C. Rose, and K. A. Feldman. 2018. Influenza A(H3N2) Variant Virus Outbreak at Three Fairs - Maryland, 2017. MMWR Morb Mortal Wkly Rep. 67(42):1169-1173. http://doi.org/10.15585/mmwr.mm6742a1.

Kaplan, B. S., C. K. Souza, P. C. Gauger, C. B. Stauft, J. Robert Coleman, S. Mueller, and A. L. Vincent. 2018. Vaccination of Pigs with a Codon-Pair Bias De-Optimized Live Attenuated Influenza Vaccine Protects from Homologous Challenge. Vaccine. 36(8):1101-1107. http://doi.org/10.1016/j.vaccine.2018.01.027.

Rajao, D. S., T. K. Anderson, P. Kitikoon, J. Stratton, N. S. Lewis, and A. L. Vincent. 2018. Antigenic and Genetic Evolution of Contemporary Swine H1 Influenza Viruses in the United States. Virology. 518:45-54. http://doi.org/10.1016/j.virol.2018.02.006.

Rajao, D. S., A. L. Vincent, and D. R. Perez. 2018. Adaptation of Human Influenza Viruses to Swine. Front Vet Sci. 5:347. http://doi.org/10.3389/fvets.2018.00347.

Souza, C. K., D. S. Rajao, M. R. Sandbulte, S. Lopes, N. S. Lewis, C. L. Loving, P. C. Gauger, and A. L. Vincent. 2018. The Type of Adjuvant in Whole Inactivated Influenza A Virus Vaccines Impacts Vaccine-Associated Enhanced Respiratory Disease. Vaccine. 36(41):6103-6110. http://doi.org/10.1016/j.vaccine.2018.08.072.

Zeller, M. A., T. K. Anderson, R. W. Walia, A. L. Vincent, and P. C. Gauger. 2018. Isu Fluture: A Veterinary Diagnostic Laboratory Web-Based Platform to Monitor the Temporal Genetic Patterns of Influenza A Virus in Swine. BMC Bioinformatics. 19(1):397. http://doi.org/10.1186/s12859-018-2408-7.

Zeller, M. A., G. Li, K. M. Harmon, J. Zhang, A. L. Vincent, T. K. Anderson, and P. C. Gauger. 2018. Complete Genome Sequences of Two Novel Human-Like H3N2 Influenza A Viruses, a/Swine/Oklahoma/65980/2017 (H3N2) and a/Swine/Oklahoma/65260/2017 (H3N2), Detected in Swine in the United States. Microbiol Resour Announc. 7(20). http://doi.org/10.1128/MRA.01203-18.

Abente, E. J., D. S. Rajao, P. C. Gauger, and A. L. Vincent. 2019. Alphavirus-Vectored Hemagglutinin Subunit Vaccine Provides Partial Protection against Heterologous Challenge in Pigs. Vaccine. 37(11):1533-1539. http://doi.org/10.1016/j.vaccine.2018.12.071.

Bolton, M. J., E. J. Abente, D. Venkatesh, J. A. Stratton, M. Zeller, T. K. Anderson, N. S. Lewis, and A. L. Vincent. 2019. Antigenic Evolution of H3N2 Influenza A Viruses in Swine in the United States from 2012 to 2016. Influenza Other Respir Viruses. 13(1):83-90. http://doi.org/10.1111/irv.12610.

Chang, J., T. K. Anderson, M. A. Zeller, P. C. Gauger, and A. L. Vincent.2019. Octoflu: Automated Classification for the Evolutionary Origin of Influenza A Virus Gene Sequences Detected in U.S. Swine. Microbiol Resour Announc. 8(32). http://doi.org/10.1128/MRA.00673-19.

Nelson, M. I., C. K. Souza, N. S. Trovao, A. Diaz, I. Mena, A. Rovira, A. L. Vincent, M. Torremorell, D. Marthaler, and M. R. Culhane. 2019. Human-Origin Influenza A(H3N2) Reassortant Viruses in Swine, Southeast Mexico. Emerg Infect Dis. 25(4):691-700. http://doi.org/10.3201/eid2504.180779.

Powell, J. D., E. J. Abente, M. K. Torchetti, M. L. Killian, and A. L. Vincent. 2019. An Avian Influenza Virus A (H7N9) Reassortant That Recently Emerged in the United States with Low Pathogenic

Phenotype Does Not Efficiently Infect Swine. Influenza Other Respir Viruses. 13(3):288-291. http://doi.org/10.1111/irv.12631.

Santos, J. J. S., E. J. Abente, A. O. Obadan, A. J. Thompson, L. Ferreri, G. Geiger, A. S. Gonzalez-Reiche, N. S. Lewis, D. F. Burke, D. S. Rajao, J. C. Paulson, A. L. Vincent, and D. R. Perez. 2019. Plasticity of Amino Acid Residue 145 near the Receptor Binding Site of H3 Swine Influenza A Viruses and Its Impact on Receptor Binding and Antibody Recognition. J Virol. 93(2). http://doi.org/10.1128/JVI.01413-18.

Walia, R. R., T. K. Anderson, and A. L. Vincent. 2019. Regional Patterns of Genetic Diversity in Swine Influenza A Viruses in the United States from 2010 to 2016. Influenza Other Respir Viruses. 13(3):262-73. http://doi.org/10.1111/irv.12559.

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Intervention Strategies to Prevent and Control Enteric Diseases of Poultry; Qingzhong Yu (P) and John Dunn

Yu, Q., *Li, Y.,* Dimitrov, K., Afonso, C. A., Spatz, S. and Zsak, L. Genetic stability of a Newcastle disease virus vectored infectious laryngotracheitis virus vaccine after serial passages in chicken embryos. Vaccine. 38:925-932, 2020. https://doi.org/10.1016/j.vaccine.2019.10.074.

Ji, Y., Liu, T., Du, Y., Cui, X., Yu, Q., Wang, Z., Li, Y., Zhu, Q. 2018. A novel genotype VII Newcastle disease virus vaccine candidate generated by mutation in the L and F genes confers improved protection in chickens. Veterinary Microbiology. 216:99-106. https://doi.org/10.1016/j.vetmic.2018.01.021.

Hu, H., Roth, J.P., Yu, Q. 2018. Generation of a recombinant Newcastle disease virus expressing two foreign genes for use as a multivalent vaccine and gene therapy vector. Vaccine. 36:4846-4850. https://doi.org/10.1016/j.vaccine.2018.06.055.

Noh, J., Lee, D., Lim, T., Lee, J., Day, J.M., Song, C. 2018. Isolation and genomic characterization of a novel avian orthoreovirus in Korea, 2014. Archives of Virology. 163:1307-1316. https://doi.org/10.1007/s00705-017-3667-8.

Eldemery, F., Li, Y., Yu, Q., Van Santen, V., Toro, H. 2017. infectious bronchitis virus S2 of 4/91 expressed from recombinant virus does not protect against ark-type challenge. Acarology International Congress Proceedings. 61(3):397-401. https://doi.org/10.1637/11632-032017-ResNoteR.

Wu, Y., He, J., Geng, J., An, Y., Ye, X., Yan, S., Yu, Q., Yin, J., Zhang, Z., Li, D. 2017. Recombinant Newcastle disease virus expressing human TRAIL as a potential candidate for hepatoma therapy. European Journal of Pharmacology. 802:85–92. http://doi:org/10.1016/j.ejphar.2017.02.042.

Ji, Y., Liu, T., Liu, B., Yu, Q., Cui, X., Guo, F., Chang, H., Zhu, Q. 2017. Two single mutations in the fusion protein of Newcastle disease virus confer hemagglutinin-neuraminidase independent fusion promotion and attenuate the pathogenicity in chickens. Virology. 509:146-151. https://doi.org/10.1016/j.virol.2017.06.021.

Hu, H., Roth, J., Zsak, L., Yu, Q. 2017. Engineered Newcastle disease virus expressing the F and G protein of AMPV-C confers protection against challenges in turkeys. Scientific Reports. 7:4025. http://doi:10.1038/s41598-017-04267-7.

Wen, G., Hu, X., Zhao, K., Wang, H., Zhang, Z., Zhang, T., Yang, J., Luo, Q., Zhang, R., Pan, Z., Shao, H., and Yu, Q. Molecular basis for the thermostability of Newcastle disease virus. Scientific Reports. 2016 Mar 3;6:22492. https://doi.org/10.1038/srep22492.

Yu, Q., Spatz, S., Li, Y, Yang, J, Zhao, W., Zhang, Z., Wen, G., Garcia, M., and Zsak, L. Newcastle disease virus vectored infectious laryngotracheitis vaccines protect commercial broiler chickens in the presence of maternally derived antibodies. Vaccine. 35:789-795. 2017. https://doi.org/10.1016/j.vaccine.2016.12.038.

Wen, G., Wang, H., Yu, Q., Zhang, W., and Shao, H. Evaluation of a thermostable Newcastle disease virus strain TS09-C as an in-ovo vaccine for chickens. PLoS One. 2017; 12(2): e0172812. https://doi.org/10.1371/journal.pone.0172812.

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Genetic and Biological Determinants of Avian Herpesviruses Pathogenicity, Transmission, and Evolution to Inform the Development of Effective Control Strategies; Stephen Spatz (P), Taejoong Kim, John Dunn and Hans Cheng **Project managed by Avian Disease and Oncology Research**

Dunn, J.R., Black Pyrkosz, A., Steep, A., Cheng, H.H. 2019. Identification of Marek's disease virus genes associated with virulence of US strains. Journal of General Virology. 100(7):1132-1139. https://doi.org/10.1099/jgv.0.001288.

Spatz, S.J., Garcia, M., Riblet, S., Ross, T.A., Volkening, J.D., Taylor, T.L., Kim, T.N., Afonso, C.L. 2019. MinION sequencing to genotype US strains of Infectious Laryngotracheitis Virus. Avian Pathology. 48(3):255-269. https://doi.org/10.1080/03079457.2019.1579298.

Umthong, S., Dunn, J.R., Cheng, H.H. 2019. Towards a mechanistic understanding of the synergistic response induced by bivalent Marek's disease vaccines to prevent lymphomas. Vaccine. 37(43):6397-6404. https://doi.org/10.1016/j.vaccine.2019.09.003.

Ahmed, H., Mays, J., Kiupel, M., Dunn, J.R. 2018. Development of reliable techniques for the differential diagnosis of avian tumor viruses by immunohistochemistry and polymerase chain reaction from formalin-fixed paraffin-embedded tissue sections. Avian Pathology. 47(4):364-374. https://doi.org/10.1080/03079457.2018.1451620.

Conrad, S.J., Silva, R.F., Hearn, C.J., Climans, M., Dunn, J.R. 2018. Attenuation of Marek's disease virus by codon pair deoptimization of a core gene. Virology. 516:219-226. https://doi.org/10.1016/j.virol.2018.01.020.

Dar, M.A., Urwat, U., Ahmad, S.M., Ahemd, R., Kushoo, Z.A., Dar, T.A., Shah, R.A., Heidari, M. 2018. Gene expression and antibody response in chicken against Salmonella Typhimurium challenge. Poultry Science. 98(5):2008-2013. https://doi.org/10.3382/ps/pey560.

Dunn, J.R., Dimitrov, K.M., Miller, P.J., Garcia, M., Turner-Alston, K., Brown, A., Hartman, A. 2018. Evaluation of protective efficacy when combining HVT vector vaccines. Avian Diseases. 63(1):75-83. https://doi.org/10.1637/11979-092818-Reg.1.

Kim, T.N., Hunt, H.D., Parcells, M.S., Van Santen, V., Ewald, S.J. 2018. Two class I genes of the chicken MHC have different functions: BF1 is recognized by NK cells while BF2 is recognized by CTLs. Immunogenetics. 70(9):599-611. https://doi.org/10.1007/s00251-018-1066-2.

Wang, D., Sun, S., Heidari, M. 2018. Marek's disease vaccine activates chicken macrophages. Journal of Veterinary Science. 19(3):375-383. https://doi.org/10.4142/jvs.2018.19.3.375.

Loncoman, C.A., Hartley, C.A., Coppo, M.C., Vaz, P.K., Diaz-Mendez, A., Browning, G.F., Garcia, M., Spatz, S.J., Devlin, J.M. 2017. Genetic diversity of infectious laryngotracheitis virus during in vivo coinfection parallels viral replication and arises from recombination hot spots within the genome. Applied and Environmental Microbiology. 83(23):e01532-17. https://doi.org/10.1128/AEM.01532-17.

Trimpert, J., Groenke, N., Jenckel, M., He, S., Kunec, D., Szpara, M.L., Spatz, S.J., Osterrieder, N., Mcmahon, D.P. 2017. A phylogenomic analysis of Marek's disease virus reveals independent paths to virulence in Eurasia and North America. Evolutionary Applications. 10(10):1091-1101. https://doi.org/10.1111/eva.12515.

Spatz, S.J., Ross, T.A., Garcia, M., Cheng, Y., Riblet, S., Schneiders, G., Volkening, J. 2016. Attenuation and protection efficacy of open reading frame C (ORF C) gene deleted recombinant of infection was laryngotracheitis virus (ILTV). Journal of General Virology. 97:2352-2362. https://doi.org/10.1099/jgv.0.000521.

Burmakina, G., Malogolovkin, A., Tulman, E., Zsak, L., Delhon, G., Diel, D., Shobogorov, N., Morgunov, Y., Morgunov, S., Kutish, G., Kolbasov, D., Rock, D. 2016. African swine fever virus serotype-specific proteins are significant protective antigens for African swine fever. Journal of General Virology. 97:1670-1675. DOI: 10.1099/jgv.0.000490. https://doi.org/10.1099/jgv.0.000490.

Conrad, S.J., Silva, R.F., Hearn, C.J., Climans, M., Dunn, J.R. 2018. Attenuation of Marek's disease virus by codon pair deoptimization of core herpesvirus genes. Virology. 516:219-226. https://doi.org/10.1016/j.virol.2018.01.020.

Dunn, J.R., Reddy, S., Niikura, M., Nair, V., Fulton, J.E., Cheng, H.H. 2017. Evaluation and identification of Marek's disease virus BAC clones as standardized reagents for research. Avian Diseases. 61(1):107-114. doi: 10.1637/0005-2086-61.1.107. https://doi.org/10.1637/0005-2086-61.1.107.

Heidari, M., Dan, W., Shuhong, S. 2017. Early immune responses to Marek's disease vaccines. Viral Immunology. doi:10.1089/vim.2016.0126. http://doi.org/10.1089/vim.2016.0126.

Teague, K.D., Graham, L.E., Dunn, J.R., Cheng, H.H., Anthony, N., Latorre, J.D., Meconi, A., Wolfeden, R.E., Wolfeden, A.D., Mahaffey, B.D., Baxter, M., Hernandez-Velasco, X., Merino-Guzman, R., Bielke, L.R., Hargis, B.M., Tellez, G. 2017. In ovo evaluation of FloraMax®-B11 on Marek´s disease HVT vaccine protective efficacy, hatchability, microbiota composition, morphometric analysis, and Salmonella Enteritidis infection in broiler chickens. Poultry Science. 96(7):2074-2082. doi:10.3382/ps/pew494. https://doi.org/10.3382/ps/pew494

6040-32000-076-00D
Integrative Strategies to Mitigate Enteric Diseases of Poultry; David Swayne (P)

Gauger, P.C., Lager, K.M., Vincent, A.L., Opriessnig, T., Kehrli, Jr., M.E., Cheung, A.K. 2011. Postweaning multisystemic wasting syndrome produced in gnotobiotic pigs following exposure to various amounts of porcine circovirus type 2a or type 2b. Veterinary Microbiology. 153(3-4):229-239. https://doi.org/10.1016/j.vetmic.2011.05.038

Wen, G., Li, L., Yu, Q., Wang, H., Luo, Q., Zhang, T., Zhang, R., Zhang, W., Shao, H. 2017. Evaluation of a thermostable Newcastle disease virus strain TS09-C as an in-ovo vaccine for chickens. PLoS One. 12(2):e0172812. doi:10.1371/journal.pone.0172812. https://doi.org/10.1371/journal.pone.0172812.

Yu, Q., Spatz, S.J., Li, Y., Yang, J., Zhao, W., Zhang, Z., Wen, G., Garcia, M., Zsak, L. 2017. Newcastle disease virus vectored infectious laryngotracheitis vaccines protect commercial broiler chickens in the presence of maternally derived antibodies. Vaccine. 35(5):789-795. doi:10.1016/j.vaccine.2016.12.038. https://doi.org/10.1016/j.vaccine.2016.12.038.

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Intervention Strategies to Prevent and Control Disease Outbreaks Caused by Emerging Strains of Avian Influenza Viruses; Mary Pantin-Jackwood (P), David Swayne, David Suarez, Erica Spackman and Darrell Kapczynski

Bertran, K., Pantin-Jackwood, M.J., Criado, M.F., Lee, D.H., Balzli, C.L. Spackman, E., Suarez, D.L., Swayne, D.E. 2019. Pathobiology and innate immune responses of gallinaceous poultry to clade 2.3.4.4A H5Nx highly pathogenic avian influenza virus infection. Veterinary Research 50:89. https://doi.org/10.1186/s13567-019-0704-5.

Chen, L.M., Donis, R.O., Suarez, D.L., Wentworth, D.E., Webby, R., Engelhardt, O.G., Swayne, D.E. Biosafety risk assessment for production and use of pandemic influenza candidate vaccine viruses for highly pathogenic avian influenza. Influenza and Other Respiratory Viruses online 10/28/2019. DOI:10.1111/irv.12698 https://doi.org/10.1111/irv.12698.

Lee, D.H., Swayne, D.E., Criado, M.F., Killmaster, L., Iqbal, S., Rashid, H.B., Chaudhry, M. 2019. Complete genome sequences of a H9N2 avian influenza virus strain found in Pakistan, 2017. Microbiological Resource Announcement 2019 Jul 11;8(28). pii: e00433-19. doi: 10.1128/MRA.00433-19. https://doi.org/10.1128/MRA.00433-19.

Leyson C., Youk, S-S., Smith, D., Dimitrov, K. Lee, D-H., Larsen, L.E., Swayne, D.E., Pantin-Jackwood, M.J. Pathogenicity and genomic changes of a 2016 European H5N8 highly pathogenic avian influenza virus (clade 2.3.4.4) in experimentally infected mallards and chickens, Virology 537:172-185. doi: 10.1016/j.virol.2019.08.020. https://doi.org/10.1016/j.virol.2019.08.020.

Ladman, B. S., J. Gelb, Jr., L. A. Sauble, M. V. Murphy and E. Spackman (2019). Protection afforded by avian influenza vaccination programmes consisting of a novel RNA particle and an inactivated avian influenza vaccine against a highly pathogenic avian influenza virus challenge in layer chickens up to 18 weeks post-vaccination. Avian Pathol 48(4): 371-381. https://doi.org/10.1080/03079457.2019.1605148.

Muzyka, D., Pantin-Jackwood, M., Spackman, E., Smith D., Rula O., Muzyka, N., Stegniy B. 2019. Isolation and genetic characterization of avian influenza viruses isolated from wild birds in the Azov-Black Sea region of Ukraine (2001-2012). Avian Diseases. 60(1 Suppl):269-78. https://doi.org/10.1637/11114-050115-Reg.

Pepin, K. M., M. W. Hopken, S. A. Shriner, E. Spackman, Z. Abdo, C. Parrish, S. Riley, J. O. Lloyd-Smith and A. J. Piaggio (2019). Improving risk assessment of the emergence of novel influenza A viruses by incorporating environmental surveillance. Philos Trans R Soc Lond B Biol Sci 374(1782): 20180346. https://doi.org/10.1098/rstb.2018.0346.

Youk, S-S., Lee, D-H., Ferreira, H.L., Afonso, C.L., Absalon, A.E., Swayne, D.E., Suarez, D.L., Pantin-Jackwood, M.J. Rapid evolution of Mexican H7N3 highly pathogenic avian influenza viruses in poultry. PlosOne. 14(9):e0222457. doi: 10.1371/journal.pone.0222457. eCollection 2019. https://doi.org/10.1371/journal.pone.0222457.

Andreychuk, D., Suarez, D.L., Andriyasov, A., Nikonova, Z., Kozlov, A., Chvala, I. 2019. Armoured exogenous internal control for real-time PCR diagnosis of avian influenza. Avian Pathology. https://doi.org/10.1080/03079457.2019.1628918.

Carnaccini, S., Santos, J.J., Obadan, A.O., Pantin Jackwood, M.J., Suarez, D.L., Rajão, D., Perez, D.R. 2019. Age-dependent pathogenesis of clade 2.3.4.4A H5N2 HPAIV in experimentally infected broad breasted white turkeys. Veterinary Microbiology. 231:183-190. https://doi.org/10.1016/j.vetmic.2019.03.011.

Criado, M.F., Bertran, K., Lee, D., Killmaster, L.F., Stephens, C.B., Spackman, E., Atkins, E., Sa E Silva, M., Mebastsion, T., Smith, R., Hughes, T., Widener, J., Pritchard, N., Swayne, D.E. 2019. Efficacy of novel recombinant fowlpox vaccine against recent Mexican H7N3 highly pathogenic avian influenza virus. Vaccine. 37(16):2232-2243. https://doi.org/10.1016/j.vaccine.2019.03.009.

Lee, D., Killian, M., Torchetti, M., Brown, I., Lewis, N., Berhane, Y., Swayne, D.E. 2019. Intercontinental spread of Asian-origin H7 avian influenza viruses by captive bird trade in 1990`s. Infection, Genetics and Evolution. 73:146-150. https://doi.org/10.1016/j.meegid.2019.04.028.

Muzyka, D., Rula, O., Tkachenko, S., Muzyka, N., Kothe, S., Stegniy, B., Pantin Jackwood, M.J., Beer, M. 2019. Highly pathogenic and low pathogenic avian influenza H5 subtype viruses in wild birds in Ukraine. Avian Diseases. 63(1):219-229. https://doi.org/10.1637/11879-042718-ResNote.1.

Ross, T.M., Dinapoli, J., Geil-Moloney, M., Bloom, C.E., Bertran, K., Balzli, C.L., Strugnell, T., Sa E Silva, M., Mebatsion, T., Bublot, M., Swayne, D.E., Kleanthous, H. 2019. A computationally designed H5 antigen shows immunological breadth of coverage and protects against drifting avian strains. Proceedings of the National Academy of Sciences. 37:2369-2376. https://doi.org/10.1016/j.vaccine.2019.03.018.

Spackman, E., Malladi, S., Ssematimba, A., Stepehns, C. 2019. Assessment of replicate numbers for titrating avian influenza virus using dose-response models. Journal of Veterinary Diagnostic Investigation. 31(4):616-619. https://doi.org/10.1177/1040638719853851.

Ssematimba, A., Malladi, S., Hagenaars, T., Bonney, P., Weaver, J., Patyk, K., Spackman, E., Halvorson, D., Cardona, C. 2019. Estimating within-flock transmission rate parameter for H5N2 highly pathogenic avian influenza virus in Minnesota turkey flocks during the 2015 epizootic. Epidemiology and Infection. 147:e179. https://doi.org/10.1017/S0950268819000633.

Stephens, C.B., Prosser, D.J., Pantin Jackwood, M.J., Berlin, A.M., Spackman, E. 2019. The pathogenesis of H7 highly pathogenic avian influenza viruses in Lesser Scaup (Aythya affinis). Avian Diseases. 63(1):230-234. https://doi.org/10.1637/11909-060118-ResNote.1.

Youk, S., Lee, D., Leyson, C., Smith, D.M., Ferreira Criado, M., DeJesus, E.G., Swayne, D.E., Pantin Jackwood, M.J. 2019. Loss of fitness of Mexican H7N3 highly pathogenic avian influenza virus in mallards after circulating in chickens. Journal of Virology. 93(14):e00543-19. https://doi.org/10.1128/JVI.00543-19.

Balzli, C.L., Bertran, K., Lee, D., Killmaster, L.F., Pritchard, N., Linz, P., Mebastsion, T., Swayne, D.E. 2018. The efficacy of recombinant turkey herpesvirus vaccines targeting the H5 of highly pathogenic avian influenza virus from the 2014/2015 North American outbreak. Vaccine. 36:84-90. https://doi.org/10.1016/j.vaccine.2017.11.026.

Bertran, K., Clark, A., Swayne, D.E. 2018. Mitigation strategies to reduce the generation and transmission of airborne highly pathogenic influenza virus particles during processing of infected poultry. International Journal of Hygiene and Environmental Health. 221(6):893-900. https://doi.org/10.1016/j.ijheh.2018.05.013.

Bertran, K., Lee, D., Criado, M.F., Balzli, C.L., Killmaster, L.F., Kapczynski, D.R., Swayne, D.E. 2018. Maternal antibody inhibition of recombinant Newcastle disease virus vectored vaccine in a primary or booster avian influenza vaccination program of broiler chickens. Vaccine. 36(43):6361-6372. https://doi.org/10.1016/j.vaccine.2018.09.015.

Bertran, K., Lee, D., Criado, M.F., Smith, D.M., Swayne, D.E., Pantin Jackwood, M.J. 2018. Pathobiology of Tennessee 2017 H7N9 low and high pathogenicity avian influenza viruses in commercial broiler breeders and specific pathogen free layer chickens. Veterinary Research. 49:82. https://doi.org/10.1186/s13567-018-0576-0.

Chen, L., Lee, D., Liu, Y., Li, W., Swayne, D.E., Chang, J., Chen, Y., Lee, F., Tu, W., Lin, Y. 2018. Reassortant clade 2.3.4.4 of highly pathogenic avian influenza A (H5N6) virus, Taiwan, 2017. Emerging Infectious Diseases. 24(6):1147-1149. https://doi.org/10.3201/eid2406.172071.

Chrzastek, K., Lee, D., Gharaineh, S., Zsak, A., Kapczynski, D.R. 2018. Characterization of H9N2 avian influenza viruses from the Middle East demonstrates heterogeneity at amino acid position 226 in the hemagglutinin and potential for transmission to mammals. Virology. 518: 195-2017. https://doi.org/10.1016/j.virol.2018.02.016.

Dharmayanti, N., Thor, S.W., Zanders, N., Hartawan, R., Ratnawati, A., Jang, Y., Rodriguez, M., Suarez, D.L., Samaan, G., Pudjiatmoko, Davis, C. 2018. Attenuation of highly pathogenic avian influenza A (H5N1) viruses in Indonesia following reassortment and acquisition of genes from low pathogenicity avian influenza (LPAI) A virus progenitors. Emerging Microbes & Infections. 7(1):1-14. https://doi.org/10.1038/s41426-018-0147-5.

Kwon, J., Jeong, S., Lee, D., Swayne, D.E., Kim, Y., Lee, S., Noh, J., Erdene-Ochir, T., Jeong, J., Song, C. 2018. New reassortant clade 2.3.4.4b avian influenza A (H5N6) virus in wild birds, South Korea, 2017-2018. Emerging Infectious Diseases. 24(10):1953-1955. https://doi.org/10.3201/eid2410.180461.

Kwon, J., Lee, D., Swayne, D.E., Noh, J., Yuk, S., Jeong, S., Lee, S.H., Woo, C., Shin, J.H., Song, C. 2018. Experimental infection of H5N1 and H5N8 highly pathogenic avian influenza viruses in Northern

Pintail (Anas acuta). Transboundary and Emerging Diseases. 65(5):1367-1371. https://doi.org/10.1111/tbed.12872.

Lee, D., Torchetti, M.K., Hicks, J., Killian, M.L., Bahl, J., Pantin Jackwood, M.J., Swayne, D.E. 2018. Transmission dynamics of highly pathogenic avian influenza virus A(H5Nx) clade 2.3.4.4, North America, 2014–2015. Emerging Infectious Diseases. 24(10):1840-1848. https://doi.org/10.3201/eid2410.171891.

Liu, Y., Lee, D., Chen, L., Lin, Y., Li, W., Hu, S., Chen, Y., Swayne, D.E., Lee, M. 2018. Detection of reassortant H5N6 clade 2.3.4.4 highly pathogenic avian influenza virus in a black-faced spoonbill (Platalea minor) found dead, Taiwan, 2017. Infection, Genetics and Evolution. 62:275-278. https://doi.org/10.1016/j.meegid.2018.04.026.

Nguyen, D., Shepard, S.S., Burke, D.F., Jones, J., Nguyen, L., Thor, S., Nguyen, T., Balish, A., Hoang, D., To, T., Iqbal, M., Wentworth, D., Spackman, E., Van Doorn, R., David, T.C., Bryant, J.E. 2018. Antigenic characterization of highly pathogenic avian influenza A(H5N1) viruses with chicken and ferret antisera reveals similar haemagglutination inhibition profiles. Emerging Microbes & Infections. 7(1):1-14. https://doi.org/10.1038/s41426-018-0100-7.

Pantin-Jackwood, M.J., DeJesus, E., Costa-Hurtado, M., Smith, D.M., Chrzastek, K., Kapczynski, D.R., Suarez, D.L. 2018. Efficacy of two licensed H5 vaccines against challenge with a 2015 United States H5N2 clade 2.3.4.4 highly pathogenic avian influenza virus in domestic ducks. Avian Diseases. 62:90-96. https://doi.org/10.1637/11895-050918-Reg.1.

Pusch, E., Suarez, D.L. 2018. The multifaceted zoonotic risk of H9N2 avian influenza. Veterinary Sciences. 5(4):82. https://doi.org/10.3390/vetsci5040082.

Spackman, E., D. J. Prosser, M. Pantin-Jackwood, C. B. Stephens and A. M. Berlin (2019). Clade 2.3.4.4 H5 North American Highly Pathogenic Avian Influenza Viruses Infect, but Do Not Cause Clinical Signs in, American Black Ducks (Anas rubripes). Avian Dis 63(2): 366-370. https://doi.org/10.1637/11950-081418-ResNote.1

Ramey, A.M., Deliberto, T.J., Berhane, Y., Swayne, D.E., Stallknecht, D.E. 2018. Lessons learned from research and surveillance directed at highly pathogenic influenza A viruses in wild birds inhabiting North America. Virology. 518:55-63. https://doi.org/10.1016/j.virol.2018.02.002.

