

COMMITTEE ON PARASITIC AND VECTOR-BORNE DISEASES

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The Committee met on October 30, 2019 at the Rhode Island Convention Center in Providence, Rhode Island, from 8:00AM to 12:00PM. There were 40 members and 35 guests present.

Diane Kitchen, Committee Chair, called the meeting to order at 8:00AM and began the Committee meeting by introducing herself and vice-chair T.R. Lansford followed by a presentation of the Committee mission. Dr. Kitchen asked attendees to sign-in, reviewed the Committee agenda, and provided instruction regarding adherence to Robert's Rules of Order during the business meeting. She also reminded attendees that only Committee members are eligible to vote on business items. Lastly, Dr. Kitchen announced that the Committee would be considering one resolution during the business meeting.

A motion (moved and seconded) to adjourn the Committee was made and passed at 12:07PM.

Presentations & Reports

Asian Longhorned Tick 2019

Thomas McKenna, D.V.M., USDA-APHIS-Veterinary Services

The Asian longhorned tick (ALHT) had no known established populations in the United States before finding it in New Jersey in 2017. It is a serious pest of livestock in the Australasian and Western Pacific Regions where it occurs. It is an aggressive biter and frequently builds intense infestations on domestic hosts causing great stress, reduced growth and production, and severe blood loss.

Since last year at this time, there is an increase from 9 to 12 states reporting ALHT infestations.

In 2019, there are the first reports in the US of probable livestock death from this tick feeding in mass, more hosts have been recorded (including chickens), and pathogen testing and transmission experiments are ongoing but no pathogens have been detected in US ALHT to date. [CDC transmission experiments show they don't transmit *Borrelia burgdorferi* \(AKA Lyme disease\)](#).

Research Update - The Arthropod-Borne Animal Diseases Research Unit (October 2019)

Leela Noronha, Steve Behan, Lee Cohnstaedt, Barbara Drolet, Dana Mitzel, Dana Nayduch, William Wilson; USDA – Agricultural Research Service, Arthropod-Borne Animal Diseases Research Unit (ABADRU), Center for Grain and Animal Health Research (CGAHR)

The research mission of the Arthropod Borne Animal Diseases Research Unit (ABADRU) is to solve major endemic, emerging, and exotic arthropod-borne disease problems in livestock. The Unit is located at the Center for Grain and Animal Health Research (CGAHR) in Manhattan, Kansas. ABADRU research falls under ARS National Research Programs NP103: Animal Health and NP104: Veterinary, Medical, and Urban Entomology. The multidisciplinary team of nine senior scientists (two vacant) lead research ranging from vector biology to virus-vector-host interactions. Significant updates in ABADRU's major research programs are highlighted below.

Adult house flies frequent microbe-rich sites such as dumpsters and animal manure for feeding and reproductive purposes. Flies become contaminated with and ingest a wide variety of bacteria which can be disseminated to other locations, including human habitation. We investigated total culturable bacteria and coliform abundance in male and female house flies collected from two environments: urban (restaurant dumpsters) and agricultural (dairy farm). We hypothesized that female flies would harbor more bacteria due to their increased association with substrates for oviposition, and that coliform abundance would be greater in agricultural flies where abundant manure is accessible. Overall, female flies harbored more bacteria than males and there was a sex by site interaction with sex effects present at the urban location. Coliform abundance did not differ by sex, site, or by sex within site. House flies carried antimicrobial resistant (AMR) strains of bacteria: 37/39 isolates were resistant to 1 or more antimicrobials and 65% of AMR strains were resistant to 4 or more antimicrobials.

Rift Valley fever (RVF) virus (RVFV) is an exotic zoonotic pathogen which poses a significant arthropod-borne animal disease threat to U.S. livestock if introduced. The rate which RVFV exchanges gene segments (reassorts) was investigated in cell-culture and sheep—a target host species. Methods to detect reassortants were developed including a reverse transcriptase-polymerase chain reaction (RT-PCR) with melt curve analysis assay to distinguish between distinct viral lineages. Plaque purified reassortants were then confirmed by sequence analysis. An additional method was developed to detect low copy genetic targets. Development and evaluation of improved diagnostic tools for Rift Valley fever have also continued. A multiplex pathogen detection assay using the Fluorescence Microsphere Immunoassay (FMIA) was evaluated in an RVFV-endemic country, Kenya, for use in diagnostics and surveillance. The RVF competitive Enzyme Linked Immunosorbent Assay (cELISA) developed through a three-way collaboration (ARS, Kansas State University, and Texas A&M) has demonstrated reliable sensitivity and specificity and has been packaged commercially. Improvements in pathological tools for RVFV were made including the establishment of methods to detect RVF viral RNA and proteins in fixed tissues. In terms of RVF countermeasures, there are currently no antiviral treatments for RVFV; however, we have identified two potential candidates. Work is ongoing to understand the mechanisms of the candidate antiviral effects and to identify other potential small molecules RVFV antivirals.

