MAY 2017

DIAGNOSTIC INSIGHTS

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Tick-borne Disease Highlight: Tularemia

By Drs. Kelli Almes and Megan Niederwerder

Warmer weather has arrived and so have the infectious diseases and vectors that go along with it. In the Midwest, especially Kansas, that means ticks and tick-borne diseases in both humans and our pets. Two of these diseases that all too often have fatal consequences for our feline patients are *Francisella tularensis*, the causative bacterium of tularemia or rabbit fever, and *Cytauxzoon felis*, the causative parasite of cytauxzoonosis or bobcat fever. Tularemia can also have serious, and occasionally fatal, consequences in humans. F. tularensis is highly infectious with only a few organisms needed for transmission, which can occur via aerosolization, ingestion, or through the bite of an infected tick. A correct and rapid diagnosis facilitates proper treatment with appropriate antibiotic therapy.

The tick vectors that transmit tularemia are all present in Kansas and are capable of transmitting other zoonotic agents¹ (Table 1). *F. tularensis* is transmitted to cats by the infected tick feeding on the cat or the cat ingesting the infected tick, typically during grooming. Cats can also become infected from feeding on an infected rabbit carcass, being bitten or scratched by another infected animal, or through mechanical transmission from other blood feeding insects. The incubation period is approximately 3-5 days. Common clinical signs and gross lesions are listed in Figure 1 and images are shown in Figures 2 and 3. Many of these same clinical signs

can be seen with *Cytauxzoonosis*; these two pathogens must be differentiated to initiate proper treatment and safety precautions.

Antemortem testing can be challenging, but submission of a lymph node aspirate from an enlarged node bacterial for culture at KSVDL yield diagnosis. The aspirate should collected aseptically and placed in preservativefree tube. Proper personal protective equipment, including gloves

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and eye protection, is recommended while performing this procedure. Lymph node biopsy could also yield a definitive diagnosis but requires anesthesia and significantly more stress on the patient. If animals have died from a suspected infection with *F. tularensis*, we recommend submitting the entire animal for necropsy,

	Tick Species and Common Name		
	Amblyomma americanum Lone Star tick	Dermacentor andersoni Mountain Wood tick	Dermacentor variabilis American Dog tick
Francisella tularensis	X	X	X
Heartland Virus	Suspected		
Rocky Mountain Spotted Fever		X	X
Ehrlichiosis	×		
Colorado tick fever		×	

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Tularemia (continued)

Figure 1: Common clinical signs and gross lesions of tularemia

Common Clinical Common Gross Signs of Tularemia **Lesions of Tularemia** Fever Multifocal splenic necrosis (Fig. 1) Lethargy Necrotizing lymphadenopathy (Fig. 2) Lymphadenopathy Pinpoint necrosis in liver and/or lungs Icterus Oral/lingual ulcerations Oral ulcerations

ancillary testing, and proper disposal. This provides the best chance of a definitive diagnosis and reduces the exposure risks for the attending veterinarian and their staff. We do not recommend performing an in-clinic necropsy for sample collection.

Francisella tularensis is a select agent due to its potential to pose a severe threat to both human and animal



Figure 2: Multifocal to coalescing splenic necrosis

health. Select agents are maintained on a federal list and specific regulations have been set for working with and disposing of these organisms. All cases of tularemia submitted to

KSVDL are confirmed by the CDC and potentially contaminated tissues are rendered sterile by autoclave on site. If a cat is suspected of having tularemia, the necropsy Figure 3: Cut section of enlarged and will be performed in a biosafety cabinet to



necrotic lymph nodes.

prevent aerosolization of the bacteria and minimize any potential exposure.

If an animal is diagnosed with tularemia, the owner or any other human with potential exposure should seek advice and possible treatment with their physician. The oral lesions present in many of these cats readily transmit the bacteria and thus, a history of saliva contact or biting should be thoroughly investigated.

References:

¹Tickborne Diseases of the United States (https://www.cdc. gov/ticks/diseases/index.html)

KSVDL Welcomes Dr. Douglas Marthaler to the Diagnostic Team

Douglas Marthaler recently joined the Veterinary Diagnostic Laboratory (VDL) at Kansas State University. Previously, he was the faculty advisor for the Molecular Research and Development section in the VDL at the University of Minnesota and focused on PCR development and implementation of new technologies into the VDL.

