Animal Health (NP 103) Annual Report for 2012

Introduction

Vision: The vision for ARS animal health research is to be a worldwide leader that delivers effective solutions to prevent and control animal diseases that impact agriculture and public health.

Mission: The mission of the Animal Health National Program (NP 103) is to conduct basic and applied research on selected diseases of economic importance to the United States livestock and poultry industries. The goals of the research mission are to produce knowledge and technology to reduce economic losses from infectious, genetic, and metabolic diseases. Cyril G. Gay and Eileen L. Thacker, National Program Leaders (NPL), Animal Health, manage the program.

The Animal Health National Program initiated the current five-year national program cycle Fiscal Year (FY) 2012. The Animal Health National Program currently includes 40 core research projects supported by 100 scientists located at 11 research sites throughout the country. The gross ARS research budget for the Animal Health Program FY 2012 was $66,682,000.

The following scientists in NP 103 received prominent awards in 2012:

Hans Cheng, Avian Diseases and Oncology Laboratory (ADOL), Lansing, Michigan, was the recipient of the 2012 Evonik Degussa Award for Achievement in Poultry Science. This prestigious award is given by the Poultry Science Association to distinctive contributions to poultry science advancement covering a period of not more than seven years preceding the annual award.

John Lippolis, National Animal Disease Center, Ames, Iowa, received the West Agro, Inc. Award, by the American Dairy Science Association (ADSA). The award is given to recognize outstanding research of milk quality as affected by control of mastitis, management of milking, and practices in production of milk.

Igseo Choi, Research Associate, Animal Parasitic Diseases Laboratory, Beltsville, Maryland, was recognized as the 2012 Young Scholar by the American Society of Animal Science (ASAS) and the Midwest Branch of the American Dairy Science Association. This award is given to recent PhD graduates who have made an outstanding contribution in research in animal and dairy science.

Mark Jenkins, Animal Parasitic Diseases Laboratory (APDL),,Beltsville, Maryland, was the recipient of the Beltsville Regional Senior Scientist Award for scientific excellence and innovative methodology in combating parasitic diseases in commercial poultry.

Donald Knowles, Animal Disease Research Unit (ADRU), Pullman, Washington, was the recipient of:

- 2012 Secretary Honor Award for Excellence (individual), for leading research on controlling re-emergent foreign animal diseases in the United States and in building a national and international research community that is training the next generation of agricultural scientists in the category of helping America promote sustainable agricultural production and biotechnology exports as America works to increase food safety.
- 2012 Federal Laboratory Consortium for Technology Transfer, Far-West Region, Outstanding Commercialization Success Award for control of a re-emergent exotic disease (Babesiosis).
- 2012 ARS Superior Sustained Effort Technology Transfer Award for "Control of a re-emergent exotic disease (Babesiosis) in the U.S. livestock through provision of modern diagnostics and chemotherapeutics."

Scientists within the National Animal Health Program were very active in their fields in FY 2012 with 88 articles published in peer-reviewed scientific journals. Many of the discoveries and findings were published in the popular press to reach our customers and stakeholders, including 79 articles in trade journals and book chapters. Technology transfer activities for the National Animal Health Program included 11 invention disclosures, 6 new Cooperative Research and Development Agreements (CRADA), 32 active Specific Cooperative Agreements (SCA), and 113 Material Transfer Agreements (MTA).

Research Results*:*
The following section of the report summarizes high impact research results addressing objectives in the current national program action plan.

Genetic Evolution of Novel Reassortant Swine Influenza Viruses with the Capability of Infecting Humans
Swine influenza A virus causes a respiratory disease in swine similar to that in humans. In collaboration with NIH scientists, ARS scientists at the National Animal Disease Center (NADC), Ames, Iowa, investigated the genetic evolution of novel reassortant swine influenza A viruses detected in the United States and Canada between 2009-2011 with a focus on H3N2 viruses. Analyses included H3N2 viruses designated A(H3N2) variant (v) because of their capability to also infect humans as recently discovered in the United States, July 2011. Analyses of samples from twelve human cases revealed that the variant swine-lineage H3N2 viruses contained the pandemic matrix (pM) gene from pandemic H1N1 viruses. The A(H3N2)v viruses are distinct from contemporary H3N2 circulating in humans and the flu viruses incorporated in the human seasonal flu vaccine, and hence represents a potential pandemic threat. Monitoring and reporting evolutionary dynamics of gene segments in swine at a detailed level is critical to understand how these

novel H3N2 viruses emerged in swine and to assess and predict the potential epidemic and/or pandemic threat of variant influenza viruses pose to humans.