Japanese encephalitis virus (JEV) is one of the most important etiologic agents for encephalitis worldwide. The virus is maintained in a cycle between culicine mosquitoes and vertebrate hosts. Work with wild-type JEV is strictly controlled and is limited to a Biosafety level-3 containment environment which is used for pathogens that can cause serious or lethal disease. The vaccine strain of JEV can be studied in lower containment lab environments which are common throughout the United States. The vaccine strain is known to grow in mosquito cells. However, the cell line that is typically used is not representative of the mosquito genus associated with most JEV vectors. The JEV vaccine strain was utilized to infect two cell lines derived from culicine mosquitoes. One cell line was derived from *Culex quinquefasciatus* which are known to be competent vectors for the virus in areas of Asia. The second cell line was derived from a species in the United States that has been shown to be a competent vector in a lab setting. Replication studies demonstrated that the virus produced peak titers after 2-3 days of infection and that the virus did not cause cytopathic effects to the cells. These data suggest that this system could be used as a surrogate for the more virulent viruses in studies examining the molecular mechanisms of the virus-vector interaction important for replication and maintenance in the culicine mosquitoes.

Female *Culicoides sonorensis* biting midges are vectors of epizootic hemorrhagic disease virus (EHDV), which causes morbidity and mortality in wild and domesticated ruminants. Key changes in female midge transcriptome profiles occurring during early infection with EHDV-2 were identified. In midges fed bloodmeals containing EHDV-2, 2401 unigenes were differentially expressed compared to midges were fed negative control bloodmeals; approximately 60% were downregulated in response to the virus (953 up; 1448 down). Downstream Gene Ontology enrichment, KEGG pathway mapping, and manual analyses were used to identify the effect of virus ingestion at both the gene and pathway levels. Downregulated unigenes were predominantly assigned to pathways related to cell/tissue structure and integrity (actin cytoskeleton, adherens junction, focal adhesion, hippo signaling), calcium signaling, eye morphogenesis and axon guidance. Unigenes attributed to sensory functions (especially vision), behavior, learning and memory were largely downregulated. Upregulated unigenes included those coding

for innate immune processes, olfaction and photoreceptor pigments. The results of this project suggested that midges respond to virus infection as soon as 36 h post-ingestion, and that EHDV-2 may have a significant phenotypic effect on sensory and neural tissues

In 2012, a major EHDV outbreak occurred in the US following a summer of severe drought and abnormally high temperatures. In addition to large losses of white-tailed deer, the Midwest and northern Plains saw a significant amount of clinical disease in cattle. Although EHDV-1, -2 and -6 were isolated, EHDV-2 was the predominant virus serotype detected. Phylogenetic analyses and sequence comparisons of newly sequenced whole genomes of 2012 EHDV-2 cattle isolates demonstrated that eight of ten EHDV-2 genomic segments showed no genetic changes that separate the cattle outbreak sequences from other EHDV-2 isolates. Two segments, VP2 and VP6, did show several unique genetic changes specific to the 2012 cattle outbreak isolates, although the impact of the genetic changes on viral fitness is unknown. The placement of isolates from 2007 and 2011 as sister group to the outbreak isolates, and the similarity between cattle and deer isolates, point to environmental variables as having a greater influence on the severity of the 2012 EHDV outbreak than viral genetic changes.

Bluetongue virus (BTV) is transmitted by biting midges (*Culicoides*) and causes disease in domestic and wild ruminants. Transmission of viruses by insects is a complex mechanism. Insects must obtain virus from an infected animal during blood feeding. The virus then must replicate itself and disseminate within the insect so that it reaches the salivary glands to be excreted into another animal when the insect feeds again. In collaboration with researchers in The Netherlands, we used genetic approaches to show that small changes in one specific protein of the virus, the NS3 protein, significantly affected its ability to replicate in insects to a point where they would not be transmitted. Such large effects from such small genetic changes helps explain why virus strains that are very similar, may not be transmitted similarly.

Vesicular stomatitis (VS) is a veterinary viral disease of cattle, horses, and swine. In the U.S., VS produces devastating economic losses, particularly in the southwestern states where the outbreaks display an occurrence pattern of 7-10-year intervals. To date, the mechanisms of geographic spread and maintenance cycles during outbreaks remain unclear. This is due, in part, to the fact that VS epidemiology has a complex of variables to consider, including a broad range of vertebrate hosts, multiple routes of transmission, and an extensive diversity of suspected vector species acting as both, mechanical and biological vectors. Infection and viral progression within vector species are highly influenced by virus serotype, as well as environmental factors including temperature and seasonality; however, the mechanisms of viral transmission, including non-conventional pathways, are yet to be fully studied. In collaboration with researchers at Kansas State University, ABADRU wrote a comprehensive review of VS transmission mechanisms, with comparisons of transmission evidence for the four most incriminated hematophagous insects: *Aedes* mosquitoes, *Lutzomyia* sand flies, *Simulium* black flies, and *Culicoides* biting midges. This provides a single, comprehensive source for livestock owners to evaluate and understand the current knowledge of VS epidemiology and insect transmission, and thereby better understand their risk of this animal disease based on vector populations present on their premises.

Currently, biting midge population management using pesticides is the best method to reduce contact between the blue tongue, VS, and EHD disease vector biting midges and domesticated animals. ABADRU has used the USDA biting midge colony in pesticide assays to determine which pesticides are the most efficacious for killing biting midges. The Centers for Disease Control bottle bioassay was used to evaluate various pyrethroids, organophosphates, and carbamates to determine the best pesticides for areal application. All the products tested worked well, although pyrethroids had the fastest knockdown. The insects did show surprising resilience to recover or regain movement 24 hours post exposure to pesticides, suggesting an ability to metabolize the active ingredients. These assays will also be used to detect pesticide resistance. A shift in the mortality curves (a loss of product efficacy) will indicate the evolution of resistance to that active ingredient. Furthermore, larval habitat treatments were tested to determine which products reduced adult emergence. Insect growth regulators worked the best and at the lowest concentrations for larval habitat treatments.