Segovia, K., Franca, M., Leyson, C.L., Kapczynski, D.R., Chrzastek, K., Bahnson, C.S., Stallknecht, D. 2018. Heterosubtypic immunity increases infectious dose required to infect mallard ducks to Influenza A Virus. PLoS One. 13(4):e0196394. https://doi.org/10.1371/journal.pone.0196394.

Sharma, P., Killmaster, L.F., Volkening, J.D., Cardenas-Garcia, S., Wajid, A., Regmani, S.F., Basharat, A., Miller, P.J., Afonso, C.L. 2018. Draft genome sequence of three novel Ochrobactrum spp. isolated from different avian hosts in Pakistan. Genome Announcements. 6(15):e00269-18. https://doi.org/10.1128/genomeA.00269-18.

Vuong, C., Chou, W., Briggs, W., Faulkner, O., Wolfenden, A., Jonas, M., Kapczynski, D.R., Hargis, B.M., Bielke, L.R., Berghman, L.R. 2018. Crude inactivated influenza A virus adjuvated with a bispecific antibody complex targeting chicken CD40 and AIV M2e confers protection against lethal HPAI challenge in chickens. Monoclonal Antibodies in Immunodiagnosis and Immunotherapy. 37(6):245-251. https://doi.org/10.1089/mab.2018.0040.

Ahmed, N., Spackman, E., Kapczynski, D.R. 2017. Immunologic evaluation of 10 different adjuvants for use in vaccines for chickens against highly pathogenic avian influenza virus. Vaccine. 35 (26):3401-3408. doi:10.1016/j.vaccine.2017.05.010. https://doi.org/10.1016/j.vaccine.2017.05.010

Suarez, D.L., Pantin-Jackwood, M.J. 2017. Recombinant viral-vectored vaccines for the control of avian influenza in poultry. Veterinary Microbiology. 206:144-151. doi:10.1016/j.vetmic.2016.11.025. https://doi.org/10.1016/j.vetmic.2016.11.025

Bertran, K., Balzli, C.L., Kwon, Y., Tumpey, T.M., Clark, A., Swayne, D.E. 2017. Airborne transmission of highly pathogenic influenza virus during processing of infected poultry. Emerging Infectious Diseases. 23(11):1806-1814. https://doi.org/10.3201/eid2311.170672.

Bertran, K., Balzli, C.L., Lee, D., Suarez, D.L., Kapczynski, D.R., Swayne, D.E. 2017. Protection of White Leghorn chickens by U.S. emergency H5 vaccines against clade 2.3.4.4 H5N2 high pathogenicity avian influenza virus. Vaccine. 35:6336-6344. http://doi.org/10.1016/j.vaccine.2017.05.051.

Bertran, K., Lee, D., Pantin Jackwood, M.J., Spackman, E., Balzli, C.L., Suarez, D.L., Swayne, D.E. 2017. Pathobiology of clade 2.3.4.4 H5Nx high pathogenicity avian influenza virus infections in minor gallinaceous poultry supports early backyard flock introductions in Western U.S., 2014-2015. Journal of Virology. 91(21):e00960-17. https://doi.org/10.1128/JVI.00960-17.

Chrzastek, K., Lee, D., Smith, D.M., Sharma, P., Suarez, D.L., Pantin Jackwood, M.J., Kapczynski, D.R. 2017. Use of Sequence-Independent, Single-Primer-Amplification (SISPA) for rapid detection, identification, and characterization of avian RNA viruses. Virology. 509:159-166. doi:10.1016/j.virol.2017.06.019. https://doi.org/10.1016/j.virol.2017.06.019

Jung-Hoon, K., Lee, D., Swayne, D.E., Jin-Yong, N., Seong-Su, Y., Tseren-Ochir, E., Woo-Tack, H., Jei-Hyun, J., Sol, J., Gyeong-Bin, G., Chang-Seon, S. 2017. Reassortant clade 2.3.4.4 Avian Influenza A(H5N6) Virus in a wild Mandarin Duck, South Korea, 2016. Emerging Infectious Diseases. 23(5):822-826. doi:10.3201/eid2305.161905. http://doi.org/10.3201/eid2305.161905

Swayne, D.E., Hill R.E., Clifford, J. 2017. Safe application of regionalization for trade in poultry and poultry products during highly pathogenic avian influenza outbreaks in the USA. Avian Pathology 46(2):125-130. URL: http://dx.doi.org/10.1080/03079457.2016.1257775.

Kapczynski, D.R., Sylte, M.J., Killian, M.L., Torchetti, M.K., Chrzastek, K., Suarez, D.L. 2017. Protection of commercial turkeys following inactivated or recombinant H5 vaccine application against the 2015 U.S. H5N2 clade 2.3.4.4 highly pathogenic avian influenza virus. Veterinary Immunology and Immunopathology. 191(2017):74-79. http://doi.org/10.1016/j.vetimm.2017.08.001.

Lee Dong-Hun, Torchetti, M.K., Killian, M.L., Berhane, Y., Swayne, D.E. 2017. Highly pathogenic avian influenza A(H7N9) virus, Tennessee, USA, March 2017. Emerging Infectious Diseases. 23(11):1860-1863. https://doi.org/10.3201/eid2311.171013.

Lee, D., Bertran, K., Kwon, J., Swayne, D.E. 2017. Evolution, global spread, and pathogenicity of highly pathogenic avian influenza H5Nx clade 2.3.4.4. Journal of Veterinary Science. 18(S1):269-280. https://doi.org/10.4142/jvs.2017.18.S1.269.

Newbury, S.P., Cigel, F., Killian, M., Leutenegger, C., Seguin, A., Crossley, B., Brennen, R., Suarez, D.L., Torchetti, M., Toohey-Kurth, K. 2017. First detection of avian lineage H7N2 in Felis catus. Genome Announcements. 5(23):e00457-17. doi:10.1128/genomeA.00457-17. https://doi.org/10.1128/genomeA.00457-17.

Pantin Jackwood, M.J., Costa-Hurtado, M., Bertran, K., Dejesus, E.G., Smith, D.M., Swayne, D.E. 2017. Infectivity, transmission and pathogenicity of H5 highly pathogenic avian influenza clade 2.3.4.4 (H5N8 and H5N2) United States index viruses in Pekin ducks and Chinese geese. Veterinary Research. 48(1):33. doi:10.1186/s13567-017-0435-4. https://doi.org/10.1186/s13567-017-0435-4

Pantin Jackwood, M.J., Stephens, C., Bertran-Dols, K., Swayne, D.E., Spackman, E. 2017. The pathogenesis of H7N8 low and highly pathogenic avian influenza viruses from the United States 2016 outbreak in chickens, turkeys and mallards. PLoS One. 12(5):1-21. doi:10.1371/journal.pone.0177265. https://doi.org/10.1371/journal.pone.0177265.

Santos, J., Obadan, A., Cardenas Garcia, S., Carnaccini, S., Pantin Jackwood, M.J., Suarez, D.L., Kapczynski, D.R., Perez, D.R. 2017. Short- and long-term protective efficacy against clade 2.3.4.4 H5N2 highly pathogenic avian influenza virus following prime-boost vaccination in turkeys. Vaccine. 35:5637-5643. http://doi.org/10.1016/j.vaccine.2017.08.059.

Spackman, E., Prosser, D., Pantin Jackwood, M.J., Berlin, A., Stephens, C.B. 2017. The pathogenesis of clade 2.3.4.4 H5 highly pathogenic avian influenza viruses in Ruddy Ducks (Oxyura jamaicensis) and Lesser Scaup (Aythya affinis). Journal of Wildlife Diseases. 53(4):1-11. doi:10.7589/2017-01-003. https://doi.org/10.7589/2017-01-003

Spackman, E., Stephens, C., Pantin Jackwood, M.J. 2017. The effect of infectious bursal disease virus induced immunosuppression on vaccination against highly pathogenic avian influenza virus. Avian Pathology. 62(1):36-44. https://doi.org/10.1637/11769-110717-Reg.1.

Stephens, C., Spackman, E. 2017. Thermal inactivation of avian influenza virus in poultry litter as a method to decontaminate poultry houses. Preventive Veterinary Medicine. 145:73–77. https://doi.org/10.1016/j.prevetmed.2017.06.012

Kapczynski, D.R., Pantin Jackwood, M.J., Spackman, E., Chrzastek, K., Suarez, D.L., Swayne, D.E. 2017. Homologous and heterologous antigenic matched vaccines containing different H5 hemagglutinins provides variable protection of chickens from the 2014 U.S. H5N8 and H5N2 clade 2.3.4.4 highly pathogenic avian influenza viruses. Vaccine. 35:6345-6353. http://doi.org/10.1016/j.vaccine.2017.04.042.

Segovia, K.M., Stalknecht, D.E., Kapczynski, D.R., Stabler, L., Berghaus, R.D., Fotjik, A., Latorre-Margalef, N., Franca, M.S. 2017. Adaptive heterosubtypic immunity to low pathogenic avian influenza viruses in experimentally infected mallards. PLoS One. 12(1):e0170335. doi:10.1371/journal.pone.0170335. https://doi.org/10.1371/journal.pone.0170335

Lee, D.H., Sharshov, K., Swayne, D.E., Kurskaya, O., Sobolev, I., Kabilov, M., Alekseev, A., Irza, V., Shestopalov, 2017. A. Novel Reassortant Clade 2.3.4.4 Avian influenza A(H5N8) virus in wild aquatic birds, Russia, 2016. Emerging Infectious Diseases 23(2):358-360. http://doi.org/10.3201/eid2302.161252.

Balzli, C.L., Lager, K., Vincent, A., Gauger, P., Brockmeier, S., Miller, L., Richt, J.A., Ma, W., Suarez, D.L., Swayne, D.E. 2016. Susceptibility and pathogenicity of swine to H5 and H7 low pathogenic avian influenza viruses. Influenza and Other Respiratory Viruses 10(4):346-52. doi: 10.1111/irv.12386. https://doi.org/10.1111/irv.12386

Bertran, K., Swayne, D.E., Pantin-Jackwood, M., Kapczynski, D.R., Spackman, E., Suarez, D.L. Lack of chicken adaptation of newly emergent Eurasian H5N8 and reassortant H5N2 high pathogenicity avian influenza viruses in the U.S. is consistent with restricted poultry outbreaks in the Pacific flyway during 2014–2015. Virology 494:190-197. doi: 10.1016/j.virol.2016.04.019. https://doi.org/10.1016/j.virol.2016.04.019

Costa-Hurtado, M., Afonso, C.L., Miller, P.J., Shepherd, E., DeJesus, E., Smith, D., and Pantin-Jackwood, M.J. Effect of infection with a mesogenic strain of Newcastle Disease Virus on infection with highly pathogenic avian influenza virus in chickens. Avian Diseases 60(1 Suppl):102-3. 2016. https://doi.org/10.1637/11171-051915-Reg

DeJesus, E., Costa-Hurtado, M., Smith, D., Lee, D.H., Spackman, E., Kapczynski, D.R., Kim-Torchetti, M., Killian, M.L., Suarez, D.L., Swayne, D.E., Pantin-Jackwood, M.J. 2016. Changes in adaptation of H5N2 highly pathogenic avian influenza H5 clade 2.3.4.4 viruses in chickens and mallards. Virology 499(2016):52-64. http://doi.org/10.1016/j.virol.2016.08.036.

Kwon, J.H., Lee, D.H., Swayne, D.E., Yuk, S.S., Ochir, T., Ochir, E., Noh, J.Y., Hong, W.T., Jeong, J.H., Jeong, S., Gwon, G.B., Song, C.S. 2016. Highly pathogenic H5N8 avian influenza viruses re-introduced to South Korea by migratory waterfowl in late 2014. Emerging Infectious Diseases 22(3):507-510. https://dx.doi.org/10.3201/eid2203.151006

Lee, D.H., Fusaro, A., Song, C.S., Suarez, D.L., Swayne, D.E. 2016. Poultry vaccination directed evolution of H9N2 low pathogenicity avian influenza viruses in Korea. Virology 488:225-231. https://doi.org/10.1016/j.virol.2015.11.023

Lee, D.H., Swayne, D.E., Sharma, P., Rehmani, S.F., Wajid, A., Suarez, D.L., Afonso, C. 2016. H9N2 Low pathogenic avian influenza in Pakistan (2012-2015). Veterinary Record Open 3:e000171, 2016. doi:10.1136/vetreco-2016-000171. http://dx.doi.org/10.1136/vetreco-2016-000171

Lee, D.H., Bahl, J., Torchetti, M.K., Killian, M.L., Ip, H.S., Deliberto, T., Swayne, D.E. 2016. Asian H5N8 HPAI virus spread into North America in late 2014 and produced novel H5N1, H5N2, and H5N8 viruses by reassortment events. Emerging Infectious Diseases 22(7):1283-1285 http://doi.org/10.3201/eid2207.160048

Muzyka, D., Pantin-Jackwood, M., Starick, E., Fereidouni, S. 2016. Evidence for genetic variation of Eurasian avian influenza viruses, subtype H15: The first report of an H15N7 virus. Archives of Virology 161(3):605-12. https://doi.org/10.1007/s00705-015-2629-2

Pantin-Jackwood, M.J., Kapczynski, D., DeJesus, E., Costa-Hurtado, M., Dauphin G., Tripodi, A., Dunn, J.R., Swayne, D.E. 2016. Efficacy of a recombinant Turkey Herpesvirus (HVT) H5 vaccine against challenge with H5N1 clades 1.1.2 and 2.3.2.1 highly pathogenic avian influenza viruses in domestic ducks (Anas platyrhynchos domesticus). Avian Diseases 60(1):22-32. https://doi.org/10.1637/11282-091615-Reg.1

Pantin-Jackwood, M.J., Costa-Hurtado, M., Shepherd, E., DeJesus, E., Smith, D., Spackman, E., Kapczynski, D.R., Suarez, D.L., Stallknecht, D., Swayne, D.E. 2016. Pathogenicity and transmission of H5 and H7 highly pathogenic avian influenza viruses in mallards, Journal of Virology 90(21):9967-9982. http://doi:10.1128/JVI.01165-16.

Reeves, A., Poulson, R.L., Muzyka, D., Ogawa, H., Imai, K., Bui, V.N., Hall, J.S., Pantin-Jackwood, M., Stallknecht, D.E., .Ramey, A.M. 2016. Limited evidence of intercontinental dispersal of avian paramyxovirus serotype 4 bymigratory birds. Infect Genet Evol. 2016. 40:104-8. https://doi.org/10.1016/j.meegid.2016.02.031

Sá e Silva, M., Moresco, K., Swayne, D.E. 2016. Very virulent infectious bursal disease virus produces more severe disease and lesions in leghorn chickens than in broiler chickens. Avian Diseases 60(1):63-66. https://doi.org/10.1637/11230-070615-ResNote.1

Sá e Silva, M., Moresco, K., Bertran, K., Jackwood, D.J., Swayne, D.E. 2016. Infection with some infectious bursal disease virus pathotypes produces virus in chicken muscle tissue and the role of humoral immunity as a mitigation strategy. Avian Diseases 60(4):758-764. https://doi.org/10.1637/11394-021716-Reg

Wang, Y., I. Davidson, R. Fouchier and E. Spackman (2016). Antigenic Cartography of H9 Avian Influenza Virus and Its Application to Vaccine Selection. Avian Diseases 60(1 Suppl): 218-225. https://doi.org/10.1637/11087-041015-Reg

Xu, Y., E. Bailey, E. Spackman, T. Li, H. Wang, L. P. Long, J. A. Baroch, F. L. Cunningham, X. Lin, R. G. Jarman, T. J. DeLiberto and X. F. Wan (2016). Limited Antigenic Diversity in Contemporary H7 Avian-Origin Influenza A Viruses from North America. Sci Rep 6: 20688. https://doi.org/10.1038/srep20688

Pearce, M.B., Gustin, K.M., Pappas, C., David, T., Pantin Jackwood, M.J., Swayne, D.E., Belser, J.A., Tumpey, T.M. 2016. Enhanced virulence of clade 2.3.2.1 highly pathogenic avian influenza A(H5N1) viruses in ferrets. Virology. 502:114-122. https://doi.org/10.1016/j.virol.2016.12.024

Dimitrov, K.M., Zarkov, I.S., Dinev, I., Goujgoulova, G.V., Miller, P.J., Suarez, D.L. 2016. Histopathological characterization and shedding dynamics of guineafowl (Numida meleagris) intravenously infected with a H6N2 low pathogenicity avian influenza virus. Avian Diseases. 60(1s):279-85. https://doi.org/10.1637/11141-050815-Reg.

Bevins, S. N., R. J. Dusek, C. L. White, T. Gidlewski, B. Bodenstein, K. G. Mansfield, P. DeBruyn, D. Kraege, E. Rowan, C. Gillin, B. Thomas, S. Chandler, J. Baroch, B. Schmit, M. J. Grady, R. S. Miller, M. L. Drew, S. Stopak, B. Zscheile, J. Bennett, J. Sengl, C. Brady, H. S. Ip, E. Spackman, M. L. Killian, M. K. Torchetti, J. M. Sleeman and T. J. Deliberto. 2016. Widespread detection of highly pathogenic H5 influenza viruses in wild birds from the Pacific Flyway of the United States. Sci Rep 6: 28980. https://doi.org/10.1038/srep28980

Pushko, P., Tretyakova, I., Hidajat, R., Zsak, A., Chrzastek, K., Tumpey, T.M., Kapczynski, D.R. 2016. Virus-like particles comprising H5, H7 and H9 hemagglutinins elicit protective immunity to heterologous avian influenza viruses in chickens. Virology. 501:176-182. doi:10.1016/j.virol.2016.12.001. https://doi.org/10.1016/j.virol.2016.12.001

Lee, D., Sharshov, K., Swayne, D.E., Kurskaya, O., Sobolev, I., Kabilov, M., Alekseev, A., Irza, V., Shestopalov, A. 2017. Novel H5N8 clade 2.3.4.4 highly pathogenic avian influenza virus in wild awuatic birds, Russia, 2016. Emerging Infectious Diseases. 23(2):358-360. doi: 10.3201/eid2302.161252. http://doi.org/10.3201/eid2302.161252

Lee, D., Torchetti, M., Killian, M., Deliberto, T., Swayne, D.E. 2017. Reoccurrence of avian influenza A(H5N2) virus clade 2.3.4.4 in wild birds, Alaska, USA, 2016. Emerging Infectious Diseases. 23(2):365-367. doi:10.3201/eid2302.161616. http://doi.org/10.3201/eid2302.161616

Lee, D., Torchetti, M., Killian, M., Swayne, D.E. 2017. Deep sequencing of H7N8 avian influenza viruses from surveillance zone supports H7N8 high pathogenicity avian influenza was limited to a single outbreak farm in Indiana during 2016. Virology. 507:216-219. doi:10.1016/j.virol.2017.04.025. https://doi.org/10.1016/j.virol.2017.04.025

Bertran, K., Lee, D., Balzli, C.L., Pantin Jackwood, M.J., Spackman, E., Swayne, D.E. 2016. Age is not a determinant factor in susceptibility of broilers to H5N2 clade 2.3.4.4 high pathogenicity avian influenza virus. Veterinary Research. 47:116. doi:10.1186/s13567-016-0401-6. https://doi.org/10.1186/s13567-016-0401-6

Lycett, S.J., Bodewes, R., Pohlmann, A., Banks, J., Banyai, K., Boni, M.F., Bouwstra, R., Breed, A.C., Brown, I.H., Chen, H., Dan, A., DeLiberto, T.J., Diep, N., Gilbert, M., Hill, S., Ip, H.S., Ke, C.W., Kida, H., Killian, M.L., Koopmans, M.P., Kwon, J., Lee, D., Lee, Y.J., Lu, L., Monne, I., Pasick, J., Pybus, O.G., Rambaut, A., Robinson, T.P., Sakoda, Y., Zohari, S., Song, C., Swayne, D.E., Torchetti, M.K., Tsai, H., Fouchier, R., Beer, M. Woolhouse, M., Kuiken, T. 2016. Role for migratory wild birds in the global spread of avian influenza H5N8. Science Magazine. 354(6309):213-217. doi:10.1126/science.aaf8852. https://science.sciencemag.org/content/sci/354/6309/213.full.pdf

Spackman, E., Cardona, C., Munoz-Aguayo, J., Fleming, S. 2016. Successes and short comings in four years of an international external quality assurance program for animal Influenza surveillance. PLoS One. 11(10):e0164261. doi:10.1371/journal.pone.0164261. https://doi.org/10.1371/journal.pone.0164261.

Spackman, E., Pantin Jackwood, M.J., Kapczynski, D.R., Swayne, D.E., Suarez, D.L. 2016. H5N2 highly pathogenic avian influenza viruses from the US 2014-2015 outbreak have an unusually long pre-clinical period in turkeys. BioMed Central (BMC) Veterinary Research. 12:260. doi:10.1186/s12917-016-0890-6. https://doi.org/10.1186/s12917-016-0890-6

Swayne, D.E., Hill, R.E., Clifford, J. 2016. Safe application of regionalization for trade in poultry and poultry products during highly pathogenic Avian Influenza outbreaks in USA. Avian Pathology. 46(2):125-130. doi:10.1080/03079457.2016.1257775 https://doi.org/10.1080/03079457.2016.1257775.

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Intervention Strategies to Predict, Prevent, and Control Disease Outbreaks Caused by Emerging Strains of Virulent Newcastle Disease Viruses; Claudio Afonso (P) and David Suarez

Ayala, A.J., Olivier, T.L., Welch, C.N., Dimitrov, K., Goraichuk, I.V., Afonso, C.L., Miller, P.J., Hernandez, S.M. 2019. Experimental Infection and Transmission of Newcastle Disease Vaccine Virus in Passerines. Avian Diseases. https://doi.org/10.1637/11980-092918-Reg.1.

Butt, S.L., Dimitrov, K.M., Zhang, J., Wajid, A., Bibi, T., Basharat, A., Brown, C.C., Rehmani, S.F., Stanton, J.B., Afonso, C.L. 2019. Enhanced phylogenetic resolution of Newcastle disease outbreaks using complete viral genome sequences from formalin-fixed paraffin-embedded tissue samples. Virus Genes. 55(4):505-512. https://doi.org/10.1007/s11262-019-01669-9.

Dimitrov, K.M., Abolnik, C., Afonso, C.L., Albina, E., Bahl, J., Berg, M., Briand, F., Brown, I.H., Choi, K., Chvala, I., Durr, P.A., Ferreira, H.L., Fusaro, A., Torcheti, M., Miller, P.J., Suarez, D.L. 2019.

Updated unified phylogenetic classification system and revised nomenclature for Newcastle disease virus. Infection, Genetics and Evolution. 74:103917. https://doi.org/10.1016/j.meegid.2019.103917.

Dimitrov, K.M., Ferreira, H.L., Pantin Jackwood, M.J., Taylor, T.L., Goraichuk, I.V., Crossley, B.M., Killian, M., Bergeson, N., Torchetti, M., Afonso, C.L., Suarez, D.L. 2019. Pathogenicity and transmission of virulent Newcastle disease virus from the 2018-2019 California outbreak and related viruses in young and adult chickens. Virology. 531:203-218. https://doi.org/10.1016/j.virol.2019.03.010.

Ferreira, H.L., Taylor, T.L., Absalon, A.E., Dimitrov, K.M., Cortes-Espinosa, Butt, S.L., Goraichuk, I.V., Volkening, J.D., Suarez, D.L., Afonso, C.L. 2019. Presence of Newcastle disease viruses of sub-genotypes Vc and VIn in backyard chickens and in apparently healthy wild birds from Mexico in 2017. Virus Genes. 55(4):479-489. https://doi.org/10.1007/s11262-019-01663-1.

Ferreira, H.L., Taylor, T.L., Sabra, M., Dimitrov, K.M., Afonso, C.L., Suarez, D.L. 2019. Virulent newcastle disease viruses from chicken origin are more pathogenic and transmissible than viruses normally maintained in wild birds. Veterinary Microbiology. 235:25-34. https://doi.org/10.1016/j.vetmic.2019.06.004.

Goraichuk, I.V., Msoffe, P.L., Chiwangad, G.H., Dimitrov, K.M., Afonso, C.L., Suarez, D.L. 2019. First complete genome sequences of a subgenotype Vd Newcastle disease virus isolate. Microbiology Resource Announcements. 8(27):e00436-19. https://doi.org/10.1128/MRA.00436-19.

Hicks, J.T., Dimitrov, K.M., Afonso, C.L., Ramey, A.M., Bahl, J. 2019. Global phylodynamic analysis of avian paramyxovirus-1 provides evidence of inter-host transmission and intercontinental spatial diffusion. BMC Evolutionary Biology. 19:108. https://doi.org/10.1186/s12862-019-1431-2.

Welch, C.N., Shittu, I., Abolnik, C., Ponman, S., Dimirov, K.M., Taylor, T.L., Williams Coplin, T.D., Goraichuk, I.V., Meseko, C.A., Ibu, J.O., Gado, D.A., Joannis, T.M., Afonso, C.L. 2019. Genomic comparison of Newcastle disease viruses isolated in Nigeria between 2002 and 2015 reveals circulation of highly diverse genotypes and spillover into wild birds. Archives of Virology. 164(8):2031-2047. https://doi.org/10.1007/s00705-019-04288-9.

Ababneh, M., Ferreira, H., Khalifeh, M., Suarez, D.L., Afonso, C.L. 2018. First genome sequence of Newcastle Disease Virus of Genotype VIIi from Jordan. Microbiology Resource Announcements. 5(23):e01136-18. https://doi.org/10.1128/MRA.01136-18.

Butt, S.L., Taylor, T.L., Volkening, J.D., Dimitrov, K., Williams Coplin, T.D., Lahmers, K.K., Suarez, D.L., Afonso, C.L., Stanton, J. 2018. Rapid virulence prediction and identification of Newcastle disease virus genotypes using third generation sequencing. Virology Journal. 15:179. https://doi.org/10.1186/s12985-018-1077-5.

He, Y., Taylor, T.L., Dimitrov, K.M., Butt, S.L., Stanton, J.B., Goraichuk, I.V., Fenton, H., Poulson, R., Zhang, J., Brown, C.C., Ip, H.S., Isidoro-Ayza, M., Afonso, C.L. 2018. Whole genome sequencing of genotype VI Newcastle disease viruses from formalin-fixed paraffin-embedded tissues from wild pigeons reveals continuous evolution and previously unrecognized genetic diversity in the U.S. Virology Journal. 15:9. https://doi.org/10.1186/s12985-017-0914-2.

Joshi, L.R., Bauermann, F.V., Hain, K.S., Kutish, G.F., Armien, A.G., Lehman, C.P., Neiger, R., Afonso, C.L., Tripathy, D.N., Diego, D.G. 2018. Detection of fowlpox virus carrying distinct genome segments of reticuloendotheliosis virus. Archives of Virology. 260:53-59. https://doi.org/10.1016/j.virusres.2018.10.017.

Sá e Silva, M., Susta, L., Moresco, K., Swayne, D.E. 2016. Vaccination of chickens decreased Newcastle disease virus contamination in eggs. Avian Pathology 45(1):38-45. http://doi.org/10.1080/03079457.2015.1112876

Sharma, P., Killmaster, L.F., Volkening, J.D., Cardenas-Garcia, S., Shittu, I., Meseko, C., Joannis, T.M., Miller, P.J., Afonso, C.L. 2018. Draft genome sequence of five novel Ochrobactrum spp. isolated from different avian hosts in Nigeria. Genome Announcements. 6:e00063-18. https://doi.org/10.1128/genomeA.00063-18.

Sharma, P., Killmaster, L.F., Volkering, J.D., Cardenas-Garcia, S., Wajid, A., Rehmani, S.F., Basharatd, A., Miller, P.J., Afonso, C.L. 2018. Draft genome sequence of three novel Ochrobactrum spp. isolated from different avian hosts in Pakistan. Genome Announcements. 6:e00269-18. https://doi.org/10.1128/genomeA.00269-18.

Susta, L., Segovia, D., Olivier, T.L., Dimitrov, K.M., Shittu, I., Marcano, V., Miller, P.J. 2018. Newcastle disease virus infection in quail. Veterinary Pathology. 55(5):682-692. https://doi.org/10.1177/0300985818767996.

Ying, H., Taylor, T.L., Dimitrov, K.M., Butt, S.L., Stanton, J.B., Goraichuk, I.V., Fenton, H., Poulson, R., Jian, Z., Brown, C.C., Ip, H.S., Isidoro-Ayza, M., Afonso, C.L. 2018. Whole-genome sequencing of genotype VI Newcastle disease viruses from formalin-fixed paraffin-embedded tissues from wild pigeons reveals continuous evolution and previously unrecognized genetic diversity in the U.S.. Virology Journal. 15(9):1-11. https://doi.org/10.1186/s12985-017-0914-2.

Dimitrov, K.M., Sharma, P., Volkening, J.D., Goraichuk, I.V., Wajid, A., Rehmani, S., Asma, B., Shittu, I., Joannis, T.M., Miller, P.J., Afonso, C.L. 2017. A robust and cost-effective approach to sequence and analyze complete genomes of small RNA viruses. Virology Journal. 14(2017):72. doi: 10.1186/s12985-017-0741-5. https://doi.org/10.1186/s12985-017-0741-5

Goraichuk, I.V., Dimitrov, K.M., Sharma, P., Miller, P.J., Swayne, D.E., Suarez, D.L., Afonso, C.L. 2017. Complete genome sequences of four avian paramyxoviruses of serotype 10 isolated from Rockhopper Penguins on the Falkland Islands. Genome Announcements. 5:e000472-17. doi:org/10.1128/genomeA.00472-17. https://doi.org/10.1128/genomeA.00472-17

Marcano, V.C., Cardenas-Garcia, S., Gogal Jr, R.M., Afonso, C.L. 2017. Intracellular fixation buffer inactivates Newcastle Disease Virus in chicken allantoic fluid, macrophages and splenocytes for immune assessment during infection. Journal of Virological Methods. 251:1-6. http://doi.org/10.1016/j.jviromet.2017.09.025.