The Discovery of a New Interferon and its Potential Application in the Control of FMD

Foot-and-mouth disease (FMD) is one of the most serious threats to the livestock industry. Despite the availability of vaccines, recent outbreaks in disease-free countries have demonstrated that development of novel FMD control strategies is imperative. ARS scientists at the Plum Island Animal Disease Center reported the identification and characterization of bovine (bo) interferon lambda 3 (IFN-λ3), a member of the type III IFN family. Expression of boIFN-λ3 using a replication-defective human adenovirus type 5 vector (Ad5-boIFN-λ3) yielded a glycosylated secreted protein with antiviral activity against FMD virus (FMDV) and vesicular stomatitis virus in bovine cell culture. Inoculation of cattle with Ad5-boIFN-λ3 induced systemic antiviral activity and up-regulation of IFN stimulated gene expression in multiple tissues susceptible to FMDV infection. The result of these studies also demonstrated that the type III IFN family is conserved in bovines and boIFN-λ3 has potential for further development as a biotherapeutic candidate to inhibit FMDV or other viruses in cattle.

Diagnostics to Detect a Newly Emerging Virus

HoBi like virus is a newly emerging type of virus distantly related to bovine viral diarrhea virus (BVDV) that has been isolated from cattle in South America, Southeast Asia, and Europe. The clinical presentation following infection with this type of virus is very similar to that seen following infection with BVDV. Like BVDV, HoBi-like viruses cause immune suppression and can establish life-long persistent infection in cattle. HoBi like viruses have not yet been detected in the United States. ARS scientists at the National Animal Disease Center (NADC), Ames, Iowa, have developed tests to provide diagnosticians and regulatory agencies with tools to screen imported animals and animal products to prevent introduction of HoBi like viruses into the United States. These tests have been transferred to APHIS and provide the means to detect and control an introduction if it were to occur in the United States.

Proposal for a Unified Nomenclature and Classification System of Newcastle Disease Virus Genotypes

Virulent Newcastle disease viruses (NDV) are found in most countries of the world. Although the United States has strict rules to prevent their entry, it is important to monitor and characterize viruses that are a potential threat to the U.S. poultry industry. ARS scientists at the Southeast Poultry Research Laboratory, Athens, Georgia, have obtained strains of NDV from Mexico, Indonesia, Malaysia, Venezuela, Pakistan, Vietnam, Belize, Dominican Republic, South Africa, Peru, and from wild birds in the United States and have sequenced and characterized them genetically. The sequences of key genes have allowed the prediction of the virulence of those viruses. This characterization has led to the identification of viruses of Asian lineages for the first time in the American continent (in Peru and in Venezuela) and to the identification of the expansion of the host range of North American virulent Newcastle disease from cormorants to other wild birds and the discovery of this type of viruses on the East Cost

of the United States (Massachusetts, Maine, New Hampshire, and Maryland). The discovery of novel NDV on the American continent provides an opportunity to improve the classification of these viruses. Historically, two systems have been simultaneously used to classify NDV isolates into lineages or genotypes, generating confusion in the nomenclature and discrepancies in the assignment of genetic groups. Based on the extensive characterization of NDV collected worldwide, ARS scientists have proposed a unified nomenclature and a classification system based on objective criteria to separate NDV into genotypes resulting in distinct taxonomic groups. Results revealed that class I viruses comprise a single genotype, while class II contains 15 genetic groups including 10 previously established (I-IX, and XI) and five new genotypes (X, XII, XIII, XIV and XV). Adoption of a unified nomenclature and of objective criteria to classify NDV isolates will facilitate studies on NDV epidemiology, evolution, disease control, and diagnostics.

