Wyoming State Veterinary Laboratory Newsletter – September 2007

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MESSAGE FROM THE DIRECTOR

As you have probably heard from the Wyoming grapevine, the WSVL has a new Director effective August 1, 2007. I have had a chance to visit with many of the practitioners who utilize our services, but as a group you are probably wondering about your new Director's background and qualifications. In short, I received the DVM and PhD (veterinary anatomic pathology) degrees from Texas A&M in 1976 and 1981, respectively. A short stint at the Collaborative Radiological Health Laboratory, Colorado State University was followed by over 21 years as Head of the Pathology Section at the Texas Veterinary Medical Diagnostic Laboratory - Amarillo. With this background, I'm sure you will appreciate that my major interests and commitments are to quality and timely diagnostic service. Along these lines, all of us at WSVL will constantly strive to update and provide the best affordable services to the practitioners, animal owners, and wildlife managers of Wyoming and elsewhere.

In the last issue of the Newsletter, the ongoing search for a fourth pathologist was mentioned with some optimism. This optimism was not misplaced. Dr. Shannon Swist will, as she put it, "join our happy band" in November. Dr. Swist received combined DVM and MS (pathobiology) degrees from Kansas State University in 2003. This was followed by a three year residency in anatomic pathology at Colorado State University. For the past year, Shannon has been honing her pathology skills at the University of Georgia, College of Veterinary Medicine. We are fortunate to have someone

of Dr. Swist's caliber join us and I hope you will welcome her. As I've told all of our pathology candidates, you are the best group of practitioners it has been my pleasure to work with. By the next Newsletter, we should be able to include a current photograph of Dr. Swist.

Our joy at the hiring of Dr. Swist is tempered by the recent loss of two faculty. Dr. Leslie Woods has resigned and will be returning to her home state of California. Although her stay was short, all have benefited from her broad knowledge and conscientious approach to diagnostic pathology. Her position will be filled, hopefully in a timely fashion. Our other loss was the retirement of Dr. William (Bill) Jolley after over twenty years of service to the University and WSVL.

Dr. Jolley will continue to be involved in teaching, at least through the spring of 2008. His experience and leadership in parasitology will be missed at WSVL. A search is in progress to fill Dr. Jolley's position. Please join us in wishing Bill the best during his retirement years.

On another note, the Departments of Veterinary Sciences and Molecular Biology have received permission to initiate an international search, led by Dr. Donal O'Toole, to fill a newly created endowed chair in prion biology. Although this position will not have a service role at WSVL, having someone of this stature will likely help in improving our knowledge and management of chronic wasting disease and other prion diseases affecting animals.

Dr. Don Montgomery

DIAGNOSTIC CASES OF INTEREST LABORATORY NOTES

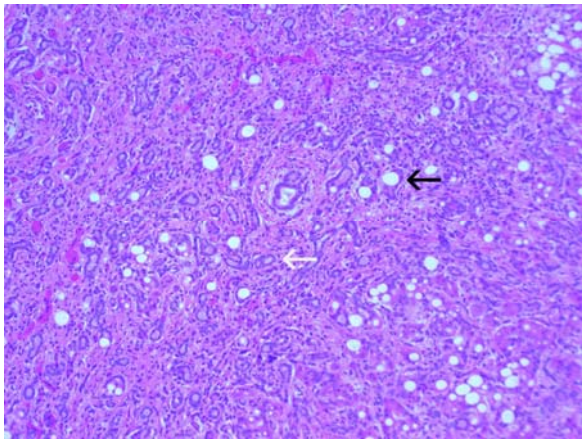
Fatal chronic hepatopathy ("hepatic lipodystrophy") in a Galloway calf

A 3-month old Galloway bull calf developed diarrhea and was treated for possible abomasal ulceration. The calf appeared to respond to treatment, but over the following two months its condition deteriorated. Dr. J. D. Fox performed a post-mortem examination. The calf had a firm yellow liver of normal size with edema around the bile duct. Samples of liver and kidney were submitted for histopathology.