His primary research interest is to understand the emergence, evolution and epidemiology of enteric and respiratory viruses in animals. Some of his previous research involved co-infections and molecular diagnostics of rotavirus A, B, C and H in swine and the global variations of porcine epidemic diarrhea virus (PEDV) and porcine deltacoronavirus (PDCoV) to understand their emergence in the United States. His future research interest involves understanding

the pig immune response to the different rotaviruses and develop a rotavirus vaccine for passive immunity in sows to protect their piglets from mortality.



Dr. Douglas Marthaler

While his previous research focused on swine pathogens, he is excited to get back to his roots and

work on bovine pathogens since he grew up on a dairy farm in central Minnesota. He's already comparing field strains of bovine rotaviruses to commercial vaccines to understand and prevent calf morbidity and mortality. With over 11 years of veterinary diagnostic experience, he's excited to share his skills and knowledge with colleagues at K-State to continue to build the diagnostic capabilities at VDL to serve the needs of our clients.

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Tick-Borne Disease Testing

Tick-Borne Disease Serology Screen

KSVDL is now offering a serology panel that screens for Canine ehrlichiosis, Lyme disease, and Rocky Mountain spotted fever.

Annually, thousands of dogs and humans are infected with tick-borne diseases and that rate is climbing. The increasing incidence of tick-transmitted diseases of dogs and people has been associated with the ever increasing range of the various tick species, encroachment of wildlife species into the traditional "urban" environments, and an increase is pet travel.

The wide variation in the disease onset, the variable clinical signs exhibited, and the response to therapy can make a definitive diagnosis of the specific tick-borne disease difficult.

- Lyme disease is transmitted by the deer tick, causing stiffness, lameness, swollen joints, loss of appetite, fatigue, and possibly fever.
- Canine ehrlichiosis is transmitted by the brown dog tick. The various symptoms include fever, loss of appetite, depression, weight loss, runny eyes and nose, swollen limbs, and possibly bleeding.
- Rocky Mountain spotted fever is carried by the American dog tick, the wood tick, and the lone star tick. The symptoms include fever, stiffness, neurological problems, and possibly skin lesions.

A screening tick-borne disease screening panel can be very helpful it identifying the causative tick-transmitted agent.

Sample: Serum in a preservative-free sterile tube

Test Schedule: Thursday and Friday (test is set up at 7:30 a.m.)

Estimated Turnaround: 2-3 days

For more information please go to www.ksvdl.org and select "Tests & Fees."

Bovine Nitrate Sampling

Recently, KSVDL has received a large number of bovine samples that were unsuitable for nitrate testing.

In the past, the submission of the entire globe had been recommended. However, the longer the fluid stays within the eye the more likely discoloration will occur. This is enhanced by the rigors the sample undergoes while being shipped to the laboratory.

KSDVL recommends that samples for nitrate testing be collected in the field, and that the entire globe not be submitted.

Aqueous humor (ocular fluid) is an excellent sample to collect in suspected cases of nitrate toxicosis. Nitrate levels in ocular fluid are relatively stable postmortem.

Samples of ocular fluid are easily collected at necropsy with a sterile needle and syringe. Begin by ensuring the surface of the eye is free from mud and other debris. A 16-18 gauge 1" needle and 3-12 cc syringe is used, depending of the size of the animal. Enter the anterior chamber through the cornea. Gently aspirate 1-2cc of ocular fluid.

After collection, the fluid should be placed in a sterile non-additive tube. The sample should be shipped on an ice pack. Following these procedures should ensure a suitable sample is obtained for nitrate testing.

Bovine Post-mortem Magnesium Testing

Vitreous humor is an excellent sample to submit for post mortem suspect hypomagnesemia cases. The sample is collected as described above, only a deeper needle penetration is required for vitreous sampling. It is important to collect vitreous and not aqueous humor as the magnesium in the vitreous humor is more stable than that present in the aqueous humor.

Samples remain stable up to 48 hours after death.

Vitreous humor magnesium level should not be used as the only diagnostic test for hypomagnesemia. Clinical signs, diet, gestation period, etc. in addition to the magnesium level should be taken into consideration.

For more information concerning testing please contact KSVDL Client Care at 866-512-5650 or clientcare@vet.k-state. edu

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KSVDL Helps Expand Capabilities of Johne's Disease Risk Assessor Mobile App By Janelle Marney

The art of writing with pen and paper is becoming a thing of the past. New technological advancements make it easier to have everything online and in the palm of your hands. With the new Johne's Disease Risk Assessor App, producers can file their Johne's Herd Risk Assessment and Management Plan (RAMP) at the touch of a button.

The University of Wisconsin School of Veterinary Medicine created the application, while individuals from the Kansas State Veterinary Diagnostics Laboratory (KSVDL) provided consultation about expanding the app to include cow-calf operations. Dr. Gregg Hanzlicek, KSVDL, was one who helped in this process.