Development of Alternative Strategies to Enhance Gut Immunity and Mitigate the Use of Antibiotics using Dietary Phytonutrients

Although widespread use of antibiotic-based growth promoters has improved the efficiency of worldwide poultry production, there is an increasing interest in developing alternative strategies to antibiotics to control infectious diseases in livestock and poultry due to the emergence of drug-resistant pathogens. ARS scientists in Beltsville, Maryland, investigated dietary phytogenics (cinnamon, garlic, and aloe vera) to enhance poultry immunity using avian coccidiosis as a disease model. Phytogenics are a group of natural growth promoters derived from herbs, spices, or other plants, and many medicinal foods and herbal products are highly effective in enhancing host defense against microbial infections. ARS scientists previously showed that phytogenics augment host immunity against infectious agents through their ability to alter gene expression. For example, Cinnamaldehyde (CINN) is a constituent of cinnamon that is widely used as a flavoring compound and has been used in some cases to treat human diseases, including inflammatory diseases. CINN has been reported to possess antioxidant, and antimicrobial activities, as well as being able to modulate T cell differentiation. In chickens fed a diet supplemented with CINN, the levels of interleukin (IL)-1 beta, IL-6, IL-15 and interferon-gamma transcripts in intestinal lymphocytes were 2- to 47-fold higher compared with chickens given a non-supplemented diet. Importantly, dietary CINN attenuated *Eimeria acervulina* and *E. maxima*-induced bodyweight loss, decreased *E. acervulina* oocyst shedding, and increased *E. tenella*-specific antibody responses compared with the non-supplemented control diet.

A Rapid Diagnostic Test for Pseudorabies Surveillance

Pseudorabies virus (PRV), the cause of Aujeszky's disease, was eradicated from U.S domestic swine herds but continues to circulate in the feral swine population and thus continues to pose a threat for the commercial swine industry. A critical need for the current PRV surveillance program in the United States is the rapid detection of PRV. ARS scientists validated a real-time PCR assay for pseudorabies virus surveillance. Real-time polymerase chain reaction (real-time PCR) is a valuable diagnostic technique that can rapidly identify infectious agents in clinical specimens. Diagnostic performance of the real-time PCR assay developed as a testing method confirmed that it is a rapid,

accurate assay that is adaptable to a variety of PCR platforms currently in use by diagnostic laboratories around the world and can provide reliable results on an array of clinical samples.

A Genetic Marker Associated with Reducing Susceptibility to Porcine Reproductive and Respiratory Syndrome (PRRS)

A genetic marker for reduced susceptibility to PRRS, the most economically significant disease in pigs, has been discovered by a research team that includes scientists from ARS in Beltsville, Maryland, Kansas State University and Iowa State University. This project was funded by the USDA National Institute for Food and Agriculture (NIFA). PRRS affects pigs at all stages of growth and is easily spread. PRRS costs the United States alone an estimated $642 million per year. The PRRS Host Genetics Consortium (PHGC) was established with funds from the U.S. National Pork Board to discover the genetic basis of host resistance or susceptibility to PRRS virus infection. Groups of 200 commercial crossbred pigs were infected with PRRS virus and followed for 42 days; blood samples and body weights were collected for detailed viral load and weight gain "phenotypes." Ear notches were used to prepare genomic DNA and using Porcine 60K SNP Beadchip generating genotypes on more than 60,000 genetic markers or single nucleotide polymorphisms (SNPs) across the genome for each pig. Using these data, the entire genome of all pigs from the first three PRRS Host Genetics Consortium **(**PHGC**)** trials were searched to identify chromosomal segments that were common to pigs that had lower virus levels and faster growth after infection. This resulted in the discovery of the genetic marker, called a quantitative trait locus (QTL), on swine chromosome 4 (SSC4) associated with improved growth of pigs that are infected with the PRRS virus. In fact this region on chromosome 4 was shown to be associated with both weight gain (WG) and viral load (VL) - 15.7% of the genetic variance for VL and 11.2% for WG. Genomic estimated breeding values (GEBV) for this SSC4 region were perfectly and favorably correlated at -1; i.e., the desired effect when virus decreased, weight increased. Now that scientists have found a genetic region, the next step is to pinpoint the gene and determine whether it shows the same effects for other strains of the PRRS virus. These results could have a major impact in the swine industry by enabling geneticists to develop plans for marker-assisted selection of pigs with improved response to PRRS.