Ramey, A.W., Goraichuk, I.V., Hicks, J.T., Dimitrov, K.M., Poulson, R.L., Stallknecht, D.E., Bahl, J., Afonso, C.L. 2017. Assessment of contemporary genetic diversity and inter-taxa/inter-region exchange of avian paramyxovirus serotype 1 in wild birds sampled in North America. Virology Journal. 14:43. doi:10.1186/s12985-017-0714-8. https://doi.org/10.1186/s12985-017-0714-8

Sabra, M., Dimitrov, K., Goraichuk, I.V., Wajid, A., Sharma, P., Rehmani, S.F., Muzyka, D.V., Williams Coplin, T.D., Miller, P.J., Afonso, C.L. 2017. Phylogenetic assessment reveals continuous evolution and circulation of pigeon-derived virulent avian avulaviruses 1 in Eastern Europe, Asia, and Africa. BMC Veterinary Research. 13(2017):291. https://doi.org/10.1186/s12917-017-1211-4.

Valdez-Gomez, H., Navarro-Lopez, R., Vazques-Mendoza, L., Zalapa-Hernandez, M., Guerrero-Hernandex, I., Fonseca-Delgado, V., Marquez-Ruiz, M., Afonso, C.L. 2017. Risk factors for the transmission of infectious diseases agents at the wild birds-commercial birds interface. A pilot study in the region of the Altos de Jalisco, Mexico. BULLETIN DE L'ACADEMIE VETERINAIRE DE FRANCE.170(2):143-150. https://doi.org/10.4267/2042/62332.

Igwe, A.O., Ezema, W.S., Afonso, C.L., Brown, C.C., Okoye, J. 2018. Pathology and distribution of velogenic viscerotropic newcastle disease virus in the reproductive system of vaccinated and unvaccinated laying hens (gallus gallus domesticus) by immunohistochemical labelling. Journal of Comparative Pathology. 159:36-48. https://doi.org/10.1016/j.jcpa.2017.12.009.

Dimitrov, K.M., Afonso, C.L., Yu, Q., Miller, P.J. 2017. Newcastle disease vaccines- a solved problem or a continuous challenge? Veterinary Microbiology. 206:126-136. https://doi.org/10.1016/j.vetmic.2016.12.019.

Isidoro-Ayza, M., Afonso, C.L., Stanton, J.B., Knowles, S., Ip, H.S., White, C.L., Fenton, H., Ruder, M.G., Dolinski, A.C., Lankton, J. 2017. Natural infections with pigeon paramyxovirus serotype 1: Pathologic changes in Eurasian collared-doves (Streptopelia decaocto) and rock pigeons (Columba livia) in the United States. Veterinary Pathology. 54(4):695-703. doi:10.1177/0300985817695782. https://pubag.nal.usda.gov/catalog/6150267

Taylor, T.L., Dimitrov, K.M., Afonso, C.L. 2017. Genome-wide analysis reveals class and gene specific codon usage adaptation in avian paramyxoviruses 1. Infection, Genetics and Evolution. 50(2017):28-37. doi:10.1016/jmeegid.2017.02.004. https://doi.org/10.1016/j.meegid.2017.02.004

Taylor, T.L., Miller, P.J., Olivier, T.L., Montiel, E., Cardenas Garcia, S., Dimitrov, K.M., Williams Coplin, T.D., Afonso, C.L. 2017. Repeated challenge with virulent Newcastle Disease Virus does not decrease the efficacy of vaccines. Avian Diseases. 61(2):245-249. doi:10.1637/11555-120816-ResNote.1. https://doi.org/10.1637/11555-120816-ResNote.1

Wajid, A., Dimitrov, K.M., Wasim, M., Rehmani, S., Basharat, A., Bibi, T., Arif, S., Yaqub, T., Tayyab, M., Ababneh, M., Sharma, P., Miller, P.J., Afonso, C.L. 2017. Repeated isolation of virulent Newcastle disease viruses in poultry and captive non-poultry avian species in Pakistan from 2011 to 2016. Preventive Veterinary Medicine. 142:1-6. https://doi.org/10.1016/j.prevetmed.2017.04.010.

Susta, L., Ying, H., Hutcheson, J.M., Lu, Y., West, F.D., Stice, S.L., Yu, P., Abdo, Z., Afonso, C.L. 2016. Derivation of chicken induced pluripotent stem cells tolerant to Newcastle disease virus-induced lysis through multiple rounds of infection. Virology Journal. 13:205. doi:10.1186/s12985-016-0659-3. https://doi.org/10.1186/s12985-016-0659-3.

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Non-antibiotic Strategies to Control Enteric Diseases of Poultry; Hyun Lillehoj (P) and Charles Li

Gu, C., Lillehoj, H.S., Sun, Z., Lee, Y., Zhao, H., Xiangyu, Z., Yan, X., Lin, S., Liu, L., Li, C.Z. 2019. Characterization of virulent netB+/tpeI+ clostridium perfringens strains from necrotic-enteritis-affected broiler chicken farms. Avian Diseases. 63(3):461-467. https://doi.org/10.1637/11973-092018.1

Kim, W., Lillehoj, H.S., Min, W. 2019. Indole treatment alleviates intestinal tissue damage induced by chicken coccidiosis through activation of the aryl hydrocarbon receptor. Frontiers in Immunology. https://doi.org/10.3389/fimmu.2019.00560

Li, C.Z., Yan, X., Lillehoj, H.S., Gu, C., Sun, Z., Oh, S., Lee, Y., Xianyu, Z., Zhao, H., Liu, L. 2019. Eimeria maxima-induced transcriptional changes in the chicken intestine. Frontiers in Immunology. 12:285. https://doi.org/10.1186/s13071-019-3534-4

Panebra, A., Lillehoj, H.S. 2019. Eimeria tenella elongation factor-1a coadministered with chicken IL-7 DNA vaccine emulsified in montanide gel 01 adjuvant enhanced the immune response to E. acervuina infection in broiler chickens. Avian Diseases. 63:342-350. https://doi.org/10.1637/11976-092418-Reg.1

Troung, A.D., Hong, Y., Lee, J., Lee, K., Lillehoj, H.S., Hong, Y. 2019. Chicken novel leukocyte immunoglobulin-like receptor subfamilies B1 and B3 are transcriptional regulators of MHC class I genes and signaling pathways. Molecular Immunology. 32:614-620. https://doi.org/10.5713/ajas.18.0561

Kurt, T., Wong, N., Fowler, H., Gay, C., Lillehoj, H., Plummer, P., Scott, H.M. and Hoelzer, K., 2019. Strategic priorities for research on antibiotic alternatives in animal agriculture-Results from an expert workshop. Frontiers in Veterinary Science, 6. P.429. https://doi.org/10.3389/fvets.2019.00429.

Oh, S., Lillehoj, H.S., Lee, Y., Bravo, D. and Lillehoj, E., 2019. Dietary antibiotic growth promoters down-regulate intestinal inflammatory cytokine expression in chickens challenged with LPS or co-infected with Eimeria maxima and Clostridium perfringens. Frontiers in Veterinary Science, 6, p.420. https://doi.org/10.3389/fvets.2019.00420.

Kim, W.H., Chaudhari, A.A. and Lillehoj, H.S., 2019. Involvement of T Cell Immunity in Avian Coccidiosis. Frontiers in Immunology, 10, p.2732. https://doi.org/10.3389/fimmu.2019.02732.

Pham, T.T., Ban, J., Lee, K., Hong, Y., Lee, J., Truong, A.D., Lillehoj, H.S. and Hong, Y.H., 2020. MicroRNA gga-miR-10a-mediated transcriptional regulation of the immune genes in necrotic enteritis afflicted chickens. Developmental & Comparative Immunology, 102, p.103472. https://doi.org/10.1016/j.dci.2019.103472.

Truong, A.D., Hong, Y., Tran, H.T.T., Dang, H.V., Nguyen, V.K., Pham, T.T., Lillehoj, H.S. and Hong, Y.H., 2019. Characterization and functional analyses of novel chicken leukocyte immunoglobulin-like receptor subfamily B members 4 and 5. Poultry science, 98(12), pp.6989-7002. https://doi.org/10.3382/ps/pez442.

Panebra, A. and Lillehoj, H.S., 2019. Eimeria tenella Elongation Factor-1α (EF-1α) Coadministered with Chicken IL-7 (chIL-7) DNA Vaccine Emulsified in Montanide Gel 01 Adjuvant Enhanced the Immune Response to E. acervulina Infection in Broiler Chickens. Avian Diseases, 63(2), pp.342-350. https://doi.org/10.1637/11976-092418-Reg.1.

Li, C., Yan, X., Lillehoj, H.S., Oh, S., Liu, L., Sun, Z., Gu, C., Lee, Y., Xianyu, Z. and Zhao, H., 2019. Eimeria maxima-induced transcriptional changes in the cecal mucosa of broiler chickens. Parasites & vectors, 12(1), p.285. https://doi.org/10.1186/s13071-019-3534-4.

de Leeuw, E.P., Lee, S.H., Kim, W.H., Kwasny, S.M., Opperman, T.J. and Lillehoj, H.S., 2018. Pleiotropic anti-infective effects of Defensin-derived antimicrobial compounds. Avian diseases, 62(4), pp.381-387. https://doi.org/10.1637/11912-061118-Reg.1.

Hong, Y., Truong, A.D., Lee, J., Lee, K., Kim, G.B., Heo, K.N., Lillehoj, H.S. and Hong, Y.H., 2019. Identification of duck liver-expressed antimicrobial peptide 2 and characterization of its bactericidal activity. Asian-Australasian Journal of Animal Sciences, 32(7), p.1052. https://doi.org/10.5713/ajas.18.0571

Afrin, F., Fernandez, C., Flores, R., Kim, W.H., Jeong, J., Chang, H., Kim, S., Lillehoj, H.S., Ming, W. 2018. Down-regulation of common cytokine receptor gamma chain inhibits inflammatory responses in macrophages stimulated with Riemerella anatipestifer. Developmental and Comparative Immunology. 81:225-234. https://doi.org/10.1016/j.dci.2017.12.009

Chaudhari, A., Kim, W., Lillehoj, H.S. 2018. Interleukin-4 (IL-4) may regulate alternative activation of macrophage-like cells in chickens: a sequential study using novel and specific neutralizing monoclonal antibodies against chicken IL-4. Developmental and Comparative Immunology. 205:72-82. https://doi.org/10.1016/j.vetimm.2018.10.011

Erik P. H. de Leeuw, Sung Hyen Lee, Woo H. Kim, Steven M. Kwasny, Timothy J. Opperman, and Hyun S. Lillehoj. 2018. Pleiotropic Anti-Infective Effects of Defensin-Derived Antimicrobial Compounds. Avian Diseases: December 2018, Vol. 62, No. 4, pp. 381-387. https://doi.org/10.1637/11912-061118-Reg.1

Fasina, Y.O., Lillehoj, H.S. 2018. Characterization of intestinal immune response to clostridium perfringens infection in broiler chickens characterization of intestinal immune tesponse to clostridium perfringens infection in broiler chickens. Poultry Science. 98:188-198. https://doi.org/10.3382/ps/pey390

Fernandez, C.P., Afrin, F., Flores, R.A., Kim, W.H., Jeong, J., Kim, W., Lillehoj, H.S., Min, W. 2018. Identification of duck IL-4 and its inhibitory effect on IL-17A expression in R. anatipestifer-stimulated splenic lymphocytes. Molecular Immunology. 95:20-29. https://doi.org/10.1016/j.molimm.2018.01.009

Gadde, U.D., Oh, S., Lillehoj, H.S., Lillehoj, E.P. 2018. Antibiotic growth promoter-induced changes in the chicken intestine: A metabolomics analysis of virginiamycin and bacitracin methylene disalicylate. Scientific Reports. 8(1):3592. https://doi.org/10.1038/s41598-018-22004-6

Grant, A., Gay, C.G., Lillehoj, H.S. 2018. Bacillus spp. as direct-fed microbial antibiotic alternatives to enhance poultry growth, immunity and intestinal health. Avian Pathology. 2:1-13. https://doi.org/10.1080/03079457.2018.1464117

Kim, W., Lillehoj, H.S. 2018. Immunity, immunomodulation, and antibiotic alternatives to maximize the genetic potential of poultry for growth and disease response. Animal Feed Science and Technology. 250:41-50. https://doi.org/10.1016/j.anifeedsci.2018.09.016

Hong, Y., Truong, A.D., Lee, J., Lee, K., Kim, G., Heo, K., Lillehoj, H.S., Hong, Y.H. 2018. Identification of duck liver-expressed antimicrobial peptide 2 and characterization of its bactericidal activity. Journal of Animal Science. 32(7): 1899-1908. https://doi.org/10.5713/ajas.18.0571

Lee, Y., Kim, W., Lee, S., Lillehoj, H.S. 2018. Detection of chicken interleukin-10 production in epithelial cells and necrotic enteritis induced by Clostridium perfringens using capture-ELISA. Veterinary Immunology and Immunopathology. https://doi.org/10.1016/j.vetimm.2018.10.001

Lee, Y., Lee, S., Gadde, U.D., Oh, S., Lee, S., Lillehoj, H.S. 2018. Allium hookeri supplementation improves intestinal immune response against necrotic enteritis. Poultry Science. 97(6):1899-1908. https://doi.org/10.3382/ps/pey031

Li, C.Z., Yan, X., Lillehoj, H.S. 2018. Complete genome sequences of Clostridium perfringens strain Del1 isolated from chickens affected by necrotic enteritis. Gut Pathogens. 9:69. https://doi.org/10.1186/s13099-017-0217-6

Oh, S., Gadde, U.D., Bravo, D., Lillehoj, E.P., Lillehoj, H.S. 2018. Growth promoting and anti-oxidant effects of magnolia bark extract in chicken uninfected or co-infected with clostridium perfringens and eimeria maxima as an experimental model of necrotic enteritis. Curr Dev Nutr. 2018 Jan 30;2(4):nzy009. doi: 10.1093/cdn/nzy009. https://doi.org/10.1093/cdn/nzy009

Rengaraj, D., Truong, A., Lillehoj, H.S., Han, J., Hong, Y. 2018. Expression and regulation of avian beta-defensin 8 protein in immune tissues and cell lines of chickens. Asian-Australasian Journal of Animal Sciences. https://doi.org/10.5713/ajas.17.0836

Troung, A.D., Hong, Y., Lee, J., Lee, K., Kil, D., Lillehoj, H.S., Hong, Y. 2018. Interlaeukin-34 regulates Th1 and Th17 cytokine production by activating multiple signaling pathways through CSF-1R in chicken cell lines. Oncotarget. 19(9):2710 E1665. https://doi.org/10.3390/ijms19061665

Truong, A., Rengaraj, D., Hong, Y., Tran, H., Dang, H., Nguyen, V., Lillehoj, H.S., Hong, Y. 2018. Leukocyte Immunoglobulin-like receptors A2 and A6 are expressed in macrophages and modulate cytokine production by activating multiple signaling pathways. International Journal of Molecular Science. https://doi.org/10.3390/ijms19092710

Fernandez, C.P., Afrin, F., Flores, R.A., Kim, W.H., Jeong, J., Kim, S., Chang, H.H., Lillehoj, H.S., Min, W. 2017. Down-regulation of inflammatory cytokines by berberine attenuates Riemerella anatipestifer infection in ducks. Developmental and Comparative Immunology. 77:121-127. https://doi.org/10.1016/j.dci.2017.07.027

Gadde, U., Oh, S.T., Lee, Y.S., Lillehoj, H.S. 2017. Dietary Bacillus subtilis-based direct-fed microbials alleviate LPS-induced intestinal immunological stress and improve intestinal barrier gene expression in commercial broiler chickens. Research in Veterinary Science. 114:236-243. https://doi.org/10.1016/j.rvsc.2017.05.004

Gadde, U.D., Lee, Y.S., Oh, S., Lillehoj, H.S. 2017. The effects of direct-fed microbial supplementation, as an alternative to antibiotics, on growth performance, intestinal immune status and epithelial barrier gene expression in broiler chickens. Probiotics and Antimicrobial Proteins. 10.1007/s12602-017-9275-9. https://doi.org/10.1007/s12602-017-9275-9

Li, C.Z., Liu, L., Zhenchao, Z., Shuai, W. 2017. Immunoproteomic analysis of proteins from unsporulated Oocysts of Eimeria tenella in MALDI TOF/TOF tandem mass spectrometry. Parasite. 24:48. https://doi.org/10.1051/parasite/2017047

Li, C.Z., Yan, X., Lillehoj, H.S. 2017. Complete genome sequence of Clostridium perfringens LLY_N11, a necrotic enteritis-inducing strain isolated from a healthy chicken intestine. Genome Announcements. 5:44. https://doi.org/10.1128/genomeA.01225-17

Lin, R.Q., Lillehoj, H.S., Lee, S.K., Oh, .S, Panebra, A., Lillehoj, E.P. 2017. Vaccination with Eimeria tenella Elongation Factor-1a Recombinant Protein Induces Protective Immunity against E. tenella and E. maxima Infections. Veterinary Parasitology. 243:79-84. https://doi.org/10.1016/j.vetpar.2017.06.003

Truong, A.D., Hoang, C., Hong, Y., Lee, J., Lee, K., Lillehoj, H.S., Hong, Y.H. 2017. Dataset on characterization of recombinant interleukin-23alpha, IL-12p40 and IL-23 complex protein, which activates JAK-STAT signaling pathway in chicken cell line using immunocytochemical. Data in Brief. 16:799-805. https://doi.org/10.1016/j.dib.2017.12.008

Truong, A.D., Hoang, C.T., Hong, Y., Lee, J., Lee, K., Lillehoj, H.S., Hong, Y.H. 2017. Functional analyses of the interaction of chicken interleukin 23 subunit p19 with IL-12 subunit p40 to form the IL-23 complex. Molecular Immunology. 92:54-67. https://doi.org/10.1016/j.molimm.2017.09.019

Truong, A.D., Rengaraj, D., Hong, Y., Hoang, C.T., Hong, Y.H., Lillehoj, H.S. 2017. Differentially expressed JAK-STAT signaling pathway genes and target microRNAs in the spleen of necrotic enteritis-afflicted chicken lines. Research in Veterinary Science. 115:235-243. https://doi.org/10.1016/j.rvsc.2017.05.018

Lee, Y., Lee, S., Han, H., Gadde, D., Oh, S., Lillehoj, H.S. 2018. Effect of dietary Allium hookeri on growth performance and anti-oxidant activity in young broiler chicken. Research in Veterinary Science. 11()118:345-350. https://doi.org/10.1016/j.rvsc.2018.03.007

Lillehoj, H., Liu, Y., Calsamiglia, S., Fernandez-Miyakawa, M.E., Chi, F., Cravens, R.L., Oh, S. and Gay, C.G. 2018. Phytochemicals as antibiotic alternatives to promote growth and enhance host health. Veterinary research, 49(1), p.76. https://doi.org/10.1186/s13567-018-0562-6

Gadde, U.D., Oh, S., Lillehoj, H.S. and Lillehoj, E.P. 2018. Antibiotic growth promoters virginiamycin and bacitracin methylene disalicylate alter the chicken intestinal metabolome. Scientific reports, 8(1), p.3592. https://doi.org/10.1038/s41598-018-22004-6

Gadde, U., Kim, W.H., Oh, S.T. and Lillehoj, H.S. 2017. Alternatives to antibiotics for maximizing growth performance and feed efficiency in poultry: a review. Animal health research reviews, 18(1), pp.26-45. https://doi.org/10.1017/S1466252316000207

Lee, Y., Lee, S.H., Gadde, U.D., Oh, S.T., Lee, S.J. and Lillehoj, H.S. 2017. Dietary Allium hookeri reduces inflammatory response and increases expression of intestinal tight junction proteins in LPS-induced young broiler chicken. Res Vet Sci, 112, pp.149-155.v https://doi.org/10.1016/j.rvsc.2017.03.019

Kim, W.H., Lillehoj, H.S., Lim, Y., Min, W., Sullivan, Y.B., Kakach, L. and LaBresh, J.W. 2017. Development and characterization of mouse monoclonal antibodies reactive with chicken CXCLi2. Developmental & Comparative Immunology, 72, pp.30-36. https://doi.org/10.1016/j.dci.2017.02.008.

Rengaraj, D., Truong, A., Ban, J., Lillehoj, H.S., Hong, Y. 2017. Distribution and differential expression of microRNAs in the intestinal mucosal layer of necrotic enteritis induced Fayoumi chickens. Asian-Australasian Journal of Animal Sciences. 30(7):1037-1047. https://doi.org/10.5713/ajas.16.0685

Lillehoj, H.S., Jang, S.I., Panebra, A., Lillehoj, E.P., Dupis, L., Ben, A.J. 2017. In ovo vaccination using Eimeria profilin and clostridium perfringens NetB proteins in montanide IMS adjuvant increases

protective immunity against experimentally-induced necrotic enteritis. Asian-Australasian Journal of Animal Sciences. 30(10):1478-1485. https://doi.org/10.5713/ajas.17.0053

Truong, A., Hong, Y., Park, B., Hoang, C., Lillehoj, H.S., Hong, Y. 2017. Analysis of differentially expressed genes in Necrotic Enteritis-infected Fayoumi chicken using RNA-Seq: next generation sequencing mRNA in Fayoumi chickens. Poultry Science. 54(2):121-133. https://doi.org/10.2141/jpsa.0160053

Truong, A.D., Rengaraj, D., Hong, Y., Hoang, C.T., Lillehoj, H.S., Hong, Y.H. 2017. Analysis of JAK-STAT signaling pathway genes and their microRNA in the intestinal mucosa of genetically disparate chicken lines induced with necrotic enteritis. Veterinary Immunology and Immunopathology. 187:1–9. doi: 10.1016/j.vetimm.2017.03.001. https://doi.org/10.1016/j.vetimm.2017.03.001

Li, C.Z., Lillehoj, H.S., Gadde, U.D., Oh, S., Ritter, D. 2017. Characterization of Clostridium perfringens strains isolated from healthy and necrotic enteritis-afflicted broiler chickens. Avian Diseases. 61:179-186. https://doi.org/10.1637/11507-093016-Reg.1

Kim, W.H., Min, W., Lillehoj, H.S. 2017. Immunomodulatory activity of chicken NK-lysin peptides. Scientific Reports. 7: 45099. https://doi.org/10.1038/srep45099

Kim, W.H., Min, W., Sullivan, Y.B., Kakach, L., Labresh, J.W., Lillehoj, H.S. 2017. Development and characterization of mouse monoclonal antibody reactive with chicken IL-8. Developmental and Comparative Immunology. 72:30-36. doi: 10.1016/j.dci.2017.02.008. https://doi.org/10.1016/j.dci.2017.02.008

Truong, A., Ban, J., Park, B., Hong, Y., Lillehoj, H.S. 2016. Characterization and functional analyses of a novel chicken CD8a variant X1 (CD8a1). Veterinary Immunology and Immunopathology. 94(7):2737-2751. doi: 10.2527/jas.2015-0133. https://doi.org/10.2527/jas.2015-0133

Lillehoj, H.S., Lee, S.H., Park, S.S., Jeong, M., Lim, Y., Mathis, G.F., Lumpkin, B., Chi, F., Ching, S., Cravens, R.L. 2016. Calcium Montmorillonite-based dietary supplement attenuates Necrotic Enteritis induced by Eimeria maxima and Clostridium perfringens in broilers. Poultry Science. 53:329-340. https://doi.org/10.2141/jpsa.0150182

Diaz, J.A., Kim, W.H., Fernandez, C.Y., Jeong, J., Afrin, F., Lillehoj, H.S., Kim, S., Kim, S., Dalloul, R.A., Min, W. 2016. Identification and expression analysis of duck interleukin-17D in Riemeralla anatipestifer infection. Developmental and Comparative Immunology. 61:190-197. doi: 10.1016/j.dci.2016.04.002. https://doi.org/10.1016/j.dci.2016.04.002

Del Cacho, E., Lillehoj, H.S., Gallego, M., Quilez, J., Lillehoj, E.P., Sanchez-Acevedo, C. 2016. The induction of protective immunity against experimental Eimeria tenella Infection using serum Exosomes. Veterinary Parasitology. 224:1–6. https://doi.org/10.1016/j.vetpar.2016.04.043

Lee, S.H., Lillehoj, H.S., Jang, S.I., Lee, K., Kim, D.K., Hong, Y.H., Kim, J.B., Choe, J., Bravo, D.M. 2016. Comparing immune and anti-oxidant effects of selenium sources against experimental necrotic enteritis model. World's Poultry Science Journal. 4(7): 420-427. https://www.cabdirect.org/cabdirect/abstract/20163285156

Matsubayashi, M., Minoura, C., Shintauro, K., Hiroyuki, T., Masaru, F., Haruo, M., Hiroshi, N., Lillehoj, H.S., Hatta, T., Tsuji, N., Sasai, K. 2016. Identification of Eimeria acervulina conoid antigen using

chicken monoclonal antibody. Parasitology Research. 115(11):4123-4128. https://doi.org/10.1007/s00436-016-5185-0

Lee, Y., Lee, S.H., Jeong, M.S., Kim, J.B., Jang, H.H., Choe, J.S., Kim, D.W., Lillehoj, H.S. 2016. In vitro analysis of the immunomodulating effects of Allium hookeri on lymphocytes, macrophages and tumour cells. Poultry Science. 54(2):142-148. https://doi.org/10.2141/jpsa.0160108

Fernandez, C.P., Kim, W.H., Diaz, J.A.R., Jeong, J., Afrin, F., Kim, S., Jang, H.K., Lee, B.H., Yim, D., Lillehoj, H.S. and Min, W. 2016. Upregulation of duck interleukin-17A during Riemerella anatipestifer infection. Developmental & Comparative Immunology, 63, pp.36-46. https://doi.org/10.1016/j.dci.2016.05.009.

Oh, S.T. and Lillehoj, H.S., 2016. The role of host genetic factors and host immunity in necrotic enteritis. Avian Pathology, 45(3):313-316. https://doi.org/10.1080/03079457.2016.1154503.

Rengaraj, D., Truong, A.D., Lee, S.H., Lillehoj, H.S. and Hong, Y.H. 2016. Expression analysis of cytosolic DNA-sensing pathway genes in the intestinal mucosal layer of necrotic enteritis-induced chicken. Veterinary immunology and immunopathology. 170:1-12. https://doi.org/10.1016/j.vetimm.2015.12.010.

Gadde, U., Rathinam, T. and Lillehoj, H.S. 2015. Passive immunization with hyperimmune egg-yolk IgY as prophylaxis and therapy for poultry diseases–a review. Animal health research reviews, 16(2), pp.163-176. https://doi.org/10.1017/S1466252315000195.

Kim, J.E., Lillehoj, H.S., Hong, Y.H., Kim, G.B., Lee, S.H., Lillehoj, E.P. and Bravo, D.M. 2015. Dietary Capsicum and Curcuma longa oleoresins increase intestinal microbiome and necrotic enteritis in three commercial broiler breeds. Research in veterinary science, 102, pp.150-158. https://doi.org/10.1016/j.rvsc.2015.07.022.

Truong, A.D., Hong, Y.H. and Lillehoj, H.S. 2015. RNA-seq profiles of immune related genes in the spleen of necrotic enteritis-afflicted chicken lines. Asian-Australasian journal of animal sciences, 28(10), p.1496. https://doi.org/10.5713/ajas.15.0143.

Truong, A.D., Hong, Y.H. and Lillehoj, H.S. 2015. High-throughput sequencing reveals differing immune responses in the intestinal mucosa of two inbred lines afflicted with necrotic enteritis. Veterinary immunology and immunopathology, 166(3-4), pp.116-124. https://doi.org/10.1016/j.vetimm.2015.06.008.

Kim, W.H., Fernandez, C.P., Diaz, J.A.R., Jeong, J., Kim, S., Lillehoj, H.S., Chang, H.H. and Min, W. 2015. Molecular cloning, characterization and mRNA expression of duck interleukin-17F. Veterinary immunology and immunopathology, 164(3-4), pp.194-200. https://doi.org/10.1016/j.vetimm.2015.02.007.

Xu, S., Lee, S.H., Lillehoj, H.S., Hong, Y.H. and Bravo, D. 2015. Effects of dietary selenium on host response to necrotic enteritis in young broilers. Research in veterinary science, 98, pp.66-73. https://doi.org/10.1016/j.rvsc.2014.12.004.