Orbivirus Activities at NVSL

Dr. Albert van Geelen, USDA/APHIS/VS/NVSL

Report provided to cover the testing summary for 2018 at NVSL for Bluetongue virus (BTV) with a discussion of the options for PCR and follow-up typing. NVSL does not perform typing of BTV unless specifically requested and CT values over 30 may require that culture is performed prior to typing.

BTV Identifications at NVSL during 2018

State	Serotype	Species	Number
AZ	BTV-13 ^{1), 2)}	Mule deer	3
AZ	BTV-17	Mule deer	1
CA	BTV-17	Mule deer	1
CA	BTV-17	Cattle	2
CA	BTV-17	Sheep	1
CA	BTV-17	Pronghorn	1
NE	BTV-17	Bighorn sheep	1
NV	BTV-17	Pronghorn	2
NV	BTV-17 ³⁾	Elk	1
OR	BTV-17	Cattle	3
OR	BTV-17	Goat	1
OR	BTV-17	Sheep	5
WA	BTV-17	Sheep	1
WY	BTV-13 ⁴⁾	Cattle	1
WV	BTV-1 ⁵⁾	White-tailed deer	1

¹⁾ All 3 animals were co-infected with EHDV-2.

²⁾ Co-infected with BTV-13 and BTV-17.

³⁾ Co-infected with EHDV-2.

⁴⁾ First detection of BTV-13 in WY although it is known from neighboring states.

⁵⁾ Isolated and submitted by Dr. Stallknecht from UGA.

2018 EHDV Identifications at NVSL

State	Serotype	Species	Number
AZ	EHDV-1 + EHDV-6	Mule deer	1
AZ	EHDV-1 ^{1), 2)}	Mule deer	5
AZ	EHDV-2	White-tailed deer	1
IA	EHDV-2	White-tailed deer	4 ³⁾
IA	EHDV-2	Elk	2
MN	EHDV-2	White-tailed deer	1
NV	EHDV-2 ⁴⁾	Pronghorn	1
NV	EHDV-2 ⁷⁾	Elk	1
OR	EHDV-2 ⁵⁾	Cattle	1
UT	EHDV-2 ⁶⁾	Cattle	2
WY	EHDV-2	White-tailed deer	1
WY	EHDV-2	Mule deer	1

¹⁾ Four of the animals were co-infected with BTV.

²⁾ First identification of EHDV-1 in AZ although it was repeatedly detected in neighboring states.

³⁾ A part of a larger outbreak in cervids with typing of randomly-selected samples.

⁴⁾ First identification of EHDV-2 in NV although present in neighboring states.

⁵⁾ First identification of EHDV-2 in OR although present in neighboring states.

⁶⁾ First identification of EHDV-2 in UT although it is present in neighboring states.

⁷⁾ Co-infected with BTV-17.



BTV Identifications (Jan-Sept 2019) at NVSL

State	Serotype	Species	Number
CA	BTV-17	Sheep	1
CA	BTV-17	White-tailed deer	1
CA	BTV-13	Cattle	1
FL	BTV-18	White-tailed deer	1 ¹⁾
FL	BTV-18	Cattle	2
FL	BTV-19	Cattle	1
FL	BTV-24	White-tailed deer	1 ¹⁾
FL	BTV-6	Goat	1
IA	BTV-11	Cattle	1

¹⁾ Isolated and submitted by Dr. Stallknecht from UGA.

EHDV Identifications at NVSL: Jan – Sept 2019

State	Serotype	Species	Number
CA	EHDV-2	Cattle	1
IA	EHDV-2	White-tailed deer	10
IA	EHDV-6	Cattle	1
MN	EHDV-2	White-tailed deer	12

EIA/EP – Updates and Illegal Movements

Angela Pelzel-McCluskey, DVM, MS; USDA-APHIS-Veterinary Services

More than 22,000 U.S. horses have been tested for EP so far during the 2019 calendar year with 65 *T. equi*-positive horses found in 6 states. All 65 EP-positives are Quarter Horse racehorses with iatrogenic transmission of the disease either suspected or confirmed. Thirteen (13) of these horses were found to be dually infected with both EP and EIA. Many of the 65 EP-positives were confirmed to be participating either currently or previously in unsanctioned racing which remains a key risk factor for exposure to the disease. Several of these horses also had a history of illegal movement from Mexico. The horses that were co-infected with both EP and EIA have been euthanized and many of the remaining EP-positive horses have been enrolled in the USDA-APHIS EP Treatment Program. All EP-positive horses will remain quarantined until permanent clearance of *T. equi* through high-dose imidocarb dipropionate treatment is achieved and the horse maintains *T. equi*-negative status on all diagnostic testing. To date, there have been 323 horses treated in the U.S. for EP with 276 horses having met the clearance and test negative criteria for quarantine release.