The U.S. Veterinary Immune Reagent Network

The U.S. Veterinary Immune Reagent Network (US VIRN, www.vetimm.org) was established to address the lack of immunological reagents specific for livestock and poultry species. Efforts are targeted at swine, ruminants, poultry, equine, and aquaculture species. ARS scientists have led the teams for swine and poultry and have successfully developed and characterized bioactive immune proteins, cloned cytokine and chemokine proteins, as well as monoclonal antibodies (mAbs) to these proteins and their receptors and immune cells. These reagents will be used to evaluate swine and poultry immune responses, changes after infections or following vaccination, and give scientists the ability to manipulate these immune proteins, and cell populations to evaluate their roles in protective immunity, immunoregulation, and immunopathology. Recombinant cytokines and chemokines for swine and poultry were cloned and expressed in yeast, purified, and shown to be bioactive. All immune proteins developed in this proposal are

available to collaborators and have been made commercially available through our U.S. VIRN partner, Kingfisher Biotech, Inc. http://www.kingfisherbiotech.com/. Another goal is to produce mAb reagents that function in different diagnostic platforms. Overall the U.S. VIRN projects are important as a means of identifying new reagents and technologies for veterinary diseases and diagnostic and vaccine discovery research.

Gold and Nanotechnology Bring Viruses to Light

Gold nanoparticles have the ability to scatter and absorb light, making them ideal in detecting virus-infected cells. Using a technology called surface-enhanced raman scattering (SERS), signals emitted from these nanoparticles can be measured using a spectrometer. ARS scientists at the Arthropod-Borne Animal Diseases Research Unit (ABADRU), Manhattan, Kansas, and collaborators at the University of Wyoming used gold nanoparticles to design tests to rapidly identify West Nile virus, a virus spread by infected mosquitoes that can cause headaches, fever, flu-like syndrome, and sometimes fatal neuroinvasive disease—aseptic meningitis, encephalitis, or acute flaccid paralysis. The goal of the project is to bring laboratory-level analytical sensitivity to the field for portable care devices. If successful, veterinarians and medical doctors will be able to take a blood sample, put it in a small vile and read it with a hand-held device. ARS scientists are also working on adapting this technology to identify multiple disease agents.

Pathologic and Biochemical Characterization of a Genetic Form of Bovine Spongiform Encephalopathy (BSE)

Transmissible spongiform encephalopathies (TSE) such as BSE are characterized by a novel transmissible "infectious" protein called a prion that converts the cellular form of the prion protein (PrP^c), normally expressed by many cells in the body, to a misfolded, disease-associated form (PrP^d) that causes pathological lesions in the central nervous system. The complete pathologic and biochemical features of a genetic form of BSE were defined and reported for the first time by ARS scientists at the National Animal Disease Center, Ames, Iowa. The genetic form of BSE is analogous to the most prevalent hereditary form of human TSE. Heritable BSE along with spontaneous BSE forms are also referred to as atypical BSE cases which have important implications in that they are not associated with the feedborne epidemic of classical BSE first recognized in the United Kingdom in the 1980s. Atypical BSE cases emphasize the need to maintain the specified risk material ruminant feed ban as a science-based policy to prevent a feedborne epidemic from developing; the feedborne nature of the classical BSE epidemic has been demonstrated to negatively impact export markets in various countries around the world, whereas atypical BSE does not connote the same concern.

Development of a Rapid Method for Detection of Disease-Associated Prions

A method for the detection of PrP^d in formalin-fixed paraffin-embedded tissue by ELISA has been developed and described by ARS scientists at the National Animal Disease Center, Ames, Iowa. Methods for diagnosis of transmissible spongiform encephalopathies (TSEs) in cattle, sheep, and cervids have traditionally depended on the availability of both frozen fresh and formalin-fixed tissues. However, in many diagnostic sample submissions only formalin-fixed samples have been available for TSE diagnosis, a situation that previously precluded analysis by rapid diagnostic procedures such as

ELISA. This work describes a method suitable for extraction of the PrP^d from formalin-fixed paraffin-embedded tissue for detection by ELISA. This represents a significant advancement for diagnostic laboratories and provides a rapid alternative method for TSE detection beyond immunohistochemistry (IHC).