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8042-31000-107-00D
Molecular Approaches to Control Intestinal Parasites that Affect the Microbiome in Swine and Small Ruminants; Joseph Urban (P), Robert Li and Dante Zarlenga

Coelho, C.H., Gazzinelli-Guimaraes, P.H., Howard, J., Barnafo, E., Alani, N.A., Muratova, O., Mccormack, A., Kelnhofer, E., Urban Jr, J.F., Narum, D., Anderson, C., Langhorne, J., Nutman, T.B., Duffy, P.E. 2019. Chronic helminth infection does not impair immune response to malaria transmission blocking vaccine Pfs230D1M-EPA/Alhydrogel® in mice. Scientific Reports. 37(8):1038-1045. https://doi.org/10.1016/j.vaccine.2019.01.027

Jarjour, N.N., Schwarzkopf, E.A., Bradstreet, T.R., Shchukina, I., Lin, C., Huang, S., Lai, C., Cook, M.E., Taneja, R., Stappenbeck, T., Randolph, G.J., Artyomov, M.N., Urban Jr, J.F., Edelson, B.T. 2019. Bhlhe40 mediates tissue-specific control of macrophage proliferation in homeostasis and type 2 immunity. Nature Immunology. 20:687-700. https://doi.org/10.1038/s41590-019-0382-5

Liu, F., Li, Z., Wang, Z., Xue, C., Tang, Q., Li, R.W. 2019. Microbial co-occurrence patterns and keystone species in the gut microbial community of mice in response to stress and chondroitin sulfate disaccharide. International Journal of Molecular Sciences. 20(9):2130. https://doi.org/10.3390/ijms20092130

Oh, S., Li, C., Baldwin, R.L., Song, S., Liu, F., Li, R.W. 2019. Temporal dynamics in meta longitudinal RNA-Seq data. Scientific Reports. 9(1):763. https://doi.org/10.1038/s41598-018-37397-7

Wang, Y., Liu, F., Urban Jr, J.F., Paerewijck, O., Geldhof, P., Li, R.W. 2019. Ascaris suum infection was associated with a worm-independent reduction in microbial diversity and altered metabolic potential in the porcine gut microbiome. International Journal for Parasitology. 49(3-4):247-256. https://doi.org/10.1016/j.ijpara.2018.10.007

Zhang, R., Liu, F., Hunt, P., Li, C., Zhang, L., Ingham, A., Li, R.W. 2019. Transcriptome analysis unraveled potential mechanisms of resistance to Haemonchus contortus infection in Merino sheep populations bred for parasite resistance. Veterinary Research. 50:7. https://doi.org/10.1186/s13567-019-0622-6

Beasely, H., Bennett, H.M., Coghlan, A., Cotton, J., Doyle, S.R., Gordon, D., Harsha, B., Huckvale, T., Lomax, J., Holroyd, N., Reid, A.J., Ribeiro, D., Rinaldi, G., Shafie, M., Stanley, E., Tracey, A., Berriman, M., Hallsworth-Pepin, K., Martin, J., Ozersky, P., Rosa, B.A., Tyagi, R., Zhang, X., Mitreva, M., Laetsch, D.R., Koutsovoulos, G., Kumar, S., Kaur, G., Blaxter, M., Howe, K.L., Leach, A.R., Mutowo, P., Rawlings, N., Kuo, T., Lee, T.J., Ke, H., Tsai, I.J., Wheeler, N.J., Day, T.A., Zamanian, M., Beech, R.N., Parkinson, J., Seshadri, S.L., Kikuchi, T., Maizels, R.M., Partono, F., Babayan, S., Allen, J.E., O'Boyle, N., Wang, L., Osuna, A., Cruz-Bustos, T., Samblas, M.G., Cuellar, C., Cooper, P.J., Devaney, E., Harcus, Y., Hodgkinson, J., Bah, G., Tanya, V.N., Eberhard, M.L., Asano, K., Rodriguez, P.F., Sato, H., Gilleard, J.S., Matthews, J.B., Cook, J., Toldeo, R., Scholz, T., Schnyder, M., Allan, F., Emery, A., Olson, P.D., Rollinson, D., Castillo, E., Kalbe, M., Eom, K.S., Horak, P., Mitreva, M., Hawdon, J.M., Urban Jr, J.F., Hill, D.E., Zarlenga, D.S., Bisset, S.A., Pfarr, K., Makepeace, B., Taylor, D.W. 2018. Comparative genomics of the major parasitic worms. Nature Genetics. 5:163-174. https://doi.org/10.1038/s41588-018-0262-1

Cao, W., Li, J., Chin, Y., Xue, C., Li, R.W., Tang, Q. 2018. Transcriptomic analysis reveals effects of fucoxanthin on intestinal glucose transport. Journal of Functional Foods. 49:205-213. 10.1016/j.jff.2018.08.018. https://doi.org/10.1016/j.jff.2018.08.018

Hu, Y., Nguyen, T., Lee, A.C., Urban Jr, J.F., Miller, M.M., Zhan, B., Koch, D.J., Noon, J., Abraham, A., Fujiwara, T. 2018. Bacillus thuringiensis Cry5B protein as a new pan-hookworm cure. International Journal for Parasitology: Drug and Drug Resistance. 8:287-294. https://doi.org/10.1016/j.ijpddr.2018.05.001

Huang, Y., Mao, K., Chen, X., Sun, M., Kawabe, T., Li, W., Usher, N., Zhu, J., Urban Jr, J.F., Paul, W.E., Germain, R.N. 2018. S1P dependent interorgan trafficking of group 2 innate lymphoid cells supports host defense. Science. 359:114–119. https://doi.org/10.1126/science.aam5809

Leroux, L., Nasr, M., Valanparamabil, R., Tam, M., Rosa, B.A., Siciliani, E., Hill, D.E., Zarlenga, D.S., Jaramillo, M., Weinstock, J.V., Geary, T.G., Stevenson, M.M., Urban Jr, J.F., Makedonka, M., Jardim, A. 2018. Analysis of the Trichuris suis excretory/secretory proteins as a function of life cycle stage and their immunomodulatory properties. Scientific Reports. 8:15921. https://doi.org/10.1038/s41598-018-34174-4

Liu, F., Horton-Sparks, K., Hull, V., Li, R.W., Martinez-Cerdeno, V. 2018. The valproic acid rat model of autism present with gut bacterial dysbiosis similar to that in human autism. Molecular Autism. 9:61. https://doi.org/10.1186/s13229-018-0251-3

Lorentsen, K., Cho, J., Luo, X., Zuniga, A., Urban Jr, J.F., Zhou, L., Gharaibeh, R., Jobin, C., Kladde, M., Avram, D. 2018. Bcl11b is essential for licensing Th2 differentiation during helminth infection and allergic asthma. Nature Communications. 9(1)1679. https://doi.org/10.1038/s41467-018-04111-0

Solano Aguilar, G., Jang, S., Lakshman, S., Gupta, R., Beshah, E., Sikaroodi, M., Vinyard, B.T., Molokin, A., Gillevet, P., Urban Jr, J.F. 2018. The effect of dietary mushroom Agaricus bisporus on intestinal microbiota composition and host immunological function. Nutrients. 10(11):1721. https://doi.org/10.3390/nu10111721

Solano Aguilar, G., Shea-Donohue, T., Madden, K., Quinones, A., Beshah, E., Lakshman, S., Xie, Y., Dawson, H.D., Urban Jr, J.F. 2018. Bifidobacterium animalis subspecies lactis modulates the local immune response and glucose uptake in the small intestine of juvenile pigs infected with the parasitic nematode Ascaris suum. Gut Microbes. 19:1-15. https://doi.org/10.1080/19490976.2018.1460014

Wang, X., Liu, F., Yuan, G., Xue, C., Li, R.W., Tang, Q. 2018. Transcriptome analysis revealed anti-obesity effects of the alginate polysaccharide in high-fat diet-induced obese mice. International Journal of Biological Macromolecules. 115:861-870. https://doi.org/10.1016/j.ijbiomac.2018.04.042

Liu, F., Zhang, N., Li, Z., Wang, X., Shi, H., Xue, C., Li, R.W., Tang, Q. 2017. Chondriotin sulfate disaccharides modified the structure and function of the murine gut microbiome under healthy and stressed conditions. Scientific Reports. 7:6783. https://doi.org/10.1038/s41598-017-05860-6

Zhang, N., Mao, X., Hou, E., Wang, Y., Xue, C., Li, R.W., Tang, Q. 2017. Neoagarotetraose protects mice against intense exercise induced stress by modulating gut microbial composition and function. Molecular Nutrition and Food Research. https://doi.org/10.1002/mnfr.201600585

Kieckens, E., Rybarczyk, Li, R.W., Vanrompay, D., Cox, E. 2016. Potential immunosuppressive effects of Escherichia coli O157:H7 experimental infection on the bovine host. Biomed Central (BMC) Genomics. 17:1049. https://doi.org/10.1186/s12864-016-3374-y

8042-32000-102-00D
Evaluation of Swine Immunity and Development of Novel Immune and Genomic Intervention Strategies to Prevent and/or Treat Respiratory Diseases of Swine; Joan Lunney (P)

Chase C, Lunney JK. 2020. Swine Immune System. Chapter 16 In: Diseases of Swine (11th Edition) Editors: Zimmerman J, Karriker L, Ramirez A, Schwartz K, Stevenson G, Zhang J. John Wiley & Sons, Inc., Hoboken, NJ. Chap. 16, pp.264-290.

Hammer SE, Ho C-S, Ando A, Rogel-Gaillard C, Charles M, Tector M, Tector AJ, Lunney JK. 2019. Importance of the porcine MHC (SLA) complex in swine health and biomedical research. Annual Review of Animal Biosciences. 8: xxx. doi: 10.1146/annurev-animal-020518-115014. https://doi.org/10.1146/annurev-animal-020518-115014.
Koltes JE, Cole JB, Clemmens R, Dilger RN, Kramer LM, Lunney JK, McCue ME, McKay S, Mateescu R, Murdoch BM, Reuter R, Rexroad C, Rosa GJM, Serão NVL, White SN, Woodward-Greene MJ, Worku M, Zhang H, James Reecy J. 2019. A vision for development and utilization of high-throughput phenotyping and big data analytics in livestock. Front Genet. 10:1197. doi: 10.3389/fgene.2019.01197. https://doi.org/10.3389/fgene.2019.01197.

Kyu-Sang, L., Dong, Q., Moll, P., Vitkovska, J., Wiktorin, G., Bannister, S., Daujotyte, D., Tuggle, C.K., Lunney, J.K., Plastow, G., Dekkers, J. 2019. The effects of a globin blocker on the resolution of 3mRNA sequencing data in porcine blood. BMC Genomics. 20:741. https://doi.org/10.1016/j.rvsc.2018.02.007

Ramos, L., Obregon-Henao, A., Henao-Tamayo, M., Bowen, R., Izzo, A., Lunney, J.K., Gonzalez-Juarrero, M. 2019. Minipigs as neonatal animal model for tuberculosis vaccine efficacy testing. Veterinary Immunology and Immunopathology. 215:109884. https://doi.org/10.1016/j.vetimm.2019.109884.

Rexroad III, C.E., Vallet, J.L., Matukumalli, L.K., Ernst, C., Van Tassell, C.P., Cheng, H.H., Reecy, J., Fulton, J., Taylor, J., Lunney, J.K., Liu, J., Cockett, N., Smith, T.P., Van Eenennaam, A., Clutter, A., Telugu, B., Purcell, C., Bickhart, D.M., Blackburn, H.D., Neibergs, H., Wells, K., Boggess, M.V., Sonstegard, T. 2019. Genome to phenome: improving animal health, production, and well-being: a new USDA blueprint for animal genome research 2018–2027. Frontiers in Genetics. 10:327. https://doi.org/10.3389/fgene.2019.00327

Dawson, H.D., Lunney, J.K. 2018. Porcine cluster of differentiation (CD) Markers 2018 Update. Research in Veterinary Science. 118:199-246. https://doi.org/10.1016/j.rvsc.2018.02.007

Hess, A.S., Lunney, J.K., Abrams, S., Choi, I., Trible, B., Hess, M.K., Rowland, R., Plastow, G., Dekkers, J. 2018. Identification of factors associated with virus level in tonsils of pigs experimentally infected with Porcine Reproductive and Respiratory Syndrome virus. Journal of Animal Science. 97:536-547. https://doi.org/10.1093/jas/sky446

Dong Q, Dekkers JCM, Rowland RRR, Lunney JK, Reecy JM, Tuggle CK, Nguyen Y. 2018. The Effects of Vaccination and WUR Genotype on Blood Gene Expression Response to Co-infection with PRRSV and PCV2 in Pigs. Proceedings of the World Congress on Genetics Applied to Livestock Production. 11: 993.

Lough G, Hess AS, Hess MK, Rashidi H, Matika O, Lunney JK, Rowland RRR, Kyriazakis I, Mulder H, Dekkers JCM, Doeschl-Wilson A. 2018. Harnessing longitudinal information to identify genetic variation in tolerance of pigs to Porcine Reproductive and Respiratory Syndrome virus infection. Genet Sel Evol. 50: 50. doi: 10.1186/s12711-018-0420-z. https://doi.org/10.1186/s12711-018-0420-z.

Hess, A.S., Trible, B., Hess, M.K., Rowland, R., Lunney, J.K., Plastow, G., Dekkers, J. 2018. Genetic relationships of antibody response, viremia level and weight gain in pigs experimentally infected with porcine reproductive and respiratory syndrome virus. Journal of Animal Science. 96:3565-3581. https://doi.org/10.1093/jas/sky229

Waide, E., Tuggle, C.K., Serao, N., Schroyen, M., Hess, A., Rowland, R., Lunney, J.K., Plastow, G., Dekkers, J. 2018. Genomic prediction of piglet response to infection with one of two porcine reproductive and respiratory syndrome virus isolates. Genetics Selection Evolution. 50:3. https://doi.org/10.1186/s12711-018-0371-4

Dunkelberger, J., Serao, N., Weng, Z., Waide, E.H., Niederwerder, M., Kerrigan, M., Lunney, J.K., Rowland, R., Dekkers, J. 2017. Genomic regions associated with host response to porcine reproductive and respiratory syndrome vaccination and co-infection in nursery pigs. BMC Genomics. 18:865. https://doi.org/10.1186/s12864-017-4182-8

Lough, G., Rashidi, H., Kyriazakis, I., Dekkers, J.C., Hess, A.S., Hess, M.K., Deeb, N., Kause, A., Lunney, J.K., Rowland, R.R., Mulder, H., Doeschl-Wilson, A. 2017. Using multi-trait and random regression models to identify genetic variation in tolerance to porcine reproductive and respiratory syndrome virus. Genetics Selection Evolution. 49:37. https://doi.org/10.1186/s12711-017-0312-7

Ramos, L., Obregon-Henao, A., Henao-Tamayo, M., Bowen, R., Lunney, J.K., Gonzalez-Juarrero, M. 2017. The minipig as an animal model to study Mycobacterium tuberculosis infection and natural transmission. Tuberculosis. 106:91-98. https://doi.org/10.1016/j.tube.2017.07.003

Dunkelberger, J., Serao, N., Neiderwerder, M., Kerrigan, M., Lunney, J.K., Rowland, R., Dekkers, J. 2017. Effect of a major quantitative trait locus for porcine reproductive and respiratory syndrome (PRRS) resistance on response to co-infection with PRRS and porcine circovirus type 2b (PCV2b) in commercial pigs, with or without prior vaccination for PRRS. Journal of Animal Science. 95:584-598. https://doi.org/10.2527/jas.2016.1071

Hong, L., Han, K., Wu, K., Lunney, J.K., Ruize, L., Huang, J., Zhao, S., Yu, M. 2017. E-cadherin and its regulator ZEB2 control the development of the placental folds by modulating trophoblast cell differentiation in pigs. Reproduction. 154:765-775. https://doi.org/10.1530/REP-17-0254

Kommadath, A., Bao, H., Choi, I., Reecy, J.M., Koltes, J.E., Fritz-Waters, E., Eisley, C.J., Grant, J.R., Rowland, R.R., Tuggle, C.K., Dekkers, J.C., Lunney, J.K., Guan, L., Stothard, P., Plastow, G.S. 2017. Genetic architecture of gene expression underlying variation in host reponse to porcine reproductive and respiratory syndrome virus infection. Scientific Reports. 7:46203. doi:10.1038/srep46203. https://doi.org/10.1038/srep46203

Waide, E.H., Tuggle, C.K., Serao, N., Schroyen, M., Hess, A., Rowland, R., Lunney, J.K., Plastow, G., Dekkers, J. 2017. Genome-wide association of piglet responses to infection with one of two porcine reproductive and respiratory syndrome virus isolates. Journal of Animal Sciences. 95:16-38. https://doi.org/10.2527/jas.2016.0874

Hay el HA, Choi I, Xu L, Zhou Y, Lunney JK, Liu GE. 2017. CNV analysis of host responses to porcine reproductive and respiratory syndrome virus infection. J Genomics. 5: 58-63. doi: 10.7150/jgen.20358. http://www.jgenomics.com/v05p0058.htm.

Tuggle CK, Giuffra E, White SN, Clarke L, Zhou H, Ross PJ, Acloque H, Reecy JM, Archibald A, Boichard M, Chamberlain A, Cheng H, Crooijmans RPMA, Delany ME, Groenen MAM, Hayes B, Lunney JK, Plastow GS, Schmidt CJ, Song J, Watson M. 2016. GO-FAANG meeting: a Gathering On Functional Annotation of ANimal Genomes. Animal Genetics. 47: 528-33. doi: 10.1111/age.12466 https://doi.org/10.1111/age.12466.

Harding JCS, Ladinig A, Novakovic P, Detmer SE, Wilkinson J, Yang T, Lunney JK, Plastow GS. 2017. Novel insights of host responses and the reproductive pathophysiology of type 2 porcine reproductive and respiratory syndrome (PRRS). Vet. Micro. 209: 114-123 doi: 10.1016/j.vetmic.2017.02.019 https://doi.org/10.1016/j.vetmic.2017.02.019

Dekkers J, Rowland B, Lunney J, Plastow G. 2017. The use of host genetics to control porcine reproductive and respiratory syndrome. Vet Microbiol. 209: 107-113 doi: 10.1016/j.vetmic.2017.03.026 https://doi.org/10.1016/j.vetmic.2017.03.026.
Rowland RRR, Lunney JK. 2017. Alternative strategies for the control and elimination of PRRS. Vet Micro. 209: 1-4. doi: 10.1016/j.vetmic.2017.09.006. https://doi.org/10.1016/j.vetmic.2017.09.006.

Lunney JK, Fang Y, Ladinig A, Chen N, Li Y, Rowland RR, Renukaradhya GJ. 2016. Porcine reproductive and respiratory syndrome virus (PRRSV): Pathogenesis and Interaction with the Immune System. Annual Review of Animal Biosciences. 4: 129-154. DOI: 10.1146/annurev-animal-022114-111025 https://doi.org/10.1146/annurev-animal-022114-111025.

Wilkinson JM, Bao H, Ladinig A, Hong L, Stothard P, Lunney JK, Plastow GS, Harding JCS. 2016. Genome-wide analysis of the transcriptional response to porcine reproductive and respiratory syndrome virus infection at the maternal/fetal interface and in the fetus. BMC Genomics. 17: 383. doi: 10.1186/s12864-016-2720-4 https://doi.org/10.1186/s12864-016-2720-4.

Schroyen M, Eisley C, Koltes JE Fritz-Waters E, Choi I, Plastow GS, Guan L, Stothard P, Bao H, Kommadath A, Reecy JM, Lunney JK, Rowland RRR, Dekkers JCM, Tuggle CK. 2016. Bioinformatic analyses in early host response to Porcine Reproductive and Respiratory Syndrome virus (PRRSV) infection reveals pathway differences between pigs with alternate genotypes for a major host response QTL. BMC Genomics. 17: 196. doi: 10.1186/s12864-016-2547-z. https://doi.org/10.1186/s12864-016-2547-z.

Hess, AS, Z Islam, Hess MK, Rowland RRR, Lunney JK, Doeschl-Wilson A, Plastow GS, Dekkers JCM. 2016. Comparison of host genetic factors influencing pig response to infection with two North American isolates of porcine reproductive and respiratory syndrome virus (PRRSV) Genet Sel Evol. 48: 43. doi: 10.1186/s12711-016-0222-0. https://doi.org/10.1186/s12711-016-0222-0.

Harding J, Ladinig A, Novakovic P, Detmer S, Wilkinson J, Yang T, Lunney J, Plastow G. 2016. PRRSV and the pregnant female. Advances in Pork Production. 27: 169-180. https://www.banffpork.ca/documents/170308043831John%20Harding_PRRSV%20&%20the%20Pregnant%20Female.pdf

Wilkinson JM, Ladinig A, Bao H, Kommadath A, Stothard P, Lunney JK, Harding JCS, Plastow GS. 2016. Differences in Whole Blood Gene Expression Associated with Infection Time-Course and Extent of Fetal Mortality in a Reproductive Model of Type 2 Porcine Reproductive and Respiratory Syndrome Virus (PRRSV) Infection. PLoS One. 11: e0153615. doi: 10.1371/journal.pone.0153615. https://doi.org/10.1371/journal.pone.0153615.

Andersson L, Archibald AL, Bottema CD, Brauning R, Burgess SC, Casas E, Cheng HH, Clarke L, Couldrey C, Dalrymple BP, Elsik CG, Foissac S, Giuffra E, Groenen MA, Hayes BJ, Huang LS, Khatib H, Kijas JW, Kim H, Lunney JK, McCarthy FM, McEwan JC, Moore S, Nanduri B, Notredame C, Palti Y, Plastow GS, Reecy JM, Rohrer GA, Sarrapoulou E, Schmidt CJ, Silverstein J, Tellam RL, Tixier-Boichard M, Tosser-Klopp G, Tuggle CK, Vilkki J, White S, Zhao S, Zhou H. 2015. Coordinated international action to accelerate Genome to Phenome- The Functional Annotation of ANimal Genomes (FAANG) Project. Genome Biology. Open Letter. 16:57 doi:10.1186/s13059-015-0622-4. https://doi.org/10.1186/s13059-015-0622-4.

Ladinig A, Detmer SE, Clarke K, Ashley C, Rowland RR, Lunney JK, Harding JC. 2015. Pathogenicity of three type 2 porcine reproductive and respiratory syndrome virus strains in experimentally inoculated pregnant gilts. Virus Res. 203: 24-35. doi: 10.1016/j.virusres.2015.03.005 https://doi.org/10.1016/j.virusres.2015.03.005.

Koltes JE, Fritz-Waters E, Eisley CJ, Choi I, Bao H, Kommadath A, Serão NVL, Boddicker NJ, Abrams SM, Schroyen M, Loyd H, Tuggle CK, Plastow GS, Guan LL, Stothard P, Lunney JK, Liu P, Carpenter S, Rowland RRR, Dekkers JCM, Reecy JM. 2015. Identification of a putative quantitative trait nucleotide in Guanylate Binding Protein 5 for host response to PRRS virus infection. BMC Genomics. 16: 412. DOI: 10.1186/s12864-015-1635-9 https://doi.org/10.1186/s12864-015-1635-9.

Schroyen M, Steibel JP, Koltes JE, Choi I, Raney NE, Eisley C, Fritz-Waters, Reecy JM, Dekkers JCM, Rowland RRR, Lunney JK, Ernst CW, Tuggle CK. 2015. Whole blood microarray analysis of pigs showing extreme phenotypes after a porcine reproductive and respiratory syndrome virus infection. BMC Genomics. 16: 516. DOI: 10.1186/s12864-015-1741-8 https://doi.org/10.1186/s12864-015-1741-8.

Ladinig A, Ashley C, Detmer SE, Lunney JK, Plastow G, Harding JCS. 2015. Maternal and fetal predictors of viral load and fetal death in porcine reproductive and respiratory syndrome virus infected pregnant gilts. Vet. Res. 46: 107. DOI: 10.1186/s13567-015-0251-7 https://doi.org/10.1186/s13567-015-0251-7.

Niederwerder M, Bawa B, Serão N, Trible B, Kerrigan M, Lunney J, Dekkers JCM, Rowland RRR. 2015. Vaccination with a porcine reproductive and respiratory syndrome (PRRS) modified live virus vaccine followed by challenge with PRRSV and porcine circovirus type 2 (PCV2) protects against PRRS but enhances PCV2 replication and pathogenesis. Clin. Vacc. Immunol. 22: 1244-54. DOI: 10.1128/CVI.00434-15 https://doi.org/10.1128/CVI.00434-15.

8042-32000-105-00D
Immune, Molecular, and Ecological Approaches for Attenuating GI Nematode Infections of Ruminants; Dante Zarlenga (P) and Wenbin Tuo

Palaniyandi, S., Rajendrakumar, A.M., Periasamy, S., Goswami, R., Tuo, W., Zhu, X., Rahaman, S.O. 2020. TRPV4 is dispensable for the development of airway allergic asthma. Laboratory Investigation. 100:265-273. https://doi.org/10.1038/s41374-019-0305-y.

Pozio, E., Zarlenga, D.S. 2019. International Commission on Trichinellosis: Recommendations for genotyping Trichinella muscle stage larvae. Food and Waterborne Parasitology. 15:e00033. https://doi.org/10.1016/j.fawpar.2018.e00033.

Sharma, R., Thompson, P., Elkin, B., Mulders, R., Branigan, M., Pongracz, J., Wagner, B., Scandrett, B., Hoberg, E., Rosenthal, B.M., Jenkins, E. 2019. Trichinella pseudospiralis in a wolverine (Gulo gulo) from

the Canadian North. International Journal for Parasitology: Parasites and Wildlife. 9:274-280. https://doi.org/10.1016/j.ijppaw.2019.06.005

Aleuy, O., Ruckstuhl, K., Hoberg, E.P., Veitch, A., Simmons, N., Kutz, S. 2018. Diversity of gastrointestinal helminths in Dall's sheep and the negative association of the abomasal nematode, Marshallagia marshalli, with fitness indicators. International Journal for Parasitology. 13:3. https://doi.org/10.1371/journal.pone.0192825

Braga, M.P., Araujo, S.B., Agosta, S., Brooks, D., Hoberg, E.P., Nylin, S., Janz, N., Boeger, W.A. 2018. Host use dynamics in a heterogeneous fitness landscape generates oscillations in host range and diversification. Evolution. https://doi.org/10.1111/evo.13557

Greiman, S., Cook, J., Tkach, V., Hoberg, E.P., Menning, D., Hope, A., Sonsthagen, S., Talbot, S. 2018. Museum metabarcoding: A novel method revealing gut helminth communities of small mammals across space and time. International Journal for Parasitology. 48(13):1061-1070. https://doi.org/10.1016/j.ijpara.2018.08.001

McBurney-Lin, S., Khorram, D., Gee, S., Hoberg, E.P., Klassen-Fischer, M., Neafie, R. 2018. A new worm infiltrating the human cornea: A report of three cases. Journal of Ophthalmology. 9:124-130. doi:10.1016/j.ajoc.2018.01.013 https://doi.org/10.1016/j.ajoc.2018.01.013

Mendez, J., Sun, D., Tuo, W., Zhengguo, X. 2018. Bovine neutrophils form extracellular traps in response to the gastrointestinal parasite Ostertagia ostertagi. Scientific Reports. 8:17598. https://doi.org/10.1038/s41598-018-36070-3

Nylin, S., Agosta, S., Bensch, S., Boeger, W.A., Braga, M., Brooks, D.R., Forister, M.L., Hamback, P.A., Hoberg, E.P., Nyman, T., Schapers, A., Stigall, A. 2018. Embracing colonizations: A new paradigm for species association dynamics. Trends in Ecology and Evolution. 33(1):4-14. https://doi.org/10.1016/j.tree.2017.10.005

Zarlenga, D.S., Mitreva, M., Thompson, P., Tyagi, R., Tuo, W., Hoberg, E.P. 2018. A tale of three kingdoms: members of the Phylum Nematoda independently acquired the detoxifying enzyme cyanase through horizontal gene transfer from plants and bacteria. Parasitology. 146(4):445-452. https://doi.org/10.1017/S0031182018001701

Cook, J.A., Galbreath, K.E., Bell, K.C., Campbell, M.L., Carriere, S., Colella, J.P., Dawson, N.G., Dunnum, J.L., Eckerlin, R.P., Geriman, S.E., Fedorov, V., Haas, G.M., Haukisalmi, V., Henttonen, H., Hope, A.G., Jackson, D., Jung, T., Koehler, A., Kinsella, M., Krejsa, D., Kutz, S.J., Liphardt, S., Macdonald, S.O., Malaney, J.L., Makarikov, A., Martin, J., Mclean, B., Mulders, R., Batsaikhan, N., Talbot, S.L., Tkach, V., Tsvetkova, A., Toman, H.M., Waltari, E., Whitman, J., Hoberg, E.P. 2016. The Beringian Coevolution Project: Holistic collections of mammals and associated parasites reveal novel perspectives on evolutionary and environmental change in the north. Arctic Science. doi.org/10.1139/as-2016-0042. https://doi.org/10.1139/as-2016-0042

Cao, L., Fetterer, R.H., Qu, G., Zhang, X., Tuo, W. 2019. Neospora caninum cytoplasmic dynein LC8 light chain 2 is differentially produced by pathogenically distinct isolates and regulates the host immune response. Parasitology. 146(5):588-595. https://doi.org/10.1017/S003118201800207X.