"The app was already an awesome tool designed for dairy veterinarians," Hanzlicek said, "We thought it would also be a great tool for cow-calf veterinarians so we asked Dr. Collins and Tom Bennett if they could redesign it for this purpose."

So far, there are approximately 700 users for the Johne's Disease Risk Assessor App. The app helps convert the RAMP into an electronic version. It is currently only available for Apple devices; however, they are currently sorting details for it to be available on Android systems.

"The goal is to reduce or eliminate risks for the introduction and/or spread of Johne's disease. Implementation of management practices aimed at Johne's disease control will increase herd biosecurity and reduce the risk for transmission of other fecal-oral transmitted pathogens," said Thomas Bennet, senior

information processing consultant at the University of Wisconsin School of Veterinary Medicine.

After downloading the app, users will see seven tabs:

- History
- Calf
- Heifer
- · Adult/Replacement
- Summary
- Recommendations
- PDF Viewer

Users should start completing the process by starting with "history" and moving along the tabs to the right. Once users complete each tab of information, they are then able to save the document as a pdf version. If users wish, they may also print the document.

With the help of the app, gathering all the information is a simpler process. Veterinarians can easily access the information and have a basic understanding of the work that has been previously done with implementations towards Johne's disease. Users will be able to enter the veterinarian's information (including name, clinic name and clinic address) into the app as well.

With the new Johne's Risk Assessor App, users can easily have their RAMP available to them at any moment and at the palm of their hands.

"It's a really cool tool and it's time to start getting the word out and doing something about the disease," Hanzlicek said.

New Fees Effective July 1st, 2017

KSVDL will have an updated fee schedule effective July 1, 2017. Most prices will increase slightly, but some will remain the same. This increased percentage is small in order to help keep our prices fair and comparable to other state diagnostic laboratories. As always, we do not charge an accession fee nor do we have in-state/out of state price differentials. In order to help our clients prepare, on June 1, 2017 the updated charges will appear alongside our current online test and fee schedule available at www.ksvdl.org.

Thank you for choosing KSVDL for your diagnostic testing needs.

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Erythema Multiforme: What do we know?

By Dr. Chanran Ganta

What is Erythema Multiforme?

Erythema Multiforme (EM) is a cutaneous reaction pattern of multifactorial etiology that is seen in dogs, cats, horses, cows and ferrets. The pathophysiology of EM is thought to be a host-specific T-cell mediated hypersensitivity. The cellular immune response is directed against various keratinocyte-associated antigens, including those associated with drugs, infections (viral, fungal, bacterial), neoplasia, various chemicals, foods and connective tissue diseases. CD8+ T lymphocytes bind to antigenically altered keratinocytes and trigger apoptosis of keratinocytes/squamous epithelial cells. Based on the extent of involvement throughout the body, it is classified as erythema minor, erythema major, Stevens-Johnson syndrome or toxic epidermal necrolysis.

What is the trigger?

The highest frequency of cases reported in dogs, cats and horses with EM are drug-induced. The drugs that are commonly associated with EM include trimethoprim potentiated sulfonamides, penicillins and cephalosporins. Other causes of EM that are less frequently reported include dyes and preservatives in food, vaccination with Bordetella bronchiseptica, parvoviral infection, and insecticidal dips.

What are the clinical signs?

There will be an acute onset of erythematous annular macules, elevated circular plaques and papules that can be

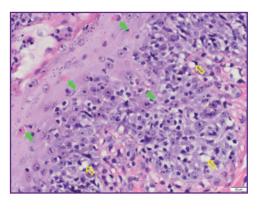
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partially symmetric. In some cases, these lesions appear as classic targetoid with concentric circular zones of erythema, blanching and exfoliation. These lesions most commonly occur on the trunk and especially involve the glabrous skin of the groin and axilla. Other common sites of involvement include mucocutaneous junctions, oral mucosa, pinna and pawpads (Figures 1-2). German Shepherd dogs and Pembroke Welsh Corgis were reported to be at increased risk. There is no breed predilection in other species.

How do you diagnose?

The differential diagnoses for this condition includes urticarial allergic eruption, superficial spreading pyoderma, bacterial folliculitis, dermatophytosis, demodicosis and early lesions of bullous autoimmune diseases. A definitive

diagnosis can be reached by histopathology. It is important to acquire a good biopsy for a definitive diagnosis of this condition. The biopsy needs to be collected from areas



of erythema without crusting or ulceration with INTACT epidermis or mucosa. Characteristic histopathological lesions include apoptotic epidermal cells/keratinocytes at all levels of the epidermis, vacuolation of the basement membrane zone, few lymphocytes surrounding apoptotic keratinocytes and interface dermatitis (Microscopic Image 400X; Solid arrows: Apoptotic keratinocytes; Open arrow: Basal cell vacuolation).

