DiagnosticInsights

Kansas State Veterinary Diagnostic Laboratory
www.ksvdl.org

Accredited by the American Association of Veterinary Laboratory Diagnosticians — November 2013

Dr. Gary Anderson Receives Prestigious Award

Dr. Gary Anderson, the Director of the KSVDL, received the E.P. Pope Award at the American Association of Laboratory Diagnosticians (AAVLD) Convention in San Diego.

This award recognizes individuals who have made outstanding contributions to the AAVLD and who have promoted and advanced the field of veterinary diagnostics.

"It's a great honor to be recognized by my peers with such a distinguished award. This award is very little about me and very much about those around me," Dr. Anderson said.

Congratulations to Dr. Anderson on this outstanding achievement!

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Canine Distemper Diagnostics

Dr. Gordon Andrews

The recent canine distemper outbreak in the animal shelter in Emporia, Kan., has prompted us to briefly review laboratory methods for establishing a definitive diagnosis of distemper virus infection.

The clinical signs of distemper are familiar to most veterinarians and often lead to a tentative clinical diagnosis, but because the clinical signs can mimic other infectious diseases, this review will focus on those laboratory tests that establish a definitive diagnosis. The clinical signs mirror the sequential pathogenesis of viral infection and an understanding of the viral pathogenesis will help determine what samples are most appropriate to select for diagnostic testing.

Exposure to the virus is generally by aerosol contact with epithelium of the upper respiratory tract, with multiplication in tissue macrophages and spread by lymphatics to tonsil, retropharyngeal and bronchial lymph nodes. By 4-6 days post infection, virus replication occurs in lymphoid follicles of the spleen, lamina propria of the stomach and intestines, mesenteric lymph nodes, and Kupfer cells of the liver. At this time there is fever and leukopenia. 8-9 days post infection there is hematogenous lymphocyte associated viremia with spread of virus to epithelial and central nervous system tissues with shedding of virus from all body excretions even in dogs with subclinical infection.

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Infection of upper respiratory epithelium results in conjunctivitis and rhinitis with serous to mucopurulent oculonasal discharge. A conjunctival or nasal swab placed in viral transport medium is an ideal sample at this time to submit for PCR testing for distemper virus. If viral transport medium swabs are unavailable, a swab moistened with sterile saline and placed in a sealed sterile tube is a good substitute. Whole blood in EDTA is an excellent sample for ante mortem PCR testing in both the acute and chronic forms of the disease. Urine is also a good sample for PCR testing.

Infection of lung epithelium results in an interstitial pneumonia, and secondary bacterial infection is common. The resulting cough can mimic infectious tracheobronchitis (kennel cough). A nasal or tracheal swab can be submitted for the canine respiratory PCR panel, which includes canine distemper virus, Mycoplasma, Bordetella bronchiseptica, canine adenovirus type 2, canine herpesvirus type 1, Influenza A, canine parinfluenza-3, and two strains of canine coronavirus.

Infection of gastrointestinal epithelium results in vomiting and diarrhea. Clinical signs of central nervous system infection are dependent on the region of the CNS involved and can include hyperesthesia, cervical rigidity, seizures, cerebellar and vestibular signs, paraparesis or tetraparesis with sensory ataxia, and myoclonus.

Gross necropsy examination often reveals pneumonia, but this is nonspecific. Some dogs develop digital hyperkeratosis (hard pad), but this is not specific for distemper infection. Dogs that have recovered from distemper may have tooth enamel hypoplasia which is considered specific for prior distemper infection. Histopathologic findings can include lymphoid depletion, interstitial pneumonia, necrosis of ameloblastic epithelium, necrosis of epithelium in the gastrointestinal tract, swelling of transitional epithelium in the renal pelvis and urinary bladder, and necrosis and inflammation in the brain. Viral inclusion bodies can be found in epithelial cells of mucous membranes, stomach, intestines, transitional epithelium of urinary pelvis and urinary bladder, neurons and astrocytes. (Figure 1) When inclusions are found in association with histologic lesions in a dog with an appropriate clinical history, they can be considered diagnostic. The presence of inclusions alone however must be interpreted with caution, because distemper inclusion-like bodies have been described in the urinary bladder and brain of normal dogs. Distemper inclusions are not always found however and may only be found late in the disease. Immunohistochemical (IHC) staining of formalin-fixed tissues for distemper virus is a sensitive and specific method