While total test numbers for EIA testing in 2019 have not yet been compiled, they are expected to be comparable to the total tests conducted in 2018 in which more than 1.2 million U.S. horses were EIA tested. So far in 2019, there have been 86 horses confirmed as EIA positive in 17 states. At least 70 of the 86 EIA-positive horses are Quarter Horse racehorses with iatrogenic transmission of the disease either suspected or confirmed. Many of the EIA-positive horses were found to be participating in unsanctioned racing. One of the 86 EIA-positive horses was a Thoroughbred racehorse from Florida participating in sanctioned racing and is suspected to have acquired the infection during a period of injury layup in which an unidentified platelet-rich plasma (PRP) product was administered to the horse by a foreign veterinarian not licensed in the U.S. It is suspected that the PRP product may have been illegally brought into the U.S. from another country. The same unlicensed foreign veterinarian is also linked to PRP treatment of a Thoroughbred racehorse found EIA-positive in Florida in 2017.

Several in-depth EP/EIA case studies, including the EIA-positive Florida Thoroughbred racehorse case described above, were presented during the committee meeting. These cases highlighted the ongoing challenges of illegal movement of horses from EP/EIA endemic countries, illegal interstate movement of unsanctioned racehorses and of quarantined horses for the purposes of continued racing, suspected illegal movement of blood products from other countries, and foreign veterinarians practicing in the U.S. without a license. Other challenges mentioned included: the need for diligent microchipping of EP/EIA infected and exposed horses in all states and the need for a searchable database to trace these microchip numbers; the lack of knowledge and interaction with unsanctioned racing venues in most states and the safety concerns inherent in those interactions; the apparent absence of involvement of sanctioned racing authorities in addressing EP/EIA positive horses and unsanctioned racing; and the ongoing concern about the potential for EP/EIA-positive horses to move into other equine industry sectors at the conclusion of their racing career.

USDA Cattle Fever Ticks 2019

Hallie Hasel, D.V.M.; USDA-APHIS-Veterinary Services

The Cattle Fever Tick Eradication Program encompasses an area of land along the Texas/Mexico border from Del Rio to Brownsville, approximately 500 miles. This strip of land was established in 1938 as the Permanent Quarantine Zone (PQZ), a border to keep the cattle fever tick from moving north following its eradication from most of the southeast US.

In FY19, the number of infested premises increased slightly, primarily in Zapata and Webb Counties within the PQZ, and due to new infestations in Jim Hogg and Jim Wells Counties north of the PQZ. The CFTEP now has 3,121 premises under quarantine, with 185 as infested premises. Fever ticks have progressed into the northern portion of Webb County and into previously fever tick free areas of Webb and Zapata Counties.

Changing demographics along the southern border, in conjunction with continued fever tick pressure from Mexico, have contributed significantly to the increase in infested premises. Mexico does not have a fever tick eradication program, and both infested livestock and wildlife continue to move across the border.

CFTEP has limited available treatments for fever ticks. Livestock treatments include CoRal spray/dip, Dectomax Injectable, and Ivermectin medicated molasses tubs. Wildlife treatment is limited to Ivermectin treated corn for whitetail deer; no other forms of treatment are available for exotic wildlife, including nilgai, axis, red deer, and other exotics now present along the southern border.

The BM86 fever tick vaccine was introduced in September 2016 and continues to be used in the PQZ. Limited herds have been injected outside of the PQZ following an epidemiological risk assessment. CFTEP has administered over 28,000 doses since the vaccine was introduced.

Fever tick research is in high demand. Alternative treatment methods and treatments with longer duration of kill in livestock are currently under research with ARS and academic collaborators. Wildlife treatment methods, including exotics, and treatment for pastures/premises/cleaning/disinfection are also required for fever tick eradication to continue.

Fever tick DNA continues to provide clues as to ongoing outbreaks, both within and outside of the PQZ. Recent outbreaks are primarily due to new DNA, thus from ticks recently introduced into Texas, most likely from south of the Rio Grande River.

The Rio Bravo Buffer Zone working group was formed in 2019, and will continue planning towards establishment of the zone on the south side of the Rio Grande River. The mission and goals are currently being established.

Rio Bravo Buffer Zone

Andy Schwartz, D.V.M.; Texas Animal Health Commission

Cattle Fever Ticks (CFT), *Rhipicephalus annulatus* and *R. microplus*, vectors of Bovine Babesiosis, are endemic to Mexico. Repeated and expansive outbreaks of CFT occur in Texas due to ticks carried by certain wildlife species and stray livestock moving northward across the US border. USDA APHIS VS (VS) and Texas Animal Health Commission (TAHC) maintain a permanent quarantine zone along a 500 mile stretch of the Rio Grande, and regularly monitor livestock and wildlife in this buffer zone to detect incursions of CFT. In order to reduce the incidence of these incursions, a bi-national cooperative effort is underway to establish a buffer zone south of the Rio Grande in Mexico, mirroring the buffer zone in Texas, and named the Rio Bravo Buffer Zone. A steering committee has been assembled with representatives of both federal governments and the states of Tamaulipas, Nuevo Leon, Coahuila, and Texas. CFT eradication measures will be cooperatively deployed in the Rio Bravo Buffer Zone with the goal of reducing and eventually eliminating the population of CFT. Expected outcomes are improvements

to the health and marketability of cattle in the zone, and reduction in the number of CFT outbreaks in Texas.

Effects of Drought and Media-Reported Violence on Cattle Fever Tick Incursions

Amy Delgado, D.V.M., M.S., Ph.D., Center for Epidemiology and Animal Health, USDA-APHIS-Veterinary Services

Ectoparasites, including cattle fever ticks, pose a risk to the global cattle population, both in reduced productivity and in livability. Cattle fever was once endemic in US cattle, but was eradicated through concerted and costly efforts. Reintroduction to US cattle could lead to substantial mortality and costs in terms of containment, eradication, and effects on producers and consumers. A permanent quarantine area provides constant surveillance for reincursions to minimize those risks.