Histologically, Dr. Ayers found remarkably extensive bile ductular hyperplasia, and disorganization and loss of hepatocellular architecture with pericellular fibrosis. Hepatocytes adjacent to (presumed) portal tracts contained large intracytoplasmic vacuoles suggestive of lipid. Renal glomeruli were large with thickened capillary loops suggestive of membranoproliferative glomerulonephritis. Terminal illness was due to hepatic failure.

Changes in this calf were consistent with a distinct hepatopathy that has been seen in Galloway cattle in England, Scotland and possibly Iceland. The first cases were seen

in 1965 and some appeared to be associated



Liver from a 3-month old Galloway calf with fibrosis and marked bile ducutular proliferation (white arrow). There is marked loss of hepatocytes. Where present, hepatocytes either contain a large clear space suggestive of lipid (black arrow) or fine vacuoles.

with the use of a particular bull. Two more recent cases have been reported from Canada. Assuming this is the same syndrome, this would be the first time the disease has been seen in the USA. Presenting signs are lethargy and - terminally - neurological signs consistent with hepatic encephalopathy. The oldest affected calf was 5 months old. Most calves die between 2 - 4 months. The disease is provisionally named "hepatic lipodystrophy." Nothing is known about its biochemical basis.

Galloway cattle are rare in Wyoming. If you have seen a Galloway calf with this presentation, please contact Donal O'Toole at the veterinary laboratory.

Stewart W, Allison CJ, Macleod NSM, Rushton B: 1982, Hepatic lipodystrophy. Vet Rec 100: 505.
Duff JP, Watson PJ, Scholes SFE: 1997, Chronic hepatopathy (hepatic lipodystrophy) of Galloway cattle. Vet Rec 141: 368
Macleod NSM, Allison CJ: 1999, Hepatic lipodystrophy of pedigree Galloway calves. Vet Rec 144: 143 – 145
Hazlett M: 2000, Hepatic lipodystrophy in 2 Galloway calves. Can Vet J 41:882

Dr. Jon Ayers/Dr. Donal O'Toole

Disseminated Histoplasmosis in a feral cat

A 2-year-old spayed female domestic shorthair cat was submitted for necropsy. This cat was one of five feral kittens rescued from a ranch cat. One of the five kittens died prior to rescue; another died after rescue. The cat's sibling developed anterior uveitis and was blind. This cat had recurrent anterior uveitis that responded to Baytril and steroids. Necropsy findings included: icterus, meningitis, anterior uveitis, pneumonia (multifoca), splenomegaly and cardiomyopathy. Histopathologic findings included multifocal granulomatous inflammation in many tissues (Fig 1).

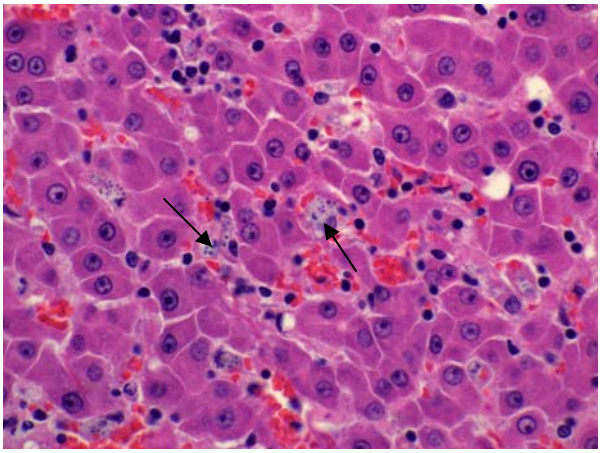


Figure 1. Clusters of *Histoplasma* organisms (arrows) are in the liver.

Most tissues had intrahistiocytic *Histoplasma spp*. Additionally, FIP virus was detected by the fluorescent antibody test.