Please contact Dr. Ganta or Dr. Bagladi if you have any questions regards sample collection and any skin diseases related questions.

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KSVDL Personnel Activities

- Dr. Kelli Almes presented Diagnostic submission hints and case examples at the Continuing Education for Veterinarians at the K-State Olathe Campus.
- Dr. Megan Niederwerder published a review article titled, Role of the microbiome in swine respiratory disease in Veterinary Microbiology, https://doi.org/10.1016/j.vetmic.2017.02.017
- Dr. Adi Wasserkrug Naor and Dr. Kelli Almes lead the KSVDL orientation program for the Class of 2018 KSU-CVM students.
- Dr. Chanran Ganta attended the National Veterinary Dermatology Forum in Orlando, Florida and presented a rare case under the session: What's Your Diagnosis titled NAVDF Cutaneous T cell Angioinvasive Lymphoma with Lung and Ocular Metastasis in a Cat.
- Dr. Gregg Hanzlicek and several USDA veterinarians, Trichomoniasis-certified 42 KSU-CVM senior veterinary students.

Field Investigations

- Dr. Gregg Hanzlicek with Dr. Matt Miesner (VHC livestock clinician) and two senior students, Taylor Nikkel and Taylor McCluskey, investigated multiple adult cow deaths on cow-calf operation.
- Dr. Gregg Hanzlicek and KSU-CVM students Mark Spare, Bobbi Ann Shanks, and Joanna Wilson assisted a local practitioner investigate respiratory issues in milk-fed calves housed in confinement.
- Dr. Gregg Hanzlicek and KSU-CVM student Jacob Nightingale assisted a local practitioner investigate an outbreak of neonatal scours in a cow-calf operation.
- Dr. Gregg Hanzlicek and KSU-CVM students Kotie Wootten, Bobbi Ann Shanks, Gina Callari, and Joanna Wilson assisted a private practitioner investigate hemorrhagic bowel syndrome in a Kansas dairy.

KSVDL on YouTube

We have posted new videos on the KSVDL YouTube® channel covering the following topics:

- · Diagnosing Giardia https://www.youtube.com/watch?v=eKg2_C-2XLE
- Fecal Centrifugation https://www.youtube.com/watch?v=mZunPcRr7C4
- Fecal Egg Count Reduction Test for Bovine and Equine Clients https://www.youtube.com/watch?v=SBYuKFbBvX8



Subscribe to the KSVDL YouTube® channel:

www.youtube.com/c/ Kansas State Veterinary Diagnostic Laboratory 1

KSVDL Laboratory Diagnosis of Canine Leptospirosis

The two tests routinely utilized for the diagnosis of canine leptospirosis are serology by the 6 serovar microscopic agglutination test (MAT) and polymerase chain reaction (PCR) testing (urine or whole blood).

Given that false negative test results may occur with both the 6 serovar MAT and the urine PCR, we recommend submitting both diagnostic tests as the combination offers the best combination of sensitivity and specificity. For more information, please call KSVDL Client Care at 866-512-5650 or visit page 6 www.ksvdl.org and choose Test and Fees.

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Developing and Delivering Accurate, Innovative Diagnostic Services

The mission of the Kansas State Veterinary Diagnostic Laboratory (KSVDL) is to develop and deliver accurate, innovative, and timely diagnostic and consultative services to the veterinary and animal health community while providing support for teaching, training and research programs.

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Continuing Education

www.vet.k-state.edu/education/continuing/

June 4-6, 2017

Annual Conference for Veterinarians Hilton Garden Inn. Manhattan, Kansas www.vet.k-state.edu/alumni/events/

July 21-25, 2017

AVMA Annual Convention Indianapolis Convention Center Indianapolis, Indiana

August 3-5, 2017

AVC Summer Conference Renaissance Denver Hotel Denver, Colorado

August 25-28, 2017

CVC Kansas City Kansas City, Missouri

For more information, call the Continuing Education Office at 785-532-4528.

Test Results and Schedules

Phone: 785.532.5650 Toll Free: 866.512.5650

Laboratory results available online, all the time!

Holiday Schedule:

Memorial Day: Closed: Monday, May 29th Independence Day: Closed: Tuesday, July 4th

To receive this newsletter by email, contact: ksvdloutreach@vet.k-state.edu.