Sharma, P., Jenkins, M.C., Zarlenga, D.S., Fetterer, R.H., Xiao, Z., Tuo, W. 2017. Characterization of an Ostertagia ostertagi annexin-like protein at different developmental stages. Parasitology Research. 116:1515-1522. https://doi.org/10.1007/s00436-017-5428-8

8042-32000-111-00D
Development of Control and Intervention Strategies for Avian Coccidiosis; Mark Jenkins (P)

Jenkins, M.C., Parker, C.C., Obrien, C.N., Ritter, D. 2019. Viable Eimeria oocysts in poultry house litter at time of chick placement. Poultry Science. https://doi.org/10.3382/ps/pez147

Jenkins, M.C., O'Brien, C.N., Parker, C.C. 2018. An optimized assay for detecting Enterocytozoon intestinalis and Encephalitozoon bieneusi in dairy calf feces using polymerase chain reaction technology. Journal of Parasitic Diseases. 43(1):75-82. https://doi.org/10.1007/s12639-018-1060-5

Jenkins, M.C., Stevens, L., Obrien, C.N., Parker, C.C., Miska, K.B., Konjufca, V. 2018. Incorporation of a recombinant Eimeria maxima IMP1 antigen into nanoparticles confers protective immunity against E. maxima challenge infection. Vaccine. 36(8):1126-1131. https://doi.org/10.1016/j.vaccine.2017.11.014

Su, S., Miska, K.B., Fetterer, R., Jenkins, M.C., Lamont, S., Wong, E. 2018. Differential expression of intestinal nutrient transporters and host defense peptides in Eimeria maxima-infected Fayoumi and Ross chickens. Poultry Science. 97:4392-4400. https://doi.org/10.3382/ps/pey286

Dubey, J.P., Jenkins, M.C. 2017. Re-evaluation of the life cycle of Eimeria maxima Tyzzer, 1929 in chickens (Gallus domesticus). Parasitology. https://doi.org/10.1017/S0031182017002153

Jenkins, M.C., Parker, C.C., Ritter, D. 2017. Eimeria oocyst concentrations and species composition in litter from commercial broiler farms during anticoccidial drug or live Eimeria oocyst vaccine control programs. Avian Diseases. 61:214-220. https://doi.org/10.1637/11578-010317-Reg.1

Jenkins, M.C., Dubey, J.P., Fetterer, R.H., Miska, K.B. 2017. Differences in fecundity of Eimeria maxima strains exhibiting different levels of pathogenicity in its avian host. Veterinary Parasitology. 236:1-6. https://doi.org/10.1016/j.vetpar.2017.01.009

Park, M., Kim, S., Fetterer, R.H., Dalloul, R.A. 2016. Functional characterization of the turkey macrophage migration inhibitory factor. Developmental and Comparative Immunology. 61:198-207. https://doi.org/10.1016/j.dci.2016.04.005

Djemai, S., Mekroud, A., Jenkins, M.C. 2016. Evaluation of ionophore sensitivity of Eimeria acervulina and Eimeria maxima isolated from the Algerian to Jijel province poultry farms. Veterinary Parasitology. 224:77-81. https://doi.org/10.1016/j.vetpar.2016.04.040

Jenkins, M.C., Obrien, C.N., Fetterer, R., Santin, M. 2015. RT-PCR specific for Cryspovirus is a highly sensitive method for detecting Cryptosporidium parvum oocysts. Food and Waterborne Parasitology. doi: 10.1016/j.fawpar.2015.11.001. https://doi.org/10.1016/j.fawpar.2015.11.001

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Genomic Intervention Strategies to Prevent and/or Treat Respiratory Diseases of Ruminants; Carol Chitko-McKown (P), Aspen Workman, Gregory Harhay, Michael Clawson and Michael Heaton

Heaton, M.P., Bassett, A.S., Whitman, K.J., Krafsur, G.M., Lee, S., Carlson, J.M., Clark, H.J., Smith, H.R., Pelster, M.C., Basnayake, V., Grotelueschen, D.M., Vander Ley, B.L. 2019. Evaluation of EPAS1 variants for association with bovine congestive heart failure. F1000Research. 8:1189. https://doi.org/10.12688/f1000research.19951.1

Workman, A.M., Kuehn, L.A., McDaneld, T.G., Clawson, M.L., Loy, J.D. 2019. Longitudinal study of humoral immunity to bovine coronavirus, virus shedding, and treatment for bovine respiratory disease in pre-weaned beef calves. BioMed Central (BMC) Veterinary Research. 15:161. https://doi.org/10.1186/s12917-019-1887-8

Yaman, Y., Keles, M., Aymaz, R., Sevim, S., Sezenler, T., Onaldi, A.T., Kaptan, C., Baskurt, A., Koncagul, S., Oner, Y., Ozturk, E., Iriadam, M., Un, C., Heaton, M.P. 2019. Association of TMEM154 variants with visna/maedi virus infection in Turkish sheep. Small Ruminant Research. 177:61-67. https://doi.org/10.1016/j.smallrumres.2019.06.006

Workman, A.M., Chitko-McKown, C.G., Smith, T.P.L., Bennett, G.L., Kalbfleisch, T.S., Basnayake, V., Heaton, M.P. 2018. A bovine CD18 signal peptide variant with increased binding activity to Mannheimia hemolytica leukotoxin. F1000Research. 7:1985. https://doi.org/10.12688/f1000research.17187.1.

Harhay, G.P., Harhay, D.M., Bono, J.L., Smith, T.P.L., Capik, S.F., DeDonder, K.D., Apley, M.D., Lubbers, B.V., White, B.J., Larson, R.L. 2018. Closed genome sequences and antibiograms of 16 pasteurella multocida isolates from bovine respiratory disease complex cases and apparently healthy controls. MICROBIOLOGY RESOURCE ANNOUNCEMENTS. 7(11):e00976-18. https://doi.org/10.1128/MRA.00976-18.

Workman, A.M., Clawson, M.L., Heaton, M.P., Dickey, A.M. 2018. First complete genome sequence of a genotype A2, subgroup 4 small ruminant lentivirus. MICROBIOLOGY RESOURCE ANNOUNCEMENTS. 7(19):e01337-18. https://doi.org/10.1128/MRA.01337-18.

Robbins, K., Dickey, A.M., Clawson, M.L., Loy, J.D. 2018. Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry identification of Moraxella bovoculi and Moraxella bovis isolates from cattle. Journal of Veterinary Diagnostic Investigation. p. 1-4. Available: jvdi.sagepub.com. https://doi.org/10.1177/1040638718789725.

Loy, J.D., Dickey, A.M., Clawson, M.L. 2018. Complete genome sequence of Moraxella bovis strain Epp63, an etiologic agent of infectious bovine keratoconjunctivitis. Microbiology Resource Announcements. 7(8):1-2. https://doi.org/10.1128/MRA.01004-18.

Dickey, A.M., Schuller, G., Loy, J.D., Clawson, M.L. 2018. Whole genome sequencing of Moraxella bovoculi reveals high genetic diversity and evidence for interspecies recombination at multiple loci. PLoS One. 13(12):e0209113. https://doi.org/10.1371/journal.pone.0209113.

Kalbfleisch, T.S., Murdoch, B.M., Smith, T.P.L., Murdoch, J.D., Heaton, M.P., Mckay, S.D. 2018. A SNP resource for studying North American moose. F1000Research. 7(40):1-17. https://doi.org/10.12688/f1000research.13501.1.

Krafsur, G.M., Neary, J.M., Garry, F., Holt, T., Gould, D.H., Mason, G.L., Thomas, M.G., Enns, R.M., Tuder, R.M., Heaton, M.P., Brown, R.D., Stenmark, K.R. 2018. Cardiopulmonary remodeling in fattened beef cattle: A naturally occurring large animal model of obesity-associated Pulmonary hypertension with left heart disease. Pulmonary Circulation. 9(1):1-13. https://doi.org/10.1177/2045894018796804.

Workman, A.M., Zhu, L., Keel, B.N., Smith, T.P.L., Jones, C. 2018. The Wnt signaling pathway is differentially expressed during the bovine herpesvirus 1 latency- reactivation cycle: evidence that two protein kinases associated with neuronal survival (Akt3 and bone morphogenetic protein receptor 2) are expressed at higher levels during latency. Journal of Virology. 92:e01937-17. https://doi.org/10.1128/JVI.01937-17

Sun, H., Workman, A., Osorio, F.A., Steffen, D., Vu, H.L.X. 2017. Development of a broadly protective modified-live virus vaccine candidate against porcine reproductive and respiratory syndrome virus. Vaccine. 36:66-73. https://doi.org/10.1016/j.vaccine.2017.11.028.

Workman, A.M., Dickey, A.M., Heaton, M.P., Clawson, M.L., Smith, T.P.L. 2017. Complete genome sequences of two genotype A2 small ruminant lentiviruses isolated from infected U.S. sheep. Genome Announcements. 5(13):e00109-17. https://doi.org/10.1128/genomeA.00109-17

Sanchez, E.G., Riera, E., Nogal, M., Gallardo, C., Fernandez, P., Bello-Morales, R., Lopez-Guerrero, J., Chitko-McKown, C.G., Richt, J.A., Revilla, Y. 2017. Phenotyping and susceptibility of established porcine cells lines to African swine fever virus infection and viral production. Scientific Reports. 7:10369. https://doi.org/10.1038/s41598-017-09948-x.

Heaton, M.P., Smith, T.P.L., Freking, B.A., Workman, A.M., Bennett, G.L., Carnahan, J.K., Kalbfleisch, T.S. 2017. Using sheep genomes from diverse U.S. breeds to identify missense variants in genes affecting fecundity. F1000Research. 6:1303. https://doi.org/10.12688/f1000research.12216.1.

Harhay, G.P., Harhay, D.M., Bono, J.L., Smith, T.P.L., Capik, S.F., DeDonder, K.D., Apley, M.D., Lubbers, B.V., White, B.J., Larson, R.L. 2017. Closed genome sequences of seven histophilus somni isolates from beef calves with bovine respiratory disease complex. Genome Announcements. 5(40):e01099-17. https://doi.org/10.1128/genomeA.01099- 17.

Clawson, M.L., Murray, R.W., Sweeney, M.T., Apley, M.D., DeDonder, K.D., Capik, S.F., Larson, R.L., Lubbers, B.V., White, B.J., Kalbfleisch, T.S., Schuller, G., Dickey, A.M., Harhay, G.P., Heaton, M.P., Capik, S.F., White, B.J., Lubbers, B.V., Apley, M.D., DeDonder, K.D., Larson, R.L., Harhay, G.P., Chitko-McKown, C.G., Harhay, D.M., Kalbfleisch, T.S., Schuller, G., Clawson, M.L. 2017. Comparison of the diagnostic performance of bacterial culture of nasopharyngeal swab and bronchoalveolar lavage fluid samples obtained from calves with bovine respiratory disease. American Journal of Veterinary Research. 78(3):350-358. https://doi.org/10.2460/ajvr.78.3.350

Workman, A.M., Kuehn, L.A., McDaneld, T.G., Clawson, M.L., Chitko-McKown, C.G., Loy, J.D. 2017. Evaluation of the effect of serum antibody abundance against bovine coronavirus on bovine coronavirus shedding and risk of respiratory tract disease in beef calves from birth through the first five weeks in a feedlot. American Journal of Veterinary Research. 78(9):1065-1076. https://doi.org/10.2460/ajvr.78.9.1065

Zhu, L., Workman, A.M., Jones, C. 2017. Potential role for a B-catenin coactivator (high mobility group AT-hook 1 protein) during the latency-reactivation cycle of bovine herpesvirus 1. Journal of Virology. 91(5):e02132-16. https://doi.org/10.1128/JVI.02132-16

Loy, J.D., Clawson, M.L. 2017. Rapid typing of Mannheimia haemolytica major genotypes 1 and 2 using MALDI-TOF mass spectrometry. Journal of Microbiological Methods. 136:30-31. https://doi.org/10.1016/j.mimet.2017.03.002

Clawson, M.L., Murray, R.W., Sweeney, M.T., Apley, M.D., DeDonder, K.D., Capik, S.F., Larson, R.L., Lubbers, B.V., White, B.J., Kalbfleisch, T.S., Schuller, G., Dickey, A.M., Harhay, G.P., Heaton, M.P., Chitko-McKown, C.G., Brichta-Harhay, D.M., Bono, J.L., Smith, T.P.L. 2016. Genomic signatures of Mannheimia haemolytica that associate with the lungs of cattle with respiratory disease, an integrative conjugative element, and antibiotic resistance genes. BMC Genomics. 17:982. doi: 10.1186/s12864-016-3316-8. https://doi.org/10.1186/s12864-016-3316-8.

Dickey, A.M., Loy, J.D., Bono, J.L., Smith, T.P.L., Apley, M.D., Lubbers, B.V., DeDonder, K.D., Capik, S.F., Larson, R.L., White, B.J., Blom, J., Chitko-McKown, C.G., Clawson, M.L. 2016. Large genomic differences between Moraxella bovoculi isolates acquired from the eyes of cattle with conjunctivitis versus the deep nasopharynx of asymptomatic cattle. Veterinary Research. 47:31. https://doi.org/10.1186/s13567-016-0316-2.

Heaton, M.P., Smith, T.P.L., Carnahan, J.K., Basnayake, V., Qiu, J., Simpson, B., Kalbfleisch, T.S. 2016. Using diverse U.S. beef cattle genomes to identify missense mutations in EPAS1, a gene associated with pulmonary hypertension. F1000Research. 5:2003. doi:10.12688/f1000research.9254.2 https://f1000research.com/articles/5-2003/v2.

DeDonder, K.D., Harhay, D.M., Apley, M.D., Lubbers, B.V., Clawson, M.L., Schuller, G., Harhay, G.P., White, B.J., Larson, R.L., Capik, S.F., Riviere, J.E., Kalbfleisch, T., Tessman, R.K. 2016. Observations on macrolide resistance and susceptibility testing performance in field isolates collected from clinical bovine respiratory disease cases. Veterinary Microbiology. 192:186-193. https://doi.org/10.1016/j.vetmic.2016.07.021.

Anton, B.P., Harhay, G.P., Smith, T.P.L., Blom, J., Roberts, R.J. 2016. Comparative methylome analysis of the occasional ruminant respiratory pathogen Bibersteinia trehalosi. PLoS One. 11(8): e0161499. https://doi.org/10.1371/journal.pone.0161499.

Workman, A.M., Smith, T.P., Osorio, F.A., Vu, H.L. 2016. Complete genome sequence of highly virulent porcine reproductive and respiratory syndrome virus variants that recently emerged in the United States. Genome Announcements. 4(4):e00772-16. https://doi.org/10.1128/genomeA.00772-16.

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Rift Valley Fever Pathogenesis, Epidemiology, and Control Measures; William Wilson (P), Leela Noronha, Dana Mitzel and Barbara Drolet

Endalew, A.D., Faburary, B., Trujillo, J.D., Gaudreault, N.N., Davis, A.S., Shivanna, V., Sunwoo, S.Y., Ma, W., Drolet, B.S., McVey, D.S., Morozov, I., Davis, A.S., Wilson, W.C. and Richt, J.A. Immunogenicity and efficacy of Schmallenberg virus envelope glycoprotein subunit vaccines. J. Vet. Sci. 20: e58, 2019.2019. doi: 10.4142/jvs.2019.20.e58. https://doi.org/10.4142/jvs.2019.20.e58.

Endalew, A.D., Faburary, Wilson, W.C. and Richt, J.A. Schmallenberg Disease - A Newly Emerged Culicoides-borne Viral Disease of Ruminants. Viruses. 11: e1065, 2019. doi: 10.3390/v11111065 https://doi.org/10.3390/v11111065.

Endalew, A., Morozov, I., Davis, A.S., Gaudreault, N.N., Wernike, K., Bawa, B., Ruder, M.G., Drolet, B.S., McVey, D.S., Faburay, B., Wilson, W.C., Richt, J.A. 2018. Virological and Serological Responses of Sheep and Cattle to Experimental Schmallenberg virus Infection. Vector-Borne and Zoonotic Diseases. 10.1089/vbz.2018.2297. https://www.liebertpub.com/doi/10.1089/vbz.2018.2297

Faburay, B., Wilson, W.C., Secka, A., Drolet, B.S., McVey, D.S. and Richt, J.A. Evaluation of an Indirect ELISA Based on Recombinant Baculovirus-Expressed Rift Valley Fever Virus Nucleoprotein as the Diagnostic Antigen. J. Clin. Microbiol. 57: e01058-19, 2019. doi: 10.1128/JCM.01058-19. https://doi.org/10.1128/JCM.01058-19.

Lang, Y., Li, Y., Jasperson, D.C., Henningson, J., Lee, J., Ma, J., Li, Y., Duff, M., Liu, H., Bai, D., McVey, D.S., Richt, J., Ikegami, T., Wilson, W.C., Ma, W. 2019. Identification and evaluation of antivirals for Rift Valley fever virus. Veterinary Microbiology. 230:110-116. https://doi.org/10.1016/j.vetmic.2019.01.027.

Lindahl, J., Ragan, I., Rowland, R., Wainaina, M., Mbotha, D., Wilson, W.C. 2019. A multiplex fluorescent microsphere immunoassay for increased understanding of Rift Valley fever epidemiology in ruminants in Kenya. PLOS Neglected Tropical Diseases. https://doi.org/10.1016/j.jviromet.2019.04.011.

Ragan, I., Bradshaw, K., Upreti, D., Odendaal, L., Richt, J., Trujillo, J.D., Wilson, W.C., Davis, A.S. 2019. Rift Valley Fever Viral RNA Detection by In Situ Hybridization in Formalin-Fixed, Paraffin-Embedded Tissues. Vector-Borne and Zoonotic Diseases. https://doi.org/10.1089/vbz.2018.2383.

Smith, M.R., Schirtzinger, E.E., Wilson, W.C., Davis, A.S. Rift Valley Fever Virus: Propagation, Quantification, and Storage. Current Protocols in Microbiology. 55: e92, 2019. doi: 10.1002/cpmc.92. https://doi.org/10.1002/cpmc.92

Gaudreault, N., Kim, I., Indran, S., Wilson, W.C., Richt, J. 2018. Molecular aspects of Rift Valley fever virus and the emergence of reassortants. Virus Genes. https://doi.org/10.1007/s11262-018-1611-y.

Neng, J., Li, Y., Driscoll, A., Wilson, W.C., Johnson, P. 2018. Detection of multiple pathogens in serum using silica-encapsulated nanotags in a surface-enhanced Raman scattering-based immunoassay. Journal of Agricultural and Food Chemistry. https://doi.org/10.1021/acs.jafc.8b00026

Ragan, I., Davis, S.A., Mcvey, D.S., Richt, J., Rowland, R.R., Wilson, W.C. 2018. Evaluation of Fluorescence Microsphere Immunoassay for the Detection of Antibodies to Rift Valley Fever Nucleocapsid Protein and Glycoproteins. Journal of Clinical Microbiology. 56: e01626-17. http://doi.org/10.1128/JCM.01626-17.

Upreti, D., Cernicchiaro, N., Richt, J.A., Wilson, W.C., Clavijo, A. and Davis, A.S. Evaluation of diagnostic accuracy and precision of a competitive ELISA for detection of antibodies to Rift Valley fever virus in cattle and sheep sera. J. Virol. Meth. 262: 6-11. 2018. doi: 10.1016/j.jviromet.2018.09.002. https://doi.org/10.1016/j.jviromet.2018.09.002.

Wilson, W.C., Kim, I., Trujillo, J., Sunwoo, S., Noronha, L.E., Urbaniak, K., Mcvey, D.S., Drolet, B.S., Morozov, I., Faburay, B., Schirtzinger, E.E., Koopman, T., Indran, S., Balaraman, V., Richt, J. 2018. Preliminary evaluation of the susceptibility of white-tailed deer (Odocoileus virginianus) to Rift Valley fever virus. Emerging Infectious Diseases. http://doi.org/10.3201/eid2409.180265.

Faburay, B., Labeaud, A., Mcvey, D.S., Wilson, W.C., Richt, J. 2017. Current status of rift valley fever vaccine development. Vaccines 5(3):2 e5030029 doi:10.3390/vaccines5030029. https://doi.org/10.3390/vaccines5030029

Hossain, M.M., Wilson, W.C., Rowland, R.R. 2017. Incorporation of antigens from whole cell lysates and purified virions from MP12 into fluorescence microsphere immunoassays for the detection of antibodies against Rift Valley fever virus. Virology Journal. 1:24-31. https://pdfs.semanticscholar.org/3103/fc5a247cfad1b1bd1aa8c7793ff5e5cc0494.pdf

Ikegami, T., Balogh, A., Nishiyama, S., Lokugamage, N., Saito, T.B., Morrill, J.C., Shivanna, V., Indran, S.V., Zhang, L., Smith, J.K., Perez, D., Juelich, T. L., Morozov, I., Wilson, W.C., Freiberg, A.N., and Richt, J.A. Distinct virulence of Rift Valley fever phlebovirus strains from different genetic lineages in a mouse model. PLOS one. 12: e0189250. 2017. doi: 10.1371/journal.pone.0189250. https://doi.org/10.1371/journal.pone.0189250.

Noronha, L. and Wilson, W.C. Comparison of two zoonotic viruses from the order Bunyavarales. Curr. Opinion Virol. 27: 36-41. 2017. doi: 10.1016/j.coviro.2017.10.007. https://doi.org/10.1016/j.coviro.2017.10.007.

Faburay, B., Gaudreault, N.N., Qifang, L., Davis, S.A., Shivanna, V., McVey, D.S., Ruder, M.G., Drolet, B.S., Wilson, W.C., Richt, J.A. 2016. Development of a sheep challenge model for Rift Valley fever. Virology. 489:128-140. doi:10.1016/j.virol.2015.12.003. https://doi.org/10.1016/j.virol.2015.12.003

Lang, Y., Henningson, J., Jasperson, D.C., Yonghai, L., Jingliao, M., Cao, N., Liu, H., Wilson, W.C., Richt, J.A., Ruder, M.G., Mcvey, D.S., Ma, W. 2016. Mouse model for the Rift Valley fever virus MP12 strain infection. Veterinary Microbiology. 195:70-77. https://doi.org/10.1016/j.vetmic.2016.09.009.

Faburay, B., Wilson, W.C., Gaudreault, N.N., Davis, A., Shivanna, V., Bawa, B., Sunwoo, S., Ma, W., Drolet, B.S., Morozov, I., McVey, D.S., Richt, J.A. 2016. A recombinant Rift Valley fever virus glycoprotein subunit vaccine confers full protection against Rift Valley fever challenge in sheep. Scientific Reports. 6(27719):1-12. doi:10.1038/srep27719. https://doi.org/10.1038/srep27719

Gaudreault, N.N. Indran, S.V, Bryant, P.K., Richt, J.A., and Wilson, W.C. Comparison of Rift Valley fever virus replication in North American livestock and wildlife cell lines. Frontiers in Microbiology 6: e664. 2015. doi: 10.3389/fmicb.2015.00664. https://doi.org/10.3389/fmicb.2015.00664.

Hossain, M.M., Wilson, W.C., Faburay, B., Richt, J.A., McVey, D.S., Rowland, R.R. 2016. Multiplex detection of IgG and IgM to Rift Valley fever virus nucleoprotein, nonstructural proteins, and glycoprotein in ovine and bovine. Vector-Borne and Zoonotic Diseases. 16(8):550-557. doi:10.1089/vbz.2014.1721. https://doi.org/10.1089/vbz.2014.1721

Shivanna, V., McDowell, C., Wilson, W.C. and Richt. J.A. 2016. Complete genome sequence of Rift Valley fever virus strains isolated from outbreaks in Saudi Arabia (2001) and Kenya (2006-07). Genome Announcements 4: e00926. 2016. doi: 10.1128/genomeA.00926-16. https://doi.org/10.1128/genomeA.00926-16.

Wilson, W.C., Davis, A., Gaudreault, N.N., Faburay, B., Trujillo, J.D., Shivanna, V., Sunwoo, S., Balogh, A., Endalew, A., Ma, W., Drolet, B.S., Ruder, M.G., Morozov, I., McVey, D.S., Richt, J.A. 2016. Experimental infection of calves by two genetically-distinct strains of rift valley fever virus. Viruses. 8(145):1-16. doi:10.3390/v8050145. https://doi.org/10.3390/v8050145.

Miller, M. M., Bennett, K. E., Drolet, B. S., Lindsay, R., Mecham, J. O., Reeves, W. K., Weingartl, H.M. Wilson. W. C. 2015. Evaluation of the efficacy, potential for vector transmission, and duration of immunity of mp-12, an attenuated rift valley fever virus vaccine candidate, in sheep. Clin. Vaccine Immunol., 22(8), 930–937. doi: 10.1128/CVI.00114-15. https://doi.org/10.1128/CVI.00114-15.

Wilson, W.C., Gaudreault, N.N.,1 Hossain, M.M.,1 McVey, D.S. Lesser-known bunyavirus infections. Rev. Sci. Tech. Off. Int. Epiz. 34: 419-430. 2015. doi: 10.20506/rst.34.2.236. https://doi.org/10.20506/rst.34.2.236

3020-32000-010-00D
Orbivirus Pathogenesis, Epidemiology, and Control Measures; Barbara Drolet (P), William Wilson, Dana Mitzel, Dana

Nayduch, D., V. Shankar, M.K. Mills, R. Robl, B.S. Drolet, M.G. Ruder, E. Scully, C.A. Saski. 2019. Transcriptome response of female Culicoides sonorensis biting midges (Diptera: Ceratopogonidae) to early infection with epizootic hemorrhagic disease virus (EHDV-2). Viruses 11 (4):473. https://doi.org/10.3390/v11050473.

Van Gennip, R., B.S. Drolet, P. Rozo Lopez, A. Roost, J. Boonstra, P. van Rijn. 2019. Vector competence is strongly affected by a small deletion or point mutations in bluetongue virus. Parasites & Vectors. 12(1):470. https://doi.org/10.1186/s13071-019-3722-2.

Maclachlan, N., Zientara, S., Wilson, W.C., Richt, J., Savini, G. 2019. Bluetongue and Epizootic Hemorrhagic Disease Viruses: Recent developments with these globally re-emerging infections of ruminant animals. Curr. Opinion Virol. 34:56-62. https://doi.org/10.1016/j.coviro.2018.12.005.

Mendiola, S.Y., Mills, M.K., Maki, E.C., Drolet, B.S., Wilson, W.C., Berghaus, R., Stallknecht, D.E., Breitenbach, J.E., Mcvey, D.S., Ruder, M.G. 2019. EHDV-2 infection prevalence varies in Culicoides sonorensis after feeding on infected white-tailed deer over the course of Viremia. Viruses. 11:371-385. https://doi.org/10.3390/v11040371.

Schirtzinger, E.E., Jasperson, D.C., Ruder, M.G., Stallknecht, D.E., Chase, C.C., Johnson, D.J., Ostlund, E.N., Wilson, W.C. 2019. Evaluation of 2012 US EHDV-2 outbreak isolates for genetic determinants of cattle infection. Journal of General Virology. 100:556-567. https://doi.org/10.1099/jgv.0.001221.

Schirtzinger, E.E., Ruder, M.G., Stallknecht, D.E., Wilson, W.C. 2019. Complete genome sequence of a 2016 Bluetongue virus serotype 3 isolate from Louisiana, USA. Microbiology Resource Announcements. 8:e00128-19. https://doi.org/10.1128/MRA.00128-19.

Wang, L., Lanka, S., Cassout, D., Mateus-Pinilla, N., Li, G., Wilson, W.C., Yoo, D., Frederickson, R. 2019. Inter-serotype reassortment among epizootic hemorrhagic disease viruses in the United States. Transboundary and Emerging Diseases. https://doi.org/10.1111/tbed.13257.

Lehiy, C.J., Reister-Hendricks, L.M., Ruder, M., McVey, D.S., Drolet, B.S. 2018. Physiological and immunological responses to Culicoides sonorensis blood feeding: A murine model. Parasites & Vectors. 11(1):358-370. https://doi.org/10.1186/s13071-018-2935-0.

Mills, M.K., Ruder, M.G., Nayduch, D., Michel, K., Drolet, B.S. 2017. Dynamics of epizootic hemorrhagic disease virus serotype 2 infection within the vector, Culicoides sonorensis (Diptera: Ceratopogonidae). PLoS One. https://doi.org/10.1371/journal.pone.0188865.

Schirtzinger, E.E., Jasperson, D.C., Ostlund, E.N., Johnson, D.J., Wilson, W.C. 2018. Recent US bluetongue virus serotype 3 isolated found outside of Florida indicate evidence of reassortment with co-circulating endemic serotypes. Journal of General Virology. 99: 157-168. https://doi.org/10.1099/jgv.0.000965.

Drolet, B.S., Reister-Hendricks, L.M., Podell, B.K., Breitenbach, J.E., McVey, D.S., Van Rijn, P.A., Bowen, R.A. 2016. European bluetongue serotype 8: Disease threat assessment for US sheep. Vector-Borne and Zoonotic Diseases. 16(6):400-407. http://doi.org/10.1089/vbz.2015.1924

Ruder, M.G., Stallknecht, D.E., Allison, A.B., Mead, D.G., Carter, D.L., Howerth, E.W. 2016. Host and Potential Vector Susceptibility to an Emerging Orbivirus in the United States: Epizootic Hemorrhagic Disease Virus Serotype 6. Veterinary Pathology. 53(3):574-584. https://doi.org/10.1177/0300985815610387.

Wilson, W.C., Ruder, M.G., Jasperson, D.C., Smith, T.P., Naraghi Arani, P., Lenhoff, R., Stallknecht, D.E., Valdivia-Granda, W.A., Sheron, D. 2016. Molecular evolution of epizootic hemorrhagic disease viruses in North America based on historical isolates using motif fingerprints. Virus Genes. 52:495-508. 2016. DOI 10.1007/s11262-016-1332-z https://doi.org/10.1007/s11262-016-1332-z.

Wilson, W.C., Ruder, M.G., Klement, E., Jasperson, D.C., Yadin, H., Stallknecht, D.E., Mead, D., and Howerth, E. Genetic characterization of epizootic hemorrhagic disease virus strains isolated from cattle in Israel. J. Gen. Virol. 96: 1400-1410. 2015. doi: 10.1099/vir.0.000089. https://doi.org/10.1099/vir.0.000089.

Drolet, B.S., Reister-Hendricks, L.M., Lehiy, C.J., Van Rijn, P.A., Bowen, R.A. 2015. Effect of Culicoides Sonorensis Salivary Proteins on Clinical Disease Outcome in Experimental Bluetongue Virus Serotype 8 Infection of Dorset Sheep. Veterinaria Italiana. 51(4):379-384. doi: 10.12834/VetIt.496.2398.1

Gaudreault, N.N., Jasperson, D.C., Dubovi, E.J., Johnson, D.J., Ostlund, E.N., and Wilson, W.C. Whole genome sequence analysis of recently circulating Bluetongue virus serotype 11 strains from the United States including two canine isolates. J. Vet. Diag. Invest. 27: 441-448. 2015. doi: 10.1177/1040638715585156. https://doi.org/10.1177/1040638715585156

McVey, D.S., Drolet, B.S. Ruder, M.G., Wilson, W.C., Nayduch, D., Pfannenstiel, R., Cohnstaedt, L.W., MacLachlan, N.J., Gay, C.G. 2015. Orbiviruses – A North American perspective. Vector-Borne and Zoonotic Diseases 18: 335-338. doi: 10.1089/vbz.2014.1699. https://doi.org/10.1089/vbz.2014.1699

Wilson, W.C., Gaudreault, N.N., Jasperson, D.C., Johnson, D., Ostlund, E., Chase, C.L., Ruder, M.G., Stallknecht, D.E. 2015. Molecular evolution of American field strains of bluetongue and epizootic hemorrhagic disease viruses. Veterinaria Italiana. 51(4):269-273. doi: 10.12834/VetIt.555.2627.1.