Factors influencing the movement of hosts and ticks through the border region are varied and complex. In addition to climate-related factors, human directed ecosystem changes can lead to instability and changes in tick habitation and pest pressure. In addition, societal factors leading to farm abandonment may increase the movement of infected cattle across the border. The purpose of this study was to examine the effects of media-reported violence on incursions of cattle fever tick infested livestock.

Violent activity was collected using a media index for search terms related to border violence adjacent to the PCFTQ. An overall media index was calculated by averaging and re-indexing indices collected for the study region of various search terms related border violence, drug cartel violence, and Mexican drug cartel activities using Google Trends. To account for environmental factors that may lead to increase in stray cattle movements or changes in optimal tick habitation, weather data were collected from the National Oceanic and Atmospheric Administration's (NOAA) National Climatic Data Center. This includes hydrological and ambient data such as maximum temperatures and precipitation indices.

Over time, the number of infested cattle apprehended by tick riders has increased. Media-reported violence was shown to have a positive effect on the number of infested cattle apprehended, even when accounting for the effects of patrol resources, climate, and ecosystem. With continued land use changes, social unrest, and changing weather patterns, the efforts to control and eradicate CFT, both in the United States and globally, will be an ongoing concern.

2019 Vesicular Stomatitis Outbreak

Angela Pelzel-McCluskey, DVM, MS; USDA-APHIS-Veterinary Services

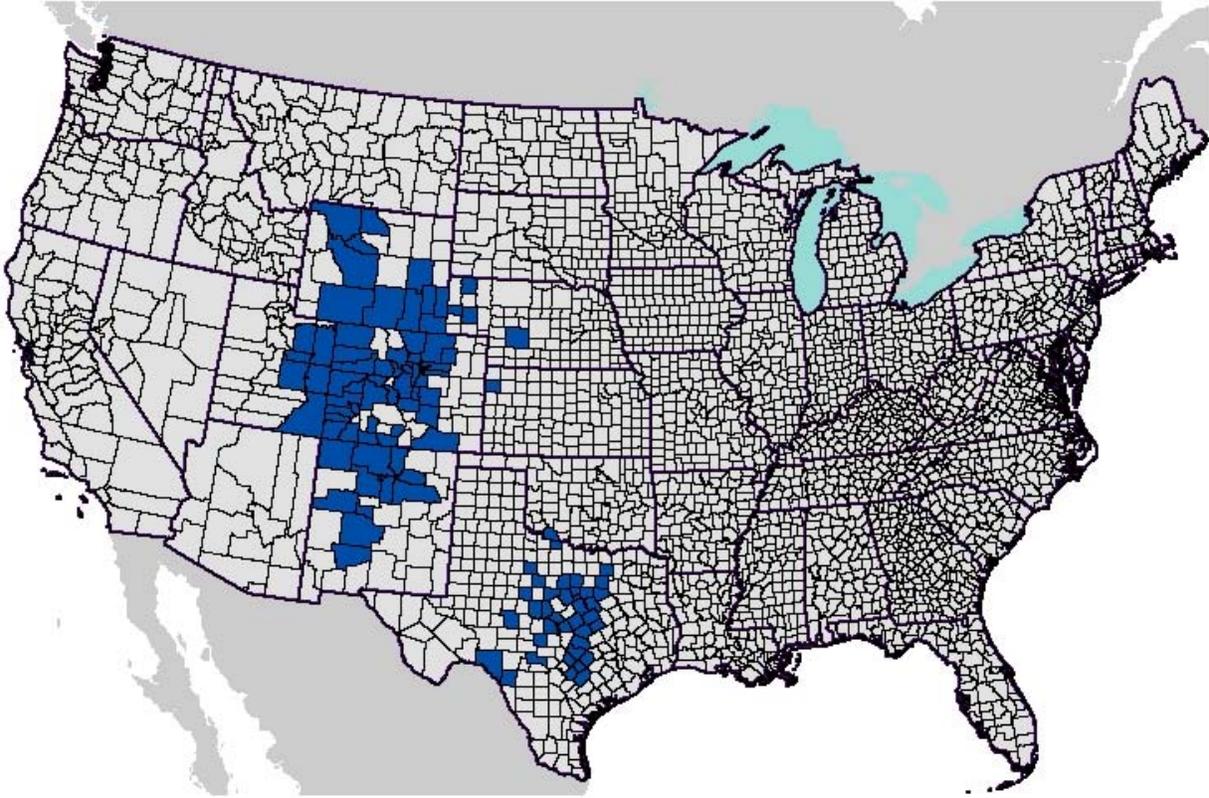
On June 21, 2019, the National Veterinary Services Laboratories in Ames, Iowa, confirmed a finding of vesicular stomatitis virus (VSV) infection (Indiana serotype) on an equine premises in Kinney County, Texas. This was the index case of VSV for the 2019 outbreak and for the state of Texas. As the outbreak progressed, 7 additional states became confirmed as VSV-affected: New Mexico on June 26, Colorado on July 3, Wyoming on July 24, Oklahoma on July 29, Nebraska on August 9, Utah on August 19, and Kansas on October 23, 2019. A total of 1,131 premises in these 8 states have been either suspected or confirmed as VSV-infected during the outbreak to date and placed under state quarantine. Quarantines remain for a period of 14 days from the onset of lesions in the last affected animal on the premises and vector mitigation strategies and enhanced biosecurity procedures are recommended on quarantined premises to reduce within-herd spread of the disease.