This is the first report of disseminated Histoplasmosis in a feral cat in Wyoming. This disease has been reported in many parts

of the United States, primarily in regions bordering the Missouri, Ohio and Mississippi Rivers. The distribution may be more widespread than originally thought and that the currently recognized regional distribution just suggests that more thorough studies have been conducted in these areas. Most cases may be underreported because a benign inapparent infection can occur and may be more common.

Dr. Leslie Woods

Famfur poisoning in a dog

A 2 year-old Heeler cross was presented to the referring veterinarian with a history of sudden onset seizures, copious salivation, vomiting and diarrhea. Physical exam revealed fixed, dilated pupils, T106.8, hemoglobinuria. Histopath was non-specific with multi-focal acute hemorrhages in multiple organs, bronchiolitis and pulmonary edema and hemorrhage. Brain was not submitted, but blood cholinesterase was normal, discounting the possibility of organophosphate/carbamate poisoning. After discussing the case with the referring vet, however, and given the classic signs of cholinesterase inhibition, we decided to look for cholinesterase inhibitors in vomitus and stomach contents, anyway. The vomitus contained significant amounts of famfur (Warbex), the stomach contents were negative for all insecticides.

As do most laboratories, we use brain and blood cholinesterase activity as a surrogate for organophosphate and carbamate insecticides. The test is relatively inexpensive, quick and covers a lot of possibilities at once. If cholinesterase activity is depressed, we know that we're going to have to spend a day or two looking at all of the individual insecticides; if not, we can move on to the next differential. In poisoning with certain carbamates, it is theoretically possible for the carbamoylated enzyme to hydrolyze the toxicant, freeing up the enzyme and resulting in a false negative result. This seems to be very rare, and in 28 years in toxicology, I've only recognized it twice. To the best of my knowledge its never been demonstrated with an organophosphate agent.

It is also possible that the physico-chemical properties of a particular cholinesterase inhibitor preclude it crossing the blood brain barrier, resulting in a normal brain cholinesterase activity despite lethal poisoning. This is the case with some of the nerve agents and the cyanobacterial toxin anatoxin A_S and may have been responsible for the initial failure to diagnose nerve gas poisoning in the Skull Valley episode 40 years ago. The converse, i.e. brain damage without blood inhibition does not seem to occur.

In this case the blood cholinesterase activity contradicts the clinical signs and chemical analysis. The stomach contents were negative at a concentration of less than 1 ppm in repeated analysis, despite confirmation of famphur in the vomitus by GC/MS. Conceivably, the dog ingested a toxic dose which inhibited critical synapses in the brain and autonomic nervous system, but expelled it quickly enough that the blood cholinesterase was recovering when it died. Conceivably, and despite repeated quality control checks, the cholinesterase test was erroneous. We have no way of knowing exactly what happened. This case does suggest, however, that it would be a good idea to order both chemical analysis for organophosphates and cholinesterase activity whenever you really need to know whether an animal was poisoned.