Wilson, W.C., Daniels, P., Ostlund, E.N., Johnson, D.E., Oberst, D., Hairgrove, T.B. Mediger, J., and McIntosh, M.T. Diagnostic tools for bluetongue and epizootic hemorrhagic disease viruses applicable to North American Veterinary Diagnosticians. Vector-Borne and Zoonotic Diseases 15: 364373. 2015. doi: 10.1089/vbz.2014.1702. https://doi.org/10.1089/vbz.2014.1702.

Drolet, B.S., P. van Rijn, E.W. Howerth, M. Beer, P.P. Mertens. 2015. A review of knowledge gaps for orbivirus research. Vector-Borne and Zoonotic Diseases. 15(6):339-347. https://doi.org/10.1089/vbz.2014.1701.

Ruder, M.G., T.J. Lysyk, D.E. Stallknecht, L.D. Foil, D,J, Johnson, C.C. Chase, D.A. Dargatz, and E.P.J. Gibbs. 2015. Transmission and epidemiology of bluetongue and epizootic hemorrhagic disease in North America: Current perspectives, research gaps, and future directions. Vector-Borne and Zoonotic Diseases 15(6):348-363. https://doi.org/10.1089/vbz.2014.1703.

Pfannenstiel, R.S., B.A. Mullens, M.G. Ruder, L. Zurek, L.W. Cohnstaedt, D. Nayduch. 2015. Management of North American Culicoides biting midges: Current knowledge and research needs. Vector-Borne and Zoonotic Diseases. 15(6):374-384. http://doi.org/10.1089/vbz.2014.1705.

McVey, D.S., and N.J. MacLachlan. 2015. Vaccines for prevention and bluetongue and epizootic hemorrhagic disease in livestock: A North American Perspective. Vector-Borne and Zoonotic Diseases. 15(6):385-396. http://doi.org/10.1089/vbz.2014.1698.

Feenstra, F., B.S. Drolet, J. Boonstra, and P.A. van Rijn. 2015. Non-structural protein NS3/NS3a is required for propagation of bluetongue virus in Culicoides sonorensis. Parasites & Vectors 8:476-84. https://doi.org/10.1186/s13071-015-1063-3.

3020-32000-013-00D
Ecology of Vesicular Stomatitis Virus (VSV) in North America; Barbara Drolet (P), Lee Cohnstaedt, Dana Mitzel and William Wilson

Rozo-Lopez, P., B. Londono-Renteria, B.S. Drolet. 2018. Vesicular stomatitis virus transmission: A comparison of incriminated vectors. Insects. 9:190-205. https://doi.org/10.3390/insects9040190.

3020-32000-014-00D
Japanese Encephalitis Virus Prevention and Mitigation Strategies; Leela Noronha (P), David McVey, Barbara Drolet, Dana Mitzel and William Wilson

Ferdousi T, Cohnstaedt LW, McVey DS, Scoglio CM. Understanding the survival of Zika virus in a vector interconnected sexual contact network. Sci Rep. 2019 May 10;9(1):7253. doi: 10.1038/s41598 019-43651-3. PubMed PMID: 31076660 https://doi.org/10.1038/s41598-019-43651-3.

Oliveira, A.R.S., Piaggio, J., Cohnstaedt, L.W., McVey, D.S., Cernicchiaro, N. 2019. Introduction of the Japanese Encephalitis Virus (JEV) in the United States – A Qualitative Risk Assessment. Transboundary and Emerging Diseases. 66(4):1558-1574. doi: 10.1111/tbed.13181. https://doi.org/10.1111/tbed.13181.

Moon, S.A., Cohnstaedt, L.W., McVey, D.S., Scoglio, C.M. 2019. A Spatio-Temporal Individual-Based Network Framework for West Nile Virus in the U.S.A.: Spreading Pattern of West Nile Virus. PLOS Computational Biology. 15(3): e1006875. https://doi.org/10.1371/journal.pcbi.1006875.

Park, S., Huang, Y., Lyons, A., Ayers, V., Hettenbach, S., McVey, D.S., Burton, K., Higgs, S., Vanlandingham, D. 2018. North American domestic pigs are susceptible to experimental infection with Japanese encephalitis virus. mBio. https://doi.org/10.1038/s41598-018-26208-8

Lyons, A., Huang, Y., Park, S., Ayers, V., Hettenbach, S., Higgs, S., McVey, D.S., Noronha, L.E., Hsu, W., Vanlandingham, D. 2018. Shedding of Japanese encephalitis virus in oral fluid of infected swine. Vector-Borne and Zoonotic Diseases. 10.1089/vbz.2018.2283. https://doi.org/10.1089/vbz.2018.2283.

Oliveira, A.R.S., Piaggio, J., Cohnstaedt, L.W., McVey, D.S., Cernicchiaro, N. 2018. A quantitative risk assessment (QRA) of the risk of introduction of the Japanese encephalitis virus (JEV) in the United States via infected mosquitoes transported in aircraft and cargo ships. Preventative Veterinary Medicine. https://doi.org/10.1016/j.prevetmed.2018.09.020

Oliveira, A., Etcheverry Hernandez, L., Strathe, E., McVey, D.S., Piaggio, J., Cohnstaedt, L.W., Cernicchiaro, N. 2018. Quantification of vector and host competence for Japanese encephalitis virus: a systematic review of the literature. Preventive Veterinary Medicine. https://doi.org/10.1016/j.prevetmed.2018.03.018.

Oliveira, A.R.S., Strathe, E., Etcheverry, L., Cohnstaedt, L.W., McVey, D.S., Piaggio, J., Cernicchiaro, N. 2018. Assessment of data on vector and host competence for Japanese encephalitis virus: A systematic review of the literature. Preventive Veterinary Medicine 154:71-89. https://doi.org/10.1016/j.prevetmed.2018.03.018.

Oliveira, A., Cohnstaedt, L.W., Strathe, E., Hernandez, L, McVey, D.S., Piaggio, J., Cernicchiaro, N.2017. Meta-analyses of the proportion of Japanese encephalitis virus infection in vectors and vertebrate hosts. Parasites and Vectors. 10:418-433. https://doi.org/10.1186/s13071-017-2354-7.

Vial, L., Ducheyne, E., Filatov, S., McVey, D.S., Sindryakova, I., Morgunov, S., Perez de Leon, A.A., Kolbasov, D., De Clercq, E. 2017. Spatial multi-criteria decision analysis for modelling suitable habitats of Ornithodoros soft ticks in the Western Palearctic region. Veterinary Parasitology. 249:2-16. https://doi.org/10.1016/j.vetpar.2017.10.022.

Huang YJ, Harbin JN, Hettenbach SM, Maki E, Cohnstaedt LW, Barrett AD, Higgs S, Vanlandingham DL. Susceptibility of a North American Culex quinquefasciatus to Japanese Encephalitis Virus. Vector Borne Zoonotic Dis. 2015 Nov;15(11):709-11. doi: 10.1089/vbz.2015.1821. https://doi.org/10.1089/vbz.2015.1821.

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6064-32000-012-00D
Transmission, Pathogenesis, and Control of Avian Mycoplasmosis; Jeffrey Evans (P), Joseph Purswell, Scott Branton and Spencer Leigh

Leigh, S., Collier, S., Branton, S., Evans, J., 2019. Antibody Pretreatment Does Not Impact Mycoplasma Gallisepticum Vaccination or Experimental Infection. Journal of Veterinary and Animal Research 2, 4. http://article.scholarena.com/Antibody-Pretreatment-Does-Not-Impact-Mycoplasma-Gallisepticum-Vaccination-or-Experimental-Infection.pdf

Nuthalapati, N., Evans, J.D., Taylor, R.L., Branton, S.L., Nanduri, B., Pharr, G.T. 2019. Transcriptomic analysis of early B-cell development in the chicken embryo. Poultry Science. 98(11):5342-5354. https://doi.org/10.3382/ps/pez354.

Elliott, K., Branton, S. L., J. D. Evans, and E. D. Peebles. 2019. Occurrence of horizontal transmission in layer chickens after administration of an in ovo strain F Mycoplasma gallisepticum. Poult Sci. 98(10):4492-4497. https://doi.org/10.3382/ps/pez306.

Leigh S.A., J.D. Evans, and S.L. Branton. 2019. The complete genome sequence of two Mycoplasma gallisepticum vaccine strains and one field isolate. Microbiol Resour Announc. 8(49):e01237. https://doi.org/10.1128/MRA.00485-19.

Nuthalapati, N., J.D. Evans, R. L. Taylor Jr., S.L. Branton, B. Nanduri, and G.T. Pharr. 2019. Transcriptomic analysis of early B-cell development in the chicken embryo. Poult. Sci. 98:5342-5354. https://doi.org/10.3382/ps/pez354

Leigh S.A., J.D. Evans, and S.L. Branton. 2019. The complete genome sequences of two Mycoplasma gallisepticum F-strain variants. Microbiol Resour Announc. 8(33):e00485. https://doi.org/10.1128/mra.00485-19

Purswell, J.L., S.L. Branton, and J.D. Evans. 2019. Performance of an automated whole-house spray vaccination system. JAPR. 28(1):215-220. https://doi.org/10.3382/japr/pfy060

Elliott, K., Branton, S. L., J. D. Evans, and E. D. Peebles. 2018. Early post-hatch survival and humoral immune response of layer chickens when in ovo vaccinated with strain F Mycoplasma gallisepticum. Poult Sci. 97(4):3860-3863. https://doi.org/10.3382/ps/pey282.

Leigh S.A., J.D. Evans, S.D. Collier, and S.L. Branton. 2018. The impact of vaccination route on Mycoplasma gallisepticum vaccine efficacy. Poult Sci. 97(7):3072-3075. https://doi.org/10.3382/ps/pey188.

Nuthalapati, N., Olanrewaju, H.A., Branton, S.L., Pharr, G.T. 2017. Evaluation of the growth of the bursa of fabricius in broilers reared under different light photoperiods. International Journal of Poultry Science. 16:481-485. https://doi.org/10.3923/IJPS.2017.481.485

Elliott, K., Branton, S.L., Evans, J.D., Gerard, P.D., Peebles, E.D. 2017. Layer chicken embryo survival to hatch when administered an in ovo vaccination of strain F Mycoplasma gallisepticum and locations of bacteria prevalence in the newly hatched chick. Poultry Science. 96:3879-3884. https://doi.org/10.3382/ps/pex196

Evans, J.D., Branton, S.L., Collier, S.D., Brooks, J.P., Purswell, J.L. 2017. Application of a micro-aerosolized disinfectant to clear Mycoplasma gallisepticum from contaminated facilities. Journal of Applied Poultry Research. 26(3):416-420. https://academic.oup.com/japr/article/26/3/416/3778201

Elliott, K., Branton, S. L., Evans, J.D., Gerard, P. D., Peebles, E.D. 2017. Layer chicken embryo survival to hatch when administered an in ovo vaccination of strain F Mycoplasma gallisepticum and locations of bacteria prevalence in the newly hatched chick. Poult Sci. 96(11):3879-3884. https://doi.org/10.3382/ps/pex196.

Leigh S.A., J.D. Evans, S.D. Collier, and S.L. Branton. 2017. The impact of vaccination route on Mycoplasma gallisepticum vaccine efficacy. Poult Sci. https://doi.org/10.3382/ps/pey188.

Evans, J.D., S.L. Branton, S.D. Collier, J.P. Brooks, and J.L. Purswell. 2016. Application of an aerosolized disinfectant to clear Mycoplasma gallisepticum from contaminated facilities. JAPR. 26(3):416-420. https://doi.org/10.3382/japr/pfx010.

Peebles, E.D., R. Jacob, S.L. Branton, J.D. Evans, S.A. Leigh, and P.D. Gerard. 2015. Effects of different vaccine combinations against Mycoplasma gallisepticum on the digestive and reproductive

organ characteristics of commercial egg-laying hens. Poult. Sci. 94(12):2898-2904. https://doi.org/10.3382/ps/pev269.

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Ecology of Vesicular Stomatitis Virus (VSV) in North America; Luis Rodriguez (P), James Zhu and Jonathan Arzt

Velazquez-Salinas, L., Pauszek, S.J., Barrera, J., Clark, B., Borca, M.V., Verdugo-Rodriguez, A., Arzt, J., Rodriguez, L.L. 2019. Validation of a site-specific recombination cloning technique for the rapid development of a full-length cDNA clone of a virulent field strain of vesicular stomatitis New Jersey virus. Journal of Virological Methods. Volume 265, March 2019, Pages 113-116. https://doi.org/10.1016/j.jviromet.2019.01.003

Velazquez-Salinas, L., Pauszek, S.J., Rodriguez, L.L. 2019. Complete genome sequence of a representative New Jersey vesiculovirus strain NJ03CPB from a region of endemicity in southern Mexico. Microbiology Resource Announcements. https://doi.org/10.1128/MRA.00499-19

Velazquez-Salinas, L., Pauszek, S.J., Stenfeldt, C., O'Hearn, E.S., Pacheco, J.M., Borca, M.V., Verdugo-Rodriguez, A., Arzt, J., Rodriguez, L.L. 2018. Increased virulence of an epidemic strain of vesicular stomatitis virus is associated with interference of the innate response in pigs. Frontiers in Microbiology. 9:1891. https://doi.org/10.3389/fmicb.2018.01891

Velazquez-Salinas, L., Pauszek, S.J., Verdugo-Rodriguez, A., Rodriguez, L.L. 2018. Complete genome sequences of two vesicular stomatitis New Jersey viruses representing the 2012 U.S. epidemic strain and its closest relative endemic strain from southern Mexico. Genome Announcements. https://doi.org/10.1128/genomeA.00049-18

Ramirez-Carvajal, L., Singh, N., De Los Santos, T.B., Rodriguez, L.L., Zhu, J.J., Long, C.R. 2015. Depletion of elongation initiation factor 4E binding proteins by CRISPR/Cas9 enhances the antiviral response in porcine cells. Antiviral Research. 125:8-13. doi: 10.1016/j.antiviral.2015.11.002. https://doi.org/10.1016/j.antiviral.2015.11.002

Velazquez-Salinas, L., Zarate, S., Eschbaumer, M., Lobo, F.P., Gladue, D.P., Arzt, J., Novella, I.S., Rodriguez, L.L. 2016. Selective factors associated with the evolution of codon usage in natural populations of arboviruses. PLoS One. PLoS ONE(7): e0159943. doi:10.1371/journal.pone.0159943. https://doi.org/10.1371/journal.pone.0159943

Fowler, V.L., Howson, E.L., Madi, M., Mioulet, V., Caiusi, C., Pauszek, S.J., Rodriguez, L.L., King, D. 2016. Development of a reverse transcription loop-mediated isothermal amplification assay for the detection of vesicular stomatitis New Jersey virus: use of rapid molecular assays to differentiate between vesicular disease viruses. Journal of Virological Methods. 234:123-131. doi: 10.1016/j.jviromet.2016.04.012. https://doi.org/10.1016/j.jviromet.2016.04.012

8064-32000-060-00D
Countermeasures to Control and Eradicate Foreign Animal Diseases of Swine; Manuel Borca (P), James Zhu, Luis Rodriguez, Jonathan Arzt and Douglas Gladue

Ramirez-Medina, E., Vuono, E., O'Donnell, V., Holinka, L.G., Silva, E., Rai, A., Pruitt, S., Carrillo, C., Gladue, D.P., Borca, M.V. 2019. Differential effect of the deletion of African swine fever virus virulence-associated genes in the induction of attenuation of the highly virulent Georgia strain. Virology. https://doi.org/10.3390/v11070599

Velazquez-Salinas, L., Verdugo-Rodriguez, A., Rodriguez, L.L., Borca, M.V. 2019. The role of interleukin 6 during viral infections. Frontiers in Microbiology. https://doi.org/10.3389/fmicb.2019.01057

Vuono, E., Ramirez-Medina, E., Holinka-Patterson, L.G., Baker-Branstetter, R., Borca, M.V., Gladue, D.P. 2019. Interaction of structural glycoprotein E2 of Classical Swine Fever Virus with protein phosphatase 1 catalytic subunit beta (PPP1CB). Virology. Viruses 2019, 11(4), 307. https://doi.org/10.3390/v11040307

Barrionuevo, F., Di Giacomo, S., Bucafusco, D., Ayude, A., Schammas, J., Miraglia, M.C., Capozzo, A., Borca, M.V., Perez-Filgueira, M. 2018. Systemic antibodies administered by passive immunization prevent generalization of the infection by foot-and-mouth disease virus in cattle after oronasal challenge. Virology. 518:143-151. https://doi.org/10.1016/j.virol.2018.02.012

Borca, M.V., Berggren, K., Ramirez, E., Vuono, E., Gladue, D.P. 2018. CRISPR/Cas gene editing of a large DNA virus: African Swine Fever Virus. Bio-protocol. 8(16). https://doi.org/10.21769/BioProtoc.2978

Borca, M.V., Holinka-Patterson, L.G., Berggren, K., Gladue, D.P. 2018. CRISPR-Cas9, a tool to efficiently increase the development of recombinant African swine fever viruses. Scientific Reports. 8(3154). https://doi.org/10.1038/s41598-018-21575-8

Borca, M.V., Holinka-Patterson, L.G., Ramirez-Medina, E., Vuono, E., Berggren, K., Gladue, D.P. 2018. Identification of structural glycoprotein E2 domain critical to mediate replicaton of Classical Swine Fever Virus in SK6 cells. Virology. 526:38-44. https://doi.org/10.1016/j.virol.2018.10.004

Borca, M.V., O'Donnell, V., Holinka-Patterson, L.G., Ramirez, E., Vuono, E., Berggren, K., Alfano, M., Carey, L.B., Richt, J.A., Risatti, G.R., Gladue, D.P. 2018. The L83L ORF of African swine fever virus strain Georgia encodes for a non-essential gene that interacts with host protein IL-1ß. Virus Research. 249:116-123. https://doi.org/10.1016/j.virusres.2018.03.017

Gladue, D.P., Largo, E., De La Arada, I., Aguilella, V., Alcaraz, A., Arrondo, J., Holinka-Patterson, L.G., Brocchi, E., Ramirez-Medina, E., Vuono, E., Berggren, K., Carrillo, C., Borca, M.V. 2018. Molecular characterization of the viroporin function of foot-and-mouth disease virus non-structural protein 2B. Journal of Virology. https://doi.org/10.1128/JVI.01360-18

Gladue, D.P., Largo, E., Holinka-Patterson, L.G., Ramirez-Medina, E., Vuono, E.A., Berggren, K., Risatti, G.R., Nieva, J.L., Borca, M.V. 2018. Classical swine fever virus p7 protein interacts with host protein CAMLG and regulates calcium permeability at the endoplasmic reticulum. Virus Research. 10(9):E460. https://doi.org/10.3390/v10090460

Largo, E., Gladue, D.P., Torralba, J., Aguilella, V.M., Alcaraz, A., Borca, M.V., Nieva, J.L. 2018. Mutation-induced changes of transmembrane pore size revealed by combined ion-channel conductance and single vesicle permeabilization analyses. Biochimica et Biophysica Acta. 1860:1015-1021. https://doi.org/10.1016/j.bbamem.2018.01.012

Velazquez-Salinas, L., Ramirez-Medina, E., Bracht, A.J., Hole, K., Brito, B., Gladue, D.P., Carillo, C. 2018. Phylodynamics of parapoxvirus genus in Mexico (2007–2011). Infection, Genetics and Evolution. https://doi.org/10.1016/j.meegid.2018.07.005

Borca, M.V., O'Donnell, V., Holinka-Patterson, L.G., Sanford, B., Azzinaro, P.A., Risatti, G., Gladue, D.P. 2017. Development of a fluorescent ASFV strain that retains the ability to cause disease in swine. Nature Scientific Reports. 7:46747. doi: 10.1038/srep46747. https://doi.org/10.1038/srep46747

Holinka-Patterson, L.G., O'Donnell, V., Risatti, G., Azzinaro, P.A., Arzt, J., Stenfeldt, C., Velazquez-Salinas, L., Gladue, D.P., Borca, M.V. 2017. Early protection events in swine immunized with an experimental live attenuated classical swine fever marker vaccine, FlagT4G. PLoS One. doi: 10.1371/journal.pone.0177433. https://doi.org/10.1371/journal.pone.0177433

O'Donnell, V., Risatti, G.R., Holinka-Patterson, L.G., Krug, P.W., Carlson, J., Velazquez-Salinas, L., Azzinaro, P., Gladue, D.P., Borca, M.V. 2017. Simultaneous deletion of the 9GL and UK genes from the African swine fever virus Georgia 2007 isolate offers increased safety and protection against homologous challenge. Journal of Virology. 91(1):e01760-16. https://jvi.asm.org/content/91/1/e01760-16.short

Carlson, J., O'Donnell, V., Alsfonso, M., Velazquez Salinas, L., Holinka-Patterson, L.G., Krug, P.W., Gladue, D.P., Higgs, S., Borca, M.V. 2016. Association of the host immune response with protection using a live attenuated african swine fever virus model. Viruses. 8(10):291. doi: 10.3390/v8100291. https://doi.org/10.3390/v8100291

Carlson, J., O'Donnell, V.K., Alfano, M., Velazquez-Salinas, L., Holinka-Patterson, L.G., Krug, P.W., Gladue, D.P., Higgs, S., Borca, M.V. 2016. Association of the host immune response with protection using a live attenuated African swine fever virus model . Viruses. 8(10):E291. https://doi.org/10.3390/v8100291

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Intervention Strategies to Support the Global Control and Eradication of Foot-and-Mouth Disease Virus (FMDV); Luis Rodriguez (P), Manuel Borca, Teresa De Los Santos, Aida Rieder, James Zhu and Jonathan Arzt

Arzt, J., Bertram, M.R., Vu, L.T., Pauszek, S.J., Hartwig, E.J., Smoliga, G.R., Palinski, R., Stenfeldt, C., Fish, I.H., Hoang, B.H., Dung, D.H. 2019. First detection and genome sequence of Senecavirus A in Vietnam. Emerging Infectious Diseases. https://doi.org/10.1128/MRA.01247-18

Arzt, J., Branan, M., Delgado, A., Yadav, S., Moreno-Torres, K., Tildesley, M., Stenfeldt, C. 2019. Quantitative impacts of incubation phase transmission of foot-and-mouth disease virus. Scientific Reports. https://doi.org/10.1038/s41598-019-39029-0

Arzt, J., Fish, I., Pauszek, S.J., Johnson, S.L., Chain, P.S., Rai, D.K., Rieder, A.E., Goldberg, T.L., Rodriguez, L.L., Stenfeldt, C. 2019. The evolution of a super-swarm of foot-and-mouth disease virus in cattle. Virology. https://doi.org/10.1371/journal.pone.0210847

Biswal, J.K., Ranjan, R., Subramaniam, S., Mohapatra, J.K., Sharma, M.K., Bertram, M.R., Brito, B., Rodriguez, L.L., Pattnaik, B., Arzt, J. 2019. Genetic and antigenic variation of foot-and-mouth disease virus during persistent infection in naturally infected cattle and Asian buffalo in India. bioRxiv. https://doi.org/10.1371/journal.pone.0214832

Fernandez-Sainz, I., Gavitt, T.D., Koster, M., Ramirez-Medina, E., Rodriguez, Y.Y., Wu, P., Silbart, L.K., De Los Santos, T.B., Szczepanek, S.M. 2019. The VP1 G-H loop hypervariable epitope contributes to protective immunity against foot and mouth disease virus in swine. Vaccine. https://doi.org/10.1016/j.vaccine.2019.05.019

Mwiine, F.N., Velazquez-Salinas, L., Ahmed, Z., Ochwo, S., Munsey, A., Kenney, M.A., Lutwama, J.J., Maree, F.F., Lobel, L., Perez, A.M., Rodriguez, L.L., Vanderwaal, K., Rieder, A.E. 2019. Serological and phylogenetic characterization of foot and mouth disease viruses from Uganda during cross sectional surveillance study in cattle between 2014 and 2017. Transboundary and Emerging Diseases. https://doi.org/10.1111/tbed.13249.

Omondi, G., Alkhamis, M., Obanda, V., Gakuya, F., Sangula, A., Pauszek, S.J., Perez, A., Ngulu, S., Van Aardt, R., Arzt, J., Vanderwaal, K. 2019. Phylogeographic and cross-species transmission dynamics of SAT1 and SAT2 foot-and-mouth disease virus in Eastern Africa. Molecular Ecology. https://doi.org/10.1111/mec.15125

Palinski, R., Bertram, M.R., Vu, L.T., Pauszek, S.J., Hartwig, E.J., Smoliga, G.R., Stenfeldt, C., Fish, I.H., Hoang, B.H., Dung, D.H., Arzt, J. 2019. First genome sequence of foot-and-mouth disease virus serotype O sublineage Ind2001e from Vietnam. Microbiology Resource Announcements. https://doi.org/10.1128/MRA.01424-18

Yadav, S., Stenfeldt, C., Branan, M.A., Moreno-Torres, K.I., Holmstrom, L., Delgado, A.H., Arzt, J. 2019. Parameterization of the durations of phases of foot-and-mouth disease in cattle. Frontiers in Veterinary Science. https://doi.org/10.3389/fvets.2019.00263

Arzt, J., Belsham, G.J., Lohse, L., Botner, A., Stenfeldt, C. 2018. Transmission of foot-and-mouth disease from persistently infected carrier cattle to naive cattle via transfer of oropharyngeal fluid. mSphere. 3(5):e00365-18. https://doi.org/10.1128/mSphere.00365-18

Bertram, M., Bravo De Rueda, C., Garabed, R., Dickmu Jumbo, S., Moritz, M., Pauszek, S.J., Abdoulkadiri, S., Rodriguez, L.L., Arzt, J. 2018. Molecular epidemiology of foot-and-mouth disease virus in the context of transboundary animal movement in the far north region of Cameroon. Frontiers in Veterinary Infectious Diseases. https://doi.org/10.3389/fvets.2018.00320

Bertram, M.R., Delgado, A., Pauszek, S.J., Smoliga, G.R., Brito, B.P., Stenfeldt, C., Hartwig, E.J., Dickmu-Jumbo, S., Abdoulmoumini, M., Abona Olivia, A., Salhine, R., Rodriguez, L.L., Gerabed, R., Arzt, J. 2018. Effect of vaccination on cattle subclinically infected with foot-and-mouth disease virus in Cameroon. Preventive Veterinary Medicine. 155:1-10. https://doi.org/10.1016/j.prevetmed.2018.04.003

Bertram, M.R., Vu, L.T., Pauszek, S.J., Brito, B.P., Hartwig, E.J., Smoliga, G.R., Hoang, B.H., Phuong, N.T., Stenfeldt, C., Fish, I.H., Hung, V.V., Delgado, A., VanderWaal, K., Rodriguez, L.L., Long, N.T., Dung, D.H., Arzt, J. 2018. Lack of transmission of foot-and-mouth disease virus from persistently infected cattle to naïve cattle under field conditions in Vietnam. Frontiers in Veterinary Science. https://doi.org/10.3389/fvets.2018.00174

Brito, B.P., Pauszek, S.J., Hartwig, E.J., Smoliga, G.R., Vu, L.T., Vu, P.P., Stenfeldt, C., Rodriguez, L.L., King, D.P., Knowles, N.J., Bachanek-Bankowska, K., Long, N.T., Dung, D.H., Arzt, J. 2018. A traditional evolutionary history of foot-and-mouth disease viruses in Southeast Asia challenged by analyses of non-structural protein coding sequences. Scientific Reports. 8:6472. https://doi.org/10.1038/s41598-018-24870-6

Chen, X., Gaglione, R., Leong, T., Bednor, L., De los Santos, T.B., Luk, E., Airola, M., Hollingsworth, N.M. 2018. Mek1 coordinates meiotic progression with DNA break repair by directly phosphorylating and inhibiting the yeast pachytene exit regulator Ndt80. PLoS Genetics. https://doi.org/10.1371/journal.pgen.1007832

Chung, C.J., Clavijo, A., Bounpheng, M., Uddowla, S., Sayed, A., Dancho, B., Olesen, I.C., Pacheco, J., Kamicker, B.J., Brake, D.A., Bandaranayaka-Mudiyanselage, C., Lee, S.S., Rai, D.K., Rieder, A.E. 2018. An improved, rapid competitive ELISA using a novel conserved 3B epitope for the detection of serum antibodies to foot-and-mouth disease virus. Journal of Veterinary Diagnostic Investigation. 30(5):699-707. https://doi.org/10.1177/1040638718779641

De Los Santos, T.B., Diaz-San Segundo, F., Rodriguez, L.L. 2018. The need for improved vaccines against foot-and-mouth disease. Current Opinion in Virology. 29:16-25. https://doi.org/10.1016/j.coviro.2018.02.005

Farooq, U., Ahmed, Z., Naeem, K., Bertram, M., Brito, B., Stenfeldt, C., Pauszek, S.J., Larocco, M.A., Arzt, J., Rodriguez, L.L. 2018. Characterization of naturally occurring, new and persistent subclinical foot-and-mouth disease virus infection in vaccinated Asian buffalo in Islamabad Capital Territory, Pakistan. Veterinary Research. https://doi.org/10.1111/tbed.12963

Gelkop, S., Sobarzo, A., Brangel, P., Vincke, C., Romao, E., Fedida-Metula, S., Strom, N., Ataliba, I., Mwiin, F.N., Ochwo, S., Velazquez-Salinas, L., McKendry, R.A., Muyldermans, S., Lutwama, J.J., Rieder, A.E., Yavelsky, V., Lobel, L. 2018. The development and validation of a novel nanobody-based competitive ELISA for the detection of foot and mouth disease 3ABC antibodies in cattle. Frontiers in Veterinary Science. https://doi.org/10.3389/fvets.2018.00250

Krug, P.W., Davis, T., O'Brien, C.C., LaRocco, M.A., Rodriguez, L.L. 2018. Disinfection of transboundary animal disease viruses on surfaces in pork packing plants. Veterinary Microbiology. 219:219-225. https://doi.org/10.1016/j.vetmic.2018.04.029

Medina, G.N., Diaz San-Segundo, F., Stenfeldt, C., Arzt, J., De los Santos, T.B. 2018. The different tactics of foot-and-mouth disease virus to evade innate immunity. Frontiers in Microbiology. https://doi.org/10.3389/fmicb.2018.02644.