The breakdown of the number of quarantined premises and affected counties by state are shown in Table 1 below and the distribution of affected premises is shown in Figure 1.

Table 1. Total number of VSV-affected premises by state as of October 23, 2019.

State	# Counties Positive	# Confirmed Positive Premises	# Suspect Premises	Total # Premises Quarantined
Colorado	38	269	417	686
Kansas	1	1	0	1
Nebraska	4	15	10	25
New Mexico	12	47	29	76
Oklahoma	1	1	0	1
Texas	37	76	96	172
Utah	6	12	11	23
Wyoming	11	41	106	147
TOTAL:	110	462	669	1,131

Figure 1. Cumulative map of VSV-affected counties: June 21 – Oct 23, 2019



Of the 1,131 VSV-affected premises identified, 1,119 premises have had only equine species clinically affected, 11 premises have had only cattle clinically affected, and 1 premises has had both equine and cattle clinically affected. Since the start of the outbreak, a total of 1,058 premises have completed the

required quarantine period and been released, but there are 73 premises still quarantined for VSV at the time of this writing.

SCWDS Update: EHDV/BTV Surveillance and Arthropod Surveys

Mark G. Ruder, Southeastern Cooperative Wildlife Disease Study (SCWDS), College of Veterinary Medicine, University of Georgia

(Other authors) Stacey Vigil, Seth White, Alec Thompson, Natalie Stilwell, Brianna Williams, Rebecca Poulson, Michael Yabsley and David Stallknecht, SCWDS, College of Veterinary Medicine, University of Georgia, Athens, GA; James Mertins, USDA-APHIS-VS, National Veterinary Services Laboratories, Ames, IA

In collaboration with the USDA-APHIS-VS and SCWDS member wildlife agencies, SCWDS conducts surveys for exotic arthropods across the United States and Caribbean region. Past and current programs include surveys for the tropical bont tick on wildlife; surveys for cattle fever ticks on wildlife in Texas; and surveys for *Haemaphysalis longicornis* and other exotic ticks on wildlife. Surveys for cattle fever ticks (*Rhipicephalus annulatus* and *R. microplus*) are ongoing in Texas, in collaboration with USDA-APHIS-VS and the Texas Animal Health Commission. SCWDS personnel examined 223 hunter-harvested animals during December 2018 and January 2019 from 20 counties in south Texas. No cattle fever ticks were found. Additional surveys are scheduled for December 2019, and January 2020.

Since the fall/winter of 2017, SCWDS has worked with numerous state, federal and private groups to conduct surveys of wildlife for *H. longicornis*. Methods have included 1) live animal trapping and environmental sampling in localized areas where *H. longicornis* has been documented, 2) passive regional surveillance of white-tailed deer and other wildlife, and 3) tick collections from wildlife presented to wildlife rehabilitation facilities in areas where *H. longicornis* has been documented. As of October 25, 2019, we have examined ticks from ~1600 individuals representing 53 species from 21 states resulting in numerous new state, county, and host records. Although the situation is dynamic, to date, we have detected *H. longicornis* in seven states (New Jersey, Maryland, West Virginia, Virginia, North Carolina, Kentucky, and Pennsylvania) on white-tailed deer, raccoons, Virginia opossum, elk, woodchuck, red fox, gray fox, coyote, eastern cottontail, and red-tailed hawk.

Annually, SCWDS processes tissue samples from throughout the United States from wild ruminants with suspected orbiviral hemorrhagic disease. Molecular detection (e.g., conventional and quantitative reverse transcription PCR). For samples that test positive by RT-PCR, virus isolation is attempted and isolates are identified to serotype. Samples with no virus isolate are not further typed. Findings from the 2018 and 2019 transmission seasons are reported here. During 2018, 102 viruses were detected from 212 tissue samples, representing 6 species of wild ruminant (183 white-tailed deer, 16 mule deer, 10 elk, 1 pronghorn, 1 bighorn sheep, and 1 moose) from 23 states. Isolations of epizootic hemorrhagic disease virus (EHDV)-2 (58), EHDV-6 (1), bluetongue virus (BTV)-1 (1), BTV-18 (1), and BTV-24 (2) were made from white-tailed deer or mule deer (see Table). An additional 25 untyped BTVs were detected in white-tailed deer, mule deer, or elk (FL, GA, ID, MD, MO, MS, NC, NE, PA, and SC), and 14 untyped EHDVs were detected in white-tailed deer, mule deer, or elk (FL, MO, MS, MT, NC, NE, PA, SC, TN, and WV). As of October 24, 2019, 196 viruses have been detected from 316 tissue samples, representing 25 states and 5 species (293 white-tailed deer, 9 mule deer, 8 elk, 4 pronghorn, and 2 cattle). To date, isolations of EHDV-1 (1), EHDV-2 (126), and bluetongue virus (BTV)-2 (1) were made from white-tailed deer, pronghorn, or cattle (see Table). An additional 16 untyped BTVs have been detected in white-tailed deer (AR, FL, GA, NC, NE, PA, and WV) and 52 untyped EHDVs have been detected in white-tailed deer, mule deer, or elk (AR, FL, GA, ID, IN, KS, KY, MO, NC, WI, and WV).