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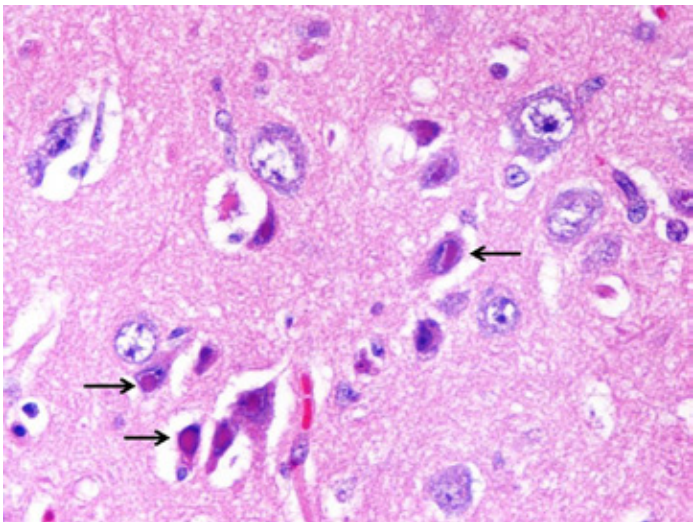

Figure 1. Brain from a dog with canine distemper showing intranuclear inclusions in astrocytes (arrows).

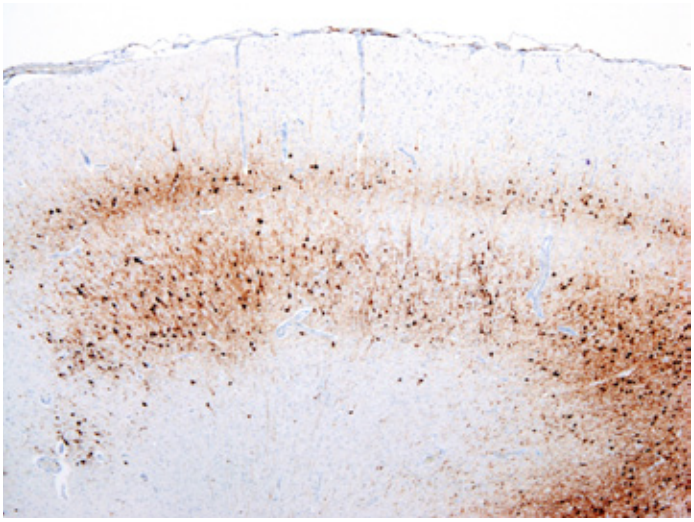

Figure 2. IHC stain of cerebral cortex from a dog with canine distemper. The brown staining is distemper virus antigen.

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of demonstrating viral antigen in tissue sections even before histologic lesions are evident. (Figure 2)

When submitting necropsy tissues for a diagnostic workup when distemper is suspected is important to include a complete set of formalin-fixed and fresh tissues even if gross lesions are not present and the patient has no clinical signs referable to a particular organ system. As an example, one dog I examined from the Emporia distemper outbreak had gross and microscopic evidence of chronic pneumonia, but PCR testing of fresh lung and IHC testing of formalin-fixed lung were both negative for distemper. There were no other microscopic lesions consistent with distemper infection in any other tissues except the brain, which did have encephalitis, and IHC staining was positive in the brain in spite of the fact that this dog was not reported to be showing any neurologic signs. Fresh necropsy tissues valuable for PCR testing include lymphoid tissues, lung, kidney, and brain.

Bovine Respiratory Bacterial Multiplex PCR Panel

A PCR to identify Mannheimia haemolytica, Pasteurella multocida, Histophilus somni, Bibersteinia trehalosi, and Mycoplasma bovis has been developed at the KSVDL. The PCR allows for a quicker turn-around time compared to bacterial culture.

This test compliments the Bovine Respiratory Viral PCR panel that is already available.

The test can be completed on either tissue or swabs.

If desiring both the Viral and Bacterial PCR, only one sample is required as a single sample can be utilized for both the viral and bacterial PCR panels.

When submitting for either the Viral PCR or Bacterial PCR or Both tests, please submit the swabs in viral transport media.

Cost: $40.00 per sample.

Contact Dr. Richard Oberst at 785.532.4411 or oberst@vet.ksu.edu for more information.

3rd Annual KSVDL Conference on Animal Diagnostics

The KSVDL will be holding our 3rd annual continuing program on Saturday, Feb. 8, 2014, from 9 a.m. to 3:30 p.m.

Program focus:

Bovine Vaccinology

Guest speaker:

Dr. Amelia Woolums

University of Georgia
College of Veterinary Medicine

Topics presented will include:

What's new about:

- bovine immunology
- vaccine timing
- modified live vs. killed product use
- how to read a titer
- autogenous vaccines

Stay tuned for more information about this timely continuing education program.

KSVDL Canine Rickettsial Diseases Testing

Ricketsial bacteria most often cause mild or unapparent disease in health dogs but can be virulent pathogens in immune-compromised patients. These pathogens are generally able to avoid complete clearance from the dog's body making diagnosis difficult. Screening for these infections prior to the dog's inclusion in a blood donation program is essential. Now there are PCR tests available at KSVDL to detect nine of these pathogens.