A Genetic Marker Associated with Resistance to Scrapie

The amino acid, lysine, at position 171 of the sheep prion protein delays development of scrapie. ARS scientists demonstrated the effect of the amino acid lysine at position 171 of the sheep prion protein on susceptibility to scrapie, a transmissible spongiform encephalopathy of sheep. Amino acid differences in the prion protein are known to play a major role in scrapie susceptibility in sheep and these genetic differences are utilized in the strategy to remove scrapie from our nation's sheep flock. Natural scrapie had previously only been described in one sheep with lysine at position 171 of the prion protein, hence not enough information was available from natural cases to determine the affect of lysine at position 171 on scrapie susceptibility. ARS scientists at the National Animal Disease Center, Ames, Iowa, demonstrated that sheep with a prion protein containing lysine at position 171 are susceptible to scrapie but have a prolonged scrapie incubation period, and that the abnormal prion protein accumulates throughout the central nervous system and lymphoid organs. Because sheep with lysine at prion amino acid position 171 develop scrapie at a slower rate than other known susceptible genotypes this information is critical to sheep breeders that want to eradicate genotypes susceptible to scrapie.

Prion infectivity in scrapie-infected sheep and goat blood

ARS researchers at the Animal Disease Research Unit in Pullman, Washington, have identified the components of sheep's and goat's blood which carry prion infectivity by using the sensitive technique of transfusion bioassay. The presence of infectious scrapie prions in the blood indicates the possibility of developing a blood-based diagnostic test but currently available immunoassays do not appear to be sensitive enough for robust detection in samples of whole blood. The insights gained are an important step toward optimizing the isolation of the blood components most relevant to early disease detection by immunoassay in both sheep and goats.

Prolonged scrapie incubation in goats linked to two genetic markers

ARS scientists performed the first oral scrapie challenge of goats heterozygous for two PrP^c polymorphisms (commonly referred to as genetic variation or alleles) of particular interest to scrapie susceptibility. Scrapie eradication in sheep is based in part on strong genetic resistance to classical scrapie. Goats may serve as a scrapie reservoir but there has been limited experimental inoculation to confirm strong genetic resistance in goats. The results confirmed that goats with mutations at two PrP^c alleles (S142 or K222) have greatly extended incubation times, indicating a need in scrapie-eradication programs for longer trace-back histories for goats bearing these alleles. Also indicated is a need to assess goats for either of these alleles for enhanced resistance to scrapie infection.

Swine influenza vaccine-associated enhanced respiratory disease (VAERD) characterized in pigs

Influenza A virus causes a respiratory disease in pigs similar to that in humans. Inactivated influenza virus vaccine use in swine has increased over the past 10 years in an effort to prevent disease and transmission of the virus. Inactivated vaccines work well when pigs are exposed to influenza viruses that are the same virus used to produce the vaccine; however, vaccine efficacy is reduced when pigs are infected with different or new strains. ARS scientists at the National Animal Disease Center in Ames, Iowa, found that pigs administered an inactivated swine influenza A vaccine followed by infection with the pandemic human influenza A virus (2009) demonstrated more severe disease compared to non-vaccinated pigs infected with the same virus. Pigs with VAERD demonstrated greater percentages of affected lungs compared to controls, the microscopic damage was more severe with distinct lesions, and had elevated immune factors associated with inflammation and disease in the lungs. Active surveillance and monitoring of the quality of match between vaccine strains and strains infecting swine herds is needed to prevent vaccine mismatch and VAERD in commercial swine. Future vaccines that stimulate improved immune responses for differing influenza viruses will be important to prevent infection and clinical disease in commercial swine production, as well as potential virus transmission to humans.