2018 SCWDS EHDV & BTV Diagnostics		
Virus Serotypes Detected		
STATE	SPECIES	VIRUS
Florida	white-tailed deer	EHDV-2
		BTV-18
		BTV-24
Georgia	white-tailed deer	EHDV-2
Idaho	white-tailed deer	EHDV-2
Kansas	white-tailed deer	EHDV-2
Kentucky	white-tailed deer	EHDV-2
		EHDV-6
Louisiana	white-tailed deer	EHDV-2
Missouri	white-tailed deer	EHDV-2
Mississippi	white-tailed deer	EHDV-2
Montana	mule deer	EHDV-2
North Carolina	white-tailed deer	EHDV-2
North Dakota	white-tailed deer	EHDV-2
	mule deer	EHDV-2
Nebraska	mule deer	EHDV-2
Pennsylvania	white-tailed deer	EHDV-2
West Virginia	white-tailed deer	EHDV-2
		BTV-1

2019 SCWDS EHDV & BTV Diagnostics		
Virus Serotypes Detected		
<i>as of October 24, 2019</i>		
STATE	SPECIES	VIRUS
Alabama	white-tailed deer	EHDV-2
Arkansas	white-tailed deer	EHDV-2
Georgia	white-tailed deer	EHDV-1
		BTV-2
Idaho	white-tailed deer	EHDV-2
	pronghorn	EHDV-2
Indiana	white-tailed deer	EHDV-2
Kansas	white-tailed deer	EHDV-2
Kentucky	white-tailed deer	EHDV-2
Louisiana	white-tailed deer	EHDV-2
Missouri	white-tailed deer	EHDV-2
North Carolina	white-tailed deer	EHDV-2
North Dakota	white-tailed deer	EHDV-2
Virginia	white-tailed deer	EHDV-2
Wisconsin	white-tailed deer	EHDV-2
West Virginia	white-tailed deer	EHDV-2
	cattle	EHDV-2

USDA-ARS Knippling-Bushland U.S. Livestock Insects Research Laboratory Update

Dr. Adalberto A. Pérez de León, et. al., USDA - Agricultural Research Service

This presentation highlights research outcomes of research by scientists with the Livestock Arthropod Pest Research Unit of the USDA-ARS Knippling-Bushland U.S. Livestock Insects Research Laboratory in collaboration with national and international collaborators. This presentation reported the results of research only. Mention of a proprietary product does not constitute an endorsement or a recommendation by the USDA for its use.

As part of a national emergency response, ARS scientists at Kerrville, Texas, collaborated with researchers at Texas A&M University System AgriLife Extension and in New Zealand to sequence the genome of the longhorned tick. The completed genome opens new avenues of research for longhorned tick control, including vaccine development and detection of pesticide resistance-associated genes. In this regard, pyrethroids comprise a class of acaricides altering the tick nervous system that are found in several products commercialized in the U.S. Mutations in the sodium channel targeted by pyrethroids are known to result in insensitivity to treatment. Thus, molecular experiments were done by ARS scientists in Kerrville, Texas and cooperators at Rutgers University to characterize the longhorned tick sodium channel to inform decisions on the use of treatments with products containing pyrethroids. No mutations previously associated to pyrethroid resistance were detected in the tested longhorned tick samples from New Jersey. This is the first characterization of a gene in the longhorned tick associated with acaricide resistance.

Cattle fever ticks (CFT) remain a real and present threat to U.S. cattle production because they are established in Mexico. Additionally, livestock-wildlife interactions in the Permanent Quarantine Zone (PQZ) established by the Cattle Fever Tick Eradication Program (CFTEP) in south Texas on the border with Mexico endanger its operations. Interactions between cattle, white tailed deer (WTD), and nilgai antelope were simulated by ARS scientists in Kerrville, Texas and collaborators at Texas A&M University to assess the risk for CFT infestations in the PQZ and beyond. This research documented the use of enhanced biosurveillance simulation tools to mitigate risk and enhance current control strategies for use in the operations of area-wide tick management programs like the CFTEP through integrated tactics for CFT suppression.

Alternative treatments with novel modes of actions are needed for the effective treatment of tick infestations in livestock because the frequency of tick populations that are resistant to conventional acaricides keeps growing worldwide. Tekko® Pro is an insect growth regulator concentrate product containing 1.3% novaluron and 1.3% pyriproxyfen as the active ingredient. This product is registered with the Environmental Protection Agency for use indoors and outdoors on furniture, carpets, and kennels. Studies by ARS scientists in Kerrville, Texas showed that Lone star tick larvae did not develop to the next stage when infesting treated cattle. This effect lasted for ~30 days. The development of cattle fever tick larvae was inhibited when the ticks were placed on cattle that had been treated on the previous day. This product could be developed to treat cattle against tick infestations.

White-tailed deer infestations threaten the viability of CFT eradication efforts. Hides from hunted white-tailed deer are systematically inspected and treated with substances to kill ticks, also known as acaricides, before they leave areas in south Texas known to be at risk of CFT infestation. However, safer acaricides are needed to treat deer hides infested with CFT, specifically the southern cattle fever tick (SCFT). The invasive SCFT is considered the most economically important external parasite of livestock worldwide. Laboratory experiments simulating infested deer hides by ARS scientists in Kerrville, Texas, showed that a commercial product containing a mixture of essential oils killed all the immature SCFT, reducing female fertility by 94%, and killing 98% of the fully engorged females.