Moreno-Torres, K.I., Brito, B.P., Branan, M.A., Rodriguez, L.L., Delgado, A.H., Stenfeldt, C., Arzt, J. 2018. Foot-and-mouth disease infection dynamics in contact-exposed pigs are determined by the estimated exposure dose. Frontiers in Veterinary Science. 5:167. https://doi.org/10.3389/fvets.2018.00167

Stenfeldt, C., Arzt, J., Pacheco, J., Gladue, D.P., Smoliga, G.R., Silva, E., Rodriguez, L.L., Borca, M.V. 2018. A partial deletion within foot-and-mouth disease virus non-structural protein 3A causes clinical attenuation in cattle but does not prevent subclinical infection. Virology. 516:115-126. https://doi.org/10.1016/j.virol.2018.01.008

Stenfeldt, C., Smoliga, G.R., Hartwig, E.J., Palinski, R., Fish, I.H., Silva, E.B., Pauszek, S.J., Arzt, J. 2018. Contact challenge of cattle with foot-and-mouth disease virus validates the role of the

nasopharyngeal epithelium as the site of primary and persistent infection. Scientific Reports. https://doi.org/10.1128/mSphere.00493-18

Visser, L.J., Medina, G.N., Rabouw, H.H., De Groot, R.J., Langereis, M.A., De los Santos, T.B., Van Kuppeveld, F.J. 2018. Foot-and-Mouth Disease Virus leader protease cleaves G3BP1 and G3BP2 and inhibits stress granule formation. Journal of Virology. 93(2):1-16. https://doi.org/10.1128/JVI.00922-18.

Ahmed, Z., Pauszek, S.J., Ludi, A., Larocco, M.A., Khan, E., Afzal, M., Arshed, M.J., Farooq, U., Arzt, J., Bertram, M., Brito, B., Naeem, K., Abubakar, M., Rodriguez, L.L. 2017. Genetic diversity and comparison of diagnostic tests for characterization of foot-and-mouth disease virus strains from Pakistan 2008-2012. Transboundary and Emerging Diseases. 65(2):534-546. https://doi.org/10.1111/tbed.12737.

Arzt, J., Brito, B.P., Pauszek, S.J., Hartwig, E.J., Smoliga, G.R., Vu, L.T., Vu, P.P., Stenfeldt, C., Rodriguez, L.L., Long, N.T., Dung, D.H. 2017. Genome sequence of foot-and-mouth disease virus serotype O lineage ind-2001d collected in Vietnam in 2015. Genome Announcements. 5(18):e00223-17. doi: 10.1128/genomeA.00223-17. https://doi.org/10.1128/GENOMEA.00223-17

Arzt, J., Pacheco Tobin, J., Stenfeldt, C., Rodriguez, L.L. 2017. Pathogenesis of virulent and attenuated foot and mouth disease virus in cattle. Virology. 14(1):89. doi: 10.1186/s12985-017-0758-9. https://doi.org/10.1186/s12985-017-0758-9

Cacciabue, M., Garcia-Nunez, M.S., Delgado, F., Curra, A., Marrero, R., Molinari, P., Rieder, A.E., Carrillo, E., Gismondi, M.I. 2017. Differential replication of foot-and-mouth disease viruses in mice determine lethality. Virology. 509:195-204. doi: 10.1016/j.virol.2017.06.012. https://doi.org/10.1016/j.virol.2017.06.012

Gutkoska, J., Larocco, M.A., Ramirez-Medina, E., De Los Santos, T.B., Lawrence, P.J. 2017. Host microRNA-203a is antagonistic to the progression of foot-and-mouth disease virus infection. Virology. doi: 10.1016/j.virol.2017.01.019. https://doi.org/10.1016/j.virol.2017.01.019

Hayder, S.S., VanderWaal, K., Ranjan, R., Biswal, J.K., Subramaniam, S., Mohapatra, J.K., Sharma, G.K., Rout, M., Dash, B., Das, B., Prusty, B., Sharma, A.K., Stenfeldt, C., Perez, A., Rodriguez, L.L., Arzt, J. 2017. Foot-and-mouth disease virus transmission dynamics and persistence in a herd of vaccinated dairy cattle in India. Transboundary and Emerging Diseases. 65(2):e404-e415. https://doi.org/10.1111/tbed.12774.

Kenney, M.A., Waters, R.A., Rieder, A.E., Pega, J., Perez-Filguera, M., Golde, W.T. 2017. Enhanced sensitivity in detection of antiviral antibody responses using biotinylation of foot-and-mouth disease virus (FMDV) capsids. Journal of Immunological Methods. 450:1-9. https://doi.org/10.1016/j.jim.2017.07.001.

Kloc, A., Diaz-San Segundo, F., Schafer, E.A., Rai, D.K., Kenney, M.A., De Los Santos, T.B., Rieder, A.E. 2017. Foot-and-mouth disease virus 5'-terminal S fragment is required for replication and modulation of the innate immune response in host cells. Virology. 512:132-143. https://doi.org/10.1016/j.virol.2017.08.036

Medina, G.N., Knudsen, G.M., Greninger, A.L., Kloc, A., Diaz-San Segundo, F., Rieder, A.E., Grubman, M., Derisi, J.L., De Los Santos, T.B. 2017. Interaction between FMDV Lpro and transcription factor ADNP is required for viral replication. Virology. 505:12-22. doi: 10.1016/j.virol.2017.02.010. https://doi.org/10.1016/j.virol.2017.02.010

Pauszek, S.J., Bertram, M.R., Vu, L.T., Hartwig, E.J., Smoliga, G.R., Brito, B., Stenfeldt, C., Hung, N.T., Hoang, B.H., Rodriguez, L.L., Dung, D.H., Arzt, J. 2017. Genome sequences of seven foot-and-mouth disease virus isolates collected from serial samples from one persistently infected carrier cow in Vietnam. Genome Announcements. 5:e00849-17. https://doi.org/10.1128/genomeA.00849-17

Rai, D.K., Diaz-San Segundo, F., Campagnola, G., Schafer, E.A., Kloc, A., Keith, A., De Los Santos, T.B., Peersen, O., Rieder, A.E. 2017. Attenuation of foot-and-mouth disease virus by engineered viral polymerase fidelity. Journal of Virology. doi: 10.1128/JVI.00081-17. https://doi.org/10.1128/JVI.00081-17

Sreenivasa, B.P., Mohapatra, J.K., Pauszek, S.J., Koster, M.J., Dhanya, V.C., Tamil Selvan, R.P., Hosamani, M., Saravanan, P., Basagoudanavar, S.H., De Los Santos, T.B., Venkataramanan, R., Rodriguez, L.L., Grubman, M.J. 2017. Recombinant human adenovirus-5 expressing capsid proteins of Indian vaccine strains of foot-and-mouth disease virus elicits effective antibody response in cattle. Veterinary Microbiology. 203:196-201. doi: 10.1016/j.vetmic.2017.03.019. https://doi.org/10.1016/j.vetmic.2017.03.019

Stenfeldt, C., Eschbaumer, M., Smoliga, G.R., Rodriguez, L.L., Zhu, J.J., Arzt, J. 2017. Clearance of a persistent Picornavirus infection is associated with enhanced pro-apoptotic and cellular immune responses. Scientific Reports. 7:17800. https://doi.org/10.1038/s41598-017-18112-4

Velazquez-Salinas, L., Niak, S., Peng, K., Pauszek, S.J., Rodriguez, L.L. 2017. Stomatitis virus (VSV) is nonpathogenic and nontransmissible in pigs, a natural host of VSV. Human Gene Therapy. 28(2):108-115. https://doi.org/10.1089/humc.2017.015

Vu, L.T., Long, N.T., Brito, B., Stenfeldt, C., Phuong, N.T., Hoang, B.H., Pauszek, S.J., Hartwig, E.J., Smoliga, G.R., Vu, P.P., Quang, L.T., Hung, V.V., Tho, N.D., Dong, P.V., Minh, P.Q., Bertram, M.R., Fish, I.H., Rodriguez, L.L., Dung, D.H., Arzt, J. 2017. First detection of foot-and-mouth disease virus O/Ind-2001d in Vietnam. PLoS One. 12(6):e0177361. https://doi.org/10.1371/journal.pone.0177361

Ranjan, R., Biswal, J.K., Subramaniam, S., Dash, B.B., Singh, K.P., Arzt, J., Rodriguez, L.L., Pattnaik, B. 2018. Evidence of subclinical foot-and-mouth disease virus infection in young calves born from clinically recovered cow under natural condition. Tropical Animal Health and Production. https://doi.org/10.1007/s11250-018-1518-6.

Brito, B., Pauszek, S.J., Eschbaumer, M., Stenfeldt, C., De Carvalho Ferreira, H., Vu, L.T., Phuong, N.T., Hoang, B.H., Tho, N.D., Dong, P.V., Minh, P.Q., Long, N.T., Dung, D.H., Rodriguez, L.L., Arzt, J. 2017. Phylodynamics of epidemic and asymptomatic foot-and-mouth disease in Vietnam 2010-2014. Veterinary Research. 48:24. https://doi.org/10.1186/s13567-017-0424-7

Hayer, S., Ranjan, R., Biswal, J., Mohapatra, J.K., Sharma, G.K., Subramaniam, S., Stenfeldt, C., Perez, A., Rodriguez, L.L., Pattnaik, B., Vanderwaal, K., Arzt, J. 2017. Quantitative characteristics of the foot-and-mouth disease carrier state under natural conditions in India. Transboundary and Emerging Diseases. doi: 10.1111/tbed.12627. https://doi.org/10.1111/tbed.12627

Stenfeldt, C., Arzt, J., Smoliga, G., Larocco, M.A., Gutkoska, J., Lawrence, P.J. 2017. Proof-of-concept study: profile of circulating micro RNAs in bovine serum harvested during acute and persistent FMDV infection. Virology Journal. 14:71. doi: 10.1186/s12985-017-0743-3. https://doi.org/10.1186/s12985-017-0743-3

Diaz-San Segundo, F., Medina, G., Stenfeldt, C., Arzt, J., De Los Santos, T.B. 2016. Foot-and-mouth disease vaccines. Veterinary Microbiology. doi: 10.1016/j.vetmic.2016.12.018. https://doi.org/10.1016/j.vetmic.2016.12.018

Pacheco, J., Stenfeldt, C., Rodriguez, L.L., Arzt, J. 2016. Infection dynamics of foot-and-mouth disease virus in cattle following intra-nasopharyngeal inoculation or contact exposure. Journal of Comparative Pathology. 155(4):314-325. doi: 10.1016/j.jcpa.2016.08.005. https://doi.org/10.1016/j.jcpa.2016.08.005

Pauszek, S.J., Eschbaumer, M., Brito, B., Ferreira De Carvalho, H.C., Vu, L.T., Phuong, N.T., Hoang, B.H., Tho, N.D., Tung, N., Thuy, N.T., Long, N.T., Dung, D.H., Rodriguez, L.L., Arzt, J. 2016. Site-specific substitution (Q172R) in the VP1 protein of subclinical FMDV isolates collected in Vietnam. Virology Reports. 6:90-96. https://doi.org/10.1016/j.virep.2016.10.001

Pedersen, L.E., Patch, J.R., Kenney, M.A., Glabman, R., Nielsen, M., Jungersen, G., Buus, S., Golde, W.T. 2016. Expanding specificity of class 1 restricted CD8+ T cells for viral epitopes following multiple inoculations of swine with a human adenivorus vectored foot-and-mouth disease virus (FMDV) vaccine. Veterinary Immunology and Immunopathology. 181:59-67. doi: 10.1016/j.vetimm.2016.07.012. https://doi.org/10.1016/j.vetimm.2016.07.012

Ranjan, R., Biswal, J.K., Subramaniam, S., Singh, K.P., Stenfeld, C., Rodriguez, L.L., Pattnaik, B., Arzt, J. 2016. Foot-and-mouth disease virus-associated abortion and vertical transmission following acute infection in cattle under natural conditions. PLoS One. 11(12):e0167163. https://doi.org/10.1371/journal.pone.0167163

Rhyan, J., Mccollum, M., Gidlewski, T., Shalev, M., Ward, G., Donahue, B., Arzt, J., Stenfeldt, C., Mohamed, F., Nol, P., Deng, M., Metwally, S., Mckenna, T., Salman, M. 2016. Foot-and-Mouth Disease in a small sample of experimentally infected pronghorn (Antilocapra americana). Journal of Wildlife Diseases. 52(4):862-873. https://doi.org/10.7589/2015-11-312

Stenfeldt, C., Pacheco, J., Brito, B.P., Moreno-Torres, K.I., Branan, M.A., Delgado, A., Rodriguez, L.L., Arzt, J. 2016. Transmission of Foot-and-mouth disease virus during the incubation period in pigs. Frontiers in Veterinary Science. 3:105. doi: 10.3389/fvets.2016.00105. https://doi.org/10.3389/fvets.2016.00105

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Genetic Impact and Improved Diagnostics for Sheep and Goat Transmissible Spongiform Encephalopathies; David Schneider (P), Stephen White, Naomi Taus, David Herndon, Lowell Kappmeyer, Cristina Cunha and Michelle Mousel

Cinar Ulas, M., Schneider, D.A., Waldron, D.F., O'Rourke, K., White, S.N. 2018. Goats singly heterozygous for PRNP S146 or K222 orally inoculated with classical scrapie at birth show no disease at ages well beyond six years. Journal of Animal Breeding and Genetics. 233:19-24. https://doi.org/10.1016/j.tvjl.2017.12.019

Madsen-Bouterse, S.A., Highland, M.A., Dassanayake, R.P., Zhuang, D., Schneider, D.A. 2018. Low-volume goat milk transmission of classical scrapie to lambs and goat kids. PLoS One. 13(9):e0204281. https://doi.org/10.1371/journal.pone.0204281

Dinkel, K.D., Schneider, D.A., Muñoz-Gutiérrez, J.F., Mcelliott, V.R., Stanton, J.B. 2017. Correlation of cellular factors and differential scrapie prion permissiveness in ovine microglia. Virus Research. 240:69-80. https://doi.org/10.1016/j.virusres.2017.07.016

Monello, R.J., Galloway, N.L., Powers, J.G., Madsen-Bouterse, S.A., Edwards, H., Wood, M., O'Rourke, K., Wild, M.A. 2017. Pathogen-mediated selection in free-ranging elk populations infected by chronic wasting disease. Proceedings of the National Academy of Sciences. 114(46):12208-12212. https://doi.org/10.1073/pnas.1707807114

Oliveira, R.D., Mousel, M.R., Pabilonia, K.L., Highland, M.A., Taylor, J.B., Knowles Jr, D.P., White, S.N. 2017. Domestic sheep show average Coxiella burnetii seropositivity generations after a sheep-associated human Q fever outbreak and lack detectable shedding by placental, vaginal, and fecal routes. PLoS One. 12(11):e0188054. https://doi.org/10.1371/journal.pone.0188054

Mousel, M.R., White, S.N., Herndon, D.R., Reynolds, J.O., Gonzalez, M.V., Johnson, W.C., Ueti, M.W., Taylor, J.B., Knowles, D.P. 2016. Ovine leukocyte profiles do not associate with variation in the prion gene, but are breed dependent. Anim Genet. 47(1):136-137. https://doi.org/10.1111/age.12381

2090-32000-036-00D
Identification of Host Factors and Immunopathogenesis of Pneumonia in Domestic and Bighorn Sheep; Stephen White (P), David Herndon, Lowell Kappmeyer and Michelle Mousel

Koltes, J.E., Cole, J.B., Serão, N.V.L., McCue, M., Woodward, J., Zhang, H., McKay, S., Lunney, J., Kramer, L., Schroeder, M., Clemmens, R., Murdoch, B., Rexroad, C.E., III, Rosa, G.J.M., Mateescu, R., White, S.N., Worku, M., Reecy, J.M. 2019. A vision for development and utilization of high-throughput phenotyping and big data analytics in livestock. Frontiers in Genetics. 10:1197. https://doi.org/10.3389/fgene.2019.01197.

Rovani, E.R., Beckman, K.B., Highland, M.A. 2019. Mycoplasma ovipneumoniae associated with polymicrobial pneumonia in a free-ranging yearling caribou (Rangifer tarandus tarandus) from Alaska, USA. Journal of Wildlife Diseases. 55(3):733-736. https://doi.org/10.7589/2018-08-188

Cinar, M., Mousel, M.R., Herndon, M.K., Taylor, J.B., White, S.N. 2018. Tenascin-XB (TNXB) amino acid substitution E2004G is associated with mature weight and milk score in rambouillet, targhee, polypay, and suffolk sheep. Small Ruminant Research. 166:129-133. https://doi.org/10.1016/j.smallrumres.2018.06.013

Davenport, K.M., Duan, M., Hunter, S.S., New, D.D., Fagnan, M.W., Highland, M.A., Murdoch, B.M. 2018. Complete mitochondrial genome sequence of bighorn sheep. Genome Announcements. https://doi.org/10.1128/genomeA.00464-18

Highland, M.A., Berglund, A.K., Knowles, D.P. 2018. Passive transfer in somestic and bighorn lambs total IgG in ewe sera and colostrum and serum IgG kinetics in lambs following colostrum ingestion are similar in domestic sheep and bighorn sheep (ovis aries and ovis canadensis). Sheep and Goat Research

Journal. 32:36-42. https://d1cqrq366w3ike.cloudfront.net/http/DOCUMENT/SheepUSA/SGRJ_V32_36-42_Highland_12-17.pdf

Highland, M.A., Herndon, D.R., Bender, S., Hansen, L., Gerlach, R.F., Beckman, K.B. 2018. Mycoplasma ovipneumoniae in wildlife species beyond the subfamily caprinae. Emerging Infectious Diseases. 24(12):2384-2386. https://doi.org/10.3201/eid2412.180632

Kise, J.N., Neupane, M., White, S.N., Neibergs, H.N. 2018. Identification of genes associated with susceptibility to mycobacterium avium ssp. paratuberculosis (Map) tissue infection in Holstein cattle using gene set enrichment analysis-SNP. Mammalian Genome. 29(7-8):539-549. https://doi.org/10.1007/s00335-017-9725-4

Batra, S.A., Shanthalingam, S., Donofrio, G., Haldorson, G.J., Chowdhury, S., White, S.N., Subramaniam, S. 2017. Immunization of bighorn sheep against Mannheimia haemolytica with a bovine herpesvirus 1-vectored vaccine. Vaccine. 35(12):1630-1636. https://doi.org/10.1016/j.vaccine.2017.02.006

Highland, M.A. 2017. Small ruminant lentiviruses: Strain variation, viral tropism, and host genetics influence pathogenesis. Veterinary Pathology. 54(3):353-354. https://doi.org/10.1177%2F0300985817695517

White, S.N., Oliveira, R.D., Mousel, M.R., Gonzalez, M.V., Highland, M.A., Herndon, M.K., Taylor, J.B., Knowles Jr, D.P. 2017. Underdominant KCC3b R31I association with blood sodium concentration in domestic sheep suggests role in oligomer function. Animal Genetics. 48(5):626-627. https://doi.org/10.1111/age.12585

Tuggle, C.K., Giuffra, E., White, S.N., Clarke, L., Zhou, H., Ross, P.J., Acloque, H., Reecy, J.M., Archibald, A., Bellone, R.R., Tixier-Boichard, M., Chamberlain, A., Cheng, H., Croojimans, R.P., Delany, M.E., Finno C.J., Groenen, M.A., Hayes, B., Lunney, J.K., Petersen, J.L., Plastow, G.S., Schmidt, C.J., Song, J., Watson, M. 2016. GO-FAANG meeting: a Gathering on Functional Annotation of Animal Genomes. Anim Genet. 47(5):528-533. https://doi.org/10.1111/age.12466

Cinar, M.U., Mousel, M.R., Herrmann-Hoesing, L.M., Taylor, J.B., White, S.N. 2016. Ovar-DRB1 haplotypes *2001 and *0303 are associated with sheep growth and ewe lifetime prolificacy. GENE 595(2):187-192. https://doi.org/10.1016/j.gene.2016.10.004

Highland, M.A., Schneider, D.A., White, S.N., Madsen-Bouterse, S.A, Knowles, D.P., Davis, W.C. 2016. Single-label flow cytometry using cross reactive monoclonal antibodies identifies leukocyte surface molecule abundance differences in domestic sheep (Ovis aries) and bighorn sheep (Ovis canadensis) neutrophils. Comp Immunol Microbiol Infect Dis. 46:40-46. https://doi.org/10.1016/j.cimid.2016.04.006

2090-32000-037-00D
Diagnostic and Control Strategies for Malignant Catarrhal Fever; Cristina Cunha (P)

Cunha, C.W., O'Toole, D., Taus, N.S., Knowles, D.P., Li, H. 2019. A rabbit model for sheep-associated malignant catarrhal fever research: from virus infection to pathogenesis studies and vaccine development. Current Clinical Microbiology Reports. https://doi.org/10.1007/s40588-019-00126-3

Cunha, C.W., Slaer, O., Macbeth, B., Warren, A., Duignan, P.J., Highland, M.A., Li, H. 2019. Domestic sheep and bighorn sheep carry distinct gammaherpesviruses belonging to the genus macavirus. Virus Research. https://doi.org/10.1016/j.virusres.2019.197729

Alhajri, S.M., Cunha, C.W., Knowles, D.P., Li, H., Taus, N.S. 2018. Evaluation of glycoprotein Ov8 as a potential antigen for an OvHV-2-specific diagnostic assay. PLoS One. 13(7):30200130. https://doi.org/10.1371/journal.pone.0200130

Ortiz, K., Javaux, J., Simon, M., Petit, T., Clavel, S., Lamglait, B., Blanc, B., Brunet, A., Myster, F., Li, H., Dewals, B. 2018. Seroprevalence of malignant catarrhal fever virus in captive wildebeest (Connochaetes sp.) in France. Transboundary and Emerging Diseases. https://doi.org/10.1111/tbed.12929

Pesavento, P., Cunha, C.W., Li, H., Jackson, K., O'Toole, D. 2018. In situ hybridization for localization of ovine herpesvirus 2, the agent of sheep-associated malignant catarrhal fever, in formalin-fixed tissues. Veterinary Pathology. 56(1):78-86. https://doi.org/10.1177/0300985818798085

Pesavento, P., Dange, R., Estrada, M., Dasjerdi, A., Pérez, V., Laroca, A., Silván, J., Diab, S., Jackson, K., Phillips, I., Li, H., Cunha, C.W., Wessels, M. 2018. Systemic necrotizing vasculitis in sheep is associated with ovine herpesvirus 2. Veterinary Pathology. 56:(1)87-92. https://doi.org/10.1177/0300985818795166

Phillips, I.L., Cunha, C.W., Galbraith, D., Highland, M.A., Bildfell, R.J., Li, H. 2018. High copy number of ovine gammaherpesvirus 2 DNA associated with malignant catarrhal fever-like syndrome in a lamb. Journal of Veterinary Diagnostic Investigation. https://doi.org/10.1177/1040638718766976

2090-32000-038-00D
Identification of Tick Colonization Mechanisms and Vaccine Development for Anaplasmosis; Susan Noh (P), David Herndon, Lowell Kappmeyer and Massaro Ueti

Futse J.E., Buami G., Kayang B.B., Koku R., Palmer G.H., Graça T., Noh S.M. 2019. Sequence and immunologic conservation of Anaplasma marginale OmpA within strains from Ghana as compared to the predominant OmpA variant. PLoS One.14(7):e0217661. https://doi.org/10.1371/journal.pone.0217661

Awada, L., Tizzani, P., Noh, S.M., Ducrot, C., Ntsama, F., Caceres, P., Chalvet-Monfray, K. 2018. Global dynamics of highly pathogenic avian influenza outbreaks between 2005 and 2016 - Focus on distance and rate of spread. Transboundary and Emerging Diseases. 65(6):2006-2016. https://doi.org/10.1111/tbed.12986

Dasilva, N.B., Ueti, M.W., Johnson, W.C., Mira, A., Schnittger, L., Valente, J.D., Vidotto, O., Masterson, H.E., Taus, N.S., Vieira, T.S. 2018. First report of Anaplasma marginale infection in goats, Brazil. PLoS One. 13(8):e0202140. https://doi.org/10.1371/journal.pone.0202140

Kaufman, E.L., Stone, N.E., Scoles, G.A., Hepp, C.M., Busch, J.D., Wagner, D.M. 2018. Range-wide genetic analysis of Dermacentor variabilis and its Francisella-like endosymbionts demonstrates phylogeographic concordance between both taxa. Parasites & Vectors. https://doi.org/10.1186/s13071-018-2886-5

Reif, K.E., Ujczo, J.K., Alperin, D.C., Noh, S.M. 2018. Differences in francisella tularensis subsp. novicida infection competence differs in cell lines derived from United States populations of Dermacentor andersoni and Ixodes scapularis. Scientific Reports. 8(1):12685. https://doi.org/10.1038/s41598-018-30419-4

Mason, K.L., Gonzalez, M.V., Chung, C., Mousel, M.R., White, S.N., Taylor, J.B., Scoles, G.A. 2017. Detection of Anaplasma ovis and validation of improved A. marginale cELISA kit for diagnostic use in domestic sheep. J Vet Diagn Invest. 29(5):763-766. https://doi.org/10.1177/1040638717709494

Capelli-Peixoto, J., Carvalho, D.D., Johnson, W.C., Scoles, G.A., Fogaça, A.C., Daffre, S., Ueti, M.W. 2017. The transcription factor relish controls anaplasma marginale infection in the bovine tick rhipicephalus microplus. Developmental and Comparative Immunology. doi: 10.1016/j.dci.2017.04.005. https://doi.org/10.1016/j.dci.2017.04.005

Manjunatha, U.H., Vinayak, S., Zambriski, J., Chao, A.T., Sy, T.L., Noble, C.G., Bonamy, G.M., Kondreddi, R.R., Zou, B., Gedeck, P., Brooks, C.F., Herbert, G.T., Sateriale, A., Tandel, J., Noh, S.M., Lakshminarayana, S.B., Lim, S.H., Goodman, L.B., Bodenreider, C., Feng, G., Zhang, L., Blasco, F., Wagner, J., Leong, J.F., Striepen, B., Diagana, T.T. 2017. A cryptosporidium PI(4)K inhibitor is a drug candidate for cryptosporidiosis. Nature. https://doi.org/10.1038/nature22337

McClure, E.E., Oliva Chavez, A.S., Shaw, D.K., Carlyon, J.A., Ganta, R.R., Noh, S.M., Wood, D.O., Bavoil, P.M., Brayton, K.A., Pedra, J.H. 2017. Engineering of obligate intracellular bacteria: progress, challenges and paradigms. Nature Reviews Microbiology. https://doi.org/10.1038/nrmicro.2017.59

Rogovskyy, A.S., Nebogatkin, I.V., Scoles, G.A. 2016. Ixodid ticks in the megapolis of Kyiv, Ukraine. Ticks and Tick Borne Diseases. 8(1):99-102. https://doi.org/10.1016/j.ttbdis.2016.10.004

Mason K.L., Gonzalez M.V., Chung C., Mousel M.R., White S.N., Taylor J.B., Scoles G.A. 2017. Validation of an improved Anaplasma antibody competitive ELISA for detection of Anaplasma ovis antibody in domestic sheep. J Vet Diagn Invest. Sep;29(5):763-766. https://doi.org/10.1177/1040638717709494

Gall A.C., Scoles G.A, Magori K., Mason K.L., Brayton K.A. Laboratory colonization stabilizes the naturally dynamic microbiome composition of field collected Dermacentor andersoni ticks. 2017. Microbiome. 5:133-143. https://doi.org/10.1186/s40168-017-0352-9

Deringer J.R., Forero-Becerra E.G., Ueti M.W., Turse J.E., Futse J.E., Noh S.M., Palmer G.H., Brown W.C. 2017. Identification of a T-Cell Epitope that is globally conserved among outer membrane proteins (OMPs) OMP7, OMP8, and OMP9 of Anaplasma marginale strains and with OMP7 from the A. marginale subsp. centrale Vaccine Strain. Clin Vaccine Immunol. 24(1). https://doi.org/10.1128/CVI.00406-16

Truchan H.K., Hebert K.S., Cockburn C.L., Magunda F., Noh S.M., Carlyon JA. 2016. The pathogen-occupied vacuoles of Anaplasma phagocytophilum and Anaplasma marginale interact with the endoplasmic reticulum. Front Cell Infect Microbiol. 6:22. https://doi.org/10.3389/fcimb.2016.00022

Gall C.A., Reif K.A., Scoles G.A., Mason K.L., Mousel M., Noh S.M., Brayton K.A. 2016. The bacterial microbiome of Dermacentor andersoni ticks influences pathogen susceptibility. ISMEJ.10:1846-1855. https://doi.org/10.1038/ismej.2015.266

Noh S.M., Dark M.J., Reif K.E., Ueti M.W., Kappmeyer L.S., Scoles G.A., Palmer G.H., Brayton K.A. Super-infection exclusion of the ruminant pathogen Anaplasma marginale in its tick vector is dependent on time between exposures to the strains. 2016. Appl Environ Microbiol. 82:3217-3224. https://doi.org/10.1128/AEM.00190-16

Beckley C.S., Shaban S., Palmer G.H., Hudak A.T., Noh S.M., Futse J.E. 2016. Disaggregating tropical disease prevalence by climatic and vegetative zones within tropical West Africa. PLoS One. 11(3):e.0152560. https://doi.org/10.1371/journal.pone.0152560

Magunda F., Thompson C.W., Schneider D.A., Noh S.M. 2016. Anaplasma marginale actively modulates vacuolar maturation during intracellular infection of its tick vector Dermacentor andersoni. Appl Environ Microbiol. 82:4715-4731. https://doi.org/10.1128/AEM.01030-16

Clayton K.A., Gall C.A., Mason K.L., Scoles G.A., Brayton K.A. 2015. The characterization and manipulation of the bacterial microbiome of the Rocky Mountain wood tick, Dermacentor andersoni. Parasit Vectors. Dec 10;8:632. https://doi.org/10.1186/s13071-015-1245-z

Araya-Anchetta A., Busch J.D., Scoles G.A., Wagner D.M. 2015. Thirty years of tick population genetics: a comprehensive review. Infect Genet Evol. Jan;29:164-79. https://doi.org/10.1177/1040638717709494

Ducken D.R., Brown W.C., Alperin D.C., Brayton K.A., Reif K.E., Turse J.E., Palmer G. H., Noh S.M. Subdominant outer membrane antigens in Anaplasma marginale: conservation, antigenicity, and protective capacity using recombinant protein. 2015. PLoS One. 10(6):e0129309. https://doi.org/10.1371/journal.pone.0129309

Lone A., Atci E., Renslow R., Beyenal H., Noh S.M., Fransson B., Abu-Lail N., Park J. J., Gang D.R., Call D.R. 2015. Staphylococcus aureus induced hypoxia and cellular damage in porcine dermal explants. Infect Immun.83:2531-41. https://doi.org/10.1128/IAI.03075-14.