Determination of an Effective Bison Brucellosis Vaccine Strategy

Brucellosis is a disease of production livestock that can cause devastating economic losses to producers and can also cause severe illness in humans. Since being initiated in 1934, billions of dollars have been spent on the U.S. Cooperative Brucellosis Eradication Program in efforts to eradicate the disease. However, the persistence of brucellosis in wildlife reservoirs (bison, elk, feral swine, and other species) and in neighboring countries poses a risk for reintroduction to domestic livestock. Although all 50 states have maintained brucellosis-free status for cattle since 2009, the infection of 3 herds in Wyoming in 2010 demonstrates the risk associated with wildlife reservoirs of *Brucella abortus* in the Greater Yellowstone Area (GYA). There is a high prevalence of brucellosis in free-ranging bison in Yellowstone National Park. Any vaccination program of bison will be difficult and expensive so determination of the most efficacious brucellosis vaccine strategy for bison is needed. ARS researchers at the National Animal Disease Center in Ames, Iowa, evaluated the safety, immunity, and protection after bison were vaccinated with a single RB51 injection, vaccinated 4 times by injection, or a single pneumatic vaccination with a needleless system. Measurements of immunity were similar across treatments and did not support the hypothesis that multiple vaccinations would induce greater responses. Experimental protection was similar for the single injection and pneumatic vaccination treatments, but data suggested that bison receiving multiple vaccinations had reduced protection. These data suggest that too frequent vaccination of bison calves with RB51 may actually reduce protection against brucellosis. In addition, results from this study suggest that pneumatic vaccination can be a safe, effective, and needle-less procedure for disease prevention in bison.

Development of Blood Based Diagnostic Tests for Bovine Tuberculosis
The USDA initiated a bovine tuberculosis eradication campaign in 1917. Significant progress has been achieved, but eradication has proved elusive. Obstacles to eradication include lack of rapid, sensitive, and specific blood based diagnostic tests for cattle and the wildlife reservoirs which serve as a source of tuberculosis infection for cattle. Development of improved tuberculosis (TB) diagnostic tests serves both the cattle and captive cervid industry. Novel diagnostic tests allowing testing for TB in a single handling event, thus decreasing stress and potential for injury to animals or humans as well as speed of results is critical in meeting this important need. ARS scientists at the National Animal Disease Center, Ames, Iowa, developed two new blood based assays including a Enzyme-Linked Immunosorbent Assay (ELISA) for diagnosing TB in cattle and white tailed deer. This will increase the ability to quickly diagnose infected animals that will assist in eradication of this important zoonotic disease.

Development of a Pasteurella multocida Vaccine for Cattle Respiratory Disease
Respiratory disease of beef and dairy cattle is considered the most costly disease facing producers today with estimated costs exceeding $1 billion annually. A recent National Animal Health Monitoring System (NAHMS) survey confirmed that respiratory disease continues as the leading cause of morbidity and mortality in U.S. feedlots and is the most common cause of weaned dairy heifer mortality. The availability of new, more effective measures to prevent and control respiratory disease threats will have a significant impact on the future of the cattle industry. The etiology of Bovine Respiratory Disease (BRD) is complicated by numerous factors of stress and multiple bacterial and viral infections. Normal cattle may carry one or more of the bacterial and viral agents in their upper respiratory passages with no apparent ill effects. Some of these agents produce only mild clinical signs by themselves, but when combined with other viral or bacterial agents and/or stress they may cause severe clinical disease and even death. Pneumonic pasteurellosis caused by *Pasteurella multocida* is an ongoing cause of multi-million dollar losses to the beef and dairy cattle industries. Two new modified-live vaccine strains of *P.multocida* were constructed by ARS researchers at the National Animal Disease Center, Ames Iowa, which may be delivered to calves by conventional injection or by an intranasal route. Effective vaccines to control *P. multocida* in susceptible cattle populations will save livestock producers millions of dollars annually while reducing therapeutic antibiotic usage and increasing the quality of beef products.

Reduced Bacterial Clearance from the Lungs of Bighorn Sheep
Respiratory pathogens of domestic sheep including *Mannheimia haemolytica* (*Mh*) lead to millions of dollars in production losses annually. In addition to production losses in domestic sheep, *Mh* is an important bacterial pathogen of bighorn sheep. Data indicate that domestic sheep transmit pathogenic subtypes of *Mh* to bighorn sheep, and a currently debated intervention is separation of domestic from bighorn sheep areas. However, the current separation strategy consisting of removing domestic sheep grazing rights in the West economically threatens a large fraction of the U.S sheep industry, and additional intervention strategies are urgently needed. Researchers from Washington State University and ARS scientists at Pullman, Washington, administered equivalent doses of *Mh* simultaneously to bighorn and domestic sheep and the bighorn sheep failed to remove