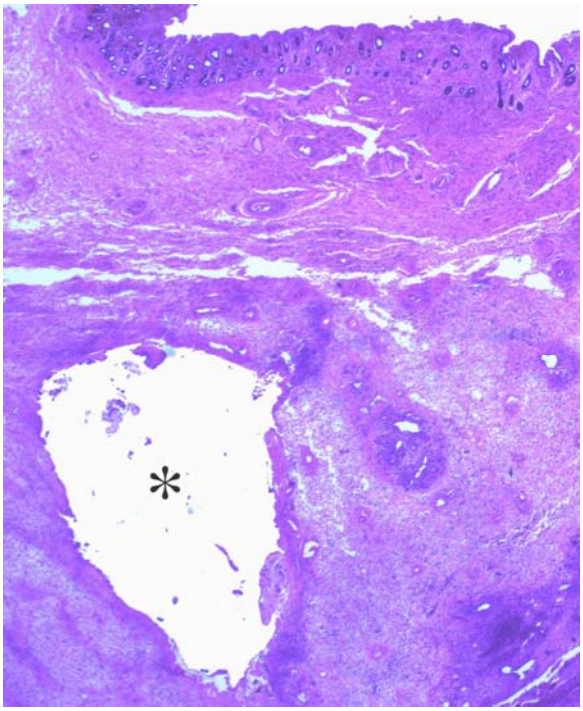
Dr. Merl Raisbeck

Cuterebral myiaisis in Wyoming dogs

Cuterebra are warble flies and dermal parasites of rodents and rabbits. They occasionally infect dogs, cats, swine, horses, mules, mink, foxes and people. In these species they generally occur in skin. On occasion they migrate to the brains or eyes, causing fatal infection.

Eggs are laid on the ground near nests or grass trails used by rodents and rabbits. Larvae enter the skin or through natural orifices, and migrate subcutaneously. After 3 - 7 weeks of migration, a discreet warble-like swelling is formed. In rodent hosts, this is near the anus, scrotum or tail. Once mature, larvae migrate through the skin, drop to the ground and pupariate. We saw several cases of cuterebriasis this summer. A typical case was a 5-year old Husky that developed a mass in the rectal area. The veterinarian found larvae in the mass and submitted the lesion. Histologically, there was a cyst-like space that contained large numbers of bacteria adherent to the wall.

Adult Cuterebra are large (20 mm) dark blue or black non-feeding flies with fine, dense hairs on the face, genae and thorax. One genus, Cuterebra, with 26 species is currently recognized in North America. Taxonomy is poorly defined. Existing keys are often inadequate for laboratory separation of species. The parasites are usually not seen by rabbit hunters since larvae drop off before hunting season.



Skin from the perineum of a dog with an intralesional Cuterebra sp. tract (asterisk). The larva was removed prior to processing. Large numbers of bacterial cocci were adherent to the fistulous tract and account for the secondary infections commonly seen in dogs with Cuterebra infections

Dr. Donal O'Toole

Corona Virus in Cats

Infection of cats with feline coronavirus is very common. Up to 90% of cats living in catteries and up to 50% of solitary cats may be positive serologically. There are many types of coronaviruses of variable virulence, some producing undetected infections which eventually lead to seroconversion. A low number of seropositive animals will develop FIP (Feline Infectious Peritonitis) which is associated with high virulence strains. The disease is characterized by a fibrinous-granulomatous serositis with protein rich

effusions into body cavities. Granulomatous necrotising phlebitis and periphlebitis and granulomatous inflammatory lesions are also observed in several organs.

Currently available serologic tests do not differentiate among strains with variable virulence. Thus, positive serology in an animal with no FIP symptoms/lesions has to be analyzed in the context of the clinical disease. On the other hand, a symptomatic animal may rarely not show antibodies against Feline Coronaviruses in which case another test like Polymerase Chain Reaction (PCR) may help to confirm the diagnosis.

The virus produces two clinical forms of the disease, one non-effusive (dry) and another effusive (wet). In both cases it has been observed that the total protein concentration is usually increased to values between 35 to 40 g/L in FIP animals. The globulin is the highest, pushing down the A:G (Albumin: Globulin) ratio. Ratio values of <0.4 indicate FIP, >0.8 rule out FIP. The sample needed to run this ratio is effusion liquid in the wet form and serum or plasma in dry FIP.

Another available test is based on the determination of AGP in blood (Alpha Acid Glycoprotein) which is an acute phase protein which consistently shows an increase in FIP cases to values up to 1500 mg/ml (normal 500 mg/ml). These laboratory determinations are very indicative of FIP and thus are highly recommended to complement the serology tests. The WSVL runs an ELISA based test for determination of antibodies against feline coronavirus. We recommend adding a determination of the A:G ratio or AGP in blood to confirm FIP in an animal. Samples for testing the A:G ratio may be referred to the WSVL Clinical Pathology Laboratory Laboratory and requests for AGP may be referred to Cardiotech Services (http://www.cardiotechservices.com).

Dr. Ana Bratanich

Brucella ovis testing

If you remember back a couple years ago the WSVL ceased to offer the ELISA test for B. ovis because of inconsistent results. The USDA did work on improving the assay so we again offered the test but it can still be a problem. This year has been especially problematic with a number of animals turning up as suspect even though they are probably uninfected. These false reactions are assumed to be antibodies in the serum to another bacteria that cross react in the ELISA. Laboratories do a number of things to try and eliminate or ignore these cross-reactive antibodies but there is not much consistency between the various laboratories that perform the test. The bottom line is that we will continue to offer the test as long as veterinarians and producers realize the limitations of the assay. Utah State University Extension service has produced an animal health fact sheet that provides useful information and can be found at http://extension.usu.edu/files/publications/factsheet/AH_Sheep_14.pdf
Again, we are doing our best with an assay that isn't the best but it is all we have.

Ken Mills/Becky Wills

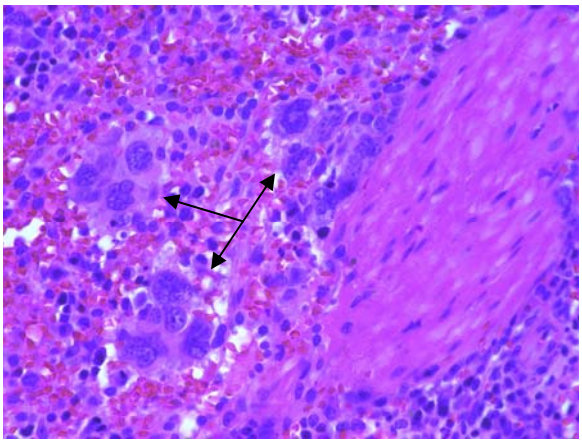
Thrombocytopenia in dogs - are you seeing it?

A veterinarian in Casper reports seeing an increase in the number of cases of thrombocytopenia in otherwise healthy dogs over the past year. We recently received

samples from one such dog, which tested negative for more common causes of infectious and immune-mediated thrombocytopenia.

Histologically there was moderate diffuse erythropoiesis, granulopoiesis and megakaryocyte hyperplasia. No infectious agents were seen. Additional testing is in progress.

If any of you think you are seeing an increase occurrence of thrombocytopenia in dogs, particularly if it seems to be geographically specific, please contact the laboratory.



Spleen from a dog with thrombocytopenia. Note the marked increase in number of megakaryocytes (arrows).

Dr. Donal O'Toole

PARTING NOTES

The world we live in has certainly changed since the current WSVL facilities were commissioned in 1984. The constant threat of bioterrorism and the emergence and re-emergence of high-impact animal pathogens including foreign animal and zoonotic diseases have dramatically changed the face of diagnostic veterinary medicine. Those of you who have recently visited WSVL have probably realized that our current facilities are crowded and have serious shortcomings in this modern era of state-of-the-art diagnostics and focus on biosecurity. During the past four months, the Veterinary Sciences Department, University, and the Wyoming Game and Fish Wildlife Disease Lab undertook an ambitious renovation and construction design program to address shortcomings in the existing facility and that would incorporate a new, secure, and much improved biosafety level-3 laboratory suite. The new BSL-3 facility would enable us to effectively and safely perform diagnostics for zoonotic and high-impact animal pathogens. Overall, the conceptual design would ease overcrowding, increase building security, and provide a safe and efficient workplace for diagnostic testing. Despite the fact that the final product was a no-frills, bare-bones effort, the final dollar figure was considerably over the projected allowable budget. This was not, however, a wasted effort. The concept envisioned by the design team is a workable solution and can be used as the basis for future planning. The importance of support from Wyoming's animal industries and veterinarians in this endeavor should not be overlooked. In turn, WSVL must maintain a reputation for excellent service and responsiveness to the animal industries of Wyoming.

Dr. Don Montgomery

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