Lone A. G., Atci E., Renslow R., Beyenal H., Noh S., Fransson B., Abu-Lail N., Park J. J., Gang D.R., Call D.R. 2015. Colonization of epidermal tissue by Staphylococcus aureus produces localized hypoxia and stimulates secretion of antioxidant and caspase-14 proteins. Infect Immun. 83:3026-34. https://doi.org/10.1128/IAI.00175-15.

Graça T., Paradiso L., Broschat S.L., Noh S.M., Palmer G.H. 2015. Primary structural variation in Anaplasma marginale Msp2 efficiently generates immune escape variants. Infect Immun. 83:4178-4184. https://doi.org/10.1128/IAI.00851-15.

2090-32000-039-00D
Development of Detection and Control Strategies for Bovine Babesiosis and Equine Piroplasmosis; Massaro Ueti, Lowell Kappmeyer, Naomi Taus, Carlos Suarez and Lindsay Fry

Abdellrazeq GS, Fry LM, Elnaggar MM, Bannantine JP, Schneider DA, Chamberlin WM, Mahmoud AHA, Park KT, Hulubei V, Davis WC. Simultaneous cognate epitope recognition by bovine CD4 and CD8 T cells is essential for primary expansion of antigen-specific cytotoxic T-cells following ex vivo stimulation with a candidate Mycobacterium avium subsp. paratuberculosis peptide vaccine. Vaccine. 2020 Jan 2. pii: S0264-410X(19)31704-9. doi: 10.1016/j.vaccine.2019.12.052. https://doi.org/10.1016/j.vaccine.2019.12.052.

Elnaggar MM, Grandoni F, Abdellrazeq GS, Fry LM, El-Naggar K, Hulubei V, Buttazzoni L, Khaliel SA, Torky HA, Davis WC. Pattern of CD14, CD16, CD163 and CD172a expression on water buffalo (Bubalus bubalis) leukocytes. Vet Immunol Immunopathol. 2019 May;211:1-5. doi: 10.1016/j.vetimm.2019.03.010. Epub 2019 Mar 25. https://doi.org/10.1016/j.vetimm.2019.03.010.

Bastos RG, Sears K, Dinkel KD, Knowles DP, Fry LM. Changes in the Molecular and Functional Phenotype of Bovine Monocytes during Theileria parva Infection. Infect Immun. 2019 Nov 18;87(12). pii: e00703-19. doi: 10.1128/IAI.00703-19. Print 2019 Dec. https://doi.org/10.1128/IAI.00703-19.

Gallego-Lopez GM, Cooke BM, Suarez CE. Interplay between Attenuation- and Virulence-Factors of Babesia bovis and Their Contribution to the Establishment of Persistent Infections in Cattle.Pathogens. 2019 Jul 4;8(3). pii: E97. doi: 10.3390/pathogens8030097. Review. https://doi.org/10.3390/pathogens8030097.

Rathinasamy V, Poole WA, Bastos RG, Suarez CE, Cooke BM. Babesiosis Vaccines: Lessons Learned, Challenges Ahead, and Future Glimpses. Trends Parasitol. 2019 Aug;35(8):622-635. doi: 10.1016/j.pt.2019.06.002. Epub 2019 Jul 4. Review. https://doi.org/10.1016/j.pt.2019.06.002.

Bohaliga GAR, Johnson WC, Taus NS, Hussein HE, Bastos RG, Suarez CE, Scoles GA, Ueti MW. Identification of proteins expressed by Babesia bigemina kinetes. Parasit Vectors. 2019 May 28;12(1):271. doi: 10.1186/s13071-019-3531-7. https://doi.org/10.1186/s13071-019-3531-7.

Mosqueda J, Hidalgo-Ruiz M, Calvo-Olvera DA, Hernandez-Silva DJ, Ueti MW, Mercado-Uriostegui MA, Rodriguez A, Ramos-Aragon JA, Hernandez-Ortiz R, Kawazu SI, Igarashi I. RON2, a novel gene in Babesia bigemina, contains conserved, immunodominant B-cell epitopes that induce antibodies that block merozoite invasion. Parasitology. 2019 Nov;146(13):1646-1654. doi: 10.1017/S0031182019001161. Epub 2019 Sep 13. https://doi.org/10.1017/S0031182019001161.

Alzan, H.F., Cooke, B.M., Suarez, C.E. 2019. Transgenic babesia bovis lacking 6-Cys sexual-stage genes as the foundation for non-transmissible live vaccines against bovine babesiosis. Ticks and Tick Borne Diseases. 10(3):722-728. https://doi.org/10.1016/j.ttbdis.2019.01.006

Fry, L.M., Bastos, R.G., Stone, B.C., Williams, L.B., Knowles, D.P., Murphy, S.C. 2019. Gene gun DNA immunization of cattle induces humoral and CD4 T-cell-mediated immune responses against the Theileria parva polymorphic immunodominant molecule. Vaccine. 37(12):1546-1553. https://doi.org/10.1016/j.vaccine.2019.02.009

Hala, H.E., Johnson, W.C., Taus, N.S., Suarez, C.E., Scoles, G.A., Ueti, M.W. 2019. Silencing expression of the Rhipicephalus microplus vitellogenin receptor gene blocks Babesia bovis transmission and interferes with oocyte maturation. Parasites & Vectors. 12(1):7. https://doi.org/10.1186/s13071-018-3270-1.

Iacobucci, E., Fritz, H., Papageorgiou, S., O'Conner, R., Taus, N.S., Ueti, M.W. 2019. Detection and genotypic characterization of toxoplasma gondii DNA within the milk of Mongolian livestock. Parasitology Research. 118(6):2005-2008. https://doi.org/10.1007/s00436-019-06306-w

Hussein, H.E., Johnson, W.C., Taus, N.S., Suarez, C.E., Scoles, G.A., Ueti, M.W. 2019. Silencing expression of the Rhipicephalus microplus vitellogenin receptor gene blocks Babesia bovis transmission and interferes with oocyte maturation. Parasites & Vectors. https://doi.org/10.1186/s13071-018-3270-1

Sears, K.P., Kappmeyer, L.S., Wise, L.N., Silva, Marta., Ueti, M.W., White, S.N., Reif, K.E., Knowles, D.P. 2019. Infection dynamics of theileria equi and theileria haneyi, a newly discovered apicomplexan of the horse. Veterinary Parasitology. 271:(68-75). https://doi.org/10.1016/j.vetpar.2019.06.009

Suarez, C.E., Alzan, A., Silva, M., Rathinasamy, V., Poole, W., Cooke, B. 2019. Unraveling the cellular and molecular pathogenesis of bovine babesiosis: is the sky the limit? International Journal for Parasitology. 49(2):183-197. https://doi.org/10.1016/j.ijpara.2018.11.002

Williams, L.B., Fry, L.M., Herndon, D.R., Franceschi, V., Schneider, D.A., Donofrio, G., Knowles, D.P. 2019. A recombinant bovine herpesvirus-4 vectored vaccine delivered via intranasal nebulization elicits viral neutralizing antibody titers in cattle. PLoS One. 14(4):e0215605. https://doi.org/10.1371/journal.pone.0215605

Wise, L.N., Kappmeyer, L.S., Knowles, D.P., White, S.N. 2019. Evolution and diversity of the EMA families of the divergent equid parasites, theileria equi and T. haneyi. Infection, Genetics and Evolution. 68:153-160. https://doi.org/10.1016/j.meegid.2018.12.020

Elnaggar MM, Grandoni F, Elnaggar KM, Abdellrazeq GS, Fry LM, Khaliel SA, Torky HA, Davis WC. Pattern of CD14, CD16, CD163A and CD172a expression on water buffalo leukocytes. Dev Comp Immunol. 2019;211:1-5. https://doi.org/10.1016/j.vetimm.2019.03.010

Bastos RG, Franceschi V, Tebaldi G, Connelley T, Morrison WI, Knowles DP, Donofrio G, Fry LM. Molecular and antigenic properties of mammalian cell-expressed Theileria parva antigen Tp9. Front Immunol. 2019;10:897. https://doi.org/10.3389/fimmu.2019.00897

Bastos RG, Sears KP, Dinkel KD, Knowles DP, and Fry LM. Changes in molecular and functional phenotype of bovine monocytes during Theileria parva infection. Infect Immun. 2019;87(12):e00703-19. https://doi.org/10.1128/IAI.00703-19

Franceschi V, Mahmoud A, Abdellrazeq G, Tebaldi G, Macchi F, Russo L, Fry LM, Elnaggar M, Bannantine J, Park KT, Hulubei V, Cavirani S, Davis WC and Donofrio G. Capacity to elicit cytotoxic CD8 T cell activity against Mycobacterium avium subsp. Paratubuerculosis is retained in a vaccine candidate 35 kDa peptide modified for expression in mammalian cells. Front Immunol. 2019;doi:10.3389/fimmu.2019.02859. https://doi.org/10.3389/fimmu.2019.02859

Abdellrazeq GS, Fry LM, Elnaggar MM, Bannantine JP, Schneider DA, Chamberlain WM, Mahmoud AHA, Park KT, Hulubei V, Davis WC. Cognate epitope recognition by bovine CD4 and CD8 T cells is essential for primary expansion of antigen-specific cytotoxic T-cells following ex vivo stimulation with a candidate Mycobacterium avium subsp. paratuberculosis peptide vaccine. Vaccine. 2019;doi:10.1016/j.vaccine.2019.12.052. https://doi.org/10.1101/555672

Olds, C.L., Mason, K.L., Scoles, G.A. 2018. Rhipicephalus appendiculatus ticks transmit theileria parva from persistently infected cattle in the absence of detectable parasitemia: implications for East Coast fever epidemiology. Parasites & Vectors. https://doi.org/10.1186/s13071-018-2727-6.

Bohaliga, G.A., Johnson, W.C., Taus, N.S., Hussein, H.E., Ueti, M.W. 2018. Identification of a putative methyltransferase gene of Babesia bigemina as a novel molecular biomarker uniquely expressed in parasite tick stages. Parasites & Vectors. 11(1):480. https://doi.org/10.1186/s13071-018-3052-9.

Gallego-Lopez, G., Lau, A., Johnson, W.C., Ueti, M.W., Suarez, C.E. 2018. Spherical body protein 2 truncated copy 11 as a specific babesia bovis attenuation marker. Parasites & Vectors. 11:169. https://doi.org/10.1186/s13071-018-2782-z

Gallego-Lopez, G., Lau, A., O'Connor, R., Ueti, M.W., Cooke, B., Laughery, J., Graca, T., Madsen-Bouterse, S., Oldiges, D., Suarez, C.E. 2018. Up-regulated expression of spherical body protein 2 truncated copy 11 is associated with reduced in vitro endothelial cell cytoadhesion by babesia bovis. International Journal for Parasitology. 49(2):127-137. https://doi.org/10.1016/j.ijpara.2018.05.015.

Gold, J.R., Knowles, D.P., Coffey, T., Bayly, W.M. 2018. Exercise-induced pulmonary hemorrhage in barrel racing horses in the Pacific Northwest region of the United States. Journal of Veterinary Internal Medicine. https://doi.org/10.1111/jvim.15066

Hernández-Silva, D., Valdez-Espinoza, U., Mercado Uriostegui, M., Aguilar-Tipacamú, G., Ramos-Aragón, J., Hernández-Ortiz, R., Ueti, M.W., Mosqueda, J. 2018. Immunomolecular characterization of MIC-1, a novel antigen in babesia bigemina, which contains conserved and immunodominant B-cell epitopes that induce neutralizing antibodies. Journal of Veterinary Science. https://doi.org/10.3390/vetsci5020032

Knowles, D.P., Kappmeyer, L.S., Haney, D., Herndon, D.R., Fry, L.M., Munro, J.B., Sears, K., Ueti, M.W., Wise, L.N., Silva, M., Schneider, D.A., Grause, J., White, S.N., Trentina, K., Bishop, R., Odongo, D.O., Pelzel-McCluske, A.M., Scoles, G.A., Mealey, R.H., Silva, J.C. 2018. Discovery of a novel species Theileria haneyi n. sp. infective to equids highlights exceptional genomic diversity within the genus Theileria: implications for apicomplexan parasite surveillance. International Journal for Parasitology. 48(9-10):127-137. https://doi.org/10.1016/j.ijpara.2018.03.010

Mohamad, S.B., Alagib, A., Abdelkareim, T.B., Haasan, M.M., Johnson, W.C., Hussein, H.E., Taus, N.S., Ueti, M.W. 2018. Molecular detection and characterization of theileria spp infecting cattle in Sennar State, Sudan. Parasitology Research. 117(4):1271-1276. https://doi.org/10.1007/s00436-018-5775-0

Ruiz, M.H., Suarez, C.E., Mercado-Uriostequi, M.A., Hernandez-Ortiz, R., Ramos, J.A., Galindo-Velazco, E., Mosqueda, J. 2018. Babesia bovis RON2 contains conserved B-cell epitopes that induce an invasion-blocking humoral immune response in immunized cattle. Parasites & Vectors. 11:575. https://doi.org/10.1186/s13071-018-3164-2

Scudiero, L., Mercado, W.J., Rudolph, A., Wang, J., Laughery, J.M., Suarez, C.E. 2018. Comparisons of the topographic characteristics and electrical charge distributions among Babesia-infected erythrocytes and extraerythrocytic merozoites using AFM. Journal of Microscopy. doi:10.1111/jmi.12697. https://doi.org/10.1111/jmi.12697

Silva, M.G., Knowles, D.P., Mazuz, M.L., Cooke, B.M., Suarez, C.E. 2018. Stable transformation of Babesia bigemina and Babesia bovis using a single transfection plasmid. Scientific Reports. 8:6096. doi:10.1038/s41598-018-23010-4. https://doi.org/10.1038/s41598-018-23010-4

Silva, M.G., Villarino, N.F., Knowles, D.P., Suarez, C.E. 2018. Assessment of Draxxin® (tulathromycin) as an anti-babesia bovis, babesia bigemina and theileria equi chemotherapeutic in cultured parasites. International Journal for Parasitology: Drug and Drug Resistance. 17:8(2):265-270. https://doi.org/10.1016/j.ijpddr.2018.04.004

Wise, L.N., Kappmeyer, L.S., Dasilva, M., White, S.N., Grause, J.F., Knowles Jr, D.P. 2017. Verification of post-chemotherapeutic clearance of theileria equi through concordance of nested PCR and immunoblot. Ticks and Tick Borne Diseases. https://doi.org/10.1016/j.ttbdis.2017.08.007.

Alzan, A., Silva, M., Davis, W., Herndon, D.R., Schneider, D.A., Suarez, C.E. 2017. Geno- and phenotypic characteristics of a transfected babesia bovis 6-Cys-E knockout clonal line. Parasites & Vectors. 10(1):214. https://doi.org/10.1186/s13071-017-2143-3

Grandoni, F., Elnaggar, M.M., Abdellrazeq, G.S., Signorelli, F., Fry, L.M., Marchitelli, C., Hulubei, V., Khaliel, S.A., Torky, H.A., Davis, W.C. 2017. Characterization of leukocyte subsets in buffalo (Bubalus bubalis) with cross-reactive monoclonal antibodies specific for bovine MHC class I and class II molecules and leukocyte differentiation molecules. Developmental and Comparative Immunology. 74:101-109. https://doi.org/10.1016/j.dci.2017.04.013

Hussein, H.E., Bastos, R.G., Schneider, D.A., Johnson, W.C., Adham, F.K., Davis, W.C., Laughery, J.M., Herndon, D.R., Alzan, H.F., Ueti, M.W., Suarez, C.E. 2017. The babesia bovis hap2 gene is not required for blood stage replication, but expressed upon in vitro sexual stage induction. PLOS Neglected Tropical Diseases. 11(10):e0005965. https://doi.org/10.1371/journal.pntd.0005965

Levi, M.M., Tirosh-Levy, S., Dahan, R., Berlin, D., Steinman, A., Edery, N., Savistsky, I., Lebovich, B., Knowles Jr, D.P., Suarez, C.E. 2017. First detection of diffuse and cerebral theileria equi infection in neonatal filly. Journal of Equine Veterinary Science. 60:23-28. https://doi.org/10.1016/j.jevs.2017.10.016

Silva, M.G., Knowles, D.P., Domingos, A., Esteves, A.M., Antunes, S., Suarez, C.E. 2017. Inhibition of the in vitro growth of babesia bigemina, babesia caballi and theileria equi parasites by trifluralin analogues. Ticks and Tick Borne Diseases. 8(4):593-597. https://doi.org/10.1016/j.ttbdis.2017.04.002.

Suarez, C.E., Bishop, R.P., Alzan, H., Poole, W., Cook, B. 2017. Advances in the application of genetic manipulation methods to apicomplexan parasites. International Journal for Parasitology. https://doi.org/10.1016/j.ijpara.2017.08.002

Elnaggar MM, Abdellrazeq GS, Venn-Watson SK, Jensen ED, Hulubei V, Fry LM, Sacco RE, Davis WC. Identification of monoclonal antibodies cross-reactive with bottlenose dolphin orthologues of the major histocompatibility complex and leukocyte differentiation molecules. Veterinary Immunology and Immunopathology. 2017;192:54-59. https://doi.org/10.1016/j.vetimm.2017.09.007

Davis, W.C., Grandoni, F., Mahmoud, E.M., Abdellrazeq, G.S., Fry, L.M., Mack, V., Marchitelli, C., Barile, V.L. 2017. Characterization of leukocyte subsets in buffalo (Bubalus bubalis) with cross-reactive monoclonal antibodies specific for bovine MHC class I and class II molecules and leukocyte differentiation molecules. Developmental and Comparative Immunology. 74:101-109. https://doi.org/10.1016/j.dci.2017.04.013.

Tebaldi, G., Williams, L.B., Verna, A.E., Macchi, F., Franceshi, V., Fry, L.M., Knowles Jr, D.P., Donofrio, G. 2017. Assessment and optimization of theileria parva sporozoite full-length p67 antigen expression in mammalian cells. PLOS Neglected Tropical Diseases. https://doi.org/10.1371/journal.pntd.0005803

Chungwon, J.C., Suarez, C.E., Bandaranayake-Mudiya, N.L., Bandaranayake-Mudiya, N., Rzepka, J., Heiniger, T.J., Chung, G., Lee, S.S., Adans, E., Yun, G. 2017. A novel modified-indirect ELISA based on spherical body protein 4 for detecting antibody during acute and long-term infections with diverse babesia bovis strains. Parasites & Vectors. 10(1):77. https://doi.org/10.1186/s13071-017-2016-9.

Johnson, W.C., Taus, N.S., Reif, K.E., Bohaliga, G., Kappmeyer, L.S., Ueti, M.W. 2017. Analysis of stage-specific protein expression during babesia bovis development within female rhipicephalus microplus. Journal of Proteome Research. 16(3):1327-1338. https://doi.org/10.1021/acs.jproteome.6b00947.

Olds, C.L., Paul, T., Scoles, G.A. 2016. Detection of theileria parva in tissues of cattle undergoing severe east coast fever disease show significant parasite accumulation in the spleen. Veterinary Parasitology. 232:32-35 https://doi.org/10.1016/j.vetpar.2016.11.012.

Olds, C.L., Mwaura, S., Odongo, D., Scoles, G.A., Bishop, R., Daubenberger, C. 2016. Induction of humoral immune response to multiple recombinant rhipicephalus appendiculatus antigens and their effect on tick feeding success and pathogen transmission. Parasites & Vectors. 9(1):484. https://doi.org/10.1186/s13071-016-1774-0.

Elnaggar MM, Abdellrazeq GS, Mack V, Knowles DP, Fry LM, Davis WC, Park KT. Characterization and use of new monoclonal antibodies to CD11c,CD14, and CD163 to analyze the phenotypic complexity of ruminantmonocyte subsets. Vet Immunol and Immunopathol. 2016;178:57-63. https://doi.org/10.1016/j.vetimm.2016.06.010.

Fry LM, Schneider DA, Frevert CW, Nelson DD, Morrison WI, Knowles DP. East Coast Fever caused by Theileria parva is characterized by macrophage activation associated with vasculitis and respiratory failure. PLOS ONE. 2016;11(5):e0156004. https://doi.org/10.1371/journal.pone.0156004

Fry LM, Snekvik KR, McBride R, Parish S. Sporadic juvenile lymphosarcoma in a set of triplet calves: Hematopoietic chimerism or chance? Vet Rec Case Rep. 2016;4:e000339. http://doi.org/10.1136/vetreccr-2016-000339.

Alzan, H.F., Lau, A., Knowles, D.P., Herndon, D.R., Ueti, M.W., Scoles, G.A., Kappmeyer, L.S., Suarez, C.E. 2016. Expression of 6-Cys gene superfamily defines babesia bovis sexual stage development within rhipicephalus microplus. PLoS One. doi:10.1371/journal.pone.0163791. https://doi.org/10.1371/journal.pone.0163791.

Suarez, C.E., Knowles, D.P., Sila, M. 2016. Identification of interchangeable cross-species function of elongation factor-1 alpha promoters in babesia bigemina and babesia bovis. Parasites & Vectors. doi:10.1186/s13071-016-1859-9. https://doi.org/10.1186/s13071-016-1859-9.

Oldiges, D., Laughery, J., Tagliari, N., Leite Filho, R., Davis, W., Da Silva, V.I., Termignoni, C., Knowles, D.P., Suarez, C.E. 2016. Transfected babesia bovis expressing a tick GST as a live vector vaccine. PLOS Neglected Tropical Diseases. doi:10.1371/journal.pntd.0005152. https://doi.org/10.1371/journal.pntd.0005152.

Rogovskyy, A.S., Nebogatkin, I.V., Scoles, G.A. 2016. Ixodid ticks in the megapolis of Kyiv, Ukraine. Ticks and Tick Borne Diseases. 8(1):99-102. https://doi.org/10.1016/j.ttbdis.2016.10.004.

Alzan HF, Knowles DP, Suarez CE. Comparative Bioinformatics Analysis of Transcription Factor Genes Indicates Conservation of Key Regulatory Domains among Babesia bovis, Babesia microti, and Theileria equi. PLoS Negl Trop Dis. 2016 Nov 10;10(11):e0004983. doi: 10.1371/journal.pntd.0004983. eCollection 2016 Nov. Review. https://doi.org/10.1371/journal.pntd.0004983.

Parasar P, Wilhelm A, Rutigliano HM, Thomas AJ, Teng L, Shi B, Davis WC, Suarez CE, New DD, White KL, Davies CJ. Expression of bovine non-classical major histocompatibility complex class I proteins in mouse P815 and human K562 cells. Res Vet Sci. 2016 Aug;107:161-170. doi: 10.1016/j.rvsc.2016.06.004. Epub 2016 Jun 8. https://doi.org/10.1016/j.rvsc.2016.06.004.

Adekanmbi EO, Ueti MW, Rinaldi B, Suarez CE, Srivastava SK. Insulator-based dielectrophoretic diagnostic tool for babesiosis. Biomicrofluidics. 2016 Jun 17;10(3):033108. doi: 10.1063/1.4954196. eCollection 2016 May. https://doi.org/10.1063/1.4954196.

Mahmoud MS, El-Ezz NT, Abdel-Shafy S, Nassar SA, El Namaky AH, Khalil WK, Knowles D, Kappmeyer L, Silva MG, Suarez CE. Assessment of Theileria equi and Babesia caballi infections in equine populations in Egypt by molecular, serological and hematological approaches Parasit Vectors. 2016 May 4;9:260. doi: 10.1186/s13071-016-1539-9. https://doi.org/10.1186/s13071-016-1539-9.

Hussein HE, Scoles GA, Ueti MW, Suarez CE, Adham FK, Guerrero FD, Bastos RG. Targeted silencing of the Aquaporin 2 gene of Rhipicephalus (Boophilus) microplus reduces tick fitness Parasit Vectors. 2015 Dec 2;8:618. doi: 10.1186/s13071-015-1226-2. https://doi.org/10.1186/s13071-015-1226-2.

APPENDIX 3

National Program 103 – Animal Health

ACCOMPLISHMENT REPORT 2016-2020

Over the last two 5-year cycles of NP 103, the following highlight NP 103 technology transfer outcomes:

Type	2011-2015	2016-2020
Inventions disclosed	65	63
Patents filed	34	45
Patents approved	8	28
Biological material inventions	8	28
Biological material licenses from inventions	8	26

APPENDIX 4

Research Collaborations

Number of Cooperative Research and Development Agreements (CRADAs): 10
Number of Trust Funds: 18
Number of Reimbursables: 21

External Grant Sources of Funding for National Program 103 Projects

2016-2020

Grants from universities usually involved cooperative research projects jointly conducted with university partners. In many cases this funding originated from third parties, such as the USDA National Institute for Food and Agriculture, National Institutes of Health, USAID, and other industry, non-profit, and government sources.

University	Government	International	Industry
29	31	18	51

Universities:

Instituto de Biotechnologia Y Biologia Molecular
Iowa State University
Kansas State University
Konkuk University
Michigan State University
Ohio State University
Pennsylvania State University
Rutgers University
Tennessee State University
University of Cambridge, United Kingdom
Universidade Estadual Paulista
University of California
University of Connecticut
University of Florida
University of Georgia
University of Illinois
University of Minnesota
University of Nebraska
Washington State University
Yale University

Research Organizations:

Bill and Melinda Gates Foundation
International Development Research Centre
John G Shedd Aquarium
National Marine Mammal Foundation
National Veterinary Research Institute, Nigeria
Sea World Parks and Entertainment, Inc
St. Jude Children's Research Hospital
Swine Health Information Center
United States-Israel Binational Agricultural Research and Development Fund (BARD)

Industries/Companies:

Adisseo
Alopexx Vaccine, LLC
Aptimune
Aviagen North America, Inc
Biocheck USA
Bio-x Diagnostics
Boehringer Ingelheim Animal Health
Codagenix, Inc
Dovetail Genomics, LLC
DSM
Elanco
Fuller Laboratories
Globion India Private, LTD
Gobroxia
HIPRA
Huvepharma
Merck and Company
Palantir Technologies
Pancosma
Park Systems, Inc.
Prevtec Microbia, Inc.
Proliant Biologicals, Inc.
Recombinetics, Inc.
Seppic
U.S. Biologic, Inc.
Zoetis

Government:

Animal and Plant Quarantine Agency, Korea
Centers for Disease Control and Prevention
Kansas Bioscience Authority
U.S. Department of Agriculture, Animal and Plant Health Inspection Service (APHIS)
U.S. Department of Agriculture, National Institute of Food and Agriculture (NIFA)
U.S. Department of Defense
U.S. Department of Health and Human Services, NIH National Institute of Allergy and Infectious Diseases
U.S. Department of Homeland Security
U.S. Department of the Interior, Fish and Wildlife Services
U.S. Navy, Office of Naval Research

Non-Government Organizations

Animal Health Research Institute, Taiwan
Egyptian Cultural and Education Bureau
Global Alliance for Livestock Veterinary Medicines (GALVMED)
National Pork Board
U.S. Poultry and Egg Association

Outgoing Funding to Support ARS Research Programs

2016-2020

Non-Assistance Cooperative Agreements

United States Universities
Auburn University
Case Western Reserve University
Iowa State University
Kansas State University
Loyola University of Chicago
Mississippi State University
Ohio State University
South Dakota State University
University of California
University of Connecticut
University of Delaware
University of Georgia
University of Illinois
University of Maryland
University of Minnesota
University of New Mexico
University of Wisconsin
Washington University, St Louis, Missouri
Washington State University

International Universities
Technical University of Denmark, National Veterinary Institute
The Royal Veterinary College of London
Universidade Del Pais Vasco
Universidade Federal do Rio Grande do Sul (UFRGS)
Universitat Pompeu Fabra

International Research Organizations
Canadian Food Inspection Agency
Centre de Recerca en Sanitat Animal
Friedrich-Loeffler Institut
Fundacion Aregeninta
Kenya Agricultural and Livestock Research Organization
Instituto Nacional de Tecnica Aerospacial
University of Copenhagen
Wildlife Science and Conservation Center, Ulaanbaatar

U.S. Companies
Conservation X Labs, Inc
Immport Therapeutics, Inc

International Companies
SAGARPA/SENASICA