the bacteria from their lungs as quickly or as thoroughly as the domestic sheep. As a result, bighorn sheep tend to have more severe lung damage associated with *Mh* pneumonia than domestic sheep, but the reasons for lack of bacterial clearance is not clear. These findings did not rule out that an overactive immune response as part of the explanation for differing lung damage between these closely related species. However, the differences in bacterial clearance between the two sheep species provide baseline data for future studies to determine potential explanations for the severity of respiratory disease in bighorn sheep that may lead to new and novel intervention strategies.

Development of a Monoclonal Antibody Specific for a Johne's Protein

Johne's disease (Paratuberculosis) is a chronic, progressive enteric disease of ruminants caused by infection with *Mycobacterium avium* subsp. *paratuberculosis* (MAP). Cattle become infected as calves, yet usually do not develop signs of diarrhea and weight loss until 2 to 5 years of age. During the subclinical phase of disease, animals may intermittently shed the organism in their feces, thereby contaminating the environment and infecting other animals within the herd. Current diagnostic tests are not able to identify these subclinically infected animals accurately. In order to prevent the further spread of this disease, improved diagnostic tools for the detection of infection and development of new vaccines to enhance control strategies are needed. ARS researchers at the National Animal Disease Center, Ames, Iowa, identified a new monoclonal antibody that selectively detects MAP and not other closely related bacterial strains. This antibody is currently the only one in the world that has this capability. Further research identified the protein, termed MAP1025 that this specific antibody binds. This antibody is the subject of a recently issued U.S. patent. The identification of this unique protein and antibody for MAP will be helpful in developing new diagnostic tools to detect infected animals.

Genetic Resistance to Nematode Infection in Cattle

Gastrointestinal nematode infections in ruminants remain a major impediment to the efficient production of both meat and dairy products and an important factor in constraining global food availability. Effective parasite control strategies are currently limited and based largely upon heavy drug usage. The treatments can be expensive and labor intensive, thus reducing profitability. There has been a recent increase in parasite resistance to anti-parasitic drugs making development of new strategies imperative. In order to develop new and novel strategies, the use of bioinformatic tools enables us to better understand host-parasite relationships. ARS scientists in Beltville, Maryland, characterized the response of the abomasal transcriptome to gastrointestinal parasites in parasite-susceptible and parasite-resistant Angus cattle using RNA-seq technology. These cattle displayed distinctly separate resistance phenotypes as assessed by fecal parasite egg counts. After assessing 15,432 bovine genes expressed in the abomasum (final stomach compartment), 64 genes were identified as significantly over-expressed in resistant cattle. Several specific biological pathways were found to be impacted in resistant animals suggesting their potential involvment in the development of parasite resistance. Understanding the mechanisms used by either the host or the parasite to resist or reduce parasites will enable the development of alternative strategies of control, one of which could include using genetic selection in the host.

Determination of a New Tick Vector for Equine Babesiosis

The insidious reemergence of equine babesiosis or piroplasmosis in 2009 in Texas is a poignant reminder of the vigilance required to maintain an infection-free status in the United States. Equine piroplasmosis has been considered a foreign animal disease in the United States since the 1980's. Ticks native to the United States such as *Dermacentor variabilis* and *Riphicephalus microplus* are known vectors of *Babesia equi,* the causative agent of equine babesiosis. ARS scientists at Pullman, Washington, in collaboration with APHIS-National Veterinary Service Laboratory, Ames, Iowa, and APHIS-Western Regional Office, Fort Collins, Colorado, identified the native U.S. tick, *Amblyomma cajennense,* as the predominant tick species found on horses at the Texas outbreak and capable of transmitting *B. equi*. In addition, it has the potential to geographically expand within the United States due to acaricide resistance and distribution by infected horses and wildlife adding an additional risk factor. Persistently infected horses are the reservoirs for tick acquisition of *B.equi* and its transmission to susceptible horses. Understanding the competency of the U.S. native ticks is critical to designing an efficient control strategy to prevent the dissemination of equine piroplasmosis within the United States and avoid great economic burdens to the U.S. horse industry.