Canine Ehrlichiosis

Pathogens detected: Ehrlichia chaffeensis, Ehrlichia canis and Ehrlichia ewingii

Sample Requested: whole (unclotted) blood or spleen

Cost: $39.00

Estimated Turnaround Time: 2 days

Canine Anaplasma species specific PCR

Pathogens detected: Anaplasma phagocytophilum and Anaplasma platys

Sample Requested: whole (unclotted) blood or spleen

Cost: $39.00

Estimated Turnaround Time: 2 days

Canine Haemoplasma PCR

Pathogens detected: Mycoplasma heamocanis and Mycoplamsa heamoparvum (formerly, Haemobartonella)

Sample Requested: whole (unclotted) blood

Cost: $ 35.00

Estimated Turnaround Time: 2 days

Canine Blood Transfusion Panel

Pathogens detected: Ehrlichia chaffeensis, E. canis, E. ewingii, Anaplasma platys, A. phagocytophilum, Mycoplasma haemocanis and M, haemoparvum

Sample Requested: whole (unclotted) blood

Cost: $74.00

Estimated Turnaround Time: 2 days

Schistosomiasis

Dr. Patricia Payne, Dr. Jamie Henningson, Dr. Jen Lehr

Schistosomiasis, caused by Heterobilharzia americana, is a parasitic disease primarily affecting dogs in the southeast United States.

This parasite has been diagnosed in dogs at Kansas State Veterinary Diagnostic Laboratory and this year was found in two horses. Raccoons are the most common natural definitive hosts. Many other domestic and wild mammals, including humans can become infected via contact with contaminated water. Dogs are able to develop patent infections; it is unknown if horses develop patent infections, but it is thought to be unlikely. The divergent geographic prevalence of this disease is thought to be related to the importation of raccoons from Texas and Florida for sporting purposes. This was a common practice in many sporting clubs throughout the 1930's- 1950's. This practice has been curtailed by the passage of law in 1984 requiring special permits for the importation of raccoons. It appears that this parasite has found a suitable intermediate host to maintain a presence in an area far from its normal area of distribution.

H. americana is a trematode that requires a snail intermediate host. Mature eggs are shed into the water via the definitive hosts feces, where they are hatch and release free-swimming miracidium. Miracidium enter the snail by penetrating its body. Once inside the snail, they multiply exponentially and are released as cercariae. The cercariae penetrate the mammalian host's skin while the host is wading or swimming in the water. This life stage can cause severe dermatitis in humans. The cercariae are destroyed in the human's skin before they are able to get into the vasculature.

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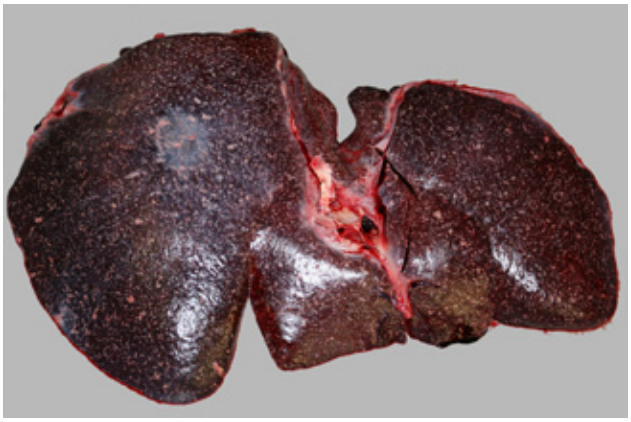
SCHISTOSOMIASIS | continued from Page 4

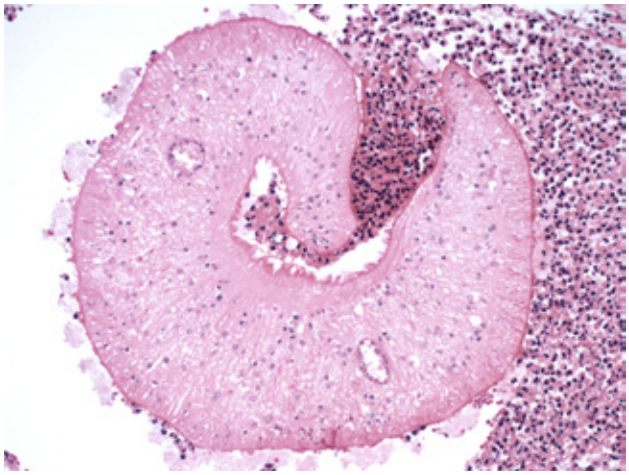
In dogs, and some other animals, this stage may also cause dermatitis. If the animal is able to support the next stage of infection, as are dogs, horses, llamas, the cercariae migrate into the vascular system. Male and female worms remain in copula and may be found in the mesenteric veins and liver vasculature of the host. The adult female sheds her eggs into these vessels and releases proteolytic enzymes into the surrounding tissues. The host's inflammatory response to the eggs results in the formation of granulomas.