Ticks are of significant One Health importance because most tick-borne diseases are zoonotic. Enhanced pathogen detection is needed to improve the diagnosis of tick-borne diseases impacting animal and public health. ARS scientists in Kerrville, Texas and collaborators at Texas A&M University developed the

TickPath Layerplex, which is an innovative molecular assay to detect several tick-borne pathogens. TickPath Layerplex detects several groups of tick-borne pathogens in a sample distinguishing the type of tick-borne pathogen in the sample. Test results guide the decision for rapid and appropriate treatment. TickPath Layerplex testing is offered by the Texas Veterinary Medical Diagnostic Laboratory. It can be used during or after treatment of some tick-borne diseases as serologic titers can be persistent despite treatment of infection.

Surveillance for acaricide resistance is critical to design strategies that mitigate risks for its development and spread. A rapid molecular test was developed by ARS scientists in Kerrville, Texas to detect different mutations simultaneously in the SCFT genome associated with resistance to pyrethroids, which is a class of pesticidal compounds commonly used for their acaricidal properties. Results from tests using this assay were combined with data sets obtained previously and analyzed to evaluate the temporal epidemiology of resistance to the pyrethroid permethrin among SCFT causing outbreaks in the US.

ARS developed an ultra-quiet nematode sprayer used previously to treat nilgai. The nematode dispensed by the sprayer kills the ticks selectively. This technology is being adapted to treat white-tailed deer. ARS scientists in Kerrville, Texas and cooperators at Texas A&M University-Kingsville determined that deer behave normally at corn feeders with the attached sprayer system. This new technology could be used during the hunting season to treat CFT infestations in deer.

ARS scientists in Kerrville, Texas analyzed infestation and environmental data, and samples collected from hunted or culled nilgai to enhance our understanding of how this exotic wildlife species complicates efforts by the CFTEP. The correlation noted between infestation and habitat with thorn scrub suggested that the vegetative canopy promotes fever tick survival, which likely impacts infestation levels of nilgai in southeastern Texas. One nilgai was seropositive for *Babesia bovis* and *B. bigemina*, the microbes causing bovine babesiosis, by complement fixation. However, it remains to be determined if productive infection with the agents of bovine babesiosis occurs in nilgai. Eleven of the nilgai tested were seropositive to antibodies against the bacterium causing bovine anaplasmosis.

Rhipicephalus annulatus is the other cattle fever tick species established in Mexico that threatens U.S. animal agriculture because of its vector ability to transmit the microbes causing bovine babesiosis. Collaborative efforts between ARS scientists in Kerrville, Texas with the Veterinary Pest Genomics Center and Texas A&M University unraveled the genome of *R. annulatus*. This offers the opportunity to translate genomic information for the innovation of technologies the CFTEP can use to keep the U.S. free of CFT in a sustainable manner. A way to do this is through comparative genomics using previously discovered sequences from the southern CFT for applied research to develop anti-fever tick vaccines.

Some arthropod disease vectors are known to modulate the immune response by susceptible host, which can promote the transmission of vector-borne pathogens. Based on previous results, it was hypothesized that the presence of an active acetylcholinesterase (AChE) in the saliva of the SCFT might be involved in the immunoregulation of the host response to tissue damage during blood feeding. ARS scientists in Kerrville, Texas obtained further evidence consistent with this hypothetical paradigm by demonstrating that multiple arthropods and biological vectors of disease including several tick species, mosquitoes, and sand flies contain AChE in their saliva. Non-biological vectors biting arthropods such as horn flies and stable flies that also feed on blood lacked salivary AChE. Science-based knowledge from confirmatory evidence that salivary AChE plays a role at the tick-host interface during blood feeding could be used to innovate tick control technologies that also block the transmission of SCFT-borne pathogens.

The use of genome editing technology CRISPR-Cas9 was validated in the New World screwworm through a research partnership between scientists with the ARS scientists at Kerrville, Texas, the University of North Carolina, and the University of Campinas in Brazil. This research technology is a key tool in developing gene drive strains and can be used to understand gene function. The technique was verified by knocking out genes for body color, olfaction, and sex determination. This method is being adapted for transgenic screwworm research.

Horn fly populations resistant to commercial products used to treat infestations in cattle is a growing problem. Safer insecticides with new modes of action are needed. ARS scientists at Kerrville, Texas determined that laboratory grade limonene, a botanical with pesticidal properties, and a commercial formulation of limonene reduced horn fly egg viability whereas contact exposure to adults caused up to 100% knockdown. Moreover, laboratory grade limonene caused adult contact mortality. Limonene was attractive to horn flies at low concentrations of less than 0.1%. This property could be used to trap horn flies away from cattle.

Committee Business:

The Committee conducted a business meeting as follows:

- 1) Call to order - 9:53AM
- 2) Review of Committee Mission
- 3) Establish quorum – 13 members – quorum established
- 4) New Business
 - a. Resolution – EIA and EP Control Strategies
 - i. Motion to adopt
 1. Moved – Katy Flynn
 2. Seconded – Andy Schwartz
 - ii. Discussion - none
 - iii. Vote – carried unanimously
- 5) Old Business - none

OTHER NOTES: