

# Lab Notes



August 2013

## Meet Our Staff

Mariann Clark, CVT joined our lab in 2001. She graduated from UCA with a bachelor of education. After graduation, she moved to New England where her husband was in the Navy. She managed a boarding kennel for three years while earning an associate degree in veterinary technology. Mariann was a vet tech for seven years at an animal hospital in New Hampshire. She currently works in our clinical pathology department.

When not working, Mariann enjoys walking her four dogs, reading and knitting. She gets her twice weekly "horse fix" at Hearts and Hooves, a therapeutic riding center in Sherwood, AR.

NEWSLETTER OF THE  
ARKANSAS LIVESTOCK  
AND POULTRY  
COMMISSION  
VETERINARY  
DIAGNOSTIC LAB

Dr. James O. Britt, Laboratory  
Director.



## Helpful Hints from the Lab-

PLEASE be sure to fill out the submission sheets completely. It is very important that the lab knows the location of samples and the species of the animal. Without the species information we cannot provide a correct range of normal values.

## Blackleg in Cattle

We have had 2 cattle herd outbreaks of blackleg (*Clostridium chauvoei*) recently, Cabot and Greenwood, AR areas. In each, 2-4 animals were found dead over the course of a day. Muscle lesions were not found in either case, but the F.A. test using spleen was positive for this bacterium on both. We are calling these presumed cases with a history that fits and lack of vaccination. Unvaccinated cattle are most susceptible from 6 months to 2 years of age. The spores live in the ground for years so it is always somewhat puzzling why we see several dying at one time, but that is typical with our lab submissions.

### Splenic Masses in Dogs

The most common splenic masses that we receive as biopsy material are subcapsular or deeper hematomas and hemangiosarcomas (HSAs) that may have hematoma formation in them, and occasional splenic hyperplastic nodules. Hematomas are probably the more common. These are sometimes submitted as the "5 lb. tumor in the 15 lb. dog." They are often caused (some think) by formation of extramedullary hematopoietic tissue in the red pulp as an often normal finding in older dogs. This makes the spleen more fragile and susceptible to trauma. In a few cases where we had information, the dog had just jumped from the sofa or had been kicked by a horse but usually there is no known recent trauma. Old or remote hematomas eventually shrink as the blood is absorbed and eventually there is a "siderotic plaque" on the capsular surface. This is an incidental lesion with a pale or yellowish tinge but it might be confused with a significant finding when doing exploratory surgery so we commonly get these as biopsy samples.

Hemangiosarcomas usually have extensive hematoma formation so they can't be distinguished from hematomas without histopathology. Send at least several sections from around the mass in formalin because the amount of actual tumor tissue can be relatively small compared to the hematoma. HSAs usually eventually metastasize to the liver, so look for possible metastases on the liver surface during surgery. The tumor vessels can cause fragmentation anemia so torn RBCs (schistocytes) with anemia and neutrophilia might be seen in a blood smear and CBC. By the way, an odd tumor is the hemangiosarcoma of the right atrium of the heart in dogs. These either rupture and cause sudden death from hemopericardium, or widely metastasize throughout the lungs and might bring on respiratory signs and a radiograph that resembles diffuse pulmonary blastomycosis. This is something we look for as an explanation of sudden death in an older dog.

Splenic hyperplastic nodules (benign "splenomas") are usually a single, discrete, somewhat raised mass. Microscopically, they consists of active lymphoid follicles with immature lymphocytes. These can be difficult to distinguish from malignant lymphomas without knowing that it is a single mass and without having some normal splenic tissue in the biopsy section. So, if you are doing exploratory surgery, finding a mass in the spleen may or may not be the answer you are looking for unless there is obvious tumor in the liver or severe blood loss from hematoma rupture or its acute expansion.

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## Cytology Exams:

Samples submitted for cytology are best prepared as thin smears on glass slides that are either quickly air dried or fixed. The goal is to prepare a thin film in which the cells are spread out into a single layer. Glass slides with frosted-ends and labeled with pencil are preferred. Glass slides can be submitted stained or unstained. Wright's Stain is used for all cytologies and blood smears stained at the lab, and is usually preferred to Diff Quik. If the sample is sufficient to prepare multiple slides, then it is best to send at least 2 or more unstained slides. The more slides submitted, the more likely the sample will be diagnostic. If the sample is scant and not readily visible on the slide, then circling the sample with a grease pencil is helpful, since it reduces the time spent locating the sample on the slide. Samples containing lipid appear wet with glistening droplets that do not dry completely. Alcohol fixatives used in Romanowsky stains dissolve lipid, often leaving these slides devoid of cells. Heat fixing lipid samples prior to shipping to the lab may increase their diagnostic yield.

Blood contamination is a common cause of non-diagnostic samples. Prolonged aspiration and the use of too large a needle are the primary causes of blood contamination. The presence of visible material within the needle hub is an indication of prolonged aspiration. Any time material becomes visible within the hub, the collection procedure should be stopped and slides made immediately. In order to minimize blood contamination, it is best to use a 22 to 25 gauge needle. Larger-bore needles are more likely to rupture small vessels and usually do not collect more cells. If blood contamination occurs, it is best to stop the procedure and reaspirate the sample using a clean syringe and needle. Some lesions are highly vascular, making it difficult to avoid blood contamination even with good technique. In these cases, the use of a non-aspiration technique may reduce blood contamination and improve diagnostic yield.

In the standard aspiration procedure, the mass is stabilized, and a 22-25 gauge needle attached to a syringe is inserted into the center of the mass. The sample is collected by pulling back on the plunger and applying negative pressure for a few seconds. In the non-aspiration technique, the sample is collected without using negative pressure. This technique can yield samples of equal or better quality than those obtained with standard aspiration. A 22-25 gauge needle is used either by itself or with a syringe attached that contains air. The needle is inserted after stabilizing the mass, and the needle is quickly moved back and forth in a stabbing motion, trying to stay along the same tract.

After collection via standard aspiration or the non-aspiration technique, a syringe containing air is used to expel the contents of the needle hub onto a glass slide. A squash prep is the most common technique used to smear tenacious or semi-solid material. A spreader slide is placed perpendicularly over the sample slide, and without the use of downward pressure, the spreader slide is lightly drawn across the length of the sample slide.

The same technique that is used to make a blood smear should be used for samples with a liquid or plasma-like consistency. This usually includes lymph node aspirates which are especially prone to rupture with the squash technique. The use of the blood smear technique with fluid-like samples will result in less cell rupture and will typically produce a thin smear with intact cells that are adequately spread out.

When generalized lymphadenopathy is present, it is best to sample lymph nodes other than the head and neck nodes, which are under constant immune stimulation with reactive change that can be difficult to distinguish from neoplasia.

Cytology can be performed on any fluid sample including urine. However, the complete fluid analysis is only appropriate for thoracic, abdominal, joint and cerebrospinal fluids. Even though a complete urinalysis includes a microscopic examination, the sediment exam in a urinalysis is not the same procedure used for cytology. Cytology of urine and other fluids of low cellularity is performed using a cytospin, which is a low-speed centrifuge that allows the sample to be concentrated directly on the slide with minimal cell destruction. Urine cytology is usually indicated when neoplasia is suspected.

All fluid samples submitted for complete fluid analysis (i.e. thoracic, abdominal, joint and cerebrospinal fluid) should include direct smears prepared at the time of sample collection along with a lavender top (EDTA) and a red top tube. EDTA prevents clot formation and also acts as a preservative, slowing down cell deterioration. If the sample clots, then an accurate cell count cannot be obtained, and if the sample deteriorates, then morphology will be almost impossible to evaluate. It is also important to place a sufficient amount of the fluid in the EDTA tube, otherwise, if the sample is too small, the total protein estimation will be artifactually elevated. A red top tube is required to run any chemistries indicated for the fluid, and a portion of the sample from the red top can also be used for culture if required.

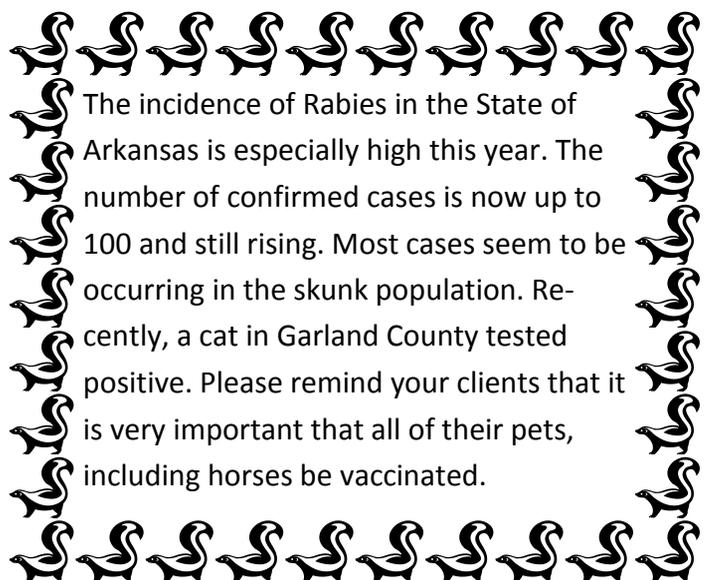
### Fast Facts-

The domestic cat is the only species of cat able to hold its tail vertically while walking. Wild cats hold their tail horizontally, or tucked between their legs while walking.

The cells which make up the antlers of a moose are the fastest growing animal cells in nature.

Mosquitoes prefer children to adults, and blondes to brunettes

Each year, Americans spend more on cat food than on baby food.



The incidence of Rabies in the State of Arkansas is especially high this year. The number of confirmed cases is now up to 100 and still rising. Most cases seem to be occurring in the skunk population. Recently, a cat in Garland County tested positive. Please remind your clients that it is very important that all of their pets, including horses be vaccinated.