The parasitized dog may become clinically ill with gastro-intestinal or hepatic disease or hypercalcemia of unknown origin. The chronic nature of the disease and the fact that H. americana are rarely found on fecal floatation make the diagnosis difficult. Fecal sedimentation or fecal PCR may be needed to diagnosis this problem in dogs. In contrast, this disease in horses is often an incidental finding at necropsy. The ultrasound appearance of the affected equine liver (starry sky liver) and the pathological appearance of the liver can be dramatic. Horses that developed granulomas in locations other than the liver, especially in the heart, were more likely to have clinical disease related to H. americana infection.

Many thanks to Wayne Corapi, DVM, PhD, DACVM, for his excellent pictures showing the diffuse hepatic granulomas in a horse with Starry Liver Disease.



1A Diffuse hepatic granulomas, are visible on gross examination



4B Microscopic view of a H. americana within one of the granulomatous lesions

Hanzlicek, A. S., Harkin, K. R., Dryden, M. W., Chun, R., Payne, P. A., Nietfeld, J. C., Debey, B. D., 2011 Canine

Schistomiasis in Kansas Five Cases (2000-2009). Journal of American Animal Hospital Association Nov/Dec 2011 47(6); e95-e102

Corapi, W. V., Snowden, K. F., Rodrigues, A., Porter, B. F., Buote, M. A., Birch, S. M., Jackson, N. D., Eden, K. B., Whitley, D. B., Mansell, J., Edwards, J., Hardy, J. and Chaffin, M. K. 2012 Natural Heterbilharzia Americana Infections in Horses in Texas. Veterinary Parasitology 49(3): 552-556

McKnowen, R. D., Veatch, J. K. and Fox, L. B. 1991 New Locality Record for Heterobilharzia Americana. Journal of Wildlife Diseases 27(1): 156-160

Acha, P. A. and Szyfres, B. Cercarial Dermatitis. Zoonoses and Communicable Diseases Common to Man and Animals, 3rd Edition Volume III. Parasitosis: 99-103

Carlson, K. L., Chaffin, M. K., Corapi, W. V., Snowden, K. F. and Schmitz, D. G. 2011 Starry Sky Hepatic Ultrasonographic Pattern in Horses. Veterinary Radiology and Ultrasound 52(5): 568-572

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KSVDL Outreach Activities

Presentations:

- Northeast, South central and North central District KVMA meetings

  Dr. Andrew's topic: Update on the shelter "mystery disease"

  Dr. Fortney's topic: 7 steps to a more successful skin biopsy result

  Dr. Hanzlicek's topic: Disease patterns observed in bovine respiratory panel results

- Northwest and Southwest District KVAM meetings

  Dr. Andrew's topic: Update on the shelter "mystery disease"

  Dr. Fortney's topic: 7 steps to a more successful skin biopsy result

  Dr. Hanzlicek's topic: A new genetic disorder observed in beef calves

- Farmer's State Bank Farmer Appreciation Banquet

  Dr. Hanzlicek's topic: Bovine Trichomoniasis in Kansas

- Animal Science Extension Agent Update: Eastern Kansas

  Dr. Hanzlicek's topic: Vaccinology

Field Investigations Completed in October:

- √ An increase in sudden death incidence occurring in a custom calf raising operation
- √ Respiratory disease outbreak in a group of single-source beef steers
- √ Reproductive issues in canines
- √ Hemorrhagic bowel syndrome epidemic in a Kansas dairy herd
- √ Johne's disease in a purebred beef operation: Risk assessment and management program

KSVDL Specializations

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COMPANION ANIMAL OUTREACH: DR. BILL FORTNEY
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FIELD INVESTIGATIONS: DR. GREGG HANZLICEK
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785-532-4457

Developing, 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.ksu.edu/CE/Conference.htm

Dec. 13 , 2013
Small Ruminant Conference and Wet Lab

Jan. 10, 2014
Bull Management Conference

Feb. 8, 2014:
Kansas State Veterinary Diagnostic Laboratory Conference: "Vaccinology"

Test Results and Schedules

Lab results may be accessed online 24 hours a day, 7 days a week!

To set up an account go to:
www.ksvdl.org

KSVDL hours:
Closed Nov. 28-29, Open Nov. 30
Closed at 3:00 p.m., Dec. 24
Closed Dec. 25

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KANSAS STATE UNIVERSITY